STRUCTURAL STUDIES ON ANTIFOLATE DRUGS

PATRICK KEVIN BRYANT

Doctor of Philosophy

THE UNIVERSITY OF ASTON IN BIRMINGHAM

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Summary

Single crystal X-ray structure determinations are reported for eleven compounds all of which are either biologically active or potentially biologically important. The compounds fall into two distinct classes:-

1. Substituted diaminopyrimides
2. Substituted diaminopyrimidinones

The first class of compounds were all selected on the basis of their common diaminopyrimide nucleus which has been demonstrated to be a vital requirement for antifolate activity. They may all be described as non-classical or small molecule lipophilic dihydrofolate reductase (DHFR) inhibitors, as opposed to the classical folate analogues, having the ability to cross the blood-brain barrier; enter cells via a rapid passive diffusion process, achieve high intracellular concentrations, thus making them an excellent choice in the search for chemotherapeutic agents with enhanced antitumour activity. The higher resolution acheived in small molecule crystallography enables the study of known enzyme substrate interactions in the solid state, providing geometrical and distance data not available from any other analytical techniques to date; supporting and enhancing data obtained in the lower resolution studies of protein crystallography. The biological importance of these compounds is discussed and an attempt is made to relate/ predict their pharmacological activity to observed structural features in the crystalline environment. Special attention is focussed on hydrogen bonding, conformational flexibility and hydrophobicity of substituents; each of which appear to make contributions to tight binding in the enzyme active site. Chapter 9 describes the use of data from the literature and the solid state modelling of an observed enzyme-substrate interaction in an attempt to define it more accurately in terms of its geometric flexibility.

Of the second class, one compound (ABPP) is reported; studied in two different crystal forms. In demonstrating both antiviral and high interferon inducing activity it is possible that this compound could be useful against cancer and also viral infections.

KEY WORDS: DHFR ANTIFOLATE CHEMOTHERAPY PYRIMIDINE
PYRIMIDINONE

To my Teachers

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Abbreviations

ASN	Asparagine
DHFR	Dihydrofolate reductase
H-bond	Hydrogen bond
ASP	Aspartate
GLU	Glutamate
NMF	N-methyl formamide
ABPP	2-amino-5-bromo-6-phenyl-4-pyrimidinine
DMF	Dimethyl formamide
DNA	Deoxyribonucleic acid
PABA	p-Aminobenzoic acid
NADPH	Nicotinamide adenine dinucleotide
dTMP	Deoxythymidylic acid monophosphate
dUMP	Deoxyuridylic acid monophosphate
FAH_4	Tetrahydrofolate
FAH_2	Dihydrofolate
TS	Thymidylate synthetase
NMR	Nuclear magnetic resonance
LCAO	Linear combination of atomic orbitals
QSAR	Quantitative structure activity relationship

CHAPTER 1

CHAPTER 1

1.1 **Introduction**

> 'Things whose textures have such mutual correspondence that cavities fit protuberances - the cavities of the first fit the protuberances of the second and vice versa form the closest possible unions.
>
> Titus Lucretius Carus 94-95 B.C.

The work of W.L. Bragg et al in developing the technique of X-ray crystallography has provided perhaps the most powerful analytical technique available to the physical sciences to date. The suggestion (1) was that crystalline solids were constituted by a three-dimensional array of atoms characterised by a regularly repeating subunit throughout the crystal. Diffraction theory applied to this model predicted that diffraction would be observed with X-rays whose wavelength (≈ 1Å) was of the same order of magnitude as the spacing (d) (2) between the scattering objects (atoms) in the array. Confirmation came when Friedrich and Knipping (3) obtained the first X-ray photograph showing diffraction by a single crystal. There then followed development of the now famous 'Bragg relationship'.

$$n\lambda = 2d\mathrm{Sin}\theta$$

where λ = incident wavelength

d = interplanar spacing

θ = angle of scattered beam.

A simple and elegant description of the phenomenon where the atoms of the crystal are considered as reflecting planes rather than point scatterers. Bragg went on to determine the internal arrangement, or structure, of the sodium chloride crystal (4). The success of this experiment fully established the technique and the possibilities of investigating structure at this level became obvious. The technique was, and still is in many cases, by no means trivial and prior to the advent of powerful computers required many months (sometimes years) of work to 'solve' a structure from its X-ray diffraction pattern.

Today advances in facilty of data collection and computing power have meant that single crystal X-ray crystallography is used almost routinely as a method of determining the three-dimensional structure and molecular dimensions of organic molecules. Using a modern four-circle diffracotmeter with automated data collection routines a dataset on a small organic molecule containing ~ 50 atoms in the asymmetric unit can be collected in around 3 days. Subsequent data reduction and the application of Direct Methods programs and least-squares refinement (see Appendix II) can result in a fully refined high resolution structure solution in a matter of weeks. (For a fuller description of the technique see Appendix I).

The observation that structure and function are intimately related and nowhere more so than in biologically functional molecules has led to much use of the technique on examining the structure of these and related compounds. Recent directions in this area of research have led to structure determination being divided into two broad catagories, both so-called small molecule crystallography and protein crystallography (this work concerns itself with the former). Protein crystallography concerns itself with medium to large structures e.g. carbohydrates, polypeptides, proteins or nucleic acids. The size and number of variables inherent in these structures together with problems of resolution and R factor (see Appendix II) has hindered progress in this area. Fortunately many biochemical reactions involve the interaction of a small molecule, or substrate, with an 'active site' formed by a large molecule (this is especially true in the case of enzymes). By structural studies on the substrate molecule much information can be deduced concerning the active site of the large molecule for which no structure, or only a low resolution structure is available.

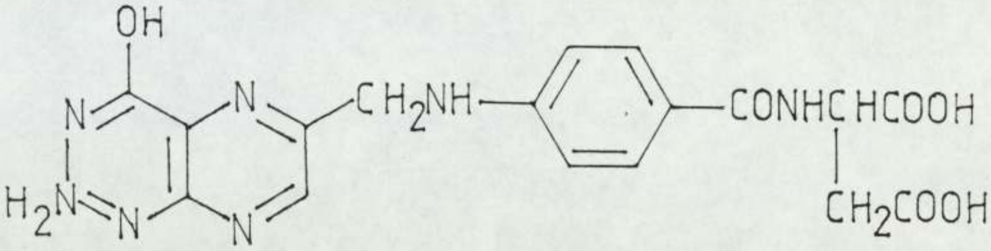
It is the purpose of this work to carry out structural studies on small molecule compounds with suggested inhibitory activity towards the enzyme dihydrofolate reductase (DHFR) to provide information which will assist in the design of improved (tightly bound and species specific) inhibitor molecules.

1.2 Chemotherapy

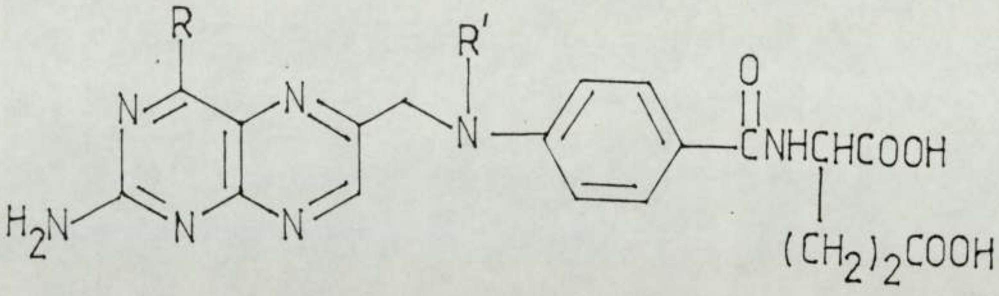
The history of cancer chemotherapy is firmly rooted in the past and there is evidence of the use of gold, silver and mercury preparations in the treatment of neoplastic disease from as early as the 14th century A.D. (5). The first non-mystical clinical use of chemotherapy appears as work by Lissauer (1865) (5) using Karsenite (Fowler's solution) in the treatment of leukeamia. Adair and Bagg (1931) (5) used sulphur mustard applied to squamous carcinoma and also by direct injection into solid human tumours. There followed the use of another alkylating agent, nitrogen mustard, (Gilman et al, 1942) (5) in clinical trials on patients with lymphosarcoma. The subsequent publication of this work in 1946 (6) heralded the beginning of modern chemotherapy, although therapy limiting effects such as bone marrow toxicity and development of drug resistance were observed. Even today one of the most commonly used chemotherapeutic agents methotrexate (MTX) (Fig. 1) (7) has a list of undesirable effects, which include: nausea, vomiting, ulceration and haemorrhage of mouth and bowel, kidney damage, bone marrow depression, fall in white blood cell and platelet counts and hair loss.

The notion, propounded by Woods (1940), of an 'antimetabolite' structually resembling the natural metabolite necessary for cellular function and interfering with its normal utilisation led to the idea of applying this principle to folate metabolism. In 1948 Sidney Farber et al (8) reported the use of aminopterin (Fig. 1) (9) a known antagonist of folic acid (10) in inhibiting the growth of Streptococcus Faecalis R. He arrived at this idea from his interpretation of counterproductive attempts to alleviate the symptoms of leukemia, observing an acceleration phenomenon in the leukemic process; as seen in the marrow and viscera of children with acute leukemia treated with injections of folic acid conjugates. Experience gained on observations of folic acid deficiency suggested that folic acid antagonists and inhibitors might be of value in the treatment of patients with acute leukemia. Farber initially used pteroyl aspartic acid (Fig. 1) and post mortem examinations of patients treated for a period of 6 days revealed a markedly hypoplastic bone marrow,

Figure 1.1 Folic acid and analogue inhibitors



I Pteroyl Aspartic Acid



II folic acid R = OH R' = H (as 4(3H)-one tautomer)

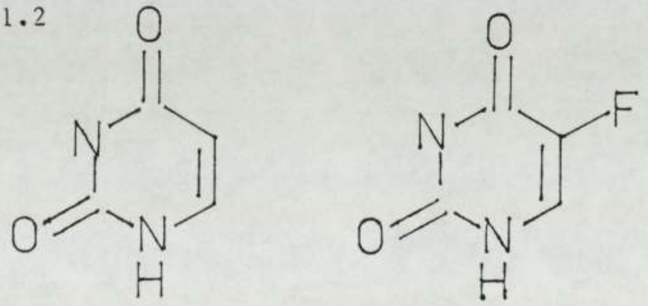
III aminopterin R = NH_2 R' = H

IV methotrexate R = NH_2 R' = CH_3

with a few immature cells, a change of this magnitude in such a short time period being hitherto unknown. Subsequent reseach using more powerful inhibitors including aminopterin showed temporary remissions in leukemic children although toxic side effects lowered the success rate considerably.

Other classes of antimetabolite have been studied, notably antipurines and antipyrimidines. Hitchings, (11) work on purine antagonists gave rise to 8-azaguanine and 2,6-diaminopurine, but these compounds proved not to be clinically useful. Elion et al (1952) (12) demonstrated the efficacy of 6-mercaptopurine (6-MP) against human leukemia and this compound along with recent active analogues 6-thioguanine and azathioprine are still used in the clinic today. Heidelberger (1960) (13), working on antipyrimidines, noted that tumours used more uracil than orotic acid (vice-versa for many normal tissues).

Fig. 1.2



uracil   5-fluorouracil

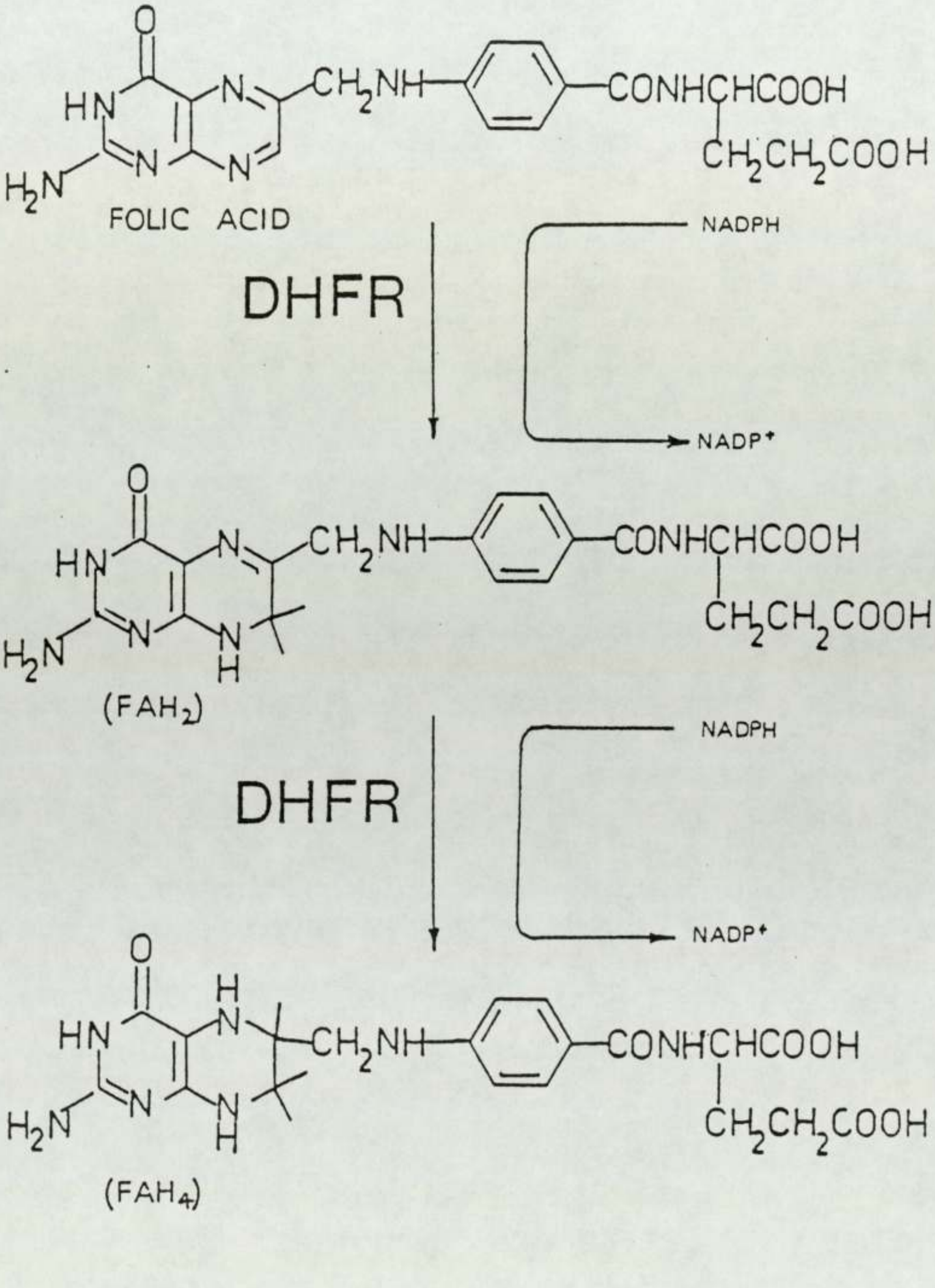
Therefore an antimetabolite resembling uracil may be preferentially incorporated by tumours (Fig. 1.2). Based on the conjecture that acetic acid is non-toxic but its fluorinated analogue is commonly used as rat poison it was thought that a fluorinated analogue might be active. This gave rise to the discovery of 5-fluorouracil which is a mainstay of anti-leukemic therapy today.

