Evidence Based Assessment of the Cardiovascular Adverse Effects of Specific Cyclooxygenase-2 Inhibitors

AIKATERINI LALATSA

Master of Philosophy

Aston University

September 2006

Aston University
SUMMARY

Evidence Based Assessment of the Cardiovascular Adverse Effects of Specific Cyclooxygenase-2 Inhibitors

A thesis submitted to Aston University for the degree of Master of Philosophy in the School of Health & Sciences

Aikaterini Lalatsa, MPharm, September 2006

Objective: To provide a quality assessment and a systematic review of all the meta-analysis and systematic reviews of the cardiovascular and related safety of rofecoxib for its licensed long-term indications.

Method: Systematically search Pubmed, Cochrane, FDA, NICE, EULAR, CCOHTA, to identify systematic reviews concerning rofecoxib's cardiovascular toxicity. The resulting set of citations was hand searched by title, MESH, and whenever available abstract. Reference lists of recovered articles were further screened for any additional citations. Quality was assessed using the QUOROM (Quality of Reporting of Meta-analyses) checklist. Quality scores were devided into quartiles (<25% i.e. very poor, 25-49% i.e. poor, 50-75% i.e. acceptable and >75% i.e. good quality) to assist judgement of quality.

Results: 15 systematic reviews were included with a total of 43,343 patients. The mean overall quality of reporting score is acceptable [63.13%, (SD: 21.41%, range = 33.3-94.4%)]. The overall score for Title (27.30%) and Results (45.46%) were poor, while overall scores for Abstracts (69.70%), Methods (71.21%) and Discussion (63.60%) were acceptable. Nine systematic reviews utilised manufacturer's files and 8 of these either received funding or the authors were employed by the manufacturer. Only 4 systematic reviews reported an end search date after 2000, when the results from the VIGOR trial became available. Cumulative meta-analysis was not performed prior to rofecoxib's withdrawal. Rofecoxib was compared with few active comparators whose cardiovascular safety is not established. Majority of patients were female, caucasians and younger than in actual practice (<65 years old) and exposed to rofecoxib for a median of only 6 weeks, a duration inadequate to assess long-term safety.

Conclusions: The worldwide withdrawal of rofecoxib emphasises further the need for adherence to standardised reporting and quality guidelines to allow researchers to synthesise available information in quantitative and unbiased manner allowing for timely and appropriate decisions. Limitation of available evidence combined with a lack of adequate pharmacovigilace independent from manufacturer's interest delayed the recognition of rofecoxib's cardiovascular adverse events. Design of prospective cardiovascular toxicity evaluations should test patients that resemble the population likely to receive the drug in clinical practice using composite outcomes for at least a year comparing the agent with placebo and established comparators.

Key words: Rofecoxib, Cardiovascular Toxicity, Systematic Review, Quality, QUOROM guidelines.

To my parents, Lisa and Stylianos,

to my brother Antonis and to my

friends Maria, Ida, Hissham,

Sophia and Kiki

ACKOWLEDGEMENTS

I would like to thank my supervisor Professor Keith Wilson for his help and guidance and willingness to aid me in carrying out and finishing this project.

Special thanks to all my research colleagues in the Centre of Evidence-Based Pharmacotherapy and especially to Ying, Shamyla, Sumiah, Rita and He Yi for all their support and encouragement during this work.

In this instance, I also would like to acknowledge Professor Helen Griffiths for all her crucial help and support.

Finally, I would like to thank the people working in the Pharmacy Department in Birmingham Children's Hospital as if it was not for them, things might have taken a different turn.

LIST OF CONTENTS

Abbreviations

Å	Angstrum
AA	Arachidonic Acid
ACR	American College of Rheumatology
ACE	Angiotensin Converting enzyme
AD	Alzheimer's Disease
ADR(s)	Adverse Drug Reaction(s)
AIMS	Arthritis Impact Measurement Scales
AMI	Acute Myocardial Infarction
APC	Adenomatous Polyposis Coli (gene)
Arg	Arginine
ARR	Absolute Risk Reduction
ASA	Aspirin, acetylosalicylic acid
A/Es	Adverse Effects
AUC	Area Under the Curve
BP	Blood Pressure
BD / bd	Twice a day
cAMP	Cyclic Adenosine Monophosphate
CB	Cerebrovascular
CCOHTA	Canadian Co-ordinating Office for Health Technology Assessment
CCTR	Cochrane Central Register of Controlled Trials (bibliographic database)
CEA	Cost-Effectiveness Analysis
GFR	Glomerular Filtration Rate
CHF	Congestive Heart Failure
CI	Confidence Interval
CINAHL	Cumulative Index to Nursing & Allied Health Literature
CLASS	Celecoxib Long-Term Arthritis Safety Study
Cmax	Maximum Plasma Concentrations
CNS	Central Nervous System
CONSORT	Consolidated Standards of Reporting Trials
COX	Cyclo-oxygenase, Cyclooxyeganse
COX-1	Cyclo-oxygenase-1, Cyclooxyeganse-1
COX-2	Cyclo-oxygenase-2, Cyclooxyeganse-2
CSM	Commission on Human Medicines
Css	Concentration at Steady State
CV	Cardiovascular
cTALH	Cortical Thick Assending Loop of Henle
DBP	Diastolic Blood Pressure
DMARDs	Disease-Modifying Anti-Rheumatic Drugs
EBM	Evidence Based Medicine
EBP	Evidence Based Pharmacotherapy

EC	Vascular Endothelium
EMBASE	Excerpta Medica (database)
ER	Endoplasmic Reticulum
EU	European Union
EULAR	European League Against Rheumatism
F	Oral Bioavailability Factor
FAP	Familiar Adenomatous Polyposis
FDA	Food Drug Administration
GI	Gastrointestinal
Gly	Glycine
H(s), h(s)	Hour(s)
HAQ	Health Assessment Questionnaire
HEED	Health Economic Evaluations Database
15-HPETE	15-hydroxyeicosatetranoeic acid
HOX	Hydroperoxidase
HT	Hypertension
HTA	Health Technology Assessment
IC 50	Inhibitory Concentration 50
IV	Intravenous
IP	Prostacyclin Receptor
kg	Kilogram(s)
L	Litre
LDL	Low-Density Lipoprotein
LOX	Lypooxygenase
LPS	Lipopolysaccharides
m	Million
μg	Microgram
MBD	Membrane Binding Domain
MEDLARS	Medical Literature Analysis and Retrieval System
MEDLINE	MEDLARS Online
MESH(s)	Medical Subject Heading(s)
MHRA	Medicines and Healthcare products Regulatory Agency
MI	Myocardial Infarction
NAG-1	NSAID-activated gene 1
N/A	Not applicable
NHS EED	National Health Service Economic Evaluation Database
NICE	National Institute of Clinical Excellence
NRR	National Research Register
NSAID(s)	Non Steroidal Anti-inflammatory Drug(s)
NTT	Number Needed to Treat
OA	Osteoarthritis
OD / od	Once a day
OMERACT	Outcome Measures in Rheumatoid Arthritis Clinical Trials
OR(s)	Odds Ratio(s)

PG(s)	Prostaglandin(s)
PGD2	Prostaglandin D2
PGE2	Prostaglandin E2
PGF2a	Prostaglandin F2a
PGG2	Prostaglandin G2
PGHS	Prostaglandin Enderoperoxidase Synthase
PGH2	Prostaglandin H2
PGI2	Prostaglandin I2; Prostacyclin
PHHP	5-phenyl-4-pentenyl-1-hydroperoxide
POX	Peroxidase
PPIs	Proton Pump Inhibitor(s)
PUB	Perforation, Ulcer and Bleeding
PubMed	Text-based search & retrieval system for MEDLINE and preMEDLINE
QDS / qds	Four times a day
RA	Rheumatoid Arthritis
RCT(s)	Randomised Controlled Trials
RF	Rheumatoid Factor
RR	Relative Risk
SBP	Systolic Blood Pressure
SD	Standard Deviation
Ser	Serine
SOP	Standard Operating Procedure
TDS / tds	Three times a day
Tmax	Time to achive Maximum Plasma Concentrations
tNSAIDs	Traditional NSAIDs
TXA2	Thromboxane A2
Tyr	Tyrosine
US(A)	United States of America
Val	Valine
VAS	Visual Analog Scale
VIGOR	Vioxx Gastrointestinal Outcomes Research
WBA(s)	Whole Blood Assay(s)
WOMAC	Western Ontario and McMaster Universities Osteoarthritis Index

List of Tables

List of Figures

Glossary

Statistics

A brief explanation of various statistical terms used in reporting of clinical trials outcomes is provided to ensure understanding of reported results of the analysed systematic reviews and / or meta-analyses.

P-value

The P-value is the result of the statistical test used to assess the probability that the result of the trial is a real effect and did not occur purely by chance. By convention a P-value below 0.05 (a one in 20 likelihood that the result occurred by chance) is accepted as indicating a true difference and is described as a statistically significant result.

95 per cent Confidence Interval (95% CI)

Trials have some degree of uncertainty because a result from a trial on a sample would not be exactly the same if the intervention were applied to a whole population. The CI around the result represents the range of values within which the true population value lies. By convention 95% CI are used, which means that you can be 95 % sure that the true result lies between a certain range.

Intention to treat analysis

An intention to treat analysis means that the results used include all the original patients, including those who have dropped out of the trial. This type of analysis reflects more closely a real life situation where some patients are not compliant with therapy.

Outcome reporting

The benefit or harm of a treatment can be expressed in various ways. With the following simple example it is easy to demonstrate their meaning:

1. Drug X produced an absolute reduction in deaths by 7.12 per cent ("absolute risk reduction")
2. Drug X reduced the death rate by 28.46 % ("relative risk reduction")
3. Drug X increased the patients' survival rate from 75 to 82 %
4. 14 people would need to be treated with drug X to prevent one death ("number needed to treat")

Some of the above statements may sound more impressive than the others, however, all of the above relate to the same results (Table G lists the outcomes of a hypothesised clinical trial as an example for the calculations). The way in which results are presented may affect the way they are perceived. An understanding of the principles underlying the expression of results in terms of relative or absolute risks is invaluable when assessing trials. In clinical practice the "number needed to treat" is the most useful expression of results.

Table G: Clinical trials results for example calculations

Groups	Total number of patients randomized to each group	Outcome at 4 years
Intervention group	**3000**	**537 dead**
(received drug X)		**2,463 alive**
Control group	**2998**	**750 dead**
(received placebo)		**2,248 alive**

Absolute Risk Reduction (ARR)

The absolute risk reduction (ARR) is the absolute amount by which drug X reduces the risk of death, calculated as:

ARR = (event rate in control group – event rate in intervention group) x 100

= (750/2998 – 537/3000) x 100

= 7.12%

i.e., drug X produced an absolute reduction in death by 7.12 %.

Relative Risk Reduction (RRR)

The relative risk of an outcome is the chances of that outcome occurring in the treatment group compared with the chances of it occurring in the control group. If the chances are the same in both groups, the relative risk is 1. The relative risk reduction (RRR) is the amount by which the risk (death in this case) is reduced by drug X as a comparative percentage of the control, calculated as:

$$RRR = \frac{[(\text{event rate in the control group} - \text{event rate in intervention group}) \times 100]}{(\text{event rate in the control group})}$$

= [(750/2998 – 537/3000) x 100] / (750/2998)

= 28.46%

i.e., drug X reduced the death rate by 28.46%

Relative comparisons make the results sound more impressive and this is a tactic often used by manufacturers. On the other hand, absolute comparisons may be used to make the risk of side effects sound smaller.

Number needed to treat (NTT)

The "number needed to treat" (NTT) is the number of people who need to be treated to produce one additional successful outcome.

NNT = 100 / ARR

= 100 / 7.12

= 14

i.e, 14 people would need to be treated with drug X to prevent one death at 4 years.

Odds Ration (OR)

The odds of an event compares the probability of the event occurring with the probability that it will not occur. If the odds are greater (or less) than 1, an event is more (or less) likely to happen. The OR is the ratio of patients in the treatment group succumbing to a particular end point of the trial to the number who do not, compared with the equivalent patients in the control group. An OR of 1 would mean that drug X had no effect i.e., there was no overall difference in outcomes between the intervention and control group.

$$OR = \frac{\text{odds of death / odds of survival in intervention group}}{\text{odds of death / odds of survival in control group}}$$

= (537/2463) / (750/ 2248) = 537 x 2248 / 750 x 2463

= 0.65

Chapter 1

Introduction

1.1 Background

The anti-inflammatory, analgesics and antipyretic drugs are a heterogeneous group of compounds, often chemically unrelated (although most of them are organic acids), which nevertheless share certain therapeutic actions and side-effects. Often they are related as aspirin-like drugs as aspirin (ASA) was the prototype for these drugs as well as due to the similarity of their therapeutic actions to ASA. However, they are widely known today as non-steroidal anti-inflammatory drugs (NSAIDs), because they were clearly distinct from the glucocorticoids (the other major group of agents used in the treatment of inflammation). NSAIDs are widely used in general practise for the treatment of musculoskeletal diseases like arthritis, representing one of the most commonly prescribed class of drugs in the U.K. and worldwide. In 1999, over 18.5 million NSAIDs treatments were prescribed in England at a cost of approximately £ 170m. (Watson, Brookes et al. 2000).

1.2 Arachidonic Acid Pathway

Among the many mediators of inflammation, the prostaglandins (PGs) are of great importance. Prostanoids are members of a large group of hormonally active, oxygenated C_{18}, C_{20}, and C_{22} fatty acids collectively known as eicosanoids that are derived from ω3 (n-3) and ω6 (n-6) polyunsaturated fatty acids and include: (i)

prostanoids formed through cyclooxygenase pathways; (ii) leukotrienes (Yokomizo, Izumi et al. 1997; Sarau, Ames et al. 1999), lipoxins (Serhan, Takano et al. 1999), hepolixins (Pace-Asciak, Reynaud et al. 1999), and monohydroxy fatty acids (Mueller, Andberg et al. 1998) produced via lipooxygenase pathways; (iii) epoxy and dihydroxy fatty acids formed from cytochrome P450s (Chen, Wang et al. 1999) and (iv) isoprostanes (Morrow, Zackert et al. 1999; Pratico, Ferro et al. 1999), isoleukotrienes, and other peroxidised fatty acid products (Khaselev and Murphy 1999), that are formed nonenzymatically (Smith, DeWitt et al. 2000).

Arachidonic acid (20:4, n-6) is the major prostanoid precursor. The biosynthesis of prostanoids involves a three-step sequence (Figure 1.1) of stimulus-initiated hydrolysis of arachidonate from glycerophospholipids involving secretory, cytoplasmic or both types of phospholipase A_2 ($sPLA_2$, $cPLA_2$) (Shinohara, Balboa et al. 1999); oxygenation of arachidonate, yielding prostaglandin endoperoxide H_2 (PGH_2) by PGHSs (prostaglandin endoperoxidase synthases); and conversion of PGH_2 to the most important biologically active end products, PGD_2, PGE_2, PGF_2a, PGI_2 (prostacyclin), or TxA_2 (thromboxane A_2) via specific synthases (Hara, Miyata et al. 1994; Kuwamoto, Inoue et al. 1997; Suzuki, Watanabe et al. 1997; Smith, DeWitt et al. 2000). These end products act as autocrine and paracrine mediators for a broad range of physiological and pathophysiological responses.

Prostaglandin endoperoxidase synthase (PGHS), also known as cyclooxygenase (COX), catalyses the first committed step in the conversion of arachidonic acid (AA) to PGs and thromboxanes (Smith and DeWitt 1995). A homogenous, enzymatically active COX or PGHS was isolated in 1976 (Hemler and

Lands 1976). This membrane-bound haemo-& glucoprotein with a molecular weight of 71kDa is found in greatest amounts in the endoplasmic reticulum of prostanoid-forming cells (Smith 1986). It exhibits COX activity which both cyclizes AA and adds the 15-hydroxyperoxy group of prostaglandin G_2 (PGG_2) (Figure 1.2) (Bazan, Botting et al. 1996). The hydroxyperoxy group of PGG_2 is reduced to the hydroxy group of PGH_2 by a peroxidase that utilises a wide variety of compounds to provide the requisite pair of electrons. Thus, PGG_2 diffuses from the cyclooxygenase active site and binds at the peroxidase active site, where it is reduced to the hydroxy endoperoxidase PGH_2, the precursor of PGs, thromboxanes and prostacyclin (Smith, DeWitt et al. 2000). Figure 1.3 illustrates the production and actions of PGs. (FitzGerald and Patrono 2001). The resulting products then exit the cells via a carrier mediated-process (Chan, Satriano et al. 1998) to activate prostanoid G-protein-linked receptors (Murata, Ushikubi et al. 1997; Sugimoto, Segi et al. 1998; Ushikubi, Segi et al. 1998), or in some cases may interact with nuclear receptors (Lim, Gupta et al. 1999).

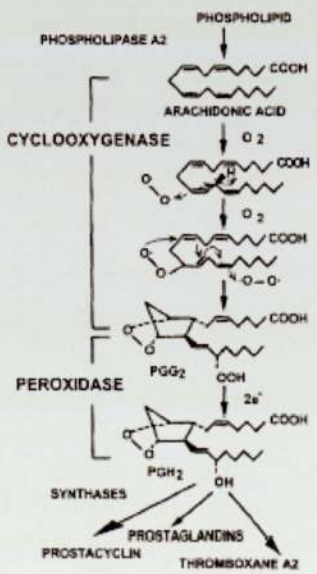
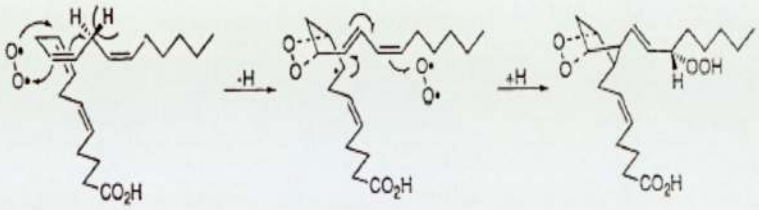


Figure1.1 Biosynthetic pathway for the formation of prostanoids derived from arachidonic acid (Smith, DeWitt et al. 2000)



Chemical steps in the conversion of arachidonic acid to PGG_2. The enzyme removes the 13-pro-*S*-hydrogen, which generates a pentadienyl radical with maximal electron density at C-11 and C-15. Trapping of the carbon radical at C-11 with O_2 produces a peroxyl radical, which adds to C-9 generating a cyclic peroxide and a carbon-centered radical at C-8. The C-8 radical adds to the double bond at C-12, generating the bicyclic peroxide and an allylic radical with maximal electron density at C-13 and C-15. Trapping of the carbon radical at C-15 with O_2 generates a peroxyl radical which is reduced to PGG_2.

Figure 1.2 Chemical steps in the conversion of AA to PGG_2 (Marnett, Rowlinson et al. 1999)

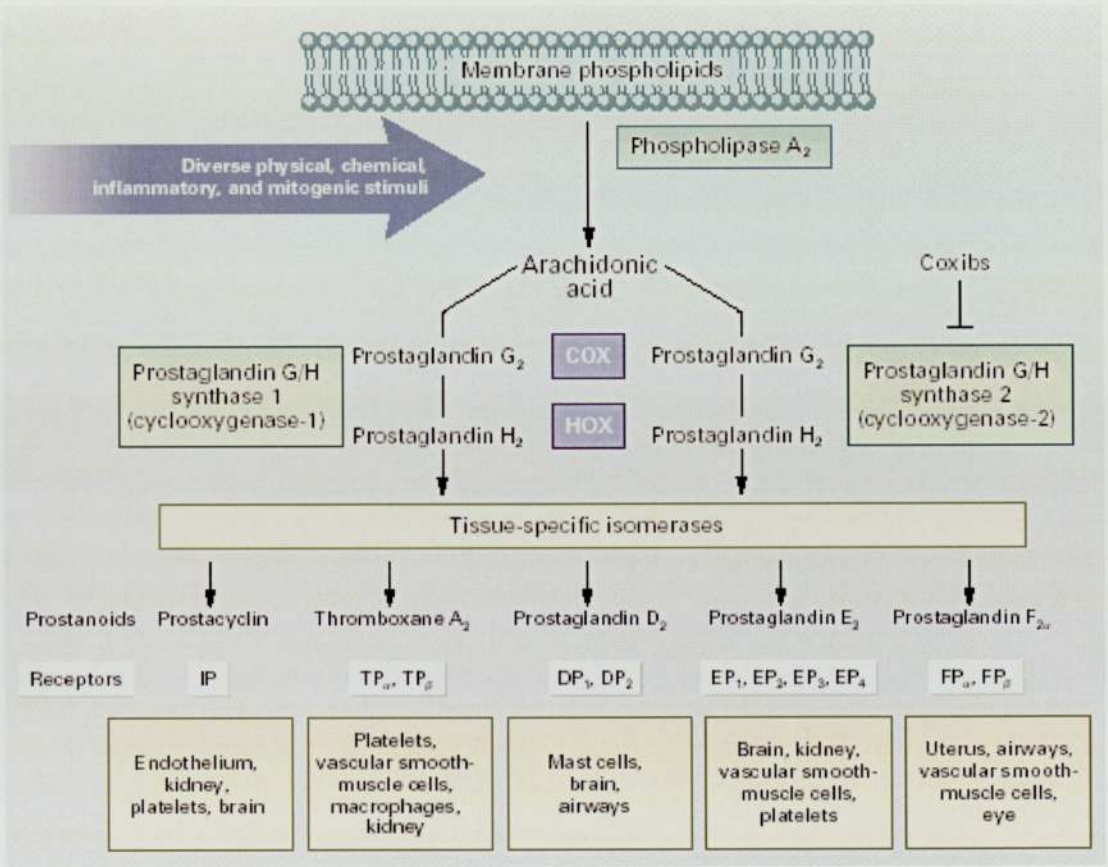


Figure 1.3 **Production & actions of PGs and Thromboxane.** AA, a 20-carbon fatty acid containing four double bonds, is liberated from the sn2 position in membrane phospholipids by phospholipase A_2, which is activated by diverse stimuli. AA is converted by cytosolic prostaglandin G/H synthases, which have both COX and hydroperoxidase (HOX) activity, to the unstable intemediate prostaglandin H_2. The synthases are colloquially termed cyclooxygenases and exist in two forms, cyclo-oxygenase-1 and cyclo-oxygenase-2. Coxibs selectively inhibit cylco-oxygenase-2.

Prostaglandin H_2 is converted by tissue-specific isomerases to multiple prostanoids. These bioactive lipids activate specific cell-membrane receptors of the superfamily of G-protein-coupled receptors. Some of the tissues in which individual prostanoids exert prominent effects are indicated. IP denotes prostacyclin receptor, TP thromboxane receptor, DP prostaglandin D_2 receptor, EP prostaglandin E_2 receptor, and FP prostaglandin F_{2a} receptor (FitzGerald and Patrono 2001).

1.2.1 Overview of the peroxidase & cyclooxygenase catalysis

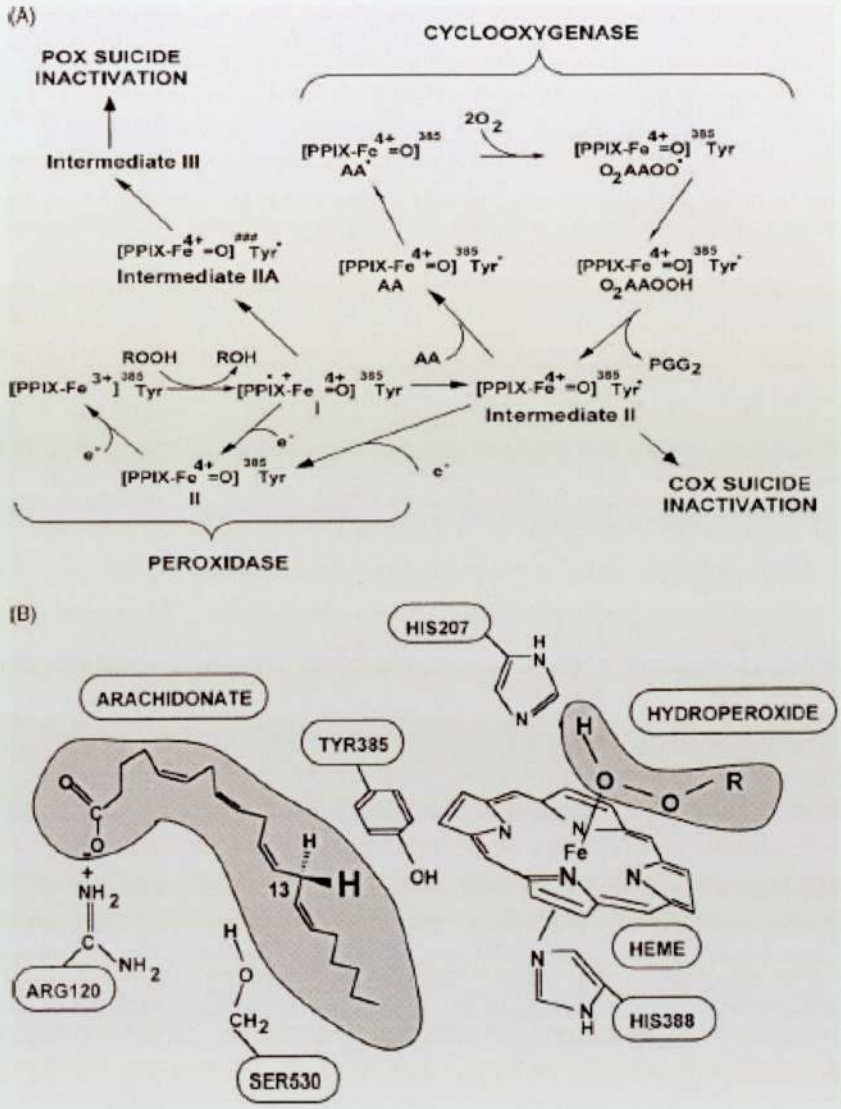
The peroxidase utilises a wide variety of compounds to provide the requisite pair of electrons. Both COX and hydroxyperoxidase activities are contained in the same dimeric protein molecule (Bazan, Botting et al. 1996). Thus, these two reactions occur at distinct but structurally and functionally interconnected sites (Figure 1.4) (Smith, DeWitt et al. 2000). The peroxidase reaction occurs at a haeme-containing active site located near the protein surface (Smith, DeWitt et al. 2000). The cyclooxygenase reaction occurs in a hydrophobic channel in the core of the enzyme (Smith, DeWitt et al. 2000). In vitro, the peroxidase activity can operate independently of the cyclooxygenase (e.g. when the cyclooxygenase site is occupied by an NSAID) (Mizuno, Yamamoto et al. 1982) or during ongoing cyclooxygenase catalysis (Koshkin and Dunford 1999). In contrast, the cyclooxygenase reaction is peroxide-dependent (Smith and Lands 1972) and requires that the haeme group at the peroxidase site undergo a two-electron oxidation (Landino, Crews et al. 1997).

The model shown on Figure 1.4 depicts the spatial interrelationships between catalytically important residues (Smith and Song 2002). The mechanistic model in figure 1.4A was developed by Ruf and co-workers (Dietz, Nastainczyk et al. 1988),

and the basis of this model remains the same today. The haeme group at the peroxidase (POX) site of PGHS undergoes a two electron oxidation by a hydroxyperoxide (e.g. PGG_2) yielding the corresponding alcohol (i.e. PGH_2) and an oxyferryl haeme radical cation (Compound I) (Smith and Song 2002). In the next step, a tyrosine residue (Tyr 385) contributes an electron to compound I producing an oxyferryl haeme and a tyrosyl radical (Intermediate II) (Smith and Song 2002). Finally, the tyrosyl radical abstracts a hydrogen from AA to begin COX cycle of oxygen insertion and cyclisation reactions (Smith and Song 2002). Neither the identity nor the source of the hydroxyperoxide necessary to initiate the first haeme oxidation in vivo is known (Smith and Song 2002). In vitro, there is typically sufficient hydroxyperoxide in commercial fatty acid substrate preparations to initiate the process, and, once started, a hydroxyperoxide (i.e. PGG2) becomes available to continue the process as necessary (Smith and Song 2002). The POX reaction requires a reducing cosubstrate to convert compound I to II, and compound II to the haeme of the resting enzyme (Smith and Song 2002). The identity of the reducing cosubstrate, in vivo, is not known (Smith and Song 2002).

Interestingly, once the COX catalytic cycle has been initiated, it can operate independently of the POX cycle (Koshkin and Dunford 1999). That is, the POX and COX reactions are not tightly coupled in the sense that there is one to one correspondence between peroxide reduction and PGG_2 formation (Wei, Kulmacz et al. 1995). Viewed from another perspective, the oxyferryl haeme group of intermediate II, which is the same as the oxyferryl haeme group of compound II of the POX cycle, can be reduced by one electron originating from a reducing cosubstrate to yield resting

haeme (Fe3+ -protoporphyrin IX) while the COX cycle continues to function (Koshkin and Dunford 1999).



Interrelationships between COX and POX. (A) Mechanistic interrelationships between the COX and POX catalytic cycles. (B) Spatial relationships among catalytically important residues in COX and POX catalysis. AA, arachidonic acid; PPIX, protoporphorin IX; ROOH, an alkyl hydroperoxide (e.g. PGG_2); ROH, an alkyl alcohol; I, compound I; II, compound II.

Figure 1.4 Interrelationships between COX & POX (Smith and Song 2002)

Although COX catalysis requires an initial oxidation of the haeme group at the POX active site, the tyrosyl radical-containing species can continue to cycle at or near maximal efficiency in the absence of COX turnover or occupancy of the COX active

site (Mizuno, Yamamoto et al. 1982; Koshkin and Dunford 1999; Song, Ball et al. 2001). When the cylcooxygenase site is occupied by an appropriate fatty acid substrate such as arachidonate, the tyrosyl radical if intermediate II initiates the cyclooxygenase reaction by abstracting the 13proS hydrogen atom to yield an arachidonate radical (Tsai, Kulmacz et al. 1995).

Although there are still unresolved issues concerning the PGHS mechanism (Tang, Copeland et al. 1997), the branched chain model is still capable to explain all the findings. The most compelling evidence for the branched chain mechanism is that PGG2 can accumulate during catalysis even in the presence of peroxidase-reducing cosubstrates (Wei, Kulmacz et al. 1995). In a clear branched chain mechanism, the tyrosyl radical once formed, would cycle continuously (Figure 1.4) (Smith, DeWitt et al. 2000; Smith and Song 2002). In fact, removal of hydroperoxides after catalysis has been initiated (e.g. upon addition of glutathione peroxidase plus reduced glutathione) stops the cyclooxygenase reaction in midstream (Lu, Tsai et al. 1999). As it can be deducted, cyclooxygenase requires the ongoing presence of hydroperoxides, presumably to regenerate compound I. The continuous need for hydroperoxides implies the intermediate II is reduced to compound II and back to haeme at a rate that competes effectively with the rate of abstraction of the hydrogen atom from arachidonate by intermediate II (Figure 1.4) (Smith, DeWitt et al. 2000). The pathway from intermediate II to compound II may be crucial in preventing untoward accumulation of enzyme radicals. Particularly when substrate is not being provided to the enzyme (Smith, DeWitt et al. 2000).

The phenomenon of suicide inactivation confounds the interpretation of the kinetics and mechanistic data on the peroxidase and cylcooxygenase reactions of PGHSs (Smith, DeWitt et al. 2000). Both the peroxidase and the cyclooxygenase activities are inactivated during catalysis by mechanism-based, first order processes (Smith, Garavito et al. 1996; Wu, Wei et al. 1999; Smith, DeWitt et al. 2000). Thus, PGHS-1 or –2 peroxide or cylcooxygenase activities fall to zero within 1-2 min even in the presence of sufficient substrates (Smith, DeWitt et al. 2000). Although it appears in figure 1.4 that suicide inactivation involves intermediate III (Wu, Wei et al. 1999), this point is not resolved. Peroxidase inactivation is independent of the nature of the oxidising peroxide (Wu, Wei et al. 1999), whereas cyclooxygenase inactivation appears to depend on the nature of the fatty acid substrate (Smith, DeWitt et al. 2000), and thus apparently on the nature of the peroxide. Suicide inactivation originates with a reaction intermediate and likely proceeds from intermediate II involving the formation of a tyrosyl radical other than the Tyr385 radical (Wu, Wei et al. 1999). It should be noted that the rates of both peroxidase and cyclooxygenase suicide inactivation are slowed markedly by peroxidase-reducing cosubstrates (Koshkin and Dunford 1999; Wu, Wei et al. 1999). Reducing cosubstrates may bias the rate of conversion of intermediate II to compound II versus intermediate III (Smith, DeWitt et al. 2000). Suicide inactivation is an interesting chemical phenomenon, but its biological relevance is unclear (Smith, DeWitt et al. 2000). In general, the amounts of PGHSs are in excess of substrate and bursts of prostanoid production by cells do not lead to major losses in PGHS activity (Smith, DeWitt et al. 2000).

1.2.2 Peroxidase Kinetics

The kinetic constants for compound I (oxyferryl haeme radical cation) and compound II/intermediate II (oxyferryl haeme) associated with heterolytic cleavage of alkyl hydroperoxides have been measured for both PGHS-1 and 2. Relatively hydrophobic alkyl hydroperoxides such as 15-HPETE (hydroxyeicosatetraenoic acid) and 5-phenyl-4-pentenyl-1-hydroperoxide (PHHP) exhibit about 10-fold higher secondary rate constants for formation of compound I (~2 x 10^7 mol^{-1} s^{-1}) versus soluble peroxides such as ethylhydroperoxide (Lu, Tsai et al. 1999) and have a lower apparent Km values (~10 μM for H_2O_2) for the peroxidase reaction as measured by rates of oxidation of reducing cosubstrates (Landino, Crews et al. 1997; Smith, DeWitt et al. 2000). Although the second order rate constants K1 for compound I formation with alkyl hydroxyperoxides are approximately the same for both isozymes (~2 x 10^7 M^{-1} s^{-1}), the first-order rate constant for the conversion of compound I to compound II/intermediate II is considerably more rapid for PGHS-2 (Lu, Tsai et al. 1999). This partly accounts for the fact that for PGHS-2, intermediate II is formed more rapidly and at lower peroxide concentrations (Smith, DeWitt et al. 2000). There is no obvious structural explanation for this property (Smith, DeWitt et al. 2000).

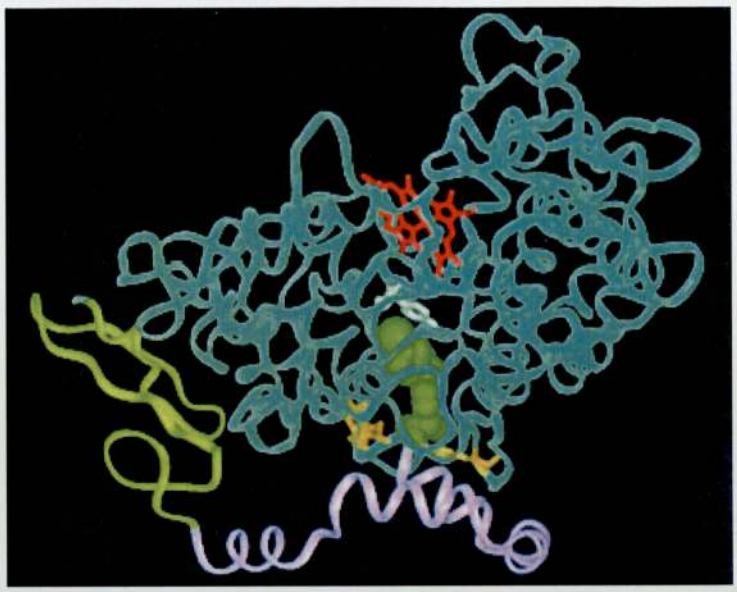
1.3 Cyclooxygenase-1 & Cyclooxygenase-2 structure and role

NSAIDs exert their major therapeutic and adverse-effects by inhibition of COX, a key enzyme in prostanoid synthesis (Vane 1971). By inhibiting PG synthesis, NSAIDs can interrupt the normal autocrine / paracrine signalling necessary for elaboration of the inflammatory response (DeWitt, Meade et al. 1993). Until recently,

all PG synthesis was thought to result from only one form of the COX enzyme (Cryer and Dubois 1998). Before the discovery of COX-2, cyclooxygenases were believed to be expressed constitutively with constant levels in individual tissues; prostaglandin synthesis was believed to increase in inflammation because of increased release of precursor (Hawkey 1999). Thus, the rate-limiting step in prostanoid biosynthesis was the availability of arachidonic acid substrate (Crofford, Lipsky et al. 2000). However, cyclooxygenase activity increases in inflammation, and this increase can be prevented by corticosteroids (Hawkey 1999). From these clues, two different approaches identified a new inducible isoform (COX-2) (Hawkey 1999). Needleman's group detected a different cyclo-oxygenase protein in monocytes stimulated by interleukin 1 (Fu, Masferrer et al. 1990). A molecular programme, designed to identify inducible immediate-early-response genes, yielded one with considerable sequence homology with the known (COX-1) gene (Kujubu, Fletcher et al. 1991).

It is now known that COX exists in at least two isoforms, while a third is postulated lately. Garavito and his colleagues have determined the three dimensional structure of COX-1 (Figure 1.5) (Picot, Loll et al. 1994). This bifunctional enzyme comprises three independent folding units: an epidermal growth factor-like domain, a membrane-binding motif and an enzymatic domain (Marnett, Rowlinson et al. 1999). The sites for peroxidase and cyclooxygenase activity are adjacent but spatially distinct (Bazan, Botting et al. 1996). The confirmation of the membrane-binding motif strongly suggests that the enzyme integrates into only a single leaflet of the lipid bilayer and is thus a monotropic membrane protein (Bazan, Botting et al. 1996). Three of the helices of the structure form the entrance to the COX channel and their insertion into the

membrane could allow arachidonic acid to gain access to the active site from the interior of the bilayer (Bazan, Botting et al. 1996).



Subunit structure of COX-1. Each COX subunit comprises three domains, an epidermal growth factor domain (*yellow*), a membrane-binding domain (*lavender*), and a catalytic domain (*blue*). The catalytic domain contains the cyclooxygenase active site and the peroxidase active site separated by the heme prosthetic group (*red*). In the present structure, the cyclooxygenase active site is occupied by a molecule of iodosuprofen (*lime*). Arg-120, Tyr-355, and Glu-524 comprise a H-bonding network that introduces a constriction at the base of the cyclooxygenase active site. They are depicted in *gold*. The volume beneath this constriction is termed the lobby and is bordered on three sides by the membrane-binding domain. The catalytically important Tyr-385 residue is depicted in *white*. The peroxidase active site is at the top of the protein in this drawing and is visible as the wide opening to the heme prosthetic group.

Figure 1.5 Subunit structure of COX-1 (Marnett, Rowlinson et al. 1999)

The primary structures of PGHS-1 and –2 from numerous species are known. Both isoforms contain signal peptides of varying lengths. Mature, processed PGHS-1 contains 576 amino acids; the mature form of PGHS-2 contains 587 amino acids (Smith, DeWitt et al. 2000). There is a 60-65% sequence identity between PGHS-1 and -2 from the same species and 85-90% identity among individual isoforms from different species (Smith, DeWitt et al. 2000). The major sequence differences between PGHS isoforms occur in the membrane binding domains (Otto and Smith 1996; Spencer, Thuresson et al. 1999). A unique difference between PGHS-1 and –2 is 18

amino acids inserted 6 residues in from the C terminus of PGHS-2 that are not present in the PGHS-1 (Smith, DeWitt et al. 2000). The function of this insert is not yet established but may mark PGHS-2 for rapid proteolysis or provide a signal for subcellular trafficking; elimination of this cassette by deletion mutagenesis has no apparent effect on PGHS-2 catalysis (Smith, DeWitt et al. 2000).

PGHSs are homodimers both functionally and structurally (Xiao, Chen et al. 1998), but the reason that dimerisation is necessary for catalysis is unknown (Smith, DeWitt et al. 2000). Each monomer as mentioned consists of three structural domains: an epidermal growth factor (EGF) domain of 50 amino acids at the N terminus, a neighbouring membrane binding domain (MBD) of about 50 amino acids, and a large C-terminal globular catalytic domain with about 460 amino acids (Picot, Loll et al. 1994; Kurumbail, Stevens et al. 1996; Luong, Miller et al. 1996). The EGF domain forms a portion of the dimmer interface and is essential for folding (Smith, DeWitt et al. 2000). The membrane binding domains (MBDs) of PGHSs contain four short, consecutive, amphipathic a helix, the last of which, helix D, merges into the catalytic domain (Smith, DeWitt et al. 2000). Hydrophobic and aromatic residues protrude from these helices and away from the hydrophilic surface of the catalytic domain to create a hydrophobic patch that interacts with the one face of the underlying lipid bilayer (Picot, Loll et al. 1994). These helices also surround an opening through which fatty acid substrates and NSAIDs are believed to enter the cyclooxygenase active site (Smith, DeWitt et al. 2000). The globular catalytic domain closely resembles that of myeloperoxidase but with a hydrophobic channel protruding into the core of this domain (Picot, Loll et al. 1994). The upper half of the tunnel is the cyclooxygenase active site and can bind fatty acid substrates and NSAIDs. PGHS-1

and –2 contain C-terminal KDEL-like i.e. (Lys-Asp-Glu-Leu)-like sequences that target PGHSs to the endoplasmic reticulum and the associated envelope (Song and Smith 1996). Both enzymes are present on the lumenal surfaces of the ER and of the inner and outer membranes of the nuclear envelope (Otto and Smith 1994; Morita, Schindler et al. 1995; Spencer, Woods et al. 1998). PGHS-2 appears to be relatively more concentrated within the nuclear envelope (Morita, Schindler et al. 1995), raising the possibility that products formed via PGHS-2 may have greater access to the nucleoplasm to affect nuclear events, perhaps via nuclear receptors (Lim, Gupta et al. 1999). PGHS-1 is N-glycosylated at 3 sites, while PGHS-2 is variably glycosylated at 2 – 4 sites (Otto, DeWitt et al. 1993). N-glycosylation is required for enzyme folding of PGHS-1 (Otto, DeWitt et al. 1993) creating difficulties in producing large quantities of this isoform (Smith, DeWitt et al. 2000). On the contrary, the expression of PGHS-2 in bacilovirus systems was successful (Barnett, Chow et al. 1994). The PGHS structures contain several water channels, including a branched channel that extends from the cyclooxygenase site near Gly 533 to dimer interface (Smith, DeWitt et al. 2000). It is not clear if the water channels are simply structural or play a direct role in catalysis (e.g. as conduits for proton flow) (Smith, DeWitt et al. 2000).

PGHS-1 and PGHS-2 have very similar active site structures, catalytic mechanisms, products, and kinetics (Smith, DeWitt et al. 2000). There are, however two structural differences between the isoenzymes that have important pharmacological and biological consequences (Smith, DeWitt et al. 2000). First, the cyclooxygenase active site of PGHS-2 is larger and more accomodating than that of PGHS-1 (Smith, DeWitt et al. 2000). This size difference has been exploited in developing COX-2-specific NSAIDs (Smith, DeWitt et al. 2000). Second, although the

gross kinetic properties (e.g. Km, Vmax) of PGHS-1 and –2 are nearly identical, PGHS-1, but not PGHS-2, exhibits negative allosterism at low arachidonate concentrations; thus, permitting PGHS-2 to compete more effectively for newly released arachidonate when the isoenzymes are expresed in the same cell (Smith, DeWitt et al. 2000).

Table 1.1 illustrates the differences and similarities between the two isoforms (Cryer and Dubois 1998; Crofford, Lipsky et al. 2000; Stichtenoth and Frolich 2003). COX-1 is a constitutive enzyme and is always present in high concentrations within tissues including platelets (Funk, Funk et al. 1991), vascular endothelial cells (Goppelt-Struebe 1995), gastric epithelial cells (catalyses the production of PGs that protect the gastric mucosa) (Fu, Masferrer et al. 1990; Smith, Meade et al. 1994), and the renal collective tubules (Smith, Meade et al. 1994; Komhoff, Grone et al. 1997; Khan, Venturini et al. 1998). COX-2 is predominately an inducible enzyme with its expression induced within inflammatory and some others cells by inflammatory mediators such as bacterial lipopolysaccharides (LPS), and cytokines, interleukin IL-1b. It catalyses production of PGs that mediate inflammation and it is almost undetectable in the absence of inflammation (de Leval, Delarge et al. 2001). Based on this traditional view, the products of COX-1 metabolism are involved in the normal regulation of physiological processes that include: stimulation of the process of haemostasis through TXA_2 synthesis (which increases platelet adhesion and aggregation), inhibition of gastric acid secretion, stimulation of protective gastric mucus production, and regulation of blood flow in various vascular beds through the synthesis of prostanoids such as PGI_2, PGE_2, i.e. this was believed to be the dominant

Table 1.1 Comparison of COX-1 and COX-2: Molecular biology and the biological roles of the cyclooxygenase isoenzymes. (Cryer and Dubois 1998; Crofford, Lipsky et al. 2000; de Leval, Delarge et al. 2001; Stichtenoth and Frolich 2003)

Table 1.1: Comparison of COX-1 and COX-2

	COX-1	COX-2
Chromosome	9	1
Homology mRNA	~60%	~60%
mRNA size	2.7kb	4.5kb
Protein size	~65kDA	~70kDA
Amino acids	576	587
Intracellular Location	Endoplasmic Reticulum Nuclear Envelope	Endoplasmic Reticulum (some) Nuclear Envelope (mostly)
Regulation	**Constitutive (mostly)**	**Inducible (mostly)**
Range of expression	Platelets Endothelial Cells Stomach Kidney Smooth Muscle Most tissues	Most tissues esp. inflammatory cells Requires stImulation by: growth factors, cytokines, phorbol esters, LPS (bacterial), mitogens, reactive O_2 metabolites macrophages, endotoxins, tumor promoters
Range of expression	**Inducible**	**Constitutive** Brain, Kidney, Reproductive system
Proposed role (main)	**"Housekeeping", Homeostasis** Gastrointestinal mucosal protection, Kidney function, Platelet aggregation, Blood flow regulation, Lung function, Bone metabolism, CNS function	**Inflammatory response** Extravasation, Pain, Fever, Proliferation
Other proposed roles	**(Inflammation)**	**Homeostasis** Kidney function and development, Blood flow regulation, CNS function, Bone metabolism, Lung function? **Tissue repair** Ulcer healing, Adaptation to vascular injury **Reproduction** Fertilisation, maintenance of pregnancy

mechanism of homeostatic regulation of glomerular filtration in the kidney (Brater, Harris et al. 2001; Wong, Wang et al. 2005). Conversely, the expression of COX-2 results in prostanoid synthesis at sites of inflammation and was seen as producing the unwanted effects arising from the inflammatory process such as pain and swelling. Thus, anti-inflammatory efficacy is believed to result from inhibition of COX-2.

COX-1 and COX-2 monomers each contain a long (25-A°) narrow hydrophobic channel with a hairpin bend at the end (Figure 1.6) (Hawkey 1999; Smith, DeWitt et al. 2000). The hydrophobic channel originates at the MBD and extends into the core of the globular domain, serving as an entrance allowing arachidonate and O_2 to enter directly from the apolar compartment of the lipid layer (Picot, Loll et al. 1994; Kurumbail, Stevens et al. 1996; Luong, Miller et al. 1996; Smith, DeWitt et al. 2000). As both isoforms are membrane-associated, the AA released from damaged membranes adjacent to the opening of the enzyme hydrophobic channel, is sucked in, twisted around the hairpin bend, two oxygens are inserted, and a free radical extracted, resulting in the five-carbon ring that characterises the PGs (Figure 1.7) (Hawkey 1999).

NSAIDs block COX-1 about halfway down the channel (Lanzo, Beechem et al. 1998). X-ray crystallography suggested that this inhibition occurs by hydrogen bonding to the polar arginine at position 120 to near tyrosine at position 385 (Hawkey 1999; Smith, DeWitt et al. 2000). Arginine (Arg) 120 is also present in COX-2. Twenty-four residues line the hydrophobic cyclooxygenase active site with only one difference between the isoenzymes; Isoleucine (Ile) at position 523 in PGHS-1 and valine (Val-smaller by a single methyl group) in PGHS-2 (Smith, DeWitt et al. 2000).

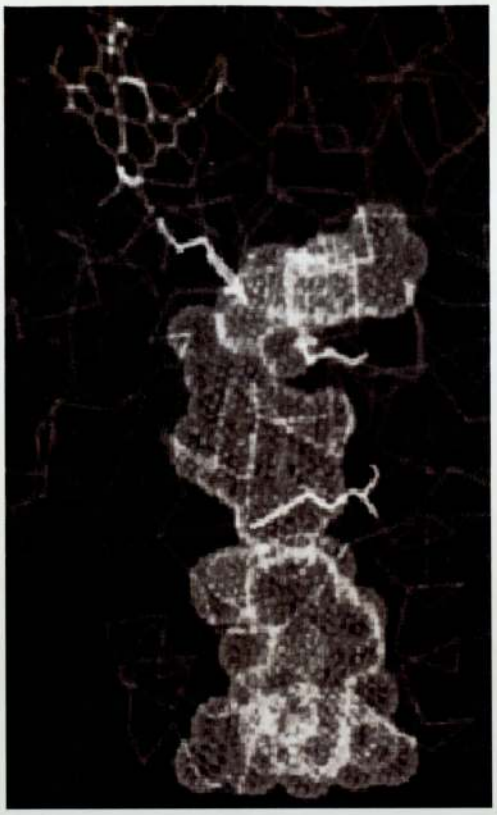


Figure 1.6 X-ray crystallography of COX-1 (Hawkey 1999)

Only 3 of the amino acids lining the hydrophobic cyclooxygenase active site channel are polar: Arg 120, Serine (Ser) 353, and Ser 530. Ser 530 is the site of irreverible acetylation by aspirin, and Arg 120 binds to the carboxylate groups of fatty acids and many NSAIDs (Smith, DeWitt et al. 2000). The smaller valine molecule in COX-2 leaves a gap in the wall of the channel, giving access to a side-pocket, which is thought to be the site of binding of many selective drugs (Figure1.7) (Hawkey 1999). The bulkier isoleucine at 523 in COX-1 is large enough to block access to the side-pocket (Hawkey 1999). Targeted single amino acid substitution of valine for isoleucine is sufficient to turn COX-1 into an enzyme that can be inhibited by COX-2 selective agents (Gierse, McDonald et al. 1996; Luong, Miller et al. 1996).

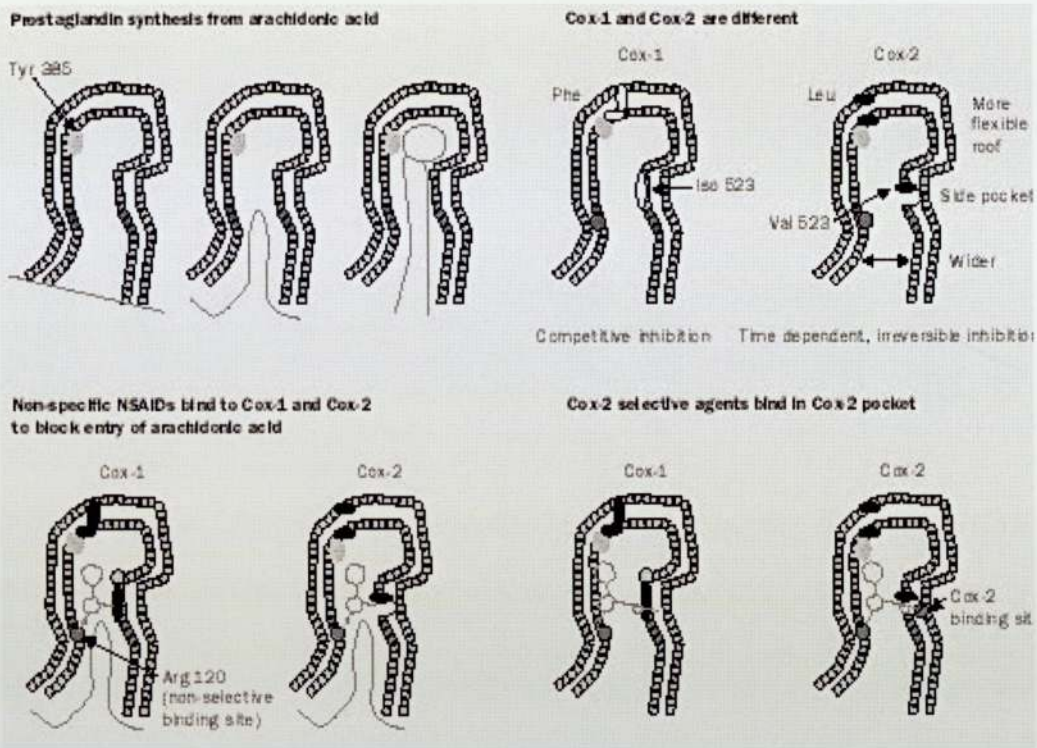


Figure 1.7 Prostaglandin synthesis and inhibition in COX-1 and COX-2 (Hawkey 1999)

1.4 COX-2 hypothesis

The discovery of two COX isoforms, a constitutive COX-1, serving homeostatic prostanoid synthesis, and an inducible COX-2, responsible for proinflammatory prostanoid production (Masferrer, Zweifel et al. 1990; Kujubu, Fletcher et al. 1991; Xie, Chipman et al. 1991; Ford-Hutchinson 1997), ushered in a new generation of NSAIDs with the promise of fewer adverse-effects: preferential and specific COX-2 inhibitors. The COX-2 hypothesis suggests that at comparable COX-2 inhibiting doses highly selective COX-2 inhibitors would be as effective as traditional NSAIDs but cause fewer gastrointestinal (GI) and renal side-effects as determined by clinical endpoints reflecting COX-1 dependent GI and renal toxicity (FitzGerald and Patrono 2001). Progressive modification and extension of flurbiprofen's methyl group

resulted in molecules that were increasingly selective in their ability to bind in the COX-2 side-pocket, but too bulky to fit within the COX-1 channel (Hawkey 1999). Many COX-2 inhibitors have structures that exploit binding within the COX-2 side-pocket (often via sulphonyl, sulphone, or sulphonamide groups) to achieve selectivity (Ford-Hutchinson 1997). Figure 1.8 illustrates some of the specific cyclooxygenase inhibitors ever marketed.

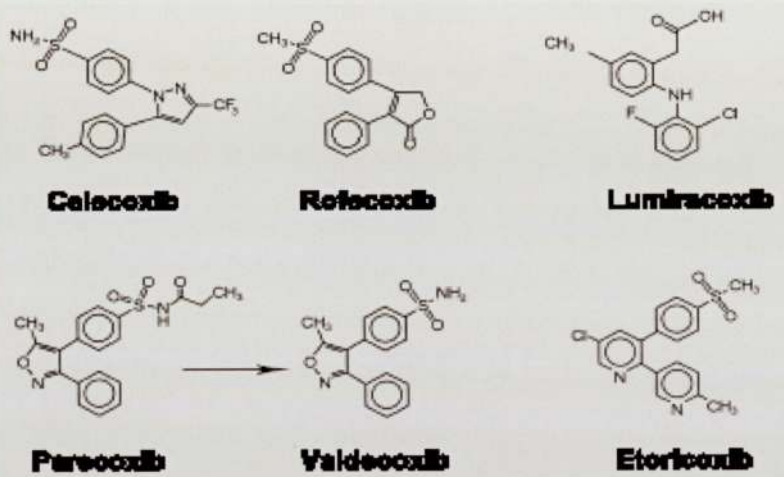


Figure 1.8 Specific COX-2 inhibitors (celecoxib, rofecoxib, lumiracoxib, parecoxib (prodrug for valdecoxib), valdecoxib and etoricoxib) (Hersh, Lally et al. 2005)

It has been proposed that the term COX-2 specific inhibitor should be used to describe agents which can inhibit COX-2, but have no effect on COX-1 over the whole range of doses used and concentrations achieved in clinical usage. (Hawkey 1999)

1.5 Selectivity: Assays and Limitations

The concept that selective COX inhibition may be a therapeutically desirable goal has led to several studies to assess the comparative COX isoform selectivity of the currently available NSAIDs and newly developed COX-2 selective and specific inhibitors. COX-2 selectivity is expressed as the ratio of the COX-2 IC_{50} (where IC_{50} represents the concentration of the drug required to achieve 50% enzyme inhibition in vitro) to the COX-1 IC_{50}, so that the more COX-2 selective an agent is the smaller is the quoted ratio [Selectivity Ratio = COX-2 IC_{50} / COX-1 IC_{50}]. Some researchers believe that the inhibitory concentration IC_{80} (80% enzyme inhibition) is more indicative of selectivity (Warner, Giuliano et al. 1999). Figure 1.9 illustrates graphically the results of a whole blood assay for a wide range of NSAIDs and selective and specific inhibitors (Warner, Giuliano et al. 1999).

No method yet commands universal support and differences in quoted selectivity can be more than 10 fold (Hawkey 1999). Four different types of in vitro assays have been used to evaluate COX-1 and COX-2 activity based on either: (i) the use of purified or recombinant COX-1 and COX-2 enzymes (de Leval, Delarge et al. 2001), (ii) the use of microsomal enzymes (Chan, Boyce et al. 1999), (iii) the use of whole cell lines that inherently express or have been tranfected to express only COX-1 or COX-2 (Mitchell, Akarasereenont et al. 1993) and (iv) the use of components derived from human (or animal) whole blood that express exclusively (~98%) either COX-1 or COX-2 (Patrignani, Panara et al. 1997; Panara, Renda et al. 1999).

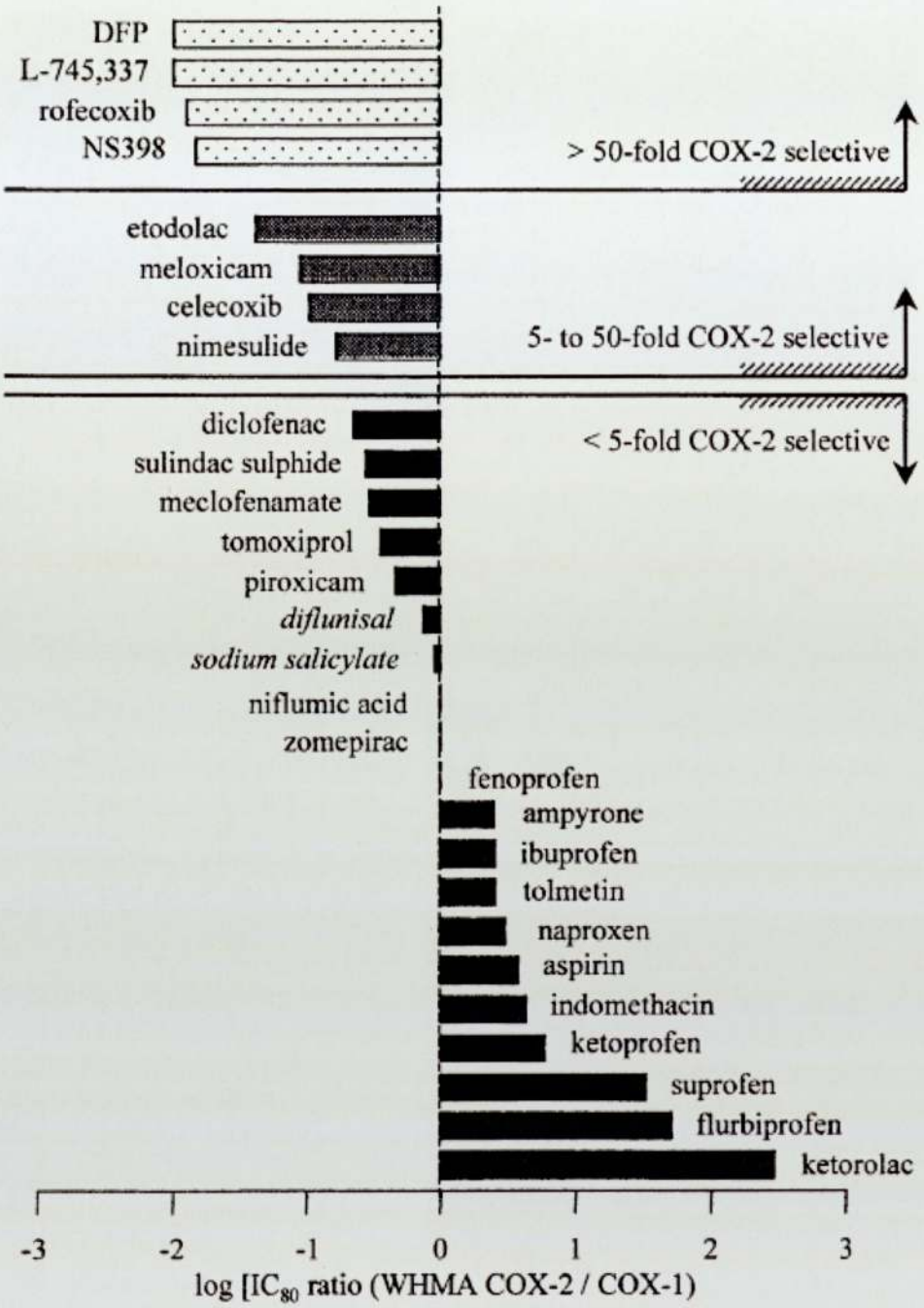


Figure 1.9 Determinable log [IC_{80} ratio (WBA-COX-2/COX-1) for all agents assayed The '0' line indicates equipotency, i.e. an IC_{80} of 1 (Warner, Giuliano et al. 1999)

Purified enzyme utilise purified enzyme and measure either the diminution of a given substrate or the amount of the product formed. However, minor differences in

the sequences between species can modify the measured potency at a cellular level. Furthermore, purified enzyme assays cannot simulate *in vivo* conditions due to differences in tissue penetration, pharmacokinetics or other factors. Normally preincubation needs to take place, which is not directly applicable *in vivo*. Purified enzyme assays take place in protein-free media, thus not taking into account the protein binding capacity of the assayed substrates *in vivo* and thus the absolute free concentration values of the assayed drug (Frolich 1997). Studies that use protein-free solutions give very low IC_{50} values, which are far removed from the *in vivo* situation (Grossman, Wiseman et al. 1995). Finally, the concentrations of substrates used are normally different (100-fold difference) in order to achieve an outcome, which can limit the generalisation of the assay results. Other variables in *in vitro* studies using recombinant enzymes include lack of glycosylation, with resulting low specific activity (Otto, DeWitt et al. 1993), and uncertainty about the relevant drug concentration in intact cells.

For the whole cell COX-2 assays, either LPS-stimulated macrophages or mammalian cells recombinantly expressing COX-2 have been described as sources for COX-2. Testing compounds for potential inhibition of COX enzyme in whole cells assays does not measure enzyme activities directly as compared to a classical enzyme assays that uses purified enzyme and measures either the diminution of a given substrate or the amount of the product formed (Berg, Christoph et al. 1997). They do not measure the direct product of COX (prostaglandin G and H) as these products are short-lived and quickly converted into eicosanoids by enzymes (Berg, Christoph et al. 1997). Furthermore, in whole cells assays, potential enzyme inhibitors do not have direct access to their target (Berg, Christoph et al. 1997). The compounds have to cross

the cell membranes and need to survive possible degradation pathways within the cell on its way to the site of action (Berg, Christoph et al. 1997). Thus, IC_{50} values from whole cell assays are not direct measure for enzyme inhibition (Berg, Christoph et al. 1997). They also reflect cellular pharmacological and cellular pharmacokinetic parameters depending on the properties of the respective compound (Berg, Christoph et al. 1997). Pharmacologically, however, the whole cell COX assays (Mitchell, Akarasereenont et al. 1993; Klein, Nusing et al. 1994; Grossman, Wiseman et al. 1995; Engelhardt, Bogel et al. 1996) have the advantage of testing compounds in vitro that resemble the in vivo application more closely than classical enzyme assays do.

Whole blood assays (WBAs) are far more relevant pharmacologically (Ehrich, Dallob et al. 1999; Cronstein 2002). These assays are performed in a physiologic medium (i.e. whole blood) with endogenous enzymes and locally derived substrates (Ahuja, Singh et al. 2003). In the whole-blood assay, synthesis of thromboxane (TXB_2) from platelets during clotting is used as an index of COX-1 activity, while synthesis of PGE_2 (principally from monocytes) in whole blood exposed to LPS (lipopolysaccharide) is an index of COX-2 activity (leukocytes COX-2 expression to bacterial endotoxins) (Patrignani, Panara et al. 1994; Patrignani, Panara et al. 1997). The secreted amounts of eicosanoids are used as a measure of for COX activities. Thus, IC_{50} values of compounds can be determined for both isoenzymes. *In vitro* WBAs are performed by the addition of drug in various concentrations to blood previously obtained. The *in vitro* WBAs take advantage of using whole cells that are pathophysiological targets for NSAIDs, of considering the intracellular transport of drugs, of providing a physiological plasma protein level and of checking for inhibition of both COX isoenzymes on a single sample. Additionally, it can be used *in vivo* to

determine the degree of COX inhibition after oral intake of therapeutic doses of the drug, which is important for COX-2 specific agents (Patrignani, Panara et al. 1997; Lipsky, Abramson et al. 1998).

As an in vitro assay is easy to perform and spares time in comparison with clinical studies, authors have proposed to use the in vitro WBA data for estimating the expected levels of NSAIDs. However, such a predictive approach is based on (a) the use of COX inhibition curves obtained by the addition of a range of concentration of NSAIDs to donated blood from few healthy subjects and (b) on the extrapolation of plasma concentrations of drugs, assuming the NSAIDs do not enter red cells and that the haematocrit is 45% (Blain, Boileau et al. 2002). Blain et al (2002) (Blain, Boileau et al. 2002) illustrated that NSAIDs partitioned differently into whole cells, although the difference remained moderate between molecules since they were acidic in nature. The factor accounting for the transformation of the whole blood into plasma concentrations followed a different rank order from the drug pKa, suggesting that it was influenced by the lipophilicity of the molecules (Blain, Boileau et al. 2002). They suggested that it could be a major flaw to neglect the ability of non-acidic molecules (all coxibs apart from lumiracoxib) to enter red cells since, for example, celecoxib is thought to be evenly distributed between erythrocytes and plasma (Blain, Boileau et al. 2002). In addition, the use of haematocrit has no influence as long as the studies are made in healthy subjects of the same sex, but it adds limitation to the predicting value of the whole blood system in patients with chronic inflammation.

Another factor to be considered is that selectivity seen in blood may not reflect selectivity at the gastric mucosa. Thus, some investigators tried to quantify the

selectivity in the gastric mucosa (Cryer and Feldman 1998). As a close correlation has been reported between the inhibitory potency of NSAIDs on thromboxane synthesis by platelets and COX-1 activity in gastric mucosa, such an ex vivo assay has a clear clinical relevance as long as the tissue concentrations of the drug are considered.

The results of in vitro assays are useful for drug screening but are difficult to interpret and are sometimes contradictory (Cronstein 2002). This may be attributed to diverse factors like the nature of the enzyme and substrate employed, incubated period (time-dependent inhibition has been demonstrated for both COX isoenzymes) and other experimental variables. Performance of *ex-vivo* WBAs on blood samples collected at a pharmacological relevant timing after systemic drug administration can give a better index for isoenzyme selectivity as it tests both the parent drug and any potential metabolites generated *in vivo* at therapeutic blood concentrations (Ahuja, Singh et al. 2003). However, the ex vivo WBA is somewhat variable as it depends clearly on the phamacokinetics of NSAIDs. As a consequence, an appropriate number of subjects (to avoid intersubject variability) might be required for a meaningful determination of NSAID selectivity and COX inhibition should be reported at pharmacologically relevant times (Ahuja, Singh et al. 2003). Thus, human WBA seems to be an interesting and most safely predictive method available currently for the evaluation of the inhibitory selectivity of COX-2 selective and specific inhibitors, although optimal investigation should be a human WBA performed *ex vivo* after intake of drugs during several days (de Leval, Delarge et al. 2001). However, such investigations cannot be performed with drugs in pre-clinical trials (de Leval, Delarge et al. 2001).

1.6 Cyclooxygenase-3

Recently, it has been suggested that there is another COX protein formed as a splice variant of COX-1 that is found in highest concentrations in the cerebral cortex and heart of the dog, which they reported to be the elusive COX-3 (Chandrasekharan, Dai et al. 2002).

While the dual COX model resolved many of the issues concerning differences between non-selective NSAIDs and highly selective COX-2 inhibitors, it still could not fully explain the pharmacologic actions of acetaminophen (Hersh, Lally et al. 2005). Many of acetaminophen's actions resemble COX-2 inhibitors (analgesic effects, antipyretic effects and a relative lack of GI toxicity (Botting 2000; Graham and Scott 2003; Hersh, Lally et al. 2005). However it lacks, or at very best possesses weak anti-inflammatory action; an important characteristic of both NSAIDs and the COX-2 selective drugs (Hersh, Lally et al. 2005). In addition, more than 50 years of clinical experience with this agent has revealed no appreciable anti-aggregatory or pro-aggregatory effects on platelets (Hersh, Lally et al. 2005).

As far back as 1972, Flower and Vane reported that acetaminophen was far more active in inhibiting COX activity in the dog brain homogenates than in homogenates from the spleen (Flower and Vane 1972). This result led them to be the first to postulate the existence of more than one COX isoform. Others have since proposed a predominately central mechanism of action for acetaminophen involving either central COX-2 inhibition, or the inhibition of a yet to be isolated COX variant termed COX-3 or the activation of desccending serotonergic pathways in the brain and

spinal cord (Botting 2000; Willoughby, Moore et al. 2000; Graham and Scott 2003; Hersh, Lally et al. 2005).

The messenger RNA that produced the COX-3 protein was derived from the same gene that coded for COX-1, except in COX-3 RNA, an intron made up of 90 nucleotides at or near the 5 prime end of the molecule was retained (Chandrasekharan, Dai et al. 2002). The retention of this intron (which is normally cleaved prior to the final synthesis of the RNA) introduces the insertion of an additional 30 amino acids into the dog COX-3 molecule (Chandrasekharan, Dai et al. 2002; Hersh, Lally et al. 2005). It was postulated that these extra amino acids would alter the folding and subsequent enzymatic properties of this newly discovered COX type (Hersh, Lally et al. 2005). In experiments performed by this group, it was demonstrated that in transfected insect cells, canine COX-3 protein was expressed and selectively, but weakly inhibited by acetaminophen, whereas transfected murine COX-1 or COX-2 was not acetaminophen sensitive (Chandrasekharan, Dai et al. 2002). In addition, other analgesic/antipyretic drugs that lacked significant anti-inflammatory activity such as phenacetin (which is metabolised to acetaminophen) and dipyrone, and classical NSAIDs with potent anti-inflammatory activity such as ibuprofen and diclofenac also displayed more potent inhibition of COX-3 than the other COX isoforms (Chandrasekharan, Dai et al. 2002). It should be noted that some scientists prefer to call this new protein as COX-1b or COX-1 variant (COX-1v) rather than COX-3 as the mRNA is encoded by the COX-1 gene, and other than the retained intron, the mRNA is indistinguishable from COX-1 (Hersh, Lally et al. 2005).

The predicted COX-3 messenger RNA protein that would be synthesised in humans would possess a completely different protein sequence and would only be about 79 amino acids long due to a stop codon (UGA) on the messenger RNA (Schwab, Beiter et al. 2003). Interestingly, two minor subtypes of this variant were also isolated, that contained only 93 nucleotides in the retained intron (Hersh, Lally et al. 2005). However, the protein they produced while able to catalyse the production of prostaglandins from AA, did not exhibit differential sensitivity to acetaminophen (Qin, Zhang et al. 2005). Furthermore, it appears, that at least in the human and the rodent, while putative COX-3 mRNA is present in a number of tissues, it does not express a functional, acetaminophen-sensitive COX protein (Kis, Snipes et al. 2005). In addition, the IC_{50} concentration of acetaminophen needed to block COX-3 in canine in vitro preparations is very high, and is unlikely to be attained in the human hypothalamus where temperature regulation occurs, following therapeutic doses of acetaminophen (Schwab, Schluesener et al. 2003; Berenbaum 2004).

1.7 COX-2 Inhibitors Pharmacokinetics

The first selective COX-2 inhibitors approved by FDA and EMEA for the treatment of osteoarthritis (OA), rheumatoid arthritis (RA), and for relief of acute pain associated with dental surgery and dysmenorrhoea, celecoxib and rofecoxib, are diaryleterocyclic derivatives containing a phenylsulphonamide and a phenylsulphone moiety (Figure 1.8), respectively, that interact with COX-2 side-pocket through slow, tight-binding kinetics (Kurumbail, Stevens et al. 1996; Patrono, Patrignani et al. 2001; Patrignani, Tacconelli et al. 2005).

The rate of absorption of rofecoxib is moderate when given orally, with peak plasma concentrations (Cmax) achieved at 2-3 hours (Tmax) (Appendix I) (Davies, Teng et al. 2003). Although the absolute extent of absorption is not known, the main oral bioavailability of rofecoxib tablets at therapeutically recommended doses of 12.5mg to 50mg is approximately 93%, based on radioactivity and metabolite urine recovery studies in normal human subjects (Halpin, Porras et al. 2002). Furthermore, pharmacokinetic studies indicate that the tablet and suspension formulations are bioequivalent (Davies, Teng et al. 2003). Both the Cmax and AUC (area under the curve) of rofecoxib are linearly related to dose within the clinical dose range of 12.5-50mg when compared in single-dose pharmacokinetic studies (Davies, Teng et al. 2003). At doses greater than 50mg there is less than proportional increase in Cmax and AUC, which is thought to be due to the limited solubility of the drug in the aqueous environment of the gastrointestinal tract (Halpin, Geer et al. 2000; Halpin, Porras et al. 2002; Davies, Teng et al. 2003). With multiple doses, steady-state plasma concentrations of rofecoxib are reached after 4 days (Depre, Ehrich et al. 2000). It has been suggested that absoprtion of poorly soluble rofecoxib varies with intestinal motility, yielding secondary absorption peaks and resulting in high variability for Tmax. (Halpin, Porras et al. 2002). Rofecoxib is extensively protein bound in plasma, primarily to albumin (~87%) and has an apparent volume of distribution of 90L (1.3L/kg) (Davies, Teng et al. 2003). The larger than expected apparent volume of distribution when compared with other traditional NSAIDs may relate to the lipophilic nature of rofecoxib (Davies, Teng et al. 2003). Interestingly, cytochrome P450 plays only a minor role in the metabolism of rofecoxib, which is mediated primarily through reduction by cytosolic enzymes, with less than 1% excreted unchanged in urine (Davies, Teng et al. 2003). Biphasic plasma rofecoxib concentration peaks are seen

after oral administration in humans (Nicoll-Griffith, Yergey et al. 2000; Halpin, Porras et al. 2002) suggestive of enterohepatic recirculation (Davies, Teng et al. 2003). Finally, following oral administration to human subjects the majority of the dose (~75%) undergoes metabolism to products that are eliminated by the kidneys into the urine (Davies, Teng et al. 2003). A small fraction of the administered radioactivity (~14%) is recovered in faeces, with a very low excretion in bile (~1.8% of the dose) (Halpin, Porras et al. 2002).