1.3 The Biological Importance of Folic Acid

Folic acid-vitamin Bc - (Fig. 1.1) is an essential requirement in the biochemistry of mammals and micro-organisms, its requirement in the human diet being in the order of 50 ug/day for adults. Symptoms of deficiency are anaemia with megalo blastic changes in the bone marrow. Certain micro-organisms, e.g. bacterial cells have the ability to synthesise folic acid from its basic subunits; a glutamoylpteridine and p-aminobenzoic acid PABA), and certain plasmodia and pathogenic bacteria synthesise folate-containing cofactors de novo (14) from the folic acid constituents. This metabolic difference between bacterial and mammalian cells may be exploited as the inhibition of folate metabolism will halt all growth in normal cells and abolish proliferation of abnormal cells such as are encountered in neoplastic diseases (cancers). In brief it presents an excellent target for cancer chemotherapy.

The importance of the role of folic acid is embodied in conversion of its reduced form FAH_4 (5,6,7,8 tetrahydrofolic acid) to a number of one-carbon unit carrying co-factors which are then involved in amino acid interconversions (Fig. 1.2) and purine and pyrimidine synthesis (Fig. 1.3), the purine and pyrimidine bases being essential precursors for DNA biosynthesis. Initially, folic acid is reduced in the cell enzymatically to FAH_4 (5,6,7,8 tetrahydrofolic acid) via 7,8 dihydrofolic acid

Figure 1.3 The Reduction of Folic Acid



by the enzyme dihydrofolate reductase (DHFR) (Fig. 1.1) (15), the reaction being driven by the conversion of NADPH to $NADP^+$ (16). FAH_4 then picks up a one-carbon group which may be a formyl, methyl or hydroxymethyl and is converted into the one-carbon unit carrying cofactors mentioned earlier. The one-carbon unit can then be transferred to other compounds which are utilised in various essential pathways, e.g. a one-carbon unit is transferred into C(2) and C(8) positions of the purine bases, the C(5) of deoxyuridylic acid (Fig. 1.3) and the methyl group of thymine.

In the cycle shown in Fig. 1.3 deoxyuridylic acid (dUMP) is converted to deoxythymidylic acid (dTMP) by the enzyme thymidylate synthetase (TS). During this cycle the continuous regeneration of FAH_4 by DHFR is required, and so-called 'thymineless' death of cells has been reported where DHFR has been irreversibly inhibited. Similarly the interconversion of glycine and serine is folate dependent (17).

It is clear that the pivotal role played by DHFR in the provision of reduced folate for interconversion of one-carbon units, principally the de novo biosynthesis of nucleic acids (10) makes it an excellent candidate for chemotherapy where control of rapidly dividing cells is important. It has been shown (Jukes and Broquist) (18) that indirect inhibition of the enzyme thymidylate synthetase, by depleting FAH_4-coenzymes is a primary mechanism by which antifolate drugs antagonise DNA synthesis.

1.4 The inhibition of Dihydrofolate Reductase

Although much effort had been directed towards antifolates it was not until a decade after the original concept that the

Figure 1.4 The conversion of dUMP to dTMP

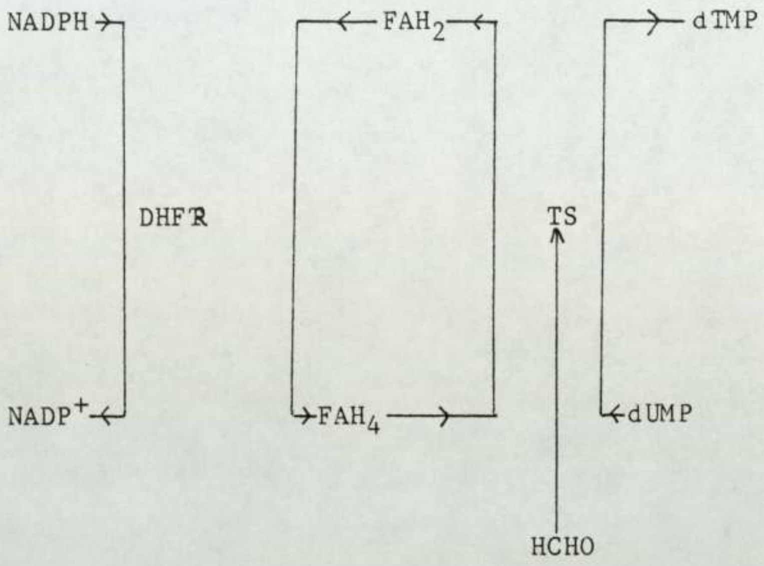


Figure 1.5 The conversion of glycine to L-serine

$$\text{5,10-methylene-FAH}_4 + \text{Gly} + H_2O \underset{\text{SHMT}}{\rightleftharpoons} \text{L-serine} + \text{FAH}_4$$

SHMT = serine - hydroxymethyl transferase

Blakely, 1954 (10).

target enzyme, DHFR, was identified (19). Once this fact was established it was possible to consider the mechanism of substrate binding at the active site of the enzyme and from a knowledge of this, attempt to tune the pharmacological properties of drug molecules to enhance binding and species specificity and minimise toxicity.

1.4.1 Dihydrofolate Reductase binding

Early theories sought to explain the enhanced binding of aminopterin in terms of the difference in hydrogen bonding potential between the 4-hydroxy and the 4-amino group. Zakrzewski (20) observed that the energy involved in promoting folic acid from the stable keto form [4-one, protonated at N(3)] to the 4-hydroxy form could explain some, but not all, of the difference in binding constants between folic acid and aminopterin. In his hydrogen bonding scheme N(1) acted as a site of hydrogen bond acceptance, although there was no thermodynamic evidence indicating the direction of the hydrogen bonding. LCAO molecular orbital calculations (21) have shown that the less direct effect of replacing 4-OH by 4-NH_2 is to shift the site of greatest basicity from N(8) to N(1). All crystallographic studies of DHFR inhibitors in protonated form have shown the site of protonation as N(1) [or its equivalent, N(3), in the case of s-triazine systems]. In summary, explanations of enhanced affinity of the folate inhibitors have focussed upon the modified pattern of hydrogen bond donors and acceptors (22) or the change in electron density in the heterocycle leading to increased basicity (23).

Information on the importance of hydrogen bonding power and basicity to DHFR inhibitors binding is available from crystal structure determinations by Matthews et al (24,25) of DHFR-methotrexate complexes one with DHFR from E. coli, the other from Lactobacillus casei which also included NADPH cofactor. Both show a number of important interactions among which is considerable hydrogen bonding between enzyme and inhibitor. Table 1.1 summarises the interactions between the drug MTX, and residues found in and around the active site for the E. coli structure.

In both structures the enzymes supply a carboxylate anion near the protonated N(1), demonstrating the importance of protonation at N(1) and thus basicity, a proton acceptor for the 2-amino group and another proton acceptor near the 4-substituent. In addition the E. coli but not the L. casei appears to furnish a proton donor in the vicinity of the 4-substituent. If the 4-amino group is capable of rehybridisation it is possible for it to act as both proton donor and acceptor thus forming two hydrogen bonds to the E. coli enzyme compared with just one for the carbonyl oxygen of the substrate (26). Acting purely as a proton donor it can form one hydrogen bond to the L. casei DHFR as compared with none for the carbonyl oxygen of the substrate. It has been suggested that the pteridine ring of bound folates may be turned over from its orientation observed in bound MTX (25); if so a ring nitrogen capable of accepting but not donating a proton could approach the carbonyl position, and the above reasoning would still hold (26).

A study of the known Asp-27 carboxylate anion to the

Table 1.1

INTERACTIONS INVOLVED IN BINDING MTX TO DHFR*

(1) Primary Hydrophobic Pocket

- (a) Van der Waals contacts between the pteridine ring and the side chains of Ile-5, Ala-7, Leu-28, Phe-31 and Ile-94.
- (b) π - π interaction between the fragment N(1)-C(2)-$N(2)H_2$-N(3) of the pteridine ring and the Ala-6 to Ala-7 peptide bond (separation distance = 3.5Å).
- (c) Hydrogen-bond donation from N(2) to the side-chain hydroxyl of Threonine - 113.
- (d) Hydrogen - bond donation from N(4) to the carboxyl oxygen of Ile-5.
- (e) Hydrogen-bond donation from N(1) to the carboxyl group of Asp-27.

(2) Secondary Hydrophobic Pocket

- (a) Van der Waals contact between the aromatic ring of the p-aminobenzoyl moiety and the side chains of Leu-28 Ile-50, Leu-54 and Ile-94.
- (b) Van der Waals contact between the β-carbon of Ser-49 and the methyl group at N(10)
- (c) Hydrogen-bond acceptance by α-carboxyl group from Arg-57.

*Matthews et al, 1977 (24).

protonated HN(1) - C(2)-NH_2 fragment of a number of 2,4-diamino pyrimidine compounds is reported in Chapter 9.

1.4.2 Hydrogen bonding of crystalline antifolate compounds

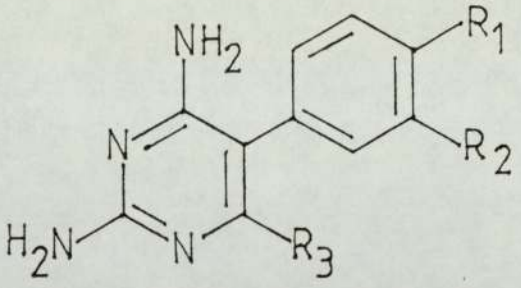
Hydrogen bonding studies of the pyrimidines and related compounds show that base-paired dimerisation about a centre or pseudo centre of symmetry in the crystal lattice using NH_2 as a proton donor, and a ring nitrogen as an acceptor is an almost universal phenomenon. Unsolvated neutral molecules (27) form two such base pairs each utilising only one of the amine hydrogens. However, when oxygen functions are available from solvent molecules in the lattice (28) N-H...O bonds are formed usually at the expense of one base pair.

In general hydrogen bonds formed by the 2 and 4 amino groups show that in the free base both tend to make the same type of interaction (base pair dimerisation); however when N(1) is protonated, the 2-amino group tends to use both protons to hydrogen bond to solvent oxygens or counter ions whilst the 4-amino group maintains a preference to form the base pair with one proton and form a hydrogen bond with a solvent oxygen or counter ion with the other (29). This effect is well exemplified by the 2,4-diaminopyrimidine structures, having the requisite moieties, in this work which are discussed later.

1.5 The rationale for non-classical inhibitors

Hitchings et al (30) reported that the cytotoxicity of a series of 2,4-diamino pyrimidines was mediated through inhibition of folate metabolism and in 1954 metoprine (DDMP) (II) (31) entered the clinic as a candidate antitumour agent; unfortunately the severe toxicity associated with this agent precluded any further evaluation. Diaminopyrimidines were subsequently found to inhibit DHFR and extensive QSAR (quantitative structure activity relationship) studies have resulted in the development of analogues with inherent species selectivity as antibacterial and antimalarial agents (32,33,34).

Figure 1.6



(I) pyrimethamine R_1 = Cl R_2 = H R_3 = Et

(II) metoprine R_1 = Cl R_2 = Cl R_3 = Me

(III) etoprine R_1 = Cl R_2 = Cl R_3 = Et

The use of pyrimethamine (I) in 1971 to produce a temporary remission in a patient with meningeal leukemia (35) stimulated a renewed interest in these agents. Hill et al (36) re-examined metoprine, a potent inhibitor of methotrexate resistant tumours and recognised the importance of concomitant folinic acid administration in alleviating toxicity (37). Problems of side-effects in metroprine and its closely related homologue etoprine (DDEP) (III), derived from action unrelated to the inhibition of folate metabolism has let to a renewed interest in the development of less toxic successors.

From the great amount of work carried out on antifolate compounds, two distinct divisions are clear:

(1) The folate analogues, or so-called classical inhibitors

(2) Small molecule, lipophilic, or non-classical inhibitors

This thesis is concerned with the latter of the two classes and a discussion of the characteristics of each class is appropriate here to demonstrate the rationale behind the pursuit of imporved antifolates of the second class.

1.5.1 Folate analogues or classical inhibitors

Methotrexate may be considered as the prototype classical antifolate. It is a potent 'pseudoirrevesible' inhibitor of DHFR from all biological sources and this has been attributed to its close structural resemblance to the natural substrate (38). As previously discussed the isosteric replacement of 4-hydroxy by an amino group on the pteridine ring increases basicity thus facilitating protonation and subsequent ionic interaction with a carboxylate residue on the enzyme (39,40). It has been argued that the predominant tautomer formed with a diaminopteridine would result in hydrogen bond formation; in fact crystallographic evidence reveals that a combination of both factors contribute to the stoichiometric binding of methotrexate to DHFR (41,42,43).

The polar nature of folate analogues (methotrexate log P=2.6 octanol/water) occurs as a consequence of the glutamate side-chain (44) and precludes any significant diffusion through cell membranes at normal concentrations. Some evidence has been obtained to suggest a limited diffusion process at extracellular drug concentrations achieved with high dose methotrexate regimes.

(45). Chemical modification of the glutamate residue has been investigated independently in an attempt to facilitate the diffusion process (46,47) and several lipophilic methotrexate esters have been synthesised and evaluated (48,49). Importantly, the hydrophilic character of methotrexate also renders the drug inaccessible to fatty tissues, including the brain and cerebrospinal fluid; such compartmentations may provide a pharmacological sanctuary in which malignant cells are spared cytotoxic drug concentrations (50).