Celecoxib is administered orally in convential release capsules, with doses of 100mg and 200mg commercially available. After a single oral 200mg dose to healthy young volunteers the mean Cmax of celecoxib were reached after between 2 and 4 hours (Appendix I) (Davies, McLachlan et al. 2000). Most traditional NSAIDs are highly (>98%) protein bound to albumin and have an apparent volume of distribution (V/F) ranging from 0.1 to 0.3 L/kg (Davies, McLachlan et al. 2000). This volume of distribution is much lower than the volume of total body water (0.6 L/kg) and is equivalent to plasma or blood volume (Davies and Morris 1993). Celecoxib is also extensively protein bound, primarily to albumin (Davies, McLachlan et al. 2000). The fraction of unbound drug remains essentially constant (mean 2.6% unbound) at total plasma celecoxib concentrations up to 4000µg/L (Davies, McLachlan et al. 2000). Based on measurement of total 14C radioactivity, celecoxib is evenly distributed between erythrocytes and plasma (red blood cell/plasma = 0.89) (Paulson, Kaprak et al. 1999). Celecoxib has an V/F of 455 +/- 166L in humans (5.7 to 7.1 L/kg) (Davies, McLachlan et al. 2000). This larger than expected V/F when compared with other NSAIDs may relate to the lipophilic nature of celecoxib or be reflectivce of a low bioavailibility (Davies, McLachlan et al. 2000). The half life if celecoxib is reported to

be between 11.2 and 15.6 hours (Davies, McLachlan et al. 2000). Celecoxib undergoes extensive hepatic metabolism in humans. Less than 2% is excreted unchanged in urine and only 2. 6% is excreted in faeces (Davies, McLachlan et al. 2000). Three metabolites of celecoxib have been found in plasma: SC-60613, SC-62087, and the glucuronide conjugate of SC-62087 (Davies, McLachlan et al. 2000). In vitro studies using human liver microsomes and heterogeneously expressed CYP protein indicate that CYP 2C9 is the major isoform responsible for celecoxib's metabolism (Davies, McLachlan et al. 2000).

Novel COX-2 inhibitors with improved biochemical selectivity over that of first generation coxibs (i.e. rofecoxib and celecoxib) have been recently developed as valdecoxib, parecoxib (the prodrug of valdecoxib), etoricoxib and lumiracoxib.

The bioavailability of orally administered valdecoxib is 83% of that of intravenously administered valdecoxib (Chavez and DeKorte 2003). In 8 healthy male subjects aged between 20-42 years who received valdecoxib 10mg once a day, the Cmax was achieved in a mean (SD) of 2. 25 (0.71) hours, and the mean terminal half-life was 8.11 (1.32) hours (Chavez and DeKorte 2003). Valdecoxib is 98% bound to plasma protein and its volume of distribution is approximately 86L (Chavez and DeKorte 2003) Valdecoxib is primarily eliminated via hepatic metabolism, and <5% is excreted unchanged in the urine and faeces. Eighty per cent of its metabolism is via the cytochrome P450 isozymes CYP3A4 and CYP2C9 (Chavez and DeKorte 2003). The remaining 20% of hepatic metabolism is through glucuronidation (Chavez and DeKorte 2003). An active hydroxylated metabolite has been identified that probably does not contribute to efficacy, as plasma concentrations are ~10% those of valdecoxib

(Chavez and DeKorte 2003). This metabolite also undergoes hepatic metabolism. Eight other metabolites have no clinically significant therapeutic effects or known toxic effects (Chavez and DeKorte 2003).

In healthy male volunteers, bioavailability of etoricoxib is 100% for the tablet (Martina, Vesta et al. 2005). Maximal concentration was 36% lower and occurred 2hours later when administered after a high-fat meal (Martina, Vesta et al. 2005). Etoricoxib has marked distribution into tissues (volume of distribution 119L) and is 92% protein bound (Martina, Vesta et al. 2005). Etoricoxib distributes rapidly, reaching peak concentration within 1-2 hours, and has an elimination half-life of approximately 22 hours (Martina, Vesta et al. 2005). Etoricoxib is metabolised to a 6'-hydorxymethyl derivative through cytochrome P450-dependent oxidation (Kassahun, McIntosh et al. 2001). CYP3A4 is the predominant isoenzyme responsible for metabolism (40-90%), with various other isoenzymes equally metabolising the remainder of etoricoxib (Martina, Vesta et al. 2005). AUC concentrations did not change remarkably with CYP P450 enzyme contributing to metabolism (Martina, Vesta et al. 2005). Etoricoxib was found to be a weak inhibitor of CYP P450 isoenzymes including 2D6, 3A4, and 2C9 (Martina, Vesta et al. 2005).

In contrast with other COX-2 selective inhibitors, lumiracoxib posesses a carboxylic acid, making it weakly acidic (Appendix I) (Scott, Rordorf et al. 2004). Studies of the steady-state pharmacokinetics of lumiracoxib have been performed in healthy subjects and patients with OA and RA (Scott, Rordorf et al. 2004). The AUC of the plasma concentration-time relationship increased in a dose-proportional manner over the dose range 50-200mg twice daily and 200-800mg once daily, and no

accumulation of drug in plasma was noted after 12, 28, or 91 days of continuous treatment (Scott, Rordorf et al. 2004). Lumiracoxib is eliminated faster with a half-life of about 5 hours (Brune and Hinz 2004).

Most coxibs are relatively lipophilic compounds. All show some structural similarities to former drugs, for example celecoxib is a sulphonamide sharing similarities with e.g. propyphenazone (Brune and Hinz 2004). Rofecoxib and etoricoxib mimic other methylsulphones, e.g. sulindac (Brune and Hinz 2004). On the other hand, lumiracoxib compared to the non-acidic other coxibs is slightly acidic and its resemblance to diclofenac indicates a phenylacetic acid derivative (Brune and Hinz 2004). The volume of distribution of the non-acidic coxibs is equal or above body weight, whereas that of lumiracoxib is (as with other acetic acid derivatives e.g. diclofenac and indomethacin) around 15% of body weight (Brune and Hinz 2004). Non-acidic coxibs distribute almost equally throughout the body, with the exception of celecoxib, which is likely to be sequestered in body fat due to its extremely high lipophilicity (Brune and Hinz 2004). On the other hand, lumiracoxib reaches high concentration in the blood stream, kidney, liver, and inflamed tissue, but comparatively lower concentration in other compartments (Brune and Hinz 2004).

Chapter 2

Licensing Indications &

Adverse Effects of Coxibs

2.1 Preface

Cyclooxygenase must be one of the most widely used therapeutic drug targets in history. Inhibitors of this enzyme have been used for more than 3,500 years, and tens of thousands tons of these compounds are consumed each year (Warner and Mitchell 2004). NSAIDs and selective COX-2 inhibitors are commonly used for their lasting analgesic and anti-inflammatory effect and this makes them particularly useful for the treatment of continuous or regular pain associated with inflammation, as in the case of arthritic disorders. Arthritis (from Greek *arthro-*, joint and *-itis*, inflammation) is a general term used to describe the inflammatory disease of one or more joints, characterised by pain, swelling, stiffness, restriction of motion and redness of the skin overlying the affected joint (NICE 2001). Osteoarthritis (OA) and rheumatoid arthritis (RA) are the most common forms of arthritis. Other arthritic disease include psoriatic arthritis, septic arthritis, juvenile arthritis, Still's disease, ankylosing spondylitis, gout and pseudogout. NSAIDS and selective COX-2 inhibitors, however, have been tested as chemopreventive agents and preventors of Alzheimer's disease. Nevertheless, they are not free of adverse-effects, especially when used long-termly.

2.2 Osteoarthritis (OA)

Osteoarthritis (OA) is currently defined by the American College of Rheumatology (ACR) as a "heterogeneous group of conditions that leads to joint symptoms and signs which are associated with defective integrity of articular cartilage, in addition to related changes in the underlying bone at the joint margins" (Sarzi-Puttini, Cimmino et al. 2005). OA is a major cause of morbidity and disability, particularly for the eldrely, while being the most common form of arthritis (Creamer and Hochberg 1997). It is a progresive disease with insidious onset and can affect single or multiple joints, with the most frequently affected joints being hands, knees, hip and spine.

2.2.1 Aetiology, Classification and Diagnosis of OA

Although the aetiology of OA remains elusive, it is no longer regarded as a simple consequence of ageing and trauma (Creamer and Hochberg 1997). Osteoarthritis diseases are a result of both mechanical and biological events that destabilise the normal coupling of degradation and synthesis of articular cartilage chondrocytes and extracellular matrix, and subchondral bone (Creamer and Hochberg 1997). The aetiology of osteoarthritis is multifactorial, with inflammatory, metabolic, genetic and mechanical causes. Risk factors such as advanced age, sex (female at higher risk), obesity, muscle weakness, trauma, depletion of sex hormones, and genetic profiles (race / ethnicity) have been identified (Srikanth, Fryer et al. 2005). Ultimately, OA diseases are manifested by morphologic, biochemical, molecular, and biomechanical changes of both cells and matrix which lead to a softening, fibrillation, ulceration, loss

of articular cartilage, sclerosis and eburnation of subchondral bone, osteophytes, and subchondral cysts (Creamer and Hochberg 1997). When clinically evident, osteoarthritic diseases are characterised by joint pain, tenderness, limitation of movement, crepitus, occasional effusion, and variable degrees of inflammation without systemic effects (Creamer and Hochberg 1997).

The American College of Rheumatology has classified OA as primary and seconadary (Table 2.1) (Altman, Asch et al. 1986) as well as provided criteria for OA of the hand, hip and knee based on clinical presentation of the disease (APPENDIX II) (Altman, Asch et al. 1986; Altman, Alarcon et al. 1991).

Table 2.1 Classification of Osteoarthritis (Altman, Asch et al. 1986)

Idiopathic / Primary	Localised (e.g. hands, feet, knees, hips, & other single sites) Generalised (three or more joint groups listed above)
Secondary	Post-traumatic Congenital or developmental diseases Localised (e.g. dysplasia) Generalised (e.g. chondrodysplasias, inherited metabolic diseases [ochronosis, haemochromatosis]) Calcium-deposition disease Other bone and joint disorders (e.g. avascular necrosis, RA, Paget's disease)
Other diseases	Endocrine diseases (e.g. acromegaly, hyperparathyroidim) Neuropathical (Charcot's) arthropathy Miscellaneous

Osteoarthritic disorders represent a group of heterogeneous conditions of multifactorial aetiology and thus are difficult to or not desirable to define with a single set of criteria (NICE 2001). Diagnosis is usually made by clinical examination and

confirmed radiographically. The cardinal radiographic features are joint space narrowing and the presence of osteophytes (NICE 2001). However, it has to be mentioned that the degree of radiographic changes poorly correlates with the clinical symptoms (NICE 2001). Additionally, the non-specific and highly subjective symptoms (such as poorly localised joint pain) that are apparent in the majority of patients and the insensitivity of diagnostic techniques in detecting changes occuring during natural courses of the diseases make clear diagnosis even more complicated (NICE 2001).

2.2.2 Prevalence of OA

OA affects roughly 43 million Americans, with associated costs of approximately $95 billions (Elders 2000). The cost of treatment in Western countries alone is 1-2% of gross national product (Reginster 2002). Its prevalence rises with age (reaching a plateau over the seventh decade) and is higher in women than in men (Creamer and Hochberg 1997; Srikanth, Fryer et al. 2005). Any estimates of overall prevalence of OA will be variable due to the differences in diagnostic criteria used in different studies (Spector and Hochberg 1994; Petersson 1996). Estimated prevalence of radiographically diagnosed OA in the U.K. is as high as 50 % in the over 55 year age group and most people over 65 years of age will have some radiological evidence of OA in at least one joint (NICE 2001). The difference between radiological evidence of OA and to the overall prevalence of symptomatic OA is also not clear, but one study estimated that around 12% of over 65 year olds are clinically affected (Watson, Brookes et al. 2000), whereas others put the prevalence of symptomatic OA between 1.6 and 3.4 million in the over 45 year age group in the UK (Lord, Victor et al. 1999).

Symptomatic knee OA occurs in about 6% of adults older than 30 years of age and 9.5 % of adults between 63-94 years of age (women 11.4% and men 6.8%) (Felson, Lawrence et al. 2000; Schnitzer, Weaver et al. 2005).

2.2.3 Clinical features of OA

The clinical features of OA are summarised in Table 2.2 (Creamer and Hochberg 1997). Pain is the most significant symptom. Usually, it is insidious in onset with mild-to-modetate intensity, and is worsened by the use of the affected joint, while on the other hand the pain improves with rest. Pain at rest or during the night are features of severe disease (Creamer and Hochberg 1997).

Table 2. 2 Symptoms & Signs in Osteoarthritic Patients (Creamer and Hochberg 1997)

Symptoms	Joint Pain
	Morning Stiffness
	Gel Phenomenon
	Bucking/ Instability
	Loss of function
Signs	Bony Enlargement
	Limitation of range of motion
	Crepitus on motion
	Tenderness on pressure
	Pain on motion
	Joint Effusion
	Malalignment, Joint Deformity, or both

2.2.4 Treatment Strategies for OA

Current treatment of OA is purely to control symptoms, because as yet there are no disease-modifying OA drugs (Creamer and Hochberg 1997). Management involves both non-pharmacological and pharmacological measures. The first include exercise, weight loss programmes, patient education, and occupational therapy. Pharmacological measures are only indicated when non-drug treatment fails to control the symptoms. Paracetamol is established today to be the first drug of choice for mild to moderate pain and is the preferred long term oral analgesic, with NSAIDs usually considered if the maximum dose of paracetamol fails to control pain and inflammation involved (Shamoon and Hochberg 2001; Jordan, Arden et al. 2003; Zhang, Doherty et al. 2005). The ACR guidelines recommend acetaminophen as first-line therapy for OA with NSAIDs and COX-2 selective and specific inhibitors available for patients who have not achieved a satisfactory response. Intra-articular corticosteroids and surgery can also be considered.

2.2.5 Outcome measures used in the assessment of OA

The most commonly used outcome measures used in the assessment of OA include patient and doctor global assessments, frequency and severity of joint pain (at rest, with movement, pain intensity, night pain, weight bearing pain, etc), stiffness (minutes/hours), functional impairment and disability. Within each of these domains, several instruments may be considered, such as a simple visual analog scale [VAS, a continuous numerical scale that ranges from 0 mm, indicative of the best outcome (e.g. no pain), to 100 mm for the worst outcome (e.g. extreme pain)] or the Likert scale (a 5

point scale in which 0 designates the best outcome and 4 designates the worst outcome) (Dougados, Leclaire et al. 2000). Measurement may be by a more complex instrument, such as the Lequense Function Severity Index (Lequesne-Algofunctional-Index, self-administer questionnaire format), the Western Ontario and McMasters Universities Osteoarthritis (WOMAC) index, the Health Assessment Questionnaire (HAQ), and the Arthritis Impact Measurement Scales (AIMS) (Lequesne, Mery et al. 1987; Bellamy, Buchanan et al. 1988; Meenan, Mason et al. 1992). The WOMAC (Western Ontario and McMaster Universities Osteoarthritis Index) is a self-administered and disease-specific health status measure developed for the assessment of the patients with OA of the hip and/or knee. This index consists of 24 questions in three subscales and probes clinically important symptoms in the areas of pain (5 questions), stiffness (2 questions) and physical function (17 questions) (Schnitzer and Hochberg 2002). The Health Assessment Questionnaire (HAQ) is another method and measures the difficulty in performing activities of daily living.

2.3 Rheumatoid Arthritis (RA)

Rheumatoid Arthritis (RA) is the most common chronic inflammatory joint disease that leads to premature functional disablity and death (Lawrence, Hochberg et al. 1989; Pincus and Callahan 1989; Lawrence, Helmick et al. 1998). It is a systemic auto-immune disorder and typified by widesread and persistent inflammation of the synovial lining of the (mainly peripheral) joints and tendon sheaths (NICE 2001). Over time, bone erosion, destruction of cartilage, and complete loss os joint integrity can occur. Its course vary markedly and is often associated with non-articular features affecting multiple organ systems (NICE 2001).

2.3.1 Aetiology, Classification and Diagnosis of RA

The cause of RA is unknown, but it is considered as a inflammatory disease because patients with RA appear to have an abnormal immune system response. Evidence points to a complex interplay between environmental and genetic factors (Rindfleisch and Muller 2005). In monozygotic twins, there is a more than 30% concordance rate for RA development , and 80% of whites with RA express the HLA-DR1 or –DR4 subtypes (Rindfleisch and Muller 2005). These two alleles, as well as others, may confer susceptibility to more severe disease by causing a specific arthrogenic peptide to be presented to the CD^{+} T cells (Rindfleisch and Muller 2005). The "heritability" of the disease has been put forward by twin, family and segregation studies.

Joint damage in RA begins with the proliferation of synovial macrophages and fibroblasts after a triggering incident, possibly autoimmune or infectious (Rindfleisch and Muller 2005). Lymphocytes infiltrate perivascular regions, and endothelial cells proliferate (Rindfleisch and Muller 2005). Neovascularization then occurs (Rindfleisch and Muller 2005). Blood vessels in the affected joint become occluded with small clots or inflammatory cells (Rindfleisch and Muller 2005). Over time, inflamed synovial tissue begins to grow irregularly, forming invasive pannus tissue (Rindfleisch and Muller 2005). Pannus invades and destroys cartilage and bone (Rindfleisch and Muller 2005). Multiple cytokines, interleukins, proteinases, and growth factors are released, causing further joint destruction and the development of systemic complications (Rindfleisch and Muller 2005). Currently it is debated whether RA is a group of conditions with common features.

Diagnosis of RA is primarily clinical and based on number of criteria such as : symmetry of affected joints, morning stiffness, the presence of subcutaneous nodules and high serum rheumatoid factor (RF) levels (NICE 2001). The revised classification criteria of the American Rheumatism Association (ARA) is given in Appendix II (Arnett, Edworthy et al. 1988; NICE 2001). In typical outpatient pratice, a definitive diagnosis using these criteria may be difficult to obtain early in the disease process (Rindfleisch and Muller 2005). Radiography may be utilised to determine the degree of joint destruction and to monitor disease progression especially in more advanced states of disease (NICE 2001).

Unlike in OA, blood tests and serology are more informative in patients with RA (NICE 2001). Erythrocyte sedimentation rate (ESR) and C-reactive protein (CRP) levels are often elevated in proportion to the inflammatory process (NICE 2001). RF is present in about 70% of the patients, although not pathognomonic (NICE 2001). The anti-nuclear antibody (ANA) is also positive in 20-30% of patients with RA and is more common in patients with extra-articular manifestations (NICE 2001). A blood test may reveal a normocytic-normocytic anaemia in some patients (NICE 2001). Aspiration of a joint which demonstrates effusion may also be useful, especially for the elimination of other conditions such as septic arthritis (NICE 2001).

2.3.2 Prevalence & Risk Factors of RA

Variations in the prevalence of RA have been observed both over time and geographically (Guillemin, Saraux et al. 2005). The hypothesis that the occurrence of RA has declined has been suggested in countries where epidemiological studies have

been conducted in previous decades, particularly in the U.K. and USA (Guillemin, Saraux et al. 2005). Studies has shown a geographical distribution in the prevalence estimates varying from 0.8-1% formerly in northern countries estimates to 0.3-0.5% recently in southern countries (Guillemin, Saraux et al. 2005). The incidense of RA is estimated to be around 20 to 60 people per 100,000, but due to the long duration of the disease, the prevalence is much higher around 500 to 1000 people per 100, 000 (0.5-1%) (Scott, Shipley et al. 1998; NICE 2001).

Women prior to menopause are affected three times more than men (NICE 2001), but after the menopause the frequency of onset is similar between sexes (NICE 2001). The typical age of onset is 20 to 45 years of age & over 75% of patients are female (Silman 1998) A positive family history, older age, silicate exposure, and smoking (Criswell, Merlino et al. 2002) are associated with an increased risk for developing RA (Rindfleisch and Muller 2005). Consumption of more than three cups of coffee daily, especially decaffeinated coffee, may also contribute (Criswell, Merlino et al. 2002).

2.3.3 Clinical features of RA

Typically (in 70 percent of cases), RA manifests as slowly progressing, symmetrical, peripheral polyarthritis, which evolves over a span of a few weeks or months, although one third of patients initially experience symptoms at just one location or a few scattered sites (NICE 2001; Rindfleisch and Muller 2005). In most patients, symptoms occur with one joint and are often accompanied by prodromal symptoms of anorexia, weakness, or fatigue (Rindfleisch and Muller 2005). However,

in the rapid-onset form of RA (15% of patients), severe symmetrical polyarthritis may develop over a few days (sometimes explosively overnight), but surprisingly these cases have better prognosis (NICE 2001). In 8 to 15 percent of patients, symptoms begin within a few days of a specific inciting event, such as an infectious illness (Rindfleisch and Muller 2005).

Joints more commonly affected are those with the highest ratio of synovium to articular cartilage (Rindfleisch and Muller 2005). The wrists are nearly always involved, as are the proximal interphalangeal and metacarpophalangeal joints (Rindfleisch and Muller 2005). The distal interphalangeal joints and sacroiliac joints tend not to be affected (Rindfleisch and Muller 2005). Rheumatoid joints typically are swollen, warm and tender due to the inflammatory activity, but they usually are not erythematous (Rindfleisch and Muller 2005). Prominent epitochlear, axillary, and cervical lymph nodes may be noted (Rindfleisch and Muller 2005). Muscles near inflamed joints often atrophy (Rindfleisch and Muller 2005). Morning stiffness lasting at least 45 minutes after initiating movement is common and patients often hold joints in flexion to minimize painful distension of joint capsules (Rindfleisch and Muller 2005). Bone destruction and permanent deformities may develop due to persistent inflammation as the disease progresses (NICE 2001).

Low-grade fever, fatigue, malaise, and other systemic complaints may arise, especially in an acute presentation (Rindfleisch and Muller 2005). Rheumatoid nodules, Sjörgen's syndrome, episcleritis and scleritis, interstitial lung disease, pericardial disease, systemic vasculitis, neuropathies, renal amylodiosis and Felty's syndrome are

extraarticular manifestations of RA and are often indicative of poor prognosis (NICE 2001).

2.3.4 Treatment strategies for RA

Optimal management of RA requires early diagnosis and treatment to control the underlying inflammatory process, and thereby, to reduce the probability of irreversible joint damage (NICE 2001). The aim of treatment is to reduce pain and stiffness as well as minimising joint functional loss by preserving joint movement, preventing deformities and preserving the patient's quality of life.

Education, dietary advice, physiotherapy and occupational therapy are the most commonly non-pharmacological management strategies employed. On the other hand, pharmacological treatment options include NSAIDS, DMARDs (disease-modifying antirheumatic drugs) like sulphasalazine, methotrexate, penicillamine, gold compounds, and steroids. Surgery may be considered if all other treatment options fail to control disease progression. Surgical procedures involve joint replacement, synovectomy, and other interventions like carpal tunnel decompression, and tendon release.

Although previously NSAIDs and analgesics were the first treatment option, with the addition of DMARDs in a stepwise fashion during the course of treatment ("pyramid approach"), the "reverse pyramid approach" is now favoured. This involves early referral to a rheumatologist and initiation of DMARDs treatment and has been shown to reduce the progression of joint damage (Weinblatt 2003). This change of approach is the result of several findings: (i) joint damage begins early in the disease,

(ii) DMARDs have significant benefits when used early, (iii) the benefits of DMARDs may be enhanced when the drugs are used in combination (Pincus, O'Dell et al. 1999; Lipsky, van der Heijde et al. 2000; Weinblatt 2003), and (iv) a number of new DMARDs are now available with good evidence of beneficial effect (Olsen and Stein 2004) (Rindfleisch and Muller 2005).

NSAIDs, salicylates and CO X-2 inhibitors are used for initial treatment of RA, because they reduce joint pain and swelling. As they do not alter the disease progression of RA, they should not only be used alone. RA patients are almost two times more likely to have serious complications from NSAIDs use than patients suffering from OA (Singh and Triadafilopoulos 1999), and they should have been observed closely for symptoms of GI side-effects (American College of Rheumatology Subcommittee 2002). The first COX-2 inhibitor to obtain license for relief of signs and symptoms of OA and RA was celecoxib. Although rofecoxib received a license for the relief of the signs and symptoms of OA in 1999, rofecoxib did not receive a license for use in RA till 2002, only after the results of the VIGOR trial were available. The VIGOR trial was a large post-marketing RCT involving 8076 RA patients at least 50 years of age (or at least 40 years of age and receiving glucocorticoid therapy) who were to receive either 50mg of rofecoxib daily or 500mg of naproxen twice daily (Bombardier, Laine et al. 2000). The primary endpoint was confirmed clinical upper gastrointestinal events (gastroduodenal perforation or obstruction, upper GI bleeding, and symptomatic gastroduodenal ulcers) (Bombardier, Laine et al. 2000). The VIGOR trial was the first RCT to clearly illustrate the GI safety advantage of COX-2 inhibitors over non-selective NSAIDs, as a previous large post-marketing trial [Celecoxib Long-Term Arthritis Safety Study (CLASS)] never managed to illustrate a statistical

significant benefit with celecoxib. The randomised, double-blind, controlled CLASS trial of 8,059 patients was designed to compare the GI safety of celecoxib with that of ibuprofen and diclofenac. The majority of the patient had OA (72%), and the rest had RA (28%) (Silverstein, Faich et al. 2000). At study entry, 60% of patients were taking corticosteroids, and approximately 10% had a history of GI bleeding or ulcer (Silverstein, Faich et al. 2000). Although the incidence of ulcer complications in the celecoxib (400mg twice daily) arm was lower than with in the NSAIDs arms taken together or separately, the differences between the observed event rates were not statistically significant for any comparison in an intent-to-treat analysis (vs ibuprofen 800mg three times a day, p=0.414; vs diclofenac 75mg twice a day, p=0.64; vs either NSAID, p=0.45) (Bombardier 2002). The incidence of ulcer complications and symptomatic ulcers with celecoxib was significantly lower compared with ibuprofen (p=0.017), but not with diclofenac (Bombardier 2002). Finally, it has to be mentioned that, although COX-2 inhibitors were welcomed for patients with RA due their gastroprotective effects, COX-2 inhibitors are no more effective than non-selective NSAIDs, while costing as much as 15-20 times more per month of therapy than generic NSAIDs (American College of Rheumatology Subcommittee 2002). Other available strategies for patients benefiting from an NSAID but who are at increased risk of serious adverse GI effects are the use of low-dose prednisolone, the use of a nonacetylated salicylate or the concomitant use of an NSAID and a gastroprotective agent (such as H2-antagonists, proton pumps inhibitors and prostaglandin analogs) (American College of Rheumatology Subcommittee 2002).

2.3.5 Outcome measures used in the assessment of RA

Clinical trials of pharmacologic agents in OA or RA employ several measures of efficacy recommended by Outcome Measures in Rheumatoid Arthitis Clinical Trials (OMERACT), a group endorsed by the International League of Associations of Rheumatology (ILAR) and the World Health Organisation (WHO) (Schnitzer and Hochberg 2002). The most commonly used outcome measures used in the assessment of RA include patient and physician global assessments, number of swollen or tender joints, pain score (usually VAS), morning stiffness (minutes or hours), time to walk 50 feet, grip strength for both hands (mm/Hg), functional status, radiological progression, articular index, Ritchie's index, changes in acute phase reactants such as C-reactive protein (CPR) and erythrocyte sedimentation rate (ESR) (NICE 2001). Comparative drug trials also utilise the rates of withdrawal from treatment (due to both insufficient response and adverse effects), compliance, concomitant paracetamol consumption, global assessment of tolerability, endoscopically detected ucler rates and also a variety of laboratory tests (liver function tests, urinalysis, complete blood count etc.) to evaluate the safety of the drug (NICE 2001).

2.3.6 Cardiovascular risk factors & Atherosclerosis in RA

As early as 1976, studies suggested that patients with RA might suffer an increased risk of CV diseases (Monson and Hall 1976). Cardiovascular disease constitutes an increasingly recognised contributor to the excess morbidity and mortality in RA patients (Solomon, Karlson et al. 2003; Wolfe, Freundlich et al. 2003). Reilly et al (1990) suggested that about half of all RA deaths can be attributed to CV disease

(Reilly, Cosh et al. 1990). It has long been known that T-cells play an important part in the pathogenesis of RA (Lee and Weinblatt 2001). However, recent data has led to the postulate that T-cell abnormalities are involved in acute coronary syndromes and atherosclerotic plaque instability (Liuzzo, Goronzy et al. 2000; Weyand, Goronzy et al. 2001). Furthermore, cytokines, C-reactive protein, and other inflammatory markers, known to be elevated in RA, are also elevated before and at times of ischaemic injuries (Liuzzo, Biasucci et al. 1994; Ridker, Cushman et al. 1997; Ridker, Buring et al. 1998). Methotrexate, known to downregulate T-cell activity, has been associated with reduced cardiovascular mortality in patients with RA (Choi, Hernan et al. 2002). Traditional risk factors such as age, sex, smoking status, diabetes mellitus, hypercholesterolaemia, systolic blood pressure, and body mass index, have been found to not adequately account for the CV disease extent oberved in RA (del Rincon, Williams et al. 2001; Dessein, Joffe et al. 2003). Markers of current and cumulative inflammation (white cell counts and radiographic joint damage, respectively) are associated with ultrasonographically determined subclinical atherosclerosis (Kumeda, Inaba et al. 2002; Nagata-Sakurai, Inaba et al. 2003), a predictor of CV events (Belcaro, Nicolaides et al. 2001) (Dessein, Joffe et al. 2005). It has to be noted that a recently published population based study of a cohort of 603 adult RA patients that fullfilled the ACR criteria for RA between 1955 and 1995, concluded that markers of systemic inflammation confer a statistically significant additional risk for CV death among RA patients, even after controlling for traditional CV risk factors and comorbidities (Maradit-Kremers, Nicola et al. 2005).

2.3.7 Juvenile RA

Rofecoxib was the first and only coxib to ever receive license for the relief of the signs and symptons of pauciarticular and polyarticular juvenile RA (JRA) in children older than 2 years of age and weighing at least 10 kilograms (22 pounds) or more with a maximum dose of 25mg daily [2-11 years (10-41kg) : 0.6mg/kg once a day, 2-11 years (>41kg) & 12-17 years: 25mg once a day]. FDA approval of rofecoxib for JRA was based on the largest JRA study ever conducted, which included 310 paediatric and adolescent patients aged between 2 to 17 years with active pauciarticular or polyarticular JRA. The contribution of COX-2 inhibitors is likely to be less significant as paediatric patients do not typically suffer significant GI problems with NSAIDs as frequently as adults (Ilowite 2002). In a database study of 702 patients with JRA, only 5 patients were found to have had a gastropathic event attributable to NSAID therapy (Keenan, Giannini et al. 1995). On the other hand, in selected populations (e.g. patients with abdominal pain and anaemia), the incidence of gastroduodenal ulcers is much higher (Mulberg, Linz et al. 1993; Len, Hilario et al. 1999).

2.4 Acute Pain

The International Association for the Study of Pain defined pain as "an unpleasant sensory and emotional experience associated with actual or potential tissue damage, or described in terms of such damage (International Association for the Study of Pain ((IASP) 1994)). Pain is devided in two types on the basis of duration, acute and chronic. Acute pain, usually results from tissue injury, ischaemia, inflammation, or visceral obstruction and normally resolves once the inciting event has passed and the

involved tissues have healed (Fine 2002). Chronic pain can be defined as pain that persists for more than 3-6 months.

Acute pain can in most of the cases be managed easily, as it usually resolves with the resolving of the cause e.g. tissue injury healing. Psychologically, patients can expect to experience improvement in pain relief. Thus, analgesia requirements decrease along the same time line limiting the patient's exposure to medication. From the latter, it can be expected that the incidence of adverse drug reactions would be minimal or at least less than long-term administration of medication. The use of traditional pain medications, including opioid analgesics, non-opioid analgesics (e. g. acetaminophen), and NSAIDs, has limitations. Some medications provide suboptimal analgesia, whereas others are hindered by adverse effects. Even short-term use of NSAIDs has been associate with GI toxicity (Gabriel, Jaakkimainen et al. 1991; Langman, Jensen et al. 1999). However, the most critical when choosing a medication to treat acute pain are time to onset of analgesia and efficacy of the analgesic.

2.4.1 Use of COX-2 inhibitors in acute pain

In May 1999, rofecoxib was the first COX-2 inhibitor to receive a license for the management of acute pain and the treatment of primary dysmenorrhoea. Only two years later (October 2001), celecoxib was licensed for the same indications by the FDA, with valdecoxib, following shortly afterwards with a license only for primary dysmenorrhoea. Parecoxib was approved in UK for the short-term relief of post-operative pain (3 days use only). The approval of COX-2 inhibitors for these indications was received with much enthusiasm, as they had the advantage of requiring

fewer administrations (rofecoxib has a once-daily dosing regimen for all indications with maximum analgesic efficacy being provided by doses of 50mg, the approved analgesic dose), they had fewer adverse-effects (avoidance of the opioid-related side-effects, and reduced GI toxicity, which is of importance in post-operative patients), while theoretically could provide analgesia equal to traditional NSAIDs. Additionally, they reduced the amount of opiates needed for pain control, while also acting synergistically by improving opiates ability to control pain (Reuben and Connelly 2000).

2.4.2 Clinical evidence of analgesics in acute pain

Traditionally, the design and contact of clinical trials of analgesics in acute pain has been good (Moore, Edwards et al. 2005). Houde, Beecher and others developed the single-dose model, which is successfully used for over 50 years (Denton and Beecher 1949; Hoode and Wallenstein 1954; Beecher 1957; Hoode 1962; Moore, Edwards et al. 2005). Selection and observer bias were minimised by the specification of randomised, double-blind trials (Moore, Edwards et al. 2005). As pain is subjective and there are no objective measures, measurement of pain must, therefore, rely on recording the patient's report. However, pain intensity and relief measurements were standardised, using categorical verbal rating scales (Figure 2.1), and later, standardised visual analogue scales (Figure 2.2). From these scales, the total pain relief (TOTPAR) or summed pain intensity difference (SPID) over 4-6 hours were usually taken as primary outcomes. For example, TOTPAR is measured by calculating the area under the curve for pain relief against time. If a patient had complete pain relief immediately, and sustained it for the full six hours of measurement, then the maximum TOTPAR would

be attained (in this case a score of 4 points times 6 hours, giving a TOTPAR of 24, the maximum achievable). Another patient who had a score of 12 would have 50% of the maximum, or 50% maxTOTPAR (Figure 2.3). The necessity for patients to have moderate or severe pain at baseline was also recognised as being crucial to produce sensitive assays (Lasagna 1962). Pain relief scales are considered to be more useful than pain intensity scales, probably because patient have the same baseline relief (zero) whereas they could start with different baseline intensity (usually moderate to severe). Relief scales results are thus easier and more sensitive to compare providing a sensitive quantitative measurement of efficacy. However, they do not provide information about the onset and peak of the analgesic effect. If the onset or peak or time to remedication / rescue medication are important, then, time to maximum pain relief (or reduction in pain intensity) or time for pain to return to baseline or time to remedication / rescue medication respectively are necessary.

Categorical Verbal Rating Scales

Pain Intensity	
Severe	3
Moderate	2
Slight	1
None	0

Pain Relief	
Complete	4
Good	3
Moderate	2
Slight	1
None	0

Figure 2.1 Categorical verbal rating scales. Categorical scales use words to describe the magnitude of the pain. The patient picks the most appropriate word.

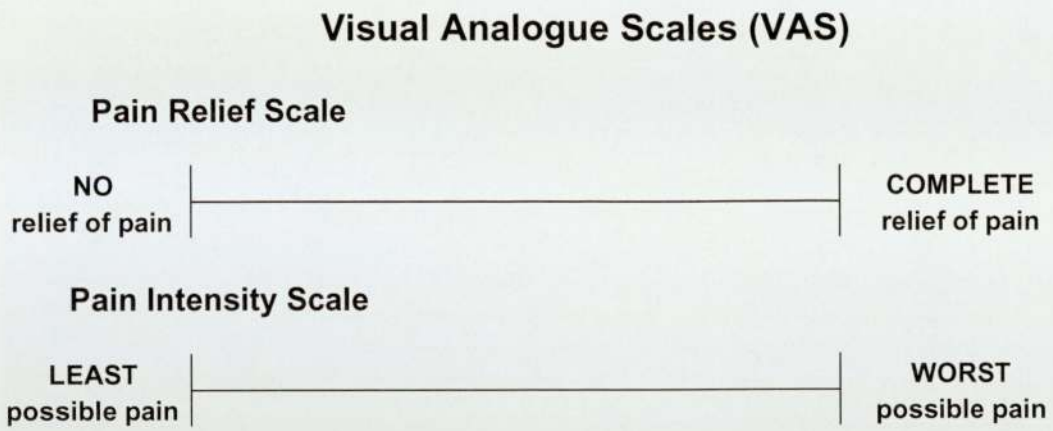
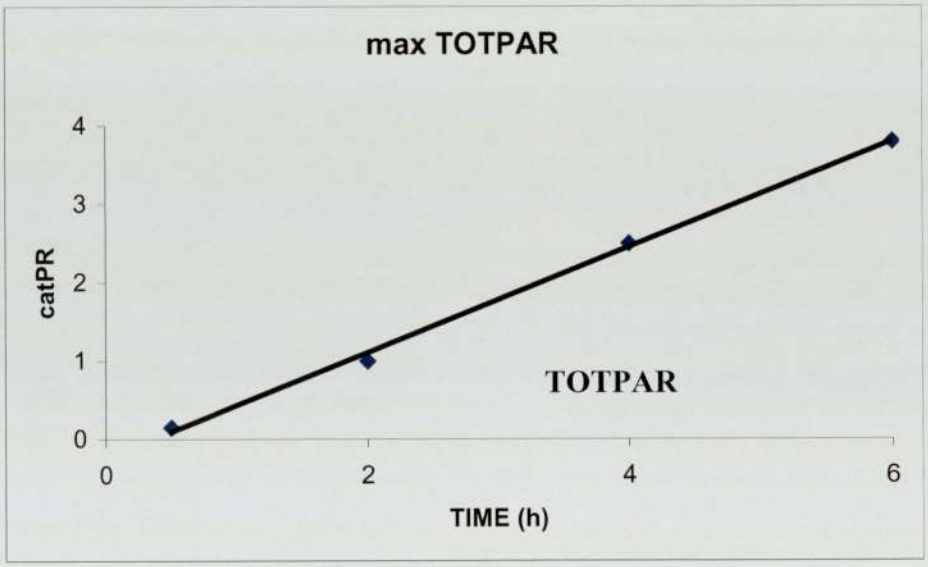


Figure 2.2 Visual analogue scales (VAS). Patients mark the line at the point corresponding to their pain. The scores are obtained by measuring the distance between the left end (i.e. "NO relief of pain" and "LEAST possible pain") and the patient's mark, usually in millimetres.



$$\frac{\text{TOTPAR}}{\text{maxTOTPAR}} \times 100 = \%\text{maxTOTPAR}$$

Figure 2.3 Calculating TOTPAR and %maxTOTPAR.

The reporting of acute pain trials has often been less than good. Outcomes reported in 160 high quality trials in acute pain were inconsistent (Barden, Edwards et al. 2004). Most trials (87%) had a measure of pain intensity, pain relief, or global outcome scale, but did not always use standard scales (Moore, Edwards et al. 2005). Reporting of other outcomes, like time to remedication and adverse effects was variably reported usually in different ways (Edwards, McQuay et al. 1999) Inconsistency in choice of outcome, poor description of outcomes, and poor quality of result reporting make difficult comparisons across trials or between drugs, as seen in other types of pain (e.g. OA trials) (Gotzsche 2001).

A number of different clinical situations, recognised as appropriate and validated models for licensing perposes, have been used to measure efficacy of analgesics in acute pain. General pain indications either acute or chronic pain, should be based on data derived from studies of visceral and somatic pains as well as of pain with different intensities e.g. mild to moderate and severe. These include third molar dental extraction, orthopaedic or general surgery usually with moderate to severe pain intensity (Table 2.3). It can be reasonably assumed that analgesics do not behave differently in different acute pain models e.g third molar dental extraction, orthopaedic surgery (Edwards, Oldman et al. 1999; Barden, Edwards et al. 2004), although in some clearly clinical situations (elderly patients, or hepatic or renal insufficiency) the metabolism and excretion of the drug can be affected. Thus, patients with severe renal and hepatic impairment are generally excluded from pain trials.

Table 2.3 Validated Pain Models for Licensing

Type of Pain	Pain Intensity	Trial Duration	Pain Model
Acute Pain	Mild to Moderate	Days (<1week)	Tooth extraction, sprain, minor surgery (e.g. cutaneous surgery, hernia, headache (other than migraine), sore throat, low back pain, primary dysmenorrhoea
Acute Pain	Moderate to Severe	Less than 48 hours to 1 week	Surgical removal of impacted teeth (third molar), renal and biliary colic, well-defined major orthopaedic surgery, well-defined major abdominal/ thoracic surgery, major skeletal trauma
Chronic Pain	Mild to Moderate	Greater than or equal to 3 months	OA, RA, low back pain
Chronic Pain	Moderate to Severe	Greater than or equal to 1 month	Cancer, skeletal metastasis with movement related pain

To obtain a marketing authorisation for acute pain management in surgery the applicant must demonstrate safety and efficacy on somatic (i.e. major orthopaedic) and visceral (abdominal, gynaecological or thoracic surgery) ((CPMP) 2002)). Appropriate studies on these populations can support a broader indication on acute pain management in general (moderate to severe pain) ((CPMP) 2002). Extrapolation, however, of results between visceral and somatic pain is not acceptable ((CPMP) 2002). An indication of mild to moderate acute pain management excluding primary dysmenorrhoea can be supported by two or more studies on mild to moderate pain using different pain models (e.g. one on tooth extraction and one on sprains) ((CPMP) 2002). Dysmenorrhoea can be used as one of the models to support an indication on mild to moderate pain management. However, dysmenorrhoea is currently the subject

of dedicated studies if the development program is to support this specific indication, while efficacy evaluations should take into account the intermittent pain conditions.

A systematic review of celecoxib (418 patients from two included studies) showed fair to good efficacy for post-operative pain with a NNT of 4.5 (95% CI, 3.3 to 7.2) compared to placebo (Barden, Edwards et al. 2003) meaning that 4.5 patients have to be treated with celecoxib before one will experience at least 50% pain relief, who would not have done with placebo. However, the recommended dose of celecoxib for acute pain relief is 400mg as a first dose, with a second dose of 200mg, if necessary, on the first day of medication. Both trials included in the systematic review assessed a single dose of 200mg to treat moderate to severe acute pain, and thus the clinical applicability of the findings of the above systematic review should be considered carefully (Barden, Edwards et al. 2003). Comparing celecoxib to the results of other systematic reviews NSAIDs or COX-2 inhibitors conducted with the methods for post-operative pain lead to less promising results. Celecoxib 200mg is less effective than rofecoxib 50mg, which has a NNT of 2.2 (95% CI 1.9 to 2.4) (667 rofecoxib 50mg patients and 315 placebo patients included in analysis with a total of 982 patients) (Barden, Edwards et al. 2005), and ibuprofen 400mg with a NNT of 2.4 (95% CI 2.3 to 2.6) (total number of subjects 4703) (Barden, Edwards et al. 2003). Analgesics comparable to celecoxib 200mg in efficacy include aspirin 600/650mg [NNT 4.4 (95% CI 4.0 to 4.9), number of patients 5061] (Edwards, Oldman et al. 1999) and paracetamol 1G [NNT 3.8 (95% CI 3.4 to 4.4), number of patients 2759] (Barden, Edwards et al. 2004).

In a systematic analysis for oral valdecoxib and injected parecoxib the NNT for one patient to experience at least 50% relief over six hours following a single oral dose of valdecoxib 20mg and 40 mg was 1.7 (95% CI 1.4 to 2.0) and 1.6 (95% CI 1.4 to 1.8) respectively, while following IV parecoxib 20mg and 40mg was 3.0 (95%CI 2.3 to 4.1) and 2.3 (95% CI 2.0 to 2.6) respectively (Barden, Edwards et al. 2003). Additionally, valdecoxib has demonstrated analgesia superior to that of placebo in post-operative knee surgery (Reynolds, Hoo et al. 2003).

The efficacy of a single dose of rofecoxib 50mg, celecoxib 400mg and 200mg, and ibuprofen 400mg was evaluated in 482 individuals after extraction of at least 2 third molars (Malmstrom, Fricke et al. 2002). The time to onset of analgesic efficacy, defined as the median time to confirmed perceptible pain relief, was shorter for rofecoxib 50mg (36 minutes) compared with celecoxib 400mg (54 minutes), although the difference did not reach statistical significance. However, patient receiving rofecoxib 50mg experienced a significantly faster onset of analgesic effect than did patients who received celecoxib 200mg (36 versus 72 minutes, respectively; $p<0.001$). Patients receiving rofecoxib 50mg and ibuprofen 400mg experienced a similar time to onset of effect (36 and 30 minutes, respectively). The onset of analgesic efficacy of all the active agents was significantly faster than that with placebo ($p<0.001$). The median time to remedication / use rescue medication was significantly longer for rofecoxib 50mg than for celecoxib 400mg (15.9 versus 10.6 hours, respectively; $p<0.05$) and than celecoxib 200mg and ibuprofen 400mg (15.9 versus 6.8 hours ($p<0.001$) and 15.9 versus 10.0 ($p<0.001$) respectively). The percentages of patients who required rescue medication were 51%, 66%, 69%, 87% and 98% in the rofecoxib 50mg, celecoxib 400mg, celecoxib 200mg, ibuprofen 400mg and placebo groups, respectively.

The weighted (by trial size) median time to remedication for valdecoxib 20mg based on 101 patients was greater than 17.5 hours, while for valdecoxib 40mg based on 199 patients was greater than 24 hours (Barden, Edwards et al. 2003). For parecoxib 20mg and 40mg IV and for placebo the mean time to remedication (weighted by trial size) was 5.6 hours (based on 170 patient) and 8.7 hours (based on 173 patients) and 1.7 to 1.8 hours respectively (Barden, Edwards et al. 2003).

Trials of COX-2 inhibitors (celecoxib, rofecoxib, valdecoxib) in acute pain were short in duration (normally around 24 hours), and adverse effects were inconsistently reported (Moore, Edwards et al. 2005). These described were generally mild and transient. The three most commonly observed adverse-effects were for rofecoxib 50mg nausea, post-extraction alveolitis (Barden, Edwards et al. 2005) and vomiting and for celecoxib 200mg nausea, vomiting and headache (Barden, Edwards et al. 2003). Nausea, vomiting, dizziness, and headache were the most common adverse effects reported for valdecoxib 20mg and 40mg. The absolute proportions of patients experiencing adverse effects was higher with placebo than with valdecoxib 20mg and 40mg, expect from vomiting in the case of valdecoxib 40mg.

2.5 Prevention of Cancer & Familiar Adenomatous Polyposis

Cyclooxygenase (COX) has recently been discussed in many clinical contexts from arthritis to Alzheimer's disease (AD) to neoplasia. Cancer prevention, at present a better option than cancer treatment is entering an era when it appears to be a realistic possibility (Kashfi and Rigas 2005). During the 1990s an association was made between regular consumption of NSAIDs (particularly aspirin) and a reduction in the

incidence of colon cancer (Dubois, Abramson et al. 1998; Warner and Mitchell 2004). The first epidemiological demonstration that NSAIDs prevent human colon cancer was published in 1988 (Kune, Kune et al. 1988). The link between NSAID consumption and cancer prevention is based on two sets of data: (i) epidemiological studies documenting an association between NSAID use and cancer risk and (ii) interventional clinical trials demonstrating that the administration of NSAIDs can actually prevent cancer (Rigas and Kashfi 2005). The epidemiological studies reported, collectively describing results on more than 1 million subjects, have pointed out the protective effect of NSAIDs against colon, oesophageal, gastric, breast (oestrogen receptor-positive) and perhaphs pancreatic and ovarian cancers (Thun, Henley et al. 2002; Rigas and Kashfi 2005). The seminal epidemiological observation that NSAIDs prevent colon, and possibly other, cancers has led to the unambiguous demonstration that aspirin does prevent colon cancer. Two randomised interventional studies using polyp recurrence as a general endpoint demonstrated the chemopreventive effect of aspirin (Baron, Cole et al. 2003; Sandler, Halabi et al. 2003). The relative risks following administration of aspirin ranged between 0.59 and 0.96, depending on the specific endpoint and aspirin dose. Several aspects of this effect seem unclear at this point, but the above studies constitute proof of chemoprevention by NSAIDs.

Current NSAIDs, however, as chemopreventive agents cannot be used without concern due to two prohibitive limitations concerning their efficacy and safety. NSAIDs can prevent at best 50% of the cases of colorectal cancer (the most thoroughly studied cancer type for prevention with NSAIDs) (Thun, Henley et al. 2002). Although no precise numbers are available for the incidence of adverse-effects of NSAIDs, it seems that among patients using NSAIDs, up to 4% per year suffer serious GI

complications (Bjorkman 1999; Rigas and Kashfi 2005). Additionally, the recent withdrawal of rofecoxib due to potential thrombotic cardiovascular adverse-effects reported in the Adenomatous Polyp Prevention on VIOXX® trial (APPROVE) of 2586 patients in total with a history of coloractal adenomas treated with rofecoxib 25mg daily or placebo (3.5% of rofecoxib patients and 1.9% of placebo suffered myocardial infarctions or strokes), have questioned the cardiovascular safety of NSAIDs and COX-2 inhibitors used long-termly, as it would be required for chemoprevention. In chemotherapy, both patient and clinician accept substantial treatment-related toxicity to save the patient's life from a fully developed cancer. In contrast, chemoprevention require agents with an efficacy approaching 100% and a safety profile that is almost perfect, as the agent is going to be prescribed oftenly to healthy subjects for a cancer that may never develop.

The first concrete clinical evidence of COX-2 selective inhibitor efficacy in humans was obtained from a phase II study of 77 patients with familiar adenomatous polyposis (FAP) (Steinbach, Lynch et al. 2000). Patients that received 400mg of celecoxib twice a day for 6 months had a 28% reduction in the mean number of colorectal polyps and a 30.7% reduction in the polyp burden (the sum of polyp diameters), as compared to with reductions of 4.5 and 4.9 % respectively, in the placebo group (Steinbach, Lynch et al. 2000). Another later study showed significant reduction in duodenal polyposis in patients with FAP after 6 months of treatment with 400mg of celecoxib twice daily (Phillips, Wallace et al. 2002). Overall, patients taking celecoxib 400 mg twice daily showed a 14.5% reduction in involved areas compared with a 1.4% for placebo ($p=0.436$) (Phillips, Wallace et al. 2002). However, patients with clinically significant disease at baseline (greater than 5% covered by polyps)

showed a 31% reduction in involved areas with celecoxib 400 mg twice daily compared with 8% on placebo (p=0.049) (Phillips, Wallace et al. 2002).

This results are of particular importance, as colon cancer has been shown to strike between 4-5% of normal individuals, independent of genetic risk, in numerous worldwide studies (Haller 2003). Celecoxib is the only selective COX-2 inhibitor ever receiving FDA approval for reducing the number of adenomatous colorectal polyps in FAP at December 1999.

2.6 Prevention and Treatment of Alzheimer's Disease

There are approximately over 750,000 people in the UK affected by dementia. Alzheimer's Disease (AD) is the most common form of dementia, making up to 55 % of all cases of dementia, and the number of cases is expected to increase as the population ages (Alzheimer's (Society 2006). AD progresses from mild memory impairment to profound dementia and eventual death, typically over a period of 8-10 years.

As the pathogenesis of AD unfolds, inflammation is believed to be involved with the process of neurodegeneration (Aisen 2002). Attention has therefore focused on the research for disease-modifying therapy and NSAIDs and COX-2 inhibitors are being evaluated for the treatment and the prevention of AD. One major strategy is to slow the rate of neuronal loss through suppression of inflammatory mechanisms in AD (Aisen 1997). A number of inflammatory processes have been identified in the brain of patients with AD, including complement activation, accumulation of activated microglial cells, and a cytokine-driven acute-phase response. Several epidemiological

studies have suggested a beneficial effect with chronic NSAID use (McGeer, Schulzer et al. 1996), and as studies have demonstrated that COX-2 may play a role in neurodegenerative mechanisms (Pasinetti and Aisen 1998), COX-2 selective inhibitors may offer some degree of neuroprotection. However, NSAIDs are not an ideal class to inhibit acute-phase response and complement activation (Aisen and Davis 1994). The NSAIDs long-term adverse-effects need also to be taken into consideration.

Although none of the selective COX-2 inhibitors received license for treatment of prevention of AD, several RCTs have been conducted to assess their effect. A double-blind, multicenter, placebo-controlled, parallel group RCT enrolling 351 patients was conducted to determine whether treatment with rofecoxib (25mg daily) or a tradiotional NSAID (naproxen 220mg daily) slows cognitive decline in patients with mild to moderate AD. The primary outcome measure was 1 year change in the Alzheimer Disease Assessment Scale-Cognitive (ADAS-Cog) subscale score. The authors concluded that rofecoxib or low-dose naproxen does not slow cognitive decline in patients with mild to moderate AD (Aisen, Schafer et al. 2003). Another one-year RCT similarly reported that COX-2 inhibitors (rofecoxib 25mg once daily) failed to slow AD progression indicating either that the disease process is too advanced to modify patients with established dementia or that COX-2 does not play a significant role in the pathogenesis of the disorder (Reines, Block et al. 2004). Recently, in a bigger RCT with patients with mild cognitive impairment, Thal et al also concluded that rofecoxib was not different from placebo and that COX-2 inhibition is not a useful approach in AD (Thal, Ferris et al. 2005). Celecoxib RCTs for AD did not manage to illustrate that COX-2 inhibitors alter the progression of AD.

2.7 NSAIDs & selective COX-2 inhibitors adverse events profile

Under normal physiological conditions the constitutive COX-1 enzyme is responsible for gastric mucosal protection, maintenance of normal kidney function, and platelet aggregation. Thus, NSAIDs, as non-selective inhibitors, can cause gastrointestinal (GI) obstruction, perforation and bleeding, renal failure, lower extremity oedema and hypertension exacerbation, and endanger haemostatic integrity.

The potential of NSAIDs to cause GI toxicity is well known; an estimated 100,000 hospitalisations occur annually in the USA due to NSAID associated serious GI complications (Wolfe, Lichtenstein et al. 1999). The incidence of symptomatic ulcers and ulcer complications associated with standard NSAID was reported in 1998 to be approximately 1 - 4% per year (Singh 1998), being a significant iatrogenic cause of morbidity and mortality. Thus, selective and specific COX-2 inhibitors held the promise of fewer adverse-effects as far as the GI tract and platelets are concerned (FitzGerald and Patrono 2001), as the do not inhibit the COX-1 isoenzyme which is believed to catalyse the synthesis of gastroprotective prostaglandins.

Specific COX-2 inhibitors received license based on the divergent incidence of a surrogate endpoint (i.e. endoscopically diagnosed ulcers) from traditional NSAIDs comparators used at similarly effective doses in patients with arthritis (Wong, Wang et al. 2005). The precision of such efficacy endpoints, typically a mixture of subjective and objective assessments, received much criticism. Despite the absence of an indication of superior efficacy or an outcome study of adverse effects, celecoxib and rofecoxib were licensed and marketed aggressively (Wong, Wang et al. 2005). This led to their broad adoption, mainly by patients not at an increased risk of GI adverse effects

from standard NSAIDs (Wong, Wang et al. 2005). The CLASS study (Celecoxib Long-Term Arthritis Safety study), a post-marketing surveillance study, suggested that GI adverse effects (A/Es) seen with celecoxib are comparable with diclofenac (justified as almost equally selective for COX-2), but lower than ibuprofen. Conversely, the VIGOR study (Vioxx Gastrointestinal Outcomes Research) revealed a significant reduction (from 4% to 2%) in serious GI A/Es with rofecoxib 50mg daily, and was the first large post-marketing trial supporting COX-2 inhibitors as a gastroprotective strategy for people requiring chronic NSAID use.

However, VIGOR also revealed a fivefold divergence in the incidence of myocardial infarction (Bombardier, Laine et al. 2000; Juni, Nartey et al. 2004). The VIGOR study group presented the myocardial infarction data exclusively as a 'reduction in the risk of myocardial infarction in the naproxen group', on the basis of the documented inhibition of platelet aggregation by naproxen, but not rofecoxib (Bombardier, Laine et al. 2000; Juni, Nartey et al. 2004). However, even if one would expect an 'aspirin' effect of naproxen, the magnitude of difference observed in VIGOR was twice as great. The suggestion of cardiovascular hazard from rofecoxib was not novel. It has been observed since the mid 1990s that COX-2 was not extant in human platelets and that structurally distinct COX-2 inhibitors (rofecoxib and celecoxib) depressed substantially the biosynthesis of PGI_2 without a concomitant effect on thromboxane (TxA_2) or platelet aggregation ex vivo (Juni, Nartey et al. 2004; Krotz, Schiele et al. 2005; Wong, Wang et al. 2005). Figure 2.4 illustrates graphically the suggested class-based mechanism of cardiovascular hazard of the coxibs.

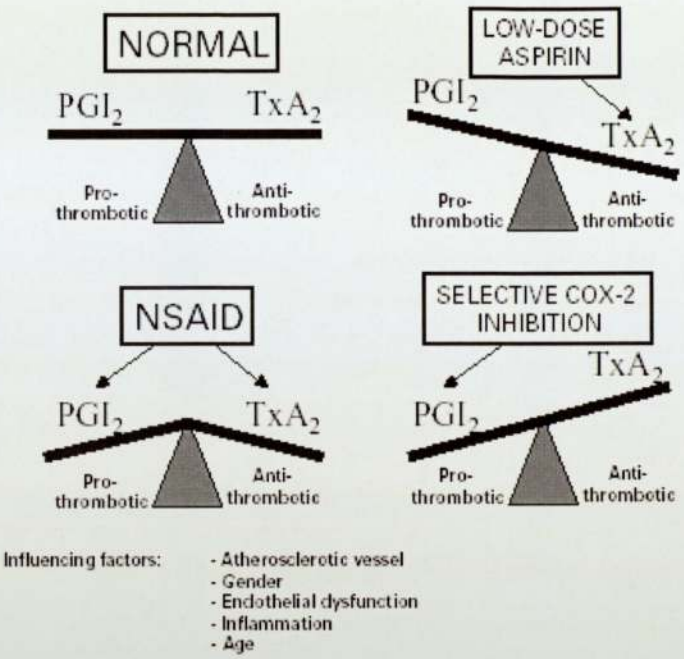


Figure 2.4 Influence of low-dose aspirin (ASA), non-selective cyclooxygenase inhibition (NSAID, high dose ASA) or selective inhibitors of COX-2 on the vascular balance of prostanoids regarding platelet activity and thus thrombosis. The effects of the respective drugs on vascular prostanoid formation and on platelets are depicted schematically. As the in vivo situation is far more complicated, this schematic panel only partly reflects a schematic of a physiological vascular situation. Whereas low-dose aspirin selectively inhibits TxA_2 formation in platelets and thus lowers systemic TxA2 levels, NSAID (or high-dose ASA) inhibit cyclooxygenases non-specifically and thus also decrease PGI_2 levels independent of the source. As it is now clear that COX-2 is constitutively expressed in the endothelium and kidney and significantly contributes to systemic PGI_2 formation even in healthy individuals, selective inhibitors of COX-2 decrease systemic levels of PGI_2, without altering TxA_2, resulting in enhanced platelet activation (Krotz, Schiele et al. 2005).

NSAID therapy can also be associated with changes in renal function, especially with respect to solute homeostasis and maintenance of renal perfusion (Brater, Harris et al. 2001). Deleterious changes in renal function are more likely with concurrent disease and medications (Brater, Harris et al. 2001). There is no clear distinction in 'physiological' constitutive COX-1 and 'inflammatory' inducible COX-2.

COX-1 expression can be increased only two- to fourfold under most circumstances, while COX-2 can be constitutively expressed in a few tissues as the kidney (particularly the macula densa in humans), the brain, the reproductive system and the lung (Katori and Majima 2000). However, considering COX-1 as the constitutive isoform and COX-2 only as the inducible is a simplification of biological complexity (Wong, Wang et al. 2005). The constitutive expression of COX-2 in the kidney raised the possibility that COX-2 inhibitors may carry the same risk of renal adverse effects, as do non-selective NSAIDs (Brater, Harris et al. 2001). On the other hand, the differential compartmental distribution of COX-1 and COX-2 might result in differences in renal effects of COX-2 inhibitors versus NSAIDs (Table 2.4) (Brater, Harris et al. 2001).

In clinical situations of decreased actual or effective circulating volume (including renal insufficiency, congestive heart failure, and cirrhosis), locally synthesised PGs play a critical role in maintaining renal perfusion. In these circumstances, endogenous renal PGs initiate counterregulatory vasodilatory mechanisms to offset the diminution in renal blood flow resulting from vasoconstriction that occurs from activation of the renin-angiotensin and adrenergic nervous system (Figure 2.5) (Brater, Harris et al. 2001). By inhibiting renal PG synthesis, NSAIDs allow unopposed vasoconstriction, potentially giving rise to acute renal failure (Brater, Harris et al. 2001).

Table 2.4. Distribution of COX-1 and COX-2 in the kidney. (Brater, Harris et al. 2001)

Tissue	Species									
	rat		rabbit		dog		monkey		human	
	COX-1	COX-2	COX-1	COX-2	COX-1	COX-2	COX-1	COX-2	COX-1	COX-2
Vasculature	C				C	C	C	C	C, C, C	C, C, C
Glomerulus		I, I C, C						C	C	C, C, C
Macula densa	C	I, I, I, I				C, I				C
Thick ascending limp of loop of Henle	C	C, C, C I, I, I, I		C, I		C, I				C
Collecting duct	C, C				C		C		C, C, C	
Interstitium	C, C	C, C, C				C	C		C, C, C	C

C denotes constitutive, I denotes inducible. Each notation represents a study wherein that isoenzyme was identified. For example, two separate studies identified COX-2 in rat glomeruli. Note that the lack of inducible COX-2 may indicate that it was not tested.

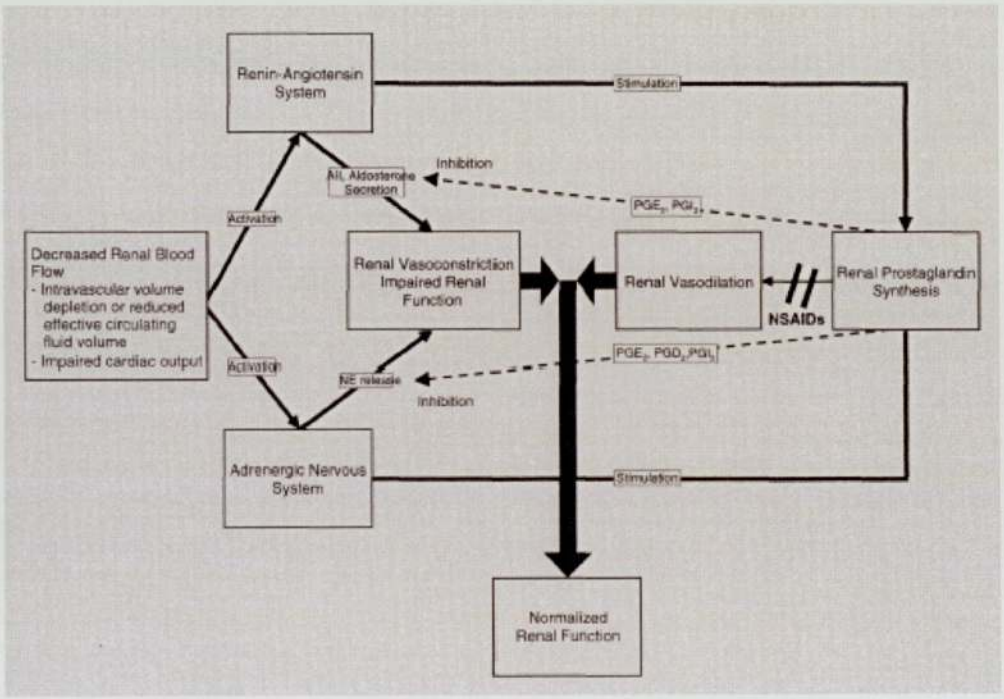


Fig. Critical role of renal PGs in mediating local counterregulatory mechanisms to offset a diminution in renal function in patients with reduced renal perfusion (e.g., volume depletion, impaired cardiac output). A fall in renal blood flow leads to synthesis of angiotensin II (AII), stimulation of aldosterone secretion, release of arginine vasopressin, increased sympathetic outflow, and release of norepinephrine (NE) which, collectively, result in renal vasoconstriction and impaired renal function. In these circumstances, renal PG synthesis is stimulated, and PGE_2, PGD_2, and PGI_2 are released, the actions of which are believed to normalize renal function by antagonizing the above vasoconstrictors. By impairing these PG-dependent, counterregulatory measures, NSAIDs may lead to a decreased GFR in patient groups. [Adapted from refs. 36 and 39].

Figure 2.5 PGs role in maintaining renal perfusion in clinical conditions of decreased actual or effective circulating volume. (Brater, Harris et al. 2001)

Collective data with both rofecoxib and celecoxib are consistent with the expectation that selective and specific inhibitors of COX-2 do not spare the kidney (Brater, Harris et al. 2001). COX-2 specific inhibitors have been found to reduce urinary sodium excretion upon onset of therapy (probably by COX-2 regulation of sodium reabsorption), although under different clinical circumstances COX-2 inhibitors can affect both solute homeostasis and renal hemodynamics (Brater, Harris et al. 2001). For both rofecoxib and celecoxib, lower-extremity peripheral oedema was reported in clinical trials (Brater, Harris et al. 2001). Aw et al (2005) have shown that coxibs (celecoxib and rofecoxib) can pause a non-significant increased risk of developing hypertension by increasing both systolic and diastolic blood pressure (Aw, Haas et al. 2005).

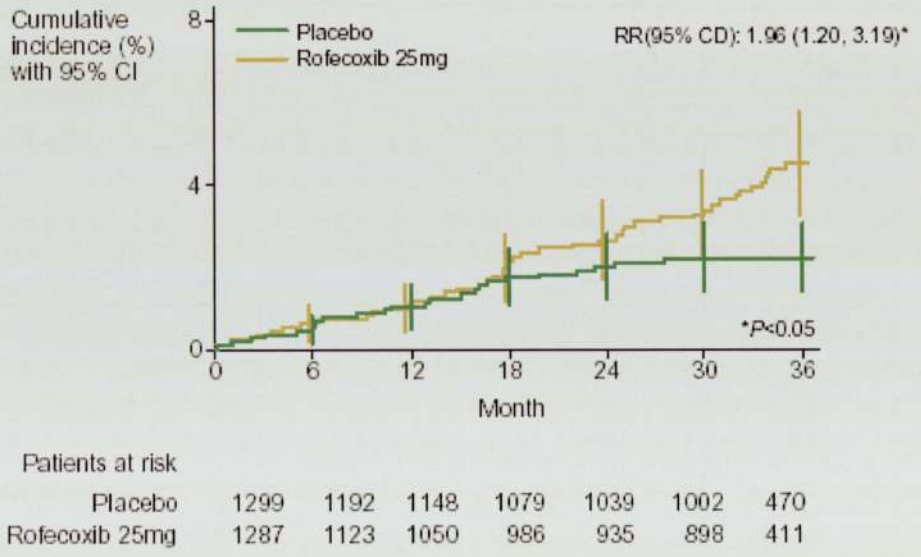


Figure 2.6 Confirmed cardiovascular events (MI and stroke) in the APPROVe trial, a randomised comparison of rofecoxib 25mg/day versus placebo in the chemoprevention of adenomatous polyp formation. Events were detected to diverge significantly between the groups after 18 months, although a numerical difference was noted as early as four months into the study (Bresalier, Sandler et al. 2005).

On the 30th of September 2004, Merck voluntary withdrew rofecoxib (VIOXX) from the market after the results of the APPROVe (Adenomatous Polyp Prevention On Vioxx) trial became available. In this placebo-controlled trial, a twofold-increased incidence of myocardial infarction (MI) and stroke in patients treated with rofecoxib versus placebo was detected after 18 months of rofecoxib therapy (Figure 2.6) (Bresalier, Sandler et al. 2005).

Rofecoxib, however, was not the only COX-2 specific inhibitor to have adverse cardiovascular effects. Meta-analysis of the two coronary artery bypass graft trials indicated a threefold elevation of the risk of MI and stroke for parecoxib / valdecoxib compared with placebo (Fitzgerald 2004). The Adenoma Prevention with Celecoxib (APC) trial, also a chemoprevention trial in patients with colonic adenomas, revealed a dose-dependent increase in the incidence of cardiovascular events with celecoxib (Solomon, McMurray et al. 2005).

Finally, hepatotoxicity, another major side-effect of all NSAIDs (the first NSAID to be withdrawn from the market (benoxaprofen) was for hepatoxicity and photosensitivity), appears still to be a significant adverse effect of coxibs (rofecoxib, lumiracoxib) (Schnitzer, Burmester et al. 2004; Rostom, Goldkind et al. 2005).

2.8 Need for evidence-based practice

In view of the well documented trends with increased individual and population ageing, the rise of chronic non-fatal conditions have important implications for society as a whole, particularly with respect to the future of heallth care. The impact of arthritic

conditions on public health and the significant costs that muskuloskeletal conditions generate will affect all societies in the future. NSAIDs, including aspirin, are now among the most widely prescribed medications in the world. Additionally, the increasing public awareness of health related issues and the increasing public expectations of quality of life is another reason emphasizing the need for evidence-based practise. The availability of newer agents, at generally considerably higher costs, although offering advantages in terms of favourable side-effects profiles and lower toxicity, may still have the potential for more serious (usually rare) adverse-effects (e.g. myocardial infarction and long-term rofecoxib use). With more threament options, but restricted budgets, along with new drugs offering marginal advantages, clinicians need to take into consideration any benefit, risk and cost of treatment aiming for optimal therapy.

Chapter 3

Evidence Based Pharmacotherapy & Systematic Reviews

3.1 Research Synthesis and Evidence Based Medicine

Research synthesis has a long history and has been developed in many spheres of scientific activity (Chalmers, Hedges et al. 2002). The rapid accumulation of medical and pharmaceutical information that need to be considered by healthcare professionals and researchers greatly complicates decision making at both the individual and the policy level (Petitti 1994). Reviews have become essential tools for anybody who wants to keep up to date with the new evidence that is accumulating in his or her field of interest (Egger, Davey Smith et al. 2001). However, reviews are also useful tools to identify areas where available evidence is insufficient and further studies are required (Egger, Davey Smith et al. 2001). Narrative reviews are unfortunately of poor quality and an unreliable source of information as pointed out by Milrow and Oxman & Guyatt (Mulrow 1987; Oxman and Guyatt 1988). Thus, there has been an increasing focus on formal methods of systematically reviewing studies, to produce explicitly formulated, reproducible, and up to date summaries of the effects of health care interventions.

3.2 EBM & EBP Definitions

"Evidence Based Medicine" (EBM) is the conscientious, explicit and judicious use of current best evidence in making decisions about the care of individual patients (Sackett, Rosenberg et al. 1996). The practice of evidence-based medicine means integrating individual clinical expertise with the best available external clinical evidence from systematic research (Sackett, Rosenberg et al. 1996). By individual clinical expertise we mean the proficiency and judgement that individual clinicians acquire through clinical experience and clinical practice (Sackett, Rosenberg et al. 1996). Increased expertise is reflected in many ways, but especially in more effective and efficient diagnosis and in the more thoughtful identification and compassionate use of individual patients' predicaments, rights, and preferences in making clinical decisions about their care (Sackett, Rosenberg et al. 1996). By best available external clinical evidence we mean clinically relevant research, often from the basic sciences of medicine, but especially from patient centred clinical research into the accuracy and precision of diagnostic tests (including the clinical examination), the power of prognostic markers, and the efficacy and safety of therapeutic, rehabilitative, and preventive regimens (Sackett, Rosenberg et al. 1996). External clinical evidence both invalidates previously accepted diagnostic tests and treatments and replaces them with new ones that are more powerful, more accurate, more efficacious, and safer (Sackett, Rosenberg et al. 1996).

"Evidence Based Pharmacotherapy" (EBP) which can be viewed as a subset of EBM has been described as the systematic, explicit and judicious use of best available evidence in making decisions about drug treatment for patients to ensure the most cost-

effective pharmacotherapy (Li Wan Po 1996). The importance of this area has been increasingly recognised as greater demands are placed on health-care resources and especially when the cost of novel pharmaceuticals is increasing dramatically. Pharmacists are called out to make formulary decisions based on their abilities to handle information and provide advice and their knowledge of pharmacotherapy. Thus, a systematic evidence-based approach is of outmost importance for prudent use of available resources.

3.2.1 EBM history

EBM is the process of systematically finding, appraising, and using contemporaneous research findings as the basis for clinical decisions (Rosenberg and Donald 1995). Although the term EBM was created in Canada by a group led by Dr. Gord Guyatt (McQueen 2001), there are various claims as to the origin of its practise. Paris in the 19th century has been suggested as the source of its philosophical origins, where clinicians like Pierre Lois rejected the pronouncements of authorities and sought the truth in systematic observation of patients (Strauss and McAlister 1999), with the 18th century staking a claim when Morgani in 1769 used the autopsy in the study of disease (Morgani 1769). In the 17th century Paris, it was noted that those who received bleeding as part of the treatment for cholera had a much higher death rate than those who were not bled. There is the suggestion of even earlier philosophical origins for the assessment of evidence in research during the reign of the Chinese emperor Qianlong (McQueen 2001). The method of 'kaozheng' (practising evidential research) was used in the interpretation of Confucian texts (McQueen 2001).

3.2.2 Driving forces leading to the development of EBM & EBP

Regardless of its origins, many factors have contributed over the past 30 years to drive the movement to EBM. Individual physicians are faced with >30,000 biomedical journals published annually and >17,000 new medical books each year (Lowe and Barnett 1994). In 1992, the ~20 English-language clinical journals dealing with adult internal medicine published >6,000 articles with abstracts; every day a physician would have to read at least 17 articles related to internal medicine alone to try to keep up to date (Haynes 1993). The overwhelming increase in medical knowledge and the inadequacy of traditional sources along with a gap between clinical research and actual practise consist the major reasons for the recent revival of EBM concepts. Lack of good evidence for many important practises as well as also practises that were taken for granted that were found to be ineffective also helped to convince physicians of the increasing need for EBM (Eddy 2005). Additionally, the global phenomenon of rising healthcare costs was also a fundamental driving force especially for EBP. The need utilise prudently available resources demanded evidence based decisions to be made. Finally, a better-educated public, which obtains information from electronic media, requires the best diagnostics and therapies without allowing disparity between diagnostic skills and clinical judgement. Thus, a deeper appreciation of the need for EBM & EBP guidelines to influence individual physician-patient decisions and pharmacist-patient advice arose (Eddy 2005).