Apart from the requirement for a specific biological transport process to gain access to cells, methotrexate also suffers from two major forms of resistance (Table 1.2); intrinsic resistance, where little or no response is observed from the beginning of therapy and acquired resistance, where an initially sensitive tumour reproliferates and is refractory to subsequent regimes with the same drug (51,52,53). The possible factors for each type of resistance are summarised in Table 1.2 and it becomes clear from this that the discovery of an effective anti-tumour drug based on DHFR inhibition is by no means a trivial exercise.

1.5.2 Non-classical, lipophilic or small-molecule inhibitors

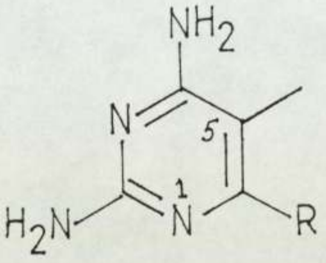
As a class, these molecules are characterised by the absence of a glutamate residue and consequently are invariably lipophilic in nature. Renewed interest in metroprine for the treatment of methotrexate resistant tumours has led to the synthesis and clinical assessment of numerous small-molecule DHFR inhibitors. These include diaminopryimidines (54,54) dihydrotriazines (40),

Table 1.2 Mechanisms of resistance to methotrexate

Intrinsic	**Acquired**
Kinetic cells in G_0 or plateau phase	Reduced capacity for MTX transport
Low capacity for MTX-poly glutamate formation	Overproduction of DHFR (gene amplification)
Poor transport of MTX	Reduced affinity of DHFR for MTX
High DHFR levels	Reduced capacity for polyglutamylation
Inadequate NADPH levels to facilitate MTX binding	
High intracellular folate levels	Opening of nucleoside salvage pathways
Rapid synthesis of DHFR	
Utilisation of salvage pathways	

quinazolines (56,57) and pyridopyrimidines. This work focuses on structural aspects of diaminopyrimidines (I).

I



R = small alkyl

The diaminopyrimidine nucleus

Favourable binding to the enzyme results from the introduction of a lipophilic group at position 5 of the pyrimidine ring. Ho et al (58) observed a correlation between the affinity of a series of 5 alkyldiamino pyrimidines for DHFR and their hydrophobicity, concluding that a bulky rigid group at this position confers maximal activity. A measure of lipophilicity (octanol/water partition coefficient) correlated well with cellular uptake and cytotoxicity (59). This is explained in terms of occupation of a hydrophobic pocket at the enzyme active site by the lipophilic 5-substituent (60,61).

Lipophilic diaminopyrimidines enter cells via a rapid passive diffusion process (62) and intracellular concentrations 30-100 fold that in the extracellular medium have been reported (59), thus overcoming resistance problems associated with active transport. The absence of a glutamate residue in metoprine precludes intracellular depletion, by metabolism to poly-y-glutamyl derivatives, thus obviating resistance mediated through a mutant folyl polyglutamyl transferase (63). Unlike methotrexate lipophilic agents may be absorbed percutaneously (64) thus substantiating the possibility of their use in treating topical neoplasias, e.g. psoriasis, where the use of methotrexate has led to hepatotoxicity (65).

The advantages of small-molecule inhibitors over classical substrate analogues are clear. Resistance to folate analogues will always emerge and be subject to carrier mechanisms responsible for transport into malignant cells. This is demonstrably so as analogues inherently more potent than methotrexate have been synthesised and evaluated with few advantages over the parent compound emerging. Given the advantages of lipophilic inhibitors and the demonstrated therapeutic potential of metoprine, unfortunately overshadowed by toxicity, the study of small molecule DHFR inhibitors is well justified.

1.6 Rational drug design

The essence of chemotherapy is the differential effect whereby the drug is more deterimental to the target organism/ tumour cell than to the host. Selective toxicity is therefore the key to the design of successful chemotherapeutic agents. Traditional methods for the discovery of chemotherapeutic agents can best be summed up in the phrase 'enlightened empiricism' (66). Success depends upon the availability of new compounds, efficient biological screening facilities, appropriate feed-back from clinicians to chemists, and hence to chemical modifications of the candidate drug. Surprisingly, the majority of the chemotherapeutic agents which are available today have arisen from this approach. The vast allocation of resources, men and materials involved in this particular strategy has given rise to much thought in an effort to rationalise the process.

In the clinical candidate discovery process one can distinguish two important steps:-

(i) The characterisation of a novel biologically active chemical class

(ii) The optimisation of these original compounds in terms of activity, specificity, bioavailability and toxicity.

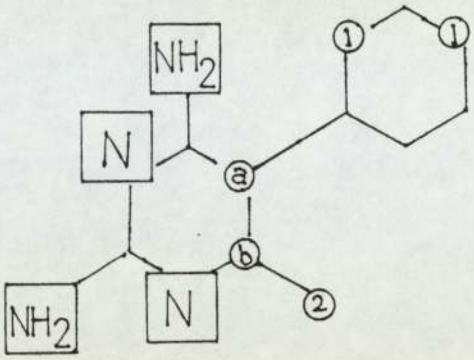
The first step remains a difficult problem and most recent work has been directed towards rational design of original putatively active structures based on an understanding of the principles underlying molecular recognition. Optimisation has been approached by QSAR studies. Hansch et al (67) characterised the observed activity of enzyme inhibitors in terms of a number of independent variables with empirically determined values for each functional group. The technique has been applied to the inhibition of DHFR by substituted pyrimidines (68). Prior to this Baker (69) had examined a variety of non-classical inhibitors for differences in activity depending on the source of the enzyme. A systematic survey of these results has elucidated some of the chemical and geometric requirements for inhibitory activity (70) (Fig. 1.6) upon which a rational design approach can be attempted. Crystallographic studies make a real contribution to this effort in providing data on:

(1) Protonation site

(2) Hydrogen bonding pattern

(3) Spatial distribution of the hydrophobic and polar

Figure 1.7



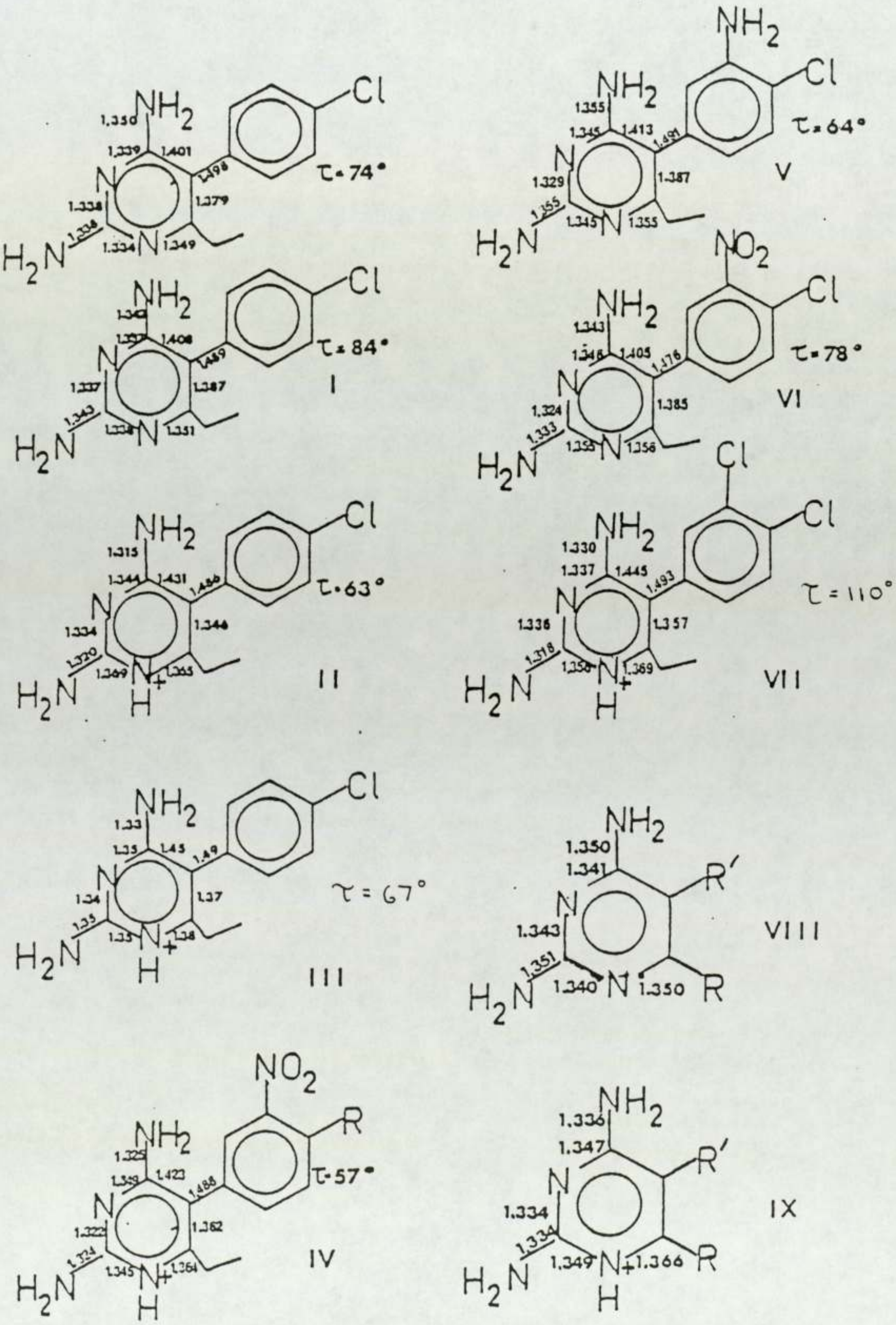
☐ primary (essential) binding sites

Either ⓐ or, more rarely, ⓑ can be nitrogen - otherwise carbon

① secondary binding sites - species differences

② steric limitations - species differences

Figure 1.8 Important bond distances [Å] for a number of derivatives of the antimalarial drug pyrimethamine.



Key to Figure 1.8

(I) Pyrimethamine (71) (2 independent molecules per asymmetric unit)

(II) Pyrimethamine hydrochloride (71)

(III) Pyrimethamine hydrobromide (72)

(IV) NPP Hydrochloride (73)

(V) m-aminopyrimethamine (74)

(VI) m-nitropyrimethamine (74)

(VII) DDMP Ethanesulphonate salt (75)

(VIII) Average values for the pyrimidin ring unprotonated (Schwalbe and Cody) (29)

(IX) Average values for the pyrimidine ring protonated at N(1) (Schwalbe and Cody) (29) components of the molecule in order to elucidate the shape of the active site and discover possible secondary sites of bonding, with particular interest in species differences.

(4) The observation of subtle changes in the geometry of the molecule due to chemical modifications and from this an evaluation of flexibility of the molecule around energy minima with respect to entry and subsequent binding at the active site.

Although today the emphasis is on molecular modelling studies using interactive, high resolution graphics devices and coordinates derived from protein crystallographic studies; small molecule crystallography is still a most powerful analytical technique. High resolution studies of small molecules are required to provide fine detail, especially in the active site of an enzyme, where protein crystallography, due to unresolved experimental considerations, can not. Although progress is being made in protein crystallography e.g. use of synchronotron radiation, area detectors, and continuing work on structure determination, the problems are great, even low resolution structures taking years to obtain and interpretation of function sometimes vague due to lack of resolution. In the foreseeable future small molecule crystallography will continue in its role of the premier structural analytical technique.

1.7 Current structural knowledge about phenylpyrimidine-based antifolates and objectives of the research described in this thesis

Because of the clinical importance of the antibacterial drug trimethoprim (VIII and IX, R = H, R' = $CH_2C_6H_2(OCH_3)_3$)

and the antimalarial drug pyrimethamine (I-III) a sizeable number of crystal structure determinations have been carried out on 2,4-diaminopyrimidine derivatives, and the geometry of this ring system has been reviewed by Schwalbe and Cody (29). Some of the most relevant structures are presented in Fig. 1.8. The studies of protonated DHFR inhibitors directly provide structural information about the form in which the inhibitor binds to the enzyme. However, many crystal structures of unprotonated DHFR inhibitors have been determined, either because this is the form in which the drugs are stored and administered or because of better availability of good-quality crystals. Comparison of data for unprotonated (VIII) and protonated (IX) diaminopyrimidines reveals several differences in bond distances. For the reasons discussed in Appendix section I.2 the increase in C(2)-N(1) and C(6)-N(1) bond distances upon protonation of this nitrogen atom is expected. The more electron-deficient character of the protonated ring elicits more electron donation by the amino groups via shorter exocyclic C-N bonds.

While much structural information is available for pyrimethamine-type antifolates, much more is needed. Interaction of the protonated drug with carboxylate anion on DHFR is thought to be an important factor in enzyme-inhibitor binding. A study of model carboxylate salts of pyrimethamine by Hill (78) showed a consistent pattern of hydrogen bonding, albeit with its utility for model-building compromised by problems with the crystallographic refinement that still needed resolution. Alteration of the relevant COO^- group of E. coli DHFR to $CONH_2$ by site-directed mutagenesis led to a surprisingly small 27-fold increase in the dissociation constant of MTX(79). This finding suggested the need for a model study of an antifolte drug complexed with an amide, in order to see if particularly favourable hydrogen-bonding interactions that compensate for the loss of the ion pair are possible in this case.

Steric hindrance that prevents coplanarity of the two rings of pyrimethamine ought to produce resolvable isomers. If one isomer can be isolated and its configuration determined crystallographically, it should become a useful probe of binding interactions. The hydrogen-bonded pyrimethamine molecule is inflexible except for twisting of the bond between rings and repositioning of the ethyl group. Although pyrimethamine can mimic the interactions of MTX with the primary hydrophobic pocket of DHFR outlined in Table 1.1, further binding interactions are unlikely. Replacement of the 4-chloro substituent on the benzene ring of pyrimethamine with a benzylamino or dibenzylamino group provides more conformational flexibility and creates the opportunity for an aromatic ring to reach either the secondary hydrophobic pocket or the cofactor binding site. Studies of the conformational preferences of such molecules therefore are important inputs for further design of such drugs.

Finally it is important to bear in mind that antifolate activity may be achievable by inhibiting folte-dependent enzymes other than DHFR. Therefore it may well have merit to study similar molecules that are unlikely to inhibit DHFR but may still have biological activity.

CHAPTER 2

CHAPTER 2

The Crystal Structure of 4[4-(2,4-diamino-6-ethyl pyrimidine-5-yl) phenyl]-1,1-dimethylpiperazinium iodide monohydrate (Primium Iodide).