3.2.3 Building blocks in EBM

EBM can be practised in any situation where there is doubt about an aspect of clinical diagnosis, prognosis, or management. There are four main steps involved in EBM: (i) formulation of a clear clinical question from a patient's problem, (ii) efficient searching of the literature for relevant clinical studies, (iii) critically appraising the identified quality evidence, and (iv) implementation of useful outcomes in clinical practise (Rosenberg and Donald 1995).

3.3 Systematic review and Meta-analysis

Traditional reviews of research or 'narrative reviews' summarise qualitatively the available studies and often deal with a broad range of issues related to a given topic rather than focusing on any particular question (Cook, Mulrow et al. 1997). All reviews, narrative and systematic alike, are retrospective, observational research studies and are therefore subject to systematic and random error (Cook 1997). Narrative reviews are subjective and lack rigorous scientific standards (Slavin 1995). Accordingly, the quality of a review depends on the extent to which scientific review methods have been used to minimise error and bias (Cook 1997). As a result, the systematic review/overview was introduced (Slavin 1995).

3.3.1 Systematic review and Meta-analysis definition

Systematic review is defined as a review that has been prepared using a systematic approach to minimising biases and random errors, which is documented in a

materials and methods section (Egger, Davey Smith et al. 2001). A systematic review may, or may not, include a meta-analysis: a statistical analysis of the results from independent studies of similar design eligible after inclusion and exclusion criteria have been defined and applied, which generally aims to produce a single estimate of the size of treatment effect and a test of homogeneity in the estimate of effect size (Egger, Davey Smith et al. 2001). Although it is always appropriate and desirable to have a concise summary of the best available evidence from primary studies using explicit, rigorous, and reproducible methods to identify, critically review, and then synthesise the evidence (Cook 1997; Greenhalgh 1997; Chalmers, Hedges et al. 2002), it may sometimes be inappropriate, or even misleading, to statistically pool results from separate studies (O'Rourke and Detsky 1989). The variability that is observed between the trials can confidently be attributed to random variation when the primary studies are carefully selected and the meta-analysis would provide an equally unbiased estimate of the treatment effect, with an increase in the precision of this estimate.

The systematic review process can be devided into six essential steps: (i) defining study objectives, (ii) defining relevant outcome measures, (iii) systematic retrieval of relevant studies, (iv) data collection, (v) summarising the evidence using statistical methods (if possible), and (vi) interpreting the results (Li Wan Po 1996).

Principally, primary and sub-objectives of the study need to be clearly defined and the questions posed should be answerable. Prior to define outcome measures, the results of available clinical trials have to be evaluated as to whether the outcomes used in assessing efficacy or safety are appropriate and valid. Instruments used for measuring clinical outcomes need to be validated in order for the results to be

meaningful. When defining outcome measures for chronic and recurrent conditions, it is important to look beyond the immediate clinical effects and attempt to evaluate the less tangible but perhaps more important aspects, such as the impact of treatment on the quality of life of those affected (Li Wan Po 1996). Evidence derived using non-validated instruments does not add much weight to the total evidence (Li Wan Po 1996). Although it is tempting to use surrogate markers, such as the use of endoscopically detected ulcers for assessing the gastrointestinal toxicity of NSAIDs and COX-2 inhibitors, extensive work is required to validate how well those surrogate measures predict the clinical outcomes of interest (i.e. perforations, ulcerations, and bleeding (PUBs)) before they can be accepted as valid evidence.

The search strategy for the identification of the relevant studies should be clearly delineated and be systematic and explicit with the aim to reduce bias and be reproducible. Retrieving studies requires expertise and unless a systematic approach is adopted many of the relevant studies may be missed (Li Wan Po 1996). Identification of published studies usually begins with a search of personal reference files and is followed be a computerised search of MEDLINE and of other computerised literature databases (e.g. EMBASE) (Petitti 1994). The title and abstract identified in the computerised search are scanned to exclude any that are clearly irrelevant (Petitti 1994). The full text of the remaining articles is retrieved, and each paper is read to determine whether it contains information on the researched topic (Petitti 1994). Numerous studies have now shown that even when a thorough computerised search is undertaken many relevant studies are missed because of poor indexing (Hetherington, Dickersin et al. 1989; Li Wan Po 1996). Thus, the electronic databases searching should be complemented by hand searching, follow-up of reference lists of articles

retrieved, searching licensing authorities websites (e.g. FDA site) & conference proceedings and writing to appropriate manufacturers and investigators known to have an interest in the drug involved (Li Wan Po 1996). Other sources unfortunately requiring a more laborious search include dissertations, trial registries, and books. In many cases, the full list of identified studies is submitted for review to a knowledgeable expert, who is asked to identify studies of the topic that have not been included on the list (Petitti 1994).

A set of inclusion and exclusion criteria must be determined for identifying the studies that are to be included in a meta-analysis. This is based on the specific hypotheses tested in the systematic review/meta-analysis and may include study design, population characteristics, intervention and outcomes (Cook, Sackett et al. 1995). Search methods and subsequent inclusion criteria may affect the results of a systematic review/meta-analysis (Cook, Sackett et al. 1995).

Study quality is also a major concern when conducting a systematic review/meta-analysis. Following the recommendations of the Cochrane Collaboration and other experts, many reviewers formally assess the quality of the primary trials (Egger, Davey Smith et al. 2001). However, the methodology for both the assessment of quality and its incorporation into systematic reviews are a matter of ongoing debate (Egger, Davey Smith et al. 2001). Although numerous (at least 25) scales and checklists have been constructed to assess quality of each study (Moher, Jadad et al. 1995; Jadad, Moore et al. 1996), the majority of them are inadequately developed (Moher, Jadad et al. 1995). The Consolidated Standards of Reporting Trials (CONSORT) statement is a continuously evolving instrument to assess the quality of

reported RCTs that was produced by a consensus meeting (Moher, Schulz et al. 2001a,b; Moher, Schulz et al. 2003).

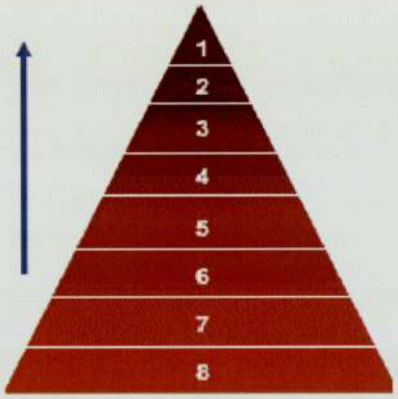
Important information regarding study design, study characteristics, study duration and study outcome need to be extracted after retrieval of all relevant studies. The use of data extraction forms is advisable. Additionally, it is recommended that two independent investigators (preferably blinded to aim of study, journal published and author names of reviewed articles) extract the required information are required in order to minimise human error and to increase reliability of data extraction (Petitti 1994). Finally, a meta-analysis should be performed only when the results of the studies were clinical homogeneous and heterogeneity can be ruled out. If the evidence are too sparse, too heterogeneous, or of low quality to proceed to a meta-analysis will not be beneficial and might lead to inappropriate outcomes. In this case, a systematic review is more appropriate.

A systematic review should clearly summarise the available evidence and provide appropriate interpretation. The degree of generalisability, and the strengths and weaknesses of the results explicitly outlined. Recommendations may be proposed based on the results and areas of future research need to be identified.

3.3.2 Hierarchy of evidence, RCTs & epidemiological studies

Randomised controlled trials (RCTs) are accepted as the gold standard of study design. They consist of 2 major components: (i) a control group, which makes sure that any outcome effects in the intervention group not due to the intervention can be

measured and adjusted for; (ii) random assignment of participants to the treatment and control groups, which ensures that bias is minimised, and potentially confounding variables are distributed evenly across the two groups. A third component oftenly considered is blinding, which makes sure that neither the participant nor the researcher knows which group the participant has been assigned to. Figure 3.1 illustrates the hierarchy of evidence. Thus, a meta-analysis of RCTs should yield an unbiased estimate of treatment effect.



1. Systematic reviews & meta-analysis
2. Randomised controlled trials (double blind)
3. Cohort Studies
4. Case-control Studies
5. Cross Sectional Studies
6. Case reports
7. Expert Opinion
8. Anecdotal

Figure 3.1 The hierarchy of evidence pyramid [The Ohio State University School of Allied Medical Professions 2005]

A fundamentally different situation arises in the case of epidemiological studies, for example case-control studies, cross-sectional studies or cohort studies. Cohort studies are observational studies of subjects with a specific disease or characteristic who are followed over a period (usually years) to detect complications or new events (Jones 2002). The group may be compared with a control group. Studies are generally concerned with what causes a disease or specific adverse drug reactions (ADRs) (Jones 2002). Case control studies share the same aim with cohort studies. Patients with a particular disease are matched with controls (people without the disease), but rather than following the subjects into the future, data on past exposure to possible causal

agents are collected (retrospectively) (Jones 2002). Case control are faster to perform but less reliable than cohort studies (Jones 2002). Cross sectional surveys is a measure of the frequency of a disease or risk factor in a defined population at a given time (Jones 2002). A case report describes the medical history of a single patient in the form of a story and a case series is a collection of similar reports, used to report or alert other healthcare professionals to rare occurrences (Jones 2002). Therefore, combining a set of epidemiological studies will thus often provide spuriously precise, but biased, estimates of association (Egger, Davey Smith et al. 2001).

It has to be noted though that RCTs are not immune to bias. Publication bias and other reporting bias may distort the evidence from both trials and observational studies (Egger, Davey Smith et al. 2001). Bias can be introduces if the methodological quality is inadequate (O'Rourke and Detsky 1989). To ensure that RCTs have been well designed, executed, and reported, the CONSORT statement was published in 1996 with a checklist of 21 items that should be included in reports or RCTs (Begg, Cho et al. 1996) and was revised in 2001 (Moher, Schulz et al. 2001a,b; Ioannidis, Evans et al. 2004).

3.3.3 Advantages of systematic reviews & meta-analyses

The major advantages of systematic reviews are (Greenhalgh 1997): (i) explicit methods limit bias in identifying and rejecting studies, (ii) conclusions are more reliable and accurate because of methods used, thus uncertainty can be resolved, (iii) large amounts of information can be assimilated quickly by healthcare providers, researchers, and policymakers forming a basis for appropriate use of healthcare

resources, (iv) the delay between research and implementation of effective diagnostic and therapeutic strategies may be reduced, (v) the results of different studies can be formally compared to establish generalisability of findings and consistency of results , (vi) reasons for heterogeneity can be identified and new hypotheses can be generated about particular subgroups, while also (vii) quantitative systematic reviews (meta-analyis) increase the power and precision of estimates of treatment effects or exposure risks without having to increase the number of patients enrolled in RCTs or contacting new RCTs.

3.3.3.1 Advantages of cumulative meta-analysis

Another advantage of conducting a meta-analysis is that it can aid extremely licensing organisations to make timely appropriate decisions. Cumulative meta-analysis can be defined as the repeated performance of meta-analysis whenever a new relevant trial is added to a series of trials (Lau, Schmid et al. 1995). One of the most significant merits of cumulative meta-analysis is that the contribution of individual studies to the cumulative pooled results can readily be determined (Lau, Schmid et al. 1995). The accumulation may proceed according to the year of completion or publication of each study, the event rate in the control group, the size of each study, the size of the difference between treatment and control groups in each study, some quality score that has been assigned to it each study, or other covariates such as drug dosages or time to treatment (Lau, Schmid et al. 1995). The results usually are presented in a graphic form as odds ratio with 95% confidence interval for the pooled data from all the trials (Figure 3.2) (McQueen 2001). Juni et al (2004) choice of a cumulative meta-analysis (Figures 3.3, 3.4) on the cardiovascular events risk reported for rofecoxib was

appropriate in order to clearly point out the earliest year at which rofecoxib cardiovascular events became statistically significant and that if the accruing data have been analysed cumulatively as soon as they became available, appropriate and timely decisions could have been taken by licensing authorities all over the world (Juni, Nartey et al. 2004).

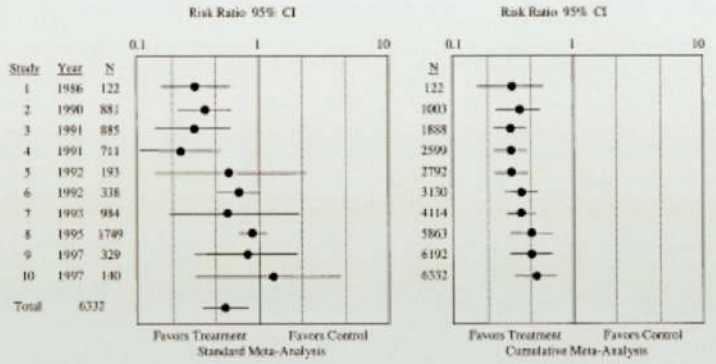


Figure 3.2 Standard meta-analysis plot of the risk ratios with their 95% confidence intervals (95% CI) for clinical studies, with the same data entered into a cumulative meta-analysis (McQueen 2001).

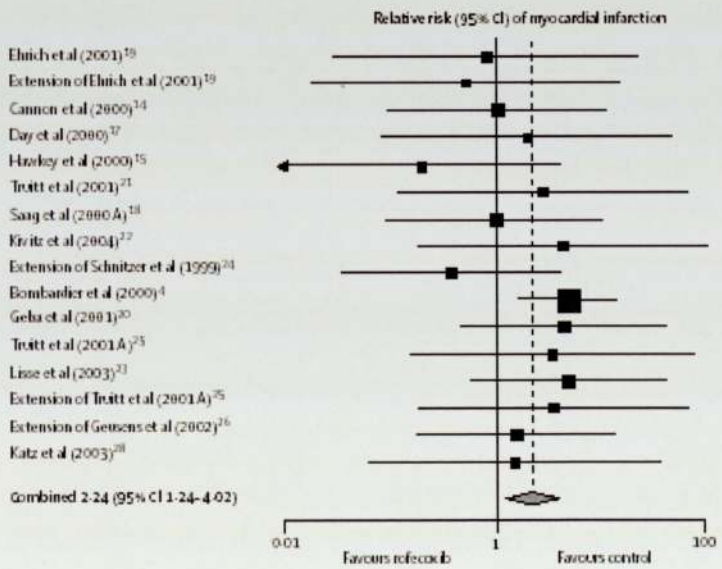


Figure 3.3 Juni's standard meta-analysis of RCTs comparing rofecoxib with control (Juni, Nartey et al. 2004)

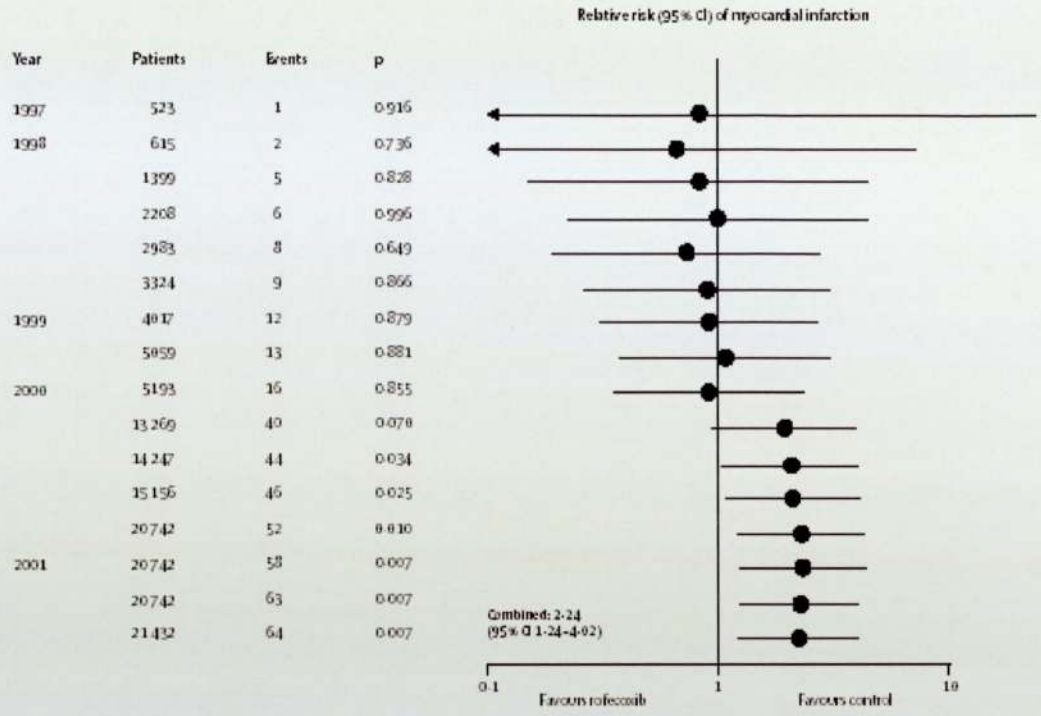


Figure 3.4 Juni's cumulative meta-analysis of RCTs comparing rofecoxib with control (Juni, Nartey et al. 2004)

3.3.4 Limitations of systematic reviews & meta-analyses

The most noticeable disadvantage is that to produce a high quality systematic review is time consuming and requires substantial resources. If the methodological quality of selected trials is inadequate, then the findings or those reviews can also be compromised. Publication bias can distort findings, because trials with statistical significant results are more likely to get published, and more likely to be published without delay, than trials without significant or negative results (Egger, Davey Smith et al. 2001). English published trials are more likely to be included which introduces language bias. Additionally, criteria for inclusion of studies into a review may be influenced by knowledge of the results of the set of potential studies (Egger, Davey

Smith et al. 2001). Finally, too strict eligibility criteria can limit the generalisability of the outcome and the potential for implementation of the outcome in clinical practise.

3.3.4.1 Publication bias in systematic reviews and meta-analysis

Publication bias derives from the selective publishing of studies with statistically significant or directionally positive results (Easterbrook, Berlin et al. 1991; Dickersin and Min 1993a; Dickersin and Min 1993b), and it can lead to inflated estimates of efficacy in meta-analysis (Moher, Cook et al. 1999). Publication bias occurs because published studies are representative of all studies that have ever been done (Petitti 1994). It has been proven that publication bias can yield significant results in favour of a therapy while when both published and those registered but unpublished studies are included the effect of the therapy was not supported (Klein, Simes et al. 1986). The problem of publication bias will be solved completely only when investigators submit and editors accept all well-conducted studies of important questions irrespective of the statistical significance of their studies (Petitti 1994). Until then, an attempt to retrieve all available evidence (published and unpublished) and the use of statistical to assess it or overcome publication may be of aid in reducing its deleterious effects.

Various approaches have been proposed for dealing with publication bias. An early method, the file-drawer method, was described by Rosenthal in 1979 (Rosenthal 1979). Rosenthal described a scenario where " the journals are filled with the 5% of the studies that show Type I errors, while the file drawers back at the lab are filled with the 95% of the studies that show nonsignificant (e.g. $p>0.05$) results (Rosenthal 1979;

Egger, Davey Smith et al. 2001). The method uses Z scores corresponding to the p-values from the individual trials included in a meta-analysis to calculate the number of unpublished nonsignificant studies that would be required to overturn the current pooled result (Rosenthal 1979). A modification of the file drawer method was produced by Klein et al (1986) (Klein, Simes et al. 1986) so that the OR scale, instead of the p-values, can be used.

Positive trials are also likely to be published more than once. Duplication of data is thus expected yielding excessively precise and inflated effect size estimates. Therefore, it is crucial to include only one report of the trial population outcomes (Berlin and Antman 1994; Naylor 1997). The inclusion of unpublished data could be of additional concerns as they may be methodologically flawed.

Including unpublished studies will be of aid in minimising publication bias. However, one cannot be certain that all such studies have been identified. Further, another problem that could arise is debate regarding the willingness of investigators to provide unpublished data (Egger and Smith 1998).

3.3.4.2 Citation Bias

The perusal of the reference lists of articles is widely used to identify additional articles that may be relevant (Egger, Davey Smith et al. 2001). The limitation of this approach is that the act of citing previous work is far from objective and retrieving literature by scanning reference lists may thus produce a biased sample of studies (Egger, Davey Smith et al. 2001). Gøetzsche et al (1999) illustrated that in an analysis

on NSAIDs in RA, trials demonstrating a superior effect of the new drug were more likely to be cited than trials with negative results (Gotzsche 1987; Egger, Davey Smith et al. 2001).

3.3.4.3 Influence of external funding and commercial interests

External funding was associated with publication independently of the statistical significance of the results (Egger, Davey Smith et al. 2001). Funding by government agencies was significantly associated with publication in three cohorts of proposals submitted to ethics committees (Easterbrook, Berlin et al. 1991; Dickersin, Min et al. 1992; Stern and Simes 1997) whereas pharmaceutical industry sponsored studies were less likely to be published in two studies (Easterbrook, Berlin et al. 1991; Dickersin, Min et al. 1992; Egger, Davey Smith et al. 2001). Indeed a large proportion of clinical trials submitted by drug companies to licensing authorities remain unpublished (Bardy 1998; Egger, Davey Smith et al. 2001). This is in agreement with a review of publications of clinical trials which separated them into those which were sponsored by the pharmaceutical industry and those supported by other means (Davidson 1986; Egger, Davey Smith et al. 2001). The results of 89% of published industry-supported trials favoured the new therapy, as compared to 61% of the other trials (Egger, Davey Smith et al. 2001). Similar results have been reported for NSAID trials and studies published in symposium proceedings (Cho and Bero 1996; Egger, Davey Smith et al. 2001). In a national survey of life-science faculty members in the United States, 20% of faculty members reported that they had experienced delays of more than six months in publication of their work and reasons for not publishing included "to delay the dissemination of undesired results" (Blumenthal, Campbell et al. 1997; Egger, Davey

Smith et al. 2001). Delays in publication were associated with involvement in commercialisation and academic-industry research relationship, as well as with male sex and higher academic rank of the investigator (Blumenthal, Campbell et al. 1997).

3.3.4.4 Language bias and Time lag bias

Along with publication bias, language bias can bias further the results of systematic reviews/meta-analysis by not including non-English (mostly) papers (Pham, Klassen et al. 2005). There is evidence that most systematic reviews/meta-analyses do not include all potential evidence (Moher, Fortin et al. 1996). It has been reported that 78% of the meta-analyses had language restrictions (Gregoire, Derderian et al. 1995) and most of these restrictions (93%) led to the exclusion of RCTs reported in languages other than English (Moher, Fortin et al. 1996). Moher et al provided evidence that non-English trials do not have significant differences with English trials regarding quality of reporting (Moher, Fortin et al. 1996). Language-restricted meta-analysis, as compared to language-inclusive meta-analysis, overestimated the treatment effect by only 2%, on average (Egger, Davey Smith et al. 2001). However, the language-inclusive meta-analysis were more precise (Moher, Pham et al. 2000; Egger, Davey Smith et al. 2001).

Additionally, the long median time between completion and publication can also affect the results and clinical practice as systematic reviews are considered the best available evidence (Egger, Davey Smith et al. 2001). However, this problem can be overcome by using cumulative meta-analysis and today by the appearance of purely electronic journals and 'ahead of publication' preview of the article.

3.3.4.5 Combinability of studies & heterogeneity

A meta-analysis attempts to gain greater objectivity, applicability and precision by including all the available evidence from randomised trials that pertain to the issue (Dickersin and Berlin 1992). Because of the broader aim of a meta-analysis, the included trials usually encompass a substantial variety of specific treatment regimens, types of patients, and outcomes (Egger, Davey Smith et al. 2001). Thus, the influence of this clinical heterogeneity needs to be explored carefully. Incompatibility in quantitative results is termed statistical heterogeneity (Egger, Davey Smith et al. 2001). It may be caused by known clinical or methodological differences between trials, or may be related to unknown or unrecorded trial characteristics (Egger, Davey Smith et al. 2001). The statistical question that needs to be answered is whether greater variation exists between the results of the trials than that caused by chance (Egger, Davey Smith et al. 2001). Homogeneity tests have low power and may fail to detect as statistically significant even at a moderate degree of genuine heterogeneity (Whitehead and Whitehead 1991). The usual test statistic (Cochran's Q) is computed by summing the squared deviations of each study's estimate from the overall meta-analytic estimate, weighing each study's contribution in the same manner in a meta-analysis (Higgins, Thompson et al. 2003). P-values are obtained by comparing the statistic with an x^2 distribution with k-1 degrees of freedom (where K is the number of studies) (Higgins, Thompson et al. 2003). Meta-analysis often include small number of trials and the power of the test in such circumstances is low (Higgins, Thompson et al. 2003). Thus, a non-significant test can never be interpreted as direct evidence in favour of the null hypothesis of homogeneity (Altman 1991), making of crucial importance to investigate the influences of the specific clinical differences between trials rather than rely on an

overall statistical test for heterogeneity (Egger, Davey Smith et al. 2001). If the heterogeneity of studies is properly handled, it may aid in the interpretation of the existing data and in planning future studies (Li Wan Po 1996).

3.4 QUOROM Guidelines

Like any research enterprise, particularly one that is observational, the meta-analysis of evidence can be flawed (Moher, Cook et al. 1999). Accordingly, the process by which meta-analysis are carried out has undergone scrutiny. A 1987 survey of 86 English-language meta-analyses (Sacks, Berrier et al. 1987) assessed each publication on 23 items from six content areas judged important in the conduct and reporting of a systematic review or meta-analysis of RCTs: study design, combinability, control of bias, statistical analysis (if applicable), sensitivity analysis, and problems of applicability. The survey results showed that only 24 (28%) of the 86 meta-analysis reported that all six content areas had been addressed (Moher, Cook et al. 1999). The updated survey, which included more recently published meta-analyses, showed little improvement in the rigour with which they were reported (Sacks, Reitman et al. 1996).

The number of published meta-analyses has increased substantially in the past decade (Chalmers and Haynes 1995). Figure 3.5 illustrates the sharp increase in the number of publications concerning systematic reviews or meta-analyses between 1986 to 1999. The increase in the number of systematic reviews or meta-analyses published has highlighted such issues as discordant systematic reviews or meta-analyses on the same topic (Jadad, Cook et al. 1997) and discordant systematic reviews/meta-analysis and randomised-trial results on the same question (LeLorier, Gregoire et al. 1997).

An important consideration in interpretation and use of meta-analyses not only report explicitly the methods they used to analyse the articles they reviewed, but also report the methods used in the research articles they analysed (Moher, Cook et al. 1999). The meta-analytical review methods used may not be provided when a paper is initially submitted; even when they are, other factors such as page limitations, peer review, and editorial decisions may change the content and format of the report before publication (Moher, Cook et al. 1999).

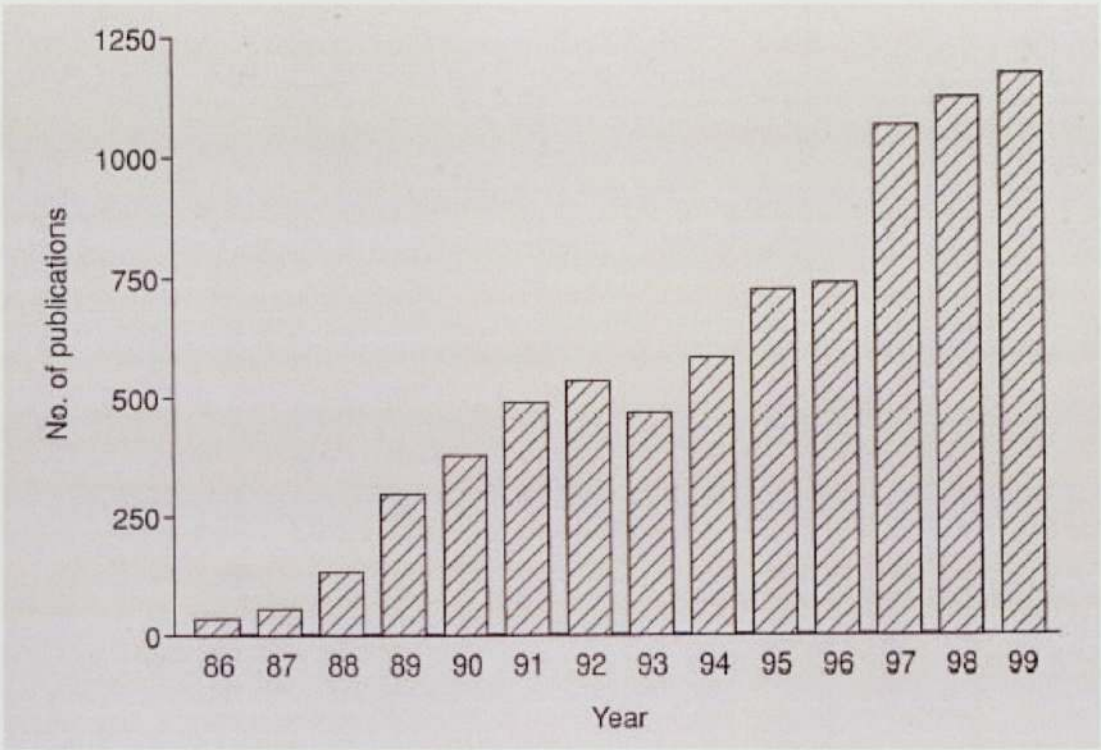


Figure 3.5 Number of publications concerning meta-analyses, 1986-1999. Results from MEDLINE search using text word and medical subject (MESH) heading "meta-analysis' and text word "systematic review" (Egger, Davey Smith et al. 2001)

Several investigators have suggested guidelines for reporting of meta-analyses (Cook, Sackett et al. 1995; Shea, Dube et al. 2001). However, a consensus across disciplines had not be developed before the Quality if Reporting of Meta-analyses (QUOROM) conference taken place (Moher, Cook et al. 1999). The QUOROM

Table 3.1 QUOROM checklist for quality reporting of systematic review or meta-analysis (Moher, Cook et al. 1999; Hemels, Vicente et al. 2004)

Heading	Subheading	Descriptor	Reported? (Y/N)	Page number
Title		Identify the report as a meta-analysis [or systematic review] of RCTs		
Abstract		Use a structured format		
		Describe		
	Objectives	The clinical question explicitly		
	Data sources	The databases (ie, list) and other information sources		
	Review methods	The selection criteria (ie, population, intervention, outcome, and study design); methods for validity assessment, data abstraction, and study characteristics, and quantitative data synthesis in sufficient detail to permit replication		
	Results	Characteristics of the RCTs included and excluded; qualitative and quantitative findings (ie, point estimates and confidence intervals); and subgroup analyses		
	Conclusion	The main results		
		Describe		
Introduction		The explicit clinical problem, biological rationale for the intervention, and rationale for review		
Methods	Searching	The information sources, in detail (eg, databases, registers, personal files, expert informants, agencies, hand-searching), and any restrictions (years considered, publication status, language of publication)		
	Selection	The inclusion and exclusion criteria (defining population, intervention, principal outcomes, and study design		
	Validity assessment	The criteria and process used (eg, masked conditions, quality assessment, and their findings)		
	Data abstraction	The process or processes used (eg, completed independently, in duplicate)		
	Study characteristics	The type of study design, participants' characteristics, details of intervention, outcome definitions, &c. and how clinical heterogeneity was assessed		
	Quantitative data synthesis	The principal measures of effect (eg, relative risk), method of combining results (statistical testing and confidence intervals), handling of missing data; how statistical heterogeneity was assessed; a rationale for any a-priori sensitivity and subgroup analyses; and any assessment of publication bias		
Results	Trial flow	Provide a meta-analysis profile summarising trial flow (see figure)		
	Study characteristics	Present descriptive data for each trial (eg, age, sample size, intervention, dose, duration, follow-up period)		
	Quantitative data synthesis	Report agreement on the selection and validity assessment; present simple summary results (for each treatment group in each trial, for each primary outcome); present data needed to calculate effect sizes and confidence intervals in intention-to-treat analyses (eg 2×2 tables of counts, means and SDs, proportions)		
Discussion		Summarise key findings; discuss clinical inferences based on internal and external validity; interpret the results in light of the totality of available evidence; describe potential biases in the review process (eg, publication bias); and suggest a future research agenda		

conference participants were clinical epidemiologists, clinicians, statisticians, and researchers who contact meta-analyses as well as editors from U. K. and North America who were interested in systematic reviews (Egger, Davey Smith et al. 2001). The QUOROM statement consists of a checklist (Table 3.1) and a flow diagram (Figure 3.6). The checklist of standards for reporting of meta-analysis describes a way to present the abstract, introduction, methods, results, and discussion sections of a report of a meta-analysis (Moher, Cook et al. 1999). The checklist is organised into 21 headings and subheadings to encourage authors to provide readers with information on searches, selection, validity assessment, data abstraction, study characteristics, quantitative data synthesis, and trial flow (Moher, Cook et al. 1999). Authors are asked

to provide a flow diagram (Figure 3.6) providing information about the number of RCTs identified, included and excluded and the reasons for excluding them (Moher, Cook et al. 1999).

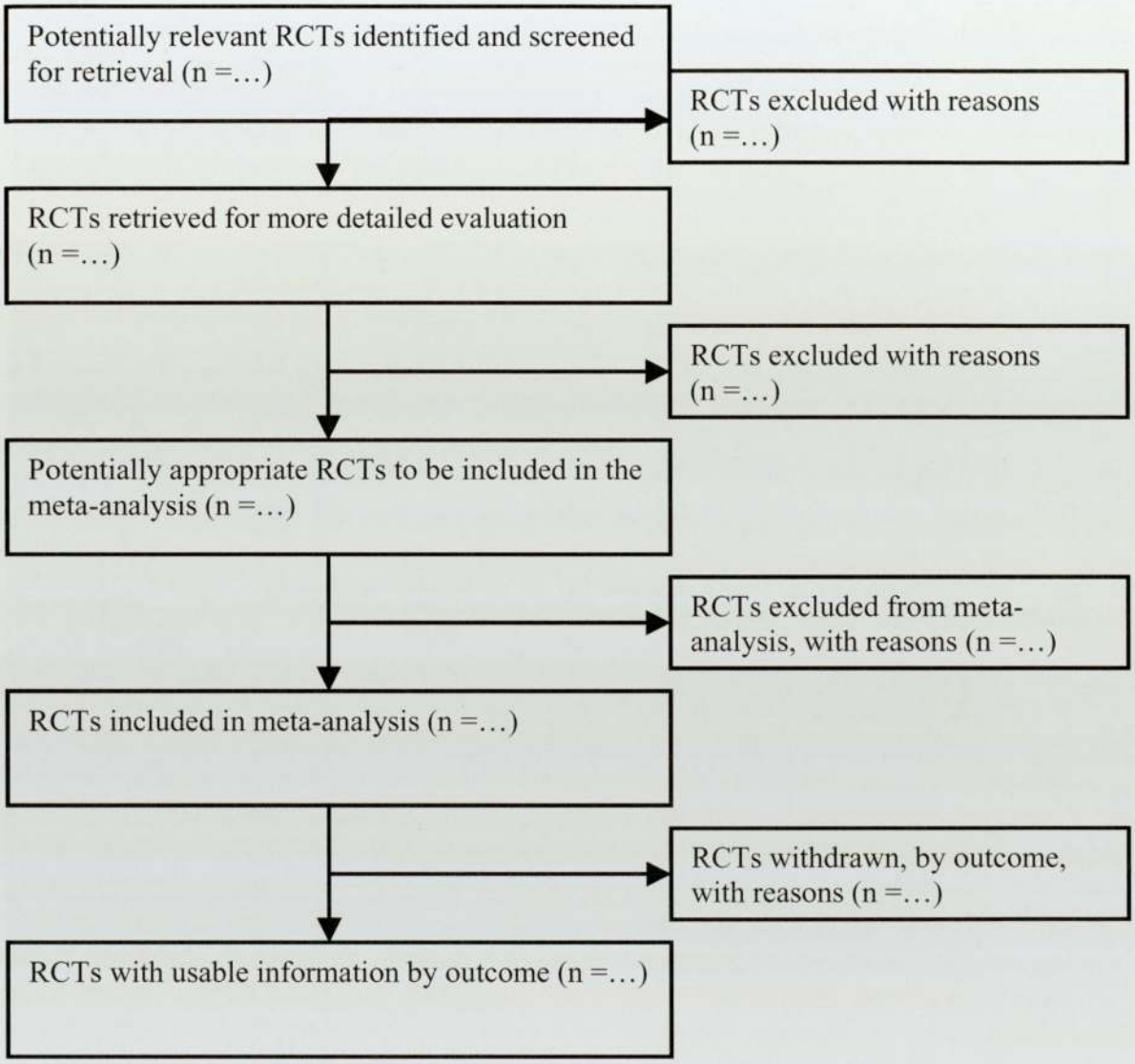


Figure 3.6 Progress through the stages of a meta-analysis for RCTs (Moher, Cook et al. 1999)

In developing the checklist, supporting scientific evidence for only eight of 18 items were identified to guide the reporting of meta-analysis of RCTs (Moher, Cook et al. 1999), which implies the need to include items that can systematically influence

estimates of treatment effects (Egger, Davey Smith et al. 2001). Some of this evidence is indirect (Moher, Cook et al. 1999). The evidence for the use of a structured abstract format, for example, were obtained by examining abstracts of original reports of individual studies (Taddio, Pain et al. 1994) and may not pertain specifically to the reporting of meta-analysis (Moher, Cook et al. 1999).

The impact of QUOROM on the editorial process had been assessed in an RCT involving eight medical journals to assess the impact of the use of QUOROM criteria on journal peer review (Moher, Cook et al. 1999). Accrual is now complete, but the results have not yet been published (Moher, Cook et al. 1999).

A systematic review to identify an inventory of published checklists and scales identified 24 instruments of which 21 were checklists and three scales (Table 3.2) (Light and Pillemer 1984; Goldschmidt 1986; L'Abbe, Detsky et al. 1987; Mullen and Ramirez 1987; Mulrow 1987; Sacks, Berrier et al. 1987; Oxman and Guyatt 1988; Meinert 1989; Smith and Stullenbarger 1989; Wilson and Henry 1992; Neely 1993; Ohlsson 1994; Oxman 1994; Taylor Halvorsen 1994; Assendelft, Koes et al. 1995; Cook, Sackett et al. 1995; Nony, Cucherat et al. 1995; Geller and Proschan 1996; Thacker, Peterson et al. 1996; Auperin, Pignon et al. 1997; Greenhalgh 1997; Pogue and Yusuf 1998; Blettner, Sauerbrei et al. 1999; Egger, Davey Smith et al. 2001). All the instruments were published except the scale proposed by Oxman et al., and can be used with all types of systematic reviews. The number of items in each instrument ranged from 5 to 101, with only two checklists having more than 35 items (Table 3.3) (Egger, Davey Smith et al. 2001). The average time required to assess the quality of a

systematic review using the checklists and the scales was 12 minutes (Table 3.3) (Egger, Davey Smith et al. 2001).

Table 3.2 Number of criteria reported by each checklist and scale (first author named) fullfilling the 17* headings and subheadings included in the QUOROM statement (Egger, Davey Smith et al. 2001)

Table 7.2 Number of criteria reported by each checklist and scale (first author named) fulfilling the 17* headings and subheadings included in the QUOROM statement.[†]

Instruments‡‡	Title	Abstract					Introduction	Method						Results			Discussion
		Objectives	Data sources	Review methods	Results	Conclusion		Searching	Selection	Validity assessment	Data abstraction	Description of study characteristics	Quantitative data synthesis	Trial flow	Description of study characteristics	Quantitative data synthesis	
Checklist																	
Bleetner	NO	NO	NO	NO	NO	NO	YES	YES	YES	NO	YES	YES	YES	NO	YES	YES	NO
Cook	NO	NO	NO	NO	NO	NO	YES	YES	YES	YES	YES	NO	YES	NO	YES	YES	YES
Geller	NO	NO	NO	NO	NO	NO	NO	YES	YES	YES	NO	NO	YES	NO	NO	YES	YES
Goldschmidt	NO	NO	NO	NO	NO	NO	YES	YES	YES	YES	YES	NO	YES	NO	NO	YES	YES
Greenhalgh	NO	NO	NO	NO	NO	NO	NO	YES	NO	YES	NO	NO	NO	NO	YES	NO	YES
Halvorsen	NO	NO	NO	NO	NO	NO	NO	YES	YES	NO	NO	YES	YES	NO	NO	NO	NO
L'Abbé	NO	NO	NO	NO	NO	NO	UN	YES	YES	YES	YES	YES	YES	NO	NO	UN	YES
Ligit	NO	NO	NO	NO	NO	NO	YES	NO	YES	NO	NO	YES	NO	NO	NO	NO	UN
Meinert	YES	YES	YES	YES	NO	YES	NO	YES	YES	NO	NO	YES	YES	NO	YES	YES	YES
Mullen	NO	NO	NO	NO	NO	NO	UN	YES	YES	YES	YES	NO	YES	NO	UN	YES	YES
Mulrow	NO	NO	NO	NO	NO	NO	YES	YES	YES	YES	NO	NO	NO	NO	YES	YES	UN
Neely	NO	YES	YES	UN	UN	YES	YES	YES	YES	YES	NO	UN	UN	NO	UN	UN	YES
Nony	NO	NO	NO	NO	NO	NO	YES	YES	YES	YES	YES	YES	YES	NO	YES	YES	YES
Ohlsson	NO	NO	NO	NO	NO	NO	YES	YES	YES	YES	YES	NO	YES	NO	YES	YES	UN
Oxman	NO	NO	NO	NO	NO	NO	NO	YES	YES	YES	NO	NO	NO	NO	NO	NO	YES
Oxman	NO	NO	NO	NO	NO	NO	NO	YES	YES	YES	YES	UN	UN	NO	YES	YES	NO
Pogue	NO	NO	NO	NO	NO	NO	YES	YES	YES	NO	YES	YES	YES	NO	YES	YES	NO
Sacks	NO	NO	NO	NO	NO	NO	NO	YES	YES	YES	YES	YES	YES	NO	YES	YES	YES
Smith	NO	NO	NO	NO	NO	NO	YES	NO	YES	YES	NO	YES	NO	NO	UN	NO	UN
Thacker	NO	NO	NO	NO	NO	NO	NO	YES	YES	YES	YES	UN	UN	NO	UN	UN	YES
Wilson	NO	NO	NO	NO	NO	NO	YES	YES	YES	YES	YES	UN	YES	NO	YES	YES	NO
Scale																	
Assendelft[46]	NO	NO	NO	NO	NO	NO	NO	YES	YES	YES	NO	YES	NO	NO	YES	YES	YES
Auperin[45]	NO	NO	NO	NO	NO	NO	NO	YES	YES	YES	YES	YES	YES	NO	UN	YES	YES
Oxman[44]	NO	NO	NO	NO	NO	NO	NO	YES	YES	YES	YES	NO	YES	NO	NO	YES	YES

* The QUORUM statement includes 18 items. One item, under the Abstract heading, "Use a structured format", was not included here as we did not evaluate the presence of this item in the other instruments.
† When items in an instrument were unclear we reviewed the complete text of the article for possible clarification.
‡ For an instrument to be included, the report had to provide a summary form, such as a checklist, of the items discussed in the text of the article.
UN = uncertain as to whether the item was reported.

None of the above checklists included all the items recommended by QUOROM (Table 3.2) (Egger, Davey Smith et al. 2001). The majority of checklists contained items about what the method section of a systematic review should include and neglected generally the other components of the report. There was considerable overlap between the content of the QUOROM checklist and the method section of the other checklists (Egger, Davey Smith et al. 2001). All but two checklists asked about

Table 3.3 Descriptive characteristics of published and unpublished checklists and scales used to assess the quality of systematic reviews or meta-analyses of RCTs (Egger, Davey Smith et al. 2001)

Instrument	Number of items	Type of quality assessed	Explicit statement regarding the purpose of tool	Time to complete*
Checklist				
Blettner	12	General	Yes	15
Cook	65	General	No	30
Geller	12	General	Yes	20
Goldschmidt	101	General	Yes	30
Greenhalgh	5	General	No	5
Halvorsen	8	General	No	5
L'Abbé	9†	General	Yes	5
Light	10	General	Yes	5
Meinert	35	General	Yes	15
Mullen	12	General	Yes	10
Mulrow	8	General	Yes	5
Neely	5†	General	No	10
Nony	30	General	No	20
Ohlsson	26	General	No	15
Oxman	11	General	Yes	5
Oxman	8	General	Yes	5
Pogue	20	General	Yes	10
Sacks	23	General	Yes	20
Smith	12	General	No	5
Thacker	15	Specific	Yes	15
Wilson	10	General	Yes	10
Scale				
Assendelft	14	Specific	No	10
Auperin	27	General	No	20
Oxman‡	9	General	Yes	5

* Approximate time which may vary depending on the operator.
† There are several sub categories within each of the questions.
‡ Unpublished.

the searching criteria and all but one asked about the selection criteria (Egger, Davey Smith et al. 2001). Sixteen included an item on validity and twelve asked about the data abstraction (Egger, Davey Smith et al. 2001). Items concerning the results and discussion sections in the QUOROM statement were definitely reported in 57% of the checklists, respectively, with the exception of the flow diagram (figure 3. 6), which was not included in any of the checklists (Egger, Davey Smith et al. 2001). The face diagram provides some face validity for the reader regarding the process used by the

authors to include studies throughout the review process (Egger, Davey Smith et al. 2001). Similar results were identified for the scales (Egger, Davey Smith et al. 2001).

From the above, it can be easily apprehended why the QUOROM statement is considered today the gold standard for the reporting of systematic reviews or meta-analyses. Additionally, its widespread acceptance allows for comparisons to be made across different systematic reviews or meta-analyses under the same or similar aim, avoiding the variability with various instruments. The use of evidence based criteria, as the QUOROM and CONSORT statement may help to improve the quality of systematic reviews or meta-analyses and RCTs, respectively.

Systematic reviews and meta-analyses are increasingly common, and when properly conducted the best estimates of treatment effect based on all available evidence are elicited. Therefore, they are valuable tools for clinical decision-making and for the production of evidence-based guidelines and policies. However, as they also possess limitations and in some occasions can be misleading, careful and critical interpretation of systematic reviews or meta-analyses may strengthen the link between best research evidence and optimal patient care.

3.5 Aims and Objectives

The aim of this study was to elucidate the reasons why available systematic reviews or meta-analyses of RCTs were unable to identify earlier the cardiovascular and renal risk (MI, stroke, hypertension, oedema, congestive heart failure, death) posed by rofecoxib treatment.

To achieve this, the study has involved a systematic review of the evidence available leading to rofecoxib's withdrawal. The objectives were:

(i) to undertake a quality assessment of the available cardiovascular safety aimed systematic reviews or meta-analyses,

(ii) to identify the methods of information retrieval and data extraction utilised in the included systematic reviews or meta-analyses

(iii) to identify the duration of RCTs included in the systematic reviews or meta-analyses as an indication of exposure to rofecoxib,

(iv) to identify all possible indications and comparators included in the systematic reviews or meta-analyses

(v) to summarise and outline the objectives, inclusion and exclusion criteria, total number of patients and design of the RCTs analysed in the included systematic reviews or meta-analyses,

(vi) to describe the population characteristics of the population of the included systematic reviews or meta-analyses and

(vii) to identify the sources of funding of the included systematic reviews or meta-analyses.

Chapter 4

Methodology

4.1 Methodology

4.1.1 Retrieval of systematic reviews and meta-analyses

Endnote 7.00 software was used to identify published systematic reviews and meta-analyses of rofecoxib included in *PubMed* irrespective of the study aim. To identify rofecoxib systematic reviews, the following search pattern was employed using medical subject headings (MESH)s and the Boolean operator 'or' and 'and' to combine terms or to increase the sensitivity and specificity of the terms:

1. Meta-analysis / Meta-analyses / Metaanalysis / Metaanalyses **or**
2. Systematic Review / systematic overview / methodologic review / methodologic overview **or**
3. Integrative research review / research integration **or**
4. Review / overview
5. Quantitative syntheses / quantitative synthesis **and**
6. Rofecoxib / Vioxx / MK-0966 / MK 0966 / *(4-[4-(Methylsulfonyl)phenyl]-3-phenylfuran-2(5H)-one)*.

Systematic searching of the following databases was also performed: Cochrane Library, Pubmed, Scirus, Food and Drug Administration (FDA) website. Additionally, websites of the National Institute of Clinical Excellence (NICE), the Canadian Co-ordinating Office for Health Technology Assessment (CCOHTA), the European League Against Rheumatism (EULAR) were exhaustively searched.

The resulting set of citations were hand searched by title, MESH, and whenever available abstract. When the study abstract had no clear reason for exclusion, the full article was obtained. Every citation that may have contained data or original information about rofecoxib's cardiovascular, cerebrovscular and renal adverse-effects was obtained for evaluation. The references given in review articles about rofecoxib and the references given in articles obtained for information were further screened for any additional citations that might identify further systematic reviews referring to rofecoxib's cardiovascular, cerebrovascular and renal adverse-effects. In addition the manufacturer's website was screened for information.

All databases were searched from the years 1966 till December 2004. A further search of the databases was performed in May 2005 to retrieve any recent publications, with the final search being conducted on the first of September 2005.

4.1.2 Inclusion & Exclusion criteria for systematic review selection

Systematic reviews or meta-analyses of an adult population were included that received any dose of rofecoxib for any stated duration for any indication versus any comparator including preferential and specific COX-2 inhibitors. For a systematic

review or meta-analysis to be included in this study, it had to be a systematic review or meta-analysis of randomised controlled trials (RCTs). The search was not restricted to the year of publication, but was restricted to the English language only. Publications cited as abstracts were only included if no corresponding full publications could be identified. Other abstracts and short reports were discarded, because they provided insufficient consistent information for meaningful interpretation. Only systematic reviews / meta-analyses including cardiovascular, cerebrovascular and renal adverse-effects were included for analysis.

Table 4.1 Inclusion & Exclusion criteria

Criteria parameter	Definition
Inclusion	
Study type	Systematic review / Meta-analysis of RCTs (integrating the results of >1 RCT of rofecoxib)
Publication date	Between 1966 and August 2005
Language	English
Patients	Adults (>18 years of age)
Indication / Disease	Any (that involves long-term exposure)
Treatment	Rofecoxib (any dose) only versus any comparator
Comparators	Placebo or active drug(s) (tNSAIDs or selective/specific COX-2 inhibitors regardless of the dose)
Exclusion	
Indication / Disease	Acute pain
Outcomes reported	Efficacy only or gastrointestinal or other safety endpoints apart from cardiovascular, cerebrovascular and renal.
Duplicates	Only updated systematic reviews / meta-analyses included
Publication	Abstracts, short reports
Treatment	Systematic review / Meta-analysis of RCTs of other COX-2 selective or specific inhibitors

4.1.3 Data extraction

Data extraction was undertaken by one independent reviewer (myself) and cross-validated by my supervisor (ALWP/ KW). Any ambiguity in data interpretation identified by us would be discussed to reach consensus. A customised spreadsheet was used to record: (i) name of first author, (ii) the year of publication, (iii) aim of systematic review / meta-analysis (primary and secondary outcomes i.e. safety, mortality), (iv) quality of reporting in systematic reviews / meta-analyses of RCTs that reported cardiovascular/cerebrovascular/renal adverse-effects of rofecoxib versus a comparator (active or placebo), (v) methods of information retrieval (e.g. databases searched), (vi) methods of data exctraction, (vii) inclusion and exclusion criteria utilised, (viii) number of RCTs included, (ix) study design of included RCTs (double-blind or single blind, cross-over or parallel), (x) indication [osteoarthritis, rheumatoid arthritis, chronic back pain, Alzheimer's disease, cancer (e.g. familiar adenomatous polyposis)], (xi) posology, (xii) population characteristics (age, sample size, ethnicity) when available, (xiii) total exposure to rofecoxib and comparators (duration of included RCTs), while noting possible omissions or errors. The total number of manufacturer's funded systematic reviews / meta-analyses was identified.

4.1.4 Derived outcome measures and statistical analysis

4.1.4.1 Quality scoring

All published articles included in the analysis were assessed for quality using the QUOROM checklist by two independent reviewers (myself and my supervisor [ALWP/KW]) (Moher, Cook et al. 1999; Moher, Cook et al. 2000).

A detailed description of this checklist is available in the literature (Moher, Cook et al. 2000) with a summary provided in Table 4.2 (Hemels, Vicente et al. 2004). The QUOROM is an 18-item checklist, which requires the reviewer to answer yes or no to each item and score each item with 1 point if the answer is yes and 0 points if the answer is no. Therefore, the highest possible score is 18. If a certain question was not applicable (N/A) for an article, the total score (in %) for that specific article was adjusted by the total number of questions that were applicable. Thus, for the two included abstracts the maximum score they could achieve was 7 and the overall quality scores are provided.

The items are separated into 6 categories, with each category representing a section within the article being evaluated. The categories (items per category) include Title (1), Abstract (6), Introduction (1), Methods (6), Results (3), and Discussion (1). It was decided that descriptors would assist in the judging of quality. Thus, the scores were devided into quartiles as follows: (a) <25% i.e. very poor, (b) 25-49% i.e. poor, (c) 50-75% i.e. acceptable and (d) >75% i.e. good quality.

Table 4.2 QUOROM checklist for quality of reporting in meta-analysis (Hemels, Vicente et al. 2004)

Category	Description of item
Title	• Have the authors identified the article as a meta-analysis of RCTs in the title?
Abstract	• Have the authors used a structured format in the abstract?
	• Has the objective/clinical question been explicitly described?
	• Have the databases and other sources been listed?
	• Has the selection criteria; methods for validity assessment, data abstraction and study characteristics, and quantitative data synthesis been described sufficiently to permit replication?
	• Have the characteristics of all RCTs, both included and excluded, been described with all qualitative and quantitative findings (i.e. point estimates and CIs), and subgroup analyses?
	• Have the main results been reported?
Introduction	• Has the clinical problem and rationale for review been described?
Methods	• Have the authors described their searching methods, information sources, e.g. databases, registers, personal files etc., including any restrictions (years, publication status, language of publication)?
	• Have the authors included the selection criteria for inclusion and exclusion?
	• Is the criteria and process used for validity assessment (e.g. masked conditions, quality assessment, and findings) described?
	• Are the processes for data abstraction described (i.e., completed independently or in duplicate?
	• Have the authors described the type of study design, participants' characteristics, details of intervention, outcome definitions, and how clinical heterogeneity was assessed?
	• Has the principle measure of effect (e.g. relative risk), method of combining results (test with CIs), handling of missing data, been described; including how statistical heterogeneity was assessed including a rationale for any *a priori* sensitivity and subgroup analyses, and any assessment of publication bias?
Results	• Have the authors provided a meta-analysis profile summarizing trial flow-chart?
	• Are descriptive data for each trial presented (e.g. age, sample size, etc.)?
	• Have the authors reported agreement on selection and validity assessment, presented sample summary results for each treatment group, presented data to calculate effect sizes and CIs in intention-to-treat analyses (e.g., 2 ×2 tables of counts, means and SDs)?
Discussion	• Have the authors summarized key findings, discussed clinical inferences based on internal and external validity, interpreted results in light of totality of available evidence, and described potential biases in the review process and suggested a future research agenda?

The mean quality score percentage for each category and item of the QUOROM scale was calculated using MINITAB^{TM} 14. Finally, each category was described based on these mean quality percentage scores as "very poor", "poor", "acceptable" and "good" as described above for each systematic review or meta-analysis. It has to be noted that the systematic review performed by the National Institute of Clinical Excellence (NICE) (NICE 2001) and the Food and Drug Administration (FDA) (FDA) meta-analyses included in the analysis will be excluded from the quality scoring analysis as they are not published systematic reviews or meta-analyses in peer review journals, and thus the QUOROM guidelines are not directly applicable to these.

4.1.4.2 Methods of Information Retrieval utilised

For a systematic retrieval of all available RCTs to be included in a systematic review / meta-analysis, a thorough search strategy of probably more than one databases is required, which is normally supplemented also with hand searching of obtained articles. Thus, it is of importance to identify in the included systematic reviews and meta-analyses of rofecoxib how the information that was reported had been identified i.e. which information sources were used.

To achieve this, all the included systematic reviews and meta-analyses were thoroughly screened - mainly the methods section, although care was taken not to omit information reported elsewhere in the published articles. The major sources of information such as the Medline, Embase, CCTR, the Cochrane Database of systematic reviews, and the FDA were classified separately as well as screening of reference lists in articles identified and/or in literature or systematic reviews. Finally, any attempts made by the authors of the systematic reviews or meta-analyses to identify 'fugitive literature' (all levels of government, academic, industry information in print and electronic format, but which is not found in databases of scientific literature e.g. government documents, theses, conference proceedings, books, internet sites, preprints) and any other sources were grouped together under 'Other' including occasionally the manufacturer's data files, as they are not freely available normally.

The end-date that the authors reported as the last date that they performed their search for retrieving RCTs was also noted to justify for any omitted RCTs from

analysis. If the authors performed an updated search on a later date, the end-date reported is the date of the last updated search.

4.1.4.3 Methods of Data Extraction utilised

The number of reviewers (independent or not), that reviewed the available RCTs on which the included systematic reviews or meta-analyses were based, was identified (if stated) by screening through the abstract and method section. Blinding of the reviewers or double-checking of the outcomes of the reviewers by different reviewers was also identified. Finally, the method that was used (if any or if stated) to resolve any discrepancies was also identified by searching the abstract and the methods section of the articles.

4.1.4.4 Duration of RCTs included in the analysed systematic reviews

The duration of the RCTs included in the analysed systematic reviews or meta-analyses is an indication of the exposure to rofecoxib, which is of importance when considering long-term adverse-effects. More specifically, an attempt was made to summarise the shorter and longer duration of exposure, by scanning the published articles for the available information. Also, if a specified cut-off duration was a reason of exclusion of RCTs in the systematic reviews or meta-analyses, this cut-off duration was noted separately for each included systematic reviews or meta-analyses. The median duration was calculated using MINITAB^{TM} 14 when possible and the upper and lower quartiles were reported. Based on these median duration results (only those that

could be calculated) the median duration of exposure to rofecoxib for all included systematic reviews /meta-analyses was calculated.

4.1.4.5 Indication of RCTs included in the analysed systematic reviews

The indication for which rofecoxib was used in the included RCTs was also taken into account, in an attempt to identify whether the systematic reviews and meta-analyses performed focused in all indications or only to a minority of them. Thus, all included systematic reviews and meta-analyses were screened to identify the indications of the RCTs that they included in their analysis. Long-term indications of rofecoxib include OA, RA and chronic pain. Although there are available RCTs for Alzheimer's disease and cancer, other long-term indications for rofecoxib, rofecoxib never received license for the prevention and treatment of Alzheimer's disease or prevention of various cancer types.

4.1.4.6 Rofecoxib's Comparators studied in the included systematic reviews

All included systematic reviews or meta-analyses were screened to identify the comparators that were used in the included RCTs. Comparators (active or placebo) were summarised along with their dosing regime [dose in miligrams (mg) and number of administrations per day i.e. OD: once a day, BD: twice daily, and TDS: three times a day]. The number of systematic reviews or meta-analyses including rofecoxib versus placebo controlled clinical trials as well as the variety of NSAIDs comparators utilised is of importance, as cardiovascular, cerebrovascular and renal adverse-effects outcomes stated in the included systematic reviews or meta-analyses can only be meaningful

when rofecoxib is compared with relevant comparators, placebo or active drugs administered in clinical practise standard doses.

4.1.4.7 Objectives, Inclusion & Exclusion Criteria and Design of included RCTs of the included systematic reviews and meta-analyses

An attempt was made to summarise in a table format the objectives / aims (primary and secondary) for all the included systematic reviews and meta-analyses, along with the inclusion and exclusion criteria utilised. For every systematic review or meta-analysis the included RCTs were summarised in detail stating the patient number, the design, the indication, the drug(s) analysed and their comparators, the duration of the RCTs in weeks and finally whether these included RCTs were given a quality score (or also a validity score). In this table, the design of the trial (i.e. whether the clinical trial, was randomised (R), whether it was double-blind (DB) or blind (B), or of parallel design (P), or single centre (SC) or multi-centre (MC) was recorded when the information was available. Quality scoring of the individual RCTs included in the systematic reviews and meta-analyses analysed was only recorded whenever available, and a note was made in the key section to identify the scale utilised. Furthermore, an attempt was made to identify patient numbers for all the included systematic reviews or meta-analyses in total as well as separately for all comparators e.g. placebo (PC), ibuprofen 800 mg three times a day (IBU / IBU 800mg TDS / IBU 800 TDS), naproxen 500mg twice daily (NAP / NAP 500mg BD / NAP 500 BD), diclofenac 50mg three times a day (DIC / DIC 50mg TDS / DIC 50 TDS), nabumetone 1000mg once a day (NAB / NAB 1000mg OD/ NAB 1000 OD), paracetamol 1g four times a day (PAR / PAR 1g QDS / PAR 1 QDS) etc. where the first three or four letters of the name of the

comparator in capitals was used, followed in the total patient number section of each systematic review or meta-analysis by the dose in milligrams (mg) or grams (g) and the frequency [once a day (OD), twice a day (BD), three times a day (TDS), four times a day (QDS)]. For rofecoxib only the first capital letter R was used which in all other columns of the table indicate rofecoxib apart from the "Design" column where it denotes a randomised clinical trial. Finally, in a section in the same table ("Key") any explanatory notes only specific to that systematic review or meta-analysis were given, while in a final "KEY" section explanatory notes that are useful for the whole table were reported.

4.1.4.8 Population characteristics in the included systematic reviews and meta-analyses

An attempt was made to summarise the age, gender distribution and ethnicity characteristics of the population included for each included systematic review or meta-analysis. If this information was not readily available, the median and the range was calculated for the age in years, and the percentage of the females as well as the percentages of the different ethnic groups was calculated.

4.1.4.9 Industrial funding of included systematic reviews and meta-analyses

It is important to explore whether the systematic reviews and meta-analyses included in our analysis received funding from external sources or to identify any conflicts of interest. Thus, all published systematic reviews and meta-analyses were scanned thoroughly to identify the sources of funding (e.g. manufacturer's of COX-2

inhibitors, government organisations). Additionally, the relationship of authors of the published papers with manufacturers of COX-2 inhibitors was explored and noted.

Chapter 5

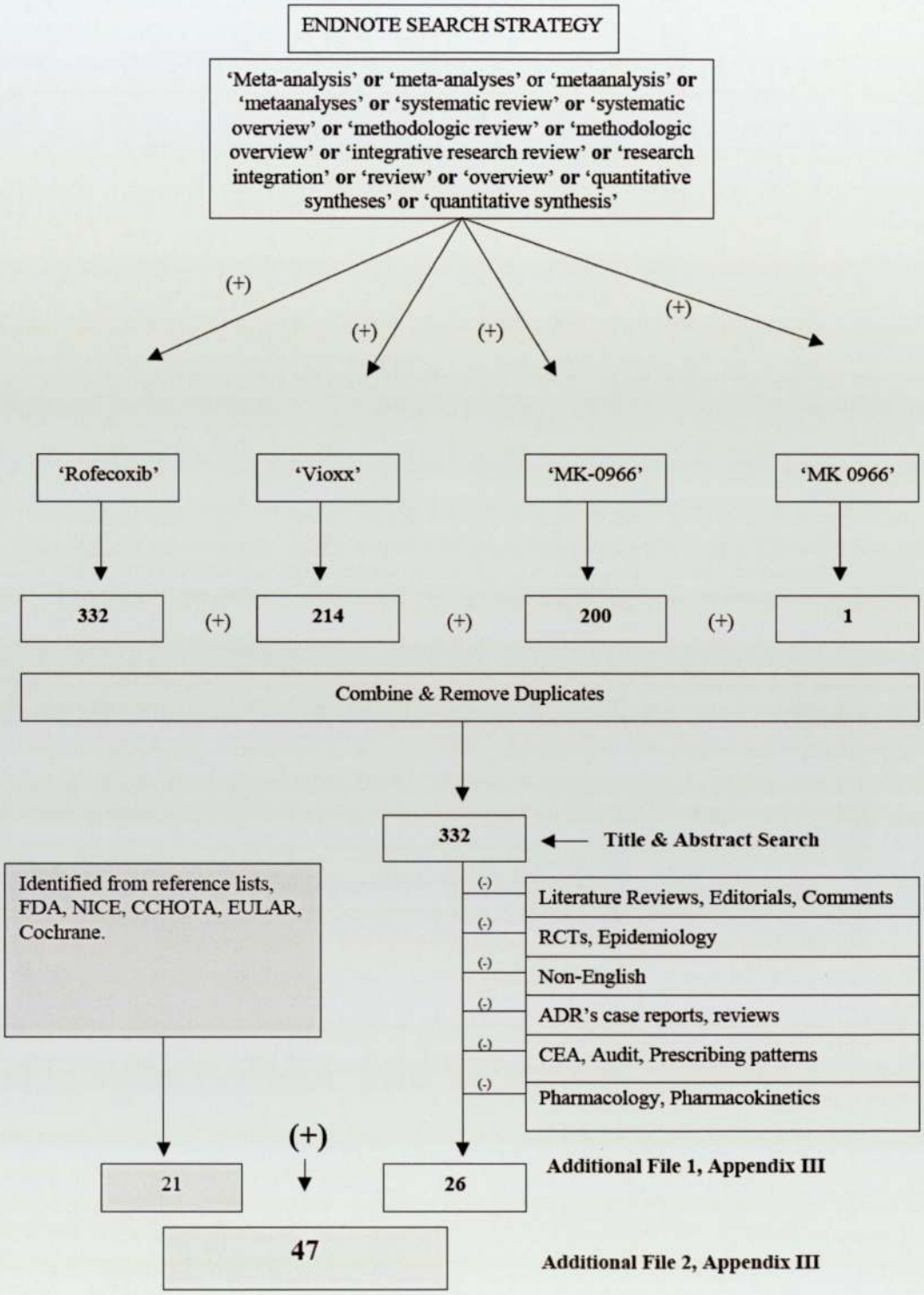
Results

5.1 Results

5.1.1 Search Strategy

Using Endnote 7.00 software, 332 articles were identified after combining and removing duplicates. The resulting set of citations were screened by title and, whenever available, by abstract. All systematic reviews and meta-analyses that may have contained RCTs comparing rofecoxib to placebo or active drug (s) were identified for further evaluation (Additional File 1, Appendix III). Articles identified from reference lists of literature reviews and other articles obtained for information, as well as articles identified from the FDA, NICE, CCHOTA, EULAR and Cochrane Database sites were screened for any additional citations and provided an additional set of citations that was added to the one obtained from the ENDNOTE searching. From the resulting set of citations (47 articles), that were fully obtained and screened (Additional File 2, Appendix III), only 15 were included in the final analysis and 32 were excluded. The reasons for exclusion were: (i) 15 presented results for acute pain trials with limited exposure to rofecoxib (usually around 24 hours only), (ii) 2 were only efficacy systematic reviews / meta-analyses, (iii) 8 focused only on gastrointestinal safety endpoints (iv) 2 focused on other safety endpoint i.e. one on liver toxicity and one on bronchoconstriction (v) 4 included only one RCT comparing rofecoxib with placebo or active drug and finally (vi) a further one was the identical systematic review of an updated systematic review that was included. A summary of the article inclusion

pathway is presented in Figure 5.1 with a summary of excluded articles presented in Table 5.1.



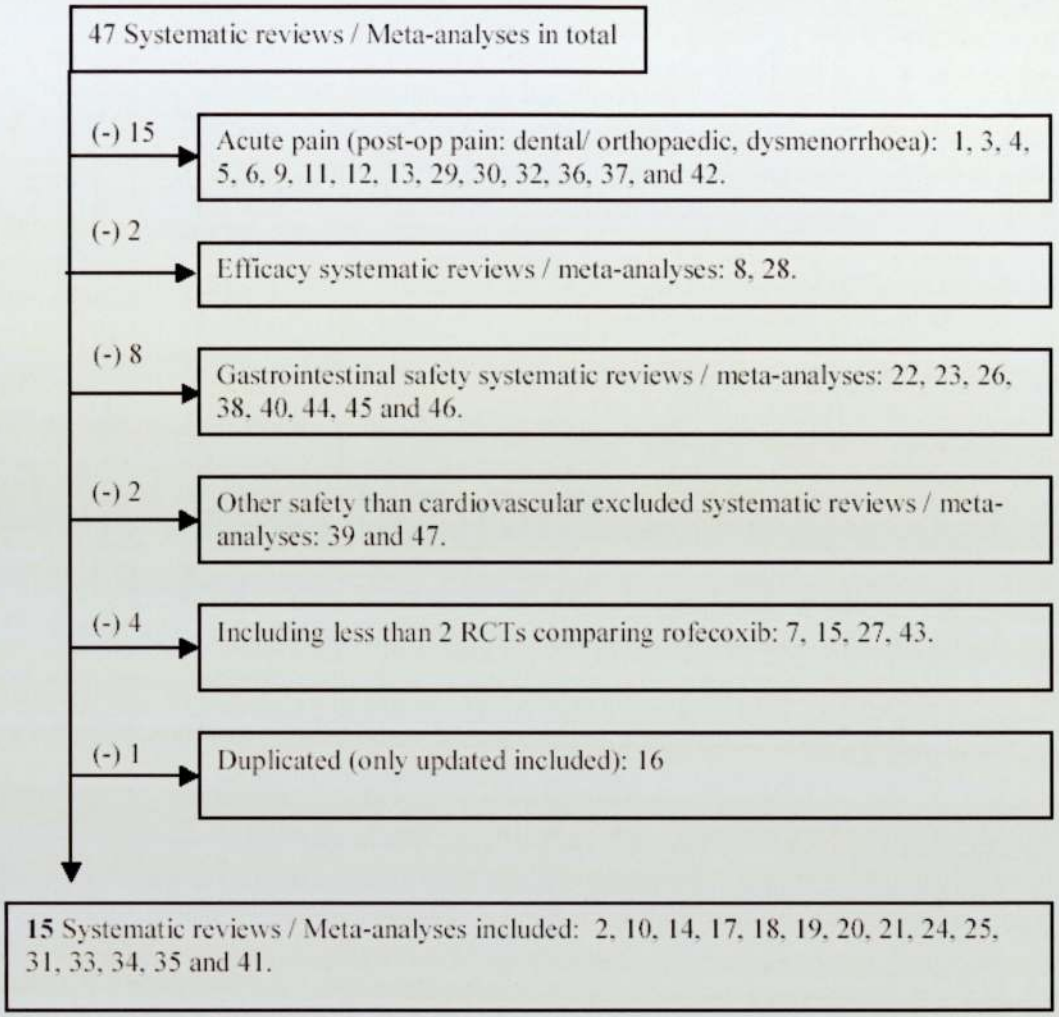


Figure 5.1 Schematic representation of search strategy employed for retrieval of systematic reviews and meta-analyses concerning cardiovascular, cerebrovascular and renal adverse-effects in RCTs comparing rofecoxib with a comparator

Table 5.1 Excluded articles and the reason of exclusion

Exclusion Criteria	Reference*
Acute pain systematic reviews / meta-analyses	No author [1], Barden 2002 [2], Barden 2004 [3], Barden 2004a [4], Barden 2005 [5], Chen [6], Desjardins [7], Edwards 2004a [8], Edwards 2004b [9], Mehlisch [10], Moore 2005b[11], Morrison [12], Romsing 2004 [13], Romsing 2005 [14], Straube [15]
Efficacy systematic reviews / meta-analyses	Bjordal [16], Lee 2005 [17]
Gastrointestinal safety systematic reviews / meta-analyses	Gomez Cerezo [18], Hooper [19], Langman [20], Rostom 2004 [21], Rostom 2005a [22], Watson 2000 [23], Watson 2001 [24], Watson 2004 [25]
Other safety systematic reviews / meta-analyses	Rostom 2005 [26], West [27]
Included only 1 RCT comparing rofecoxib	Bassett [28], Gagnier [29], Lee 2004 [30], Towheed [31]
Updated	Garner 2002 [32]
*	Numbers refer to Additional File 3, Appendix III

5.1.2 Results for derived outcome measures

5.1 2.1 Quality scoring

The mean (SD) overall quality scoring, using the 18-item QUOROM checklist, was 63.13 % (21.41%) with a range of 44.40 - 83.30 % and a median score of 61.10 % (Table 5.3). Using our pre-determined definition of quality, no article received a grade of 'very poor quality', 4 were 'poor', 3 were 'acceptable', and 4 were 'good'. Individual systematic reviews or meta-analyses scores are given in detail in Table 5.2, while overall category scores were also examined, and are presented in Table 5.3. The worst performing categories were *Title* (mean = 27.3 %, SD = 46.7 %), and *Results*

(mean = 45.46 %, SD = 26.97 %). The overall *Abstracts* score (mean = 69.70 %, SD = 20.84), *Methods* score (mean = 71.21 %, SD = 26.97 %), and *Discussion* score (mean = 63.60 %, SD = 50.50 %) were acceptable. Good quality scores were found for the *Introduction* (mean = 81.80 %, SD = 40.5 %).

The mean (SD) overall quality scoring, using the QUOROM checklist only for the title and abstract for the two included abstracts (Daniels and Seidenberg 1999; Geba, Polis et al. 2003), was 42.9 % (20.2%) with a range of 28.6 –57.1 % and a median score of 42.9 %. Using our pre-determined definition of quality, no abstract received a grade of 'very poor quality', 1 was 'poor', and 1 was 'acceptable'. Individual systematic reviews or meta-analyses (reported only as abstracts) scores are given in detail in Table 5.2, while overall category scores were also examined and are presented in Table 5.4. The worst performing category was *Title* (mean = 0.0%, SD = 0.0%). The overall *Abstracts* score (mean = 50.0%, SD = 23.6%) was acceptable.

Comparing quality scores before and after publication of the QUOROM statement would not have been beneficial, as all papers were published after the QUOROM statement was published.