2.1 Experimental

A crystalline sample of the compound was obtained (76) and from it a suitable crystal was selected. The quality of the specimen crystal was confirmed by examinining its birefringent properties with a polarising microscope. The single cyrstal was then mounted onto a glass filament using an epoxy adhesive, the filament having been secured to a goniometer head stud by the hot wax method.

2.2 Crystal Data

$C_{18}H_{27}N_6I.\ H_2O$, M = 470.375, F(000) = 480, Triclinic a = 9.977(3), b = 10.380(5), c = 11.798(3) Å,α = 74.06(3), β = 62.30(4), γ = 76.89(6)°, V = 1033, Å^3 (by least squares refinement on diffractometer angles for 25 atuomatically centred reflections, λ = 0.71069 Å), space group P$\bar{1}$, Z,= 2, Dx = 1.518 gcm^{-3} (method of flotation in solvent mixture). Off white needles from which a cube was taken. Crystal dimensions 0.20mm on a side , μ (Mo-Kα) = 15.55 cm^{-1}.

2.3 Data Collection and Processing

CAD4 diffractometer, W/2θ mode with W scan width - 1.0 + 0.35 tanθ , W scan speed 0.9-3.3 degmm^{-1}, graphite monochromated Mo-Kα radiation 4194 reflections measured

($2.0 \leqslant \theta \leqslant 29°$ ±h,±k,+l), 3968 unique (merging R= 0.0299, no correction for absorption), giving 2969 with $I > 3\sigma(I)$ number of refining parameters; 351. Linear and approximately isotropic crystal decay ca. 1% (for 3 standard reflections checked every 7200s X-ray exposure time), corrected during processing.

2.4 Structure Determination and Refinement

Data reduction was followed by direct methods phase determination (SHELX76) and electron density synthesis. All hydrogen atoms were located by difference Fourier analysis during least squares refinement of non-hydrogen atoms. Full matrix least squares refinement of all positional and thermal parameters, with all non-hydrogen atoms anisotropic, using the weighting scheme W $= 1{\cdot}02/[\sigma^2 (Fo) + 0.0008F^2]$ with $\sigma(Fo)$ from counting statistics gave satisfactory agreement analyses. Final R and R_w values are 0.0376 and 0.0451 $(\Delta/\sigma)_{max}$ in the final refinement cycle was 0.024 this for x/a of atom H11. Of the peaks $0.5e\text{Å}^{-3}$, the four largest peaks are all about 1A from I1 and may be ascribed to lone pair anisotropy around the anion; the fifth peak is 2Å from I1. After this density there was no feature in the map > 0.5 $e\text{Å}^{-3}$.

Programs, computers used and sources of atomic scattering factor data are given in Appendix II.

Table 2.1 Positional parameters (fractional coordinates * 10**4, hydrogens * 10**3) with estimated standard deviations in parentheses.

ATOM	x/a	y/b	z/c	
I(1)	33165(4)	20770(4)	-35991(3)	I
N(1)	1622(4)	1712(3)	3286(3)	
C(2)	3022(4)	1108(4)	2662(4)	
N(21)	3963(5)	0891(4)	3250(4)	
N(3)	3596(4)	0769(3)	1496(3)	
C(4)	2687(4)	1099(4)	0880(4)	
N(41)	3269(5)	0781(4)	-0306(4)	
C(5)	1198(4)	1762(4)	1428(4)	
C(6)	0725(4)	2012(4)	2654(4)	
C(61)	-0878(5)	2603(5)	3413(4)	
C(62)	-1842(6)	1505(7)	4417(6)	
C(1')	0217(4)	2181(4)	0706(4)	
C(2')	0467(5)	3303(6)	-0253(5)	
C(3')	-0492(6)	3823(6)	-0870(5)	
C(4')	-1745(4)	3214(4)	-0530(4)	
C(5')	-1981(6)	2044(5)	0422(6)	
C(6')	-1008(6)	1536(5)	1016(6)	
N(1")	-2820(4)	3799(4)	-1049(3)	
C(2")	-3513(6)	2854(5)	-1284(6)	
C(3")	-4858(6)	3562(6)	-1560(6)	
N(4")	-4452(4)	4751(3)	-2685(3)	
C(4"1)	-3418(7)	4284(7)	-3947(5)	
C(4"2)	-5871(6)	5484(6)	-2790(6)	
C(5")	-3730(6)	5657(5)	-2410(6)	
C(6")	-2377(6)	4934(5)	-2144(5)	
O(1)	0023(7)	2628(10)	5813(8)	
H(1)	462(6)	034(6)	305(5)	
H(2)	342(6)	095(5)	416(6)	
H(3)	278(5)	101(5)	-074(5)	
H(4)	403(6)	050(5)	-062(5)	
H(5)	-128(5)	319(5)	277(5)	
H(6)	-090(4)	331(4)	386(4)	
H(7)	-193(8)	088(8)	403(7)	
H(8)	-297(9)	182(7)	493(7)	
H(9)	-148(8)	095(7)	518(7)	
H(10)	130(6)	372(5)	-048(5)	
H(11)	-029(6)	457(5)	-147(5)	
H(12)	-291(5)	151(5)	077(4)	
H(13)	-116(5)	079(5)	153(4)	
H(14)	-267(6)	246(5)	-205(5)	
H(15)	-381(7)	221(7)	-055(7)	
H(16)	-533(6)	300(5)	-176(5)	
H(17)	-555(7)	387(6)	-085(6)	
H(18)	-335(7)	503(7)	-448(6)	
H(19)	-403(8)	379(7)	-404(7)	
H(20)	-246(6)	374(5)	-389(5)	
H(21)	-562(6)	616(6)	-343(6)	
H(22)	-638(6)	485(5)	-288(5)	
H(23)	-646(7)	576(6)	-196(6)	
H(24)	-342(6)	639(6)	-317(6)	
H(25)	-453(6)	606(6)	-165(6)	

Table 2.1 contd.

H(26)	-146(6)	462(5)	-294(5)	
H(27)	-213(6)	555(6)	-180(5)	
H(28)	05(1)	24(1)	51(1)	II
H(29)	040(9)	249(9)	614(8)	

I * 10**5
II * 10**2

Table 2.2 Anisotropic temperature factors, and isotropic temperature factors where shown; with standard deviations in parentheses.

ATOM	U11	U22	U33	U23	U13	U12
I(1)	.0440(2)	.0560(2)	.0449(2)	-.0131(1)	-.0243(1)	.0043(1)
N(1)	.029(2)	.034(2)	.028(2)	-.010(2)	-.011(1)	.004(1)
C(2)	.025(2)	.027(2)	.029(2)	-.006(2)	-.009(2)	.002(2)
N(21)	.033(2)	.045(2)	.039(2)	-.014(2)	-.020(2)	.010(2)
N(3)	.025(2)	.030(2)	.028(2)	-.008(1)	-.010(1)	.003(1)
C(4)	.028(2)	.024(2)	.030(2)	-.005(2)	-.012(2)	-.002(2)
N(41)	.029(2)	.047(2)	.033(2)	-.018(2)	-.015(2)	.010(2)
C(5)	.026(2)	.031(2)	.031(2)	-.008(2)	-.011(2)	.001(2)
C(6)	.025(2)	.031(2)	.030(2)	-.006(2)	-.012(2)	.003(2)
C(61)	.028(2)	.041(3)	.038(2)	-.015(2)	-.012(2)	.012(2)
C(62)	.033(3)	.063(4)	.066(4)	-.024(3)	.001(3)	-.006(3)
C(1′)	.028(2)	.034(2)	.032(2)	-.011(2)	-.014(2)	.005(2)
C(2′)	.031(2)	.077(4)	.058(3)	.012(3)	-.022(2)	-.020(3)
C(3′)	.040(3)	.069(4)	.055(3)	.028(3)	-.028(2)	-.023(3)
C(4′)	.028(2)	.031(2)	.037(2)	-.010(2)	-.017(2)	.004(2)
C(5′)	.064(3)	.035(3)	.078(4)	.007(3)	-.052(3)	-.016(2)
C(6′)	.070(4)	.028(3)	.073(4)	.007(3)	-.051(3)	-.012(2)
N(1")	.038(2)	.033(2)	.041(2)	-.003(2)	-.023(2)	-.002(2)
C(2")	.047(3)	.035(2)	.057(3)	.002(2)	-.033(3)	-.010(2)
C(3")	.039(3)	.050(3)	.054(3)	.003(3)	-.027(3)	-.013(2)
N(4")	.032(2)	.033(2)	.037(2)	-.012(2)	-.019(2)	.005(2)
C(4"1)	.052(3)	.068(4)	.043(3)	-.023(3)	-.023(3)	.011(3)
C(4"2)	.042(3)	.054(3)	.051(3)	-.011(3)	-.029(3)	.006(3)
C(5")	.059(3)	.028(2)	.063(3)	-.001(2)	-.043(3)	-.004(2)
C(6")	.049(3)	.036(3)	.060(3)	-.001(2)	-.038(3)	-.009(2)
O(1)	.078(4)	.27(1)	.138(6)	-.146(7)	-.080(4)	.095(5)

	U ISO
H(1)	.05(2)
H(2)	.05(1)
H(3)	.03(1)
H(4)	.04(1)
H(5)	.04(1)
H(6)	.014(9)
H(7)	.10(3)
H(8)	.10(2)
H(9)	.09(2)
H(10)	.05(1)
H(11)	.05(1)
H(12)	.04(1)
H(13)	.04(1)
H(14)	.04(1)
H(15)	.08(2)
H(16)	.05(1)
H(17)	.06(2)
H(18)	.07(2)
H(19)	.08(2)
H(20)	.05(1)
H(21)	.06(2)
H(22)	.04(1)
H(23)	.06(2)

Table 2.2 contd.

	U ISO
H(24)	.07(2)
H(25)	.06(2)
H(26)	.04(1)
H(27)	.05(1)
H(28)	.19(5)
H(29)	.10(3)

Table 2.3 Bond Lengths(A) with standard deviations

N(1) - C(2)	1.337(6)	C(5') -C(6')	1.382(9)
N(1) - C(6)	1.349(6)	C(5') -H(12)	1.04(5)
C(2) -N(21)	1.355(6)	C(6') -H(13)	0.84(5)
C(2) - N(3)	1.337(6)	N(1") -C(2")	1.467(7)
N(21) - H(1)	0.76(6)	N(1") -C(6")	1.457(7)
N(21) - H(2)	0.97(6)	C(2") -C(3")	1.508(9)
N(3) - C(4)	1.343(6)	C(2") -H(14)	1.02(6)
C(4) -N(41)	1.347(6)	C(2") -H(15)	0.90(7)
C(4) - C(5)	1.409(6)	C(3") -N(4")	1.509(7)
N(41) - H(3)	0.81(5)	C(3") -H(16)	0.96(6)
N(41) - H(4)	0.71(6)	C(3") -H(17)	0.88(7)
C(5) - C(6)	1.378(6)	N(4") -C(4"1)	1.503(8)
C(5) -C(1')	1.500(6)	N(4") -C(4"2)	1.490(7)
C(6) -C(61)	1.504(7)	N(4") -C(5")	1.484(7)
C(61) -C(62)	1.524(8)	C(4"1)-H(18)	0.85(7)
C(61) - H(5)	1.02(5)	C(4"1)-H(19)	0.94(8)
C(61) - H(6)	1.00(4)	C(4"1)-H(20)	1.01(6)
C(62) - H(7)	0.93(8)	C(4"2)-H(21)	0.85(6)
C(62) - H(8)	1.02(8)	C(4"2)-H(22)	0.98(5)
C(62) - H(9)	1.09(8)	C(4"2)-H(23)	0.96(7)
C(1') -C(2')	1.358(7)	C(5") -C(6")	1.513(8)
C(1') -C(6')	1.377(8)	C(5") -H(24)	0.98(7)
C(2') -C(3')	1.391(8)	C(5") -H(25)	1.00(6)
C(2') -H(10)	0.91(6)	C(6") -H(26)	1.03(5)
C(3') -C(4')	1.370(7)	C(6") -H(27)	0.97(6)
C(3') -H(11)	0.88(6)	O(1) -H(28)	0.81(14)
C(4') -C(5')	1.388(8)	O(1) -H(29)	0.62(9)
C(4') -N(1")	1.416(6)		

Table 2.4 Angles(degrees) with standard deviations

C(2) - N(1) - C(6)	115.7(4)	C(1') -C(6') -H(13)	121.4(36)
N(1) - C(2) -N(21)	115.9(4)	C(5') -C(6') -H(13)	116.9(36)
N(1) - C(2) - N(3)	126.4(4)	C(4') -N(1") -C(2")	116.1(4)
N(21) - C(2) - N(3)	117.6(4)	C(4') -N(1") -C(6")	116.2(4)
C(2) -N(21) - H(1)	115.1(46)	C(2") -N(1") -C(6")	111.1(4)
C(2) -N(21) - H(2)	112.4(36)	N(1") -C(2") -C(3")	110.9(5)
H(1) -N(21) - H(2)	119.1(58)	N(1") -C(2") -H(14)	105.4(31)
C(2) - N(3) - C(4)	116.6(4)	N(1") -C(2") -H(15)	106.7(47)
N(3) - C(4) -N(41)	116.9(4)	C(3") -C(2") -H(14)	112.5(31)
N(3) - C(4) - C(5)	122.2(4)	C(3") -C(2") -H(15)	110.4(47)
N(41) - C(4) - C(5)	120.9(4)	H(14) -C(2") -H(15)	110.7(57)

C(4) -N(41) - H(3)	120.3(37)	C(2") -C(3") -N(4")	112.6(5)
C(4) -N(41) - H(4)	122.8(46)	C(2") -C(3") -H(16)	114.6(35)
H(3) -N(41) - H(4)	116.3(58)	C(2") -C(3") -H(17)	107.5(43)
C(4) - C(5) - C(6)	115.4(4)	N(4") -C(3") -H(16)	106.1(35)
C(4) - C(5) -C(1′)	121.8(4)	N(4") -C(3") -H(17)	107.9(43)
C(6) - C(5) -C(1′)	122.8(4)	H(16) -C(3") -H(17)	107.8(55)
N(1) - C(6) - C(5)	123.6(4)	C(3") -N(4") -C(4"1)	110.5(4)
N(1) - C(6) -C(61)	114.8(4)	C(3") -N(4") -C(4"2)	109.1(4)
C(5) - C(6) -C(61)	121.6(4)	C(3") -N(4") -C(5")	108.1(4)
C(6) -C(61) -C(62)	110.9(4)	C(4"1)-N(4") -C(4"2)	107.8(4)
C(6) -C(61) - H(5)	108.3(30)	C(4"1)-N(4") -C(5")	111.7(4)
C(6) -C(61) - H(6)	111.5(23)	C(4"2)-N(4") -C(5")	109.6(4)
C(62) -C(61) - H(5)	115.9(30)	N(4") -C(4"1)-H(18)	100.3(48)
C(62) -C(61) - H(6)	109.9(23)	N(4") -C(4"1)-H(19)	103.3(46)
H(5) -C(61) - H(6)	99.8(38)	N(4") -C(4"1)-H(20)	109.4(32)
C(61) -C(62) - H(7)	112.4(52)	H(18) -C(4"1)-H(19)	107.4(66)
C(61) -C(62) - H(8)	115.7(47)	H(18) -C(4"1)-H(20)	119.6(58)
C(61) -C(62) - H(9)	115.9(42)	H(19) -C(4"1)-H(20)	114.8(56)
H(7) -C(62) - H(8)	100.2(70)	N(4") -C(4"2)-H(21)	107.7(44)
H(7) -C(62) - H(9)	107.8(66)	N(4") -C(4"2)-H(22)	109.1(32)
H(8) -C(62) - H(9)	103.3(63)	N(4") -C(4"2)-H(23)	102.7(40)
C(5) -C(1′) -C(2′)	119.4(4)	H(21) -C(4"2)-H(22)	114.0(54)
C(5) -C(1′) -C(6′)	123.5(4)	H(21) -C(4"2)-H(23)	112.6(59)
C(2′) -C(1′) -C(6′)	117.0(5)	H(22) -C(4"2)-H(23)	110.1(51)
C(1′) -C(2′) -C(3′)	122.3(5)	N(4") -C(5") -C(6")	112.4(5)
C(1′) -C(2′) -H(10)	117.3(37)	N(4") -C(5") -H(24)	106.4(39)
C(3′) -C(2′) -H(10)	120.4(37)	N(4") -C(5") -H(25)	107.7(37)
C(2′) -C(3′) -C(4′)	120.9(5)	C(6") -C(5") -H(24)	110.9(39)
C(2′) -C(3′) -H(11)	118.5(38)	C(6") -C(5") -H(25)	111.1(37)
C(4′) -C(3′) -H(11)	120.6(38)	H(24) -C(5") -H(25)	108.0(53)
C(3′) -C(4′) -C(5′)	117.1(5)	N(1") -C(6") -C(5")	110.9(5)
C(3′) -C(4′) -N(1")	121.8(4)	N(1") -C(6") -H(26)	110.1(30)
C(5′) -C(4′) -N(1")	121.0(4)	N(1") -C(6") -H(27)	102.5(35)
C(4′) -C(5′) -C(6′)	121.2(5)	C(5") -C(6") -H(26)	114.3(30)
C(4′) -C(5′) -H(12)	123.5(28)	C(5") -C(6") -H(27)	105.6(35)
C(6′) -C(5′) -H(12)	115.3(28)	H(26) -C(6") -H(27)	112.9(47)
C(1′) -C(6′) -C(5′)	121.5(5)	H(28) - O(1) -H(29)	115.7(**)

Table 2.5 Torsion angles(degrees) with standard deviations

C(6) - N(1) - C(2) -N(21)	175.5(4)	C(2′) -C(3′) -C(4′) -C(5′)	-1.8(8)
C(6) - N(1) - C(2) - N(3)	-0.7(6)	C(2′) -C(3′) -C(4′) -N(1")	173.5(5)
C(2) - N(1) - C(6) - C(5)	-1.8(6)	H(11) -C(3′) -C(4′) -C(5′)	-180.0(44)
C(2) - N(1) - C(6) -C(61)	176.0(4)	H(11) -C(3′) -C(4′) -N(1")	-4.6(45)
N(1) - C(2) -N(21) - H(1)	158.3(51)	C(3′) -C(4′) -C(5′) -C(6′)	1.3(8)

N(1) - C(2) -N(21) - H(2)	17.5(39)	C(3') -C(4') -C(5') -H(12)	178.5(33)
N(3) - C(2) -N(21) - H(1)	-25.2(51)	N(1") -C(4') -C(5') -C(6')	-174.1(5)
N(3) - C(2) -N(21) - H(2)	-165.9(38)	N(1") -C(4') -C(5') -H(12)	3.2(34)
N(1) - C(2) - N(3) - C(4)	1.8(6)	C(3') -C(4') -N(1") -C(2")	145.0(5)
N(21) - C(2) - N(3) - C(4)	-174.4(4)	C(3') -C(4') -N(1") -C(6")	11.6(7)
C(2) - N(3) - C(4) -N(41)	178.9(4)	C(5') -C(4') -N(1") -C(2")	-39.8(6)
C(2) - N(3) - C(4) - C(5)	-0.4(6)	C(5') -C(4') -N(1") -C(6")	-173.3(5)
N(3) - C(4) -N(41) - H(3)	-175.3(42)	C(4') -C(5') -C(6') -C(1')	1.2(9)
N(3) - C(4) -N(41) - H(4)	-4.3(55)	C(4') -C(5') -C(6') -H(13)	-174.5(40)
C(5) - C(4) -N(41) - H(3)	3.9(43)	H(12) -C(5') -C(6') -C(1')	-176.3(31)
C(5) - C(4) -N(41) - H(4)	174.9(54)	H(12) -C(5') -C(6') -H(13)	8.0(51)
N(3) - C(4) - C(5) - C(6)	-1.8(6)	C(4') -N(1") -C(2") -C(3")	168.0(4)
N(3) - C(4) - C(5) -C(1')	177.2(4)	C(4') -N(1") -C(2") -H(14)	-70.0(33)
N(41) - C(4) - C(5) - C(6)	179.0(4)	C(4') -N(1") -C(2") -H(15)	47.8(50)
N(41) - C(4) - C(5) -C(1')	-2.0(7)	C(6") -N(1") -C(2") -C(3")	-56.2(6)
C(4) - C(5) - C(6) - N(1)	3.0(6)	C(6") -N(1") -C(2") -H(14)	65.8(33)
C(4) - C(5) - C(6) -C(61)	-174.7(4)	C(6") -N(1") -C(2") -H(15)	-176.5(49)
C(1') - C(5) - C(6) - N(1)	-176.1(4)	C(4') -N(1") -C(6") -C(5")	-167.4(4)
C(1') - C(5) - C(6) -C(61)	6.3(7)	C(4') -N(1") -C(6") -H(26)	65.2(33)
C(4) - C(5) -C(1') -C(2')	-77.8(6)	C(4') -N(1") -C(6") -H(27)	-55.1(36)
C(4) - C(5) -C(1') -C(6')	106.6(6)	C(2") -N(1") -C(6") -C(5")	57.0(6)
C(6) - C(5) -C(1') -C(2')	101.2(6)	C(2") -N(1") -C(6") -H(26)	-70.4(33)
C(6) - C(5) -C(1') -C(6')	-74.4(6)	C(2") -N(1") -C(6") -H(27)	169.2(36)
N(1) - C(6) -C(61) -C(62)	-78.6(5)	N(1") -C(2") -C(3") -N(4")	55.8(6)
N(1) - C(6) -C(61) - H(5)	153.1(32)	N(1") -C(2") -C(3") -H(16)	177.2(39)
N(1) - C(6) -C(61) - H(6)	44.3(25)	N(1") -C(2") -C(3") -H(17)	-62.9(45)
C(5) - C(6) -C(61) -C(62)	99.3(5)	H(14) -C(2") -C(3") -N(4")	-62.0(34)
C(5) - C(6) -C(61) - H(5)	-29.0(32)	H(14) -C(2") -C(3") -H(16)	59.5(52)
C(5) - C(6) -C(61) - H(6)	-137.9(25)	H(14) -C(2") -C(3") -H(17)	179.3(56)
C(6) -C(61) -C(62) - H(7)	-61.8(56)	H(15) -C(2") -C(3") -N(4")	173.8(50)
C(6) -C(61) -C(62) - H(8)	-176.0(52)	H(15) -C(2") -C(3") -H(16)	-64.8(64)
C(6) -C(61) -C(62) - H(9)	62.8(46)	H(15) -C(2") -C(3") -H(17)	55.0(67)
H(5) -C(61) -C(62) - H(7)	62.3(65)	C(2") -C(3") -N(4") -C(4"1)	68.2(6)
H(5) -C(61) -C(62) - H(8)	-51.9(62)	C(2") -C(3") -N(4") -C(4"2)	-173.4(5)
H(5) -C(61) -C(62) - H(9)	-173.1(57)	C(2") -C(3") -N(4") -C(5")	-54.3(6)
H(6) -C(61) -C(62) - H(7)	174.5(61)	H(16) -C(3") -N(4") -C(4"1)	-58.0(37)
H(6) -C(61) -C(62) - H(8)	60.3(58)	H(16) -C(3") -N(4") -C(4"2)	60.4(37)
H(6) -C(61) -C(62) - H(9)	-60.9(52)	H(16) -C(3") -N(4") -C(5")	179.5(37)
C(5) -C(1') -C(2') -C(3')	-173.4(5)	H(17) -C(3") -N(4") -C(4"1)	-173.3(45)
C(5) -C(1') -C(2') -H(10)	6.1(42)	H(17) -C(3") -N(4") -C(4"2)	-54.9(45)
C(6') -C(1') -C(2') -C(3')	2.5(8)	H(17) -C(3") -N(4") -C(5")	64.2(45)
C(6') -C(1') -C(2') -H(10)	-178.0(42)	C(3") -N(4") -C(4"1)-H(18)	175.3(49)
C(5) -C(1') -C(6') -C(5')	172.7(5)	C(3") -N(4") -C(4"1)-H(19)	64.5(47)
C(5) -C(1') -C(6') -H(13)	-11.8(43)	C(3") -N(4") -C(4"1)-H(20)	-58.2(34)
C(2') -C(1') -C(6') -C(5')	-3.0(8)	C(4"2)-N(4") -C(4"1)-H(18)	56.1(49)
C(2') -C(1') -C(6') -H(13)	172.5(42)	C(4"2)-N(4") -C(4"1)-H(19)	-54.6(47)
C(1') -C(2') -C(3') -C(4')	0.0(9)	C(4"2)-N(4") -C(4"1)-H(20)	-177.3(34)
C(1') -C(2') -C(3') -H(11)	178.1(44)	C(5") -N(4") -C(4"1)-H(18)	-64.3(49)
H(10) -C(2') -C(3') -C(4')	-179.5(43)	C(5") -N(4") -C(4"1)-H(19)	-175.1(47)

H(10) -C(2') -C(3') -H(11)	-1.4(62)
C(3") -N(4") -C(4"2)-H(21)	179.6(46)
C(3") -N(4") -C(4"2)-H(22)	-56.3(34)
C(3") -N(4") -C(4"2)-H(23)	60.5(41)
C(4"1)-N(4") -C(4"2)-H(21)	-60.3(46)
C(4"1)-N(4") -C(4"2)-H(22)	63.8(34)
C(4"1)-N(4") -C(4"2)-H(23)	-179.4(41)
C(5") -N(4") -C(4"2)-H(21)	61.5(46)
C(5") -N(4") -C(4"2)-H(22)	-174.4(34)
C(5") -N(4") -C(4"2)-H(23)	-57.6(41)
C(3") -N(4") -C(5") -C(6")	55.0(6)
C(3") -N(4") -C(5") -H(24)	176.6(40)
C(3") -N(4") -C(5") -H(25)	-67.8(38)
C(4"1)-N(4") -C(5") -C(6")	-66.8(6)
C(4"1)-N(4") -C(5") -H(24)	54.8(41)
C(5") -N(4") -C(4"1)-H(20)	62.2(34)
C(4"1)-N(4") -C(5") -H(25)	170.4(38)
C(4"2)-N(4") -C(5") -C(6")	173.7(5)
C(4"2)-N(4") -C(5") -H(24)	-64.7(41)
C(4"2)-N(4") -C(5") -H(25)	50.9(38)
N(4") -C(5") -C(6") -N(1")	-57.8(6)
N(4") -C(5") -C(6") -H(26)	67.3(34)
N(4") -C(5") -C(6") -H(27)	-168.1(37)
H(24) -C(5") -C(6") -N(1")	-176.8(41)
H(24) -C(5") -C(6") -H(26)	-51.8(53)
H(24) -C(5") -C(6") -H(27)	72.9(55)
H(25) -C(5") -C(6") -N(1")	63.0(39)
H(25) -C(5") -C(6") -H(26)	-171.9(51)
H(25) -C(5") -C(6") -H(27)	-47.3(54)