Table 5. 2 QUOROM checklist results for CV* systematic reviews / meta-analyses for rofecoxib

Heading	Subheading	Descriptor	Aw, T-J et al 2005	Daniels, B. 1999 [A]	Garner, S. et al 2005 CD005115	Garner, S. et al 2005 CD003685	Geba, G 2003 [A]	Gertz, B. J. 2002	Goldstein, J. L. et al 2005
Title		Identify the report as a meta-analysis [or systematic review] of RCTs	1	0	0	0	0	0	0
Abstract		Use a structured format	1	1	1	1	0	1	1
	Objectives	The clinical question explicitly	1	1	1	1	1	1	1
	Data Sources	The databases (i.e. list) & other information sources	0	1	1	1	0	0	0
	Review Methods	The selection criteria (i.e.population, intervention, outcome, & study design); methods for validity assessment, data abstraction, & study characteristics, and quantitative data synthesis in sufficient detail to permit replication	0	0	1	1	0	0	0
	Results	Characteristics of the RCTs included & excluded;quantitative findings (i.e. point estimates and confidence intervals); & subgroup analyses	1	0	1	1	0	0	1
	Conclusion	The main results	1	1	0	1	1	1	1
Introduction		The explicit clinical problem, biological rationale for the intervention, and rationale for review	1	NA	1	1	NA	1	1

Table 5. 2 QUOROM checklist results for CV* systematic reviews / meta-analyses for rofecoxib

Heading	Subheading	Descriptor	Aw, T-J et al 2005	Daniels, B. 1999 [A]	Garner, S. et al 2005 CD005115	Garner, S. et al 2005 CD003685	Geba, G 2003 [A]	Gertz, B. J. 2002	Goldstein, J. L. et al 2005
Methods	Searching	The information sources, in detail (e.g. databases, registers, personal files, expert informants, agencies, hand-searching), & any restrictions (years considered, publication status, language of publication)	1	NA	1	1	NA	1	1
	Selection criteria	The inclusion & exclusion criteria (defining population, intervention, principal outcomes, & study design)	1	NA	1	1	NA	0	1
	Validity assessment	The criteria & process used (e.g masked conditions, quality assessment, & their finding)	0	NA	1	1	NA	0	0
	Data abstraction	The process or processes used (e.g. completed independently, in duplicate)	1	NA	1	1	NA	0	0
	Study characteristics	The type of study design, participants' characteristics, details of intervention, outcome definitions, & how clinical heterogeneity was assessed	1* heterogeneity	NA	1*	1*	NA	1*	1
	Quantitative data synthesis	The principal measures of effect (e.g relative risk), method of combining results (statistical testing & CI), handling of missing data; how statistical heterogeneity was assessed; a rationale for any a-priory sensitivity and subgroup analysis; and any assessment of publications bias	1*	NA	1*	1*	NA	1*	1

Table 5. 2 QUOROM checklist results for CV* systematic reviews / meta-analyses for rofecoxib

Heading	Subheading	Descriptor	Aw, T-J et al 2005	Daniels, B. 1999 [A]	Garner, S. et al 2005 CD005115	Garner, S. et al 2005 CD003685	Geba, G 2003 [A]	Gertz, B. J. 2002	Goldstein, J. L. et al 2005
Results	Trial flow	Provide a meta-analysis profile summarising trial flow	1	NA	0	0	NA	0	0
	Study characteristics	Provide descriptive data for each trial (e.g. age, sample size, intervention, dose, duration, follow-up period)	1	NA	1	1	NA	1	1
	Quantitative data synthesis	Report agreement on the selection and validity assessment; present simple summary results (for each treatment group in each trial, for each primary outcome); present data needed to calculate effect sizes and confidence intervals in ITT analysis (e.g. 2 x 2 tables of counts, means & SDs, proportions)	0	NA	1	1	NA	0	1
Discussion		Summarise key findings; discuss clinical inferences based on internal and external validity; interpret the results in light of the totality of available evidence; describe potential biases in the review process (e.g. publication bias); and suggest a future research agenda)	1	NA	1	1	NA	0	0
Total score (%)		(Maximum score = 18)	13 (72.2%)	4 of 7 (57.1%)	15 (83.3%)	16 (88.9%)	2 of 7 (28.6%)	8 (44.4%)	10 (55.6%)

Table 5. 2 QUOROM checklist results for CV* systematic reviews / meta-analyses for rofecoxib

Heading	Subheading	Descriptor	Juni, P. et al 2004	Konstam, M. A. et al 2001	Moore, R. A. et al 2005	Mukherjee, D. et al 2001	Reicin, A. S. et al 2002	Schnitzer, T. J. et al 2005
Title		Identify the report as a meta-analysis [or systematic review] of RCTs	1	0	1	0	0	0
Abstract		Use a structured format	1	1	0	0	0	1
	Objectives	The clinical question explicitly	1	1	1	1	1	1
	Data Sources	The databases (i.e. list) & other information sources	1	1	1	1	0	0
	Review Methods	The selection criteria (i.e.population, intervention, outcome, & study design); methods for validity assessment, data abstraction, & study characteristics, and quantitative data synthesis in sufficient detail to permit replication	1	0	1	0	0	0
	Results	Characteristics of the RCTs included & excluded;quantitative findings (i.e. point estimates and confidence intervals); & subgroup analyses	1	1	0	1	0	0
	Conclusion	The main results	1	1	1	1	1	1
Introduction		The explicit clinical problem, biological rationale for the intervention, and rationale for review	1	1	0	1	0	1

Table 5. 2 QUOROM checklist results for CV* systematic reviews / meta-analyses for rofecoxib

Heading	Subheading	Descriptor	Juni, P. et al 2004	Konstam, M. A. et al 2001	Moore, R. A. et al 2005	Mukherjee, D. et al 2001	Reicin, A. S. et al 2002	Schnitzer, T. J. et al 2005
Methods	Searching	The information sources, in detail (e.g. databases, registers, personal files, expert informants, agencies, hand-searching), & any restrictions (years considered, publication status, language of publication)	1	1	1	1	1	0
	Selection criteria	The inclusion & exclusion criteria (defining population, intervention, principal outcomes, & study design)	1	1	1	0	0	1
	Validity assessment	The criteria & process used (e.g masked conditions, quality assessment, & their finding)	1	0	1	0	0	0
	Data abstraction	The process or processes used (e.g. completed independently, in duplicate)	1	0	1	0	0	0
	Study characteristics	The type of study design, participants' characteristics, details of intervention, outcome definitions, & how clinical heterogeneity was assessed	1	1	1	0	0	1*
	Quantitative data synthesis	The principal measures of effect (e.g relative risk), method of combining results (statistical testing & CI), handling of missing data; how statistical heterogeneity was assessed; a rationale for any a-priory sensitivity and subgroup analysis; and any assessment of publications bias	1	1	1	1*	1	1*

Table 5. 2 QUOROM checklist results for CV* systematic reviews / meta-analyses for rofecoxib

Heading	Subheading	Descriptor	Juni, P. et al 2004	Konstam, M. A. et al 2001	Moore, R. A. et al 2005	Mukherjee, D. et al 2001	Reicin, A. S. et al 2002	Schnitzer, T. J. et al 2005
Results	Trial flow	Provide a meta-analysis profile summarising trial flow	1	0	0	0	0	0
	Study characteristics	Provide descriptive data for each trial (e.g. age, sample size, intervention, dose, duration, follow-up period)	1	1	1	0	0	0
	Quantitative data synthesis	Report agreement on the selection and validity assessment; present simple summary results (for each treatment group in each trial, for each primary outcome); present data needed to calculate effect sizes and confidence intervals in ITT analysis (e.g. 2 x 2 tables of counts, means & SDs, proportions)	0	0	1	0	1	0
Discussion		Summarise key findings; discuss clinical inferences based on internal and external validity; interpret the results in light of the totality of available evidence; describe potential biases in the review process (e.g. publication bias); and suggest a future research agenda)	1	0	1	1	1	0
Total score (%)		(Maximum score = 18)	17 (94.4%)	11 (61.1%)	14 (77.8%)	8 (44.4%)	6 (33.3%)	7 (38.9%)

Table 5.3 Overall quality score by category and item (N = 11)

Category	Item	Mean (range)	Quality grade
Title	Identified as a meta-analysis of RCTs in the title?	27.30% (0-100%)	Poor
Abstract		69.70% (33.3-100%)	Acceptable
	Structured format?	72.73%	
	Objective/clinical question described?	100.00%	
	Databases and sources listed?	54.55%	
	Selection criteria, etc. been described sufficiently?	36.36%	
	Have characteristics been described?	63.64%	
	Main results reported?	90.91%	
Introduction	Clinical problem desribed?	81.80% (0-100%)	Good
Methods		71.21% (33.3-100%)	Acceptable
	Described searching methods and sources?	90.91%	
	Inclusion and exclusion?	72.73%	
	Validity assessment described?	36.36%	
	Processes for data abstraction described?	45.45%	
	Study design, and clinical heterogeneity assessed?	81.82%	
	Principal measure of effect, etc. been described?	100.00%	
Results		45.46% (0-66.7%)	Poor
	Trial flow-chart provided?	18.18%	
	Descriptive data presented?	72.73%	
	Authors reported agreement, effect sizes, and CIs, etc.?	45.45%	
Discussion	Summarised key findings, etc?	63.60% (0-100%)	Acceptable
Overall		63.13% (33.3-94.4%)	Acceptable

Table 5.4 Overall quality score by category and item (N = 2) [Abstracts only]

Category	Item	Mean (range)	Quality grade
Title	Identified as a meta-analysis of RCTs in the title?	0.00% (0-0.0%)	Very poor
Abstract		69.70% (33.3-100%)	Acceptable
	Structured format?	50.00%	
	Objective/clinical question described?	100.00%	
	Databases and sources listed?	50.00%	
	Selection criteria, etc. been described sufficiently?	0.00%	
	Have characteristics been described?	0.00%	
	Main results reported?	100.00%	

5.1.2.2 Methods of Information Retrieval utilised

Table 5.5 summarises the different databases or available sources of information searched to provide for RCTs included in the systematic reviews or meta-analyses analysed. Nine of the included systematic reviews / meta-analyses (FDA 2001; Daniels and Seidenberg 1999; Konstam, Weir et al. 2001; Gertz, Krupa et al. 2002; Reicin, Shapiro et al. 2002; Geba, Polis et al. 2003; Goldstein, Bello et al. 2005; Moore, Derry et al. 2005a; Schnitzer, Weaver et al. 2005) utilised the manufacturer's data files as their solely source of information [8 of them Merck Co., Inc. files and 1 Pfizer (Pharmacia), Inc. files (Moore, Derry et al. 2005a)]. All of the remaining (six) systematic reviews or meta-analyses (Mukherjee, Nissen et al. 2001; NICE 2001; Juni, Nartey et al. 2004; Aw, Haas et al. 2005; Garner, Fidan et al. 2005a; Garner, Fidan et al. 2005b) that were analysed had used Medline. An Embase search, that is normally done to supplement the Medline search, was performed in 4 systematic reviews or meta-analyses (NICE 2001; Juni, Nartey et al. 2004; Garner, Fidan et al. 2005a;

Garner, Fidan et al. 2005b). The CCTR and the Cochrane database of systematic reviews was searched by another three systematic reviews or meta-analyses (NICE 2001; Juni, Nartey et al. 2004; Aw, Haas et al. 2005) in addition to the two included Cochrane systematic reviews (Garner, Fidan et al. 2005a; Garner, Fidan et al. 2005b). The FDA site was searched by only two systematic reviews or meta-analyses (Juni, Nartey et al. 2004; Aw, Haas et al. 2005). Systematic hand searching of reference lists and bibliographies was only performed in 5 included systematic reviews or meta-analyses (NICE 2001; Juni, Nartey et al. 2004; Aw, Haas et al. 2005; Garner, Fidan et al. 2005a; Garner, Fidan et al. 2005b).

The final date that a search was conducted was stated in only 3 (FDA 2001; Konstam, Weir et al. 2001; Moore, Derry et al. 2005) out of the 9 systematic reviews or meta-analyses that utilised manufacturer's files. All the remaining published systematic reviews or meta-analyses reported the final date a search was performed or updated (Table 5.5) (Mukherjee, Nissen et al. 2001; NICE 2001; Juni, Nartey et al. 2004; Aw, Haas et al. 2005; Garner, Fidan et al. 2005a; Garner, Fidan et al. 2005b).

5.1.2.3 Methods of Data Extraction utilised

The 8 systematic reviews or meta-analyses that utilised Merck's data files (FDA 2001; Daniels and Seidenberg 1999; Konstam, Weir et al. 2001; Gertz, Krupa et al. 2002; Reicin, Shapiro et al. 2002; Geba, Polis et al. 2003; Goldstein, Bello et al. 2005; Schnitzer, Weaver et al. 2005) did not provide information about the data abstraction process (Table 5.6). In 4 of the remaining systematic reviews or meta-analyses (Juni, Nartey et al. 2004; Aw, Haas et al. 2005; Garner, Fidan et al. 2005a; Garner, Fidan et

al. 2005b) a minimum of two independent reviewers abstracted the required information, while for the National Institute of Clinical Excellence (NICE) systematic review one independent reviewer filled in a prepared extraction form that was checked by a second independent reviewer (Table 5.6). This method was subsequently modified to involve two independent reviewers in the updated version including data till December 2000 (NICE 2001). One systematic review (Mukherjee, Nissen et al. 2001) did not give any information on the data abstraction process. In only one systematic review (Aw, Haas et al. 2005) was one of the reviewers identified to be blinded, and in only one meta-analysis was the abstracted information checked by two different reviewers (Juni, Nartey et al. 2004).

Consensus is normally achieved by discussion. In the two Cochrane systematic reviews, consensus was achieved by contacting the authors of the RCTs (Garner, Fidan et al. 2005a; Garner, Fidan et al. 2005b). However, in 10 of the 13 fully published systematic reviews or meta-analyses (FDA 2001; Konstam, Weir et al. 2001; Mukherjee, Nissen et al. 2001; NICE 2001; Gertz, Krupa et al. 2002; Reicin, Shapiro et al. 2002; Juni, Nartey et al. 2004; Aw, Haas et al. 2005; Goldstein, Bello et al. 2005; Schnitzer, Weaver et al. 2005) a formal method to achieve consensus was not described. However, for the updated version of NICE, discussion was the main method to resolve any discrepancies, while contacting the authors was possible, if more clarification was required. None of the systematic reviews or meta-analyses included in this study employed a third reviewer as the method to achieve consensus.

Table 5.5 Methods of information (RCTs) retrieval included in systematic reviews / meta-analyses concerning Rofecoxib CV* safety

Sources	Aw, T-J et al 2005	Daniels, B. et al 1999 [A]	FDA	Garner, S. E. et al 2005 CD005115	Garner, S. E. et al 2005 CD003685	Geba, G 2003 [A]	Gertz, B. J. 2002	Goldstein, J. L. et al 2005	Juni, P. et al 2004	Konstam, M. A. et al 2001	Moore, R. A. et al 2005	Mukherjee, D. et al 2001	NICE	Reicin, A. S. et al 2002	Schnitzer, T. J. et al 2005
Medline	•			•	•				•			•	•		
Embase				•	•				•				•		
CCTR				•	•				•				•		
Cochrane	•			•	•										
FDA	•		NA						•						
Ref Lists	•			•	•				•				•		
Reviews					•										
Other / Fugitive Literature	[A], bibliographies	Merck	Merck	Experts, NRR, NHS EED, Health Technology Assessment Database	NRR, NHS EED, Health Technology Assessment Database, World Wide Web, NICE+	Merck	Merck	Merck	CINAHL, Science Citation Index, Conference proceedings, Experts	Merck	Pfizer - Pharmacia	World wide web	NRR, NHS EED, Merck, Pfizer, Boerhinger Ingelheim, Shire, HTA, HEED, World wide web	Merck	Merck
End Date	May 2004	NS- OA dev prog	February 2001	August 2004	December 2000	NS	NS	NS	Sep 2004	15th Sep 2000	Dec 2003	Jan 98 till Feb 01	October 2000/ December 2000	NS	NS

Key: CV*: Cardiovascular, renal cerebrovascular adverse-effects; NS: Not stated; NA: Not applicable; [A]: Abstract (in conferencec proceedings); Merck / Pfizer / Boerhinger Ingelheim / Shire: Used manufacturer's files; Ref lists: Check reference lists of published articles; Fugitive Literature: Attempt to identify RCTs in government / licensing authorities sites, organisations, conference reports, etc.; *: (Searched only for systematic reviews & policies using the Cochrane Collaboration trial & CRD economic evaluation filter; NRR: National Research Register, NHS EED: National Health Service Economic Evaluation Database; NICE+: Merck submissions to NICE of any additional/unpublished information; HTA: Health Technology Assessment database; OA dev prog: Included only the 9 RCTs forming the basis for licensing rofecoxib for the relief and treatment of pain in osteoarthritis patients.

Table 5.6 Data abstraction techniques utilised in systematic reviews / meta-analyses concerning rofecoxib's CV safety

Meta-analysis / Systematic Review	Aw, T-J et al 2005	Daniels, B. et al 1999 [A]	FDA	Garner, S. E. et al 2005 CD 005115	Garner, S. E. et al 2005 CD 003685	Geba, G 2003 [A]	Gertz, B. J. 2002	Goldstein, J. L. et al 2005	Juni, P. et al 2004	Konstam, M. A. et al 2001	Moore, R. A. et al 2005	Mukherjee, D. et al 2001	NICE	Reicin, A. S. et al 2002	Schnitzer, T. J. et al 2005
Independent reviewers	2	NS	NS	2	2	NS	NS	NS	2	NS	3	NS	1*	NS	NS
Blinded reviewers	1	NS	NS	NS	NS	NS	NS	NS	No	NS	No	NS	NS	NS	NS
Checked	No	NS	NS	No	No	NS	NS	NS	Other 2	NS	No	NS	Yes (1)	NS	NS
Discussion	NS	NS	NS	Yes	Yes	NS	NS	NS	NS	NS	Yes	NS	NS*	NS	NS
Third reviewer	No	NS	NS	No	No	NS	NS	NS	NS	NS	No	NS	No	NS	NS
Other				Contact authors to resolve	Contact authors to resolve								*		

Key; CV: Cardiovascular, NS: Not Stated, NA: Not Applicable, Other: Other ways of achieving concensus, *: NICE update used 2 independent reviewers while all discrepancies were resolved by discussion or by contacting the authors.

5.1.2.4 Duration of RCTs included in the analysed systematic reviews

One week (Garner, Fidan et al. 2005b) was the shortest duration for a RCT to be included for analysis in the included systematic reviews or meta-analyses and 86 weeks was the longest (Konstam, Weir et al. 2001) (Table 5.7). Six systematic reviews or meta-analyses excluded RCTs with duration shorter than 4 weeks (Konstam, Weir et al. 2001; NICE 2001; Juni, Nartey et al. 2004; Aw, Haas et al. 2005; Garner, Fidan et al. 2005a) and one exluded RCTs with duration shorter than 2 weeks (Moore, Derry et al. 2005a).

Table 5.7 Duration of included RCTs in CV safety systematic reviews / meta-analyses identified

Meta-analysis / Systematic Reviews	Excluded if	Shorter included	Longer Included	Average duration (Median)
Aw, T-J. et al 2005	< 4 weeks	4 weeks	24 weeks	12 W
Daniels, B. et al 1999 [A]		NS	86 weeks	22 W*
FDA		4 weeks	52 weeks	UC
Garner, S. E. et al 2005 CD005115		1 week (7day cross over)	52 weeks	6 W
Garner, S. E. et al 2005 CD003685	< 4 weeks	8 weeks	36weeks	30 W
Geba, G 2003 [A]			6 weeks	6 W
Gertz, B. J. et al 2002		6 weeks	24 weeks	6 W
Goldstein, J. L. et al 2005		6 weeks	6 weeks	6 W
Juni, P. et al 2004	< 4 weeks	4 weeks	56 weeks	12 W
Konstam, M. A. et al 2001	< 4 weeks	4 weeks	86 weeks	24.5 W
Moore, R. A. et al 2005	< 2 weeks	2 weeks	52 weeks	6 W (for R trials) *1
Mukherjee, D. et al 2001		6 weeks	52 weeks	UC
NICE	< 4 weeks	6 weeks	52 weeks	6 W
Reicin, A. S. et al 2002	NS	NS	88weeks	14 W
Schnitzer, T. J. et al 2005		6 weeks	6 weeks	6 W
Total Duration [Median, (Q1-Q3)]				6W (6-13)

Key; NS: Not Stated, N/A: Not Applicable, W: weeks, UC: Unable to calculate based on published data, R: Rofecoxib, *: Mean reported, median could not be calculated, *1: 12 W or more (77% of observations), Q1: Lower quartile, Q3: Upper quartile.

Table 5.7 shows that the median duration of exposure to rofecoxib in the included systematic reviews or meta-analyses [apart from the two (FDA 2001; Mukherjee, Nissen et al. 2001) that it was not possible to calculate or obtain the value

from the published articles] is 6 weeks (lower quartile: 12, upper quartile: 13), which is relatively short for long-term indications as OA or RA.

5.1.2.5 Indication of RCTs included in the analysed systematic reviews

Fourteen systematic reviews or meta-analyses included OA RCTs and both the two abstracts focused solely on OA (Table 5.8). Only one systematic review focused solely on RA RCTs of rofecoxib (Garner, Fidan et al. 2005a), while another 7 included RCTs of rofecoxib indicated for both OA and RA (FDA; Konstam, Weir et al. 2001; Mukherjee, Nissen et al. 2001; NICE 2001; Juni, Nartey et al. 2004; Aw, Haas et al. 2005; Moore, Derry et al. 2005). Two meta-analyses included additionally to OA and RA chronic low back pain (FDA; Juni, Nartey et al. 2004), while two included also RCTs for the prevention and treatment of Alzheimer's disease (unlicensed indication) (FDA; Konstam, Weir et al. 2001). Finally, no systematic review or meta-analysis included any RCTs for the prevention of any type of cancer, for which rofecoxib never received license.

Table 5.8 Indications for Rofecoxib use in the RCTs included in the systematic reviews / meta-analyses analysed

	Indications				
Meta-analysis / Systematic Review	**Osteoarthritis**	**Rheumatoid Arthritis**	**Chronic Low Back Pain**	**Alzheimer's Disease***	**Cancer***
Aw, T-J. et al 2005	•	•			
Daniels, B. et al 1999 **[A]**	•				
FDA	•	•	•	•	
Garner, S. E. et al 2005 CD005115	•				
Garner, S. E. et al 2005 CD003685		•			
Geba, G 2003 **[A]**	•				
Gertz, B. J. et al 2002	•				
Goldstein, J. L. et al 2005	•				
Juni, P. et al 2004	•	•	•		
Konstam, M. A. et al 2001	•	•		•	
Moore, R. A. et al 2005	•	•			
Mukherjee, D. et al 2001	•	•			
NICE	•	•			
Reicin, A. S. et al 2002	•				
Schnitzer, T. J. et al 2005	•				

Key: Cancer: colon adenoma prevention, lung cancer, breast cancer etc., *: not licensed indication.

5.1.2.6 Rofecoxib's Comparators studied in the included systematic reviews

Twelve systematic reviews or meta-analyses contained information comparing rofecoxib to placebo (Table 5.9) (FDA 2001; Daniels and Seidenberg 1999; Konstam, Weir et al. 2001; Mukherjee, Nissen et al. 2001; NICE 2001; Gertz, Krupa et al. 2002; Geba, Polis et al. 2003; Juni, Nartey et al. 2004; Aw, Haas et al. 2005; Garner, Fidan et al. 2005a; Garner, Fidan et al. 2005b; Moore, Derry et al. 2005a). All licensed dosing regimes of comparators utilised were within UK licensed doses ((BNF) March 2005) apart from nabumetone 1500mg once daily that was higher than the maximum recommended dose (1000mg once a day). In the Cochrane systematic review that included a trial comparing rofecoxib with valdecoxib 10mg (Garner, Fidan et al. 2005b), no analysis was performed as only one trial was included. The same systematic review was the only one to include RCTs comparing rofecoxib to Nimesulide and Nimesulide Retard (slow release formulation of Nimesulide) (Garner, Fidan et al. 2005). Naproxen, the most popular NSAID comparator, was included in 9 out of 15 systematic reviews or meta-analyses (FDA 2001; Konstam, Weir et al. 2001; Mukherjee, Nissen et al. 2001; NICE 2001; Juni, Nartey et al. 2004; Aw, Haas et al. 2005; Garner, Fidan et al. 2005a; Garner, Fidan et al. 2005b; Moore, Derry et al. 2005), followed by diclofenac (8 out of 15, (FDA 2001; Konstam, Weir et al. 2001; NICE 2001; Gertz, Krupa et al. 2002; Reicin, Shapiro et al. 2002; Juni, Nartey et al. 2004; Aw, Haas et al. 2005; Garner, Fidan et al. 2005b)) and ibuprofen (7 out of 15, (FDA 2001; Konstam, Weir et al. 2001; NICE 2001; Gertz, Krupa et al. 2002; Reicin, Shapiro et al. 2002; Juni, Nartey et al. 2004; Garner, Fidan et al. 2005b)). Rofecoxib was only compared to one specific COX-2 inhibitor (Celecoxib). The dose of celecoxib used was the lower recommended dose for OA. In one of the abstracts (Daniels and Seidenberg

1999) included in this analysis the NSAID comparators utilised was not stated (Table 5.9).

Table 5.9 Rofecoxib's comparators studied in the included systrematic reviews / meta-analyses

Comparators vs rofecoxib	Aw, T-J.et al 2005	Daniels, B. 1999 [A]	FDA	Garner, S. E. et al 2005 CD005115	Garner, S. E. et al 2005 CD003685	Geba, G. 2003 [A]	Gertz, B. J. et al 2002	Goldstein, J. L. et al 2005	Juni, P. et al 2004	Konstam, M. A. et al 2001	Moore, R. A. et al 2005	Mukherjee, D. et al 2001	NICE	Reicin, A. S. et al 2002	Schnitzer, T. J. et al 2005
Placebo	•	•	•	•	•	•	•		•	•	•	•	•		
Naproxen 500mg BD	•		•	•	•				•	•	•	•	•		
Ibuprofen 800mg TDS			•	•			•		•	•			•	•	
Diclofenac 50mg TDS	•		•	•*			•		•	•			•*2	•	
Nimesulide 100mg OD				•											
Nimesulide 300mg Retard OD				•											
Nabumetone 500mg BD									•	•		•			
Nabumetone 1000mg OD				•						•		•	•	•	
Nabumetone 1500mg OD			•	•			•		•				•		
Celecoxib 200mg OD	•			•		•		•			•				•
Valdecoxib 10mg				•*1											
Paracetamol 1000mg QDS				•		•									•
NSAID (not specified)		•													

Key; *: with or without Misoprostol 200mg TDS, *1:only one study in the systematic review, not compared, Nimesulide Retard: Slow release formulation of Nimesulide, *2: Diclofenac 50mg TDS or Diclofenac 50mg with Misoprostol 200mg BD.

5. 1. 2. 7 Objectives, Inclusion & Exclusion Criteria and Design of included RCTs of the included systematic reviews and meta-analyses

Table 5.10 summarises information about each included systematic review or meta-analyses with respect to objectives, inclusion / exclusion criteria and gives a brief outline of the RCTs that were used in the analysis of the included systematic reviews or meta-analysis. In the included 15 systematic reviews and meta-analyses, a total of 39 RCTs were included of which only 15 are analysed uniquely in one systematic review or meta-analysis (Table 5.11). The two abstracts (Daniels and Seidenberg 1999; Geba, Polis et al. 2003) were probably later fully published, as exactly the same RCTs and population was analysed with almost identical aims in two identified (Gertz, Krupa et al. 2002; Schnitzer, Weaver et al. 2005) systematic reviews or meta-analyses. Efficacy was the primary aim for 6 systematic reviews and meta-analyses (NICE 2001; Geba, Polis et al. 2003; Garner, Fidan et al. 2005a; Garner, Fidan et al. 2005b; Moore, Derry et al. 2005a; Schnitzer, Weaver et al. 2005). Blood pressure effects was the primary aim of 3 meta-analyses (Geba, Polis et al. 2003; Aw, Haas et al. 2005; Schnitzer, Weaver et al. 2005). Renovascular safety was the primary aim of 1 systematic review (Gertz, Krupa et al. 2002) but was also assessed in one more meta-analysis (Goldstein, Bello et al. 2005). Thromboembolic adverse effects associated with rofecoxib use were the aim of 8 systematic reviews and meta-analyses (FDA 2001; Daniels and Seidenberg 1999; Konstam, Weir et al. 2001; Mukherjee, Nissen et al. 2001; Reicin, Shapiro et al. 2002; Juni, Nartey et al. 2004; Garner, Fidan et al. 2005a; Garner, Fidan et al. 2005b). Finally, total adverse effects and withdrawals was the main safety aim of two systematic reviews (NICE 2001; Moore, Derry et al. 2005a)

The total number of patients if allowances are made for the inclusion of a trial in more than one systematic reviews or meta-analyses was 44343. However, there are some discrepancies between different meta-analyses particularly about the number of patient analysed in the RCT included or the number of patients it is still valid to use in a systematic review or meta-analysis with a specific aim.

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring
Aw, T-J. et al (2005) Arch. Int. Med. 165: 1-7	WMD SBP & DBP	Prospecti-ve	<50 Pts	BOMBARDIER, C 2000 NEJM 343:1520-8	8076	RA	R, C, B, P, MC	**R50**	NAP 500mg BD	36	
	RR of Pts developing HT (coxibs vs PC, coxibs vs NSAIDs)	Outcome described in coxibs & compa-rators	<4 weeks duration	CANNON, GW 2000 ARTHRITIS RHEUM 43: 978-987	784	OA	R, C, DB, P	**R12. 5/ 25**	DIC 50mg TDS	52	
	RR of Pts developing a clinical important increase in SBP/ DBP (trials comparing **R** & C)	RCTs	Healthy volunteers	COLLANTES, E 2002 BMC FAM PRACT 3:10	871	RA	R, C, DB, P, MC	ETOR 90mg OD	PC / NAP 500mg BD	12	
		Parallel	No outcome described (BP)	EHRICH, EW 1999 J RHEUMATOL 26: 2438-47	219	OA	R, C, DB, P, MC	**R25 / 125**	PC	6	
		Published		EMERY, P LANCET 1999 354: 2106-2111	655	RA	R, C, DB, P, DD	C 200mg BD	DIC SR 75mg BD	24	
		English only		GEBA, GP 2001 16 06 ANN EUR CONG RHEUM PRAGUE	1082	OA	R, C, P	**R25** / C200mg OD	PC	6	
				HAWKEY, CJ 2003 GUT 52: 820-26	660	RA	R, DB, PC, P, MC	**R50**	PC / NAP 500mg BD	12	
				HUNT, RH 2003 ALIMENT PHARMACOL THER 17: 201-210	742	OA,RA	R, DB, P, PC	ETOR 120mg OD	PC / NAP 500mg BD	12	
				LEUNG, AT 2002 CURR MED RES OPIN 18: 49-58	501	OA	R, DB, P, PC	ETOR 60mg OD	PC / NAP 500mg BD	12	

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring
Aw, T-J. et al (2005) Arch. Int. Med. 165: 1-7				LISSE, JR 2003 ARCH INT MED 139: 539-46	5557	OA	R, C, B, P, MC	**R25**	NAP 500mg BD	12	
				MATSUMOTO, AK 2002 J RHEUMATOL 29: 1623-30	816	RA	R, DB, P, PC	ETOR 90mg OD	PC / NAP 500mg BD	12	
				SCHNITZER, TJ 1999 CLIN THER 21: 1688-1702	658	RA	R, DB, P, PC	**R5/25/50**	PC	8	
				SILVERSTEIN, FE 2000 JAMA 284: 1247-55	8059	OA,RA	R, C, DB, P, MC	C 400mg BD	IBU 800mg TDS / DIC 75mg BD	24	
				SIMON, LS JAMA 1999 282: 1921-8	1149	RA	R, DB, P, PC, MC	C 100mg/200mg/400mg BD	PC / NAP 500mg BD	12	
				SOWERS, J 2003 20 06 AM COLL RHEUM LISBON CONF	268	OA,DM, HT	R, C, DB, P	**R25** / C 200mg OD	NAP 500mg BD	6	
				WHELTON, A 2001 AM J THER 8: 85-95	810	OA,HT	R, C, DB, P, MC	**R25**	C 200mg OD	6	
				WHELTON, A 2001 ANN EUR CONG RHEUM PRAGUE	13274	OA	R, C, DB, MC	C 200mg/day or 400mg/day	DIC 100mg/day/ NAP 1000mg/day	NA	
				WHELTON, A 2002 AM J CARDIOL 90: 959-963	1092	OA,HT	R,C,DB	**R25**	C 200mg OD	6	
				WHITE, WB 2002 HYPERTENSION 39: 929-934	178	Arhtritis/ HT	R, DB, P, PC	C 200mg BD	PC	4	
Total Pt					**45451**						

Key; WMD: Weighted mean difference, SBP: Systolic Blood Pressure, DBP: Diastolic Blood Pressure, BP: Blood Pressure, DM: Diabetes mellitus, HT: Hypertension, RR: Relative Risk, NA: Not Available.

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring
Daniels, B. et al (1999) Arthritis & Rheum. 42 SUPPL S143, 435 [A]	Incidence of thromboembolic CV A/Es in pts treated with PC, **R**, & NSAIDs	OA develop-ment progra-mme		1	NS	OA	NS	**R**	NS		
	Analysis of deaths			2	NS	OA	NS	**R**	NS	Max. Duration 86, Mean duration 22	
				3	NS	OA	NS	**R**	NS		
				4	NS	OA	NS	**R**	NS		
				5	NS	OA	NS	**R**	NS		
				6	NS	OA	NS	**R**	NS		
				7	NS	OA	NS	**R**	NS		
				8	NS	OA	NS	**R**	NS		
				9	NS	OA	NS	**R**	NS		
Total Pt					**5943: 3595R, 1565 NSAIDs, 783PC**						

Key; 1-9: indicates the number of clinical trials included, CV: Cardiovascular

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring
FDA	Incidence of thromboembolic CV A/Es in pts treated with R			1							
				2							
				3							
				4							
				5							
				6							
				7							
				8							
				9							
				10							
				11							
				12							
				13							
				14							
				15							
				16							
				17							
				18							
				19							
				20							
				21							
				22							
				23							
				24							
				25							
Total Pts					**28349**						

Key; 1-25: indicates the number of clinical trials included, CV: Cardiovascular

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring*
Garner, S. E. et al (2005) The Cochrane Database of Systematic Reviews, Issue 1, CD005115	Efficacy & safety of **R** in OA manage-ment by systematic review of available evidence	RCT	Unpubli-shed	ACEVEDO, E 2001 SCAND J RHEUMATOL 30:19-24	483: R12.5 242, DIC+MIS 241	OA HIP/ KNEE / SPINE	R, DB, DD, MC	**R12.5**	DIC 50mg +MIS 200mg BD	6	5,B
		Published		BIANCI, M 2003 DRUGS 63 SUPPL 1: 37-46	30	OA KNEE	R, DB, SC, Cross-Over	**R25**	NIM 100mg OD / C 200mg OD	3 (7 day cross over)	4,B
		Prospecti-ve		CANNON, GW 2000 ARTHRITIS & RHEUM 43(5): 978-987	784: R 12.5 259, R25 257, DIC 268	OA HIP/ KNEE	R, DB, DD, MC (USA)	**R12,5/25**	DIC 50mg TDS	52	5,B
		Parallel		DAY, RO 2000 ARCH INT MED 160 (12): 1781-87	809: R12.5 244, R25 242, IBU 249	OA HIP/ KNEE	R, DB, DD, PC, MC	**R 12.5/25**	PC / IBU 800mg TDS	6	5,A
		Accepted Outcome Measure I		EHRICH, EW 1999 J RHEUMATOL 26 (11): 2438-447	219-	OA KNEE	R, DB, PC, MC (USA)	**R25/125**	PC	6	5,A
		Any language		EHRICH, EW 2001 AM J MANAG CARE 7 (6): 609 616	672: R5 149, R12.5 144, R25 137, R50 97, PC 145	OA HIP/ KNEE	R, DB, PC, MC (USA)	**R 5/12.5/25**	PC	6	3,B
		Any age		GEBA, GP 2001 AM GERIATR SOC 49: S126 [A] (WEAVER, AL 2006 J. CLIN. RHEUMATOL. 12 (1): 17-25)	978: R12.5 390, NAB 392, 196PC	OA KNEE	R, DB, DD, PC, MC (USA)	**R12.5**	PC / NAB 500mg BD	6	N/A, D
		Any sex		GEBA, GP 2002 JAMA 287 (1): 64-71	381	OA KNEE	R, DB, DD, PC, MC (USA)	**R12.5/25**	C 200mg OD/ PAR 1g QDS	6	5,B

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring
Garner, S. E. et al (2005) The Cochrane Database of Systematic Reviews, Issue 1, CD005115				GEBA, G 2003 EULAR HYPER	1578: 12.5R 259, 25R 527, 200C 523, PAR 269	OA KNEE	R, DB	**R12.5/25**	C 200mg OD/ PAR 1g QDS	6	N/A, D
				GIBOFSKY, A 2003 ARTHRITIS & RHEUM 48 (11): 3102-3111	475: R25 190, C200 189, PC 96	OA KNEE	R, DB, DD, PC, MC	**R25**	PC / C200mg OD	6	5,B
				HAWKEY, C 2000 ARTHRITIS & RHEUM 43 (2): 370-77	775: R25 195, R50 193, IBU 193, PC 194	OA	R, DB, PC, MC	**R 25/50**	PC / IBU 800mg TDS	16-24	3,B
				HERRERA, JA 2003 AM J THER 10(6): 468-72	114	OA KNEE	R,DB, SC	**R25**	NIM Retard 300mg OD	4 (30days)	3,B
				KIVITZ, AJ 2004 J AM GERIATR SOC 52: 666-674	1042: R12.5 424, NAB 410, PC 208	OA KNEE	R, DB, DD, PC, MC (USA)	**R12.5**	PC / NAB 1000mg OD	6	5,A
				LAINE, L 1999 GASTROENTEROLOGY 117 (4): 776-783	742: R25 195, R50 186, IBU 184, 177PC	OA	R, DB, DD, PC, MC (USA)	**R 25/50**	PC / IBU 800mg TDS	12 (16-24)	4,A
				LISSE, JR 2003 ARCH INT MED 139: 539-546	5557: R25 2799, NAP 2787	OA	R, DB, DD, MC	**R25**	NAP 500mg BD	12	5,B
				McKENNA, F 2001 J CLIN RHEUM 7: SUPPL 3: 151-159	182: R25 59, C200 63, PC 60	OA KNEE	R, DB, DD, PC, MC (USA)	**R25**	PC / C 200mg OD	6	4,B

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring
Garner, S. E. et al (2005) The Cochrane Database of Systematic Reviews, Issue 1, CD005115				MOSKOWITZ, RW 2000 EULAR FR10277	530: R25 208, VAL 212, PC 110	OA KNEE	R, DB, DD, PC, MC (USA)	**R25**	PC / VAL 10mg OD	2	N/A,B
				MYLLYKANGAS-LUOSUJARVI, R 2002 SCAND J RHEUM 31: 337-344	2 STUDIES (944); A:482: R12.5 242 + NAP 240, B: 462: R12.5 229 + NAP 233	OA HIP/ KNEE	R, DB, DD, MC	**R12.5**	NAP 500mg BD	6	5,B
				NICCOLI, L 2002 CLIN & EXP RHEUM 20: 201-7	90	OA HIP/ KNEE / HAND	B, SC	**R25**	DIC150 / AMG2	2	3,B
				SAAG, K 1998 ARTHRITIS & RHEUM 41 SUPPL 9: A242	736: R12.5 219, R25 227, IBU 221, PC 69	OA HIP/ KNEE	R, DB, DD, PC, MC	**R12.5/25**	PC / IBU 800mg TDS	6	N/A,B
				SAAG, K 2000 ARCH FAM MED 9 (10): 1124-34	693: R12.5 231, R25 232, DIC 230	OA HIP/ KNEE	R, DB, DD, MC	**R12.5/25**	DIC 50mg TDS	52	5,B
				SCHNITZER, TJ 2001 EULAR A	1082	OA HIP/ KNEE	DB,PC	**R25**	PC / C 200mg OD	6	N/A,B
				SOWERS, J 2003 AM J HYPERTENSION 16 SUPPL (5,2):11A	404: R25 138, C200 136, NAP 130	OA	R, DB, MC (USA)	**R25**	C 200mg OD/ NAP500mg BD	12	N/A,B
				TRUITT,K 2001 AGING CLIN EXP RES 13: 112-21	341	OA HIP/ KNEE	R, DB?, PC, MC (USA)	**R12.5/25**	PC / NAB 1500mg OD	6	5,B

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring
Garner, S. E. et al (2005) The Cochrane Database of Systematic Reviews, Issue 1, CD005115				WHELTON, A 2002 AM J MANAG CARE 8 (15 SUPPL): S371-82	810: R25 399, C200 411	OA HIP/ KNEE / HAND	R, DB, DD, MC	**R25**	C 200mg OD	6	5,A
				WHELTON, A 2002 AM J CARDIOL 90 (9): 959-63	1092: R25 543, C200 549	OA HIP/ KNEE	R, DB, DD, MC	**R25**	C 200mg OD	6	5,B
Total Pt					**21543**						

Key; Accepted Outcome Measure I:OMERACT, WOMAC, HAQ, LEQUESNE INDEX, Physician global assessment and quality of life, *: Jadad 1996 and Schulz 1995 assessment tools were used, AMG2: Amtolmetin guacyl 600mg BD for 3 days and then 600mg /day, DB?: Unsure whether it is DB, N/A: Not applicable.

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring*
Garner, S. E. et al (2006) The Cochrane Database of Systematic Reviews, Issue 1, CD003685	Efficacy & toxicity of **R** for treating RA	RCTs	Healthy volunteers	BOMBARDIER, C 2000 NEJM 343 (21): 1520-8	8076: R50 4047,NAP 4029	RA	R#, DB, MC	**R50**	NAP 500mg BD	52	4,B
		Parallel	Acute Pain	SCHNITZER, TJ 1999 CLIN THER 21 (10): 1688-1702	658: 158R5, 171R25, 161R50, 168PC	RA	R, DB, PC, MC (USA)	**R5/25/50**	PC	8	3,B
		Open trials inlcuded only if results could be explained with respect to inherent bias	Concomitant use of intra-articular corticosteroids								
		RA	<50pts /arm								
		Outcome Measures: ACR, OMERACT	<4weeks in duration								
		Any age Any sex									
Total Pt						8734					

Key; *: Jadad 1996 and Schulz 1995 assessment tools were used, R#: Randomised from 2 to 52 weeks, ACR: American College of Rheumatology, OMERACT: Outcome Measures for Rheumatoid Arthritis Clinical Trials.

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring
Geba, P. G. et. al 2003 AJH 16 (5): Part 2, 39A [A]	Hypertension A/Es in pts treated with **R** or C versus Par			GEBA, GP 2002 JAMA 287 (1): 64-71	381: R12.5 95, R25 95, C200 94, PAR 97	OA KNEE	R, DB, P	**R12.5/25**	C 200mg OD/ PAR 1g QDS	6	
				SCHNITZER, T. J. 2005 J. RHEUMATOL 32: 1093-1105	1579: R12.5 260, R25 527, C200 523, PAR 269	OA KNEE	R, DB, P	**R12.5/25**	C 200mg OD/ PAR 1g QDS	6	
					1960: R12.5 355, R25 622, C200 620, PAR 363						
Total Pt											

Key;

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring
Gertz, B. J. et al 2002 Curr Med. Res. Opin. 18 (2): 82-91	Renovascular safety profile of **R** versus NSAIDs (Phase IIB/III clinical develop-ment program)	OA develop-ment program (1995-98)	Not comply with ACR criteria	Merck 010	219: R25 73, R125 74, PC 72	OA	R, DB, PC	**R25/125**	PC	6	
		RCT	R doses <12.5mg & >25mg	Merck 029	672: R5 149, R12.5 144, R25 137, R50 97, PC 145	OA	R, DB, PC	**R5/12.5/ 25/50**	PC	6	
		DB	NAB trial was not included in analysis (1 only)	Merck 033	736: R12.5 219, R25 227, IBU 221, PC 69	OA	R, DB, PC	**R12.5/25**	PC / IBU 800mg TDS	6	
		PC?		Merck 034	693: R12.5 231, R25 232, DIC 230	OA	R, DB	**R12.5/25**	DIC 50mg TDS	24	
				Merck 035	784: R12.5 259, R25 257, DIC 268	OA	R, DB	**R12.5/25**	DIC 50mg TDS	24	
				Merck 040	809: R12.5 244, R25 242, IBU 249, PC 74	OA	R, DB, PC	**R12.5/25**	PC / IBU 800mg TDS	6	
				Merck 044	742: R25 195, R50 186, IBU 184, PC 177	OA	R, DB, PC	**R25/50**	PC / IBU 800mg TDS	24	

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring
Gertz, B. J. et al 2002 Curr Med. Res. Opin. 18 (2): 82-91				Merck 045	775: R25 195, R50 193, IBU 193, PC 194	OA	R, DB, PC	**R25/50**	PC / IBU 800mg TDS	24	
				Merck 058	341: R12.5 118, R25 56, NAB 115, PC 52	OA	R, DB, PC	**R12.5/25**	PC / NAB 1500 OD	6	
					5771: R5 149, R12.5 1215, R25 1614, R50 476, R125 74, 847 IBU 800 TDS, DIC 50 TDS 498, NAB 1500 OD 115, PC 783						

Total Pt

Key; PC?: Alhtough stated all 9 trials were PC in the analysis two of them (034, 035) had no PC arm, ACR: American College of Rheumatology

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring
Goldstein, J. L et al 2005 J. Rheumatol 32: 111-7	GI tolerability of C & **R** in elderly hypertensive pts with OA (with or without co-administration of Aspirin (<326mg)	RCT	On Aspirin >325MG	WHELTON, A 2001 AM J THER 8: 85-95	810: R25 399, C 200 411	OA HIP / KNEE/ HAND	R, DB, MC	**R25**	C 200mg OD	6	
		DB	C >200mg OD	WHELTON, A 2002 J CARDIOL 90: 959-63	1092: R25 543, C 200 549	OA HIP / KNEE/ HAND	R, DB, MC	**R25**	C 200mg OD	6	
		ELDERLY >66 YOA	R <25mg, >25mg OD								
	[Each trial was designed to assess cardiorenal safety apart from GI tolerability]	OA (KNEE, HIP, HAND)	Esophageal, gastric, pyloric channel / duodenal ulceration prior 30 days of inclusion								
		On Hypertensive med.	Active GI Dis., also using antiulcer med. (antacids, H2, PPIs, PGS)								
Total Pt					**1902: R25 942, C 200 OD 960**						

Key; YOA: Years of age, GI Dis: Gastrointestinal disease, Med.: Medications, H2: H2 receptor antagonists, PPIs: Proton Pump Inhibitors; PGs: prostaglandins,

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring
Juni, P. et al 2004 JAMA 364 (9450): 2021-9	Assess robustness of evidence on A/Es of **R** prior withdrawal.	RCT	<4 weeks duration	EHRICH, EW 1999 H RHEUMATOL 26: 2438-47	145: R25 73, PC 72	OA	R, DB, PC	**R25**	PC	6	Adequate allocation
	Fatal / Non fatal AMI	Active or PC	Alzheimer's Disease$	LAINE, L 1999 GASTROENTEROL 117 (4): 776-83	742: R25 195, R50 186, PC 177, IBU 184	OA	R, DB, PC	**R25/50**	PC / IBU 800mg TDS	24	
	Fatal / Non fatal strokes (Thromb & Haem) & CV mortality (including deaths from unknown causes)	DB?	Colon Adenoma$	SCHNITZER, TJ 1999 CLIN THER 21: 1688-702	500: R25 171, R50 161, PC 168	RA	R, DB, PC	**R25/50**	PC	8	
	Composite outcomes of serious CV events (Non fatal AMI, Non fatal iscaemic haem. stroke, death from a vascular cause, death from unknown cause)	Quality: Allocation Concealment, external review of events	TRUITT 2001 entension: No CV information	EXTENSION TO SCNITZER: SCNITZER, TJ 1999 CLIN THER 21: 1688-702	544: R25 235, R50 223, NAP 86	RA	R, DB	**R25/50**	NAP 500mg BD	44	
		OA	R <12.5mg, >50mg OD	BOMBARDIER, C 2000 NEJM 343: 1520-28	8076:R50 4047, NAP 4029	RA	R, DB	**R50**	NAP 500mg BD	UP TO 56	
		RA		CANNON, GW 2000 ARTHRITIS RHEUM 43 (5): 978-87	784:R12.5 259, R25 257, DIC 268	OA	R, DB	**R12.5/25**	DIC 50mg TDS	52	

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring
Juni, P. et al 2004 JAMA 364 (9450): 2021-9		Low Back Chronic Pain		DAY, R 2000 ARCH INT MED 160 (12): 1781-87	809: R12.5 244, R25 242, IBU 249, PC 74	OA	R, DB, PC	**R12.5/25**	PC / IBU 800mg TDS	6	Adequate allocation
				HAWKEY, C 2000 ARTHRITIS RHEUM 43 (2): 370-7	775: R25 195, R50 193, IBU 193, PC 194	OA	R, DB, PC	**R25/50**	PC / IBU 800mg TDS	24	
				SAAG, K 2000 ARCH FAM MED 9: 1124-34	736: R12.5 219 ,R25 277, IBU 221, PC 69	OA	R, DB, PC	**R12.5/25**	PC / IBU 800mg TDS	6	
				SAAG, K 2000 ARCH FAM MED 9: 1124-34	693: R12.5 231, R25 232, DIC 230	OA	R, DB	**R12.5/25**	DIC 50mg TDS	52	
				EHRICH, EW 2001 AM J MANAG CARE 7: 609-16	523: R12.5 144, R25 137, R50 97, PC 145	OA	R, DB, PC	**R12.5/25/50**	PC	6	
				UNPUBLISHED EHRICH EXTENSION EHRICH, EW 2001 AM J MANAG CARE 7: 609-16	438: R12.5 102, R25 146, R50 100, DIC 90	OA	R, DB	**R12.5/25/50**	DIC 50mg TDS	26	
				GEBA, GP 2001 J. AM GERIATR SOC 49: S126 [A] (WEAVER, AL 2006 J. CLIN. RHEUMATOL. 12 (1): 17-25)	978: R12.5 390, PC 196, NAB 392	OA	R, DB, PC	**R12.5**	PC / NAB 500mg BD	6	

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring
Juni, P. et al 2004 JAMA 364 (9450): 2021-9				TRUITT, KE 2001 AGING CLIN EXP RES 13:112-21	341: R12.5 118, R25 56, NAB 115, PC 52	OA	R, DB, PC	**R12.5/25**	PC / NAB 1500mg OD	6	Adequate allocation
				UNPUBLISHED EXTENSION OF TRUITT. TRUITT, KE 2001 ARTHRITIS RHEUM 44: S369	673: R12.5 335 ,R25 114, NAP 224	RA	R, DB	**R12.5/25**	NAP 500mg BD	40	
				GEUSENS, PP 2002 SCAND J RHEUMATOL 31: 230-38	1058: R25 315, R50 297, NAP 147, PC 299	RA	R, DB, PC	**R25/50**	PC / NAP 500mg BD	12	
				UNPUBLISHED EXTENSION OF GEUSENS. GEUSENS, PP 2002 SCAND J RHEUMATOL 31: 230-38	893: R25 253, R50 392, NAP 248	RA	R, DB	**R25/50**	NAP 500mg BD	40	
				HAWKEY, CJ 2003 GUT 52: 820-6	660: R50 219, NAP 220, PC 221	RA	R, DB, PC	**R50**	PC / NAP 500mg BD	12	
				KATZ, N 2004 CURR MED RES OPIN 20 (5): 651-658	690: R25 233, R50 229, PC 228	CRONIC LOW BACK PAIN	R, DB, PC	**R25/50**	PC	4	Adequate allocation
				LISSE, JR 2003 ANN INT MED 139: 539-46	5586: R25 2799, NAP 2787	OA	R, DB	**R25**	NAP 500mg BD	12	

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring
Juni, P. et al 2004 JAMA 364 (9450): 2021-9				KIVITZ, AJ 2004 J AM GERIATR SOC 52: 666 74	1042: R12.5 424, NAB 410, PC 208	OA	R, DB, PC	**R12.5**	PC / NAB 1000mg OD	6	
					27595: R12.5 2279, R25 6412, R50 6258, IBU 800 TDS 847, NAP 500 BD 7890, DIC 50 TDS 588, NAB 500 BD 392, NAB 1000 OD 410, NAB 1500 OD 115, PC						
Total Pt					**2404**						

Key; AMI: Acute Myocardial Infarction, Thromb.: Haem.: CV: Cardiovascular, $: FDA files are unavailble

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W) !	Quality Scoring
Konstam, M. A. et al 2001 Circulation 104: 2280-8	Pooled CV safety analysis of all relevant data of R till 09/2000	Merck data	< 4WKS Duration	PHASE IIB DOSE FINDING	634	RA	NS	**R25/50**	PC / NAP	104	
	Assess the RR of CV thrombotic events among pts rendomised to R compared to PC, NSAIDs (DIC, IBU, NAB) & NAP	PHASE IIB TO V	R <12.5 mg	PHASE III: DOMESTIC USA [TRUITT, KE 2001 ARTHRITIS RHEUM 44: S369]	909	RA	NS	**R12.5/25/50**	PC / NAP	52	
		All completed & unblinded in early Septe-mber 2000.	Healthy Volunteers	PHASE III: INTERNATIONAL [GEUSENS, PP 2002 SCAND J RHEUMATOL 31: 230-38]	1058	RA	NS	**R25/50**	PC / NAP	52	
		APTC composite outcome	Head to head comparisons with C	PHASE III: ENDOSCOPY [HAWKEY, CJ 2003 GUT 52: 820-6]	660	RA	NS	**R50**	PC / NAP	13	
		Alzheimer Trials	Events after 14 days period after completion of study	PHASE IIB/III OA STUDIES [BOMBARDIER, C 2000 NEJM 343: 1520-28]	8076	RA	NS	**R50**	NAP	36	
				PHASE IIB/III OA STUDIES	5505	OA	NS	**R12.5/25/50**	PC / DIC / IBU / NAB	86	
				BONE METABOLISM STUDY	305	OA	NS	**R25**	PC / IBU	60	
				NABUMETONE STUDY 1 [KIVITZ, AJ 2004 J AM GERIATR SOC 52: 666-74]	1042	OA	NS	**R12.5**	PC / NAB	6	

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W) !	Quality Scoring
Konstam, M. A. et al 2001 Circulation 104: 2280-8				NABUMETONE STUDY 2 [GEBA,GP 2001 J AM GERIATR SOC 49: S126]	978	OA	NS	**R12.5**	PC / NAB	6	
				NAPROXEN STUDY	481	OA	NS	**R12.5**	NAP	6	
				ARTHROTEC STUDY	483	OA	NS	**R12.5**	DIC	6	
				ADVANTAGE [LISSE, JR 2003 ANN INT MED 139: 439-46]	5556	OA	NS	**R25**	NAP	12	
				ALZHEIMER PREVENTION [COMBINED DATA FROM 8 STUDIES]	1406	ALZHEIMER	NS	**R25**	PC	112	
				ALZHEIMER TREATMENT [AISEN, PS 2003 JAMA 289: 2819-26]	682	ALZHEIMER	NS	**R25**	PC	48	
				PHASE III CHRONIC LOW BACK PAIN [KATZ, N 2004 CURR MED RES OPIN 20 (5): 651-658 (2 STUDIES)]	380	CRONIC LOW BACK PAIN	NS	**R25/50**	PC	4	ADEQ CONCEAL
				PHASE III CHRONIC LOW BACK PAIN [KATZ, N 2004 CURR MED RES OPIN 20 (5): 651-658 (2 STUDIES)]	310	CRONIC LOW BACK PAIN	NS	**R25/50**	PC	4	ADEQ CONCEAL
Total Pt					**28465**						

Key; !: Planned duration of patient follow-up, Comparators doses were not stated in the publication, ADEQ CONCEAL: Adequate concealment of allocation.

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring*
Moore, R. A. 2005 Arthritis Research & Therapy 7: R644-665	Tolerability and A/Es in clinical trials of C in OA & RA	RCTs	<2 weeks duration	C-002	396: C 200 OD 136, R25 132, NAP 500 BD 128	OA	R, DB	C 200 OD	**R25** / NAP 500mg BD	12	5, 16
		DB	Open label extensions	C-003	475: C 200 OD 189, R25 190, PC 96	OA	R, DB	C 200 OD	**R25** / PC	6	5, 16
		Any C dose	<18YOA	C-010	524	OA	R, DB	C 200 OD	PAR 1g QDS	6	5, 16
		Any co-mparator	>Aspirin 326mg	C-013	291	OA	R, DB	C 40/100/200 BD	PC	2	5, 16
		All completed by December 2003	Analgesics in general & PAR up to 2g/day for max. of 3 days but not within 48h of arthritis assessment	C-020	1092	OA	R, DB	C 50/100/200 BD	PC / NAP 500mg BD	12	5, 16
		Symptoma-tic OA / RA of 3 months duration prior to entry.	Antibiotics for H. pylori eradication, Metronida-zole, Lithium, Anticoagu-lants	C-021	1214	OA	R, DB	C 50/100/200 BD	PC / NAP 500mg BD	12	5, 16

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring*
Moore, R. A. 2005 Arthritis Research & Therapy 7: R644-665			Anti-ulcer med (PPIs, H2, anta-cids, sucra-lfate, & misoprostol	C-042	687	OA	R, DB	C 200 OD	DIC 100mg/day	6	5, 16
				C-047	401	OA	R, DB	C 50/200/800 OD	PC	4	5, 16
				C-054	1060	OA	R, DB	C 100/200/ 400 OD	PC / NAP 500mg BD	12	5, 16
				C-060	684	OA	R, DB	C 100 BD/ C 200 OD	PC	6	5, 16
				C-087	715	OA	R, DB	C 100 BD/ C 200 OD	PC	6	5, 16
				C-096	13194	OA	R, DB	C 100/200 BD	NAP 500mg BD / DIC 50mg BD	12	5, 16
				C-118	598	OA	R, DB	C 200 OD	PC / DIC 50mg TDS	6	5, 16
				C-149	810: C 200 OD 411: R25 399	OA	R, DB	C 200 OD	**R25**	6	5, 16
				C-152	182: C 200 OD 63, R25 59, PC 60	OA	R, DB	C 200 OD	**R25** / PC	6	5, 16
				C-181	1092: C 200 OD 549, R25 543	OA	R, DB	C 200 OD	**R25**	6	5, 16
				C-209	316	OA	R, DB	C 200 OD	PC / NAP 500mg BD	6	5, 16
				C-210	362	OA	R, DB	C 200 OD	PC / NAP 500mg BD	6	5, 16
				C-211	315	OA	R, DB	C 200 OD	NAP 500mg BD	6	5, 16

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring*
Moore, R. A. 2005 Arthritis Research & Therapy 7: R644-665				C-216	959	OA	R, DB	C 200 BD	PC / LOXO 60mg TDS	4	5, 16
				C-249	977	OA	R, DB	C 200 OD	PC / PAR 1g QDS	2 x 6 crossover	5, 16
				C-012	327	RA	R, DB	C 40/100/ 200 BD	PC	4	5, 16
				C-022	1149	RA	R, DB	C 100/200/ 400 BD	PC	12	5, 16
				C-023	1102	RA	R, DB	C 100/200/ 400 BD	PC / NAP 500mg BD	12	5, 16
				C-041	655	RA	R, DB	C 200 BD	DIC SR 75mg BD	24	5, 16
				C-062	536	OA/ RA	R, DB	C 200 BD	NAP 500mg BD	12	5, 16
				C-071	1097	OA / RA	R, DB	C 200 BD	IBU 800mg TDS / DIC 75mg BD	12	5, 16
				C-102	7968	OA/ RA	R, DB	C 400 BD	IBU 800mg TDS / DIC 75mg BD	52	5, 16
				C-105	657	OA / RA	R, DB	C 100 BD	DIC 50mg BD	12	5, 16
				C-106	124	OA/ RA	R, DB	C 100 BD	DIC 50mg BD	12	5, 16
				C-107	88	OA / RA	R, DB	C 100 BD	DIC 50mg BD	12	5, 16
Total Pt					**39605**						

Key; *: Quality by Jadad 1996 (1st number), Validity by Smith 2000 (2nd number) scales respectively , YOA: Year of age, H. pylori: Helicobacter pylori infection, Med: medications, H2: H2 receptor antagonists, PPIs: Proton Pump Inhibitors, LOXO: Loxoprofen

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs #	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring*
NICE				VIGOR (see Bombardier in NICE update)	NS	RA	NS	**R**	NS	NS	NS
	Overall efficacy of rofecoxib, celecoxib, etodolac, meloxicam	UK licensed daily doses of active drug for efficacy	Lower than licensed doses for safety of active drug assessment	SCHNITZER, T. J. 1999 CLIN THER 21 (10): 1688-702	658	RA	R, DB, MC (USA)	**R5/25/50**	PC	8	3
	Total number of A/Es reported	Higher doses allowed only for safety assess-ment		SAAG, K 1998 ARTHRITIS RHEUM 41 (SUPPL 9): A242	736	OA	R, DB	**R12.5/25**	IBU 800mg/day / PC	6	NC
	Total number of GI A/Es reported			TRUITT, KE 1999 XIV EULAR CONGRESS, GLASGOW, [A]	341	OA	R, DB, MC (USA)	**R12.5/25**	NAB 1500mg OD / PC	6	NC
	Total numbers of withdrawals through A/Es reported			HAWKEY, C 2000 ARTHRITIS RHEUM 43 (2): 370-7	775	OA	R, B, MC	**R25/50**	IBU 800mg TDS / PC	24	5
	Total numbers of PUBs			CANNON, GW 2000 ARTHRITIS & RHEUM 43(5): 978-987	784	OA	R, DB, MC (USA)	**R12.5/25**	DIC 50mg TDS	52	4
	Separate analysis for NSAIDs and PC			DAY, R 2000 ARCH INT MED 160 (12): 1781-87	809	OA	R, DB, MC	**R12.5/25**	IBU 800mg TDS / PC	6	3
	Cost-effectiveness			GEBA, G 1999 ARTHRITIS RHEUM 42 (SUPPL 9): A440	1042	OA	R, DB	**R12.5**	NAB 1000mg OD / PC	6	NC
				LAINE, L 1999 GASTROENTEROL 117 (4): 776-83	742	OA	R, DB	**R25/50**	IBU 800mg TDS / PC	24	5
				EHRICH, E W 1997 ARTHRITIS RHEUM 40 (SUPPL 9): A330	672	OA	R, DB, MC (USA)	**R5/12.5/25 /50**	PC	6	NC
				EHRICH, E W 1999 J RHEUMATOL 26 (11): 2438-47	219	OA	R, DB, MC (USA)	**R25/125**	PC	6	5
Total					**6778**						

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs #	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring*
NICE update	As above		<50 pts in each arm	BOMBARDIER, C 2000 NEJM 343: 1520-28	8076	RA	R, DB, MC	**R50**	NAP 500mg BD	52	
Inlcudes all the above trials plus these			<4 weeks duration	ACEVEDO, E 2001 SCAND J RHEUMATOL 30:19-24	693	OA	R, DB, MC	**R12.5/25**	DIC 50mg TDS	52	
			Other pain than OA, RA	GEBA, G A 2000 EULAR [A]	382	OA	R, DB, MC (USA)	**R12.5/25**	C 200mg OD / PAR IG QDS	6	
				LAURENZI 2000 a EULAR [A]	482	OA	R, DB, MC (USA)	**R12.5**	NAP 500mg BD / NAB 1000mg/day	6	
				LAURENZI 2000 b EULAR [A]	483	OA	R, DB, MC (USA)	**R12.5**	DIC 50mg + MIS 200mg BD	6	
				WHELTON, A 2000 EULAR [A]	810	OA	R, DB, MC (USA)	**R25**	C 200mg OD	6	
				McKENNA, F 2000 EULAR [A]	182	OA	R, DB, MC (USA, UK)	**R25**	C 200mg OD / PC	6	
Total Pt					**17886**						

Key; #: Only Rofecoxib RCTs included, *: Quality by Jadad 1996 while also quality assessment was based on the poportion of patients that were lost to follow-up, NICE: National Institute of Clinical Excellence (UK), NC: Not calculable,PUB: Perforation, Ulceration & Bleeding.

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring
Reicin, A. S. 2002 Am J. Cardiol. 89: 204-9	To assess the comparative CV thrombogenic risks of specific COX-2 inhibitors with that of non selective NSAIDs	OA deve-lopment program	Events after the 14 days period of trial completion	1 EHRICH, EW 2001 AM J MANAG CARE 7: 609-16 [JUNI /LANGMAN 029]	NS	OA	R,DB	**R?**	IBU 800mg TDS / DIC 50mg TDS?	NS	
		RCTs	Other indications	2 TRUITT, KE 2001 AGING CLIN EXP RES 13:112-21 [058]	NS	OA	R,DB	**R12.5/25**	NAB 1500mg OD	6	
		DB	Non Merck	3	NS	OA	R,DB	**R?**	IBU 800mg TDS / DIC 50mg TDS?	NS	
		PC	R 5mg dose	4	NS	OA	R,DB	**R?**	IBU 800mg TDS / DIC 50mg TDS?	NS	
			Aspirin & Anticoagu-lants users	5	NS	OA	R,DB	**R?**	IBU 800mg TDS / DIC 50mg TDS?	NS	
				6	NS	OA	R,DB	**R?**	IBU 800mg TDS / DIC 50mg TDS?	NS	
				7	NS	OA	R,DB	**R?**	IBU 800mg TDS / DIC 50mg TDS?	NS	
				8	NS	OA	R,DB	**R?**	IBU 800mg TDS / DIC 50mg TDS?	NS	

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring
Reicin, A. S. 2002 Am J. Cardiol. 89: 204-9											
					5435, R 3357: R12.5 1209, R25 1603, R50 545, NSAIDS 564: IBU 847, DIC 590, NAB						
Total Pt					**127**						

Key; CV: Cardiovascular, ?Dosing and frequency of comparators and active drug not clearly stated in published article.

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring
Schnitzer, T. J. 2005 J Rheumatol. 32: 1093-105	To compare the efficacy in OA taking PAR1g QDS, C200mg OD, **R**12.5mg or **R**25mg	Non Pregnant	Concurrent medical disease or arthritic disease	GEBA, GP 2002 JAMA 287:64-71 [VACT-1]	1960-1578 =382: R12.5 96, R25 95, C200 97, PAR 94	OA KNEE	R,DB,P, MC(29 SITES IN USA)	R12.5, 25	C200OD/ PAR1gQD S	6	
	Outcome Measures: PGART, WOMAC	Older than 40 YOA	Abnormal LAB clinical significant results	SNITZER, TJ 2005 J RHEUMATOL 32(6): 1093-105	1578: R12. 5 259, R25 527, C200 523, PAR 269	OA KNEE	R,DB,P, MC (97 SITES IN USA)	R12.5, 25	C200OD/ PAR1gQD S	6	
	To compare the safety in OA taking PAR1g QDS, C200mg OD, **R**12.5mg or **R**25mg	Pain disability for MIN. of 6 minutes	Allergies: ASA, sulfonami-de, NSAIDs								
	To confirm the results of VACT 1 trial in a larger patient population using the same clinical endpoints	Functional class I, II, III	Pregnancy								
	Incidence of overall A/Es, Serious A/Es, Drug related A/Es leading to DC / edema or hypertenion related A/Es , or to DC / congestive heart failure or GI A/Es	Using NSAIDs, COX-2, PAR to control pain & disability	<40YOA								
		OA of the knee only									

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring
Schnitzer, T. J. 2005 J Rheumatol. 32: 1093-105		VAS <80mm pain score on flat surface walking (visit 1) Washout period >5 t1/2 i.e. 4-15 days MIN. VAS >40mm pain on flat surface walking (visit 2) Worse-ning of 1 point in Likert scale (0-4) IGADS (visit 2) PAR pts were allowed to use PAR as rescue (only worsening of IGADS)									

Table 5. 10 Summary of systematic reviews / meta-analyses included

Systematic review / Meta-analysis	Outcomes / Aims	Inclusion Criteria	Exclusion Criteria	Trials/RCTs	Patient Number	Indication	Design	Drug	Compara-tors	Duration (W)	Quality Scoring
Schnitzer, T. J. 2005 J Rheumatol. 32: 1093-105					**1960: R12.5 355, R25 622, C200 620, PAR 363**						
Total Pt											

Key; VAS: Visual Analogue Scale, PGART: Patient Global Assessment of Response to Therapy, WOMAC: Western Ontario & McMaster Universitites Index, YOA: Years of age, pts: patients, IGADS: Investigators Global Assessment of Disease Status, t1/2: Half-life, MIN: minimum, LAB: laboratory, DC: Discontinuation.

KEY (Table 5.10): R: Rofecoxib, C: Celecoxib, ETOR: Etoricoxib, VAL: Valcecoxib, NAP: Naproxen, IBU: Ibuprofen, DIC: Diclofenac, MIS: Misoprostol, NAB: Nabumetone, NIM: Nimesulide, LOXO: Loxoprofen, PAR: Paracetamol, PC: Placebo, NSAID: Non-steroidal anti-inflammatory drugs, CV: Cardiovascular, GI: Gastrointestinal, RCTs: Randomised Clinical Trial(s), R (Design): Randomised, C (Design): Controlled, PC (Design): Placebo-controlled, DB: Double-blind, B: Blind, P (Design): Parallel, DD: Double dummy, MC: Multicenter, USA: United States of America, OD: Once a day, BD: Twice a day, TDS: Three times a day, QDS: Four times a day, W: Weeks, Pt(s): Patient(s), ACR: Americal College of Rheumatology, WOMAC: Western Ontario & McMaster Universitites Index, OMERACT: Outcomes Measures in Rheumatology, HAQ: Health Assessment Questionnaire, OA: Osteoarthritis, RA: Rheumatoid arthritis, A/Es: Adverse events, NS: Not stated, NC: Not Calculable, [A] or A: Abstract, YOA: Years of Age, NICE: National Institute of Clinical Excellence, FDA: Food and Drug Administration (USA). Only the name of the first author is stated.

Table 5.11 Correlations between rofecoxib's RCTs and the included systematic reviews / meta-analyses

RCTs	Total Patient number	Aw, T-J et al 2005	Daniels, B. et al 1999 [A] ^	FDA^	Garner, S. E. et al 2005 CD005115	Garner, S. E. et al 2005 CD003685	Geba, G 2003 [A]	Gertz, B. J. 2002	Goldstein, J. L. et al 2005	Juni, P. et al 2004	Konstam, M. A. et al 2001	Moore, R. A. et al 2005	Mukherjee, D. et al 2001	NICE / NICE update	Reicin, A. S. et al 2002^	Schnitzer, T. J. et al 2005
BOMBARDIER, C 2000 NEJM 343:1520-8	8076	•				•				•	•		•	•		
CANNON,GW 2000 ARTHRITIS RHEUM 43: 978-987	784	•			•			•		•				•		
EHRICH, EW 1999 J RHEUMATOL 26: 2438-47	219	•			•			•		•				•		
GEBA,GP 2001 16 06 ANN EUR CONG RHEUM PRAGUE	1082	•			•											
HAWKEY,CJ 2003 GUT 52: 820-26	660	•								•	•					
LISSE,JR 2003 ARCH INT MED 139: 539-46	5557	•			•					•	•					
SCHNITZER,TJ 1999 CLIN THER 21: 1688-1702	658	•				•				•				•		
EXTENSION TO: SCNITZER, TJ 1999 CLIN THER 21: 1688-702	544									•						
SOWERS, J 2003 20 06 AM COLL RHEUM LISBON CONF (1) / SOWERS, J 2003 AM J HYPERTENSION 16 SUPPL (5,2):11A (2)	268 /404	•(1)			•(2)											
WHELTON,A 2001 AM J THER 8: 85-95	810	•			•				•			•				
WHELTON, A 2002 AM J CARDIOL 90: 959-963	1092	•			•				•			•				

Table 5.11 Correlations between rofecoxib's RCTs and the included systematic reviews / meta-analyses

RCTs	Total Patient number	Aw, T-J et al 2005	Daniels, B. et al 1999 [A] *	FDA*	Garner, S. E. et al 2005 CD005115	Garner, S. E. et al 2005 CD003685	Geba, G 2003 [A]	Gertz, B. J. 2002	Goldstein, J. L. et al 2005	Juni, P. et al 2004	Konstam, M. A. et al 2001	Moore, R. A. et al 2005	Mukherjee , D. et al 2001	NICE / NICE update	Reicin, A. S. et al 2002*	Schnitzer, T. J. et al 2005
ACEVEDO,E 2001 SCAND J RHEUMATOL 30:19-24	483				•						•					
BIANCI, M 2003 DRUGS 63 SUPPL 1: 37-46	30/31				•											
DAY, RO 2000 ARCH INT MED 160 (12): 1781-87	809				•			•		•				•		
EHRICH, EW 2001 AM J MANAG CARE 7 (6): 609-616	672				•			•		•				•		
UNPUBLISHED EXTENSION TO: EHRICH, EW 2001 AM J MANAG CARE 7: 609-16	438									•						
GEBA, GP 2001 AM GERIATR SOC 49: S126 [A] (WEAVER, AL 2006 J. CLIN. RHEUMATOL. 12 (1): 17-25)	978				•					•	•		•			
GEBA, GP 2002 JAMA 287 (1): 64-71	381				•		•									•
GEBA, G 2003 EULAR HYPER	1578				•		•									•
GIBOFSKY, A 2003 ARTHRITIS & RHEUM 48 (11): 3102-3111	475				•							•				
HAWKEY, C 2000 ARTHRITIS & RHEUM 43 (2): 370-77	775				•			•		•				•		

Table 5.11 Correlations between rofecoxib's RCTs and the included systematic reviews / meta-analyses

RCTs	Total Patient number	Aw, T-J et al 2005	Daniels, B. et al 1999 [A] *	FDA*	Garner, S. E. et al 2005 CD005115	Garner, S. E. et al 2005 CD003685	Geba, G 2003 [A]	Gertz, B. J. 2002	Goldstein, J. L. et al 2005	Juni, P. et al 2004	Konstam, M. A. et al 2001	Moore, R. A. et al 2005	Mukherjee, D. et al 2001	NICE / NICE update	Reicin, A. S. et al 2002*	Schnitzer, T. J. et al 2005
HERRERA, JA 2003 AM J THER 10(6): 468 72	114				•											
KIVITZ, AJ 2004 J AM GERIATR SOC 52: 666-674	1042				•					•	•		•	•		
LAINE, L 1999 GASTROENTEROLOGY 117 (4): 776-783	742				•			•		•				•		
McKENNA, F 2001 J CLIN RHEUM 7: SUPPL 3: 151-159	182				•											
MOSKOWITZ, RW 2000 EULAR FRI0277	530				•											
MYLLYKANGAS-LUOSUJARVI, R 2002 SCAND J RHEUM 31: 337-344	944; A: 482, B: 462				•						•					
NICCOLI, L 2002 CLIN & EXP RHEUM 20: 201-7	90?				•											
SAAG, K 1998 ARTHRITIS & RHEUM 41 SUPPL 9: A242	736				•			•		•				•		
SAAG, K 2000 ARCH FAM MED 9 (10): 1124-34	693				•			•		•						
TRUITT,K 2001 AGING CLIN EXP RES 13: 112-21	341				•			•		•	•			•		
UNPUBLISHED EXTENSION TO: TRUITT, KE 2001 ARTHRITIS RHEUM 44: S369	673									•	•					

Table 5.11 Correlations between rofecoxib's RCTs and the included systematic reviews / meta-analyses

RCTs	Total Patient number	Aw, T-J et al 2005	Daniels, B. et al 1999 [A] *	FDA*	Garner, S. E. et al 2005 CD005115	Garner, S. E. et al 2005 CD003685	Geba, G 2003 [A]	Gertz, B. J. 2002	Goldstein, J. L. et al 2005	Juni, P. et al 2004	Konstam, M. A. et al 2001	Moore, R. A. et al 2005	Mukherjee, D. et al 2001	NICE / NICE update	Reicin, A. S. et al 2002*	Schnitzer, T. J. et al 2005
GEUSENS, PP 2002 SCAND J RHEUMATOL 31: 23038	1058									•	•					
UNPUBLISHED EXTENSION TO: GEUSENS, PP 2002 SCAND J RHEUMATOL 31: 23038	893									•						
KATZ, N 2004 CURR MED RES OPIN 20 (5): 651-658	690									•	•					
PHASE IIB DOSE FINDING	634										•					
PHASE IIB/III OA STUDIES	5505										•					
BONE METABOLISM STUDY	305										•					
ALZHEIMER PREVENTION [COMBINED DATA FROM 8 STUDIES]	1406										•					
ALZHEIMER TREATMENT [AISEN, PS 2003 JAMA 289: 2819-26]	682										•					
C-002	396											•				
C-152	182											•				
Total Patient Number	44343	45451 (19206)	5943	28349	21543	8734	1960	5771	1902	27595	28465	39605 (2945)	18061 (10096)	(6778 / 17886)	5435	1960

Key; RCTs: Randomised Controlled Trial(s). *: Published articles did not provide with full details to identify the RCTs utilised , ( ): Only rofecoxib patients.

5.1.2.8 Population characteristics in the included systematic reviews or meta-analyses

Table 5.12 summarises the available information concerning the age, sex and ethnic characteristics of the population included in the systematic reviews or meta-analyses analysed in this study. In some cases, it was not possible to calculate a total mean or median age of the included patients, because information was given as a percentage of patients aged older or younger than 66 years old. Thus, due to the lack of information and diversity in the types of reporting, it was decided not to present a total about the age of the whole population.

The percentage of female patients - in all systematic reviews and meta-analyses that provided gender information - was always greater (almost in a female to male ratio of 2.5:1). This is coherent with the gender distribution of patient in the majority of rofecoxib's RCTs.

Only three systematic reviews or meta-analyses (Gertz, Krupa et al. 2002; Reicin, Shapiro et al. 2002; Moore, Derry et al. 2005a) out of the 15 included in this study provided information about the ethnicity of the population included inspite of the importance of ethnicity in practise.

Table 5.12 Population characteristics of subjects analysed in the included sustematic reviews / meta-analyses

	Age (years)		Sex (%)		Race (%)				
Systematic reviews / Meta-analyses	Mean (SD)	Median (range)	Females *	Males*	Cauca-sian	Asian	Black	Hispa-nic	Other
Aw, T-J. et al 2005		60 (51.7-74)	71 (44-82)				NS		
Daniels, B. et al 1999 [A]	62 (NS)		NS				NS		
FDA	NS	NS							
Garner, S. E. et al 2005 CD005115		62.9 (61.3-83)	NS				NS		
Garner, S. E. et al 2005 CD003685		56.5 (55-58)	78.85 (78-79.7)				NS		
Geba, G 2003 [A]		62	68		87				
Gertz, B. J. et al 2002#	NC		73.44	26.56	80.88	0.43	5.05	12.38	1.26
Goldstein, J. L. et al 2005	73.6 (NS)		63.84				NS		
Juni, P. et al 2004	NS	NS	NS				NS		
Konstam, M. A. et al 2001	NS	NS	RA: 79.61, OA: 71.21, AD/LBP: 44.84						
Moore, R. A. et al 2005		>60 (17-96)	70.65		48.90	0.92	0.41	0.17%	NS
Mukherjee, D. et al 2001	NS	NS	NS				NS		
NICE		NC	NC				NS		
Reicin, A. S. et al 2002*3	NC	63 (38-94)	73				NS		
Schnitzer, T. J. et al 2005		62 (39-93)	67.55	32.45	87.24		6.73	4.59	1.43

Key; *: Median (range), #: Mean of 5072 patients (rofecoxib 5mg, 50mg and 125mg arms were excluded), NS: Not Stated, NC: Not able to be calculated by the data available in the systematic review/meta-analysis, *3: 197 patients were doubled counted (total patients number: 5632).

5.1.2.9 Industrial funding of included systematic reviews and meta-analyses

Only two out of the fifteen systematic reviews and meta-analyses reported no conflict of interest (Table 5.13) [(Mukherjee, Nissen et al. 2001; Garner, Fidan et al. 2005b)]. Five of the analysed systematic reviews and meta-analyses were sponsored by manufacturer's of COX-2 inhibitors [3 by Merck Inc, Co. (Daniels and Seidenberg 1999; Reicin, Shapiro et al. 2002; Schnitzer, Weaver et al. 2005), and 2 by Pfizer / Pharmacia (Goldstein, Bello et al. 2005; Moore, Derry et al. 2005a)]. Some or all of the

Table 5.13 Funding Sources & Financial Conflicting Interests declared in the included systematic reviews / meta-analyses

Meta-analysis / Systematic Review	Funding & Conflicting Interests: Sponsored by Manufacturer	Authors employed by Manufacturer	Government Support	University Support	None	Funding / Conflict of Interests
Aw, T-J. et al 2005		Pfizer, Merck, Novartis				NHMRC Public Health Postgraduate Research Scholarship ID No 237059, Royal Australasian College of Physicians Postdoctoral Fellowship, Some of the authors have been consultants for Pfizer, Merck, Novartis
Daniels, B. et al 1999 [A]	Merck					Authors are employees of Merck
FDA			USA			No sources of commercial support
Garner, S. E. et al 2005 CD005115					•	No sources of support
Garner, S. E. et al 2005 CD003685			NICE (UK)*	University of Ottawa (CANADA)*		Multiple sources of external funding (GMSG-Transnational) and also internal support from NICE (UK) and Institute for Population Health, University of Ottawa (Canada)
Geba, G 2003 [A]	NS	Merck	NS	NS		Authors are employees of Merck
Gertz, B. J. et al 2002		Merck				Authors are employees of Merck
Goldstein, J. L. et al 2005	Pfizer	Pfizer / Pharmacia				Authors are employees of Pfizer
Juni, P. et al 2004			Swiss National Science Foundation (Switzerland)			Swiss National Science Foundation's National Research Programme 53 (Grant Number: 3200-066278.01 and 405340-104762). Authors report no conflict of interest
Konstam, M. A. et al 2001		Merck / Pfizer				Authors are employees of Merck
Moore, R. A. et al 2005	Pfizer *1	Pfizer				One author is a Pfizer employee - Unrestricted educational grant from Pfizer to support this work
Mukherjee, D. et al 2001					•	No sources of support
NICE			UK			No sources of commercial support
Reicin, A. S. et al 2002	Merck	Merck				Funded by Merck
Schnitzer, T. J. et al 2005	Merck	Merck				Authors are employees of Merck - Merck supported this work

Key; NS: Not stated, *: Internal sources of support, *1: Financial support from Pfizer included freedom for authors to reach their own conclusions, and an absolute right to publish the results of their research, irrespective of any conclusions reached -Pfizer had the right to view the final manuscript before publication, and did so, NICE: National Institute of Clinical Excellence.

authors of eight of the published systematic reviews or meta-analyses were employed or had previously been employed by manufacturer's of COX-2 inhibitors (Konstam, Weir et al. 2001; Gertz, Krupa et al. 2002; Reicin, Shapiro et al. 2002; Geba, Polis et al. 2003; Aw, Haas et al. 2005; Goldstein, Bello et al. 2005; Moore, Derry et al. 2005a; Schnitzer, Weaver et al. 2005). Finally, four out of the fifteen were conducted by government organisations or received funding / support by governements (United Kigdom (UK), United States of America (USA), Switzerland) [(FDA 2001; NICE 2001; Juni, Nartey et al. 2004; Garner, Fidan et al. 2005a)].

Chapter 6

Discussion

6.1 Preface

The present study is a systematic review of all the systematic reviews and meta-analyses available on September 2005 relating to the cardiotoxicity of the COX-2 inhibitor rofecoxib. From the 47 published articles that were identified only fifteen systematic reviews or meta-analyses fulfilled the inclusion criteria. The aim was to summarise the available evidence and postulate possible reasons why rofecoxib's cardiotoxicity was not identified earlier. As part of this study, a quality of reporting assessment was carried out for all the included systematic reviews and meta-analyses.

Since millions of elderly people take selective and nonselective NSAIDs for their analgesic and anti-inflammatory benefits (FitzGerald and Patrono 2001), there remain many questions about the CV safety of coxibs as well as nonselective NSAIDs, particularly in older adults (Solomon, Avorn et al. 2006).

Rofecoxib had total sales of US $4.5 billion in 2001 and was the most prescribed arthritis pain medication across Europe, Canada & Latin America (Merck & Co. 2002). The fourth Vioxx ® (rofecoxib) case to be heard in the US concluded on the 5th of April 2006 with a jury from New Jersey, the home state of the manufacturer, delivering a split decision (Booth 2006). The jury concluded that Vioxx ® was the cause of plaintiff John McDarby's heart attack and ordered the company to pay US

$4.5 million in compensation (Booth 2006). However, the jury decided not to award compensation to the second plaintiff Thomas Cona, who claimed he used rofecoxib for just under two years before his heart attack, but had only 3 prescriptions for rofecoxib over that time (Booth 2006). This is the second time that a jury has awarded punitive damages in cases concerning rofecoxib (Tanne 2006). The first was a case in Texas, in which the widow of a 59 year old man who died of an arrhythmia (Tanne 2005). The jury awarded his widow US $253.4 million (US $24.4 million for economic loss and emotional anguish and US $229 million in punitive damages) (Tanne 2005). However, the award was reduced under state law to about US $26.1 million and Merck appealed (Tanne 2005). Merck faces nearly 13,000 cases in the US relating to rofecoxib. Nearly half a million people in the UK have taken rofecoxib, and several hundred are considering suing (Tanne 2006).

From 1999 to September 2004, an estimated 106.7 million rofecoxib prescriptions were dispensed in the US, of which 17.6% were high-dose (greater than 25mg per day) (Graham, Campen et al. 2005). In two Merck-sponsored RCTs (Bombardier, Laine et al. 2000; Bresalier, Sandler et al. 2005), relative risks for acute myocardial infarction of 5 for high-dose rofecoxib and 2 for the standard dose were recorded (Graham, Campen et al. 2005). The background rate for acute myocardial infarction among control groups from studies of CV risk in NSAID users varied from 7.9 per 1000 person-years in CLASS (Silverstein, Faich et al. 2000) to 12.4 per 1000 person-years in TennCare (Graham, Campen et al. 2005). Using the relative risks from the above-mentioned randomised clinical trials and the background rates seen in NSAID risk studies, an estimated 88,000-140,000 excess cases of serious coronary heart disease probably occurred in US over the market-life of rofecoxib (McAlister,

Straus et al. 2000; Graham, Campen et al. 2005). The US national estimate of case-fatality rate (fatal myocardial infarction plus sudden cardiac death) was 44% [American Heart (Association 2003)], which suggests that many of the excess cases attributed to rofecoxib use were fatal (Graham, Campen et al. 2005).

Rofecoxib is an example of a drug that was very successfully marketed and prescribed for thousands of patients without applying the necessary caution that is imperative for all new drugs and especially for those for which there is limited information about their efficacy and safety over existing medication. Whether the cardiovascular and cerebrovascular toxicity associated with rofecoxib is a class effect of all selective COX-2 selective inhibitors or specific to rofecoxib remains unclear as the mechanism of these adverse-effects remains unknown. These unanswered questions await further research.

6.2 Reasons for late recognition of rofecoxib's CV toxicity

After the largest prescription-drug withdrawal from the market in history, a need to identify the potential reasons for its late withdrawal was apparent to the research and medical community in order to learn from this block-buster drug withdrawal and avoid future repetition of the same mistakes. Although the reasons why rofecoxib's CV toxicity was not identified earlier are still widely debated, it is relevant to identify some major points.

6.2.1 Quality of reporting of systematic reviews and meta-analyses assessing rofecoxib's CV safety

The assessment of the quality of systematic reviews and meta-analses is intertwined with the quality of reporting, that is the extent to which a systematic review or meta-analysis provides information about the inclusion and exclusion criteria, the sources utilised, the abstraction process, the design, conduct and analysis of the individual RCTs as well as a providing a single estimate of the size of treatment effect and a test of homogeneity in the estimate of effect size. If the results of systematic reviews are to be used by health care providers and health care consumers, it is necessary that they are as free as possible from bias (i.e. systematic error). Although the available data addressing quality of reporting are sparse, it appears that a scientific report is a reasonable marker for how the project was conducted. In an assessment of the quality of meta-analyses in major depressive disorder through the use of the QUOROM checklist, the overall quality of reporting was 50.2% (Hemels, Vicente et al. 2004). Past quality analyses suggested that quality results of 56% were "embarassing" (Narine, Yee et al. 1991; Squires 1991). In a follow-up analysis 3 years later, the same group concluded that there is "still need for improvement" with scores of 57% for non-structured abstracts and 74% for structured abstracts (Squires 1994; Taddio, Pain et al. 1994). No other quality of reporting comparable data were possible to be identified for systematic reviews or meta-analyses.

Despite quality guidelines (Moher, Cook et al. 1999), the present study has shown that the average quality of published systematic reviews and meta-analyses for the cardiovascular and renal safety of rofecoxib is only acceptable [63.13% (21.41%),

see chapter 5, Table 5.3, p139]. The quality scores in this analysis were considered acceptable based on the pre-study classification. Only four of the included systematic reviews or meta-analyses received an overall grade of "good" quality (>75%) in the 18-item QUOROM checklist. This checklist was designed to provide authors of systematic reviews and meta-analyses with objective criteria to ensure that the reporting of their analysis is performed in a manner which results in a transparent report upon which to base conclusions. Great emphasis is given on the *Abstract* section of the published article (6 out of the 18 items of the checklist). The *Abstract* section received an acceptable overall score of 69.70% (see chapter 5, Table 5.3, p139) that was lower than the overall score for the *Introduction* (81.80%) and *Methods* (71.21%) sections. However, the *Abstract* should be a microcosm of the article as a whole through the use of a structured format (Moher, Cook et al. 1999). The structured format item of the checklist received an overall score of 72.73% (see chapter 5, Table 5.3, p139). Notably, the selection criteria utilised QUOROM checklist item of the *Abstract* section received a score of 36.36%. Clearly, the trials included in systematic reviews or meta-analyses should ideally be relevant and carefully selected to be of high methodological quality and free of bias such that the differences in outcomes observed between groups of patients can confidently be attributed to the intervention under investigation. Additionally, the marginally acceptable reporting of the databases and sources utilised for information in the *Abstract* section (54.55%) is of note. Although in the *Methods* section most authors described in great detail the sources utilised (90.91%), it was not considered to be of crucial importance to include in the *Abstract* section or the word limit did not allow authors to go into detail. Similar are the findings for the reporting of the selection criteria between the *Abstract* section (36.36%) and the *Methods* section (72.73%). This is also visible in the separate

analysis of the 2 included abstracts. Only one of them followed a structured format and reported the sources utilised, while both did not report the methodology utilised adequately to provide replication. All of the above illustrate a weakness in reporting adequately the methodology of the systematic review or meta-analysis in the *Abstract* section, which is a crucial tool for the original screening of eligible published articles. When the full version of a paper is not always available or great resources would be required to screen all the available published papers in that area of research, the abstract is the only information a researcher may be able to access freely. The abstract is always the first point of contact with the research reported for most and especially busy practitioners and healthcare professionals, so great care needs to be taken in the quality of its reporting.

The poor quality of reporting identified in the *Title* (27.30%) and *Results* sections (45.46%) is alarming. Only 3 out of the 11 published systematic reviews and meta-analyses and neither of the 2 abstracts analysed separately stated in the *Title* section of the published paper that the paper is a systematic review or a meta-analysis. Unlike the QUOROM checklist, almost none of the scales previously available (see chapter 3, Table 3.2, p112) included a question on the *Title*, the *Introduction*, or the *Abstract* (Shea, Dube et al. 2001). Although, in developing the QUOROM checklist the supporting scientific evidence was sometimes indirect, the QUOROM group judged this as a reasonable approach, because further evidence about the merits of indentifying in the *Title* that this published article is a systematic review or meta-analysis are pending (Moher, Cook et al. 1999).

The overall quality score for presenting a trial flow diagram in the *Results* section was 18.18% in this analysis (see chapter 5, Table 5.3, p139). Apart from the QUOROM checklist, no scale has previously suggested producing a trial flow diagram in the *Results* section (Shea, Dube et al. 2001). If a systematic method of searching the available sources was utilised, it is reasonably easy to construct this flow diagram and can be of great value when other research groups are trying to update the search or to compare their results with that of another group. As "fugitive" literature cannot be directly assessed, it provides a measure of the validity of the search to the reader if the process used by the authors to include studies throughout the review process was described. However, although the nessecity of this flow diagram, or at least the reporting of the excluded trials along with the reasons, is clear, they are still largely unreported. Notably, even the two included Cochrane systematic reviews, that were of high reporting quality, did not illustrate this process with a trial flow diagram, although they included a description of the inclusion and exclusion criteria. Furthermore, although descriptive data for each RCTs included in the systematic reviews or meta-analyses was reported in an acceptable manner (72.73%), the report of the quantitative data synthesis was rated as poor (45.45%). Quantitative data synthesis is the overall aim of a meta-analysis and when appropriately conducted should form the basis of clinical decisions. Thus, excellent reporting is required to allow for an unambiguous interpretation of the synthesised data.

The quality of the *Discussion* section was acceptable (63.60%), providing with a relative acceptable summary of findings, while interpreting the results in light of the totality of available evidence. However, there is still room for improvement especially if the results for the *Discussion* section of this analysis is compared with the good

quality of reporting of the *Discussion* section in other research areas (Hemels, Vicente et al. 2004).

The only section where the overall quality of reporting was rated as good was the *Introduction* (81.80%) signifying that the description of the clinical problem identified requiring to perform the systematic review or meta-analysis is good.

Although the overall results of this analysis are more promissing than results previously reported in other research areas as mentioned above, there still remains a great need for adherence to standardised reporting to improve the quality of reporting, especially of systematic reviews or meta-analyses focusing on adverse-effects. Safety aimed systematic reviews or meta-analyses are increasingly becoming more important in risk-benefit analysis performed by policy makers and healthcare professionals for the provision of care due to the increased costs of treatment (especially with marginally more effective new agents versus older alternatives) combined with the unwillingness of manufacturers to perform large safety aimed RCTs.

However, it is possible that the quality of reporting identified for the included systematic reviews or meta-analsyses may be attributed to limitations put forth by the journal of publication, i.e. word limits or preset abstract structure, rather than lack of quality in reporting by the authors of the systematic review or meta-analysis. Nevertheless, it has to be mentioned that the structured format of the *Abstract* is not necessarily endangered by the word limit (Mulrow, Thacker et al. 1988). Furthermore, the availability today of peer-reviewed electronic journals allows for an in depth

reporting of research outcomes with fewer limitations in style and word limit than traditional published journals.

It is also relevant to note that the QUOROM checklist was designed to assess the quality of reporting and not necessarily the quality of the research performed. Studies performed at the highest degree of scientific merit may have been graded poorly as a result of inadequate reporting style. (Hemels, Vicente et al. 2004) Thus, a poor quality score does not necessarily imply that inappropriate research was perfomed, but that authors are in need of improved reporting of (presumambly) excellent research. (Hemels, Vicente et al. 2004)

Finally, a limitation of the analysis in the present study is the lack of any assessment of inter-rater reliability as only one reviewer (myself) undertook this task.

6.2.2 Sources of information retrieval and abstraction process

Nine of the 15 CV safety systematic reviews and meta-analyses utilised only manufacturer's data files (see chapter 5, Table 5.5, p143). Multiple reasons can explain this finding. Firstly, manufacturer's data files are more detailed than published RCTs and usually provide all the required data for a systematic review or meta-analysis. Secondly, the drug company especially for a newly marketed agent like rofecoxib would be aware of all available RCTs (published or unpublished) that had been performed or were still ongoing. A combination of the above factors necessitates ties between those conducting the systematic review or meta-analysis and the pharmaceutical industry. Five out of these 9 systematic reviews or meta-analyses were

funded by manufacturers of COX-2 inhibitors (see chapter 5, Table 5.12, p187) and in 3 of the remaining four the authors were employed by manufacturers of coxibs. The FDA conducted meta-analysis could only be based on the data provided by the manufacturers, as this meta-analysis was performed by a licensing authority.

All of the remaining 6 included systematic reviews and meta-analyses that were analysed in this study performed a MEDLINE search (see chapter 5, Table 5.5, p143). Of these 6, only 4 supplemented the MEDLINE search with an EMBASE search. The majority of journals indexed in MEDLINE are published in USA, whereas EMBASE has a better coverage of european journals (Egger and Davey Smith 2001). It is usually important to examine both MEDLINE and EMBASE, because the overlap in journals covered by the two database is only 34% (Egger and Davey Smith 2001). The same four systematic reviews or meta-analyses were the only ones to also search the Cochrane Controlled Trials Register (CCTR), which currently includes over 250,000 records and is clearly the best single source of published trials for inclusion in systematic reviews or meta-analyses (Egger and Davey Smith 2001). Hand searching of the reference lists of the identified papers was performed by the majority of authors of the included systematic reviews or meta-analyses, while some attempts were made to identify "fugitive" literature. The latter varied from searching the World Wide Web (without specifying a method) to searching conference abstracts, bibliographies and government databases (with a vaguely reported method). It can be concluded that the majority of the systematic reviews or meta-analyses that did not base their analysis on the manufacturer's data files utilised a reasonable variety of data sources and at least attempted to identify fugitive literature, although not in a structured, clearly documented and reproducible way.

It is interesting to note that only in 3 out of the 9 systematic reviews or meta-analyses that utilised manufacturer's data files reported an end date for inclusion of RCTs. From the remaining 6 systematic reviews or meta-analyses, all of which reported an end date, only 4 included RCTs after 2000, the year when the results of the VIGOR trial were published, 3 of which reported an end date after May 2004 and were published after rofecoxib's withdrawal from the market. Thus, although the VIGOR trial results questioned the cardiovascular and cerebrovascular safety of rofecoxib, no adequate further attempts were made to summarise the available evidence in a cumulative manner or update these systematic reviews with the results of new RCTs.

Safety post-marketing trials normally required for pharmacovigilance did not take place for rofecoxib. Currently, licensing authorities need to mandate that a trial be performed in the post-marketing phase of a drug by confronting the manufacturer that the drug in question may be withdrawn from the market (Topol 2005). Manufacturers of coxibs were unwilling to initiate dedicated CV trials on their own accord (Topol 2005). With early results of coxibs trials that brought out their prothrombotic potential, rapid initiation of follow-up RCTs was absolutely necessary, especially when half of real world patients with arthritis suffer from coexisting CV disease (Cox, Frisse et al. 2004) (Topol 2005).

The abstraction process was not documented adequately (see chapter 5, Table 5.6, p144). Six out of the 15 systematic reviews or meta-analyses report to use independent reviewers. Furthermore, only one systematic review reports of using one blinded reviewer and only two of double checking the results of the abstraction process. Although blinding of already published articles can be demanding (e.g. even

style of the written text can provide information about the journal of publication to an experienced researcher), it aids significantly in the objectivity of the result. Double-checking of the results of the abstraction process is useful to eliminate potential errors introduced due to human error or misinterpratation. Data extraction is an important methodological step in any systematic review affecting directly the research outcome, but at least the reporting of it was inadequate especially in those systematic reviews or meta-analyses obtaining information directly from the manufacturer's data files. Only the Cochrane collaboration systematic reviews, contacted the authors of the RCTs included in their analysis for further clarification.

6.2.3 Underpowered systematic reviews and meta-analyses including short RCTs assesed the CV safety of COX-2 inhibitors versus comparators and placebo utilising composite endpoints

The present study identified only 15 systematic reviews or meta-analyses aimed at cardiovascular and renal safety of rofecoxib with an estimated total number of patient of 44343 (see chapter 5, Table 5.11, p181) including in total 39 RCTs. The event rate of CV adverse events observed for rofecoxib in APPROVe is smaller than 1 in a 1,000, and RCTs conducted for rofecoxib were not designed to pick up these adverse events. The 1994 International Drug Safety Standard for evaluating adverse events related to long-term use of a drug for a non-life threatening disease recommends that 1500 patients be treated during drug development and that 600 patients be treated for 6 months and 300 for 12 months. This should detect adverse events occuring in 1/300 to 1/500 patients (Ravaud and Tubach 2005). Although development programs often include larger numbers of patients, they are clearly

inadequate for reliably detecting adverse events seen with a rate of 1 / 10,000, particularly when the risk increase is small compared to the general population. Sample sizes of 20,000 to 80,000 would be needed to show the unexpected increase in cardiovascular events in populations without high-risk groups (Bombardier, Laine et al. 2000; Silverstein, Faich et al. 2000; Woodworth, Furst et al. 2001; Tugwell, Judd et al. 2005). VIGOR was the largest RCT conducted for rofecoxib and it contained only 8076 patients. Furthermore, to detect an increase in an adverse event that occurs in 0.1% of controls (the occurence rate of myocardial infarction in the VIGOR trial placebo arm) and 0.15% of treated patients (a 50% risk increase), 210,000 patients must be treated (Ravaud and Tubach 2005). However, this implies the necessity that all future coxib studies need to be adequately powered to detect cardiovascular differences, requiring a vast number of patients.

The ATPC (Antiplatelet Trialists' Collaboration) endpoint includes CV, haemorrhagic, and unknown deaths, nonfatal myocardial infarctions, and nonfatal strokes (Antithrombotic Trialists Collaboration 2002). This composite endpoint has been widely used to assess the overall CV impact of antithrombotic compounds, since it summarises the irreversibly morbid and fatal CV sequelae that may accompany therapy with antiplatelet agents (Weir, Sperling et al. 2003). However, the use of this composite cardiovascular endpoint could have diluted any increase risk of myocardial infarction (Juni, Nartey et al. 2004), explaining partly the difference between earlier meta-analysis (Konstam, Weir et al. 2001) and those published after rofecoxib's withdrawal. The relative risk of cardiovascular thrombotic events occuring with rofecoxib compared to nonselective NSAIDs (RR: 1.01, 95%CI: 0.59-1.77) is higher than relative risk of APTC composite endpoint (RR: 0.80, 95%CI: 0.3-1.7) and

remains higher for the comparison made to placebo (rofecoxib RR: 1.06, 95%CI: 0.34-3.23 versus placebo RR: 0.70, 95%CI: 0.16-4.17) in the rofecoxib development program (Weir, Sperling et al. 2003).

Although the review of the phase IIB/III OA program revealed no adverse CV safety signals, the theoretical concerns about the possible effects of selective COX-2 inhibition on the balance of vasoactive eicosanoids led Merck (in 1998) to introduce a new standard operating procedure (SOP) to assess CV safety in the rofecoxib development program (Weir, Sperling et al. 2003). In studies initiated after the SOP was introduced, investigators submitted all potential CV events for review by an external expert committee that remained blinded to treatment assignment (Weir, Sperling et al. 2003). Events were categorised by using prespecified case definitions as cardiac events (acute myocardial infarctions (AMI), unstable angina pectoris, sudden and / or unexplained death, resuscitated cardiac arrest, cardiac thrombus), peripheral vascular events (pulmonary embolism, peripheral arterial thrombosis, peripheral venous thrombosis), and cerebrovascular events (ischaemic cerebrovascular stroke, cerebrovascular venous thrombosis, transient ischaemic attack). CV events not confirmed as having a thrombotic cause were confirmed as nonthrombotic, haemorrhagic (included haemorrhagic cerebrovascular stroke), or deemed nonconfirmable as the result of insufficient data (Weir, Sperling et al. 2003). The adjudication process, thus, ensured uniform diagnoses of CV events among different trials, as well as improved diagnostic accuracy of investigator-reported events (Weir, Sperling et al. 2003). Adjudicated data were to be used for all prospectively defined analyses (after 1998), and adjudicated data were not available for the OA development program (8 phase IIB/III trials between 1995 to 1998) (Weir, Sperling et al. 2003).

However, Juni et al (Juni, Nartey et al. 2004) showed that the reported increase in risk was greater in trials with external endpoints committee (relative risk 3.9), suggesting that misclassification of coronary events could have biased results in trials that did not include external appraisal of safety outcomes (OA development program trials based on which rofecoxib received license worldwide). Thus, it is reasonable to conclude that the inclusion of an independent endpoints committee should be the rule, and exceptions to this rule should be justified (Juni, Nartey et al. 2004). The systematic review by Konstam et al (Konstam, Weir et al. 2001) compares adjudicated data with unadjudicated data basing the event rate with NSAIDs other than naproxen on short-term trials with a small number of events, which is flawed (Wright 2002). However, it is interesting to point out that the thromboembolic events in celecoxib's trials were not adjudicated by blinded observers (White, Faich et al. 2002; Wright 2002). A crucial lesson to be learned is that data on adverse events from industry-sponsored RCTs are trustworthy only if an independent endpoints committee is involved (Juni, Reichenbach et al. 2005).

Furthermore, it is worthwhile to mention that none of the coxibs trials reported the NNT but only the relative risk (Wright 2002). However, RR on its own has been shown to increase the willingness to use a new drug (Wright 2002).

6.2.3.1 Confounding by indication

Rofecoxib had received a license for the relief of pain and treatment of OA and RA and for acute pain and chronic back pain. However, available long RCTs with a placebo arm were available for other unlicensed indications like Alzheimer's disease

and various types of cancer. As far as safety issues are concerned ideally all indications have to be included. Only two (FDA 2001; Juni, Nartey et al. 2004) out of the 15 systematic reviews or meta-analyses (see chapter 5, Table 5.8, p146) have included chronic low back pain RCTs, one of which (FDA 2001) included also Alzheimer's disease (total number of systematic reviews or meta-analyses including Alzheimer's Disease RCTs was 2 in this analysis). The meta-analysis by Juni et al (Juni, Nartey et al. 2004) received much critisicm as the authors did not include the data from the 3 Alzheimer's Disease trials (Kim and Reicin 2005). However, these trials neither met the prespecified inclusion criteria, nor had the data presented on the FDA's website been reviewed by an independent endpoints committee. However, including only the trials for chronic musculoskeletal pain is reflecting the indications of the clinical use of rofecoxib in actual clinical practise.

Further, although the events reported in Alzheimer's studies have been included in a recent meta-analysis (Kearney, Baigent et al. 2006), the events reported in the early terminated (30th September 2004) ViP (a double-blinded, randomised, placebo-controlled trial evaluating the effects of rofecoxib 25mg compared with placebo in decreasing the risk incidence of capsular and extracapsular prostate cancer in men with initially elevated risk) and VICTOR (Vioxx® in Colorectal Therapy, a phase III randomised double-blind, placebo-controlled of rofecoxib in colorectal cancer patients following potentially curative therapy) trials have not been analysed in a meta-analysis.

6.2.3.2 Need for placebo trials and comparators with an established cardiovascular profile

Apart from the fact that there is still only some speculative mechanisms proposed for the CV toxicity of coxibs, the safety of traditional NSAID comparators have been questioned. Naproxen appears to be the only NSAID with some cardioprotective effect, although in the ADAPT trial naproxen was shown to increase CV (cardiovascular) and CB (cerebrovascular) events. On the other hand, ibuprofen appears to increase myocardial infarctions by approximately 10% based on the results of the TARGET trial that included both ibuprofen and naproxen (Farkouh, Kirshner et al. 2004; Topol 2005). Additionally, it has been convincingly shown that certain NSAIDs antagonise the irreversible platelet inhibition induced by aspirin (Catella-Lawson, Reilly et al. 2001; Konstantinopoulos and Lehmann 2005). Administration of ibuprofen, but not rofecoxib, acetaminophen or diclofenac, with or prior to enteric-coated aspirin negates the aspirin antiplatelet effect by binding to a serine 529 residue in the hydrophobic channel that aspirin must transverse before it can access platelet COX-1. (Catella-Lawson, Reilly et al. 2001; Konstantinopoulos and Lehmann 2005) Long-term placebo- and active-controlled trials are generally not available for the NSAIDs, with the exception of the trials where certain NSAIDs were used as active controls in studies of COX-2 selective drugs (Jenkins and Seligman 2005). The FDA has issued new supplemented labeling request letters for over-the-counter NSAIDs (15th July 2005) and is currently reviewing the CV safety of already licensed NSAIDs ((FDA) 2005a).

Baring the above in mind, rofecoxib was only compared with placebo, naproxen, ibuprofen, diclofenac, nabumetone and nimesulide as well as with paracetamol and celecoxib. As VIGOR did not contain a placebo arm, the interpretation of the adverse cardiovascular event could also be interpreted by a protective effect of naproxen. As the cardiovascular safety of the comparators (traditional NSAIDs) were not clearly established in RCTs, it would have been beneficial to have a placebo arm. However, this could be unethical as unecessary pain would be caused to the trial participants.

6.2.3.3 Duration of included RCTs

The average median duration of included RCTs in the analysed systematic reviews and meta-analyses was 6 weeks (6-13 weeks) [see chapter 5, Table 5.7, p145]. The shortest trial included was one week and the longest 86 weeks (see chapter 5, Table 5.7, p145). Rofecoxib was prescribed for long-term indications as OA or RA, and thus a median duration of 6 weeks is not adequate to assess the safety of this agent. A recently published meta-analysis of the risk of atherothrombosis with all COX-2 inhibitors including lumiracoxib (Kearney, Baigent et al. 2006) reported that out of 121 placebo controlled trials, nine only were long-term trials with one year or longer scheduled treatment (mean 139 weeks), while the remaining were shorter (mean 11 weeks). Around two thirds of the vascular events had occurred in the nine long-term trials (Kearney, Baigent et al. 2006).

When rofecoxib was withdrawn from the market, Merck used the preliminary results of the APPROVe trial to conclude that estimated cumulative incidence curves

for adjudicated serious thrombotic events in the rofecoxib and placebo groups were similar for approximately the first 18 months of treatment, and only diverged after this time. The estimated relative risk calculated with the use of Cox model represents a time-averaged hazard ratio and thus may not adequately describe the difference between the treatment and placebo groups when the proportional-hazards assumption does not hold, and one could not conclude from the data that a shorter course of rofecoxib is safe (Lagakos 2006). All the intention-to-treat analyses in a newly released report show that the confirmed thrombotic event curves begin to diverge much earlier, generally within four to six months, whereas the APTC event curves show divergence after only 3 months of exposure to rofecoxib (Nissen 2006). Furthermore, Juni et al reported an increased risk of myocardial infarction in trials of both short and long duration (Juni, Nartey et al. 2004).

6.2.3.4 Population characteristics

The population analysed by the systematic reviews and meta-analyses included in this study were fairly young (see chapter 5, Table 5.12, p186) compared to the people that actually were using rofecoxib in clinical practice mostly for the the relief and treatment of signs and symptoms of OA or RA. COX-2 prescribing was subject to "chanelling bias" in practice by policy makers. The increased cost of therapy with coxibs and the lack of superiority of efficacy of COX-2 inhibitors compared to traditional NSAIDs, imposed the need to policy makers to reserve these agents for people at an increased risk of GI complications. Age is considered a risk factor for GI complications. Also, a large group of patients hospitalised due to adverse GI events of NSAIDs are elderly. Thus, COX-2 inhibitors were reserved for elderly patients, a

group which also carries an increased risk for cardiovascular and renal morbidity and mortality. Furthermore, the incidence (1995, USA) of symptomatic hand, hip, and knee OA (identified using radiography and joint symptoms) increased with age and women had higher rates than men, especially after age 50 (Oliveria, Felson et al. 1995; Merck & Co. 2006). Around age 80, there was a leveling off or a decrease in the incidence of OA in both groups and all joint sites (Oliveria, Felson et al. 1995; Merck & Co. 2006). This is of course translated in an increase in the number of elderly patients treated with NSAIDs and thus most likely with a COX-2 inhibitor. Hence, the population of the cardiovascular and renal safety aimed systematic reviews or meta-analyses is younger and not descriptive of the population that was using rofecoxib in actual practice.

Although more women than men suffer from OA, the ratio is not as high as that illustrated in RCTs included in the systematic reviews or meta-analyses (~2.5:1 female to male ratio, see chapter 5, Table 5.12, p186). On the other hand, however, this ratio is meaningful for RA patients. RA has an earlier onset than OA, and women prior to menopause are affected three times more than men, although after the menopause the frequency of onset is similar between sexes (NICE 2001).

OA is worldwide in distribution, geographic and ethnic differences have been reported (Merck & Co. 2006). For example, the prevalence of hand and knee OA is similar among Europeans and Americans (Dequeker and Dieppe 1998). There is greater variation in the distribution of hip OA with markedly lower rates in African Blacks, Asian Indians, and Hong Kong Chinese (Dequeker and Dieppe 1998). In 3 out of the four systematic reviews or meta-analyses that reported the ethnicity of the analysed population (see chapter 5, Table 5.12, p186), more than 80% of the

participants were caucasians. Therefore, the outcomes of the systematic reviews or meta-analyses could be applicable in countries with a high percentage of caucasians, but they cannot be translated in clinical practice worldwide. Clinical determined differences in drug-metabolising enzyme activity can lead to a wide interindividual variability in drug response, resulting in altered efficacy or toxicity in the affected individuals. (Evans and Relling 1999)

6.2.3.5 Identified errors in the analysed systematic reviews or meta-analyses

Certain discrepancies or errors were identified in the published articles of the included systematic reviews or meta-analyses that are worthwhile to note.

In the meta-analysis by Aw et al (Aw, Haas et al. 2005) two errors were identified in the table providing characteristics of the 19 included RCTs meeting their inlcusion criteria. The phase II RCT (Schnitzer, Truitt et al. 1999) of rofecoxib that included 658 RA patients did not include a celecoxib arm. These patients were randomly allocated to receive placebo or rofecoxib 5mg, 25mg, or 50mg once daily. Furthermore, the Simon et al (Simon, Weaver et al. 1999) RCT for celecoxib apart from comparing celecoxib with placebo also included a naproxen 500mg twice daily arm that was missed in the table. Whether these errors were just publication errors or affect their analysis is unknown.

The Merck sponsored systematic review by Daniels et al (Daniels and Seidenberg 1999) that was reported only in abstract form reported safety data from the rofecoxib OA development program (9 RCTs) for all patients enrolled into placebo (N

=783), rofecoxib (N=3595) and NSAID (N=1565) treatment groups (total N: 5943). However, in the Cochrane systematic review of rofecoxib in RA (Garner, Fidan et al. 2005), it is mentioned that the Daniels et al (Daniels and Seidenberg 1999) systematic review analysed data from 7535 patients with OA, including 5943 patients from 9 RCTs plus unpublished data from 1592 patients. Thromboembolic CV event per 100 patients years reported in both publications were the same as well as the incidence of cardiac, central nervous and peripheral system events. However, in the abstract publication the inclusion of the unpublished data from 1592 patients is not mentioned.

In the second systematic review that was included in this analysis and was only reported in abstract format (Geba, Polis et al. 2003), an error considering the number of patients included in the VACT-1 and VACT-2 RCTs was observed. In the abstract publication of this systematic review, VACT-1 included 381 patients and VACT-2 included 1579 patients with OA of the knee (Geba, Polis et al. 2003). A total of 1960 patients randomly allocated to receive rofecoxib 12.5mg (N=355), rofecoxib 25mg (N=622), celecoxib 200mg (N=620) and acetaminophen 4000 mg (N=363) daily (Geba, Polis et al. 2003; Schnitzer, Weaver et al. 2005). Although the total number of patients included in each arm and in total is correct, the total number of patients in each trial separately is wrong. In the full publication of the VACT-1 trial a total number of patient was reported of 382 (Geba, Weaver et al. 2002). Furthermore, a more recent full publication of the pooled analysis of the VACT trials reports 382 in VACT-1 and 1578 in VACT-2 (Schnitzer, Weaver et al. 2005). Thus, there was clearly an error in the reported abstract of this pooled analysis. The full publication of the pooled analysis of the VACT studies (Schnitzer, Weaver et al. 2005) only became

available after rofecoxib was withdrawn from the market and 2 years later than the abstract was published.

6.2.3.6 Cumulative meta-analysis not part of Vioxx®'s pharmacovigilance

One identified reason (Juni, Nartey et al. 2004) why rofecoxib's adverse cardiovascular and cerebrovascular adverse events were not identified earlier is that the available RCTs with data on the CV safety of rofecoxib were not analysed in a cumulative manner. Cumulative meta-analysis provides a framework for updating the summary results from all relevant trials as evidence accumulates (Lau, Antman et al. 1992). A cumulative meta-analysis, was published just a month after rofecoxib's withdrawal from the market, indicated that an increased risk of myocardial infarction was evident from 2000 onwards and that this effect was both substantial and unlikely to be a chance finding (Juni, Nartey et al. 2004). An increased risk was evident in 2000, when 14247 patients had been randomised and 44 events had occurred (Juni, Nartey et al. 2004). At the end of 2000 (52 myocardial infarctions, 20742 patients) the relative risk was 2.30 (95% CI 1.22-4.33, P: 0.010). Subsequent trials only brought the number of events to 64 (21432 patients), narrowing the CI, while the point estimates remained similar (Juni, Nartey et al. 2004).

There was, however, much criticism of the results from this cumulative meta-analysis. The difference in myocardial infarction risk for rofecoxib regardless of comparator was driven by the difference between rofecoxib and a single comparator, naproxen, especially by the results of VIGOR (Merck & Co. 2004; Kim and Reicin 2005). Thus, in Merck's response, the meta-analysis of Juni et al violates the basic

principle of meta-analaysis to combine "like with like" and these conclusions are driven by their choice of method, involving pooling of results for placebo, non-naproxen NSAIDs (ibuprofen, diclofenac, nabumetone), and naproxen ignoring pharmacodynamic differences between naproxen and other NSAIDs, and placebo (Kim and Reicin 2005). Juni et al justified combining the data across the comparators, because CI against individual comparators were wide and the statistical test for interaction was not significant (Juni, Nartey et al. 2004). However, the limiting power of the heterogeneity test and the use of random-effects model (that decreases the probability of finding an interaction) does justify the combination of the comparator groups (Hardy and Thompson 1998; Lievre and Abadie 2005).

Additionally, this cumulative meta-analysis did not combine all available papers. In particular, 3 studies in Alzheimer's disease (AD) were not included, although being available and contributing 28 myocardial infarctions compared with a total of 64 in the cumulative meta-analysis (including 24 from VIGOR) (Merck & Co. 2004; Lievre and Abadie 2005). It has also to be noted that in the studied of AD, the population was in much higher risk of myocardial infarction (8.2/1000 patient-years in the placebo group) than did those included in Juni's meta-analysis (1.45/1000 patient-years in the control groups) (Lievre and Abadie 2005). However, the inclusion criteria of the meta-analysis by Juni and co-workers specified that only all RCTs in adult patients with chronic musculoskeletal disorders comparing rofecoxib 12.5-50mg daily with other NSAIDs or placebo would be considered (Juni, Nartey et al. 2004). AD, which is not a licensed indication for rofecoxib or other coxibs for that reason, was, therefore, left out. Furthermore, the increased risk of myocardial infarction associated with the use of rofecoxib in trials with an external endpoint committee is also evident

after the exclusion of VIGOR (pooled RR : 2.5, 95% CI 1.1-6.0) (Juni, Reichenbach et al. 2005).

Recently, a meta-analysis of RCTs focusing on renal and arrhytmia adverse events of COX-2 inhibitors (Zhang, Ding et al. 2006), following Juni's and co-workers' (Juni, Nartey et al. 2004). example, showed that these adverse events were evident by the end of the year 2000 that rofecoxib was associated with overall adverse renal events (P: <0.001; and for all subsequent years), as well as specific events of hypertension and peripheral oedema (P: <0.01 for both; and for all subsequent years).

Although it is known that cumulative meta-analysis is a valuable tool that can be a significant aid for timely and appropriate decision-making, it is not currently always used as part of the regular pharmacovigilance program established in the majority of countries. Relying on company performed meta-analysis because the company has access to all available data is not necessarily the best available practice. The FDA and other licensing authorities should review their procedures, and identify and remove the obstacles to making continuously updated summary information available to decision makers (Dieppe, Ebrahim et al. 2004).

6.3 Availability of data and publication bias

Another point highlighted after rofecoxib's withdrawal was the difficulty for independent researchers to access the original full trial results. Reporting bias may be particularly important for adverse-effects (Egger, Dickersin et al. 2001). Hemminki examined reports of clinical trials submitted by drug companies to licensing authorities

in Finland and Sweden and found that unpublished trials gave information on adverse effects more often than published trials (Hemminki 1980). Thus, in most of the cases independent researchers can only access the FDA available information or the already published reports of the trials.This clearly underscores the need for free access to the FDA full data files. In some instances important discrepancies were noted between published data and figures in FDA files. The VIGOR Study Group (Bombardier, Laine et al. 2000) reported a four-fold increased risk of myocardial infarction, whereas the figures available from the FDA files indicated a five-fold increase in risk (Juni, Nartey et al. 2004). In the published report of the APPROVe trial (Bresalier, Sandler et al. 2005) the methods section referred to the use of the logarithm of time. However, this description of the method used for the report of the p-value (p-value = 0.01) for the test of proportionality of hazards was in error (Kim 2006). The results of an overall test of the proportional-hazards assumption for the entire 36-month observation period did not reach statistical significance (P = 0.07) (Kim 2006). Even if these discrepancies did not occur, in most of the cases a publication due to limitation of space in a journal will lack the full wealth of knowledge of the original trial data. Thus, all future meta-analyses by independent researchers will have to rely on the degree of rigour of the published report increasing the necessity of improving the quality of reporting. Additionally, reliance on published studies tends to introduce a bias from over-representation of those which showed positive findings. On the other hand, it has to be clearly outlined that by making important safety data accessible to interested researchers and the public at large does not of course absolve authorities from their duty to carefully and continuously monitor the evidence on the adverse effects of the drugs (Juni, Nartey et al. 2004).

Notably, prior to the withdrawal of rofecoxib (30th of September 2004) only a handful of articles raised concern about it's safety, although the Fitzerald's hypothesis (an inbalance of the vasoactive prostanoids PGI_2 and TxA_2 can lead to an increased CV toxicity of coxibs) was known at the time of their launch. Before the 30th of September 2004, most publications would have hinted towards a potential risk, but would finish with the cliché that further trials are required. However, a great plethora of publications in almost all journals appeared asserting the CV toxicity of rofecoxib and questioning the CV safety of the other coxibs after rofecoxib's withdrawal. A quick pubmed search using only the term "rofecoxib" would give 738 hits till the 30th of September 2004, while on the 11th of July 2006 (almost two years after its discontinuation) would give 1704 hits, more than double of the publications available when rofecoxib was on the market.

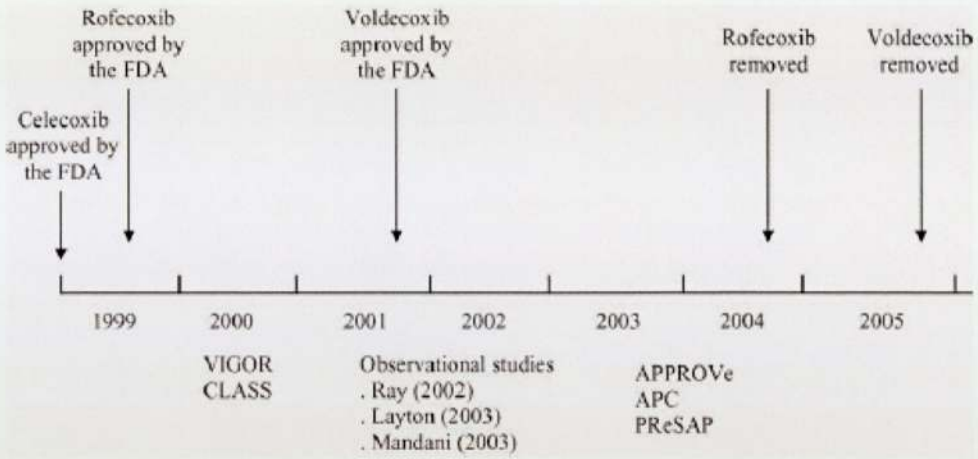
External funding was associated with publication independently of the statistical significance of the results (Egger, Dickersin et al. 2001). However, results were heterogeneous and the effect appears to depend on the source of funding (Egger, Dickersin et al. 2001). Research funded by industry is less likely to be published (Hemminki 1980; Bardy 1998). Additionally, the results of 89% of published industry-supported trials favoured the new therapy, as compared to 61% of the other trials (Davidson 1986). Similar results have been reported for NSAIDs trials (Rochon, Gurwitz et al. 1994) and drug studies published in symposium proceedings (Cho and Bero 1996). The implication is that the pharmaceutical industry tends to discourage the publication of negative studies which it has funded (Egger, Dickersin et al. 2001). Keeping the above in mind is important when considering that 5 out of the 15 systematic reviews / meta-analyses were sponsored by industry while in 8 out of the 15

systematic reviews / meta-analyses the authors were or had been employees of coxib manufacturer's (see chapter 5, Table 5.13, p187).

Although obtaining and including data from unpublished trials appears to be the obvious way of avoiding publication bias, the inclusion of data from unpublished studies can itself introduce bias (Egger, Dickersin et al. 2001). The trials that can be located may be an unrepresentative sample of all unpublished trials (Egger, Dickersin et al. 2001). Unpublished trials maybe of lower methodological quality than published trials. A recent study of 60 meta-analyses that included published and unpublished trials found that unpublished trials were less likely to adequately conceal treatment allocation and blind outcome assessments (Sterne, Bartlett et al. 2000). Thus, the notion that meta-analyses of individual patient's data are always superior to meta-analyses of published work might have to be revised (Horton 1999).

6.4 Observational studies versus randomised trials

Regulatory agencies and experts give less weight to observational studies than to randomised trials. Several retrospective cohort studies were published between the publication of the VIGOR study, which provided the first warning signals about rofecoxib, and the recommendation by the independent committee of the APPROVe trial that prompted the removal of rofecoxib. A timeline for reported toxicity with rofecoxib has been taken from a paper by (Ravaud and Tubach 2005)



Ray et al (Ray, Stein et al. 2002a) and Layton et al (Layton, Hughes et al. 2003) reported that the relative risk for myocardial infarction in patients taking rofecoxib was 1.7 (95%CI: 0.98-2.95, P: nonsignificant) with dosages greater than 25mg.day, compared to non-users, and that the relative risk for cardiovascular events was 1.38 (95%CI: 0.71-2.67, P: nonsignificant) compared to meloxicam users. Mamdani et al (Mamdani, Rochon et al. 2003) showed no relation between rofecoxib and CV risk when compared with either non-use of NSAIDs, traditional NSAIDs, or celecoxib. Thus, observational data were inconsistent and were dismissed by regulatory authorities and experts again for being inherently biased. Small relative risks (i.e. <2.0) in observational studies may easily arise due to confounding or bias (Temple 1999). However, although RCTs are considered to be optimal for establishing efficacy, this is not necessarily true for safety, because of the inherent bias of RCTs (Ravaud and Tubach 2005). RCTs are conducted in highly selected patients that in most cases do not mirror the true population using the drug in clinical practise.

Randomised trials and observational studies although different attempt to compare an event in exposed to drug in question group with one that does not receive

the intervention. Randomisation, the most important difference between the two, seeks to obtain two groups almost identical for all known and unknown baseline factors that could potentially influence the study outcomes. In observational studies baseline differences may exist between the control and study groups (Ravaud and Tubach 2005). An example can be that patients utilising COX-2 inhibitors maybe be older than those prescribed with a traditional NSAID (Ravaud and Tubach 2005). If an increased is detected in the COX-2 treated group, the respective contributions to this increase of COX-2 inhibitor treatment and older age cannot be determined (Ravaud and Tubach 2005). On the other hand, patients enroled in RCTs are usually higly selected, and this may exclude certain important groups or lead to their underrepresentation (i.e. older patients or patients with multiple co-morbidities or drug treatments). RCTs focus on internal validity (the extent on which differences in effects between two arms can be ascribed to the study drug), whereas observational studies focus on external validity (the degree to which the findings can be generalised) (Ravaud and Tubach 2005).

Several studies have compared the results of observational studies and RCTs in a number of areas and close correlation were found (correlation between odds ratio: $r = 0.75$, $P < 0.001$) (Ioannidis, Dixon et al. 1999; Ravaud and Tubach 2005). However, the treatment effects were usually larger in the observational studies (Ravaud and Tubach 2005). Even when the comparison was restricted to prospective observational studies (in theory less biased than other observational designs), in over one-third of cases the odds ratios were more than twice those in RCTs (Ravaud and Tubach 2005). The likely explanation is that exposure and confounding factors are difficult to eliminate, and appropriate adjustment for potential confounders is challenging (McKee, Britton et al. 1999; Ravaud and Tubach 2005; Rochon, Gurwitz et al. 2005).

Selection of cases and controls is challenging in observational studies identifying prevalent users rather than new users (incident users) (Ravaud and Tubach 2005). Furthermore, accurate information on exposure is usually lacking along with an accurate assessment of treatment duration (Ravaud and Tubach 2005). It is also difficult to exclude concomitant exposure to other agents and especially OTC analgesics (Ravaud and Tubach 2005). Diagnosis of cardiovascular events was usually made by the patient's general practioner without review by an independent committee blinded to the exposure data (Ravaud and Tubach 2005). Finally potential confounding factors are often unclear as data on the cardiovascular risk factors (as obesity, smoking status etc.) are lacking (Ravaud and Tubach 2005). Data were analysed under the hypothesis that the risk remains constant over time, which was not true (Ravaud and Tubach 2005). Recall bias in retrospective studies is associated with better recollection of COX-2 inhibitor than therapy with traditional NSAIDs (Ravaud and Tubach 2005).

Although observational studies are not ideal, the most sensible approach probably consists in improving the quality of observational studies and meta-analyses of observational data, and using the results to generate hypotheses (Ravaud and Tubach 2005). Then, these hypotheses need to be tested in large pragmatic postmarketing RCTs conducted in conditions as close as possible to real-life clinical practice, and on sufficiently large test groups to find evidence for rare side-effects (Ravaud and Tubach 2005).

6.5 ADRs signals ignored

The Yellow Card system is a "spontaneous" reporting system for suspected ADRs that was introduced in the UK following the thalidomide tragedy. The name arises from the colour of the report forms completed by a variety of healthcare professionals and lately also directly from patients. In the UK a total of 4,069 yellow cards were filled reporting a total of 7,189 suspected adverse drug reactions for rofecoxib of which 105 were reported as fatal (Medicines and Healthcare products Regulatory Agency 2006). This information was obtained from the Drug Analysis Prints, which is the collection of the information of the Yellow Cards submitted by healthcare professionals jointly to the Medicines and Healthcare products Regulatory Agency (MHRA) and the Commission on Human Medicines (CSM) in the UK, that process, evaluate and take any necessary action on the suspected ADRs. Although conclusions cannot be made on the safety and risk of a medicine based solely on the Drug Analysis Prints, a useful signal can be generated.

A large number of reports were received for the COX-2 inhibitor including rofecoxib. For example, in Mersey CSM regional centre, a total of 143 reports were received for rofecoxib and 40 for celecoxib by 2002 (Dingle 2002). Cardiovascular events accounted for 13% of the total for COX-2 inhibitors and it is interesting to note the 35% were gastrointestinal (Dingle 2002). These include 70 serious reactions (38.2%), four of which were fatal (Dingle 2002). Although a high number of ADRs is expected for black-triangled newly marketed drugs as the COX-2 inhibitors, a signal about rofecoxib was emerging through the yellow card system. Although the MHRA

informed the healthcare providers of this signal, maybe further action (i.e. requesting manufacturer's of coxibs to launch further adequately powered trials) was required.

A limitation of the yellow card scheme is that a yellow card does not imply causality (as all suspected adverse drug reactions should be reported), and a true signal or false positive is left to the MHRA to be decided. After January 2005, months after rofecoxib was withdrawn, the MHRA introduced a pilot scheme to welcome yellow cards by patients in an attempt to reduce the amount of undereported ADRs (Crombie 1984; Belton 1997) and improve pharmacovigilance services in the UK. This, however, can make signal detection more difficult as events would be diluted by the reporting of minor adverse drug reactions, unless these reports are analysed separately.

6.6 Marketing & the need to differentiate coxibs from tNSAIDs

Abdominal pain, dyspepsia or nausea, which are among the most commonly reported symptoms in NSAID users (Brogden, Heel et al. 1984; Giercksky, Husby et al. 1988; Ofman, MacLean et al. 2002), have been reported to occur early-most commonly within the first 6 weeks of treatment in contrast to the risk of NSAID ulcer complications which remains constant over time of NSAID exposure (Goldstein, Bello et al. 2005). As the incidence of symptomatic ulcers and ulcer complications are a significant iatrogenic cause of morbidity and mortality (Singh 1998), the COX-2 inhibitors were marketed as agents that held the promise of fewer adverse-effects as far at least as the GI tract and the platelets are concerned. Despite the absence of an indication of superior efficacy or an outcome study of adverse effects, celecoxib and rofecoxib were licensed and marketed aggressively (Wang, Wang et al. 2005). In the

US, more than 100 million dollars direct-to-consumer advertising with unrealistic expecations about pain relief (never proven to be have enhanced efficacy compared to traditional NSAIDs), marked GI protection and safety (never proven for celecoxib or valdecoxib) took place. As arthritis is one of the most common conditions requiring medication, this direct public and aggressive marketing further exacerbated the problem. It has been shown (Ray, Stein et al. 2002) that patients using rofecoxib were considerably more likely to have a history of major cardiovascular disease than a history of major GI bleeding (Juni, Reichenbach et al. 2005). The relative risks of myocardial infarction and ulcer complications observed in the RCTs included in the analysed systematic reviews or meta-analysis are therefore unlikely to translate into a favourable benefit-risk ratio in clinical practise: the estimated number needed to treat (NNT) for 1 year to cause one myocardial infarction is 70 patients whereas the NNT to avoid one hospitalisation for peptic ulcer disease is 157 (Juni, Reichenbach et al. 2005).

Thus, it is necessary that healthcare professionals to resist being "seduced by mechanisms" (Petitti 2004) that would suspend healthy scepticism when interpreting data (Juni, Nartey et al. 2004).

6.7 Clinical evidence for the CV adverse-effects of coxibs

The recall of rofecoxib was decided by the analysis of the safety data from the APPROVe (Adenomatous Polyp Prevention on Vioxx) trial that was intended primarily to test for rofecoxib (25mg daily) protection of the recurrence of colorectal polyps, but which also led to the premature cessation of the trial. In this study of 2586

patients, an excess incidence of cardiovascular events was statistically significant for rofecoxib versus placebo (Table 6.1B) (Bresalier, Sandler et al. 2005). A total of 46 patients in the rofecoxib group had a confirmed thrombotic event during 3059 patient-years of follow-up (1.50 events per 100 patient-years), as compared with 26 patients in the placebo group during 3327 patient-years of follow-up (0.78 event per 100 patient-years); the corresponding relative risk was 1.92 (95 percent confidence interval, 1.19 to 3.11; P=0.008) (Bresalier, Sandler et al. 2005). Hypertension was evident early in the rofecoxib arm, but the increased relative risk of myocardial infarctions and thrombotic strokes achieved statistical significance after 18 months of treatment (Bresalier, Sandler et al. 2005). The APPROVe trial permitted the use of aspirin doses up to 325mg but did not include aspirin administration in the protocol. These results prompted Merck to withdraw rofecoxib voluntarily on the 30th of September 2004 (Table 6.1A). Six months later (7th of April 2005), the FDA decided that it would carefully review any new proposal from Merck for resumption of marketing of rofecoxib and that this review will be discussed with the new FDA Drug Safety Oversight Board and an advisory committee prior to a final decision (Table 6.1A) ((FDA) 2005).

Rofecoxib was approved by the FDA in May 1999 (Table 6.1A) based on the results of small, short-term phase III trials that did not demonstrate a statistically significant risk of adverse cardiovascular outcomes (Konstantinopoulos and Lehmann 2005; Villalba 2005). Despite the fact these trials, which involved approximately 5000 patients, were adequately powered only to examine outcomes such as pain relief, safety concerns were still raised as thromboembolic events were more frequent in the

rofecoxib arm compared to placebo after 6 weeks (0.67% versus 0.24%) (Konstantinopoulos and Lehmann 2005; Psaty and Furberg 2005; Villalba 2005)

Table 6.1A COX-2 Inhibitors: Characteristics and Status in the US and the EU

Generic (Brand) Name	Chemistry	COX-1/ COX-2 ratio*	Indications	Licensing date (FDA)	Status in US	Status in EU
Celecoxib (Celebrex)	Sulfonamide	30	OA signs & symptoms	31.12.98	Label revision (07.04.05) & inclusion of a medication guide	Label revision (Feb 2005)-Ongoing safety review
			RA signs & symptoms	31.12.98		
			Analgesia / Primary Dysmenorrhoea	10.10.01		
			FAP	23.12.99		
Valdecoxib (Bextra)	Sulfonamide	261	OA signs & symptoms	16.11.01	Withdrawn voluntarily (07.04.05): Serious skin reactions & increased CV toxicity in CABG patients	Withdrawn voluntarily (07.04.05): Serious skin reactions & increased CV toxicity in CABG patients
			RA signs & symptoms	16.11.01		
			Primary Dysmenorrhoea		Not licensed in US for this indication	
Parecoxib (Dynastat)	Sulfonamide	261	Short-term post-op pain		Not licensed in US	Ongoing safety review
Rofecoxib (Vioxx)	Sulfone	276	OA signs & symptoms	20.05.99	Withdrawn voluntarily (30.09.04): Increased CV & CB toxicity	Withdrawn voluntarily (30.09.04): Increased CV & CB toxicity
			RA signs & symptoms	April 02		
			Analgesia / Primary Dysmenorrhoea	20.05.99		
			Migraine	March 04		
			JRA	10.09.04		
Etoricoxib (Arcoxia)	Sulfone	344	OA and RA signs & symptoms			Ongoing safety review
			Acute Gout			
Lumiracoxib (Prexige)	Phenyl acetic acid	433	OA signs & symptoms		Awaiting License-Novartis withhold progress after 30.09.04	Ongoing safety review -Post Launch Enhanced Pharmacovigilance and Risk Management Plan
			RA signs & symptoms			
			Short-term relief of post-op pain			
			Primary Dysmenorrhoea			
Imrecoxib					Not licensed	Not licensed
Cimicoxib					Not licensed	Not licensed

Key: OA: Osteoarthritis, RA: Rheumatoid Arthritis, FAP: Familiar adenomatous polyposis, JRA: Juvenile Rheumatoid Arthritis, Post-op: post-operative, US: United States of America, FDA: Food and Drug Administration, EU: European Union, CV: Cardiovascular, CABG: Coronary artery bypass graft. * Konstantinopoulos, P.A and Lehmann, D. F (2005) J. Clin. Pharmacol. 45 (7): 742-50.

Table 6.1B Major Clinical Trials (RCTs) evaluating COX-2 inhibitors

RCT	Drug/dose	Endpoints	Design	Population	ASA Use/Notes
VIGOR	Rofecoxib 50mg OD Naproxen 500mg BD	Assess the GI toxicity of rofecoxib compared to naproxen	R, DB, C, MC	4047 4029 **8076 RA**	No
APPROVe	Rofecoxib 25.g OD Placebo	Assess effects of rofecoxib on the risk of recurrent neoplasia of polyps	R, DB, PC, MC	1287 1299 **2586 CA**	Allowed (<326mg) -no inclusion in protocol
CLASS	Celecoxib 400mg BD Ibuprofen 800mg TDS Diclofenac 75mg BD	Assess the GI toxicity of celecoxib compared to other NSAIDs	R, DB, C, MC	3987 1985 1996 **7968 OA/RA (27.4%)**	Allowed (<326mg): 21.8% of population (1739 patients)/ 57% withdrawal
APC	Celecoxib 200mg BD Celecoxib 400mg BD Placebo	Assess celecoxib on prevention of adenomatous polyps in patients after polypectomy	R, DB, PC, MC	685 671 679 **2035 CA**	Allowed: 30% of patients
PreSAP	Celecoxib 400mg BD Placebo	Assess celecoxib on prevention of APs in patients after polypectomy	R, DB, PC	933 628 **1561 CA**	Allowed
ADAPT	Naproxen 220mg BD Celecoxib 200mg Placebo	Assess naproxen & celecoxib on delay of prevention of onset of AD & age-related cognitive decline	R, DB, PC, (double placebo)	**2625 AD**	Allowed use of low-dose ASA/ Patients older than 70 years
TARGET	Lumiracoxib 400mg OD Naproxen 500mg BD Ibuprofen 800mg TDS	Assess GI and CV toxicity of lumiracoxib compared to other NSAIDs	R, DB, PC, MC	9156 4754 4415 **18325 OA**	Allowed use of low-dose ASA (25%)/ Patients older than 50years/ 39% withdrawal

Key: RCT: Randomised Clinical Trial, OD: Once daily, BD: Twice daily, TDS, Three times daily, R: randomised, DB: Double-blind, C: controlled, PC: Placebo Controlled, MC: Multicenter, ASA: Aspirin, OA: Osteoarthritis, RA: Rheumatoid Arthritis, CA: Colorectal adenomas, APs: Adenomatous polyps, AD: Alzheimer's Disease, GI: Gastrointestinal, CV: Cardiovascular, NSAIDs: Non-steroidal anti-inflammatory drugs

However, more evidence indicating an association between rofecoxib and cardiovascular events was provided by the VIGOR (Vioxx Gastrointestinal Outcomes Research) trial. VIGOR compared rofecoxib (50mg daily) with naproxen (500mg twice daily) in 8076 patients with RA. (Table 6.1B) (Bombardier, Laine et al. 2000). The occurrence of cardiovascular effects was not a pre-specified end-point; VIGOR was designed to compare primarily the GI events of randomised RA patients (patient

with inherent increased risk to CV disease) to rofecoxib or naproxen. However, cardiovascular events were also monitored. Notably, this trial lacked a placebo arm, precluded the use of aspirin and excluded patients with recent cardiovascular events (Konstantinopoulos and Lehmann 2005). The VIGOR trial demonstrated a significantly higher incidence of myocardial infarctions in the rofecoxib arm as compared to the naproxen arm (0.4% versus 0.1%, respectively) (Bombardier, Laine et al. 2000). Although the VIGOR study had not been designed to investigate side-effects of rofecoxib, it brought about the alarming result of a nearly 5-fold increased risk of myocardial infarction in those patients that received rofecoxib (Krotz, Schiele et al. 2005). The lack of placebo arm raised the question of whether the cardiovascular risk observed in the rofecoxib arm was due to rofecoxib itself or to a cardioprotective effect of naproxen.

The authors of the VIGOR study related this difference to a potential antiplatelet effect of naproxen. Even if naproxen is able to inhibit platelet COX-1-dependent TxA_2 production, it also inhibits systemic PHI_2 production *in vivo*, which is a critical difference when compared to low-dose aspirin (Capone, Tacconelli et al. 2004). The possible cardioprotective effect of naproxen has also been examined in several observational, pharmacoepidemiological studies (Jick 2000; Rahme, Pilote et al. 2002; Ray, Stein et al. 2002; Ray, Stein et al. 2002; Schlienger, Jick et al. 2002; Watson, Rhodes et al. 2002; Mamdani, Rochon et al. 2003; Garcia Rodriguez, Varas-Lorenzo et al. 2004; Kimmel, Berlin et al. 2004; Graham, Campen et al. 2005). Firstly, in observational studies of the effects of treatment, non-randomised comparisons can be affected by confounding by indication (Grobbee and Hoes 1997). Secondly, a recent meta-analysis of observational studies of naproxen (Juni, Nartey et al. 2004) illustrated

that patient taking naproxen were 0.86 times as likely as patients taking other agents to experience a myocardial infarction (95%CI 0.75-0.99). Thus, even if we consider that naproxen possessed a protective effect, it is probably small, and as pointed out (Juni, Dieppe et al. 2002; Ray, Stein et al. 2002), not large enough to account for a 5 to 1 difference observed in VIGOR (Konstantinopoulos and Lehmann 2005).

No large placebo-controlled randomised trials addressing the cardioprotective potential of naproxen are available (Dieppe, Ebrahim et al. 2004). However, the ADAPT (Alzheimer Disease Anti-inflammatory Prevention Trial) was stopped after an average follow-up of 3 years due to an apparent increase in cardiovascular and cerebrovascular events in the naproxen arm compared to placebo (Table 6.1B) ((NIH) 2005). The purpose of this large, randomised, double-blind, placebo-controlled study, sponsored by the National Institute of Aging, was to test the ability of the non-selective NSAID naproxen and the COX-2 inhibitor celecoxib to delay or prevent the onset of Alzheimer's disease (AD) and age-related cognitive decline (Martin, Meinert et al. 2002; Konstantinopoulos and Lehmann 2005). No increase in cardiovascular and cerebrovascular events was identified in the celecoxib arm.

The forty-six patient of 4097 (1.1%) in the rofecoxib arm of the VIGOR trial and 20 of 4029 (0.5%) in the naproxen arm had serious cardiovascular events (Absolute risk reduction [ARR]: 0.006, Number needed to treat [NNT]: 167, or 1 serious CV event for every 167 patients treated with rofecoxib compared to naproxen; relative risk [RR]: 2.2), and this adverse outcome cannot be attributed solely to naproxen (Konstantinopoulos and Lehmann 2005). If the estimate of the magnitude of the difference in VIGOR proved accurate, it was twice what one would expect from an

"aspirin" effect of naproxen (Wong, Chowienczyk et al. 2005). These results led to a label change for Vioxx® in April 2002 contra-indicating the use of rofecoxib in patients with obvious ischaemic heart disease (Konstantinopoulos and Lehmann 2005).

The clinical picture observed with other coxibs was similar to the one observed for rofecoxib. Although celecoxib did not demonstrate an increase in CV and cerebrovascular (CB) disease in the ADAPT trial, the Adenoma Prevention with Celecoxib (APC) trial was suspended only 3 days after the cessation of the ADAPT trial. The APC (Table 6.1B) was a prospective, randomised, double-blind, multicenter trial sponsored by the National Cancer Institute that assessed the efficacy of celecoxib for the prevention of adenomatous polyps in patients who had undergone polypectomy (Solomon, McMurray et al. 2005). It involved 2035 patients randomised to placebo, celecoxib 200mg twice daily (bd) and celecoxib 400mg bd with a planned follow up of 60 months (Solomon, McMurray et al. 2005). However, after 33 months of follow up, due to an increased risk of fatal and nonfatal CV events associated with celecoxib use, the safety board decided to suspend the trial (Solomon, McMurray et al. 2005). A dose-response effect regarding the incidence of adverse-effect regarding the incidence of adverse CV outcomes was demonstrated: 7 of 679 patients (1%) died of CV causes in the placebo group, as compared with 16 of 685 patients (2.3%) receiving 200mg of celecoxib bd (ARR: 1.3%, NNT: 77, RR: 2.3) and 23 of 671 patients (3.4%) receiving 400mg bd (ARR: 2.4%, NNT: 42, RR: 3.4) (Konstantinopoulos and Lehmann 2005). Notably, this tendency was observed for both aspirin and non-aspirin users (Konstantinopoulos and Lehmann 2005).

Parallel to the APC trial, another randomised controlled trial, the Prevention of Spontaneous Adenomatous Polyps (PreSAP) trial, was conducted by the manufacturer (Pfizer) including 1561 patients randomised to celecoxib 400mg daily or placebo (Pfizer Inc. 2004; Konstantinopoulos and Lehmann 2005). Identical methods of analysis was used in PreSAP as with the APC trial and the same independent committee as in the APC trial. However, preliminary data from this study, which was suspended after 32 months of follow-up due to findings of the APC trial, did not show any increased risk in CV events in the celecoxib arm compared to placebo.

Celecoxib was licensed in 1999 based on evidence from small, short-term phase III trials whose primary endpoints were pain relief and endoscopically visualised gastric ulceration. The Celecoxib Long-term Arthritis Safety Study (CLASS) trial involved 8059 patients who received celecoxib 400mg bd, ibuprofen 800mg three times daily (tds) or diclofenac 75mg bd (Table 6.1B) (Silverstein, Faich et al. 2000). In CLASS the use of aspirin was permitted, unlike in VIGOR, and 21% of the participants (1739 patients) received low-dose aspirin (Silverstein, Faich et al. 2000). Although there were more events in the celecoxib group than in the ibuprofen and diclofenac group, this difference did not achieve statistical significance in patients receiving or not receiving aspirin (Silverstein, Faich et al. 2000). Cardiovascular toxicity, however, was not the primary endpoint of the CLASS trial, which was not adequately powered to detect differences in the rates of cardiovascular events (Konstantinopoulos and Lehmann 2005).

Shortly after the withdrawal of rofecoxib, the FDA issued a "black box" warning (FDA label change that requires the warnings about the agent to appear in a

black box in the patient information leaflet) for valdecoxib for life-threatening skin reactions and cardiovascular risk ((FDA) 2004). This warning was elicited by the results of two placebo controlled RCTs in patient immediately after coronary bypass grafting revealed an increased risk of serious CV events associated with the use of valdecoxib and its intravenous pro-drug parecoxib (Ott, Nussmeier et al. 2003; Nussmeier, Whelton et al. 2005). In a meta-analysis of these two RCTs, although the treatment-placebo difference did not reach conventional levels of statistical significance for the individual trials, valdecoxib was associated with a 3-fold higher risk of CV events than placebo (RR: 3.08, 95%CI: 1.20-7.87, P: 0.019), while no statistical evidence of heterogeneity was apparent (P: 0.86) (Furberg, Psaty et al. 2005). Of note, no study assessing the GI effect of valdecoxib has ever been reported (Konstantinopoulos and Lehmann 2005). On the other hand, both prodrug and active drug are sulphonamide derivatives, and life-threatening hypersensitivity disorders including anaphylaxis, angioedema, erythema multiforme, exfoliative dermatitis, Stevens-Johnson syndrome, and toxic epidermal necrolysis have been documented (Konstantinopoulos and Lehmann 2005). Based on the increased risk of adverse cardiovascular events in the short-term coronary artery bypass surgery trials, the reports of serious skin and potentially life-threatening skin reactions, and the absence of any demonstrated benefit of valdecoxib compared to already established NSAIDs, the FDA and EMEA decided to withdraw Bextra from the market and Pfizer agreed to voluntarily suspend sales and marketing of Bextra in the US and European Union. ((FDA) 2005b).