Figure 2.1 Primium Iodide. Molecular plot and number scheme

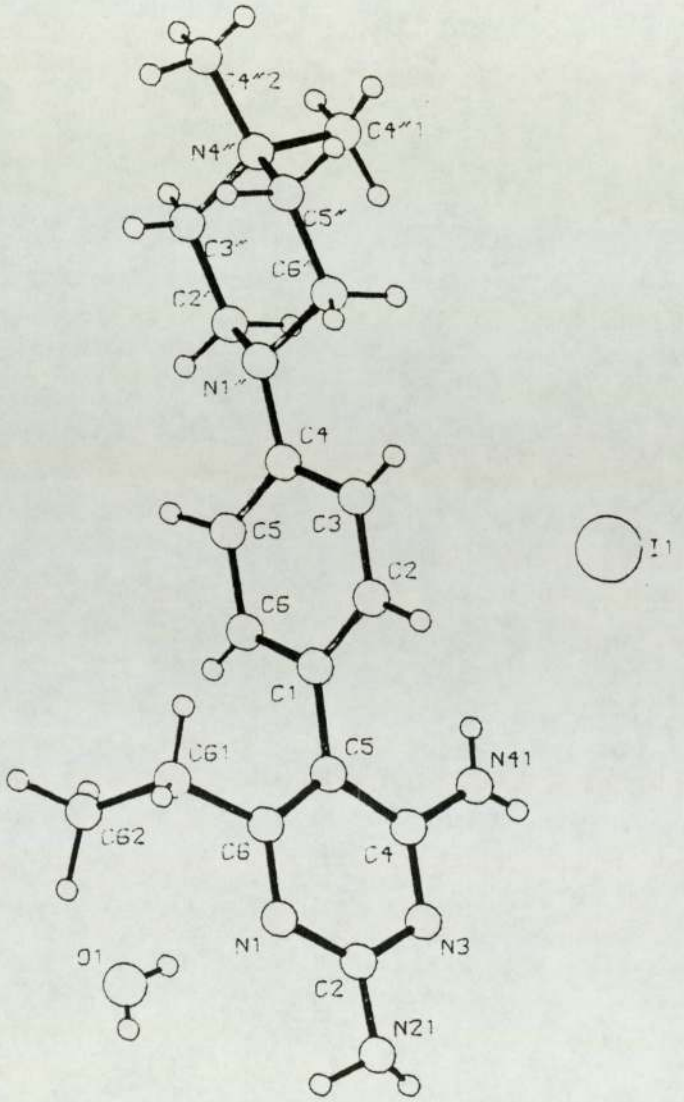


Figure 2.2 Primium Iodide. Unit cell packing diagram

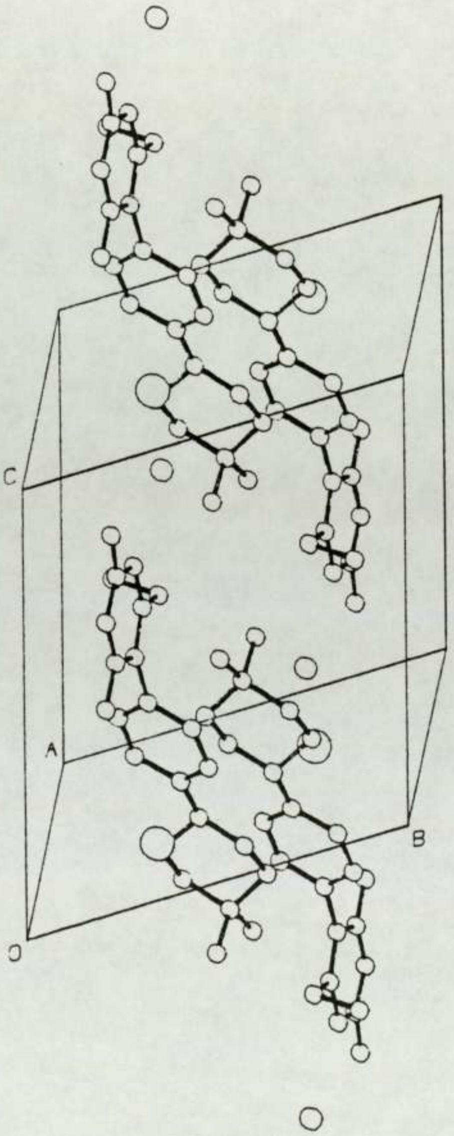


Table 2.6 Hydrogen bond contact distances

Hydrogen Bond	Donor-Acceptor Distance (Å)
$O(1) - H(01)_I --- N(1)_I$	2.964
$N(41) - H(4)_I --- N(3)_{II}$	3.036
$O(1) - H(02)_I --- I(1)_{III}$	3.557
$N(21) - H(1)_I --- I(1)_{II}$	3.685
$N(41) - H(3)_I --- I(1)_I$	3.727

The subscripts I, II and III refer to the equivalent positions X, Y, Z; 1-X, -Y, -Z; X, Y, 1 + Z.

2.5 Results and Discussion

The crystal structure of 4[4-(2,4-diamino-6-ethylpyrimidine-5-yl) phenyl]-1,1-dimethylpiperazinium iodide. H_2O (primium iodide), as determined crystallographically, is depicted in Figure 2.1 together with the numbering scheme used; positional parameters for all atoms are given in Table 2.1, with anisotropic thermal parameters for non-hydrogens and isotropic tempterature factors for hydrogens in Table 2.2.

The site of methylation is confirmed with the methyl disubstituted quaternary ammonium nitrogen of the cation at N(4"). Bond distances [Å] and interatomic angles [°] are given in Tables 2.3 and 2.4. A comparison of bond lengths for the pyrimidine moiety and its exocyclic amino groups with average values for the unprotonated pyrimidine ring (29) (Figure 1.7 viii) shows close agreement with only C(2) - N(3) differing by more than 0.01 A from mean values.

The classic base pair dimerisation is observed in this case with hydrogen bonding in the crystal (table 2.6) between the 4-amino group and N(3) of centrosymmetrically related molecules. Hydrogen bonds are observed involving both hydrogen atoms of the water O(1) - H(01) --- N(1) [2.964A] and O(1) - H(02) --- I [3.557 Å], the latter being considerably longer than the normal hydrogen bond length of approximately 3 Å; however Jellinek (77) reports a length of 3.57 Å for O-H --- I in the structure of muscarine iodide. The remaining amino hydrogen atoms participate in N - H --- I hydrogen bonds which, as expected from the larger radius and lower electronegativity of nitrogen, are 0.1 - 0.2 Å longer than the O - H --- I hydrogen bond.

The torsion angle around the bridge bond between pyrimidine and phenyl rings is -75.0°, which is not unusual for compounds of this type (see Figure 7.1). It is likely that the deviation from coplanarity results from a minimisations of Van der Waals interaction with the 6-ethyl side chain, which itself is rotated -78.2° away from the plane of the pyrimidine ring.

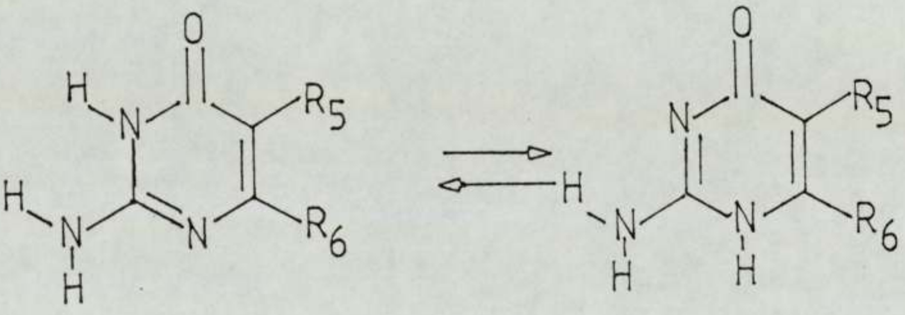
CHAPTER 3

CHAPTER 3

The Crystal Structures of two solvates of the Putative Anti-tumour Drug 2-amino-5-bromo-6-phenyl-4-(1/3 H)-pyrimidinone (ABPP)

3.1 Introduction

The title compound has demonstrated antiviral activity (80). Unusually for such a small molecule it is a highly active interferon inducing agent (81), and this property could make it useful against cancer as well as viral infections. In principle it is capable of existing either as the 3H tautomer or as the 1H.



The former resembles guanine and could form a base-pair with cytosine, whilst the latter resembles cytosine and could pair up with guanine. It is possible that a polymer of suitable derivatives of this basic structural unit might interact with an oncogene sequence and block its replication. Before rational design studies can be carried out on such molecules it is necessary to ascertain the preferred tautomeric form and modes of intermolecular association.

Ab initio calculations with extended basis sets show that the 3H tautomer of 4-pyrimidinone is considerably more stable that any other (82). It is not surprising that the 3H carbonyl oxygen atom bears a partial negative charge, and the hydrogen atom attached to N(3) or N(1) of the ring is positive. These unlike charges are nearer to each other in the 3H than in the 1H tautomer. The situation ought to be similar in dilute solution in a nonpolar solvent. However, in a polar solvent or in the solid state it is possible that dipole interactions could make the 1H tautomer more stable. In crystalline 6-methylisocytosine (R_5 = H, R_6 = CH_3) only the 1H tautomer is present (83). ABPP itself is reported to crystallize from acetic acid as the 1H tautomer with half of the molecules protonated (84). For the present study crystals of ABPP were grown from N-methylformamide and dimethylformamide in order to provide more biologically relevant conditions of neutrality and moderately high polarity.

3.2 The Crystal Structure of 2-amino-5-bromo-6-phenyl-4-(1/3H)-pyrimidinone. N-methylformamide solvate (ABPP.NMF).

3.2.1 Experimental

A crystalline sample of the compound (76) was obtained and from it a suitable crystal was selected. The quality of the specimen crystal was confirmed by examining its birefringent properties with a polarising microscope. The single crystal was then mounted onto a glass filament using an epoxy adhesive; the filament having been secured to a goniometer head stud by the hot wax method.

3.2.2 Crystal Data

$C_{10}H_8N_3OBr.C_{10}H_8N_3OBr.\ C_2H_5NO$, M = 296.64 F(000) = 592. Triclinic a = 9.147(3), b = 9.971(5), c = 14.590(4) Å, $\alpha = 70.00(3)$, $\beta = 71.14(3)$, $\gamma = 75.31(3)°$, V = 1168 $Å^3$ (by least-squares refinement on diffractometer angles for 25 automatically centred reflections, $\lambda = 0.71069$ Å), space group $P\bar{1}$, Z = 2, Dx = 1.68 gcm^{-3}, Dm = 1.71 gcm^{-3} (method of flot-ation in solvent mixture). White plates. Crystal dimensions, 0.075 x 0.05 x 0.35 mm, μ (Mo-Kα) = 34.15 cm^{-1}.

3.2.3 Data Collection and Processing

CAD4 diffractometer, w/2θ made with w scan width = 1.35 + 0.35 tanθ, w scan speed 0.7 - 2.5 deg min^{-1}, graphite monochromated Mo-Kα 4378 reflections measured ($2° \leq \theta \leq 25°$ + $h \pm k \pm l$), 4095 unique (merging R = 0.0270, no correction for absorption), giving 1951 with $I > 3\sigma(I)$, number of parameters refined 310. Linear and approximately isotropic crystal decay ca. 4% (for 3 reference reflections checked every 7200s x-ray exposure time), corrected during processing.

3.2.4 Structure Determination and Refinement

Data reduction followed by direct methods (SHELX76) and electron density synthesis. All hydrogen atoms were located by difference Fourier analysis during isotropic least squares refinement of non-hydrogen atoms. Due to the poor quality of the data and the presence of bromine in the structure it was found to be necessary to constrain all hydrogen atoms during full matrix least squares refinement with all non-hydrogen atoms anisotropic. Using the AFIX option in SHELX76 all hydrogens were refined with fixed geometry and common temperature factor; except pyrimidinone ring and amino hydrogens which were refined under a common temperature factor only. The weighting scheme $W = 1.4992/[\sigma^2(F_o) + 0.001\ F_o^2]$, with $\sigma(F_o)$ from counting statistics, gave satisfactory agreement analyses. Final R and R_w values are 0.0659 and 0.0746 after 15 cycles of least squares refinement. Examination of a final electron density synthesis showed no feature greater than $\sim 2/3$ eÅ^{-3}. At convergence (Δ/σ) max = 0.001 [C(4A)x/A].

Table 3.1 Positional parameters (fractional coordinates * 10**3 or 10**4) with estimated standard deviations in parentheses.

ATOM	x/a	y/b	z/c	
N(1A)	673(1)	1054(1)	725(1)	
C(2A)	713(1)	1004(1)	646(1)	
N(21A)	616(1)	024(1)	591(1)	
N(3A)	859(1)	935(1)	616(1)	
C(4A)	976(2)	908(1)	665(1)	
O(41A)	108(1)	845(1)	629(1)	
C(5A)	925(1)	965(1)	750(1)	
BR(1A)	0818(2)	9370(2)	8170(1)	
C(6A)	777(1)	031(1)	778(1)	
C(1'A)	717(1)	1079(2)	871(1)	
C(2'A)	634(2)	219(2)	862(1)	
C(3'A)	568(2)	268(2)	949(1)	
C(4'A)	584(2)	178(2)	042(1)	
C(5'A)	661(2)	037(2)	050(1)	
C(6'A)	728(2)	987(2)	965(1)	
N(1B)	1103(1)	622(1)	373(1)	
C(2B)	1066(1)	690(1)	447(1)	
N(21B)	1173(1)	669(1)	494(1)	
N(3B)	928(1)	770(1)	473(1)	
C(4B)	816(1)	783(1)	426(1)	
O(41B)	686(1)	863(1)	448(1)	
C(5B)	845(2)	713(1)	348(1)	
BR(1B)	6821(2)	7203(2)	2995(1)	
C(6B)	994(1)	633(1)	322(1)	
C(1'B)	054(2)	567(2)	239(1)	
C(2'B)	1140(2)	427(2)	255(1)	
C(3'B)	1200(2)	371(2)	172(1)	
C(4'B)	1171(2)	453(2)	078(1)	
C(5'B)	1088(2)	587(2)	065(1)	
C(6'B)	1029(2)	644(2)	145(1)	
N(1C)	1384(2)	667(1)	678(1)	
C(11C)	1360(3)	590(2)	786(2)	
C(2C)	1495(2)	626(2)	605(1)	
O(21C)	1603(1)	521(1)	620(1)	
H(1A)	628	956	560	*
H(2A)	515	1009	633	*
H(3A)	863	874	576	*
H(4A)	620(2)	289(2)	789(1)	
H(5A)	507(2)	377(2)	942(1)	
H(6A)	537(2)	217(2)	108(1)	
H(7A)	667(2)	964(2)	123(1)	
H(8A)	788(2)	877(2)	972(1)	
H(1B)	1208	531	375	*
H(2B)	278	644	480	*
H(3B)	175	679	541	*
H(4B)	1160(2)	365(2)	328(1)	
H(5B)	1270(2)	265(2)	180(1)	
H(6B)	1216(2)	406(2)	016(1)	
H(7B)	1069(2)	649(2)	-008(1)	
H(8B)	960(2)	750(2)	135(1)	
H(1C)	1491(2)	684(2)	528(1)	
H(2C)	1308(2)	768(1)	657(1)	

H(3C)	1263(3)	650(2)	828(2)
H(4C)	1464(3)	577(2)	811(2)
H(5C)	1334(3)	484(2)	798(2)

* Hydrogen atoms unrefined

Table 3.2 Anisotropic temperature factors, and isotropic temperature factors where shown; with standard deviations in parentheses.