Finally, another coxib, only licensed in the UK, lumiracoxib, was studied in patients mostly at low CV risk in the Therapeutic Arthritis Research and

Gastrointestinal Event Trial (TARGET) study (Farkouh, Kirshner et al. 2004). A total of 18,325 OA patients were randomised to lumiracoxib 400mg daily, naproxen 500mg bd, or ibuprofen 800mg tds for 1 year, in 2 substudies of identical design (lumiracoxib versus ibuprofen or naproxen) (Konstantinopoulos and Lehmann 2005). Randomisation was stratified for low-dose aspirin use and age (Konstantinopoulos and Lehmann 2005). Incidence of myocardial infarction (clinical and silent) in the overall population in the individual substudies was 0.38% with lumiracoxib (18 events) versus 0.21% with naproxen (10 events) and 0.11% with lumiracoxib (5 events) versus 0.16% with ibuprofen (7 events) (Farkouh, Kirshner et al. 2004). In the naproxen substudy, rates of myocardial infarction (clinical and silent) did not differ significantly compared with lumiracoxib in the population not taking low-dose aspirin (hazard ratio 2.37 [95% CI: 0.74-7.55], P: 0.1454), overall (1.77 [0.82-3.84], P: 0.1471), and in patients taking aspirin (1.36 [0.47-3.93], P: 0.5658). In the ibuprofen substudy, these rates did not differ between lumiracoxib and ibuprofen in the population not taking low-dose aspirin (0.75 [0.20-2.79], P: 0.6669), overall (0.66 [0.21-2.09], P: 0.4833), and in patients taking aspirin (0.47 [0.04-5.14], P: 0.5328) (Farkouh, Kirshner et al. 2004). Hence the number of events was small, but the RR in non-aspirin users was 1.47, although it did not attain significance (Wong, Chowienczyk et al. 2005). However, the follow up for the TARGET study was only 1 year, and this trial was clearly not adequately powered to detect the difference in CV events in non-aspirin users (Konstantinopoulos and Lehmann 2005). Thus, as it can be concluded from all the above RCTs, most of the coxibs exhibit a tendency for cardiovascular toxicity.

6.8 Postulated mechanisms explaining the coxibs's cardiotoxicity

Although not an aim of this study, it is important to discuss the postulated mechanisms based on which the CV and renal adverse events of COX-2 inhibitors are elicited. Understanding the pharmacological evidence is basic for the prediction of the efficacy but also toxicity of new agents. Some of these mechanisms and the experimental data to support them were available even before the first COX-2 inhibitor received license.

6.8.1 Imbalance in the production of vasoactive prostanoids

The recall of rofecoxib followed the publication of an extensive series of articles that argued in favor of an enhanced CV risk. The mechanism originally presented that has now achieved widespread acceptance is that these agents suppress prostacyclin PGI_2 production of the endothelium, while letting the generation of thromboxane TxA_2 from platelets unaffected (McAdam, Catella-Lawson et al. 1999).

NSAIDs inhibit the activity of the prostaglandin H synthase (PGHS) by preventing access of arachidonic acid to the catalytic site of the cyclooxygenase located inside a hydrophobic channel formed by PGHS, without affecting peroxidase activity, which is located outside this hydrophobic channel (Smith, DeWitt et al. 2000; FitzGerald 2003). Following the production of PGH_2, a second enzymatic process is needed to ultimately form the different biologically active prostanoids, catalysed by tissue-specific enzymes (Helliwell, Adams et al. 2004). These enzymes show some specificity with respect to the tissue they are expressed in and also generate specific

prostanoids, which also determines the name of the enzyme, and have numerous functions in the vascular system (see chapter 1, Figure 1.3, p19) (Kaplan-Machlis and Klostermeyer 1999; Krotz, Schiele et al. 2005).

Inhibition of cyclooxygenases results in decreased substrate availability for such tissue-specific prostanoid synthases and subsequently decreases the production of the specific prostanoid (Krotz, Schiele et al. 2005). Formation of a specific prostanoid from arachidonic acid, seems to be determined by the tissue of interest and by the specific pathophysiological situation (Helliwell, Adams et al. 2004). In platelets for example, which only contain the COX-1 isoform, the major PGH_2-metabolising isomerase coupled to COX-1 is TxA_2 synthase, which leads to the production of the major arachidonic acid product of cyclooxygenase activity in platelets, TxA_2 (FitzGerald 1991; Helliwell, Adams et al. 2004). As platelets as well as vascular smooth muscle cells express TxA_2 receptors, the release of TxA_2 from platelets results in platelet aggregation and to a lesser extent in vasoconstriction . (Armstrong 1996). When platelet COX-1 is inhibited by NSAID or by aspirin, the resulting inhibition of TxA_2 mediates the desired antiplatelet effect (see chapter 1, Figure 1.3, p19). Thus, aspirin remains the most importance substance counteracting platelet aggregation, as firstly it binds irreversibly acetylating a serine residue at position 529 in the cyclooxygenase hydrophobic channel (Catella-Lawson, Reilly et al. 2001; Patrono, Coller et al. 2001) and secondly low-dose aspirin only effectively inhibits platelet cyclooxygenase activity (Krotz, Schiele et al. 2005). Although a single dose of only 100mg/day already shows an inhibitory effect on COX-1, it is further increased by repetition of this dose, and low-dose aspirin ultimately blocks TxA_2 synthesis through accumulation in platelets (Krotz, Schiele et al. 2005). As platelets are anuclate

structures, they are unable to sufficiently resynthesize cyclooxygenase (the substrate's access to its active site is impeded for the lifetime of the platelet), so the inhibitory effect of aspirin can only be reversed by novel platelet synthesis from megakaryotes (Krotz, Schiele et al. 2005). Platelets are regenerated at a daily rate of approximately 10 percent (Di Minno, Silver et al. 1983; Patrono, Ciabattoni et al. 1985). For the platelet aggregation to be impaired, the capacity of platelets to generate TxA_2 must be inhibited more than 95% (Van Hecken, Schwartz et al. 2000). Clinically doses of NSAIDs also have an impact on the activity of cyclooxygenase, these drugs do not bind irreversibly and usually dissociate from their binding sites at cyclooxygenase (Patrono, Patrignani et al. 2001). Thus, only aspirin effectively inhibits platelet COX-1 activity as well as resulting TxA_2 synthesis, whereas NSAIDs inhibit all cyclooxygenases and resulting prostanoids, but only do so reversibly (Krotz, Schiele et al. 2005). Even naproxen with a more extended pharmacodynamic half-time, leading to more sustained platelet inhibition, was reported to reduce myocardial infarction by 14% compared to 23% reduction by aspirin (Juni, Nartey et al. 2004; Konstantinopoulos and Lehmann 2005).

On the other hand higher doses of aspirin or NSAIDs do not result in more effective inhibition of platelet aggregation. Prostacyclin (PGI_2), a potent platelet inhibitor, is formed in intact vascular endothelium through cyclooxygenase coupled to PGI_2 synthase. Whereas repeated administration of low doses of aspirin has little effect on immediate or long-term cyclooxygenase activity in the endothelium due to the aforementioned trasncriptional novel synthesis of cyclooxygenases because endothelial COX-2 has limited sensitivity to drug (Patrono, Coller et al. 2001; Patrono, Patrignani et al. 2001), high doses of aspirin or NSAIDs have similar effects on endothelial PGI_2

and platelet TxA_2 synthesis, thus theoretically exerting antithrombotic as well as prothrombotic effects (see chapter 2, Figure 2.4, p82) (Krotz, Schiele et al. 2005). This combined with the limited time span and the reversibility of NSAIDs binding to cyclooxygenases form the pharmacological basis for several observations reporting that NSAID are not effective as low-dose aspirin in inhibiting platelet aggregation (Krotz, Schiele et al. 2005).

Furthermore, cyclooxygenase isoforms are differentially expressed and regulated throughout the vascular system. Although COX-1 is expressed almost ubiquisly and regarded as a housekeeping enzyme (Lipsky 1999; Buttar and Wang 2000), while COX-2 seems to be an inducible enzyme upregulated by stimulation with pro-inflammatory mediators (e.g. cytokines, growth factors, lipopolysaccharides) (Hinz, Brune et al. 2000; Hinz and Brune 2002), there is evidence for COX-2 being constitutively expressed in a variety of tissues with important physiological functions in some (Iseki 1995; Zimmermann, Sarbia et al. 1998; Nantel, Meadows et al. 1999) (Chakraborty, Das et al. 1996);(Slater, Dennes et al. 1999a; Slater, Dennes et al. 1999b); (Tegeder, Neupert et al. 2000; Damm, Rau et al. 2001)) (Krotz, Schiele et al. 2005). COX-2 is constitutively expressed in some cells of the vascular system, e.g. endothelial cells, or cells of the renal medulla, renal vasculature or the macula densa, and participates in the regulation of vessel function through paracrine or autocrine release of certain prostanoids (Hinz and Brune 2002; FitzGerald 2003). Moreover, it has been shown that COX-2 constitutively binds to PGI_2 synthase in endothelial cells (Liou, Shyue et al. 2000), and numerous data suggest that it is a physiological source of PGI_2 *in vivo* (Gimbrone, Topper et al. 2000; Krotz, Schiele et al. 2005). Multiple studies have demonstrated that any form of vascular stimulation (such as laminar shear stress) upregulates COX-2 gene expression in the endothelial cells, leading to

increased production of PGI_2 (Fitzgerald, Roy et al. 1986). Under these circumstances, the increased production of PGI_2 occurs concomitantly with the enhanced production of TxA_2 by the platelets (FitzGerald, Smith et al. 1984; Funk, Funk et al. 1991). This concomitant increase in production of PGI2 and TxA2 reflects a homeostatic response to any vascular stimuli that cause platelet activation (i.e. rupture of the atherosclerotic plaque) (Funk, Funk et al. 1991).

Hence, *in vivo*, there is a fine-tuned balance between certain prostanoids produced by specific coupling of cyclooxygenases with tissue-dependent prostanoid synthases which is influenced by the differential expression of cyclooxygenase isoforms (Krotz, Schiele et al. 2005). In addition factors like age, the stage of atherosclerosis diseases, or the extent of preexisting endothelial function, gender, and the interaction of prostanoids with the production of other autacoids contribute to the role of COX-2 dependent prostanoid formation *in vivo* (Krotz, Schiele et al. 2005).

Concern about the depression of vascular PGI_2 production in the absence of concomitant platelet inhibition (McAdam, Catella-Lawson et al. 1999; FitzGerald and Patrono 2001) had enhanced awareness of the need for adjuvant antiplatelet therapy in appropriate patients who are receiving COX-2 selective inhibitors. It is easy to perceive that any drug that selectively reduces plasma levels of a physiological antiplatelet substance like PGI_2, without altering levels of the corresponding platelet activator, TxA_2, theoritically has an intrinsic likeliness of increasing the activity of circulating platelets, predisposing patients to adverse CV outcomes (Krotz, Schiele et al. 2005).

Even though this mechanism of an intrinsic likeliness of prothrombotic effects of selective COX-2 inhibitors was postulated even before their launch in 1999, the first evidence for an enhanced thrombotic risk under selective inhibition of COX-2 was gathered by Hennan et al. in dogs in 2001 (Hennan, Huang et al. 2001). In this study, high-dose aspirin had no effect in coronary artery thrombotic occlusion unless it was withdrawn and a recovery time for the endothelium to resynthesize cyclooxygenase was allowed for. After the endothelium had recovered cyclooxygenase (but not platelets because of the irreversible binding of aspirin), there was an increased time to thrombotic occlusion, but this antithrombotic effect was prevented by the administration of celecoxib during recovery (Hennan, Huang et al. 2001). Recently, two experimental studies clearly proved that there is thrombotic risk under selective inhibition of COX-2 *in vivo*. The first of these studies that was published just before Vioxx® was withdrawn from global markets, used a highly sensitive *in vivo* microcirculatory model (Buerkle, Lehrer et al. 2004; Krotz, Schiele et al. 2005). It revealed that selective inhibition of COX-2 enhanced platelet activation, leading to increased platelet rolling at the intact arterioral wall. Moreover, firm platelet adhesion was increased and ultimately a markedly reduced time to thrombotic occlusion upon vessel wall damage resulted (Buerkle, Lehrer et al. 2004). The other study, appearing just after rofecoxib's withdrawal, showed that during hypoxia in the pulmonary circulation of rats, there was enhanced platelet activation under selective inhibition of COX-2 (Pidgeon, Tamosiuniene et al. 2004). Interestingly, in all these studies, selective inhibitors of COX-2 have not been reported to cause spontaneous thrombosis (Belton, Duffy et al. 2003; Buerkle, Lehrer et al. 2004; Pidgeon, Tamosiuniene et al. 2004). Nevertheless, these studies could prove what already was theoretically

plausible, i.e. that selective COX-2 inhibitors enhance platelet activation and thus are able to trigger the onset of thrombotic events (Krotz, Schiele et al. 2005).

6.8.2 Sulfone COX-2 inhibitors pro-oxidant activity

The observed differences in thrombotic risk among COX-2 inhibitors may be due to trial design (aspirin use, comparative NSAID, underpowered trials) or their distinct physico-chemical properties (Walter, Jacob et al. 2004). Distinct physico-chemical properties of COX-2 inhibitors underlie differences in their pharmacology, pharmacokinetics and potentially their cardiovascular safety (Vane 2002). These actions may be independent of COX-2 inhibition (Vane 2002).

The mechanistic basis for the pro-oxidant activity associated with the sulfone COX-2 inhibitors is attributed to physico-chemical properties and specific interactions with phospholipid molecules (Walter, Jacob et al. 2004). The sulfone COX-2 inhibitors, etoricoxib and rofecoxib, exhibited pro-oxidant activity in human plasma and isolated LDL (Walter, Jacob et al. 2004). The pro-oxidant effects of these agents were unrelated to COX-2 inhibition as they were reproduced in pure lipid vesicles enriched with arachidonic acid. Additionally, no changes were observed in lipid peroxidation rates with sulfonamide COX-2 selective inhibitors (celecoxib and valdecoxib), other NSAIDs (naproxen, ibuprofen, diclofenac), or even other sulfone-containing compounds (methyl phenyl sulfone) (Walter, Jacob et al. 2004). The lack of activity for celecoxib was observed even at suprapharmacological doses (Walter, Jacob et al. 2004). By contrast, the pro-oxidant effects of rofecoxib were dose-dependent (Walter, Jacob et al. 2004).

Rofecoxib has been shown to increase the susceptibility of human low-density lipoprotein and cellular membrane lipids to oxidative modification, a contributing factor to plaque instability and thrombus formation (Preston Mason, Walter et al. 2006). Independently of COX-2 inhibition, rofecoxib also promoted the nonenzymatic formation of isoprostanes and reactive aldehydes from biologic lipids (Preston Mason, Walter et al. 2006). The basis for these observations is that rofecoxib alters lipid structure and readily forms a reactive maleic anhydride in the presence of oxygen (Preston Mason, Walter et al. 2006). By contrast, other selective (celecoxib, valdecoxib) and nonselective (naproxen, diclofenac) inhibitors did not influence rates of low-density lipoprotein and membrane lipid oxidation (Preston Mason, Walter et al. 2006). Recent evidence have confirmed these findings by demonstrating that the prooxidant activity of rofecoxib can be blocked by the potent antioxidant astaxanthin in homochiral form (all-trans 3S, 3'S) (Preston Mason, Walter et al. 2006). These findings provide a mechanistic rationale for differences in cardiovascular risk among COX-selective inhibitors because of their intrinsic physicochemical properties (Preston Mason, Walter et al. 2006).

6.8.3 Renal mechanism of CV adverse-effects of coxibs

Prostaglandins as PGE_2, PGF_{2a}, PGD_2, PGI_2, generated at various intrarenal sites, are known to modulate a variety of aspects of renal physiology, including renal tubular function, renal haemodynamics, and secretion of renin from the juxtaglomerular apparatus, with attendant effects on aldosterone and potassium homeostasis and tubular sodium, water, and urea transport (Roman and Lechene 1981; Kramer, Stinnesbeck et al. 1985; Conte, Cianciaruso et al. 1992; Frazier and Yorio

1992; Navar, Inscho et al. 1996; Breyer, Zhang et al. 1998; Brater, Harris et al. 2001). PGI_2 and PGD_2, synthesised by glomerular and medullary interstitial cells, have been shown to redistribute blood flow from the renal cortex to the juxtamedullary region by dilating renal vascular beds, thereby lowering vascular resistance (Oates, FitzGerald et al. 1988). In experimental models, PGE_2 produces a diuretic and natriuretic action through inhibition of sodium chloride transport in the thick ascending limp of the loop of Henle and in the collecting duct (Stokes and Kokko 1977; Stokes 1979). Renal prostaglandin effects can, thus, be devided in those that are physiologically important at all times even in healthy individuals and those that occur during settings of renal "stress" (Brater, Harris et al. 2001).

In the mammalian kidney, the macula densa is involved in regulating renin release (Persson, Salomonsson et al. 1991) by sensing alterations in luminal chloride via changes in the rate of sodium-potassium-2-chloride co-transport (Schlatter, Salomonsson et al. 1989). The induction of a high renin state, induced by the imposition of a salt-deficient diet, significantly increased macula densa/cortical thick assending loop of Henle (cTALH) COX-2 mRNA and immunoreactive protein (Harris, McKanna et al. 1994). A selective COX-2 inhibitor, NS398, inhibited increases in renal renin expression in response to a low salt diet (Harding, Sigmon et al. 1997). Increases in renin mRNA expression and renal renin activity in response to angiotensin-converting enzyme inhibition were also blunted by the highly selective COX-2 inhibitor, SC58236 (Cheng, Wang et al. 1999). Furthermore, in experimental renovascular hypertension, in which macula densa COX-2 expression is increased, COX-2 inhibition blunted increases in renin expression and lowered blood pressure (BP) (Hartner, Goppelt-Struebe et al. 1998; Wang, Cheng et al. 1999). Direct evidence

for a role for COX-2 were provided, as in an isolated perfused juxtaglomerular preparation increased renin release in response to lowering the prefusate sodium chloride concentration was blocked by NS398 (Traynor, Smart et al. 1999).

Accumulating evidence suggests that COX-2 plays an important role in maintaining renal function in the setting of physiological stress, in which renal function becomes dependent upon prostaglandins, including volume depletion, after radiocontrast administration and congestive heart failure (Schlondorff 1993). Medullary COX-2 mRNA and protein expression is significantly increased after dehydration (Yang, Schnermann et al. 1999; Hao, Yull et al. 2000). In vitro, shifting to hypertonic media (using either sodium chloride or mannitol) directly induces COX-2 expression in renal medullary interstitial cells, and suggests that ambient tonicity is a major factor regulating COX-2 expression in the medulla (Breyer and Harris 2001). The signalling mechanisms activated by different osmolytes may depend on the cell type examined. In contrast to cultured renal epithelial cells (Yang, Schnermann et al. 1999), where hyperosmolarity achieved using the cell permeable solute, urea, increased COX-2, urea did not increases COX-2 expression in interstitial cells (Hao, Yull et al. 2000) (Breyer and Harris 2001).

Osmotic induction of COX-2 expression in medullary interstitial cells plays an important role in the ability of these cells to survive hypertonic stress after dehydration (Breyer and Harris 2001). Normally, near 100% of medullary interstitial cells survive an abrupt increase in ambient tonicity; however, in the presence of sub-micromolar concentrations of the COX-2 inhibitor SC58236, only approximately 50% of cells survive (Hao, Yull et al. 2000) (Breyer and Harris 2001). Similarly, in-vivo water

deprivation in the setting of a COX-2 inhibitor induced dramatic medullary interstitial cell apoptosis, whereas simple water deprivation had no effect on interstitial cell survival (Hao, Yull et al. 2000) (Breyer and Harris 2001). The mechanism by which COX-2 action promotes medullary interstitial cell survival and osmotic tolerance remains uncertain (Breyer and Harris 2001). Nonetheless, these observations may have important implications for understanding the pathogenesis of NSAID-associated renal medullary injury (Breyer and Harris 2001).

COX-2 derived prostanoids may also play a critical role in maintaining renal medullary blood supply, renal salt excretion and systemic blood pressure (Breyer and Harris 2001). COX-2 rich medullary interstitial cells span the area between the vasa rectae and medullary tubules including thick loops (Lemley and Kriz 1991). Cultured medullary interstitial cells produce abundant PGE_2, which has been shown directly to dilate vasa rectae, counteracting the constrictor effect of angiotensin and endothelin, thereby helping to maintain renal medulary blood flow (Silldorff, Yang et al. 1995). (Breyer and Harris 2001)

Prostaglandins play an important role in maintaining medullary blood supply, particularly in the setting of volume depletion (Roman and Lianos 1990), and after radio-contrast (Agmon, Peleg et al. 1994) (Breyer and Harris 2001). Other studies (Hla and Maciag 1991; Komhoff, Grone et al. 1997) suggested that COX-2 may be expressed in the endothelial cells of the renal medulla, where it could also influence vascular tone (Breyer and Harris 2001). The regulation of renal medullary blood flow has significant implications for regulating salt excretion and systemic blood pressure (Cowley, Mattson et al. 1995). Reduced medullary interstitial pressure increases renal

salt absorption, and could contribute to sodium retention in the setting of NSAID use and after the ingestion of COX-2 selective inhibitors (Brater 1999; Catella-Lawson, McAdam et al. 1999).

Medullary interstitial prostanoids not only have the capacity to modulate vascular tone, but also epithelial solute and water readsorption (Breyer and Breyer 2000). Loss of tonic inhibitory effect of COX-2 derived PGE_2 on salt absorption by the medullary thick limb and collecting duct may also contribute to sodium retention (Breyer and Breyer 2000). Taken together, these data suggest that COX-2 inhibition in the renal medulla might not only contribute to salt retention and hypertension, but also compromise medullary blood flow, risking anoxic injury to the cellular elements in the renal medulla as well as directly risking medullary interstitial cell viability (Breyer and Breyer 2000).

COX metabolites have been implicated in functional and structural alterations in glomerular and tubulointerstitial alterations in glomerular and tubulointerstitial inflammatory diseases (Feng, Sun et al. 1993; Klahr and Morrissey 1998) Several studies (Stahl, Kudelka et al. 1986; Nath, Chmielewski et al. 1987; Pelayo and Shanley 1990; Schmitz, Krupa et al. 1994) have suggested that prostanoids may also modify renal function and glomerular damage after subtotal renal ablation, and glomerular prostaglandin production may be altered in such conditions (Breyer and Harris 2001). After subtotal renal ablation, there were selective increases in renal cortical COX-2 mRNA and immunoreactive protein expression, without significant alterations in COX-1 expression (Breyer and Harris 2001). This increased COX-2 expression was most prominent in the macula densa and surrounding cTALH, the site of expression of

cortical COX-2 in the normal rat kidney (Wang, Cheng et al. 2000). In addition, there was detectable COX-2 immunoreactivity in some glomeruli from remnant kidneys, with increased expression in visceral epithelial cells and mesangial cells (Breyer and Harris 2001).

6.8.3.1 Renal clinical evidence supporting a renal mechanism of cardiotoxicity observed with COX-2 inhibitors

NSAID use is associated with at least four major renal syndromes in man, including acute haemodynamically mediated renal insufficiency, sodium retention with hypertension or oedema, hyperkalaemia, and papillary necrosis (Schlondorff 1993; Brater 1999). It was originally hoped that renal safety would be improved by using COX-2 selective inhibitors. However, soon after their launch it was obvious by case reports that COX-2 inhibitors do not spare the kidney (Perazella and Eras 2000).

Recent reports suggest that COX-2 inhibitors will reduce glomerular filtration in susceptible patients (Brater 1999; Breyer and Harris 2001). Although rare, NSAID-associated renal insufficiency occurs in a significant proportion of patients with underlying congestive heart failure, diabetes and old age (Schlondorff 1993; Brater 1999). These risk factors are additive and patients with multiple risk factors are rarely included in clinical studies of these drugs (Breyer and Harris 2001). Thus, it is relevant that celecoxib caused a transient decrease in glomerular fltration rate (GFR) in salt-depleted but otherwise healthy young male subjects (Rossat, Maillard et al. 1999). In another clinical study, elderly patients on a salt-restricted diet (two risk factors), demonstrated that rofecoxib decreased GFR by approximately 15% (Swan, Rudy et al.

2000). Although these clinical studies have suggested a severe decline in renal function in subjects treated with COX-2 specific inhibitors occurs as observed with those treated with NSAIDs, the subjects were not at high risk of renal insufficiency (Breyer and Harris 2001).

In the CLASS study, 1% of the celecoxib patients compared with 1.6% of the NSAID patients (ibuprofen and diclofenac arms) (P=0.03) shared an increase in serum creatinine >1.8mg/dL and/or serum urea nitrogen >40mg/dL (Silverstein, Faich et al. 2000). In the VIGOR study, the incidence of renal function adverse-effects was 1.2% in the rofecoxib and 0.9% in the naproxen group, while 0.2% of patients in each arm discontinued treatment due to these side-effects (Bombardier, Laine et al. 2000; Kramer, Kammerl et al. 2004). In the SUCCESS VI study, 1.5% of hypertensive patients with OA developed clinically significant serum renal laboratory values during 6 weeks of treatment with both 200mg celecoxib or 25mg of rofecoxib (Whelton, Fort et al. 2001). So, in patient with diminished intravascular volume (i.e. pre-existing heart and liver failure, nephrotic syndrome, diminished fluid intake in elderly patients), COX-2 inhibitors have been shown to cause acute renal failure as often as conventional NSAIDs (Kramer, Kammerl et al. 2004).

Sodium retention, oedema and hypertension is a well known side-effects of NSAIDs (Murray, Greene et al. 1992; Brater 1999), but also apparent in COX-2 selective inhibitors (Catella-Lawson, McAdam et al. 1999; Rossat, Maillard et al. 1999; Swan, Rudy et al. 2000; Whelton, Schulman et al. 2000). These studies have shown a reduction of urinary sodium excretion with rofecoxib and celecoxib, while associated with modest sodium retention in otherwise healthy individuals. A reduced

GFR may limit the filtered sodium load and salt excretion (Rossat, Maillard et al. 1999; Swan, Rudy et al. 2000). Additionally, PGE_2 directly inhibits sodium absorption in the thick ascending limb and collecting duct (Breyer and Breyer 2000). The relative abundance of COX-2 in medullary interstitial cells places this enzyme adjacent to both these nephron segments, allowing for COX-2-derived PGE_2 to modulate salt absorption. COX-2 inhibitors decrease renal PGE_2 production (Catella-Lawson, McAdam et al. 1999; Whelton, Schulman et al. 2000), and thereby may enhance renal sodium retention (Breyer and Harris 2001).

Furthermore, reduction in renal medullary blood flow by the inhibition of vasodilator prostanoids may significantly reduce renal salt excretion and promote the development of oedema and hypertension (Cowley, Mattson et al. 1995). COX-2 selective inhibitors have been shown to exacerbate salt-dependent hypertension in rats (Muscara, Vergnolle et al. 2000), and also similarly in humans (Brater 1999).

In a study (Whelton, Fort et al. 2001) specifically set up to look at the effects of coxibs on oedema formation and hypertension, patients with OA and hypertension were randomised to receive celecoxib 200mg or rofecoxib 25mg for 6 weeks with unchanged antihypertensive medication. The primary endpoint, increase in oedema from baseline of at least 1 grade in peripheral oedema plus 3% weight gain or increase from baseline of 2 or more grades) occurred more often in rofecoxib treated patients than celecoxib treated patients (9.5 vs 4.9%, $P<0.05$) (Kramer, Kammerl et al. 2004). This is in accordance with a higher gain of relative body weight in the rofecoxib versus celecoxib group (0.6 versus 0.1% at week 1, 0.6 versus 0.2% at week 6; 0.6% would be roughly equivalent to a weight gain of 0.5 kg) (Kramer, Kammerl et al. 2004).

Significantly more patients also reached a systolic blood pressure (SBP) endpoint (>20mHg increase, >140mmHg absolute value) during rofecoxib than celecoxib treatment (16.5 versus 10.9%, $P<0.05$) (Whelton, Fort et al. 2001). Mean SBP was unchanged during celecoxib treatment, but increased significantly by 2.4, 2.8, and 3.1mmHg at weeks 1, 2, and 6 after rofecoxib treatment. Finally, 4 patients in the rofecoxib group versus none in the celecoxib developed congestive heart failure during the trial (Kramer, Kammerl et al. 2004). These results were also confirmed in a similarly designed large trial (SUCCESS VII) (Whelton, White et al. 2002). However, these trials have been critisised with regard to the doses of COX-2 inhibitors compared [celecoxib 200mg OD (half-maximal dose) and rofecoxib 25mg OD (maximal daily dose)], differences in plasma half-lives (rofecoxib longer than celecoxib), and the fact that significantly more patients were pretreated with ACE inhibitors in the celecoxib group in the smaller trial of the two (Kramer, Kammerl et al. 2004).

When reviewing data on spontaneous reported oedema formation in the 12-week North American arthritis trials, a frequency of 2.1% was observed for both celecoxib and NSAID administration in comparison to 1.1% during placebo (Whelton, Maurath et al. 2000). No-dose dependency of oedema formation was observed with daily doses of celecoxib ranging from 100-800mg (Whelton, Maurath et al. 2000). Nonetheless, the incidence of oedema formation after celecoxib administration in patients on diuretics was expecially high (~6.5%) (Whelton, Maurath et al. 2000). Finally, in the 6-month CLASS trial, a 2.8% of oedema was observed versus a 3.5% in patients treated with NSAIDs (ibuprofen, diclofenac) (Silverstein, Faich et al. 2000).

The frequency of celecoxib-induced severe congestive heart failure (CHF) was reported to very low with 0.03% with celecoxib and 0.07% with NSAID use and 0.05% with placebo in the North American arthritis trials (Whelton, Maurath et al. 2000), while 1.7% of patients using celecoxib and 2.3% using NSAIDs in the 6-month CLASS trial (Silverstein, Faich et al. 2000).

The frequency of celecoxib-induced hypertension was reported to be 0.8% with celecoxib and 0.7% with NSAID use while only 0.3% with placebo in the 12 weeks North American arthritis trial (Whelton, Maurath et al. 2000). Higher rates of hypertension was observed in the 6-month CLASS trial of 1.7% for celecoxib-treated patients and 2.3% for NSAIDs treated patients (Silverstein, Faich et al. 2000).

A meta-analysis of 19 RCTs involving a total of 45451 participants treated with coxibs in RCTs that provided BP data was recently conducted (Aw, Haas et al. 2005). Primarily, a disproportionate rise in SBP compared to diastolic BP (DBP), on average, with coxib use. This potential widened pulse pressure could have significant CV risk implications as demonstrated in the Framingham study (Kannel 2000), which observed a very steep relationship between SBP and CV risk (Aw, Haas et al. 2005). Interestingly, for each defined level of SBP, the lower the DBP, the greater the CV risk (Kannel 2000; Leonetti, Cuspidi et al. 2000; Aw, Haas et al. 2005). Among the trials analysed, coxibs caused a weighted mean difference point estimate increase in SBP and DBP compared with placebo (3.85/1.06 mmHg) and NSAIDs (2.83/1.34 mmHg) (Aw, Haas et al. 2005). Coxibs were associated with a nonsignificantly higher RR of causing hypertension compared with placebo (RR: 1.61, 95%CI [0.91-2.84,P: 0.10]) and NSAIDs (RR: 1.25, [0.87-1.78, P: 0.23]) (Aw, Haas et al. 2005). Another major

observation of this meta-analysis was that of a consistent increase in systolic and diastolic blood pressure (BP) with rofecoxib in head-to head trials versus celecoxib (Aw, Haas et al. 2005). Rofecoxib induced a weighted mean difference point estimate increase in SBP (2.83 mmHg) and a nonsignificant higher risk of developing clinically important SBP elevation (RR: 1.50, [1.00-2.26, P: 0.05]) compared with celecoxib (Aw, Haas et al. 2005). There also appears to be a different effect when reviewing the individual contributions of each coxib with respect to the development of hypertension (Aw, Haas et al. 2005). These differential effects on BP may relate to differences in pharmacokinetic and pharmacodynamic properties. Celecoxib has a shorter half-life than rofecoxib, with differential effects on BP still evident during 24-hour ambulatory BP monitoring (Liew and Krum 2002; White, Kent et al. 2002; Aw, Haas et al. 2005). Rofecoxib is metabolised by cytosol reductase, which may (particularly at high doses) competitively inhibit the metabolism of aldosterone (Aw, Haas et al. 2005). This may further exacerbate fluid and sodium retention as well as vascular remodelling (Liew and Krum 2002). Alternatively, celecoxib may also inhibit carbonic anhydrase (originally developed as an antiglaucoma agent), leading to a diuretic action that would offset some of the BP-elevating effect of COX-2 inhibition within the kidney (FitzGerald and Patrono 2001).

Thus, from all the above, COX-2 inhibitors very early after their launch have been shown to cause volume overload with oedema formation, deterioration of cardiac function, and hypertension in frequencies close to those of NSAID use (Kramer, Kammerl et al. 2004).

6.9 Ethical considerations & faults

Neither Merck, Co. Inc or licensing authorities fullfilled its responsibilities to the public. Data from the VIGOR trial were not submitted to a peer-reviewed journal until November 2000, while the reported cardiovascular data were incomplete. The reason for the latter was partly because the design and execution of the trial had not anticipated the occurrence of adverse cardiovascular events (Topol 2004). It was not until the 2001 (8^{th} of February) that the FDA Arthritis Advisory Committee met to discuss concern about the potential CV risks associated with rofecoxib. It remains unclear why the FDA waited for two years after its review and approval of rofecoxib to conduct this meeting. It was, then, mandatory to conduct a trial specifically assessing CV risk and benefit of these agents (Mukherjee, Nissen et al. 2001). Given the very high coinidence of coronary disease and arthritis, this group may represent the largest segment of the population for whom rofecoxib was prescribed (Topol 2004). Unfortunately such a trial was never done. Licensing authorities have the authority to mandate that a trial be conducted, but they never took the initiative (Topol 2004).

Furthermore, in USA, rofecoxib's manufacturer company was spending more than 100 million dollars per year in direct-to-consumer advertising, an activity regulated by the FDA (Topol 2004). For the past few years, every month has seen more than 10 million prescriptions for rofecoxib in the USA alone. (Topol 2004) Although the FDA could have stopped Merck from using direct-to-consumer advertising of this block-buster drug , the only significant action taken was to on 11^{th} of April 2002, when the FDA instructed Merck to include certain precautions about CV risks (black box) in its package insert (Topol 2004).

The FDA sponsored large epidemiologic studies in a cohort of Kaiser Permanente patients (Topol 2004). Over the lifespan of rofecoxib, several large epidemiological studies took place with a population reaching the 1,4 million patients amplifying the concerns about the risk of myocardial infarction and serious cardiovascular adverse events (Topol 2004). Merck responded to these epidemiological findings by pointing the inherent bias of observational studies and that the only convencing evidence should come from a large RCT. However, Merck was unwilling to initiate such a trial to assess the CV toxicity of rofecoxib, and the licensing authorities failed to demand one.

Although direct-to-consumer advertising of prescription only medicines is not allowed in Europe, rofecoxib sales were analogous. As a consequence of the publication of the VIGOR trial results, the Committee for Proprietary Medicinal Products announced that a Europe wide safety review of COX-2 inhibitors was being launched (French referral under Article 31 of the Directive 2001/83/EC), with particular emphasis on GI and CV safety due to concenrs about the frequency of the GI and CV events ((EMEA) 2002). The EMEA did not mandate either the conduct of safety aimed RCTs for these agents. The NICE guidance on the use of COX-2 inhibitors for OA and RA issued on July 2001 was issued before the full trial data for CLASS and VIGOR were available. However, after CV concerns were raised, a review of this guidance was initiated in August 2003 almost two years later. This review was suspended in February 2005 following the withdrawal of rofecoxib and pending the outcome of the EMEA review on the safety of coxibs. At the request of the EMEA, Pfizer agreed to withdraw voluntarily valdecoxib on 7th of April 2005 due to CV and fatal skin reactions associated with its use. The EMEA safety review

concluded in July 2005 that COX-2 inhibitors are contra-indicated in patients with established ischaemic heart disease and/or cerebrovascular disease (stroke), and also in patients with peripheral arterial disease resulting in changes in the summary of product characteristics of coxibs ((EMEA) 2005).

Commercial interests play an ever larger role in directing medical practice. A large portion of the budget of regulatory authorities is derived directly from drug companies. Furthermore, medical journals are ill equipped to withstand the drug companies' financial pressure, research and statistical capacity, commercial ties with most recognised experts, and lack of transparency in the research they fund (Abramson and Starfield 2005). Universities have become dependent on pharmaceutical industry funding, while also enganging in industry's entrepreneurial activities (Abramson and Starfield 2005). Most specialty medical societies and large nonprofit health organisations e.g. the Arthritis Foundation receive a large part of their funding from drug manufacturers (Abramson and Starfield 2005).

Celecoxib use is restricted at the moment by the EMEA in Europe while being under constant ongoing review. However, this passive position of waiting for data to accrue is no longer acceptable. Careful monitoring of the pharmacovigilance signals whether they are obtained from observational studies, adverse drug reactions reporting schemes or RCTs should be carefully considered and acted upon. The public trusts national regulatory authorities to perform research sufficient to protect patients from unecessary harm, even if the task of balancing between benefit and harm seem impossible. Although one can make a case that the purpose of an industry is to make profit and not necessarily to serve the public good, it is difficult to accept this as a

justification for the behavior of medical scientists and regulatory authorities (Abramson and Starfield 2005).

6.10 Current practice & the possibility of rofecoxib being re-marketed

The MHRA, and the European Medicines Agency (EMEA), and the Commission on Safety of Medicines (CSM) have all formulated guidelines on the prescribing of coxibs. The CSM have sent their guidelines to all UK doctors (Sooriakumaran 2006). Patients with established ischaemic heart disease or cerebrovascular disease should be switched to alternative treatment as COX-2 inhibitors are contra-indicated. In addition, the existing contraindication for severe heart failure is now extended to include moderate heart failure (NHYA class II-IV) (MHRA 2005). Caution is advised to doctors when prescribing COX-2 inhibitors in patients with risk factors for heart disease such as hypertension, hyperlipidaemia, diabetes and smoking ((EMEA) 2005). For all patients the balance of GI and CV risk should be considered before prescribing a COX-2 selective inhibitor, particularly for those with risk factors for heart disease and those taking low-dose aspirin for whom GI benefit has not been clearly demonstrated (MHRA 2005). The lowest effective dose of COX-2 selective inhibitor should be used for the shortest necessary period (MHRA 2005). Periodic re-evaluation is recommended, especially for OA patients who may only require intermittent treatment (MHRA 2005). Gastroprotective agents should be considered for patients switched to non-selective NSAIDs (MHRA 2005). Paracetamol and non-drug interventions should be always the first to consider, which should be effective for most patients. In order to achieve to implement this guidance, most Primary Care Trusts (PCTs) in the UK shortly after the guidance became available

initiated audits to review the use of the coxibs in practice after the withdrawal of rofecoxib and to switch patients to non-selective NSAIDs when appropriate (with the use of a gastroprotective agent when required) with relatively success (Figure 6.1).

Prescription Pricing Authority data shows the effect of safety concerns following the withdrawal of rofecoxib on the prescribing of COX-2 selective inhibitors (NHS National Prescribing Centre 2005/2006). The number of prescription items dispensed in England for NSAIDs, excluding celecoxib, etoricoxib, rofecoxib, valdecoxib, parecoxib, etodolac, and meloxicam, has remained roughly constant at around 1.2 million items monthly. Most NSAID prescribing was for ibuprofen and diclofenac, which accounted for 55% of all NSAID prescription items dispensed in September 2004, rising to 68% of all NSAID prescription items dispensed in February 2005 (NHS National Prescribing Centre 2005/2006).

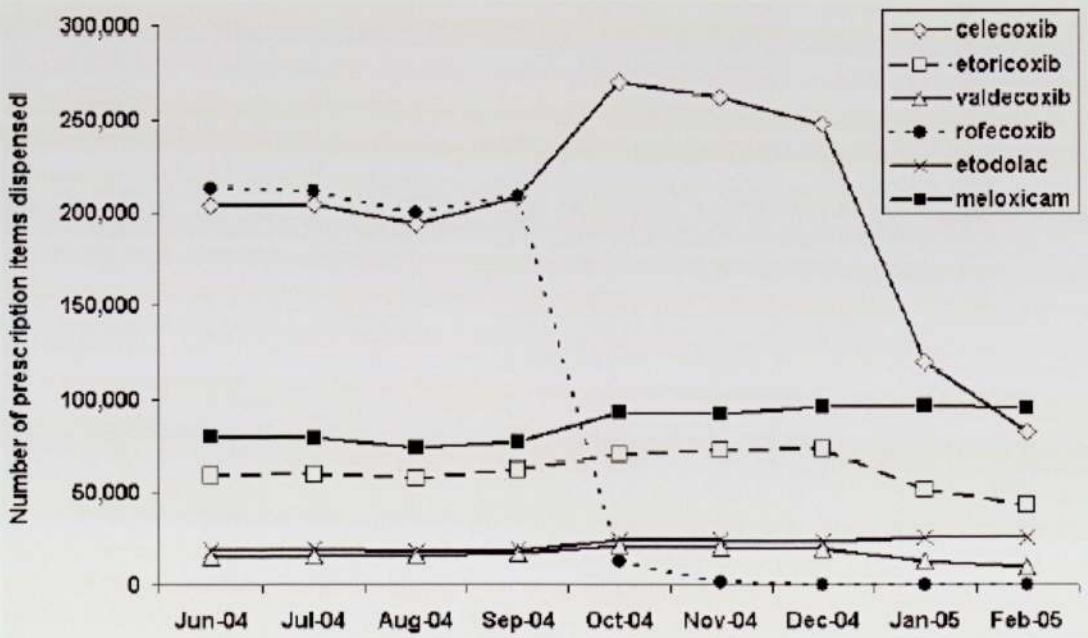


Figure 6.1 Trends in prescribing of COX-2 selective inhibitors in England (NHS National Prescribing Centre 2005/2006)

Recently it has been reported that Health Canada's Expert Advisory Panel has launched a review of the cardiovascular risk of rofecocib, valdecoxib, celecoxib and meloxicam (Sibbald 2006). A suggestion was made to attempt to lauch rofecoxib back on the basis of "patients would benefit from having a variety of drugs to choose from" (Sibbald 2006). However, rofecoxib will remain off the market until a new drug submission is received and approved. Merck has not yet decided whether to resubmit a new license application (Sibbald 2006).

To summarise, the above study provides a quality of reporting assessment and a systematic review of the 15 identified CV safety aimed systematic reviews or meta-analyses for a specific COX-2 inhibitor, rofecoxib in an attempt to identify possible reasons for the late recognition of its cardiotoxicity. The overall quality of reporting was assessed using the QUOROM checklist and found acceptable, although there is room for improvement particularly in abstract reporting. A summary of the objectives, inclusion and exclusion criteria, total number of patients and design of RCTs included in the analysed systematic reviews or meta-analyses was provided. Exploring the methods of information retrieval and data extraction, it was identified that the majority of the systematic reviews or meta-analyses utilised solely the manufacturer's data files and reveived funding from COX-2 manufacturers. The mean duration of exposure to rofecoxib was calculated to be 6 weeks, which was inadequate to assess CV safety in long-term use for indications like OA or RA. A few active comparators were used in these systematic reviews and the number of patients included was not adequate to assess rare or unexpected adverse events. Moreover, a description of the population of the analysed systematic reviews or meta-analyses was provided and the potential influences of funding explored. Finally, a brief description of the postulated

mechanisms for the cardiotoxicity of rofecoxib was provided linking them to available clinical evidence along with ethical considerations arising today and the current use of COX-2 inhibitors currently.

Chapter 7

Conclusions / Recommendations

7.1 Conclusions

Limitation of available evidence combined with a lack of adequate pharmacovigilace independent from manufacturers interest was a combination of factors that delayed the recognition of rofecoxib's CV adverse events.

The small number of CV safety aimed systematic reviews or meta-analyses performed by independent researchers or licensing authoririte prior to rofecoxib's world wide voluntary withdrwawal indicates that although signals were available to alert healthcare professionals they were ignored or not acted upon appropriately. The quality of reporting of available systematic reviews or meta-analyses needs to be improved further by following current quality guidelines. Even if the quality of the reseach was excellent, the quality of reporting can be a hinder in comparing results obtained from different groups if it is not of a high standard. Most of RCTs included were underpowered and too short to detect rare or unexpected adverse-effects. Furthermore, the results from available observational data were inconclusive and not meta-analysed even if they are considered to be inherently prone to bias. The inability to answer questions as whether naproxen was cardioprotective and if so in what extent combined with the lack of placebo trials (considered unethical) and RCTs lacking multiple active comparators contributed in the inability to attribute the CV effects to rofecoxib. Finally, the population of patients inlcuded in RCTs and thus systematic

reviews did not mirror the age and health status of patients treated with rofecoxib in actual practice.

Advertising, hopes of a new class of GI protective NSAIDs and the inability to understand the caveats of EBM especially by practitioners are also to blame for the widespread use of this agent. Action taken by licensing authrorities in the form of conta-indicating rofecoxib in high-risk patients of developing CV and CB adverse-events, while mandating a CV safety aimed long-term (more than 1 year duration) RCT was necessary. Cumulative meta-analysis of available RCTs should have been performed as part of rofecoxib's surveillance especially during the time the agent was black-triangled and questions were raised about its CV safety.

7.2 **Recomendations**

This study is a systematic review of the available evidence leading to the voluntarily withdrawal of rofecoxib due to cardiovascular toxicity. As in recent years several agents were withdrawn from the market or severely restricted due to unexpected cardiovascular toxicity (Gussak, Litwin et al. 2004), a number of recommendations can be made about the design of propsective cardiovascular safety evaluations as well as actions that need to be taken after rofecoxib's withdrawal.

The population tested should resemble the population likely to receive the drug in clinical practice (Konstam 2003). Thus, manufacturer's designing a propsective CV safety trial or independent researchers should broadly include the population targeted for an indication while including patients with common comorbidities, those receiving

commonly coadministered drugs, and those who are at increased risk of CV events. Secondly, the power of the analysis should be adequate to exclude a major adverse event. A reasonable approach to avoid impractical sample-sizes is to power such an evaluation to exclude, at most, a 50% increase in serious CV adverse events (Konstam 2003). Power sufficient to detect smaller differences may be warranted if there is particular reason for concern (Konstam 2003). For agents planned for chronic use, there should be adequate patient exposure for at least 1 year of treatment (Konstam 2003). There is a clear need to control for effects on elevation of BP as well as to include more than one active comparators or placebo. The results of the VIGOR trial were misinterpreted because of a lack of placebo group and the availibility of only one active comparator, the impact of which could not be clarified.

One approach for decreasing sample size requirements in safety aimed trials is the use of composite outcomes where different clinically relevant endpoints are combined. By virtue of increasing the event rate, fewer patients are required to detect a relative treatment effect of 50% (Tugwell, Judd et al. 2005). Composite outcomes are more effective if the components are relevant by themselves, but are not correlated with each other (or only weakly) (Tugwell, Judd et al. 2005). Combining outcomes could logically be taken further to designate serious endpoints in each organ system and to agree on an inclusion frequency criteria such as 1% greater in the experimental group than in the control group (Tugwell, Judd et al. 2005). However, if endpoints are not related, then statistical noise is added that may obscure the real outcome differences (Tugwell, Judd et al. 2005). If the endpoints are not all equally important, this may result in a too small sample size for the more important but less common endpoints (Tugwell, Judd et al. 2005). With CV events, a full index might be required

to be looked at, but if there are no effects for some adverse events for example arrhythmias, then including them will not increase the power to detect CV events (Tugwell, Judd et al. 2005). Thus, the all-cause and all single-organ cause alarm systems for the unexpected are required, but groupings of problems with putative common mechanisms to preserve some power and some specificity are also required (Tugwell, Judd et al. 2005). Future COX-2 inhibitors trials should include a cluster of MI-related events such as MI death, nonfatal MI, and angina in order to increase power, while maintaining a particular mechanism, that of thrombosis (Tugwell, Judd et al. 2005). Another cluster might be oedema, hypertension, and congestive heart failure, combining fatal and non fatal events under the same mechanism of fluid retention (Tugwell, Judd et al. 2005).

Furthermore, the inclusion of an independent endpoints committee should be the rule, and exceptions to this rule should be justified. (Juni, Nartey et al. 2004) The use of Data Monitoring Committees or Data Safety Monitoring Boards is probably one of the main breakthroughs in patient protection achieved in the recent years. Virtually every drug trial now has an independent committee of four of five members, including at least one clinician with extensive experience in the area of the study and one statistician (Ravaud and Tubach 2005). The other members should have experience wih clinical trials and may represent the other specialties potentially concerned with the trial (i.e. a cardiologist and a gastroenterologist in trials of COX-2 inhibitors) (Ravaud and Tubach 2005). Experts forming this committee should not have any other role in the trial and no potential conflicts of interest.

In new drug development, the burden of proof for overall and CV safety rests with the developer. For an agent that is going to be used extensively and chronically, the question of CV safety must be carefully considered. It is essential to establish the level of concern as early in a drug's development program for CV safety analysis to be developed prospectively (Konstam 2003). Licensing authorities are clearly obliged to request proof of long-term safety from drug developers, to avoid a repetition of rofecoxib example.

Today there is a clear need to regain the public trust to licensing authorities of fulfilling the aim of safeguarding public health. Careful consideration of observational data need to be taken. If summaries of product characteristics are routinely updated from spontaneous ADRs data, which are considered less "robust", then epidemiologic data should not be just ignored due to lack of "robustness" (Arellano 2006).

Increase of the number of reported ADRs can also provide with data to aid licensing authorities in practicing pharmacovigilance. Electronic submission mostly from doctors, hospital and community pharmacist, as well as reports from patients will make the process faster and hopefully will increase the number of ADRs reported. This will aid licesning authorities to analyse ADR data faster and take timely decisions.

The late detection of COX-2 inihibitor CV toxicity illustrates the difficulties raised by drug safety assessments. Available tools for evaluating drug safety, although imperfect, complement each other (Ravaud and Tubach 2005). Careful thinking of all possible parameters is required in developing licensing studies and RCTs, observational cohort studies, and conventional drug surveillance efforts. Postmarketing

studies should include a number of patients proportional to the total number treated in order to ensure the detection of small risks that may impact public health if present in large populations (Ravaud and Tubach 2005). A systematic review of the results from these various approaches is probably the most effective strategy also performed in a cumulative manner. Toxicity of new agents, however, may escape detection for several years even if all of the above are conducted and the licensing authorities have fulfilled their promise.

Whether rofecoxib will receive again license or not, the COX-2 inhibitors still continue to be an option in the treatment armamentarium. Multiple lines of evidence indicate that COX-2 inhibitors are associated with an increased risk of adverse CV outcomes, which is mostly evident in patients with established atherogenic disease. Assessment of individual risk-benefit is probably the most critical point in deciding when or if to prescribe these drugs. Patient education of the potential CV, GI and skin disease risks is obviously important. These notions are exemplified in the current EMEA recommendations about the coxibs.

Chapter 8

References

8.1 References

Abramson, J. and B. Starfield (2005). "The effect of conflict of interest on biomedical research and clinical practice guidelines: can we trust the evidence in evidence-based medicine?" J Am Board Fam Pract **18**(5): 414-8.

Agmon, Y., H. Peleg, Z. Greenfeld, S. Rosen and M. Brezis (1994). "Nitric oxide and prostanoids protect the renal outer medulla from radiocontrast toxicity in the rat." J Clin Invest **94**(3): 1069-75.

Ahuja, N., A. Singh and B. Singh (2003). "Rofecoxib: an update on physicochemical, pharmaceutical, pharmacodynamic and pharmacokinetic aspects." J Pharm Pharmacol **55**(7): 859-94.

Aisen, P. S. (1997). "Inflammation and Alzheimer's disease: mechanisms and therapeutic strategies." Gerontology **43**(1-2): 143-9.

Aisen, P. S. (2002). "Evaluation of selective COX-2 inhibitors for the treatment of Alzheimer's disease." J Pain Symptom Manage **23**(4 Suppl): S35-40.

Aisen, P. S. and K. L. Davis (1994). "Inflammatory mechanisms in Alzheimer's disease: implications for therapy." Am J Psychiatry **151**(8): 1105-13.

Aisen, P. S., K. A. Schafer, M. Grundman, E. Pfeiffer, M. Sano, K. L. Davis, M. R. Farlow, S. Jin, R. G. Thomas and L. J. Thal (2003). "Effects of rofecoxib or naproxen vs placebo on Alzheimer disease progression: a randomized controlled trial." Jama **289**(21): 2819-26.

Akil, M. and R. S. Amos (1995). "ABC of rheumatology. Rheumatoid arthritis--I: Clinical features and diagnosis." Bmj **310**(6979): 587-90.

Altman, D. G. (1991). Practical statistics for medical research. London, Chapman and Hall: 167-70.

Altman, R., G. Alarcon, D. Appelrouth, D. Bloch, D. Borenstein, K. Brandt, C. Brown, T.

D. Cooke, W. Daniel, D. Feldman and et al. (1991). "The American College of Rheumatology criteria for the classification and reporting of osteoarthritis of the hip." Arthritis Rheum **34**(5): 505-14.

Altman, R., E. Asch, D. Bloch, G. Bole, D. Borenstein, K. Brandt, W. Christy, T. D. Cooke, R. Greenwald, M. Hochberg and et al. (1986). "Development of criteria for the classification and reporting of osteoarthritis. Classification of osteoarthritis of the knee. Diagnostic and Therapeutic Criteria Committee of the American Rheumatism Association." Arthritis Rheum **29**(8): 1039-49.

Alzheimer's Society. (2006) Facts about dementia London, Available from: (http://www.alzheimers.org.uk/Facts_about_dementia/Statistics/index.htm) [Accessed on: 31st January 2006]

American College of Rheumatology Subcommittee. (2002). "Guidelines for the management of rheumatoid arthritis: 2002 Update." Arthritis Rheum **46**(2): 328-46.

American Heart Association. (2003) Heart Disease and Stroke Statistics: 2004 update Dalas, Texas, Available from: (http://www.americanheart.org/downloadable/heart/1079736729696HDSStats2004UpdateREV3-19-04.pdf) [Accessed on: 9th July 2006]

Anon (2003). "[Meta-analysis on acute pain. Rofecoxib helped three quarters of patients]." MMW Fortschr Med **145**(31-32): 57.

Arellano, F. M. (2006). "Observational studies and the withdrawal of rofecoxib." Pharmacoepidemiol Drug Saf **15**(3): 203-5.

Armstrong, R. A. (1996). "Platelet prostanoid receptors." Pharmacol Ther **72**(3): 171-91.

Arnett, F. C., S. M. Edworthy, D. A. Bloch, D. J. McShane, J. F. Fries, N. S. Cooper, L. A. Healey, S. R. Kaplan, M. H. Liang, H. S. Luthra and et al. (1988). "The American Rheumatism Association 1987 revised criteria for the classification of rheumatoid arthritis." Arthritis Rheum **31**(3): 315-24.

Assendelft, W. J., B. W. Koes, P. G. Knipschild and L. M. Bouter (1995). "The relationship between methodological quality and conclusions in reviews of spinal manipulation." Jama **274**(24): 1942-8.

Athirakul, K., H. S. Kim, L. P. Audoly, O. Smithies and T. M. Coffman (2001). "Deficiency of COX-1 causes natriuresis and enhanced sensitivity to ACE

inhibition." Kidney Int **60**(6): 2324-9.

Auperin, A., J. P. Pignon and T. Poynard (1997). "Review article: critical review of meta-analyses of randomized clinical trials in hepatogastroenterology." Aliment Pharmacol Ther **11**(2): 215-25.

Aw, T. J., S. J. Haas, D. Liew and H. Krum (2005). "Meta-analysis of Cyclooxygenase-2 Inhibitors and Their Effects on Blood Pressure." Arch Intern Med.

Barden, J., J. Edwards, A. Moore and H. McQuay (2004). "Single dose oral paracetamol (acetaminophen) for postoperative pain." Cochrane Database Syst Rev(1): CD004602.

Barden, J., J. Edwards, R. Moore and H. McQuay (2005). "Single dose oral rofecoxib for postoperative pain." Cochrane Database Syst Rev(1): CD004604.

Barden, J., J. Edwards, R. A. Moore and H. J. McQuay (2004). "Single dose oral rofecoxib for postoperative pain." Cochrane Database Syst Rev(1): CD004604.

Barden, J., J. E. Edwards, L. Mason, H. J. McQuay and R. A. Moore (2004). "Outcomes in acute pain trials: systematic review of what was reported?" Pain **109**(3): 351-6.

Barden, J., J. E. Edwards, H. J. McQuay and R. Andrew Moore (2004). "Pain and analgesic response after third molar extraction and other postsurgical pain." Pain **107**(1-2): 86-90.

Barden, J., J. E. Edwards, H. J. McQuay and R. A. Moore (2002). "Single-dose rofecoxib for acute postoperative pain in adults: a quantitative systematic review." BMC Anesthesiol **2**(1): 4.

Barden, J., J. E. Edwards, H. J. McQuay and R. A. Moore (2003). "Single dose oral celecoxib for postoperative pain." Cochrane Database Syst Rev(2): CD004233.

Barden, J., J. E. Edwards, H. J. McQuay and R. A. Moore (2003). "Oral valdecoxib and injected parecoxib for acute postoperative pain: a quantitative systematic review." BMC Anesthesiol **3**(1): 1.

Barden, J., J. E. Edwards, H. J. McQuay, P. J. Wiffen and R. A. Moore (2004). "Relative efficacy of oral analgesics after third molar extraction." Br Dent J **197**(7): 407-11; discussion 397.

Bardy, A. H. (1998). "Bias in reporting clinical trials." Br J Clin Pharmacol **46**: 147-150.

Barkin, R. L. and A. Buvanendran. (2004) Utilization of the Cox-2 Inhibitors (Coxibs) in

the Management of Acute Postoperative Pain and an Overview of Cox-1 NSAID Agents FreeMedCME, The Dannenmiller Memorial Educational Foundation San Antonio, Texas, December 2004 Available from: (http://www.freemedcme.com/cme/articles/article.cfm?mode=accreditation_view&cme_id=10) [Accessed on: 25th August 2006]

Barnett, J., J. Chow, D. Ives, M. Chiou, R. Mackenzie, E. Osen, B. Nguyen, S. Tsing, C. Bach, J. Freire and et al. (1994). "Purification, characterization and selective inhibition of human prostaglandin G/H synthase 1 and 2 expressed in the baculovirus system." Biochim Biophys Acta **1209**(1): 130-9.

Baron, J. A., B. F. Cole, R. S. Sandler, R. W. Haile, D. Ahnen, R. Bresalier, G. McKeown-Eyssen, R. W. Summers, R. Rothstein, C. A. Burke, D. C. Snover, T. R. Church, J. I. Allen, M. Beach, G. J. Beck, J. H. Bond, T. Byers, E. R. Greenberg, J. S. Mandel, N. Marcon, L. A. Mott, L. Pearson, F. Saibil and R. U. van Stolk (2003). "A randomized trial of aspirin to prevent colorectal adenomas." N Engl J Med **348**(10): 891-9.

Bassett, K., J. M. Wright, L. Puil, T. L. Perry, B. Heran and C. Cole (2002). "Cyclooxygenase-2 inhibitors update: Journal articles fail to tell the whole story." Can. Fam. Physician **48**: 1455-60.

Bazan, N., J. Botting and J. Vane (1996). New targets in inflammation: inhibitors of COX-2 or adhesion molecules (Proceedings of a conference held on 15-16 April 1996, New Orleans, USA. London, Kluwer Academic Publishers (Dordrecht/Boston/London) William Harvey Press.

Beecher, H. K. (1957). "The measurement of pain; prototype for the quantitative study of subjective responses." Pharmacol Rev **9**(1): 59-209.

Begg, C., M. Cho, S. Eastwood, R. Horton, D. Moher, I. Olkin, R. Pitkin, D. Rennie, K. F. Schulz, D. Simel and D. F. Stroup (1996). "Improving the quality of reporting of randomized controlled trials. The CONSORT statement." Jama **276**(8): 637-9.

Belcaro, G., A. N. Nicolaides, G. Ramaswami, M. R. Cesarone, M. De Sanctis, L. Incandela, P. Ferrari, G. Geroulakos, A. Barsotti, M. Griffin, S. Dhanjil, M. Sabetai, M. Bucci and G. Martines (2001). "Carotid and femoral ultrasound morphology screening and cardiovascular events in low risk subjects: a 10-year

follow-up study (the CAFES-CAVE study(1))." Atherosclerosis **156**(2): 379-87.

Bellamy, N., W. W. Buchanan, C. H. Goldsmith, J. Campbell and L. W. Stitt (1988). "Validation study of WOMAC: a health status instrument for measuring clinically important patient relevant outcomes to antirheumatic drug therapy in patients with osteoarthritis of the hip or knee." J Rheumatol **15**(12): 1833-40.

Belton, K. J. (1997). "Attitude survey of adverse drug-reaction reporting by health care professionals across the European Union. The European Pharmacovigilance Research Group." Eur J Clin Pharmacol **52**(6): 423-7.

Belton, O. A., A. Duffy, S. Toomey and D. J. Fitzgerald (2003). "Cyclooxygenase isoforms and platelet vessel wall interactions in the apolipoprotein E knockout mouse model of atherosclerosis." Circulation **108**(24): 3017-23.

Berenbaum, F. (2004). "COX-3: fact or fancy?" Joint Bone Spine **71**(6): 451-3.

Berg, J., T. Christoph, M. Widerna and A. Bodenteich (1997). "Isoenzyme-specific cyclooxygenase inhibitors: a whole cell assay system using the human erythroleukemic cell line HEL and the human monocytic cell line Mono Mac 6." J Pharmacol Toxicol Methods **37**(4): 179-86.

Berlin, J. A. and E. M. Antman (1994). "Advantages and limitations of metaanalytic regressions of clinical trials data." Online J Curr Clin Trials **Doc No 134**: [8425 words; 84 paragraphs].

Bjordal, J. M., A. E. Ljunggren, A. Klovning and L. Slordal (2004). "Non-steroidal anti-inflammatory drugs, including cyclo-oxygenase-2 inhibitors, in osteoarthritic knee pain: meta-analysis of randomised placebo controlled trials." Bmj **329**(7478): 1317.

Bjorkman, D. J. (1999). "Current status of nonsteroidal anti-inflammatory drug (NSAID) use in the United States: risk factors and frequency of complications." Am J Med **107**(6A): 3S-8S; discussion 8S-10S.

Blain, H., C. Boileau, F. Lapicque, E. Nedelec, D. Loeuille, C. Guillaume, A. Gaucher, C. Jeandel, P. Netter and J. Y. Jouzeau (2002). "Limitation of the in vitro whole blood assay for predicting the COX selectivity of NSAIDs in clinical use." Br J Clin Pharmacol **53**(3): 255-65.

Blettner, M., W. Sauerbrei, B. Schlehofer, T. Scheuchenpflug and C. Friedenreich (1999). "Traditional reviews, meta-analyses and pooled analyses in epidemiology." Int J

Epidemiol **28**(1): 1-9.

Blumenthal, D., E. G. Campbell, M. S. Anderson, N. Causino and K. S. Louis (1997). "Withholding research results in academic life science. Evidence from a national survey of faculty." Jama **277**(15): 1224-8.

(BNF), British National Formulary (March 2005). London, British Medical Association & Royal Pharmaceutical Society of Great Britain.

Bombardier, C. (2002). "An evidence-based evaluation of the gastrointestinal safety of coxibs." Am J Cardiol **89**(6A): 3D-9D.

Bombardier, C., L. Laine, A. Reicin, D. Shapiro, R. Burgos-Vargas, B. Davis, R. Day, M. B. Ferraz, C. J. Hawkey, M. C. Hochberg, T. K. Kvien and T. J. Schnitzer (2000). "Comparison of upper gastrointestinal toxicity of rofecoxib and naproxen in patients with rheumatoid arthritis. VIGOR Study Group." N Engl J Med **343**(21): 1520-8, 2 p following 1528.

Booth, S. (2006) Press releases: Latest US Vioxx trial ends in a split decision, but gives fresh hope to UK Vioxx victims Available from: (http://www.irwinmitchell.com/PressOffice/PressReleases/LatestUSVioxxTrialEndsInASplitDecisionButGivesFreshHopeToUKvictims.htm) [Accessed on: 8th July 2006]

Botting, R. M. (2000). "Mechanism of action of acetaminophen: is there a cyclooxygenase 3?" Clin Infect Dis **31 Suppl 5**: S202-10.

Brater, D. C. (1999). "Effects of nonsteroidal anti-inflammatory drugs on renal function: focus on cyclooxygenase-2-selective inhibition." Am J Med **107**(6A): 65S-70S; discussion 70S-71S.

Brater, D. C., C. Harris, J. S. Redfern and B. J. Gertz (2001). "Renal effects of COX-2-selective inhibitors." Am J Nephrol **21**(1): 1-15.

Bresalier, R. S., R. S. Sandler, H. Quan, J. A. Bolognese, B. Oxenius, K. Horgan, C. Lines, R. Riddell, D. Morton, A. Lanas, M. A. Konstam and J. A. Baron (2005). "Cardiovascular events associated with rofecoxib in a colorectal adenoma chemoprevention trial." N Engl J Med **352**(11): 1092-102.

Breyer, M. D. and R. M. Breyer (2000). "Prostaglandin E receptors and the kidney." Am J Physiol Renal Physiol **279**(1): F12-23.

Breyer, M. D. and R. C. Harris (2001). "Cyclooxygenase 2 and the kidney." Curr Opin Nephrol Hypertens **10**(1): 89-98.

Breyer, M. D., Y. Zhang, Y. F. Guan, C. M. Hao, R. L. Hebert and R. M. Breyer (1998). "Regulation of renal function by prostaglandin E receptors." Kidney Int Suppl **67**: S88-94.

Brogden, R. N., R. C. Heel, T. M. Speight and G. S. Avery (1984). "Piroxicam. A reappraisal of its pharmacology and therapeutic efficacy." Drugs **28**(4): 292-323.

Brune, K. and B. Hinz (2004). "Selective cyclooxygenase-2 inhibitors: similarities and differences." Scand J Rheumatol **33**(1): 1-6.

Buerkle, M. A., S. Lehrer, H. Y. Sohn, P. Conzen, U. Pohl and F. Krotz (2004). "Selective inhibition of cyclooxygenase-2 enhances platelet adhesion in hamster arterioles in vivo." Circulation **110**(14): 2053-9.

Buttar, N. S. and K. K. Wang (2000). "The "aspirin" of the new millennium: cyclooxygenase-2 inhibitors." Mayo Clin Proc **75**(10): 1027-38.

Capone, M. L., S. Tacconelli, M. G. Sciulli, M. Grana, E. Ricciotti, P. Minuz, P. Di Gregorio, G. Merciaro, C. Patrono and P. Patrignani (2004). "Clinical pharmacology of platelet, monocyte, and vascular cyclooxygenase inhibition by naproxen and low-dose aspirin in healthy subjects." Circulation **109**(12): 1468-71.

Catella-Lawson, F., B. McAdam, B. W. Morrison, S. Kapoor, D. Kujubu, L. Antes, K. C. Lasseter, H. Quan, B. J. Gertz and G. A. FitzGerald (1999). "Effects of specific inhibition of cyclooxygenase-2 on sodium balance, hemodynamics, and vasoactive eicosanoids." J Pharmacol Exp Ther **289**(2): 735-41.

Catella-Lawson, F., M. P. Reilly, S. C. Kapoor, A. J. Cucchiara, S. DeMarco, B. Tournier, S. N. Vyas and G. A. FitzGerald (2001). "Cyclooxygenase inhibitors and the antiplatelet effects of aspirin." N Engl J Med **345**(25): 1809-17.

Chakraborty, I., S. K. Das, J. Wang and S. K. Dey (1996). "Developmental expression of the cyclo-oxygenase-1 and cyclo-oxygenase-2 genes in the peri-implantation mouse uterus and their differential regulation by the blastocyst and ovarian steroids." J Mol Endocrinol **16**(2): 107-22.

Chalmers, I. and R. B. Haynes (1995). Reporting, updating and correcting systematic reviews of the effects of health care. Systematic reviews. I. Chalmers and D. G.

Altman. London, BMJ Publishing Group: 86-95.

Chalmers, I., L. V. Hedges and H. Cooper (2002). "A brief history of research synthesis." Eval Health Prof **25**(1): 12-37.

Chan, B. S., J. A. Satriano, M. Pucci and V. L. Schuster (1998). "Mechanism of prostaglandin E2 transport across the plasma membrane of HeLa cells and Xenopus oocytes expressing the prostaglandin transporter "PGT"." J Biol Chem **273**(12): 6689-97.

Chan, C. C., S. Boyce, C. Brideau, S. Charleson, W. Cromlish, D. Ethier, J. Evans, A. W. Ford-Hutchinson, M. J. Forrest, J. Y. Gauthier, R. Gordon, M. Gresser, J. Guay, S. Kargman, B. Kennedy, Y. Leblanc, S. Leger, J. Mancini, G. P. O'Neill, M. Ouellet, D. Patrick, M. D. Percival, H. Perrier, P. Prasit, I. Rodger and et al. (1999). "Rofecoxib [Vioxx, MK-0966; 4-(4'-methylsulfonylphenyl)-3-phenyl-2-(5H)-furanone]: a potent and orally active cyclooxygenase-2 inhibitor. Pharmacological and biochemical profiles." J Pharmacol Exp Ther **290**(2): 551-60.

Chandrasekharan, N. V., H. Dai, K. L. Roos, N. K. Evanson, J. Tomsik, T. S. Elton and D. L. Simmons (2002). "COX-3, a cyclooxygenase-1 variant inhibited by acetaminophen and other analgesic/antipyretic drugs: cloning, structure, and expression." Proc Natl Acad Sci U S A **99**(21): 13926-31.

Chavez, M. L. and C. J. DeKorte (2003). "Valdecoxib: a review." Clin Ther **25**(3): 817-51.

Chen, J. K., D. W. Wang, J. R. Falck, J. Capdevila and R. C. Harris (1999). "Transfection of an active cytochrome P450 arachidonic acid epoxygenase indicates that 14,15-epoxyeicosatrienoic acid functions as an intracellular second messenger in response to epidermal growth factor." J Biol Chem **274**(8): 4764-9.

Chen, L. C., R. A. Elliott and D. M. Ashcroft (2004). "Systematic review of the analgesic efficacy and tolerability of COX-2 inhibitors in post-operative pain control." J Clin Pharm Ther **29**(3): 215-29.

Chenevard, R., D. Hurlimann, M. Bechir, F. Enseleit, L. Spieker, M. Hermann, W. Riesen, S. Gay, R. E. Gay, M. Neidhart, B. Michel, T. F. Luscher, G. Noll and F. Ruschitzka (2003). "Selective COX-2 inhibition improves endothelial function in coronary artery disease." Circulation **107**(3): 405-9.

Cheng, H. F., J. L. Wang, M. Z. Zhang, Y. Miyazaki, I. Ichikawa, J. A. McKanna and R.

C. Harris (1999). "Angiotensin II attenuates renal cortical cyclooxygenase-2 expression." J Clin Invest **103**(7): 953-61.

Cho, M. and L. Bero (1996). "The quality of drug studies published in symposium proceedings." Ann Intern Med **124**: 485-9.

Cho, M. K. and L. A. Bero (1996). "The quality of drug studies published in symposium proceedings." Ann Intern Med **124**(5): 485-9.

Choi, H. K., M. A. Hernan, J. D. Seeger, J. M. Robins and F. Wolfe (2002). "Methotrexate and mortality in patients with rheumatoid arthritis: a prospective study." Lancet **359**(9313): 1173-7.

Cochrane, D. J., B. Jarvis and G. M. Keating (2002). "Etoricoxib." Drugs **62**(18): 2637-51; discussion 2652-3.

Collaboration, A. T. (2002). "Collaborative meta-analysis of randomised trials of antiplatelet therapy for prevention of death, myocardial infarction, and stroke in high risk patients." Bmj **324**(7329): 71-86.

Conte, G., B. Cianciaruso, L. De Nicola, V. Sepe, G. Romano, R. Domenico, A. Caglioti, G. Fuiano and A. Dal Canton (1992). "Inhibition of urea tubular reabsorption by PGE1 infusion in man." Nephron **60**(1): 42-8.

Cook, D. (1997). "Systematic reviews: the case for rigorous methods and rigorous reporting." Can J Anaesth **44**(4): 350-3.

Cook, D. J., C. D. Mulrow and R. B. Haynes (1997). "Systematic reviews: synthesis of best evidence for clinical decisions." Ann Intern Med **126**(5): 376-80.

Cook, D. J., D. L. Sackett and W. O. Spitzer (1995). "Methodologic guidelines for systematic reviews of randomized control trials in health care from the Potsdam Consultation on Meta-Analysis." J Clin Epidemiol **48**(1): 167-71.

Cowley, A. W., Jr., D. L. Mattson, S. Lu and R. J. Roman (1995). "The renal medulla and hypertension." Hypertension **25**(4 Pt 2): 663-73.

Cox, E. R., M. Frisse, A. Behm and K. A. Fairman (2004). "Over-the-counter pain reliever and aspirin use within a sample of long-term cyclooxygenase 2 users." Arch Intern Med **164**(11): 1243-6.

(CPMP), Committee For Proprietary Medicinal Products (2002) Note For Guidance on Clinical Investigation Of Medicinal Products For Treatment On Nociceptive Pain

EMEA London, Available from: (www.emea.eu.int/pdfs/human/ewp/061200eu.pdf) [Accessed on: 5th January 2006]

Creamer, P. and M. C. Hochberg (1997). "Osteoarthritis." Lancet **350**(9076): 503-8.

Criswell, L. A., L. A. Merlino, J. R. Cerhan, T. R. Mikuls, A. S. Mudano, M. Burma, A. R. Folsom and K. G. Saag (2002). "Cigarette smoking and the risk of rheumatoid arthritis among postmenopausal women: results from the Iowa Women's Health Study." Am J Med **112**(6): 465-71.

Crofford, L. J., P. E. Lipsky, P. Brooks, S. B. Abramson, L. S. Simon and L. B. van de Putte (2000). "Basic biology and clinical application of specific cyclooxygenase-2 inhibitors." Arthritis Rheum **43**(1): 4-13.

Crombie, I. (1984). "Inherent limitations of the yellow card system for the detection of unsuspected adverse drug reactions." Hum Toxicol **3**(4): 261-9.

Cronstein, B. N. (2002). "Cyclooxygenase-2-selective inhibitors: translating pharmacology into clinical utility." Cleve Clin J Med **69 Suppl 1**: SI13-9.

Cryer, B. and A. Dubois (1998). "The advent of highly selective inhibitors of cyclooxygenase--a review." Prostaglandins Other Lipid Mediat **56**(5-6): 341-61.

Cryer, B. and M. Feldman (1998). "Cyclooxygenase-1 and cyclooxygenase-2 selectivity of widely used nonsteroidal anti-inflammatory drugs." Am J Med **104**(5): 413-21.

Damm, J., T. Rau, C. Maihofner, A. Pahl and K. Brune (2001). "Constitutive expression and localization of COX-1 and COX-2 in rabbit iris and ciliary body." Exp Eye Res **72**(6): 611-21.

Daniels, B. and B. Seidenberg (1999). "Cardiovascular safety profile of rofecoxib in controlled clinical trials." Arthritis Rheum **42**(Suppl): S143.

Davidson, R. A. (1986). "Source of funding and outcome of clinical trials." J Gen Intern Med **1**: 155-8.

Davies, B. and T. Morris (1993). "Physiological parameters in laboratory animals and humans." Pharm Res **10**(7): 1093-5.

Davies, N. M., A. J. McLachlan, R. O. Day and K. M. Williams (2000). "Clinical pharmacokinetics and pharmacodynamics of celecoxib: a selective cyclo-oxygenase-2 inhibitor." Clin Pharmacokinet **38**(3): 225-42.

Davies, N. M., X. W. Teng and N. M. Skjodt (2003). "Pharmacokinetics of rofecoxib: a specific cyclo-oxygenase-2 inhibitor." Clin Pharmacokinet **42**(6): 545-56.

de Leval, X., J. Delarge, P. Devel, P. Neven, C. Michaux, B. Masereel, B. Pirotte, J. L. David, Y. Henrotin and J. M. Dogne (2001). "Evaluation of classical NSAIDs and COX-2 selective inhibitors on purified ovine enzymes and human whole blood." Prostaglandins Leukot Essent Fatty Acids **64**(4-5): 211-6.

del Rincon, I. D., K. Williams, M. P. Stern, G. L. Freeman and A. Escalante (2001). "High incidence of cardiovascular events in a rheumatoid arthritis cohort not explained by traditional cardiac risk factors." Arthritis Rheum **44**(12): 2737-45.

Denton, J. and H. K. Beecher (1949). "New analgesics." J. Am. Med. Assoc. **141**: 1051-7.

Depre, M., E. Ehrich, A. Van Hecken, I. De Lepeleire, A. Dallob, P. Wong, A. Porras, B. J. Gertz and P. J. De Schepper (2000). "Pharmacokinetics, COX-2 specificity, and tolerability of supratherapeutic doses of rofecoxib in humans." Eur J Clin Pharmacol **56**(2): 167-74.

Dequeker, J. and P. A. Dieppe (1998). Disorders of bone cartilage and connective tissue. London, Mosby.

Desjardins, P. J., D. R. Mehlisch, D. J. Chang, D. Krupa, A. B. Polis, R. A. Petruschke, K. Malmstrom and G. P. Geba (2005). "The time to onset and overall analgesic efficacy of rofecoxib 50 mg: a meta-analysis of 13 randomized clinical trials." Clin J Pain **21**(3): 241-50.

Dessein, P. H., B. I. Joffe and S. Singh (2005). "Biomarkers of endothelial dysfunction, cardiovascular risk factors and atherosclerosis in rheumatoid arthritis." Arthritis Res Ther **7**(3): R634-43.

Dessein, P. H., B. I. Joffe and A. E. Stanwix (2003). "Inflammation, insulin resistance, and aberrant lipid metabolism as cardiovascular risk factors in rheumatoid arthritis." J Rheumatol **30**(7): 1403-5.

DeWitt, D. L., E. A. Meade and W. L. Smith (1993). "PGH synthase isoenzyme selectivity: the potential for safer nonsteroidal antiinflammatory drugs." Am J Med **95**(2A): 40S-44S.

Di Minno, G., M. J. Silver and S. Murphy (1983). "Monitoring the entry of new platelets into the circulation after ingestion of aspirin." Blood **61**(6): 1081-5.

Dickersin, K. and J. A. Berlin (1992). "Meta-analysis: state-of-the-science." Epidemiol Rev **14**: 154-76.

Dickersin, K. and Y. I. Min (1993a). "Publication bias: the problem that won't go away." Ann N Y Acad Sci **703**: 135-46; discussion 146-8.

Dickersin, K. and Y. I. Min (1993b). "NIH clinical trials and publication bias." Online J Curr Clin Trials **Doc No 50**: [4967 words; 53 paragraphs].

Dickersin, K., Y. I. Min and C. L. Meinert (1992). "Factors influencing publication of research results. Follow-up of applications submitted to two institutional review boards." Jama **267**(3): 374-8.

Dieppe, P. A., S. Ebrahim, R. M. Martin and P. Juni (2004). "Lessons from the withdrawal of rofecoxib." Bmj **329**(7471): 867-8.

Dietz, R., W. Nastainczyk and H. H. Ruf (1988). "Higher oxidation states of prostaglandin H synthase. Rapid electronic spectroscopy detected two spectral intermediates during the peroxidase reaction with prostaglandin G2." Eur J Biochem **171**(1-2): 321-8.

Dingle, H. (2002) Mersey ADR Newsletter. Winter 2002. No. 19 CSM, Mersey Liverpool, UK, Available from: (http://www.liv.ac.uk/~druginfo/csm/ADR_Bulletin19.pdf#search=%22mersey%20newsletter%22) [Accessed on: 25th August 2006]

Dougados, M., P. Leclaire, D. van der Heijde, D. A. Bloch, N. Bellamy and R. D. Altman (2000). "Response criteria for clinical trials on osteoarthritis of the knee and hip: a report of the Osteoarthritis Research Society International Standing Committee for Clinical Trials response criteria initiative." Osteoarthritis Cartilage **8**(6): 395-403.

Dubois, R. N., S. B. Abramson, L. Crofford, R. A. Gupta, L. S. Simon, L. B. Van De Putte and P. E. Lipsky (1998). "Cyclooxygenase in biology and disease." Faseb J **12**(12): 1063-73.

Easterbrook, P. J., J. A. Berlin, R. Gopalan and D. R. Matthews (1991). "Publication bias in clinical research." Lancet **337**(8746): 867-72.

Eddy, D. M. (2005). "Evidence-based medicine: a unified approach." Health Aff (Millwood) **24**(1): 9-17.

Edwards, J. E., H. J. McQuay, R. A. Moore and S. L. Collins (1999). "Reporting of

adverse effects in clinical trials should be improved: lessons from acute postoperative pain." J Pain Symptom Manage **18**(6): 427-37.

Edwards, J. E., R. A. Moore and H. J. McQuay (2004a). "Individual patient meta-analysis of single-dose rofecoxib in postoperative pain." BMC Anesthesiol **4**(1): 3.

Edwards, J. E., R. A. Moore and H. J. McQuay (2004b). "Rofecoxib for dysmenorrhoea: meta-analysis using individual patient data." BMC Womens Health **4**(1): 5.

Edwards, J. E., A. D. Oldman, L. A. Smith, D. Carroll, P. J. Wiffen, H. J. McQuay and R. A. Moore (1999). "Oral aspirin in postoperative pain: a quantitative systematic review." Pain **81**(3): 289-97.

Egger, M. and G. Davey Smith (2001). Principles of and procedures for systematic reviews. Systematic Reviews in Health Care: Meta-analysis in context. M. Egger, G. Davey Smith and D. G. Altman. London, BMJ Publishing Group**:** 23-42.

Egger, M., G. Davey Smith and D. G. Altman (2001). Systematic reviews in Health Care: Meta-analysis in context. London, BMJ Publishing Group.

Egger, M., K. Dickersin and G. D. Smith (2001). Problems and limitation in conducting systematic reviews. Systematic Reviews in Health Care: Meta-analysis in context. M. Egger, G. Davey Smith and D. G. Altman. London, BMJ Publishing Group**:** 43-68.

Egger, M. and G. D. Smith (1998). "Bias in location and selection of studies." Bmj **316**(7124): 61-6.

Ehara, S., M. Ueda, T. Naruko, K. Haze, A. Itoh, M. Otsuka, R. Komatsu, T. Matsuo, H. Itabe, T. Takano, Y. Tsukamoto, M. Yoshiyama, K. Takeuchi, J. Yoshikawa and A. E. Becker (2001). "Elevated levels of oxidized low density lipoprotein show a positive relationship with the severity of acute coronary syndromes." Circulation **103**(15): 1955-60.

Ehrich, E. W., A. Dallob, I. De Lepeleire, A. Van Hecken, D. Riendeau, W. Yuan, A. Porras, J. Wittreich, J. R. Seibold, P. De Schepper, D. R. Mehlisch and B. J. Gertz (1999). "Characterization of rofecoxib as a cyclooxygenase-2 isoform inhibitor and demonstration of analgesia in the dental pain model." Clin Pharmacol Ther **65**(3): 336-47.

Elders, M. J. (2000). "The increasing impact of arthritis on public health." J Rheumatol

Suppl **60**: 6-8.

(EMEA), The European Agency for the Evaluation of Medicinal Products. (2002) Committee for Proprietary Medicinal Products 23-25 July 2002 Plenary Meeting Monthly Report London, Available from: (http://www.emea.eu.int/pdfs/human/press/pr/347302en.pdf) [Accessed on: 1st September]

(EMEA), The European Agency for the Evaluation of Medicinal Products. (2005) Press Release: European Medicines Agency concludes action on COX-2 Inhibitors London, Available from: (http://www.emea.eu.int/pdfs/human/press/pr/20776605en.pdf) [Accessed on: 1st September 2006]

Engelhardt, G., R. Bogel, C. Schnitzer and R. Utzmann (1996). "Meloxicam: influence on arachidonic acid metabolism. Part 1. In vitro findings." Biochem Pharmacol **51**(1): 21-8.

Evans, W. E. and M. V. Relling (1999). "Pharmacogenomics: translating functional genomics into rational therapeutics." Science **286**(5439): 487-91.

Farkouh, M. E., H. Kirshner, R. A. Harrington, S. Ruland, F. W. Verheugt, T. J. Schnitzer, G. R. Burmester, E. Mysler, M. C. Hochberg, M. Doherty, E. Ehrsam, X. Gitton, G. Krammer, B. Mellein, A. Gimona, P. Matchaba, C. J. Hawkey and J. H. Chesebro (2004). "Comparison of lumiracoxib with naproxen and ibuprofen in the Therapeutic Arthritis Research and Gastrointestinal Event Trial (TARGET), cardiovascular outcomes: randomised controlled trial." Lancet **364**(9435): 675-84.

FDA, US Food and Drug Administration. (2001) Vioxx Medical Review, Rockville, Maryland, Available from: (http://www.fda.gov/) [Accessed on: 11th July 2006]

(FDA), US Food and Drug Administration. (2004) Bextra Label Updated with Boxed Warning Concerning Severe Skin Reactions and Warning Regarding Cardiovascular Risk, Rockville, Maryland, Available from: (http://www.fda.gov/bbs/topics/answers/2004/ans01331.html) [Accessed on: 11th July 2006]

(FDA), US Food and Drug Administration. (2005a) COX-2 Selective (includes Bextra, Celebrex, and Vioxx) and Non-Selective Non-Steroidal Anti-Inflammatory Drugs

(NSAIDs) Rockville, Maryland, 18th July 2005 Available from: (http://www.fda.gov/cder/drug/infopage/cox2/) [Accessed on: 17th July 2006]

(FDA), US Food and Drug Administration. (2005b) FDA announces series of changes to the class of marketed non-steroidal anti-inflammatory drugs (NSAIDs), Rockville, Maryland, Available from: (http://www.fda.gov/bbs/topics/news/2005/NEW01171.html) [Accessed on: 11th July 2006]

Felson, D. T., R. C. Lawrence, P. A. Dieppe, R. Hirsch, C. G. Helmick, J. M. Jordan, R. S. Kington, N. E. Lane, M. C. Nevitt, Y. Zhang, M. Sowers, T. McAlindon, T. D. Spector, A. R. Poole, S. Z. Yanovski, G. Ateshian, L. Sharma, J. A. Buckwalter, K. D. Brandt and J. F. Fries (2000). "Osteoarthritis: new insights. Part 1: the disease and its risk factors." Ann Intern Med **133**(8): 635-46.

Feng, L., W. Sun, Y. Xia, W. W. Tang, P. Chanmugam, E. Soyoola, C. B. Wilson and D. Hwang (1993). "Cloning two isoforms of rat cyclooxygenase: differential regulation of their expression." Arch Biochem Biophys **307**(2): 361-8.

Fine, P. G. (2002). "The role of rofecoxib, a cyclooxygenase-2-specific inhibitor, for the treatment of non-cancer pain: a review." J Pain **3**(4): 272-83.

Fitzgerald, D. J., L. Roy, F. Catella and G. A. FitzGerald (1986). "Platelet activation in unstable coronary disease." N Engl J Med **315**(16): 983-9.

FitzGerald, G. A. (1991). "Mechanisms of platelet activation: thromboxane A2 as an amplifying signal for other agonists." Am J Cardiol **68**(7): 11B-15B.

FitzGerald, G. A. (2003). "COX-2 and beyond: Approaches to prostaglandin inhibition in human disease." Nat Rev Drug Discov **2**(11): 879-90.

Fitzgerald, G. A. (2004). "Coxibs and cardiovascular disease." N Engl J Med **351**(17): 1709-11.

FitzGerald, G. A. and C. Patrono (2001). "The coxibs, selective inhibitors of cyclooxygenase-2." N Engl J Med **345**(6): 433-42.

FitzGerald, G. A., B. Smith, A. K. Pedersen and A. R. Brash (1984). "Increased prostacyclin biosynthesis in patients with severe atherosclerosis and platelet activation." N Engl J Med **310**(17): 1065-8.

Flower, R. J. and J. R. Vane (1972). "Inhibition of prostaglandin synthetase in brain

explains the anti-pyretic activity of paracetamol (4-acetamidophenol)." Nature **240**(5381): 410-1.

Ford-Hutchinson, A. W. (1997). New highly selective COX-2 inhibitors. Selective COX-2 inhibitors: pharmacology, clinical effects and therapeutic potential. J. R. Vane and J. Botting. London, Kluwer Academic Publishers and William Harvey Press**:** 117-25.

Frazier, L. W. and T. Yorio (1992). "Eicosanoids: their function in renal epithelia ion transport." Proc Soc Exp Biol Med **201**(3): 229-43.

Frolich, J. C. (1997). "A classification of NSAIDs according to the relative inhibition of cyclooxygenase isoenzymes." Trends Pharmacol Sci **18**(1): 30-4.

Fu, J. Y., J. L. Masferrer, K. Seibert, A. Raz and P. Needleman (1990). "The induction and suppression of prostaglandin H2 synthase (cyclooxygenase) in human monocytes." J Biol Chem **265**(28): 16737-40.

Funk, C. D., L. B. Funk, M. E. Kennedy, A. S. Pong and G. A. Fitzgerald (1991). "Human platelet/erythroleukemia cell prostaglandin G/H synthase: cDNA cloning, expression, and gene chromosomal assignment." Faseb J **5**(9): 2304-12.

Furberg, C. D., B. M. Psaty and G. A. FitzGerald (2005). "Parecoxib, valdecoxib, and cardiovascular risk." Circulation **111**(3): 249.

Gabriel, S. E., L. Jaakkimainen and C. Bombardier (1991). "Risk for serious gastrointestinal complications related to use of nonsteroidal anti-inflammatory drugs. A meta-analysis." Ann Intern Med **115**(10): 787-96.

Gagnier, J. J., S. Chrubasik and E. Manheimer (2004). "Harpgophytum procumbens for osteoarthritis and low back pain: a systematic review." BMC Complement Altern Med **4**: 13.

Garcia Rodriguez, L. A., C. Varas-Lorenzo, A. Maguire and A. Gonzalez-Perez (2004). "Nonsteroidal antiinflammatory drugs and the risk of myocardial infarction in the general population." Circulation **109**(24): 3000-6.

Garner, S., D. Fidan, R. Frankish, M. Judd, T. Towheed, G. Wells and P. Tugwell (2002). "Rofecoxib for the treatment of rheumatoid arthritis." Cochrane Database Syst Rev(3): CD003685.

Garner, S., D. Fidan, R. Frankish, M. Judd, T. Towheed, G. Wells and P. Tugwell (2005a).

"Rofecoxib for rheumatoid arthritis." Cochrane Database Syst Rev(1): CD003685.

Garner, S., D. Fidan, R. Frankish and L. Maxwell (2005b). "Rofecoxib for osteoarthritis." Cochrane Database Syst Rev(1): CD005115.

Geba, G., A. Polis, A. Petruschke and W. Keane (2003). "Hypertension among osteoarthritis patients treated with rofecoxib, celecoxib, or acetaminophen: an analysis of the data from pooled VACT studies." American Journal of Hypertension **16**(5,part 2): 39A.

Geba, G. P., A. L. Weaver, A. B. Polis, M. E. Dixon and T. J. Schnitzer (2002). "Efficacy of rofecoxib, celecoxib, and acetaminophen in osteoarthritis of the knee: a randomized trial." Jama **287**(1): 64-71.

Geller, N. L. and M. Proschan (1996). "Meta-analysis of clinical trials: a consumer's guide." J Biopharm Stat **6**(4): 377-94.

Gertz, B. J., D. Krupa, J. A. Bolognese, R. S. Sperling and A. Reicin (2002). "A comparison of adverse renovascular experiences among osteoarthritis patients treated with rofecoxib and comparator non-selective non-steroidal anti-inflammatory agents." Curr Med Res Opin **18**(2): 82-91.

Giercksky, K. E., G. Husby, H. E. Rugstad and A. Revhaug (1988). "Epidemiology of NSAID-induced gastrointestinal problems and the role of cimetidine in their prevention." Aliment Pharmacol Ther **2 Suppl 1**: 33-41.

Gierse, J. K., J. J. McDonald, S. D. Hauser, S. H. Rangwala, C. M. Koboldt and K. Seibert (1996). "A single amino acid difference between cyclooxygenase-1 (COX-1) and -2 (COX-2) reverses the selectivity of COX-2 specific inhibitors." J Biol Chem **271**(26): 15810-4.

Gimbrone, M. A., Jr., J. N. Topper, T. Nagel, K. R. Anderson and G. Garcia-Cardena (2000). "Endothelial dysfunction, hemodynamic forces, and atherogenesis." Ann N Y Acad Sci **902**: 230-9; discussion 239-40.

Goldschmidt, P. G. (1986). "Information synthesis: a practical guide." Health Serv Res **21**(2 Pt 1): 215-37.

Goldstein, J. L., A. E. Bello, W. Spalding, S. Suh and J. G. Fort (2005). "Cyclooxygenase-2 specific inhibitors and upper gastrointestinal tolerability in patients with osteoarthritis receiving concomitant low dose aspirin: pooled analysis of 2 trials." J

Rheumatol **32**(1): 111-7.

Gomez Cerezo, J., R. Lubomirov Hristov, A. J. Carcas Sansuan and J. J. Vazquez Rodriguez (2003). "Outcome trials of COX-2 selective inhibitors: global safety evaluation does not promise benefits." Eur J Clin Pharmacol **59**(2): 169-75.

Goppelt-Struebe, M. (1995). "Regulation of prostaglandin endoperoxide synthase (cyclooxygenase) isozyme expression." Prostaglandins Leukot Essent Fatty Acids **52**(4): 213-22.

Gotzsche, P. C. (1987). "Reference bias in reports of drug trials." Br Med J (Clin Res Ed) **295**(6599): 654-6.

Gotzsche, P. C. (2001). "Reporting of outcomes in arthritis trials measured on ordinal and interval scales is inadequate in relation to meta-analysis." Ann Rheum Dis **60**(4): 349-52.

Graham, D. J., D. Campen, R. Hui, M. Spence, C. Cheetham, G. Levy, S. Shoor and W. A. Ray (2005). "Risk of acute myocardial infarction and sudden cardiac death in patients treated with cyclo-oxygenase 2 selective and non-selective non-steroidal anti-inflammatory drugs: nested case-control study." Lancet **365**(9458): 475-81.

Graham, G. G. and K. F. Scott (2003). "Mechanisms of action of paracetamol and related analgesics." Inflammopharmacology **11**(4): 401-13.

Greenberg, S. G., J. N. Lorenz, X. R. He, J. B. Schnermann and J. P. Briggs (1993). "Effect of prostaglandin synthesis inhibition on macula densa-stimulated renin secretion." Am J Physiol **265**(4 Pt 2): F578-83.

Greenhalgh, T. (1997). "Papers that summarise other papers (systematic reviews and meta-analyses)." Bmj **315**(7109): 672-5.

Gregoire, G., F. Derderian and J. Le Lorier (1995). "Selecting the language of the publications included in a meta-analysis: is there a Tower of Babel bias?" J Clin Epidemiol **48**(1): 159-63.

Grobbee, D. E. and A. W. Hoes (1997). "Confounding and indication for treatment in evaluation of drug treatment for hypertension." Bmj **315**(7116): 1151-4.

Grossman, C. J., J. Wiseman, F. S. Lucas, M. A. Trevethick and P. J. Birch (1995). "Inhibition of constitutive and inducible cyclooxygenase activity in human platelets and mononuclear cells by NSAIDs and Cox 2 inhibitors." Inflamm Res **44**(6): 253-

7.

Guillemin, F., A. Saraux, P. Guggenbuhl, C. H. Roux, P. Fardellone, E. Le Bihan, A. Cantagrel, I. Chary-Valckenaere, L. Euller-Ziegler, R. M. Flipo, R. Juvin, J. M. Behier, B. Fautrel, C. Masson and J. Coste (2005). "Prevalence of rheumatoid arthritis in France: 2001." Ann Rheum Dis **64**(10): 1427-30.

Gussak, I., J. Litwin, R. Kleiman, S. Grisanti and J. Morganroth (2004). "Drug-induced cardiac toxicity: emphasizing the role of electrocardiography in clinical research and drug development." J Electrocardiol **37**(1): 19-24.

Haller, D. G. (2003). "COX-2 inhibitors in oncology." Semin Oncol **30**(4 Suppl 12): 2-8.

Halpin, R. A., L. A. Geer, K. E. Zhang, T. M. Marks, D. C. Dean, A. N. Jones, D. Melillo, G. Doss and K. P. Vyas (2000). "The absorption, distribution, metabolism and excretion of rofecoxib, a potent and selective cyclooxygenase-2 inhibitor, in rats and dogs." Drug Metab Dispos **28**(10): 1244-54.

Halpin, R. A., A. G. Porras, L. A. Geer, M. R. Davis, D. Cui, G. A. Doss, E. Woolf, D. Musson, C. Matthews, R. Mazenko, J. I. Schwartz, K. C. Lasseter, K. P. Vyas and T. A. Baillie (2002). "The disposition and metabolism of rofecoxib, a potent and selective cyclooxygenase-2 inhibitor, in human subjects." Drug Metab Dispos **30**(6): 684-93.

Hao, C. M., F. Yull, T. Blackwell, M. Komhoff, L. S. Davis and M. D. Breyer (2000). "Dehydration activates an NF-kappaB-driven, COX2-dependent survival mechanism in renal medullary interstitial cells." J Clin Invest **106**(8): 973-82.

Hara, S., A. Miyata, C. Yokoyama, H. Inoue, R. Brugger, F. Lottspeich, V. Ullrich and T. Tanabe (1994). "Isolation and molecular cloning of prostacyclin synthase from bovine endothelial cells." J Biol Chem **269**(31): 19897-903.

Harding, P., D. H. Sigmon, M. E. Alfie, P. L. Huang, M. C. Fishman, W. H. Beierwaltes and O. A. Carretero (1997). "Cyclooxygenase-2 mediates increased renal renin content induced by low-sodium diet." Hypertension **29**(1 Pt 2): 297-302.

Hardy, J. R. and S. G. Thompson (1998). "Detecting and describing heterogeneity in meta-analysis." Stat. Med. **17**: 841-56.

Harris, R. C., J. A. McKanna, Y. Akai, H. R. Jacobson, R. N. Dubois and M. D. Breyer (1994). "Cyclooxygenase-2 is associated with the macula densa of rat kidney and

increases with salt restriction." J Clin Invest **94**(6): 2504-10.

Hartner, A., M. Goppelt-Struebe and K. F. Hilgers (1998). "Coordinate expression of cyclooxygenase-2 and renin in the rat kidney in renovascular hypertension." Hypertension **31**(1 Pt 2): 201-5.

Hawkey, C. J. (1999). "COX-2 inhibitors." Lancet **353**(9149): 307-14.

Haynes, R. B. (1993). "Where's the meat in clinical journals?" ACP J Club **119**: A23-4.

Helliwell, R. J., L. F. Adams and M. D. Mitchell (2004). "Prostaglandin synthases: recent developments and a novel hypothesis." Prostaglandins Leukot Essent Fatty Acids **70**(2): 101-13.

Hemels, M. E., C. Vicente, H. Sadri, M. J. Masson and T. R. Einarson (2004). "Quality assessment of meta-analyses of RCTs of pharmacotherapy in major depressive disorder." Curr Med Res Opin **20**(4): 477-84.

Hemler, M. and W. E. Lands (1976). "Purification of the cyclooxygenase that forms prostaglandins. Demonstration of two forms of iron in the holoenzyme." J Biol Chem **251**(18): 5575-9.

Hemminki, E. (1980). "Study of information submitted by drug companies to licensing authorities." Br Med J **280**(6217): 833-6.

Hennan, J. K., J. Huang, T. D. Barrett, E. M. Driscoll, D. E. Willens, A. M. Park, L. J. Crofford and B. R. Lucchesi (2001). "Effects of selective cyclooxygenase-2 inhibition on vascular responses and thrombosis in canine coronary arteries." Circulation **104**(7): 820-5.

Hermann, M., G. Camici, A. Fratton, D. Hurlimann, F. C. Tanner, J. P. Hellermann, M. Fiedler, J. Thiery, M. Neidhart, R. E. Gay, S. Gay, T. F. Luscher and F. Ruschitzka (2003). "Differential effects of selective cyclooxygenase-2 inhibitors on endothelial function in salt-induced hypertension." Circulation **108**(19): 2308-11.

Hersh, E. V., E. T. Lally and P. A. Moore (2005). "Update on cyclooxygenase inhibitors: has a third COX isoform entered the fray?" Curr Med Res Opin **21**(8): 1217-26.

Hetherington, J., K. Dickersin, I. Chalmers and C. L. Meinert (1989). "Retrospective and prospective identification of unpublished controlled trials: lessons from a survey of obstetricians and pediatricians." Pediatrics **84**(2): 374-80.

Higgins, J. P., S. G. Thompson, J. J. Deeks and D. G. Altman (2003). "Measuring

inconsistency in meta-analyses." Bmj **327**(7414): 557-60.

Hinz, B. and K. Brune (2002). "Cyclooxygenase-2--10 years later." J Pharmacol Exp Ther **300**(2): 367-75.

Hinz, B., K. Brune and A. Pahl (2000). "Cyclooxygenase-2 expression in lipopolysaccharide-stimulated human monocytes is modulated by cyclic AMP, prostaglandin E(2), and nonsteroidal anti-inflammatory drugs." Biochem Biophys Res Commun **278**(3): 790-6.

Hla, T. and T. Maciag (1991). "Cyclooxygenase gene expression is down-regulated by heparin-binding (acidic fibroblast) growth factor-1 in human endothelial cells." J Biol Chem **266**(35): 24059-63.

Hoode, R. W. (1962). On appraising pain ans analgesic drugs. The assessment of pain in man & animals. K. C. A. and R. Smith. Edinburgh, Livingstone**:** 2002-12.

Hoode, R. W. and S. Wallenstein (1954). "Comparisons of oral vs parenteral effectiveness of some analgesics in patients with pain due to cancer." Fed. Proc. **13**: 367-8.

Hooper, L., T. J. Brown, R. Elliott, K. Payne, C. Roberts and D. Symmons (2004). "The effectiveness of five strategies for the prevention of gastrointestinal toxicity induced by non-steroidal anti-inflammatory drugs: systematic review." Bmj **329**(7472): 948.

Horton, R. (1999). "The information wars." Lancet **353**(9148): 164-5.

(IASP), International Association for the Study of Pain. (1994) Classification of Chronic Pain IASP Press Seattle, 09th November 2004, Available from: (http://www.iasp-pain.org/terms-p.html) [Accessed on: 19th December 2005]

Ilowite, N. T. (2002). "Current treatment of juvenile rheumatoid arthritis." Pediatrics **109**(1): 109-15.

Ioannidis, J. P., D. O. Dixon, M. McIntosh, J. M. Albert, S. A. Bozzette and S. M. Schnittman (1999). "Relationship between event rates and treatment effects in clinical site differences within multicenter trials: an example from primary Pneumocystis carinii prophylaxis." Control Clin Trials **20**(3): 253-66.

Ioannidis, J. P., S. J. Evans, P. C. Gotzsche, R. T. O'Neill, D. G. Altman, K. Schulz and D. Moher (2004). "Better reporting of harms in randomized trials: an extension of the CONSORT statement." Ann Intern Med **141**(10): 781-8.

Iseki, S. (1995). "Immunocytochemical localization of cyclooxygenase-1 and cyclooxygenase-2 in the rat stomach." Histochem J **27**(4): 323-8.

Jadad, A. R., D. J. Cook and G. P. Browman (1997). "A guide to interpreting discordant systematic reviews." Cmaj **156**(10): 1411-6.

Jadad, A. R., R. A. Moore, D. Carroll, C. Jenkinson, D. J. Reynolds, D. J. Gavaghan and H. J. McQuay (1996). "Assessing the quality of reports of randomized clinical trials: is blinding necessary?" Control Clin Trials **17**(1): 1-12.

Jenkins, J. K. and P. J. Seligman 2005 Memorandum: Analysis and recommendations for Agency action regarding non-steroidal anti-inflammatory drugs and cardiovascular risk. Rockville, Maryland, Available from: (http://www.fda.gov/cder/drug/infopage/cox2/NSAIDdecisionMemo.pdf) [Accessed on: 17th July 2006]

Jick, S. S. (2000). "The risk of gastrointestinal bleed, myocardial infarction, and newly diagnosed hypertension in users of meloxicam, diclofenac, naproxen, and piroxicam." Pharmacotherapy **20**(7): 741-4.

Jones, C. (2002). "Research methods." The Pharmaceutical Journal **268**: 839-877.

Jordan, K. M., N. K. Arden, M. Doherty, B. Bannwarth, J. W. Bijlsma, P. Dieppe, K. Gunther, H. Hauselmann, G. Herrero-Beaumont, P. Kaklamanis, S. Lohmander, B. Leeb, M. Lequesne, B. Mazieres, E. Martin-Mola, K. Pavelka, A. Pendleton, L. Punzi, U. Serni, B. Swoboda, G. Verbruggen, I. Zimmerman-Gorska and M. Dougados (2003). "EULAR Recommendations 2003: an evidence based approach to the management of knee osteoarthritis: Report of a Task Force of the Standing Committee for International Clinical Studies Including Therapeutic Trials (ESCISIT)." Ann Rheum Dis **62**(12): 1145-55.

Juni, P., P. Dieppe and M. Egger (2002). "Risk of myocardial infarction associated with selective COX-2 inhibitors: questions remain." Arch Intern Med **162**(22): 2639-40; author reply 2640-2.

Juni, P., L. Nartey, S. Reichenbach, R. Sterchi, P. A. Dieppe and M. Egger (2004). "Risk of cardiovascular events and rofecoxib: cumulative meta-analysis." Lancet **364**(9450): 2021-9.

Juni, P., S. Reichenbach, P. Dieppe and M. Egger (2005). "Discontinuation of Vioox:

Author's reply." Lancet **365**(9453): 26-27.

Kannel, W. B. (2000). "Fifty years of Framingham Study contributions to understanding hypertension." J Hum Hypertens **14**(2): 83-90.

Kaplan-Machlis, B. and B. S. Klostermeyer (1999). "The cyclooxygenase-2 inhibitors: safety and effectiveness." Ann Pharmacother **33**(9): 979-88.

Kashfi, K. and B. Rigas (2005). "Is COX-2 a 'collateral' target in cancer prevention?" Biochem Soc Trans **33**(Pt 4): 724-7.

Kassahun, K., I. S. McIntosh, M. Shou, D. J. Walsh, C. Rodeheffer, D. E. Slaughter, L. A. Geer, R. A. Halpin, N. Agrawal and A. D. Rodrigues (2001). "Role of human liver cytochrome P4503A in the metabolism of etoricoxib, a novel cyclooxygenase-2 selective inhibitor." Drug Metab Dispos **29**(6): 813-20.

Katori, M. and M. Majima (2000). "Cyclooxygenase-2: its rich diversity of roles and possible application of its selective inhibitors." Inflamm Res **49**(8): 367-92.

Kearney, P. M., C. Baigent, J. Godwin, H. Halls, J. R. Emberson and C. Patrono (2006). "Do selective cyclo-oxygenase-2 inhibitors and traditional non-steroidal anti-inflammatory drugs increase the risk of atherothrombosis? Meta-analysis of randomised trials." Bmj **332**(7553): 1302-8.

Keenan, G. F., E. H. Giannini and B. H. Athreya (1995). "Clinically significant gastropathy associated with nonsteroidal antiinflammatory drug use in children with juvenile rheumatoid arthritis." J Rheumatol **22**(6): 1149-51.

Khan, K. N., C. M. Venturini, R. T. Bunch, J. A. Brassard, A. T. Koki, D. L. Morris, B. F. Trump, T. J. Maziasz and C. L. Alden (1998). "Interspecies differences in renal localization of cyclooxygenase isoforms: implications in nonsteroidal antiinflammatory drug-related nephrotoxicity." Toxicol Pathol **26**(5): 612-20.

Khaselev, N. and R. C. Murphy (1999). "Susceptibility of plasmenyl glycerophosphoethanolamine lipids containing arachidonate to oxidative degradation." Free Radic Biol Med **26**(3-4): 275-84.

Kim, P. S. (2006) An Open Letter from Merck Whitehouse Station, NJ, USA, Available from: (http://www.merck.com/newsroom/vioxx/pdf/Open_Letter_Concerning_VIOXX_June_26_2006.pdf) [Accessed on: 25th August 2006]

Kim, P. S. and A. S. Reicin (2005). "Discontinuation of Vioxx." Lancet **365**(9453): 23; author reply 26-7.

Kimmel, S. E., J. A. Berlin, M. Reilly, J. Jaskowiak, L. Kishel, J. Chittams and B. L. Strom (2004). "The effects of nonselective non-aspirin non-steroidal anti-inflammatory medications on the risk of nonfatal myocardial infarction and their interaction with aspirin." J Am Coll Cardiol **43**(6): 985-90.

Kis, B., J. A. Snipes and D. W. Busija (2005). "Acetaminophen and the Cyclooxygenase-3 Puzzle: Sorting out Facts, Fictions, and Uncertainties." J Pharmacol Exp Ther **315**(1): 1-7.

Klahr, S. and J. J. Morrissey (1998). "The role of growth factors, cytokines, and vasoactive compounds in obstructive nephropathy." Semin Nephrol **18**(6): 622-32.

Klein, S., J. Simes and G. L. Blackburn (1986). "Total parenteral nutrition and cancer clinical trials." Cancer **58**(6): 1378-86.

Klein, T., R. M. Nusing, J. Pfeilschifter and V. Ullrich (1994). "Selective inhibition of cyclooxygenase 2." Biochem Pharmacol **48**(8): 1605-10.

Komhoff, M., H. J. Grone, T. Klein, H. W. Seyberth and R. M. Nusing (1997). "Localization of cyclooxygenase-1 and -2 in adult and fetal human kidney: implication for renal function." Am J Physiol **272**(4 Pt 2): F460-8.

Konstam, M. A. (2003). "Matters of the heart: assessing the cardiovascular safety of new drugs." Am Heart J **146**(4): 561-2.

Konstam, M. A., M. R. Weir, A. Reicin, D. Shapiro, R. S. Sperling, E. Barr and B. J. Gertz (2001). "Cardiovascular thrombotic events in controlled, clinical trials of rofecoxib." Circulation **104**(19): 2280-8.

Konstantinopoulos, P. A. and D. F. Lehmann (2005). "The cardiovascular toxicity of selective and nonselective cyclooxygenase inhibitors: comparisons, contrasts, and aspirin confounding." J Clin Pharmacol **45**(7): 742-50.

Koshkin, V. and H. B. Dunford (1999). "Coupling of the peroxidase and cyclooxygenase reactions of prostaglandin H synthase." Biochim Biophys Acta **1430**(2): 341-8.

Kramer, B. K., M. C. Kammerl and M. Komhoff (2004). "Renal cyclooxygenase-2 (COX-2). Physiological, pathophysiological, and clinical implications." Kidney Blood Press Res **27**(1): 43-62.

Kramer, H. J., B. Stinnesbeck, G. Klautke, J. Kipnowski, D. Klingmueller, K. Glaenzer and R. Duesing (1985). "Interaction of renal prostaglandins with the renin-angiotensin and renal adrenergic nervous systems in healthy subjects during dietary changes in sodium intake." Clin Sci (Lond) **68**(4): 387-93.

Krotz, F., T. M. Schiele, V. Klauss and H. Y. Sohn (2005). "Selective COX-2 inhibitors and risk of myocardial infarction." J Vasc Res **42**(4): 312-24.

Kujubu, D. A., B. S. Fletcher, B. C. Varnum, R. W. Lim and H. R. Herschman (1991). "TIS10, a phorbol ester tumor promoter-inducible mRNA from Swiss 3T3 cells, encodes a novel prostaglandin synthase/cyclooxygenase homologue." J Biol Chem **266**(20): 12866-72.

Kumeda, Y., M. Inaba, H. Goto, M. Nagata, Y. Henmi, Y. Furumitsu, E. Ishimura, K. Inui, Y. Yutani, T. Miki, T. Shoji and Y. Nishizawa (2002). "Increased thickness of the arterial intima-media detected by ultrasonography in patients with rheumatoid arthritis." Arthritis Rheum **46**(6): 1489-97.

Kune, G. A., S. Kune and L. F. Watson (1988). "Colorectal cancer risk, chronic illnesses, operations, and medications: case control results from the Melbourne Colorectal Cancer Study." Cancer Res **48**(15): 4399-404.

Kurumbail, R. G., A. M. Stevens, J. K. Gierse, J. J. McDonald, R. A. Stegeman, J. Y. Pak, D. Gildehaus, J. M. Miyashiro, T. D. Penning, K. Seibert, P. C. Isakson and W. C. Stallings (1996). "Structural basis for selective inhibition of cyclooxygenase-2 by anti-inflammatory agents." Nature **384**(6610): 644-8.

Kuwamoto, S., H. Inoue, Y. Tone, Y. Izumi and T. Tanabe (1997). "Inverse gene expression of prostacyclin and thromboxane synthases in resident and activated peritoneal macrophages." FEBS Lett **409**(2): 242-6.

L'Abbe, K. A., A. S. Detsky and K. O'Rourke (1987). "Meta-analysis in clinical research." Ann Intern Med **107**(2): 224-33.

Lagakos, S. W. (2006). "Time-to-event analyses for long-term treatments--the APPROVe trial." N Engl J Med **355**(2): 113-7.

Landino, L. M., B. C. Crews, J. K. Gierse, S. D. Hauser and L. J. Marnett (1997). "Mutational analysis of the role of the distal histidine and glutamine residues of prostaglandin-endoperoxide synthase-2 in peroxidase catalysis, hydroperoxide

reduction, and cyclooxygenase activation." J Biol Chem **272**(34): 21565-74.

Langman, M. J., D. M. Jensen, D. J. Watson, S. E. Harper, P. L. Zhao, H. Quan, J. A. Bolognese and T. J. Simon (1999). "Adverse upper gastrointestinal effects of rofecoxib compared with NSAIDs." Jama **282**(20): 1929-33.

Lanzo, C. A., J. M. Beechem, J. Talley and L. J. Marnett (1998). "Investigation of the binding of isoform-selective inhibitors to prostaglandin endoperoxide synthases using fluorescence spectroscopy." Biochemistry **37**(1): 217-26.

Lasagna, L. (1962). "The psychophysics of clinical pain." Lancet **2**: 572-5.

Lau, J., E. M. Antman, J. Jimenez-Silva, B. Kupelnick, F. Mosteller and T. C. Chalmers (1992). "Cumulative meta-analysis of therapeutic trials for myocardial infarction." N Engl J Med **327**(4): 248-54.

Lau, J., C. H. Schmid and T. C. Chalmers (1995). "Cumulative meta-analysis of clinical trials builds evidence for exemplary medical care." J Clin Epidemiol **48**(1): 45-57; discussion 59-60.

Lawrence, R. C., C. G. Helmick, F. C. Arnett, R. A. Deyo, D. T. Felson, E. H. Giannini, S. P. Heyse, R. Hirsch, M. C. Hochberg, G. G. Hunder, M. H. Liang, S. R. Pillemer, V. D. Steen and F. Wolfe (1998). "Estimates of the prevalence of arthritis and selected musculoskeletal disorders in the United States." Arthritis Rheum **41**(5): 778-99.

Lawrence, R. C., M. C. Hochberg, J. L. Kelsey, F. C. McDuffie, T. A. Medsger, Jr., W. R. Felts and L. E. Shulman (1989). "Estimates of the prevalence of selected arthritic and musculoskeletal diseases in the United States." J Rheumatol **16**(4): 427-41.

Layton, D., K. Hughes, S. Harris and S. A. Shakir (2003). "Comparison of the incidence rates of thromboembolic events reported for patients prescribed celecoxib and meloxicam in general practice in England using Prescription-Event Monitoring (PEM) data." Rheumatology (Oxford) **42**(11): 1354-64.

Lee, C., E. Hunsche, R. Balshaw, S. X. Kong and T. J. Schnitzer (2005). "Need for common internal controls when assessing the relative efficacy of pharmacologic agents using a meta-analytic approach: case study of cyclooxygenase 2-selective inhibitors for the treatment of osteoarthritis." Arthritis Rheum **53**(4): 510-8.

Lee, C., W. L. Straus, R. Balshaw, S. Barlas, S. Vogel and T. J. Schnitzer (2004). "A

comparison of the efficacy and safety of nonsteroidal antiinflammatory agents versus acetaminophen in the treatment of osteoarthritis: a meta-analysis." Arthritis Rheum **51**(5): 746-54.

Lee, D. M. and M. E. Weinblatt (2001). "Rheumatoid arthritis." Lancet **358**(9285): 903-11.

LeLorier, J., G. Gregoire, A. Benhaddad, J. Lapierre and F. Derderian (1997). "Discrepancies between meta-analyses and subsequent large randomized, controlled trials." N Engl J Med **337**(8): 536-42.

Lemley, K. V. and W. Kriz (1991). "Anatomy of the renal interstitium." Kidney Int **39**(3): 370-81.

Len, C., M. O. Hilario, E. Kawakami, M. T. Terreri, D. J. Becker, J. Goldenberg and U. Fagundes Neto (1999). "Gastroduodenal lesions in children with juvenile rheumatoid arthritis." Hepatogastroenterology **46**(26): 991-6.

Leonetti, G., C. Cuspidi, M. Facchini and M. Stramba-Badiale (2000). "Is systolic pressure a better target for antihypertensive treatment than diastolic pressure?" J Hypertens Suppl **18**(3): S13-20.

Lequesne, M. G., C. Mery, M. Samson and P. Gerard (1987). "Indexes of severity for osteoarthritis of the hip and knee. Validation--value in comparison with other assessment tests." Scand J Rheumatol Suppl **65**: 85-9.

Li Wan Po, A. (1996). "Evidence-Based Pharmacotherapy." The Pharmaceutical Journal **256**: 308-312.

Lievre, M. and E. Abadie (2005). "Discontinuation of Vioxx." Lancet **365**(9453): 23-4; author reply 26-7.

Liew, D. and H. Krum (2002). "The role of aldosterone receptor blockade in the management of cardiovascular disease." Curr Opin Investig Drugs **3**(10): 1468-73.

Light, R. J. and D. B. Pillemer (1984). The science of reviewing research. Cambridge, MA: Harvard University Press.

Lim, H., R. A. Gupta, W. G. Ma, B. C. Paria, D. E. Moller, J. D. Morrow, R. N. DuBois, J. M. Trzaskos and S. K. Dey (1999). "Cyclo-oxygenase-2-derived prostacyclin mediates embryo implantation in the mouse via PPARdelta." Genes Dev **13**(12): 1561-74.

Liou, J. Y., S. K. Shyue, M. J. Tsai, C. L. Chung, K. Y. Chu and K. K. Wu (2000).

"Colocalization of prostacyclin synthase with prostaglandin H synthase-1 (PGHS-1) but not phorbol ester-induced PGHS-2 in cultured endothelial cells." J Biol Chem **275**(20): 15314-20.

Lipsky, P. E. (1999). "Role of cyclooxygenase-1 and -2 in health and disease." Am J Orthop **28**(3 Suppl): 8-12.

Lipsky, P. E., S. B. Abramson, L. J. Crofford, R. N. DuBois, L. S. Simon and L. B. van de Putte (1998). "The classification of cyclooxygenase inhibitors." J Rheumatol **25**: 2298-303.

Lipsky, P. E., D. M. van der Heijde, E. W. St Clair, D. E. Furst, F. C. Breedveld, J. R. Kalden, J. S. Smolen, M. Weisman, P. Emery, M. Feldmann, G. R. Harriman and R. N. Maini (2000). "Infliximab and methotrexate in the treatment of rheumatoid arthritis. Anti-Tumor Necrosis Factor Trial in Rheumatoid Arthritis with Concomitant Therapy Study Group." N Engl J Med **343**(22): 1594-602.

Liuzzo, G., L. M. Biasucci, J. R. Gallimore, R. L. Grillo, A. G. Rebuzzi, M. B. Pepys and A. Maseri (1994). "The prognostic value of C-reactive protein and serum amyloid a protein in severe unstable angina." N Engl J Med **331**(7): 417-24.

Liuzzo, G., J. J. Goronzy, H. Yang, S. L. Kopecky, D. R. Holmes, R. L. Frye and C. M. Weyand (2000). "Monoclonal T-cell proliferation and plaque instability in acute coronary syndromes." Circulation **101**(25): 2883-8.

Loll, P. J., D. Picot, O. Ekabo and R. M. Garavito (1996). "Synthesis and use of iodinated nonsteroidal antiinflammatory drug analogs as crystallographic probes of the prostaglandin H2 synthase cyclooxygenase active site." Biochemistry **35**(23): 7330-40.

Lord, J., C. Victor, P. Littlejohns, F. M. Ross and J. S. Axford (1999). "Economic evaluation of a primary-care based education programme for patients with osteoarthritis of the knee." Health Technol Assess **3**(23): iii-55.

Lowe, H. J. and G. O. Barnett (1994). "Understanding and using the medical subject headings (MeSH) vocabulary to perform literature searches." Jama **271**(14): 1103-8.

Lu, G., A. L. Tsai, H. E. Van Wart and R. J. Kulmacz (1999). "Comparison of the peroxidase reaction kinetics of prostaglandin H synthase-1 and -2." J Biol Chem

274(23): 16162-7.

Luong, C., A. Miller, J. Barnett, J. Chow, C. Ramesha and M. F. Browner (1996). "Flexibility of the NSAID binding site in the structure of human cyclooxygenase-2." Nat Struct Biol **3**(11): 927-33.

Lyseng-Williamson, K. A. and M. P. Curran (2004). "Lumiracoxib." Drugs **64**(19): 2237-46; discussion 2247-8.

Malmstrom, K., J. R. Fricke, P. Kotey, B. Kress and B. Morrison (2002). "A comparison of rofecoxib versus celecoxib in treating pain after dental surgery: a single-center, randomized, double-blind, placebo- and active-comparator-controlled, parallel-group, single-dose study using the dental impaction pain model." Clin Ther **24**(10): 1549-60.

Mamdani, M., P. Rochon, D. N. Juurlink, G. M. Anderson, A. Kopp, G. Naglie, P. C. Austin and A. Laupacis (2003). "Effect of selective cyclooxygenase 2 inhibitors and naproxen on short-term risk of acute myocardial infarction in the elderly." Arch Intern Med **163**(4): 481-6.

Mancini, J. A., D. Riendeau, J. P. Falgueyret, P. J. Vickers and G. P. O'Neill (1995). "Arginine 120 of prostaglandin G/H synthase-1 is required for the inhibition by nonsteroidal anti-inflammatory drugs containing a carboxylic acid moiety." J Biol Chem **270**(49): 29372-7.

Mangold, J. B., H. Gu, L. C. Rodriguez, J. Bonner, J. Dickson and C. Rordorf (2004). "Pharmacokinetics and metabolism of lumiracoxib in healthy male subjects." Drug Metab Dispos **32**(5): 566-71.

Maradit-Kremers, H., P. J. Nicola, C. S. Crowson, K. V. Ballman and S. E. Gabriel (2005). "Cardiovascular death in rheumatoid arthritis: a population-based study." Arthritis Rheum **52**(3): 722-32.

Marnett, L. J., S. W. Rowlinson, D. C. Goodwin, A. S. Kalgutkar and C. A. Lanzo (1999). "Arachidonic acid oxygenation by COX-1 and COX-2. Mechanisms of catalysis and inhibition." J Biol Chem **274**(33): 22903-6.

Martin, B. K., C. L. Meinert and J. C. Breitner (2002). "Double placebo design in a prevention trial for Alzheimer's disease." Control Clin Trials **23**(1): 93-9.

Martina, S. D., K. S. Vesta and T. L. Ripley (2005). "Etoricoxib: a highly selective COX-2

inhibitor." Ann Pharmacother **39**(5): 854-62.

Masferrer, J. L., B. S. Zweifel, K. Seibert and P. Needleman (1990). "Selective regulation of cellular cyclooxygenase by dexamethasone and endotoxin in mice." J Clin Invest **86**(4): 1375-9.

Mason, R. P., M. F. Walter and P. E. Mason (1997). "Effect of oxidative stress on membrane structure: small-angle X-ray diffraction analysis." Free Radic Biol Med **23**(3): 419-25.

McAdam, B. F., F. Catella-Lawson, I. A. Mardini, S. Kapoor, J. A. Lawson and G. A. FitzGerald (1999). "Systemic biosynthesis of prostacyclin by cyclooxygenase (COX)-2: the human pharmacology of a selective inhibitor of COX-2." Proc Natl Acad Sci U S A **96**(1): 272-7.

McAlister, F. A., S. E. Straus, G. H. Guyatt and R. B. Haynes (2000). "Users' guides to the medical literature: XX. Integrating research evidence with the care of the individual patient. Evidence-Based Medicine Working Group." Jama **283**(21): 2829-36.

McGeer, P. L., M. Schulzer and E. G. McGeer (1996). "Arthritis and anti-inflammatory agents as possible protective factors for Alzheimer's disease: a review of 17 epidemiologic studies." Neurology **47**(2): 425-32.

McKee, M., A. Britton, N. Black, K. McPherson, C. Sanderson and C. Bain (1999). "Methods in health services research. Interpreting the evidence: choosing between randomised and non-randomised studies." Bmj **319**(7205): 312-5.

McQueen, M. J. (2001). "Overview of evidence-based medicine: challenges for evidence-based laboratory medicine." Clin Chem **47**(8): 1536-46.

Medicines and Healthcare products Regulatory Agency, (MHRA) (2005) Updated advice on the safety of selective COX-2 inhibitors. Letter to healthcare professionals (incorporating a questions and answers document). London, Available from: (http://medicines.mhra.gov.uk) [Accessed on: 25th August 2006]

Medicines and Healthcare products Regulatory Agency, (MHRA) (2006) Data Analysis Print: Rofecoxib London, Available from: (www.mhra.gov.uk/home/groups/public/documents/sentineldocuments/dap_1130236049248.pdf) [Accessed on: 25th August 2006]

Meenan, R. F., J. H. Mason, J. J. Anderson, A. A. Guccione and L. E. Kazis (1992).

"AIMS2. The content and properties of a revised and expanded Arthritis Impact Measurement Scales Health Status Questionnaire." Arthritis Rheum **35**(1): 1-10.

Mehlisch, D., P. Desjardins, D. Krupa, R. Polis, R. Petruschke, G. Geba and D. Chang (2004). "Efficacy of rofecoxib 50mg in patients with moderate or severe pain: A meta-analysis of 13 clinical trials." The Journal of Pain **5**(3): S63.

Meinert, C. L. (1989). "Meta-analysis: science or religion?" Control Clin Trials **10**(4 Suppl): 257S-263S.

Merck (2005) Osteoarthritis: Definition and Classification Whitehouse Station, NJ, USA, March 2001 Available from: (http://www.merckmedicus.com/pp/us/hcp/diseasemodules/osteoarthritis/default.jsp) [Accessed on: 04th November 2005]

Merck & Co., Inc. (2002) Annual report 2002 Available from: (http://www.merck.com/finance/annualreport/ar2002/merck_financial_section.pdf) [Accessed on: 8th July 2006]

Merck & Co., Inc. (2004) Response to Article by Juni et al. Published in The Lancet on Nov. 5 Whitehouse Station, NJ, USA, Available from: (http://www.merck.com/statement_2004_1105/lancet.pdf) [Accessed on: 1st February 2005]

Merck & Co., Inc. (2004) Overview Chart of Major Vioxx Research Studies N.J. USA, Available from: (http://www.merck.com/newsroom/vioxx/pdf/vioxx_timeline_1013.pdf) [Accessed on: 25th August 2006]

Merck & Co., Inc. (2006) OA: Epidemiology Whitehouse Station, NJ, USA, March 2001 Available from: (http://www.merckmedicus.com/pp/us/hcp/diseasemodules/osteoarthritis/epidemiology.jsp) [Accessed on: 25th August 2006]

Mikuls, T. R., J. R. Cerhan, L. A. Criswell, L. Merlino, A. S. Mudano, M. Burma, A. R. Folsom and K. G. Saag (2002). "Coffee, tea, and caffeine consumption and risk of rheumatoid arthritis: results from the Iowa Women's Health Study." Arthritis Rheum **46**(1): 83-91.

Mitchell, J. A., P. Akarasereenont, C. Thiemermann, R. J. Flower and J. R. Vane (1993).

"Selectivity of nonsteroidal antiinflammatory drugs as inhibitors of constitutive and inducible cyclooxygenase." Proc Natl Acad Sci U S A **90**(24): 11693-7.

Mizuno, K., S. Yamamoto and W. E. Lands (1982). "Effects of non-steroidal anti-inflammatory drugs on fatty acid cyclooxygenase and prostaglandin hydroperoxidase activities." Prostaglandins **23**(5): 743-57.

Moher, D., D. J. Cook, S. Eastwood, I. Olkin, D. Rennie and D. F. Stroup (1999). "Improving the quality of reports of meta-analyses of randomised controlled trials: the QUOROM statement. Quality of Reporting of Meta-analyses." Lancet **354**(9193): 1896-900.

Moher, D., D. J. Cook, S. Eastwood, I. Olkin, D. Rennie and D. F. Stroup (2000a). "Improving the Quality of Reports of Meta-Analyses of Randomised Controlled Trials: The QUOROM Statement." Onkologie **23**(6): 597-602.

Moher, D., D. J. Cook, S. Eastwood, I. Olkin, D. Rennie and D. F. Stroup (2000b). "Improving the quality of reports of meta-analyses of randomised controlled trials: the QUOROM statement. QUOROM Group." Br J Surg **87**(11): 1448-54.

Moher, D., P. Fortin, A. R. Jadad, P. Juni, T. Klassen, J. Le Lorier, A. Liberati, K. Linde and A. Penna (1996). "Completeness of reporting of trials published in languages other than English: implications for conduct and reporting of systematic reviews." Lancet **347**(8998): 363-6.

Moher, D., A. R. Jadad, G. Nichol, M. Penman, P. Tugwell and S. Walsh (1995). "Assessing the quality of randomized controlled trials: an annotated bibliography of scales and checklists." Control Clin Trials **16**(1): 62-73.

Moher, D., B. Pham, T. P. Klassen, K. F. Schulz, J. A. Berlin, A. R. Jadad and A. Liberati (2000). "What contributions do languages other than English make on the results of meta-analyses?" J Clin Epidemiol **53**(9): 964-72.

Moher, D., K. F. Schulz and D. G. Altman (2001a). "The CONSORT statement: revised recommendations for improving the quality of reports of parallel-group randomized trials." Ann Intern Med **134**(8): 657-62.

Moher, D., K. F. Schulz and D. G. Altman (2001b). "The CONSORT statement: revised recommendations for improving the quality of reports of parallel-group randomised trials." Lancet **357**(9263): 1191-4.

Moher, D., K. F. Schulz and D. G. Altman (2003). "The CONSORT statement: revised recommendations for improving the quality of reports of parallel-group randomised trials." Clin Oral Investig **7**(1): 2-7.

Monakier, D., M. Mates, M. W. Klutstein, J. A. Balkin, B. Rudensky, D. Meerkin and D. Tzivoni (2004). "Rofecoxib, a COX-2 inhibitor, lowers C-reactive protein and interleukin-6 levels in patients with acute coronary syndromes." Chest **125**(5): 1610-5.

Monson, R. R. and A. P. Hall (1976). "Mortality among arthritics." J Chronic Dis **29**(7): 459-67.

Moore, R. A., S. Derry, G. T. Makinson and H. J. McQuay (2005). "Tolerability and adverse events in clinical trials of celecoxib in osteoarthritis and rheumatoid arthritis: systematic review and meta-analysis of information from company clinical trial reports." Arthritis Res Ther **7**(3): R644-65.

Moore, R. A., J. E. Edwards and H. J. McQuay (2005). "Acute pain: individual patient meta-analysis shows the impact of different ways of analysing and presenting results." Pain **116**(3): 322-31.

Morgani, J. B. (1769). The seats and causes of diseases investigated by anatomy. London.

Morita, I., M. Schindler, M. K. Regier, J. C. Otto, T. Hori, D. L. DeWitt and W. L. Smith (1995). "Different intracellular locations for prostaglandin endoperoxide H synthase-1 and -2." J Biol Chem **270**(18): 10902-8.

Morrison, B. W., J. Fricke, J. Brown, W. Yuan, P. Kotey and D. Mehlisch (2000). "The optimal analgesic dose of rofecoxib: Overview of six randomized controlled trials." J Am Dent Assoc **131**(12): 1729-37.

Morrow, J. D., W. E. Zackert, J. P. Yang, E. H. Kurhts, D. Callewaert, R. Dworski, K. Kanai, D. Taber, K. Moore, J. A. Oates and L. J. Roberts (1999). "Quantification of the major urinary metabolite of 15-F2t-isoprostane (8-iso-PGF2alpha) by a stable isotope dilution mass spectrometric assay." Anal Biochem **269**(2): 326-31.

Mueller, M. J., M. Andberg and J. Z. Haeggstrom (1998). "Analysis of the molecular mechanism of substrate-mediated inactivation of leukotriene A4 hydrolase." J Biol Chem **273**(19): 11570-5.

Mukherjee, D., S. E. Nissen and E. J. Topol (2001). "Risk of cardiovascular events

associated with selective COX-2 inhibitors." Jama **286**(8): 954-9.

Mulberg, A. E., C. Linz, E. Bern, L. Tucker, M. Verhave and R. J. Grand (1993). "Identification of nonsteroidal antiinflammatory drug-induced gastroduodenal injury in children with juvenile rheumatoid arthritis." J Pediatr **122**(4): 647-9.

Mullen, P. D. and G. Ramirez (1987). "Information synthesis and meta-analysis." Advan Health Educat Promot **2**: 201-39.

Mulrow, C. D. (1987). "The medical review article: state of the science." Ann Intern Med **106**(3): 485-8.

Mulrow, C. D., S. B. Thacker and J. A. Pugh (1988). "A proposal for more informative abstracts of review articles." Ann Intern Med **108**(4): 613-5.

Murata, T., F. Ushikubi, T. Matsuoka, M. Hirata, A. Yamasaki, Y. Sugimoto, A. Ichikawa, Y. Aze, T. Tanaka, N. Yoshida, A. Ueno, S. Oh-ishi and S. Narumiya (1997). "Altered pain perception and inflammatory response in mice lacking prostacyclin receptor." Nature **388**(6643): 678-82.

Murray, M. D., P. K. Greene, D. C. Brater, A. K. Manatunga and S. D. Hall (1992). "Effects of flurbiprofen on renal function in patients with moderate renal insufficiency." Br J Clin Pharmacol **33**(4): 385-93.

Muscara, M. N., N. Vergnolle, F. Lovren, C. R. Triggle, S. N. Elliott, S. Asfaha and J. L. Wallace (2000). "Selective cyclo-oxygenase-2 inhibition with celecoxib elevates blood pressure and promotes leukocyte adherence." Br J Pharmacol **129**(7): 1423-30.

Nagata-Sakurai, M., M. Inaba, H. Goto, Y. Kumeda, Y. Furumitsu, K. Inui, H. Koyama, M. Emoto, E. Ishimura, T. Shoji and Y. Nishizawa (2003). "Inflammation and bone resorption as independent factors of accelerated arterial wall thickening in patients with rheumatoid arthritis." Arthritis Rheum **48**(11): 3061-7.

Nantel, F., E. Meadows, D. Denis, B. Connolly, K. M. Metters and A. Giaid (1999). "Immunolocalization of cyclooxygenase-2 in the macula densa of human elderly." FEBS Lett **457**(3): 475-7.

Narine, L., D. S. Yee, T. R. Einarson and A. L. Ilersich (1991). "Quality of abstracts of original research articles in CMAJ in 1989." Cmaj **144**(4): 449-53.

Nath, K. A., D. H. Chmielewski and T. H. Hostetter (1987). "Regulatory role of

prostanoids in glomerular microcirculation of remnant nephrons." Am J Physiol **252**(5 Pt 2): F829-37.

Navar, L. G., E. W. Inscho, S. A. Majid, J. D. Imig, L. M. Harrison-Bernard and K. D. Mitchell (1996). "Paracrine regulation of the renal microcirculation." Physiol Rev **76**(2): 425-536.

Naylor, C. D. (1997). "Meta-analysis and the meta-epidemiology of clinical research." Bmj **315**(7109): 617-9.

Neely, J. G. (1993). "Literature review articles as a research form." Otolaryngol Head Neck Surg **108**(6): 743-8.

NHS National Prescribing Centre, NPC. (2005/2006) What's been happening with COX-2 prescribing? Liverpool, UK, Available from: (http://www.npc.co.uk/MeReC_Extra/2005/no17_2005.pdf) [Accessed on: 25th August 2006]

NICE, National Institute for Clinical Excellence. (2001) Guidance on the use of cyclo-ocygenase (COX) II selective inhibitors, celecoxib, rofecoxib, meloxicam, and etodolac for osteoarthritis and rheumatoid arthritis. National Institute for Clinical Excellence London, Available from: (www.nice.org.uk) [Accessed on: 29th July 2006]

NICE, National Institute for Clinical Excellence. (2001a) The Clinical Effectiveness and Cost Effectiveness of Celecoxib, Rofecoxib, Meloxicam and Etodolac (COX-II Inhibitors) for Rheumatoid Arthritis and Osteoarthritis. London, Available from: (http://www.nice.org.uk/pdf/coxiihtareport.pdf) [Accessed on: 05th November 2005]

Nicoll-Griffith, D. A., J. A. Yergey, L. A. Trimble, J. M. Silva, C. Li, N. Chauret, J. Y. Gauthier, E. Grimm, S. Leger, P. Roy, M. Therien, Z. Wang, P. Prasit, R. Zamboni, R. N. Young, C. Brideau, C. C. Chan, J. Mancini and D. Riendeau (2000). "Synthesis, characterization, and activity of metabolites derived from the cyclooxygenase-2 inhibitor rofecoxib (MK-0966, Vioxx)." Bioorg Med Chem Lett **10**(23): 2683-6.

(NIH), US Department of Health and Human Services. (2005) Use of non-steroidal anti-inflammatory drugs suspended in large Alzheimer's disease prevention trial

Bethesda, Maryland, USA, Available from: (http://www.nih.gov/news/pr/dec2004/od-20.htm) [Accessed on: 11th July 2006]

Nishi, K., H. Itabe, M. Uno, K. T. Kitazato, H. Horiguchi, K. Shinno and S. Nagahiro (2002). "Oxidized LDL in carotid plaques and plasma associates with plaque instability." Arterioscler Thromb Vasc Biol **22**(10): 1649-54.

Nissen, S. E. (2006). "Adverse cardiovascular effects of rofecoxib." N Engl J Med **355**(2): 203-4; author reply 203-5.

Nony, P., M. Cucherat, M. C. Haugh and J. P. Boissel (1995). "Critical reading of the meta-analysis of clinical trials." Therapie **50**: 339-51.

Nussmeier, N. A., A. A. Whelton, M. T. Brown, R. M. Langford, A. Hoeft, J. L. Parlow, S. W. Boyce and K. M. Verburg (2005). "Complications of the COX-2 inhibitors parecoxib and valdecoxib after cardiac surgery." N Engl J Med **352**(11): 1081-91.

Oates, J. A., G. A. FitzGerald, R. A. Branch, E. K. Jackson, H. R. Knapp and L. J. Roberts, 2nd (1988). "Clinical implications of prostaglandin and thromboxane A2 formation (1)." N Engl J Med **319**(11): 689-98.

Ofman, J. J., C. H. MacLean, W. L. Straus, S. C. Morton, M. L. Berger, E. A. Roth and P. Shekelle (2002). "A metaanalysis of severe upper gastrointestinal complications of nonsteroidal antiinflammatory drugs." J Rheumatol **29**(4): 804-12.

Ohlsson, A. (1994). "Systematic reviews--theory and practice." Scand J Clin Lab Invest Suppl **219**: 25-32.

Oliveria, S. A., D. T. Felson, J. I. Reed, P. A. Cirillo and A. M. Walker (1995). "Incidence of symptomatic hand, hip, and knee osteoarthritis among patients in a health maintenance organization." Arthritis Rheum **38**(8): 1134-41.

Olsen, N. J. and C. M. Stein (2004). "New drugs for rheumatoid arthritis." N Engl J Med **350**(21): 2167-79.

O'Rourke, K. and A. S. Detsky (1989). "Meta-analysis in medical research: strong encouragement for higher quality in individual research efforts." J Clin Epidemiol **42**(10): 1021-4.

Ott, E., N. A. Nussmeier, P. C. Duke, R. O. Feneck, R. P. Alston, M. C. Snabes, R. C. Hubbard, P. H. Hsu, L. J. Saidman and D. T. Mangano (2003). "Efficacy and safety of the cyclooxygenase 2 inhibitors parecoxib and valdecoxib in patients undergoing

coronary artery bypass surgery." J Thorac Cardiovasc Surg **125**(6): 1481-92.

Otto, J. C., D. L. DeWitt and W. L. Smith (1993). "N-glycosylation of prostaglandin endoperoxide synthases-1 and -2 and their orientations in the endoplasmic reticulum." J Biol Chem **268**(24): 18234-42.

Otto, J. C. and W. L. Smith (1994). "The orientation of prostaglandin endoperoxide synthases-1 and -2 in the endoplasmic reticulum." J Biol Chem **269**(31): 19868-75.

Otto, J. C. and W. L. Smith (1996). "Photolabeling of prostaglandin endoperoxide H synthase-1 with 3-trifluoro-3-(m-[125I]iodophenyl)diazirine as a probe of membrane association and the cyclooxygenase active site." J Biol Chem **271**(17): 9906-10.

Ovid Technologies, Inc. (2005) Ovid Technologies Field Guide: Cochrane Central Register of Controlled Trials (CCTR), The Evidence Based Reviews Collection. New York, USA, 26th May, 2005 Available from: (http://www.ovid.com/site/products/ovidguide/cctrdb.htm) [Accessed on: 9th April 2006]

Oxman, A. D. (1994). "Checklists for review articles." Bmj **309**(6955): 648-51.

Oxman, A. D. and G. H. Guyatt (1988). "Guidelines for reading literature reviews." Cmaj **138**(8): 697-703.

Pace-Asciak, C. R., D. Reynaud, P. Demin and S. Nigam (1999). "The hepoxilins. A review." Adv Exp Med Biol **447**: 123-32.

Panara, M. R., G. Renda, M. G. Sciulli, G. Santini, M. Di Giamberardino, M. T. Rotondo, S. Tacconelli, F. Seta, C. Patrono and P. Patrignani (1999). "Dose-dependent inhibition of platelet cyclooxygenase-1 and monocyte cyclooxygenase-2 by meloxicam in healthy subjects." J Pharmacol Exp Ther **290**(1): 276-80.

Pasinetti, G. M. and P. S. Aisen (1998). "Cyclooxygenase-2 expression is increased in frontal cortex of Alzheimer's disease brain." Neuroscience **87**(2): 319-24.

Patrignani, P., M. R. Panara, A. Greco, O. Fusco, C. Natoli, S. Iacobelli, F. Cipollone, A. Ganci, C. Creminon, J. Maclouf and et al. (1994). "Biochemical and pharmacological characterization of the cyclooxygenase activity of human blood prostaglandin endoperoxide synthases." J Pharmacol Exp Ther **271**(3): 1705-12.

Patrignani, P., M. R. Panara, M. G. Sciulli, G. Santini, G. Renda and C. Patrono (1997).

"Differential inhibition of human prostaglandin endoperoxide synthase-1 and -2 by nonsteroidal anti-inflammatory drugs." J Physiol Pharmacol **48**(4): 623-31.

Patrignani, P., S. Tacconelli, M. G. Sciulli and M. L. Capone (2005). "New insights into COX-2 biology and inhibition." Brain Res Brain Res Rev **48**(2): 352-9.

Patrono, C., G. Ciabattoni, P. Patrignani, F. Pugliese, P. Filabozzi, F. Catella, G. Davi and L. Forni (1985). "Clinical pharmacology of platelet cyclooxygenase inhibition." Circulation **72**(6): 1177-84.

Patrono, C., B. Coller, J. E. Dalen, G. A. FitzGerald, V. Fuster, M. Gent, J. Hirsh and G. Roth (2001). "Platelet-active drugs : the relationships among dose, effectiveness, and side effects." Chest **119**(1 Suppl): 39S-63S.

Patrono, C., P. Patrignani and L. A. Garcia Rodriguez (2001). "Cyclooxygenase-selective inhibition of prostanoid formation: transducing biochemical selectivity into clinical read-outs." J Clin Invest **108**(1): 7-13.

Paulson, S. K., T. A. Kaprak, C. J. Gresk, D. M. Fast, M. T. Baratta, E. G. Burton, A. P. Breau and A. Karim (1999). "Plasma protein binding of celecoxib in mice, rat, rabbit, dog and human." Biopharm Drug Dispos **20**(6): 293-9.

Pelayo, J. C. and P. F. Shanley (1990). "Glomerular and tubular adaptive responses to acute nephron loss in the rat. Effect of prostaglandin synthesis inhibition." J Clin Invest **85**(6): 1761-9.

Perazella, M. A. and J. Eras (2000). "Are selective COX-2 inhibitors nephrotoxic?" Am J Kidney Dis **35**(5): 937-40.

Persson, A. E., M. Salomonsson, P. Westerlund, R. Greger, E. Schlatter and E. Gonzalez (1991). "Macula densa cell function." Kidney Int Suppl **32**: S39-44.

Petersson, I. F. (1996). "Occurrence of osteoarthritis of the peripheral joints in European populations." Ann Rheum Dis **55**(9): 659-61.

Petitti, D. (2004). "Commentary: hormone replacement therapy and coronary heart disease: four lessons." Int J Epidemiol **33**(3): 461-3.

Petitti, D. B. (1994). Meta-Analysis, Decision Analysis, and Cost-Effectiveness Analysis: Methods for Quantitative Synthesis in Medicine. New York, Oxford University Press Inc.

Pfizer Inc. (2004) Pfizer Statement on New Information Regarding Cardiovascular Safety

of Celebrex New York, Available from: (http://www.pfizer.com/pfizer/are/investors_releases/2004pr/mn_2004_1217.jsp) [Accessed on: 11th July 2006]

Pham, B., T. P. Klassen, M. L. Lawson and D. Moher (2005). "Language of publication restrictions in systematic reviews gave different results depending on whether the intervention was conventional or complementary." J Clin Epidemiol **58**(8): 769-76.

Phillips, R. K., M. H. Wallace, P. M. Lynch, E. Hawk, G. B. Gordon, B. P. Saunders, N. Wakabayashi, Y. Shen, S. Zimmerman, L. Godio, M. Rodrigues-Bigas, L. K. Su, J. Sherman, G. Kelloff, B. Levin and G. Steinbach (2002). "A randomised, double blind, placebo controlled study of celecoxib, a selective cyclooxygenase 2 inhibitor, on duodenal polyposis in familial adenomatous polyposis." Gut **50**(6): 857-60.

Picot, D., P. J. Loll and R. M. Garavito (1994). "The X-ray crystal structure of the membrane protein prostaglandin H2 synthase-1." Nature **367**(6460): 243-9.

Pidgeon, G. P., R. Tamosiuniene, G. Chen, I. Leonard, O. Belton, A. Bradford and D. J. Fitzgerald (2004). "Intravascular thrombosis after hypoxia-induced pulmonary hypertension: regulation by cyclooxygenase-2." Circulation **110**(17): 2701-7.

Pincus, T. and L. F. Callahan (1989). "Reassessment of twelve traditional paradigms concerning the diagnosis, prevalence, morbidity and mortality of rheumatoid arthritis." Scand J Rheumatol Suppl **79**: 67-96.

Pincus, T., J. R. O'Dell and J. M. Kremer (1999). "Combination therapy with multiple disease-modifying antirheumatic drugs in rheumatoid arthritis: a preventive strategy." Ann Intern Med **131**(10): 768-74.

Pogue, J. and S. Yusuf (1998). "Overcoming the limitations of current meta-analysis of randomised controlled trials." Lancet **351**(9095): 47-52.

Pratico, D., D. Ferro, L. Iuliano, J. Rokach, F. Conti, G. Valesini, G. A. FitzGerald and F. Violi (1999). "Ongoing prothrombotic state in patients with antiphospholipid antibodies: a role for increased lipid peroxidation." Blood **93**(10): 3401-7.

Pratico, D., L. Iuliano, A. Mauriello, L. Spagnoli, J. A. Lawson, J. Rokach, J. Maclouf, F. Violi and G. A. FitzGerald (1997). "Localization of distinct F2-isoprostanes in human atherosclerotic lesions." J Clin Invest **100**(8): 2028-34.

Preston Mason, R., M. F. Walter, H. P. McNulty, S. F. Lockwood, J. Byun, C. A. Day and

R. F. Jacob (2006). "Rofecoxib Increases Susceptibility of Human LDL and Membrane Lipids to Oxidative Damage: A Mechanism of Cardiotoxicity." J Cardiovasc Pharmacol **47 Suppl 1**: S7-S14.

Psaty, B. M. and C. D. Furberg (2005). "COX-2 inhibitors--lessons in drug safety." N Engl J Med **352**(11): 1133-5.