ATOM	U11	U22	U33	U23	U13	U12
N(1A)	.033(6)	.033(7)	.036(7)	-.020(5)	-.015(5)	.001(5)
C(2A)	.025(7)	.031(8)	.039(8)	-.020(7)	-.005(6)	.006(6)
N(21A)	.030(6)	.069(9)	.061(8)	-.051(7)	-.018(6)	.012(6)
N(3A)	.038(6)	.030(6)	.040(7)	-.020(6)	-.022(5)	.009(5)
C(4A)	.035(8)	.036(8)	.027(8)	-.010(7)	-.011(6)	.008(7)
O(41A)	.023(5)	.039(6)	.057(6)	-.029(5)	-.015(5)	.016(4)
C(5A)	.031(7)	.030(8)	.031(8)	-.007(6)	-.024(6)	.003(6)
BR(1A)	.044(1)	.041(1)	.053(1)	-.019(1)	-.029(1)	.006(1)
C(6A)	.027(7)	.035(8)	.026(7)	-.015(6)	-.001(6)	-.009(6)
C(1′A)	.027(7)	.034(8)	.05(1)	-.023(8)	-.012(7)	.001(6)
C(2′A)	.037(9)	.04(1)	.06(1)	-.023(9)	-.017(8)	-.006(8)
C(3′A)	.07(1)	.07(1)	.07(1)	-.06(1)	.02(1)	-.00(1)
C(4′A)	.07(1)	.11(2)	.04(1)	-.05(1)	.00(1)	-.01(1)
C(5′A)	.040(9)	.11(2)	.04(1)	-.03(1)	-.011(8)	-.01(1)
C(6′A)	.047(9)	.05(1)	.040(9)	-.015(8)	-.012(8)	-.002(8)
N(1B)	.028(6)	.032(6)	.022(6)	-.011(5)	-.004(5)	.004(5)
C(2B)	.041(8)	.017(7)	.035(8)	-.008(6)	-.020(7)	.001(6)
N(21B)	.026(6)	.057(9)	.055(8)	-.032(7)	-.022(6)	.014(6)
N(3B)	.029(6)	.023(6)	.029(6)	-.013(5)	-.012(5)	.011(5)
C(4B)	.025(7)	.037(8)	.040(8)	-.020(7)	-.013(6)	.004(6)
O(41B)	.024(5)	.053(6)	.051(6)	-.034(5)	-.012(4)	.010(5)
C(5B)	.037(8)	.032(8)	.045(8)	-.021(7)	-.024(7)	.007(6)
BR(1B)	.035(1)	.063(1)	.063(1)	-.040(1)	-.020(1)	.005(1)
C(6B)	.029(7)	.020(7)	.038(8)	-.001(6)	-.015(6)	-.005(6)
C(1′B)	.042(8)	.041(9)	.029(8)	-.020(7)	-.013(7)	.001(7)
C(2′B)	.040(8)	.030(0)	.038(9)	-.020(6)	.002(7)	.008(7)
C(3′B)	.046(9)	.028(9)	.07(1)	-.014(8)	-.013(9)	.003(7)
C(4′B)	.047(9)	.06(1)	.04(1)	-.036(9)	.005(8)	-.018(9)
C(5′B)	.06(1)	.06(1)	.032(8)	-.028(8)	-.014(7)	.014(8)
C(6′B)	.052(9)	.036(9)	.048(9)	-.020(8)	-.018(8)	-.001(7)
N(1C)	.063(9)	.036(8)	.10(1)	-.036(8)	-.046(9)	.029(7)
C(11C)	.17(2)	.10(2)	.09(2)	-.05(1)	-.05(2)	.08(2)
C(2C)	.035(9)	.07(1)	.07(1)	-.02(1)	-.012(9)	.015(9)
O(21C)	.044(6)	.057(7)	.097(9)	-.042(7)	-.031(6)	.025(6)

	U ISO
H(1A)	.03(1)
H(2A)	.03(1)
H(3A)	.03(1)
H(4A)	.09(1)
H(5A)	.09(1)
H(6A)	.09(1)
H(7A)	.09(1)
H(8A)	.09(1)
H(1B)	.03(1)
H(2B)	.03(1)
H(3B)	.03(1)
H(4B)	.09(1)
H(5B)	.09(1)
H(6B)	.09(1)
H(7B)	.09(1)

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Table 3.2 contd.

	U ISO
H(8B)	.09(1)
H(1C)	.3(1)
H(2C)	.3(1)
H(3C)	.22(8)
H(4C)	.22(8)
H(5C)	.22(8)

Table 3.3 Bond Lengths(A) with standard deviations

N(1A) -C(2A)	1.309(18)	C(2B) -N(3B)	1.334(17)
N(1A) -C(6A)	1.346(17)	N(21B)-H(2B)	0.903(12)
C(2A) -N(21A)	1.322(19)	N(21B)-H(3B)	0.735(12)
C(2A) -N(3A)	1.342(18)	N(3B) -C(4B)	1.360(17)
N(21A)-H(1A)	0.903(13)	C(4B) -O(41B)	1.264(16)
N(21A)-H(2A)	0.949(13)	C(4B) -C(5B)	1.450(19)
N(3A) -C(4A)	1.393(18)	C(5B) -Br(1B)	1.827(14)
N(3A) -H(3A)	0.972(11)	C(5B) -C(6B)	1.395(20)
C(4A) -O(41A)	1.244(17)	C(6B) -C(1'B)	1.459(19)
C(4A) -C(5A)	1.440(19)	C(1'B)-C(2'B)	1.398(20)
C(5A) -Br(1A)	1.898(13)	C(1'B)-C(6'B)	1.390(21)
C(5A) -C(6A)	1.346(19)	C(2'B)-H(4B)	1.080(21)
C(6A) -C(1'A)	1.481(20)	C(2'B)-C(3'B)	1.394(22)
C(1'A)-C(2'A)	1.397(22)	C(3'B)-H(5B)	1.080(23)
C(1'A)-C(6'A)	1.381(22)	C(3'B)-C(4'B)	1.400(24)
C(2'A)-H(4A)	1.080(23)	C(4'B)-H(6B)	1.080(25)
C(2'A)-C(3'A)	1.41(3)	C(4'B)-C(5'B)	1.347(23)
C(3'A)-H(5A)	1.08(3)	C(5'B)-H(7B)	1.080(22)
C(3'A)-C(4'A)	1.38(3)	C(5'B)-C(6'B)	1.360(22)
C(4'A)-H(6A)	1.08(3)	C(6'B)-H(8B)	1.079(22)
C(4'A)-C(5'A)	1.39(3)	N(1C) -H(2C)	1.080(20)
C(5'A)-H(7A)	1.08(3)	N(1C) -C(11C)	1.46(3)
C(5'A)-C(6'A)	1.391(25)	N(1C) -C(2C)	1.317(23)
C(6'A)-H(8A)	1.080(23)	C(11C)-H(3C)	1.08(3)
N(1B) -C(2B)	1.376(17)	C(11C)-H(4C)	1.08(3)
N(1B) -C(6B)	1.387(17)	C(11C)-H(5C)	1.08(3)
N(1B) -H(1B)	1.147(10)	C(2C) -H(1C)	1.08(3)
C(2B) -N(21B)	1.305(18)	C(2C) -O(21C)	1.249(21)

Table 3.4 Angles(degrees) with standard deviations

C(2A) -N(1A) -C(6A)	119.4(12)	C(2B) -N(21B)-H(2B)	135.3(13)
N(1A) -C(2A) -N(21A)	122.0(13)	C(2B) -N(21B)-H(3B)	136.4(15)
N(1A) -C(2A) -N(3A)	120.9(12)	H(2B) -N(21B)-H(3B)	88.1(12)
N(21A)-C(2A) -N(3A)	117.1(13)	C(2B) -N(3B) -C(4B)	118.4(11)
C(2A) -N(21A)-H(1A)	117.3(13)	N(3B) -C(4B) -O(41B)	119.0(12)
C(2A) -N(21A)-H(2A)	110.1(13)	N(3B) -C(4B) -C(5B)	121.8(12)
H(1A) -N(21A)-H(2A)	95.8(12)	O(41B)-C(4B) -C(5B)	119.2(12)
C(2A) -N(3A) -C(4A)	124.5(12)	C(4B) -C(5B) -Br(1B)	118.5(10)
C(2A) -N(3A) -H(3A)	112.2(11)	C(4B) -C(5B) -C(6B)	117.4(12)
C(4A) -N(3A) -H(3A)	119.4(11)	Br(1B)-C(5B) -C(6B)	123.8(11)
N(3A) -C(4A) -O(41A)	118.9(12)	N(1B) -C(6B) -C(5B)	118.7(12)

N(3A) -C(4A) -C(5A)	112.1(12)	N(1B) -C(6B) -C(1'B)	114.2(11)
O(41A)-C(4A) -C(5A)	128.9(13)	C(5B) -C(6B) -C(1'B)	127.0(13)
C(4A) -C(5A) -Br(1A)	114.4(10)	C(6B) -C(1'B)-C(2'B)	119.9(13)
C(4A) -C(5A) -C(6A)	120.9(12)	C(6B) -C(1'B)-C(6'B)	120.1(13)
Br(1A)-C(5A) -C(6A)	124.7(10)	C(2'B)-C(1'B)-C(6'B)	120.0(13)
N(1A) -C(6A) -C(5A)	122.1(12)	C(1'B)-C(2'B)-H(4B)	121.4(16)
N(1A) -C(6A) -C(1'A)	115.8(12)	C(1'B)-C(2'B)-C(3'B)	117.4(14)
C(5A) -C(6A) -C(1'A)	122.0(12)	H(4B) -C(2'B)-C(3'B)	121.2(16)
C(6A) -C(1'A)-C(2'A)	117.1(13)	C(2'B)-C(3'B)-H(5B)	119.6(17)
C(6A) -C(1'A)-C(6'A)	122.6(13)	C(2'B)-C(3'B)-C(4'B)	120.3(15)
C(2'A)-C(1'A)-C(6'A)	120.0(14)	H(5B) -C(3'B)-C(4'B)	120.1(17)
C(1'A)-C(2'A)-H(4A)	120.1(17)	C(3'B)-C(4'B)-H(6B)	119.0(18)
C(1'A)-C(2'A)-C(3'A)	119.5(15)	C(3'B)-C(4'B)-C(5'B)	121.5(16)
H(4A) -C(2'A)-C(3'A)	120.5(18)	H(6B) -C(4'B)-C(5'B)	119.6(18)
C(2'A)-C(3'A)-H(5A)	119.6(20)	C(4'B)-C(5'B)-H(7B)	120.3(17)
C(2'A)-C(3'A)-C(4'A)	120.3(18)	C(4'B)-C(5'B)-C(6'B)	119.0(15)
H(5A) -C(3'A)-C(4'A)	120.0(21)	H(7B) -C(5'B)-C(6'B)	120.7(17)
C(3'A)-C(4'A)-H(6A)	120.1(22)	C(1'B)-C(6'B)-C(5'B)	121.8(14)
C(3'A)-C(4'A)-C(5'A)	119.4(19)	C(1'B)-C(6'B)-H(8B)	118.8(16)
H(6A) -C(4'A)-C(5'A)	120.5(22)	C(5'B)-C(6'B)-H(8B)	119.4(17)
C(4'A)-C(5'A)-H(7A)	119.5(20)	H(2C) -N(1C) -C(11C)	117.9(17)
C(4'A)-C(5'A)-C(6'A)	121.1(17)	H(2C) -N(1C) -C(2C)	117.0(16)
H(7A) -C(5'A)-C(6'A)	119.5(20)	C(11C)-N(1C) -C(2C)	125.0(16)
C(1'A)-C(6'A)-C(5'A)	119.6(15)	N(1C) -C(11C)-H(3C)	108.8(23)
C(1'A)-C(6'A)-H(8A)	120.3(17)	N(1C) -C(11C)-H(4C)	111.4(23)
C(5'A)-C(6'A)-H(8A)	120.1(18)	N(1C) -C(11C)-H(5C)	108.3(23)
C(2B) -N(1B) -C(6B)	120.7(11)	H(3C) -C(11C)-H(4C)	109.4(27)
C(2B) -N(1B) -H(1B)	114.7(10)	H(3C) -C(11C)-H(5C)	109.5(27)
C(6B) -N(1B) -H(1B)	121.8(10)	H(4C) -C(11C)-H(5C)	109.4(27)
N(1B) -C(2B) -N(21B)	116.9(12)	N(1C) -C(2C) -H(1C)	117.9(19)
N(1B) -C(2B) -N(3B)	123.0(12)	N(1C) -C(2C) -O(21C)	123.7(16)
N(21B)-C(2B) -N(3B)	120.1(12)	H(1C) -C(2C) -O(21C)	118.3(19)

Table 3.5 Torsion angles(degrees) with standard deviations

C(6A) -N(1A) -C(2A) -N(21A)	-179.6(13)	H(1B) -N(1B) -C(2B) -N(3B)	-162.3(11)
C(6A) -N(1A) -C(2A) -N(3A)	-1.9(20)	C(2B) -N(1B) -C(6B) -C(5B)	-0.8(19)
C(2A) -N(1A) -C(6A) -C(5A)	3.3(20)	C(2B) -N(1B) -C(6B) -C(1'B)	174.7(12)
C(2A) -N(1A) -C(6A) -C(1'A)	-174.8(12)	H(1B) -N(1B) -C(6B) -C(5B)	159.1(11)
N(1A) -C(2A) -N(21A)-H(1A)	-150.4(14)	H(1B) -N(1B) -C(6B) -C(1'B)	-25.4(17)
N(1A) -C(2A) -N(21A)-H(2A)	-42.3(19)	N(1B) -C(2B) -N(21B)-H(2B)	23.7(24)
N(3A) -C(2A) -N(21A)-H(1A)	31.9(20)	N(1B) -C(2B) -N(21B)-H(3B)	-164.0(18)
N(3A) -C(2A) -N(21A)-H(2A)	139.9(13)	N(3B) -C(2B) -N(21B)-H(2B)	-158.1(15)
N(1A) -C(2A) -N(3A) -C(4A)	0.8(21)	N(3B) -C(2B) -N(21B)-H(3B)	14.1(28)
N(1A) -C(2A) -N(3A) -H(3A)	158.2(12)	N(1B) -C(2B) -N(3B) -C(4B)	1.7(19)