Qi, Z., C. M. Hao, R. I. Langenbach, R. M. Breyer, R. Redha, J. D. Morrow and M. D. Breyer (2002). "Opposite effects of cyclooxygenase-1 and -2 activity on the pressor response to angiotensin II." J Clin Invest **110**(1): 61-9.

Qin, N., S. P. Zhang, T. L. Reitz, J. M. Mei and C. M. Flores (2005). "Cloning, Expression and Functional Characterization of Human COX-1 Splicing Variants: Evidence for Intron 1 Retention." J Pharmacol Exp Ther.

Rahme, E., L. Pilote and J. LeLorier (2002). "Association between naproxen use and protection against acute myocardial infarction." Arch Intern Med **162**(10): 1111-5.

Ravaud, P. and F. Tubach (2005). "Methodology of therapeutic trials: lessons from the late evidence of the cardiovascular toxicity of some coxibs." Joint Bone Spine **72**(6): 451-5.

Ray, W. A., C. M. Stein, J. R. Daugherty, K. Hall, P. G. Arbogast and M. R. Griffin (2002a). "COX-2 selective non-steroidal anti-inflammatory drugs and risk of serious coronary heart disease." Lancet **360**(9339): 1071-3.

Ray, W. A., C. M. Stein, K. Hall, J. R. Daugherty and M. R. Griffin (2002b). "Non-steroidal anti-inflammatory drugs and risk of serious coronary heart disease: an observational cohort study." Lancet **359**(9301): 118-23.

Reddy, L. R. and E. J. Corey (2005). "Facile air oxidation of the conjugate base of rofecoxib (Vioxx), a possible contributor to chronic human toxicity." Tetrahedron Lett. **46**: 927-9.

Reginster, J. Y. (2002). "The prevalence and burden of arthritis." Rheumatology (Oxford) **41 Supp 1**: 3-6.

Reicin, A. S., D. Shapiro, R. S. Sperling, E. Barr and Q. Yu (2002). "Comparison of cardiovascular thrombotic events in patients with osteoarthritis treated with rofecoxib versus nonselective nonsteroidal anti-inflammatory drugs (ibuprofen, diclofenac, and nabumetone)." Am J Cardiol **89**(2): 204-9.

Reilly, P. A., J. A. Cosh, P. J. Maddison, J. J. Rasker and A. J. Silman (1990). "Mortality and survival in rheumatoid arthritis: a 25 year prospective study of 100 patients." Ann Rheum Dis **49**(6): 363-9.

Reines, S. A., G. A. Block, J. C. Morris, G. Liu, M. L. Nessly, C. R. Lines, B. A. Norman and C. C. Baranak (2004). "Rofecoxib: no effect on Alzheimer's disease in a 1-year, randomized, blinded, controlled study." Neurology **62**(1): 66-71.

Reuben, S. S. and N. R. Connelly (2000). "Postoperative analgesic effects of celecoxib or rofecoxib after spinal fusion surgery." Anesth Analg **91**(5): 1221-5.

Reynolds, L. W., R. K. Hoo, R. J. Brill, J. North, D. P. Recker and K. M. Verburg (2003). "The COX-2 specific inhibitor, valdecoxib, is an effective, opioid-sparing analgesic in patients undergoing total knee arthroplasty." J Pain Symptom Manage **25**(2): 133-41.

Ridker, P. M., J. E. Buring, J. Shih, M. Matias and C. H. Hennekens (1998). "Prospective study of C-reactive protein and the risk of future cardiovascular events among apparently healthy women." Circulation **98**(8): 731-3.

Ridker, P. M., M. Cushman, M. J. Stampfer, R. P. Tracy and C. H. Hennekens (1997). "Inflammation, aspirin, and the risk of cardiovascular disease in apparently healthy men." N Engl J Med **336**(14): 973-9.

Rigas, B. and K. Kashfi (2005). "Cancer prevention: a new era beyond cyclooxygenase-2." J Pharmacol Exp Ther **314**(1): 1-8.

Rindfleisch, J. A. and D. Muller (2005). "Diagnosis and management of rheumatoid arthritis." Am Fam Physician **72**(6): 1037-47.

Rochon, P. A., J. H. Gurwitz, R. W. Simms and et.al. (1994). "A study of manufacturer-supported trials of nonsteroidal anti-inflammatory drugs in the treatment of arthritis." Arch Intern Med **154**: 157-63.

Rochon, P. A., J. H. Gurwitz, K. Sykora, M. Mamdani, D. L. Streiner, S. Garfinkel, S. L. Normand and G. M. Anderson (2005). "Reader's guide to critical appraisal of cohort studies: 1. Role and design." Bmj **330**(7496): 895-7.

Rodrigues, C. R., M. P. Veloso, H. Verli, C. A. Fraga, A. L. Miranda and E. J. Barreiro (2002). "Selective PGHS-2 inhibitors: a rational approach for treatment of the inflammation." Curr Med Chem **9**(8): 849-67.

Roman, R. J. and C. Lechene (1981). "Prostaglandin E2 and F2 alpha reduces urea reabsorption from the rat collecting duct." Am J Physiol **241**(1): F53-60.

Roman, R. J. and E. Lianos (1990). "Influence of prostaglandins on papillary blood flow and pressure-natriuretic response." Hypertension **15**(1): 29-35.

Romsing, J. and S. Moiniche (2004). "A systematic review of COX-2 inhibitors compared with traditional NSAIDs, or different COX-2 inhibitors for post-operative pain." Acta Anaesthesiol Scand **48**(5): 525-46.

Romsing, J., S. Moiniche, O. Mathiesen and J. B. Dahl (2005). "Reduction of opioid-related adverse events using opioid-sparing analgesia with COX-2 inhibitors lacks documentation: a systematic review." Acta Anaesthesiol Scand **49**(2): 133-42.

Rosenberg, W. and A. Donald (1995). "Evidence based medicine: an approach to clinical problem-solving." Bmj **310**(6987): 1122-6.

Rosenthal, R. (1979). "The 'file drawer problem' and tolerance for null results." Psychological Bulletin **86**: 638-641.

Rossat, J., M. Maillard, J. Nussberger, H. R. Brunner and M. Burnier (1999). "Renal effects of selective cyclooxygenase-2 inhibition in normotensive salt-depleted subjects." Clin Pharmacol Ther **66**(1): 76-84.

Rostom, A., C. Dube, M. Boucher, J. Joyce, T. J. Brown, L. Hooper and E. Jolicoeur (2005). "Adverse gastrointestinal effects of COX-2 inhibitors for inflammatory diseases." The Cochrane Database of Systematic Reviews **CD004519**(3).

Rostom, A., C. Dube, E. Jolicoeur, M. Boucher and J. Joyce (2004). "Gastroduodenal ulcers associated with the use of non-steroidal anti-inflammatory drugs : a systematic review of preventive pharmacological interventions." Ottawa: Canada Coordinating Office for Health Technology Assessment ; 2004. Technology overview no12 **Technology overview no12**.

Rostom, A., L. Goldkind and L. Laine (2005). "Nonsteroidal anti-inflammatory drugs and hepatic toxicity: a systematic review of randomized controlled trials in arthritis patients." Clin Gastroenterol Hepatol **3**(5): 489-98.

Sackett, D. L., W. M. Rosenberg, J. A. Gray, R. B. Haynes and W. S. Richardson (1996). "Evidence based medicine: what it is and what it isn't." Bmj **312**(7023): 71-2.

Sacks, H. S., J. Berrier, D. Reitman, V. A. Ancona-Berk and T. C. Chalmers (1987).

"Meta-analyses of randomized controlled trials." N Engl J Med **316**(8): 450-5.

Sacks, H. S., D. Reitman, D. Pagano and B. Kupelnick (1996). "Meta-analysis: an update." Mt Sinai J Med **63**(3-4): 216-24.

Sandler, R. S., S. Halabi, J. A. Baron, S. Budinger, E. Paskett, R. Keresztes, N. Petrelli, J. M. Pipas, D. D. Karp, C. L. Loprinzi, G. Steinbach and R. Schilsky (2003). "A randomized trial of aspirin to prevent colorectal adenomas in patients with previous colorectal cancer." N Engl J Med **348**(10): 883-90.

Sarau, H. M., R. S. Ames, J. Chambers, C. Ellis, N. Elshourbagy, J. J. Foley, D. B. Schmidt, R. M. Muccitelli, O. Jenkins, P. R. Murdock, N. C. Herrity, W. Halsey, G. Sathe, A. I. Muir, P. Nuthulaganti, G. M. Dytko, P. T. Buckley, S. Wilson, D. J. Bergsma and D. W. Hay (1999). "Identification, molecular cloning, expression, and characterization of a cysteinyl leukotriene receptor." Mol Pharmacol **56**(3): 657-63.

Sarzi-Puttini, P., M. A. Cimmino, R. Scarpa, R. Caporali, F. Parazzini, A. Zaninelli, F. Atzeni and B. Canesi (2005). "Osteoarthritis: an overview of the disease and its treatment strategies." Semin Arthritis Rheum **35**(1 Suppl 1): 1-10.

Schlatter, E., M. Salomonsson, A. E. Persson and R. Greger (1989). "Macula densa cells sense luminal NaCl concentration via furosemide sensitive Na+2Cl-K+ cotransport." Pflugers Arch **414**(3): 286-90.

Schlienger, R. G., H. Jick and C. R. Meier (2002). "Use of nonsteroidal anti-inflammatory drugs and the risk of first-time acute myocardial infarction." Br J Clin Pharmacol **54**(3): 327-32.

Schlondorff, D. (1993). "Renal complications of nonsteroidal anti-inflammatory drugs." Kidney Int **44**(3): 643-53.

Schmitz, P. G., S. M. Krupa, P. H. Lane, J. C. Reddington and L. Salinas-Madrigal (1994). "Acquired essential fatty acid depletion in the remnant kidney: amelioration with U-63557A." Kidney Int **46**(4): 1184-91.

Schnitzer, T. J., G. R. Burmester, E. Mysler, M. C. Hochberg, M. Doherty, E. Ehrsam, X. Gitton, G. Krammer, B. Mellein, P. Matchaba, A. Gimona and C. J. Hawkey (2004). "Comparison of lumiracoxib with naproxen and ibuprofen in the Therapeutic Arthritis Research and Gastrointestinal Event Trial (TARGET), reduction in ulcer complications: randomised controlled trial." Lancet **364**(9435):

665-74.

Schnitzer, T. J. and M. C. Hochberg (2002). "COX-2-selective inhibitors in the treatment of arthritis." Cleve Clin J Med **69 Suppl 1**: SI20-30.

Schnitzer, T. J., K. Truitt, R. Fleischmann, P. Dalgin, J. Block, Q. Zeng, J. Bolognese, B. Seidenberg and E. W. Ehrich (1999). "The safety profile, tolerability, and effective dose range of rofecoxib in the treatment of rheumatoid arthritis. Phase II Rofecoxib Rheumatoid Arthritis Study Group." Clin Ther **21**(10): 1688-702.

Schnitzer, T. J., A. L. Weaver, A. B. Polis, R. A. Petruschke and G. P. Geba (2005). "Efficacy of rofecoxib, celecoxib, and acetaminophen in patients with osteoarthritis of the knee. A combined analysis of the VACT studies." J Rheumatol **32**(6): 1093-105.

Schwab, J. M., T. Beiter, J. U. Linder, S. Laufer, J. E. Schulz, R. Meyermann and H. J. Schluesener (2003). "COX-3--a virtual pain target in humans?" Faseb J **17**(15): 2174-5.

Schwab, J. M., H. J. Schluesener and S. Laufer (2003). "COX-3: just another COX or the solitary elusive target of paracetamol?" Lancet **361**(9362): 981-2.

Schwartz, J. I., K. Vandormael, M. P. Malice, R. N. Kalyani, K. C. Lasseter, G. B. Holmes, B. J. Gertz, K. M. Gottesdiener, M. Laurenzi, K. J. Redfern and K. Brune (2002). "Comparison of rofecoxib, celecoxib, and naproxen on renal function in elderly subjects receiving a normal-salt diet." Clin Pharmacol Ther **72**(1): 50-61.

Scott, D. L., M. Shipley, A. Dawson, S. Edwards, D. P. Symmons and A. D. Woolf (1998). "The clinical management of rheumatoid arthritis and osteoarthritis: strategies for improving clinical effectiveness." Br J Rheumatol **37**(5): 546-54.

Scott, G., C. Rordorf, C. Reynolds, J. Kalbag, M. Looby, S. Milosavljev, M. Weaver, J. P. Huff and D. A. Ruff (2004). "Pharmacokinetics of lumiracoxib in plasma and synovial fluid." Clin Pharmacokinet **43**(7): 467-78.

Serhan, C. N., T. Takano and J. F. Maddox (1999). "Aspirin-triggered 15-epi-lipoxin A4 and stable analogs on lipoxin A4 are potent inhibitors of acute inflammation. Receptors and pathways." Adv Exp Med Biol **447**: 133-49.

Shamoon, M. and M. C. Hochberg (2001). "The role of acetaminophen in the management of patients with osteoarthritis." Am J Med **110 Suppl 3A**: 46S-9S.

Shea, B., C. Dube and D. Moher (2001). Assessing the quality of reports of meta-analyses: a systematic review of scales and checklists. Systematic Reviews in Health Care: Meta-analysis in context. M. Egger, G. Davey Smith and D. G. Altman. London, BMJ Publishing Group: 122-142.

Shinohara, H., M. A. Balboa, C. A. Johnson, J. Balsinde and E. A. Dennis (1999). "Regulation of delayed prostaglandin production in activated P388D1 macrophages by group IV cytosolic and group V secretory phospholipase A2s." J Biol Chem **274**(18): 12263-8.

Sibbald, B. (2006). "Vioxx should be allowed back on the market advises expert panel." Cmaj **175**(3): 234.

Silldorff, E. P., S. Yang and T. L. Pallone (1995). "Prostaglandin E2 abrogates endothelin-induced vasoconstriction in renal outer medullary descending vasa recta of the rat." J Clin Invest **95**(6): 2734-40.

Silman, A. J. (1998). Rheumatoid Arthritis. Oxford Textbook of Rheumatology. P. J. Maddison, D. Isenberg, P. Woo and D. Glass. Oxford, Oxford University: 811-828.

Silverstein, F. E., G. Faich, J. L. Goldstein, L. S. Simon, T. Pincus, A. Whelton, R. Makuch, G. Eisen, N. M. Agrawal, W. F. Stenson, A. M. Burr, W. W. Zhao, J. D. Kent, J. B. Lefkowith, K. M. Verburg and G. S. Geis (2000). "Gastrointestinal toxicity with celecoxib vs nonsteroidal anti-inflammatory drugs for osteoarthritis and rheumatoid arthritis: the CLASS study: A randomized controlled trial. Celecoxib Long-term Arthritis Safety Study." Jama **284**(10): 1247-55.

Simes, R. J. (1986). "Publication bias: the case for an international registry of clinical trials." J Clin Oncol **4**(10): 1529-41.

Simon, L. S., A. L. Weaver, D. Y. Graham, A. J. Kivitz, P. E. Lipsky, R. C. Hubbard, P. C. Isakson, K. M. Verburg, S. S. Yu, W. W. Zhao and G. S. Geis (1999). "Anti-inflammatory and upper gastrointestinal effects of celecoxib in rheumatoid arthritis: a randomized controlled trial." Jama **282**(20): 1921-8.

Singh, G. (1998). "Recent considerations in nonsteroidal anti-inflammatory drug gastropathy." Am J Med **105**(1B): 31S-38S.

Singh, G. and G. Triadafilopoulos (1999). "Epidemiology of NSAID induced gastrointestinal complications." J Rheumatol Suppl **56**: 18-24.

Slater, D., W. Dennes, R. Sawdy, V. Allport and P. Bennett (1999a). "Expression of cyclo-oxygenase types-1 and -2 in human fetal membranes throughout pregnancy." J Mol Endocrinol **22**(2): 125-30.

Slater, D. M., W. J. Dennes, J. S. Campa, L. Poston and P. R. Bennett (1999b). "Expression of cyclo-oxygenase types-1 and -2 in human myometrium throughout pregnancy." Mol Hum Reprod **5**(9): 880-4.

Slaughter, D., N. Takenaga, P. Lu, C. Assang, D. J. Walsh, B. H. Arison, D. Cui, R. A. Halpin, L. A. Geer, K. P. Vyas and T. A. Baillie (2003). "Metabolism of rofecoxib in vitro using human liver subcellular fractions." Drug Metab Dispos **31**(11): 1398-408.

Slavin, R. E. (1995). "Best evidence synthesis: an intelligent alternative to meta-analysis." J Clin Epidemiol **48**(1): 9-18.

Smith, M. C. and E. Stullenbarger (1989). "Meta-analysis: an overview." Nurs Sci Q **2**(3): 114-5.

Smith, W. L. (1986). "Prostaglandin biosynthesis and its compartmentation in vascular smooth muscle and endothelial cells." Annu Rev Physiol **48**: 251-62.

Smith, W. L. and D. L. DeWitt (1995). "Biochemistry of prostaglandin endoperoxide H synthase-1 and synthase-2 and their differential susceptibility to nonsteroidal anti-inflammatory drugs." Semin Nephrol **15**(3): 179-94.

Smith, W. L., D. L. DeWitt and R. M. Garavito (2000). "Cyclooxygenases: structural, cellular, and molecular biology." Annu Rev Biochem **69**: 145-82.

Smith, W. L., R. M. Garavito and D. L. DeWitt (1996). "Prostaglandin endoperoxide H synthases (cyclooxygenases)-1 and -2." J Biol Chem **271**(52): 33157-60.

Smith, W. L. and W. E. Lands (1972). "Oxygenation of polyunsaturated fatty acids during prostaglandin biosynthesis by sheep vesicular gland." Biochemistry **11**(17): 3276-85.

Smith, W. L., E. A. Meade and D. L. DeWitt (1994). "Interactions of PGH synthase isozymes-1 and -2 with NSAIDs." Ann N Y Acad Sci **744**: 50-7.

Smith, W. L. and I. Song (2002). "The enzymology of prostaglandin endoperoxide H synthases-1 and -2." Prostaglandins Other Lipid Mediat **68-69**: 115-28.

Solomon, D. H., J. Avorn, T. Sturmer, R. J. Glynn, H. Mogun and S. Schneeweiss (2006).

"Cardiovascular outcomes in new users of coxibs and nonsteroidal antiinflammatory drugs: high-risk subgroups and time course of risk." Arthritis Rheum **54**(5): 1378-89.

Solomon, D. H., E. W. Karlson, E. B. Rimm, C. C. Cannuscio, L. A. Mandl, J. E. Manson, M. J. Stampfer and G. C. Curhan (2003). "Cardiovascular morbidity and mortality in women diagnosed with rheumatoid arthritis." Circulation **107**(9): 1303-7.

Solomon, S. D., J. J. McMurray, M. A. Pfeffer, J. Wittes, R. Fowler, P. Finn, W. F. Anderson, A. Zauber, E. Hawk and M. Bertagnolli (2005). "Cardiovascular risk associated with celecoxib in a clinical trial for colorectal adenoma prevention." N Engl J Med **352**(11): 1071-80.

Song, I., T. M. Ball and W. L. Smith (2001). "Different suicide inactivation processes for the peroxidase and cyclooxygenase activities of prostaglandin endoperoxide H synthase-1." Biochem Biophys Res Commun **289**(4): 869-75.

Song, I. and W. L. Smith (1996). "C-terminal Ser/Pro-Thr-Glu-Leu tetrapeptides of prostaglandin endoperoxide H synthases-1 and -2 target the enzymes to the endoplasmic reticulum." Arch Biochem Biophys **334**(1): 67-72.

Sooriakumaran, P. (2006). "COX-2 inhibitors and the heart: are all coxibs the same?" Postgrad Med J **82**(966): 242-5.

Spector, T. D. and M. C. Hochberg (1994). "Methodological problems in the epidemiological study of osteoarthritis." Ann Rheum Dis **53**(2): 143-6.

Spencer, A. G., E. Thuresson, J. C. Otto, I. Song, T. Smith, D. L. DeWitt, R. M. Garavito and W. L. Smith (1999). "The membrane binding domains of prostaglandin endoperoxide H synthases 1 and 2. Peptide mapping and mutational analysis." J Biol Chem **274**(46): 32936-42.

Spencer, A. G., J. W. Woods, T. Arakawa, Singer, II and W. L. Smith (1998). "Subcellular localization of prostaglandin endoperoxide H synthases-1 and -2 by immunoelectron microscopy." J Biol Chem **273**(16): 9886-93.

Squires, B. (1991). "Abstracts: the need for improvement." Canadian Medical Association Journal **144**: 421.

Squires, B. (1994). "Evaluating structure abstracts." Canadian Medical Association Journal **150**: 1535.

Srikanth, V. K., J. L. Fryer, G. Zhai, T. M. Winzenberg, D. Hosmer and G. Jones (2005). "A meta-analysis of sex differences prevalence, incidence and severity of osteoarthritis." Osteoarthritis Cartilage **13**(9): 769-81.

Stahl, R. A., S. Kudelka, M. Paravicini and P. Schollmeyer (1986). "Prostaglandin and thromboxane formation in glomeruli from rats with reduced renal mass." Nephron **42**(3): 252-7.

Steinbach, G., P. M. Lynch, R. K. Phillips, M. H. Wallace, E. Hawk, G. B. Gordon, N. Wakabayashi, B. Saunders, Y. Shen, T. Fujimura, L. K. Su and B. Levin (2000). "The effect of celecoxib, a cyclooxygenase-2 inhibitor, in familial adenomatous polyposis." N Engl J Med **342**(26): 1946-52.

Steinberg, D. (1997). "Low density lipoprotein oxidation and its pathobiological significance." J Biol Chem **272**(34): 20963-6.

Steinberg, D., S. Parthasarathy, T. E. Carew, J. C. Khoo and J. L. Witztum (1989). "Beyond cholesterol. Modifications of low-density lipoprotein that increase its atherogenicity." N Engl J Med **320**(14): 915-24.

Steinbrecher, U. P. and M. Lougheed (1992). "Scavenger receptor-independent stimulation of cholesterol esterification in macrophages by low density lipoprotein extracted from human aortic intima." Arterioscler Thromb **12**(5): 608-25.

Stern, J. M. and R. J. Simes (1997). "Publication bias: evidence of delayed publication in a cohort study of clinical research projects." Bmj **315**(7109): 640-5.

Sterne, J. A. C., C. Bartlett, P. Juni and M. Egger (2000). Do we need comprehensive literature searches? A study of publication and language bias in meta-analyses of controlled trials. 3rd Symposium on Systematic Reviews: beyond the basics., Oxford.

Stichtenoth, D. O. and J. C. Frolich (2003). "The second generation of COX-2 inhibitors: what advantages do the newest offer?" Drugs **63**(1): 33-45.

Stokes, J. B. (1979). "Effect of prostaglandin E2 on chloride transport across the rabbit thick ascending limb of Henle. Selective inhibitions of the medullary portion." J Clin Invest **64**(2): 495-502.

Stokes, J. B. and J. P. Kokko (1977). "Inhibition of sodium transport by prostaglandin E2 across the isolated, perfused rabbit collecting tubule." J Clin Invest **59**(6): 1099-

104.

Straube, S., S. Derry, H. J. McQuay and R. A. Moore (2005). "Effect of preoperative Cox-II-selective NSAIDs (coxibs) on postoperative outcomes: a systematic review of randomized studies." Acta Anaesthesiol Scand **49**(5): 601-13.

Strauss, S. E. and F. A. McAlister (1999). "Evidence-based medicine: past, present, and future." Ann R Coll Physicians Surg Can **32**: 260-4.

Sugimoto, Y., E. Segi, K. Tsuboi, A. Ichikawa and S. Narumiya (1998). "Female reproduction in mice lacking the prostaglandin F receptor. Roles of prostaglandin and oxytocin receptors in parturition." Adv Exp Med Biol **449**: 317-21.

Suzuki, T., K. Watanabe, Y. Kanaoka, T. Sato and O. Hayaishi (1997). "Induction of hematopoietic prostaglandin D synthase in human megakaryocytic cells by phorbol ester." Biochem Biophys Res Commun **241**(2): 288-93.

Swan, S. K., D. W. Rudy, K. C. Lasseter, C. F. Ryan, K. L. Buechel, L. J. Lambrecht, M. B. Pinto, S. C. Dilzer, O. Obrda, K. J. Sundblad, C. P. Gumbs, D. L. Ebel, H. Quan, P. J. Larson, J. I. Schwartz, T. A. Musliner, B. J. Gertz, D. C. Brater and S. L. Yao (2000). "Effect of cyclooxygenase-2 inhibition on renal function in elderly persons receiving a low-salt diet. A randomized, controlled trial." Ann Intern Med **133**(1): 1-9.

Taddio, A., T. Pain, F. F. Fassos, H. Boon, A. L. Ilersich and T. R. Einarson (1994). "Quality of nonstructured and structured abstracts of original research articles in the British Medical Journal, the Canadian Medical Association Journal and the Journal of the American Medical Association." Cmaj **150**(10): 1611-5.

Tang, M. S., R. A. Copeland and T. M. Penning (1997). "Detection of an Fe2+-protoporphyrin-IX intermediate during aspirin-treated prostaglandin H2 synthase II catalysis of arachidonic acid to 15-HETE." Biochemistry **36**(24): 7527-34.

Tanne, J. H. (2005). "Merck faces ongoing claims after Texan ruling on rofecoxib." Bmj **331**(7515): 471.

Tanne, J. H. (2006). "Court awards claimant 13.5m dollars in rofecoxib lawsuit." Bmj **332**(7547): 927.

Taylor Halvorsen, K. (1994). The reporting format. The handbook of research synthesis. H. Cooper and L. V. Hedges. New York, Russell Sage Foundation**:** 425-37.

Tegeder, I., W. Neupert, H. Guhring and G. Geisslinger (2000). "Effects of selective and unselective cyclooxygenase inhibitors on prostanoid release from various rat organs." J Pharmacol Exp Ther **292**(3): 1161-8.

Temple, R. (1999). "Meta-analysis and epidemiologic studies in drug development and postmarketing surveillance." Jama **281**(9): 841-4.

Thacker, S. B., H. B. Peterson and D. F. Stroup (1996). "Meta-analysis for the obstetrician-gynecologist." Am J Obstet Gynecol **174**: 1403-7.

Thal, L. J., S. H. Ferris, L. Kirby, G. A. Block, C. R. Lines, E. Yuen, C. Assaid, M. L. Nessly, B. A. Norman, C. C. Baranak and S. A. Reines (2005). "A randomized, double-blind, study of rofecoxib in patients with mild cognitive impairment." Neuropsychopharmacology **30**(6): 1204-15.

The John Hopkins University, D. o. R. 2006 Education: diagnostic and therapeutic guidelines (Osteoarthritis) John Hopkins Arhtitis Webmaster Baltimore-Washington, USA, 2006 Available from: (http://www.hopkins-arthritis.com/edu/acr/acr.html#class_hand) [Accessed on: 6th of February 2006]

The Ohio State University School of Allied Medical Professions. (2005) The model project Ohio, Available from: (http://www.amp.osu.edu/modelproject/) [Accessed on: 9th September 2006]

Thun, M. J., S. J. Henley and C. Patrono (2002). "Nonsteroidal anti-inflammatory drugs as anticancer agents: mechanistic, pharmacologic, and clinical issues." J Natl Cancer Inst **94**(4): 252-66.

Topol, E. J. (2004). "Failing the public health--rofecoxib, Merck, and the FDA." N Engl J Med **351**(17): 1707-9.

Topol, E. J. (2005). "Arthritis medicines and cardiovascular events--"house of coxibs"." Jama **293**(3): 366-8.

Towheed, T. E. (2005). "Systematic review of therapies for osteoarthritis of the hand." Osteoarthritis Cartilage **13**(6): 455-62.

Traynor, T. R., A. Smart, J. P. Briggs and J. Schnermann (1999). "Inhibition of macula densa-stimulated renin secretion by pharmacological blockade of cyclooxygenase-2." Am J Physiol **277**(5 Pt 2): F706-10.

Tsai, A., R. J. Kulmacz and G. Palmer (1995). "Spectroscopic evidence for reaction of

prostaglandin H synthase-1 tyrosyl radical with arachidonic acid." J Biol Chem **270**(18): 10503-8.

Tugwell, P., M. G. Judd, J. F. Fries, G. Singh and G. A. Wells (2005). "Powering our way to the elusive side effect: a composite outcome 'basket' of predefined designated endpoints in each organ system should be included in all controlled trials." J Clin Epidemiol **58**(8): 785-90.

Ushikubi, F., E. Segi, Y. Sugimoto, T. Murata, T. Matsuoka, T. Kobayashi, H. Hizaki, K. Tuboi, M. Katsuyama, A. Ichikawa, T. Tanaka, N. Yoshida and S. Narumiya (1998). "Impaired febrile response in mice lacking the prostaglandin E receptor subtype EP3." Nature **395**(6699): 281-4.

Van Hecken, A., J. I. Schwartz, M. Depre, I. De Lepeleire, A. Dallob, W. Tanaka, K. Wynants, A. Buntinx, J. Arnout, P. H. Wong, D. L. Ebel, B. J. Gertz and P. J. De Schepper (2000). "Comparative inhibitory activity of rofecoxib, meloxicam, diclofenac, ibuprofen, and naproxen on COX-2 versus COX-1 in healthy volunteers." J Clin Pharmacol **40**(10): 1109-20.

Vane, J. R. (1971). "Inhibition of prostaglandin synthesis as a mechanism of action for aspirin-like drugs." Nat New Biol **231**(25): 232-5.

Vane, J. R. (2002). "Biomedicine. Back to an aspirin a day?" Science **296**(5567): 474-5.

Villalba, M. L. (2005) Vioxx Excerpts from primary review of NDA 21-042-Osteoarthritis US Food and Drug Administration Rockville, Maryland, Available from: (http://www.fda.gov/ohrms/dockets/ac/05/briefing/2005-4090B1_05_F-FDA-Tab-D-1.pdf) [Accessed on: 11th July 2006]

Walter, M. F., R. F. Jacob, C. A. Day, R. Dahlborg, Y. Weng and R. P. Mason (2004). "Sulfone COX-2 inhibitors increase susceptibility of human LDL and plasma to oxidative modification: comparison to sulfonamide COX-2 inhibitors and NSAIDs." Atherosclerosis **177**(2): 235-43.

Wang, D., M. Wang, Y. Cheng and G. A. Fitzgerald (2005). "Cardiovascular hazard and non-steroidal anti-inflammatory drugs." Curr Opin Pharmacol **5**(2): 204-10.

Wang, J. L., H. F. Cheng and R. C. Harris (1999). "Cyclooxygenase-2 inhibition decreases renin content and lowers blood pressure in a model of renovascular hypertension." Hypertension **34**(1): 96-101.

Wang, J. L., H. F. Cheng, S. Shappell and R. C. Harris (2000). "A selective cyclooxygenase-2 inhibitor decreases proteinuria and retards progressive renal injury in rats." Kidney Int **57**(6): 2334-42.

Warner, T. D., F. Giuliano, I. Vojnovic, A. Bukasa, J. A. Mitchell and J. R. Vane (1999). "Nonsteroid drug selectivities for cyclo-oxygenase-1 rather than cyclo-oxygenase-2 are associated with human gastrointestinal toxicity: a full in vitro analysis." Proc Natl Acad Sci U S A **96**(13): 7563-8.

Warner, T. D. and J. A. Mitchell (2004). "Cyclooxygenases: new forms, new inhibitors, and lessons from the clinic." Faseb J **18**(7): 790-804.

Watson, D. J., S. E. Harper, P. L. Zhao, J. A. Bolognese and T. J. Simon (2001). "Gastrointestinal medications and procedures in osteoarthritis patients treated with rofecoxib compared with nonselective NSAIDs." MedGenMed **3**(4): 6.

Watson, D. J., S. E. Harper, P. L. Zhao, H. Quan, J. A. Bolognese and T. J. Simon (2000). "Gastrointestinal tolerability of the selective cyclooxygenase-2 (COX-2) inhibitor rofecoxib compared with nonselective COX-1 and COX-2 inhibitors in osteoarthritis." Arch Intern Med **160**(19): 2998-3003.

Watson, D. J., T. Rhodes, B. Cai and H. A. Guess (2002). "Lower risk of thromboembolic cardiovascular events with naproxen among patients with rheumatoid arthritis." Arch Intern Med **162**(10): 1105-10.

Watson, D. J., Q. Yu, J. A. Bolognese, A. S. Reicin and T. J. Simon (2004). "The upper gastrointestinal safety of rofecoxib vs. NSAIDs: an updated combined analysis." Curr Med Res Opin **20**(10): 1539-48.

Watson, M. C., S. T. Brookes, J. R. Kirwan and A. Faulkner (2000). "Non-aspirin, non-steroidal anti-inflammatory drugs for osteoarthritis of the knee." Cochrane Database Syst Rev(2): CD000142.

Wei, C., R. J. Kulmacz and A. L. Tsai (1995). "Comparison of branched-chain and tightly coupled reaction mechanisms for prostaglandin H synthase." Biochemistry **34**(26): 8499-512.

Weinblatt, M. E. (2003). "Rheumatoid arthritis in 2003: where are we now with treatment?" Ann Rheum Dis **62 Suppl 2**: ii94-6.

Weir, M. R., R. S. Sperling, A. Reicin and B. J. Gertz (2003). "Selective COX-2 inhibition

and cardiovascular effects: a review of the rofecoxib development program." Am Heart J **146**(4): 591-604.

West, P. M. and C. Fernandez (2003). "Safety of COX-2 inhibitors in asthma patients with aspirin hypersensitivity." Ann Pharmacother **37**(10): 1497-501.

Weyand, C. M., J. J. Goronzy, G. Liuzzo, S. L. Kopecky, D. R. Holmes, Jr. and R. L. Frye (2001). "T-cell immunity in acute coronary syndromes." Mayo Clin Proc **76**(10): 1011-20.

Whelton, A., J. G. Fort, J. A. Puma, D. Normandin, A. E. Bello and K. M. Verburg (2001). "Cyclooxygenase-2--specific inhibitors and cardiorenal function: a randomized, controlled trial of celecoxib and rofecoxib in older hypertensive osteoarthritis patients." Am J Ther **8**(2): 85-95.

Whelton, A., C. J. Maurath, K. M. Verburg and G. S. Geis (2000). "Renal safety and tolerability of celecoxib, a novel cyclooxygenase-2 inhibitor." Am J Ther **7**(3): 159-75.

Whelton, A., G. Schulman, C. Wallemark, E. J. Drower, P. C. Isakson, K. M. Verburg and G. S. Geis (2000). "Effects of celecoxib and naproxen on renal function in the elderly." Arch Intern Med **160**(10): 1465-70.

Whelton, A., W. B. White, A. E. Bello, J. A. Puma and J. G. Fort (2002). "Effects of celecoxib and rofecoxib on blood pressure and edema in patients > or =65 years of age with systemic hypertension and osteoarthritis." Am J Cardiol **90**(9): 959-63.

White, W. B., G. Faich, A. Whelton, C. Maurath, N. J. Ridge, K. M. Verburg, G. S. Geis and J. B. Lefkowith (2002). "Comparison of thromboembolic events in patients treated with celecoxib, a cyclooxygenase-2 specific inhibitor, versus ibuprofen or diclofenac." Am J Cardiol **89**(4): 425-30.

White, W. B., J. Kent, A. Taylor, K. M. Verburg, J. B. Lefkowith and A. Whelton (2002). "Effects of celecoxib on ambulatory blood pressure in hypertensive patients on ACE inhibitors." Hypertension **39**(4): 929-34.

Whitehead, A. and J. Whitehead (1991). "A general parametric approach to the meta-analysis of randomized clinical trials." Stat Med **10**(11): 1665-77.

Willoughby, D. A., A. R. Moore and P. R. Colville-Nash (2000). "COX-1, COX-2, and COX-3 and the future treatment of chronic inflammatory disease." Lancet

355(9204): 646-8.

Wilson, A. and D. A. Henry (1992). "Meta-analysis. Part 2: Assessing the quality of published meta-analyses." Med J Aust **156**(3): 173-4, 177-80, 184-7.

Witztum, J. L. and J. A. Berliner (1998). "Oxidized phospholipids and isoprostanes in atherosclerosis." Curr Opin Lipidol **9**(5): 441-8.

Wolfe, F., B. Freundlich and W. L. Straus (2003). "Increase in cardiovascular and cerebrovascular disease prevalence in rheumatoid arthritis." J Rheumatol **30**(1): 36-40.

Wolfe, M. M., D. R. Lichtenstein and G. Singh (1999). "Gastrointestinal toxicity of nonsteroidal antiinflammatory drugs." N Engl J Med **340**(24): 1888-99.

Wong, D., M. Wang, Y. Cheng and G. A. Fitzgerald (2005). "Cardiovascular hazard and non-steroidal anti-inflammatory drugs." Curr Opin Pharmacol **5**(2): 204-10.

Wong, M., P. Chowienczyk and B. Kirkham (2005). "Cardiovascular issues of COX-2 inhibitors and NSAIDs." Aust Fam Physician **34**(11): 945-8.

Woodworth, T. G., D. E. Furst, V. Strand, J. Kempeni, H. Fenner, C. S. Lau, F. Miller, R. Day, J. Lipani and P. Brooks (2001). "Standardizing assessment of adverse effects in rheumatology clinical trials. Status of OMERACT Toxicity Working Group March 2000: towards a common understanding of comparative toxicity/safety profiles for antirheumatic therapies." J Rheumatol **28**(5): 1163-9.

Wright, J. M. (2002). "The double-edged sword of COX-2 selective NSAIDs." Cmaj **167**(10): 1131-7.

Wu, G., C. Wei, R. J. Kulmacz, Y. Osawa and A. L. Tsai (1999). "A mechanistic study of self-inactivation of the peroxidase activity in prostaglandin H synthase-1." J Biol Chem **274**(14): 9231-7.

Xiao, G., W. Chen and R. J. Kulmacz (1998). "Comparison of structural stabilities of prostaglandin H synthase-1 and -2." J Biol Chem **273**(12): 6801-11.

Xie, W. L., J. G. Chipman, D. L. Robertson, R. L. Erikson and D. L. Simmons (1991). "Expression of a mitogen-responsive gene encoding prostaglandin synthase is regulated by mRNA splicing." Proc Natl Acad Sci U S A **88**(7): 2692-6.

Yang, T., J. B. Schnermann and J. P. Briggs (1999). "Regulation of cyclooxygenase-2 expression in renal medulla by tonicity in vivo and in vitro." Am J Physiol **277**(1 Pt

2): F1-9.

Yokomizo, T., T. Izumi, K. Chang, Y. Takuwa and T. Shimizu (1997). "A G-protein-coupled receptor for leukotriene B4 that mediates chemotaxis." Nature **387**(6633): 620-4.

Zhang, J., E. L. Ding and Y. Song (2006). "Adverse Effects of Cyclooxygenase 2 Inhibitors on Renal and Arrhythmia Events: Meta-analysis of Randomized Trials." Jama.

Zhang, W., M. Doherty, N. Arden, B. Bannwarth, J. Bijlsma, K. P. Gunther, H. J. Hauselmann, G. Herrero-Beaumont, K. Jordan, P. Kaklamanis, B. Leeb, M. Lequesne, S. Lohmander, B. Mazieres, E. Martin-Mola, K. Pavelka, A. Pendleton, L. Punzi, B. Swoboda, R. Varatojo, G. Verbruggen, I. Zimmermann-Gorska and M. Dougados (2005). "EULAR evidence based recommendations for the management of hip osteoarthritis: report of a task force of the EULAR Standing Committee for International Clinical Studies Including Therapeutics (ESCISIT)." Ann Rheum Dis **64**(5): 669-81.

Zimmermann, K. C., M. Sarbia, K. Schror and A. A. Weber (1998). "Constitutive cyclooxygenase-2 expression in healthy human and rabbit gastric mucosa." Mol Pharmacol **54**(3): 536-40.

Chapter 9

Publications

Poster presented in the National Institute of Clinical Excellence (NICE) 2006: Tackling Health Priorities Annual Conference and Exhibition on the 6-7th of December 2006 at the ICC, Birmingham.

QUALITY ASSESSMENT OF SYSTEMATIC REVIEWS REPORTING CARDIOVASCULAR, RENAL OR CEREBROVASCULAR ADVERSE EVENTS OF A SPECIFIC CYCLOOXYGENASE-2 INHIBITOR, ROFECOXIB

A. LALATSA (lalatsaa@aston.ac.uk) , K.A. WILSON (K.A.Wilson@aston.ac.uk)
SCHOOL OF LIFE AND HEALTH SCIENCES, ASTON UNIVERSITY, BIRMINGHAM, B4 7ET, UK

BACKGROUND AND OBJECTIVES

Systematic reviews and meta-analyses of randomised controlled trials (RCTs) provide the highest level of evidence, but the quality of published systematic reviews and meta-analyses has not been adequately assessed. Therefore, the quality of reporting of all the systematic reviews and meta-analyses providing information concerning the cardiovascular, renal and cerebrovascular safety of rofecoxib for its long-term indications in adults was assessed.

DESIGN

Quality Assessment

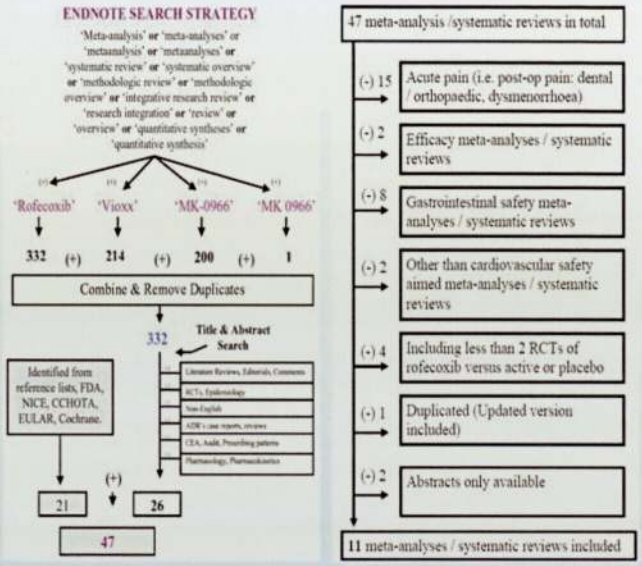
METHODS

Medline / Pubmed, the Cochrane Collaboration Database, as well as the Food and Drug Administration (FDA), the National Institute of Clinical Excellence (NICE), the European League Against Rheumatism (EULAR), the Canadian Coordinating Office of Health Technology Assessment (CCHOTA) websites were systematically searched by an independent reviewer to identify systematic reviews or meta-analyses reporting the cardiovascular, renal and cerebrovascular toxicity of rofecoxib. The resulting set of citations was then hand searched by title, MESH, and whenever available abstract. All identified literature reviews as well as the reference lists of recovered articles were further screened for any additional citations (Fig. 1). Quality was assessed using the QUOROM (Quality of Reporting of Meta-analyses) checklist (1). Quality scores were divided into quartiles (<25% i.e. very poor, 25-49% i.e. poor, 50-75% i.e. acceptable and >75% i.e. good quality) to assist judgement of quality (2).

INCLUSION & EXCLUSION CRITERIA

INCLUSION CRITERIA: Systematic reviews and meta-analyses of randomised controlled trials (RCTs) provide the highest level of evidence, but the quality of published systematic reviews and meta-analyses has not been adequately assessed. Therefore, the quality of reporting of all the systematic reviews and meta-analyses providing information concerning the cardiovascular, renal and cerebrovascular safety of rofecoxib for its long-term indications in adults was assessed.

EXCLUSION CRITERIA: Articles concerning solely other specific or selective COX-2 inhibitors or traditional NSAIDs, children, cancer indications, acute pain, or including information only on other adverse-effects apart from cardiovascular, renal or cerebrovascular. Literature or narrative reviews were excluded.



(Figure 1: Search strategy results and flow diagram)

Table 1: Overall quality score by category and item (N = 11)

Category	Item	Mean (range)	Quality grade
Title	Identified as a meta-analysis or RCTs in the title?	27.30% (0-100%)	Poor
Abstract		69.70% (33.3-100%)	Acceptable
	Structured format?	72.73%	
	Objective/clinical question described?	100.00%	
	Databases and sources listed? Selection criteria, etc. been described sufficiently?	54.55%	
		36.36%	
	Have characteristics been described?	63.64%	
	Main results reported?	90.91%	
Introduction	Clinical problem described?	81.80% (0-100%)	Good
Methods		71.21% (33.3-100%)	Acceptable
	Described searching methods and sources?	90.91%	
	Inclusion and exclusion?	72.73%	
	Validity assessment described?	36.36%	
	Processes for data abstraction described?	45.45%	
	Study design, and clinical heterogeneity assessed?	81.82%	
	Principa measure of effect, etc. been described?	100.00%	
Results		45.46% (0-66.7%)	Poor
	Trial flow chart provided?	18.18%	
	Descriptive data presented?	72.73%	
	Authors reported agreement, effect sizes, and CIs, etc.?	45.45%	
		63.64%	
Discussion	Summarised key findings, etc?	(0-100%)	Acceptable
		63.12%	
Overall		(33.3-94.4%)	Acceptable

RESULTS

47 systematic reviews and meta-analyses were identifiec from the preliminary search by title, MESH and whenever available abstract; 36 were excluded [acute pain (15), efficacy only endpoints (2), gastrointestinal safety (8), hepatotoxicity (1), bronchospasm (1), duplicated (1), included less than 2 randomised controlled trials comparing rofecoxib (4), abstracts available only (2), published only online (2)] and 11 were included (Fig. 1). The mean overall quality score is 63.13% (SD: 21.41% range: 33.3-94.4%). The overall score for Title (27.30% and Results (45.46%) were poor, while overall scores fo Abstracts (69.70%), Methods (71.21%) and Discussion (63.60%) were acceptable. Good quality scores were only found for Introduction (81.80%) [Table 1]. All systematic reviews and meta-analyses were published after the QUOROM quality guidelines were published.

CONCLUSION

Despite quality guidelines, the average quality c published systematic reviews or meta-analyses of COX-2 specific inhibitor is barely acceptable (63.12%). The worldwide withdrawal of rofecoxib from the marke on the 30th of September 2004 emphasises further th need for adherence to standardised reporting an quality guidelines to allow researchers to be able t synthesise available information in a quantitative an unbiased manner that would allow for timely an appropriate decisions.

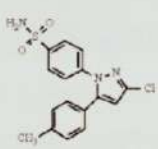
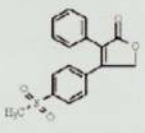
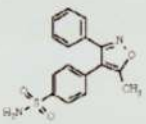
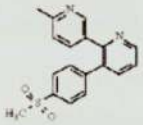
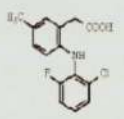
REFERENCES:

1. Moher, D., D. J. Cook, S. Eastwood, I. Olkin, D. Rennie and D. F. Stroup (1999). "Improving the quality of reports of meta-analyses of randomised controlled trials: the QUOROM statement. Quality of Reporting of Meta-analyses." Lancet 354 (9193): 1896-900.
2. Hemels, M. E., C. Vicente, H. Sadri, M. J. Masson, and T. R. Einarson (2004). "Quality assessment of meta-analyses of RCT of pharmacotherapy in major depressive disorder." Curr. Med. Res. Opin. 20 (4): 477-84.

Appendix I

Pharmacodynamics & Pharmacokinetics of Coxibs

Table A.1 Pharmacodynamic and pharmacokinetics of orally administered COX-2 inhibitors (Cochrane, Jarvis et al. 2002; Barkin and Buvanendran 2004; Lyseng-Williamson and Curran 2004; Patrignani, Tacconelli et al. 2005)

	Celecoxib	Rofecoxib	Valdecoxib	Etoricoxib	Lumiracoxib
Chemistry	Sulphonamide 	Sulphonyl 	Sulphonamide 	Sulphonyl 	Phenylacetic acid 
COX-1 / COX-2 ratio	30	276	61	344	433

Pharmacokinetics

Oral Bioavailability (F,%)	22-40	92-93	83	100	74
Tmax (h)	2-4	2-3	2.3	1	2-3
Cmax (µg/ml) peak plasma level	0.7 (>65 years 40% higher)	0.2 (53% ↑ in hepatic insufficiency)	-	3.6	4.18
Half-life (h)	11 (fasting)	10-17	8-11	22	3-6
Vol.Distribution (L)	455	86-91	86	120	9
Plasma Protein Binding (%)	97	87	98	92	>98
Css (days)	≥5	4	4	7	2-3 hours
Kinetics	Linear	Non-linear (saturable)	Non-linear	Linear	Linear

Metabolism / Excretion

Main Pathway	Oxidation CYP450 (2C9, 3A4)	Cytosolic reduction	Oxidation CYP450 (2C9, 3A4)	Oxidation CYP450 (3A4)	Oxidation CYP450 (2C9)
Urinary excretion (%)	29	72	70	70	54
Faeces excretion (%)	57	14	Hepatic (primary)	20	43

Appendix II

OA & RA Classification Criteria

ACR Clinical Classification Criteria for OA of the Hip
(Altman, Alarcon et al. 1991)

Using history, physical examination & laboratory findings:

Pain in the hip
AND

Internal hip rotation $<15^{o}$ AND ESR $\subseteq$ 45 mm/hour or hip flexion $\subseteq 115^{o}$ if ESR unavailable	OR	Internal hip rotation $\geq 15^{o}$ AND Pain associated with internal hip AND Morning stiffness of hip $\subseteq$ 1 hour AND Over 50 years of age

Using history, physical examination, laboratory & radiographic finding:
Traditional format:

Pain in the hip
AND 2 OF THE FOLLOWING

ESR < 20mm/hour

Radiographic femoral
and/or acetabular osteophytes

Radiographic joint space narrowing
(superior, axial, and/or medial)

Classification tree:

Pain in the hip
AND

Radiographic femoral and/or Acetabular osteophytes	OR	ESR$\subseteq$ 20mm/hour and Radiographic axial joint space narrowing

ACR Clinical Classification Criteria for OA of the Knee
(Altman, Asch et al. 1986; Altman, Alarcon et al. 1991)

Using history and physical examination:

Pain in the knee

AND 3 OF THE FOLLOWING

Over 50 years of age
Less than 30 minutes of morning stiffness
Crepitus on active motion
Bony tenderness
Bony enlargement
No palpable warmth of synovium

Using history, physical examination & radiographic findings:

Pain in the knee

AND 1 OF THE FOLLOWING

Over 50 years of age
Less than 30 minutes of morning stiffness
Crepitus on active motion and osteophytes

Using history, physical examination & laboratory findings:

Pain in the knee

AND % OF THE FOLLOWING

Over 50 years
Less than 30 minutes of morning stiffness
Crepitus on active motion and osteophytes
Bony tenderness
Bony enlargement
No palpable warmth of synovium
ESR < 40mm/hour
Rheumatoid Factor (RF) < 1:40
Synovial Fluid (SF) signs of OA

ACR Clinical Classification Criteria for OA of the Hand (The John Hopkins University 2006)

Pain, aching or stiffness in the hand

AND 3 OF THE FOLLOWING

Hard tissue enlargement of 2 or more of the following joints:
2nd and 3rd distal interphalangeal,
the 2nd and 3rd proximal interphalangeal.
and the 1st carpometacarpal joints of both hands *

Hard tissue enlargement of 2 or more distal interphalangeal joints

Less than 3 swollen MCP joints

* Deformity of at least one of the joints listed above.
* MCP = metacarpophalangeal

Revised ARA Clinical Classification Criteria for RA (2000) (NICE 2001)

For classification purposes, a patient is said to have RA if he or she has satisfied at least 4 of the following 7 criteria. Criteria 1 through 4 must have been present for at least 6 weeks. Patients with 2 clinical diagnoses are not excluded. Designation as classic, definite, or probable RA is not to be made.

1. Morning stiffness: Morning stiffness in and around the joints, lasting at least 1 hour before maximal improvement.
2. Arthritis of 3 or more joint areas: At least 3 joints areas simultaneously have had soft tissue swelling or fluid (not bony overgrowth alone) observed by a doctor; the 14 possible joint areas are right or left proximal interphalangeal (PIP) joints, metacarpophalangeal (MCP) joints, wrist, elbow, knee, ankle, and metatarsophalangeal (MPT) joints.

3. Arthritis of hand joints: At least 1 area swollen (as defined above) in a wrist, MCP, or PIP joint.
4. Symmetric arthritis: Simultaneous involvement of the same joint areas (see 2 above) on both sides of the body (bilateral involvement of PIPs, MCPs, or MTPs is acceptable without absolute symmetry).
5. Rheumatoid nodules: Subcutaneous nodules, over bony prominences, or extensor surfaces, or in juxta-articular regions, observed by a doctor.
6. Serum rheumatoid factor: Demonstration of abnormal amounts of serum rheumatoid factor by any method for which the result has been positive in <5% of normal control subjects.

Radiographic changes

Radiographic changes typical of RA on posteroanterior hand and wrist radiographs, which must include erosions or unequivocal bony decalcification localised to or most marked adjacent to the involved joints (osteoarthritis changes alone do not qualify).

Appendix III

Search Results

Additional File 1

1. Aw, T.J., et al., 2005. Meta-analysis of Cyclooxygenase-2 Inhibitors and Their Effects on Blood Pressure. *Arch Intern Med.*
2. Barden, J., et al., 2002. Single-dose rofecoxib for acute postoperative pain in adults: a quantitative systematic review. *BMC Anesthesiol*, **2**(1): p. 4.
3. Barden, J., et al., 2004. Single dose oral rofecoxib for postoperative pain. *Cochrane Database Syst Rev*, (1): p. CD004604.
4. Barden, J., et al., 2005. Single dose oral rofecoxib for postoperative pain. *Cochrane Database Syst Rev*, (1): p. CD004604.
5. Chen, L.C., R.A. Elliott, and D.M. Ashcroft, 2004. Systematic review of the analgesic efficacy and tolerability of COX-2 inhibitors in post-operative pain control. *J Clin Pharm Ther*, **29**(3): p. 215-29.
6. Desjardins, P.J., et al., 2005. The time to onset and overall analgesic efficacy of rofecoxib 50 mg: a meta-analysis of 13 randomized clinical trials. *Clin J Pain*, **21**(3): p. 241-50.
7. Edwards, J.E., R.A. Moore, and H.J. McQuay, 2004. Rofecoxib for dysmenorrhoea: meta-analysis using individual patient data. *BMC Womens Health*, **4**(1): p. 5.
8. Edwards, J.E., R.A. Moore, and H.J. McQuay, 2004. Individual patient meta-analysis of single-dose rofecoxib in postoperative pain. *BMC Anesthesiol*, **4**(1): p. 3.
9. Gagnier, J.J., S. Chrubasik, and E. Manheimer, 2004. Harpgophytum procumbens for osteoarthritis and low back pain: a systematic review. *BMC Complement Altern Med*, **4**: p. 13.
10. Garner, S., et al., 2002. Rofecoxib for the treatment of rheumatoid arthritis. *Cochrane Database Syst Rev*, (3): p. CD003685.
11. Garner, S., et al., 2005b. Rofecoxib for osteoarthritis. *Cochrane Database Syst Rev*, (1): p. CD005115.
12. Garner, S., et al., 2005a. Rofecoxib for rheumatoid arthritis. *Cochrane Database Syst Rev*, (1): p. CD003685.
13. Juni, P., et al., 2004. Risk of cardiovascular events and rofecoxib: cumulative meta-analysis. *Lancet*, **364**(9450): p. 2021-9.
14. Konstam, M.A., et al., 2001. Cardiovascular thrombotic events in controlled, clinical trials of rofecoxib. *Circulation*, **104**(19): p. 2280-8.
15. Langman, M.J., et al., 1999. Adverse upper gastrointestinal effects of rofecoxib compared with NSAIDs. *Jama*, **282**(20): p. 1929-33.
16. Lee, C., et al., 2005. Need for common internal controls when assessing the relative efficacy of pharmacologic agents using a meta-analytic approach: case

study of cyclooxygenase 2-selective inhibitors for the treatment of osteoarthritis. *Arthritis Rheum*, **53**(4): p. 510-8.

17. Moore, R.A., J.E. Edwards, and H.J. McQuay, 2005. Acute pain: individual patient meta-analysis shows the impact of different ways of analysing and presenting results. *Pain*, **116**(3): p. 322-31.
18. Moore, R.A., et al., 2005. Tolerability and adverse events in clinical trials of celecoxib in osteoarthritis and rheumatoid arthritis: systematic review and meta-analysis of information from company clinical trial reports. *Arthritis Res Ther*, **7**(3): p. R644-65.
19. Morrison, B.W., et al., 2000. The optimal analgesic dose of rofecoxib: Overview of six randomized controlled trials. *J Am Dent Assoc*, **131**(12): p. 1729-37.
20. Mukherjee, D., S.E. Nissen, and E.J. Topol, 2001. Risk of cardiovascular events associated with selective COX-2 inhibitors. *Jama*, **286**(8): p. 954-9.
21. Romsing, J. and S. Moiniche, 2004. A systematic review of COX-2 inhibitors compared with traditional NSAIDs, or different COX-2 inhibitors for post-operative pain. *Acta Anaesthesiol Scand*, **48**(5): p. 525-46.
22. Rostom, A., L. Goldkind, and L. Laine, 2005. Nonsteroidal anti-inflammatory drugs and hepatic toxicity: a systematic review of randomized controlled trials in arthritis patients. *Clin Gastroenterol Hepatol*, **3**(5): p. 489-98.
23. Straube, S., et al., 2005. Effect of preoperative Cox-II-selective NSAIDs (coxibs) on postoperative outcomes: a systematic review of randomized studies. *Acta Anaesthesiol Scand*, **49**(5): p. 601-13.
24. Watson, D.J., et al., 2000. Gastrointestinal tolerability of the selective cyclooxygenase-2 (COX-2) inhibitor rofecoxib compared with nonselective COX-1 and COX-2 inhibitors in osteoarthritis. *Arch Intern Med*, **160**(19): p. 2998-3003.
25. Watson, D.J., et al., 2004. The upper gastrointestinal safety of rofecoxib vs. NSAIDs: an updated combined analysis. *Curr Med Res Opin*, **20**(10): p. 1539-48.
26. West, P.M. and C. Fernandez, 2003. Safety of COX-2 inhibitors in asthma patients with aspirin hypersensitivity. *Ann Pharmacother*, **37**(10): p. 1497-501.

Additional File 2

1. 2003. [Meta-analysis on acute pain. Rofecoxib helped three quarters of patients]. *MMW Fortschr Med*, **145**(31-32): p. 57.
2. Aw, T.J., et al., 2005. Meta-analysis of Cyclooxygenase-2 Inhibitors and Their Effects on Blood Pressure. *Arch Intern Med*.
3. Barden, J., et al., 2002. Single-dose rofecoxib for acute postoperative pain in adults: a quantitative systematic review. *BMC Anesthesiol*, **2**(1): p. 4.
4. Barden, J., et al., 2004. Single dose oral rofecoxib for postoperative pain. *Cochrane Database Syst Rev*, (1): p. CD004604.
5. Barden, J., et al., 2004. Relative efficacy of oral analgesics after third molar extraction. *Br Dent J*, **197**(7): p. 407-11; discussion 397.
6. Barden, J., et al., 2005. Single dose oral rofecoxib for postoperative pain. *Cochrane Database Syst Rev*, (1): p. CD004604.
7. Bassett, K., et al., 2002. Cyclooxygenase-2 inhibitors update: Journal articles fail to tell the whole story. *Can. Fam. Physician*, **48**: p. 1455-60.
8. Bjordal, J.M., et al., 2004. Non-steroidal anti-inflammatory drugs, including cyclo-oxygenase-2 inhibitors, in osteoarthritic knee pain: meta-analysis of randomised placebo controlled trials. *Bmj*, **329**(7478): p. 1317.
9. Chen, L.C., R.A. Elliott, and D.M. Ashcroft, 2004. Systematic review of the analgesic efficacy and tolerability of COX-2 inhibitors in post-operative pain control. *J Clin Pharm Ther*, **29**(3): p. 215-29.
10. Daniels, B. and B. Seidenberg, 1999. Cardiovascular safety profile of rofecoxib in controlled clinical trials. *Arthritis Rheum*, **42**(Suppl): p. S143.
11. Desjardins, P.J., et al., 2005. The time to onset and overall analgesic efficacy of rofecoxib 50 mg: a meta-analysis of 13 randomized clinical trials. *Clin J Pain*, **21**(3): p. 241-50.
12. Edwards, J.E., R.A. Moore, and H.J. McQuay, 2004. Individual patient meta-analysis of single-dose rofecoxib in postoperative pain. *BMC Anesthesiol*, **4**(1): p. 3.
13. Edwards, J.E., R.A. Moore, and H.J. McQuay, 2004. Rofecoxib for dysmenorrhoea: meta-analysis using individual patient data. *BMC Womens Health*, **4**(1): p. 5.
14. (FDA), US Food and Drug Administration (2001) Vioxx Medical Review Rockville, Maryland, Available from: (http://www.fda.gov) [Accessed on: 11th July 2006]
15. Gagnier, J.J., S. Chrubasik, and E. Manheimer, 2004. Harpgophytum procumbens for osteoarthritis and low back pain: a systematic review. *BMC Complement Altern Med*, **4**: p. 13.
16. Garner, S., et al., 2002. Rofecoxib for the treatment of rheumatoid arthritis. *Cochrane Database Syst Rev*, (3): p. CD003685.
17. Garner, S., et al., 2005a. Rofecoxib for osteoarthritis. *Cochrane Database Syst Rev*, (1): p. CD005115.
18. Garner, S., et al., 2005b. Rofecoxib for rheumatoid arthritis. *Cochrane Database Syst Rev*, (1): p. CD003685.
19. Geba, G., et al., 2003. Hypertension among osteoarthritis patients treated with rofecoxib, celecoxib, or acetaminophen: an analysis of the data from pooled vact studies. *American Journal of Hypertension*, **16**(5,part 2): p. 39A.

20. Gertz, B.J., et al., 2002. A comparison of adverse renovascular experiences among osteoarthritis patients treated with rofecoxib and comparator non-selective non-steroidal anti-inflammatory agents. *Curr Med Res Opin*, **18**(2): p. 82-91.
21. Goldstein, J.L., et al., 2005. Cyclooxygenase-2 specific inhibitors and upper gastrointestinal tolerability in patients with osteoarthritis receiving concomitant low dose aspirin: pooled analysis of 2 trials. *J Rheumatol*, **32**(1): p. 111-7.
22. Gomez Cerezo, J., et al., 2003. Outcome trials of COX-2 selective inhibitors: global safety evaluation does not promise benefits. *Eur J Clin Pharmacol*, **59**(2): p. 169-75.
23. Hooper, L., et al., 2004. The effectiveness of five strategies for the prevention of gastrointestinal toxicity induced by non-steroidal anti-inflammatory drugs: systematic review. *Bmj*, **329**(7472): p. 948.
24. Juni, P., et al., 2004. Risk of cardiovascular events and rofecoxib: cumulative meta-analysis. *Lancet*, **364**(9450): p. 2021-9.
25. Konstam, M.A., et al., 2001. Cardiovascular thrombotic events in controlled, clinical trials of rofecoxib. *Circulation*, **104**(19): p. 2280-8.
26. Langman, M.J., et al., 1999. Adverse upper gastrointestinal effects of rofecoxib compared with NSAIDs. *Jama*, **282**(20): p. 1929-33.
27. Lee, C., et al., 2004. A comparison of the efficacy and safety of nonsteroidal antiinflammatory agents versus acetaminophen in the treatment of osteoarthritis: a meta-analysis. *Arthritis Rheum*, **51**(5): p. 746-54.
28. Lee, C., et al., 2005. Need for common internal controls when assessing the relative efficacy of pharmacologic agents using a meta-analytic approach: case study of cyclooxygenase 2-selective inhibitors for the treatment of osteoarthritis. *Arthritis Rheum*, **53**(4): p. 510-8.
29. Mehlisch, D., et al., 2004. Efficacy of rofecoxib 50mg in patients with moderate or severe pain: A meta-analysis of 13 clinical trials. *The Journal of Pain*, **5**(3): p. S63.
30. Moore, R.A., J.E. Edwards, and H.J. McQuay, 2005. Acute pain: individual patient meta-analysis shows the impact of different ways of analysing and presenting results. *Pain*, **116**(3): p. 322-31.
31. Moore, R.A., et al., 2005. Tolerability and adverse events in clinical trials of celecoxib in osteoarthritis and rheumatoid arthritis: systematic review and meta-analysis of information from company clinical trial reports. *Arthritis Res Ther*, **7**(3): p. R644-65.
32. Morrison, B.W., et al., 2000. The optimal analgesic dose of rofecoxib: Overview of six randomized controlled trials. *J Am Dent Assoc*, **131**(12): p. 1729-37.
33. Mukherjee, D., S.E. Nissen, and E.J. Topol, 2001. Risk of cardiovascular events associated with selective COX-2 inhibitors. *Jama*, **286**(8): p. 954-9.
34. NICE.
35. Reicin, A.S., et al., 2002. Comparison of cardiovascular thrombotic events in patients with osteoarthritis treated with rofecoxib versus nonselective nonsteroidal anti-inflammatory drugs (ibuprofen, diclofenac, and nabumetone). *Am J Cardiol*, **89**(2): p. 204-9.
36. Romsing, J. and S. Moiniche, 2004. A systematic review of COX-2 inhibitors compared with traditional NSAIDs, or different COX-2 inhibitors for post-operative pain. *Acta Anaesthesiol Scand*, **48**(5): p. 525-46.

37. Romsing, J., et al., 2005. Reduction of opioid-related adverse events using opioid-sparing analgesia with COX-2 inhibitors lacks documentation: a systematic review. *Acta Anaesthesiol Scand*, **49**(2): p. 133-42.
38. Rostom, A., et al., 2004. Gastroduodenal ulcers associated with the use of non-steroidal anti-inflammatory drugs : a systematic review of preventive pharmacological interventions. *Ottawa: Canada Coordinating Office for Health Technology Assessment ; 2004. Technology overview no12*, **Technology overview no12**.
39. Rostom, A., L. Goldkind, and L. Laine, 2005. Nonsteroidal anti-inflammatory drugs and hepatic toxicity: a systematic review of randomized controlled trials in arthritis patients. *Clin Gastroenterol Hepatol*, **3**(5): p. 489-98.
40. Rostom, A., et al., 2005. Adverse gastrointestinal effects of COX-2 inhibitors for inflammatory diseases. *The Cochrane Database of Systematic Reviews*, **CD004519**(3).
41. Schnitzer, T.J., et al., 2005. Efficacy of rofecoxib, celecoxib, and acetaminophen in patients with osteoarthritis of the knee. A combined analysis of the VACT studies. *J Rheumatol*, **32**(6): p. 1093-105.
42. Straube, S., et al., 2005. Effect of preoperative Cox-II-selective NSAIDs (coxibs) on postoperative outcomes: a systematic review of randomized studies. *Acta Anaesthesiol Scand*, **49**(5): p. 601-13.
43. Towheed, T.E., 2005. Systematic review of therapies for osteoarthritis of the hand. *Osteoarthritis Cartilage*, **13**(6): p. 455-62.
44. Watson, D.J., et al., 2000. Gastrointestinal tolerability of the selective cyclooxygenase-2 (COX-2) inhibitor rofecoxib compared with nonselective COX-1 and COX-2 inhibitors in osteoarthritis. *Arch Intern Med*, **160**(19): p. 2998-3003.
45. Watson, D.J., et al., 2001. Gastrointestinal medications and procedures in osteoarthritis patients treated with rofecoxib compared with nonselective NSAIDs. *MedGenMed*, **3**(4): p. 6.
46. Watson, D.J., et al., 2004. The upper gastrointestinal safety of rofecoxib vs. NSAIDs: an updated combined analysis. *Curr Med Res Opin*, **20**(10): p. 1539-48.
47. West, P.M. and C. Fernandez, 2003. Safety of COX-2 inhibitors in asthma patients with aspirin hypersensitivity. *Ann Pharmacother*, **37**(10): p. 1497-501.

Additional File 3

1. Anon, 2003. [Meta-analysis on acute pain. Rofecoxib helped three quarters of patients]. *MMW Fortschr Med*, **145**(31-32): p. 57.
2. Barden, J., et al., 2002. Single-dose rofecoxib for acute postoperative pain in adults: a quantitative systematic review. *BMC Anesthesiol*, **2**(1): p. 4.
3. Barden, J., et al., 2004. Single dose oral rofecoxib for postoperative pain. *Cochrane Database Syst Rev*, (1): p. CD004604.
4. Barden, J., et al., 2004. Relative efficacy of oral analgesics after third molar extraction. *Br Dent J*, **197**(7): p. 407-11; discussion 397.
5. Barden, J., et al., 2005. Single dose oral rofecoxib for postoperative pain. *Cochrane Database Syst Rev*, (1): p. CD004604.
6. Chen, L.C., R.A. Elliott, and D.M. Ashcroft, 2004. Systematic review of the analgesic efficacy and tolerability of COX-2 inhibitors in post-operative pain control. *J Clin Pharm Ther*, **29**(3): p. 215-29.
7. Desjardins, P.J., et al., 2005. The time to onset and overall analgesic efficacy of rofecoxib 50 mg: a meta-analysis of 13 randomized clinical trials. *Clin J Pain*, **21**(3): p. 241-50.
8. Edwards, J.E., R.A. Moore, and H.J. McQuay, 2004a. Individual patient meta-analysis of single-dose rofecoxib in postoperative pain. *BMC Anesthesiol*, **4**(1): p. 3.
9. Edwards, J.E., R.A. Moore, and H.J. McQuay, 2004b. Rofecoxib for dysmenorrhoea: meta-analysis using individual patient data. *BMC Womens Health*, **4**(1): p. 5.
10. Mehlisch, D., et al., 2004. Efficacy of rofecoxib 50mg in patients with moderate or severe pain: A meta-analysis of 13 clinical trials. *The Journal of Pain*, **5**(3): p. S63.
11. Moore, R.A., J.E. Edwards, and H.J. McQuay, 2005b. Acute pain: individual patient meta-analysis shows the impact of different ways of analysing and presenting results. *Pain*, **116**(3): p. 322-31.
12. Morrison, B.W., et al., 2000. The optimal analgesic dose of rofecoxib: Overview of six randomized controlled trials. *J Am Dent Assoc*, **131**(12): p. 1729-37.
13. Romsing, J. and S. Moiniche, 2004. A systematic review of COX-2 inhibitors compared with traditional NSAIDs, or different COX-2 inhibitors for post-operative pain. *Acta Anaesthesiol Scand*, **48**(5): p. 525-46.
14. Romsing, J., et al., 2005. Reduction of opioid-related adverse events using opioid-sparing analgesia with COX-2 inhibitors lacks documentation: a systematic review. *Acta Anaesthesiol Scand*, **49**(2): p. 133-42.
15. Straube, S., et al., 2005. Effect of preoperative Cox-II-selective NSAIDs (coxibs) on postoperative outcomes: a systematic review of randomized studies. *Acta Anaesthesiol Scand*, **49**(5): p. 601-13.
16. Bjordal, J.M., et al., 2004. Non-steroidal anti-inflammatory drugs, including cyclo-oxygenase-2 inhibitors, in osteoarthritic knee pain: meta-analysis of randomised placebo controlled trials. *Bmj*, **329**(7478): p. 1317.
17. Lee, C., et al., 2005. Need for common internal controls when assessing the relative efficacy of pharmacologic agents using a meta-analytic approach: case

study of cyclooxygenase 2-selective inhibitors for the treatment of osteoarthritis. *Arthritis Rheum*, **53**(4): p. 510-8.

18. Gomez Cerezo, J., et al., 2003. Outcome trials of COX-2 selective inhibitors: global safety evaluation does not promise benefits. *Eur J Clin Pharmacol*, **59**(2): p. 169-75.
19. Hooper, L., et al., 2004. The effectiveness of five strategies for the prevention of gastrointestinal toxicity induced by non-steroidal anti-inflammatory drugs: systematic review. *Bmj*, **329**(7472): p. 948.
20. Langman, M.J., et al., 1999. Adverse upper gastrointestinal effects of rofecoxib compared with NSAIDs. *Jama*, **282**(20): p. 1929-33.
21. Rostom, A., et al., 2004. Gastroduodenal ulcers associated with the use of non-steroidal anti-inflammatory drugs : a systematic review of preventive pharmacological interventions. *Ottawa: Canada Coordinating Office for Health Technology Assessment ; 2004. Technology overview no12*, **Technology overview no12**.
22. Rostom, A., et al., 2005. Adverse gastrointestinal effects of COX-2 inhibitors for inflammatory diseases. *The Cochrane Database of Systematic Reviews*, **CD004519**(3).
23. Watson, D.J., et al., 2000. Gastrointestinal tolerability of the selective cyclooxygenase-2 (COX-2) inhibitor rofecoxib compared with nonselective COX-1 and COX-2 inhibitors in osteoarthritis. *Arch Intern Med*, **160**(19): p. 2998-3003.
24. Watson, D.J., et al., 2001. Gastrointestinal medications and procedures in osteoarthritis patients treated with rofecoxib compared with nonselective NSAIDs. *MedGenMed*, **3**(4): p. 6.
25. Watson, D.J., et al., 2004. The upper gastrointestinal safety of rofecoxib vs. NSAIDs: an updated combined analysis. *Curr Med Res Opin*, **20**(10): p. 1539-48.
26. Rostom, A., L. Goldkind, and L. Laine, 2005. Nonsteroidal anti-inflammatory drugs and hepatic toxicity: a systematic review of randomized controlled trials in arthritis patients. *Clin Gastroenterol Hepatol*, **3**(5): p. 489-98.
27. West, P.M. and C. Fernandez, 2003. Safety of COX-2 inhibitors in asthma patients with aspirin hypersensitivity. *Ann Pharmacother*, **37**(10): p. 1497-501.
28. Bassett, K., et al., 2002. Cyclooxygenase-2 inhibitors update: Journal articles fail to tell the whole story. *Can. Fam. Physician*, **48**: p. 1455-60.
29. Gagnier, J.J., S. Chrubasik, and E. Manheimer, 2004. Harpgophytum procumbens for osteoarthritis and low back pain: a systematic review. *BMC Complement Altern Med*, **4**: p. 13.
30. Lee, C., et al., 2004. A comparison of the efficacy and safety of nonsteroidal antiinflammatory agents versus acetaminophen in the treatment of osteoarthritis: a meta-analysis. *Arthritis Rheum*, **51**(5): p. 746-54.
31. Towheed, T.E., 2005. Systematic review of therapies for osteoarthritis of the hand. *Osteoarthritis Cartilage*, **13**(6): p. 455-62.
32. Garner, S., et al., 2002. Rofecoxib for the treatment of rheumatoid arthritis. *Cochrane Database Syst Rev*, (3): p. CD003685.