N(21A)-C(2A) -N(3A) -C(4A)	178.6(13)
N(21A)-C(2A) -N(3A) -H(3A)	-24.0(18)
C(2A) -N(3A) -C(4A) -O(41A)	-178.6(13)
C(2A) -N(3A) -C(4A) -C(5A)	-0.9(19)
H(3A) -N(3A) -C(4A) -O(41A)	25.5(19)
H(3A) -N(3A) -C(4A) -C(5A)	-156.8(12)
N(3A) -C(4A) -C(5A) -Br(1A)	-178.0(9)
N(3A) -C(4A) -C(5A) -C(6A)	2.1(19)
O(41A)-C(4A) -C(5A) -Br(1A)	-0.6(20)
O(41A)-C(4A) -C(5A) -C(6A)	179.5(14)
C(4A) -C(5A) -C(6A) -N(1A)	-3.4(21)
C(4A) -C(5A) -C(6A) -C(1'A)	174.5(13)
Br(1A)-C(5A) -C(6A) -N(1A)	176.7(10)
Br(1A)-C(5A) -C(6A) -C(1'A)	-5.3(19)
N(1A) -C(6A) -C(1'A)-C(2'A)	-49.5(18)
N(1A) -C(6A) -C(1'A)-C(6'A)	124.6(15)
C(5A) -C(6A) -C(1'A)-C(2'A)	132.4(15)
C(5A) -C(6A) -C(1'A)-C(6'A)	-53.4(21)
C(6A) -C(1'A)-C(2'A)-H(4A)	-2.4(24)
C(6A) -C(1'A)-C(2'A)-C(3'A)	177.1(15)
C(6'A)-C(1'A)-C(2'A)-H(4A)	-176.7(17)
C(6'A)-C(1'A)-C(2'A)-C(3'A)	2.8(24)
C(6A) -C(1'A)-C(6'A)-C(5'A)	-176.1(15)
C(6A) -C(1'A)-C(6'A)-H(8A)	3.7(26)
C(2'A)-C(1'A)-C(6'A)-C(5'A)	-2.1(24)
C(2'A)-C(1'A)-C(6'A)-H(8A)	177.6(17)
C(1'A)-C(2'A)-C(3'A)-H(5A)	178.5(20)
C(1'A)-C(2'A)-C(3'A)-C(4'A)	-0.6(27)
H(4A) -C(2'A)-C(3'A)-H(5A)	-2.1(31)
H(4A) -C(2'A)-C(3'A)-C(4'A)	178.9(20)
C(2'A)-C(3'A)-C(4'A)-H(6A)	178.1(22)
C(2'A)-C(3'A)-C(4'A)-C(5'A)	-2.3(29)
H(5A) -C(3'A)-C(4'A)-H(6A)	-1.0(36)
H(5A) -C(3'A)-C(4'A)-C(5'A)	178.7(21)
C(3'A)-C(4'A)-C(5'A)-H(7A)	-176.4(21)
C(3'A)-C(4'A)-C(5'A)-C(6'A)	3.0(30)
H(6A) -C(4'A)-C(5'A)-H(7A)	3.2(35)
H(6A) -C(4'A)-C(5'A)-C(6'A)	-177.4(22)
C(4'A)-C(5'A)-C(6'A)-C(1'A)	-0.8(27)
C(4'A)-C(5'A)-C(6'A)-H(8A)	179.5(20)
H(7A) -C(5'A)-C(6'A)-C(1'A)	178.6(19)
H(7A) -C(5'A)-C(6'A)-H(8A)	-1.1(31)
C(6B) -N(1B) -C(2B) -N(21B)	177.1(12)
C(6B) -N(1B) -C(2B) -N(3B)	-1.0(19)
H(1B) -N(1B) -C(2B) -N(21B)	15.8(16)
H(2C) -N(1C) -C(2C) -H(1C)	-11.3(28)
H(2C) -N(1C) -C(2C) -O(21C)	170.6(17)
N(21B)-C(2B) -N(3B) -C(4B)	-176.3(12)
C(2B) -N(3B) -C(4B) -O(41B)	-177.6(12)
C(2B) -N(3B) -C(4B) -C(5B)	-0.7(19)
N(3B) -C(4B) -C(5B) -Br(1B)	173.4(10)
N(3B) -C(4B) -C(5B) -C(6B)	-1.1(20)
O(41B)-C(4B) -C(5B) -Br(1B)	-9.6(18)
O(41B)-C(4B) -C(5B) -C(6B)	175.9(12)
C(4B) -C(5B) -C(6B) -N(1B)	1.8(19)
C(4B) -C(5B) -C(6B) -C(1'B)	-173.1(13)
Br(1B)-C(5B) -C(6B) -N(1B)	-172.4(10)
Br(1B)-C(5B) -C(6B) -C(1'B)	12.7(21)
N(1B) -C(6B) -C(1'B)-C(2'B)	47.8(18)
N(1B) -C(6B) -C(1'B)-C(6'B)	-130.9(14)
C(5B) -C(6B) -C(1'B)-C(2'B)	-137.1(15)
C(5B) -C(6B) -C(1'B)-C(6'B)	44.1(21)
C(6B) -C(1'B)-C(2'B)-H(4B)	2.3(24)
C(6B) -C(1'B)-C(2'B)-C(3'B)	-177.8(13)
C(6'B)-C(1'B)-C(2'B)-H(4B)	-179.0(16)
C(6'B)-C(1'B)-C(2'B)-C(3'B)	1.0(21)
C(6B) -C(1'B)-C(6'B)-C(5'B)	177.7(14)
C(6B) -C(1'B)-C(6'B)-H(8B)	-2.6(24)
C(2'B)-C(1'B)-C(6'B)-C(5'B)	-1.1(23)
C(2'B)-C(1'B)-C(6'B)-H(8B)	178.6(16)
C(1'B)-C(2'B)-C(3'B)-H(5B)	178.7(17)
C(1'B)-C(2'B)-C(3'B)-C(4'B)	-1.0(23)
H(4B) -C(2'B)-C(3'B)-H(5B)	-1.4(27)
H(4B) -C(2'B)-C(3'B)-C(4'B)	178.9(17)
C(2'B)-C(3'B)-C(4'B)-H(6B)	-178.5(18)
C(2'B)-C(3'B)-C(4'B)-C(5'B)	1.2(25)
H(5B) -C(3'B)-C(4'B)-H(6B)	1.8(29)
H(5B) -C(3'B)-C(4'B)-C(5'B)	-178.5(18)
C(3'B)-C(4'B)-C(5'B)-H(7B)	178.8(18)
C(3'B)-C(4'B)-C(5'B)-C(6'B)	-1.3(25)
H(6B) -C(4'B)-C(5'B)-H(7B)	-1.5(29)
H(6B) -C(4'B)-C(5'B)-C(6'B)	178.5(18)
C(4'B)-C(5'B)-C(6'B)-C(1'B)	1.2(24)
C(4'B)-C(5'B)-C(6'B)-H(8B)	-178.5(18)
H(7B) -C(5'B)-C(6'B)-C(1'B)	-178.9(17)
H(7B) -C(5'B)-C(6'B)-H(8B)	1.5(28)
H(2C) -N(1C) -C(11C)-H(3C)	1.6(30)
H(2C) -N(1C) -C(11C)-H(4C)	-119.1(25)
H(2C) -N(1C) -C(11C)-H(5C)	120.5(24)
C(2C) -N(1C) -C(11C)-H(3C)	178.2(22)
C(2C) -N(1C) -C(11C)-H(4C)	57.5(30)
C(2C) -N(1C) -C(11C)-H(5C)	-62.9(29)
C(11C)-N(1C) -C(2C) -H(1C)	172.0(20)
C(11C)-N(1C) -C(2C) -O(21C)	-6.1(29)

Table 3.6 Hydrogen bond contact distances

Hydrogen Bond	Donor-Acceptor Distance (Å)
$N(1B)-H(1B)_{I}\ldots O(21C)_{II}$	2.731
$N(1C)-H(2C)_{I}\ldots O(41A)_{I}$	2.839
$N(21A)-H(1A)_{I}\ldots O(41B)_{I}$	2.866
$N(21A)-H(2A)_{I}\ldots O(41B)_{III}$	2.871
$N(21B)-H(3B)_{I}\ldots O(41A)_{I}$	2.897
$N(21B)-H(2B)_{I}\ldots O(21C)_{II}$	2.921
$N(3A)-H(3A)_{I}\ldots N(3B)_{I}$	2.901

The subscripts I, II and III refer to the equivalent positions X, Y, Z; 3-X, 1-Y, 1-Z; 1-X, 2-Y, 1-Z.

Figure 3.1 ABPP.NMF Molecular plot and numbering scheme

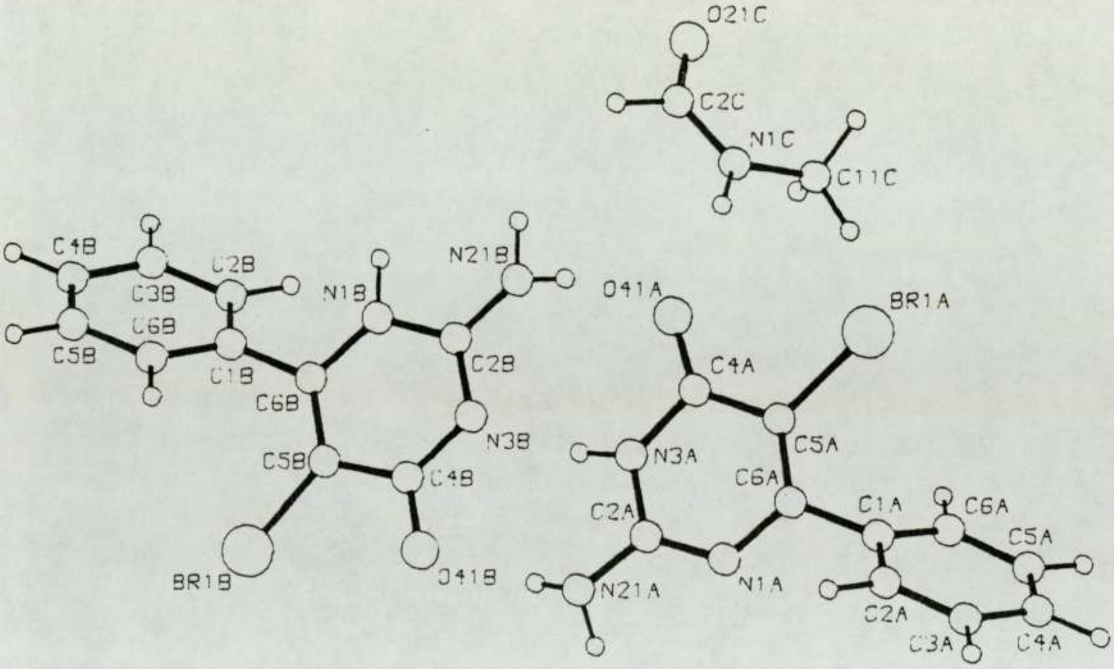
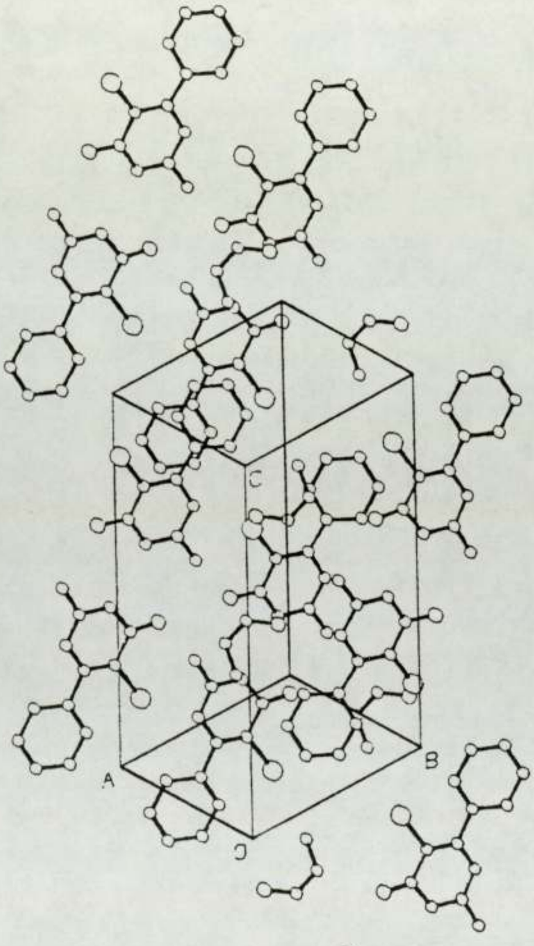


Figure 3.2 ABPP.NMF Unit cell packing diagram



3.3 The Crystal Structure of 2-amino-5-bromo-6-phenyl-4-(1H)-pyrimidinone.dimethyl formamide solvate (ABPP.DMF)

3.3.1 Experimental

A crystalline sample of the compound was obtained (76) and from it a suitable crystal was selected. The quality of the specimen crystal was confirmed by examining its birefringent properties with a polarising microscope. The single crystal was then monted onto a glass filament using epoxy adhesive; the filament having been secured to a goniometer head stud by the hot wax method.

3.3.2 Crystal Data

$C_{10}H_8N_3OBr.C_3H_7NO$, M = 340.174, F(000)=692. Orthorhombic a = 32.715(6), b = 6.663(2), c = 6.774(3) Å, V = 1477 $Å^3$ (by least squares refinement on diffractometer angles for 25 automatically centred reflections, λ = 0.71069 Å), space group Pnn2, Z = 4, Dx = 1.53 gcm^{-3}, Dm = 1.527 gcm^{-3} (method of floatation in solvent mixture). White opalescent trapezoids. Crystal dimensions 0.6 x 0.4 x 0.2 mm, μ (Mo-Kα) = 27.09 cm^{-1}.

3.3.3 Data Collection and Processing

CAD4 diffractometer, w/2θ mode with w scan width = 1.0 + 0.35 tan θ, w scan speed 0.3 - 3.3 deg min^{-1}, graphite monochromated Mo-Kα radiation; 3754 reflections measured (2 $\geqslant \theta \geqslant$ 27° + h + k $\pm$ l), 1732 unique (merging R = 0.0667, no correction for absorption) giving 1353 with I > 3 σ(I), number of parameters refined 150. Linear and approximately isotropic crystal decay ca. 2.4% (for 3 reference reflections checked every 7200 s x-ray exposure time) corrected during processing.

3.3.4 Structure Determination and Refinement.

Data reduction was followed by direct methods phase determination and electron density synthesis (MULTAN80) revealed all non-hydrogen atoms. Difference Fourier analysis during isotropic least squares refinement of non-hydrogen atoms did not clearly reveal hydrogen atoms, therefore it was decided to calculate all hydrogens using AFIX options in SHELX76 then to refine them in fixed geometries and under a common temperature factor. The 2-amino hydrogens were located and these were refined normally. Due to the lack of data $>3\sigma$ from the small sample crystal, the phenyl ring substituent was refined as a rigid group with a common isotropic temperature factor for both hydrogen and non-hydrogen atoms. The Z/C parameter for Br(51) was fixed as the origin. The weighting scheme $W = 7.5983/\sigma^2(F_o)$, with $\sigma(F_o)$ from counting statistics, gave reasonable agreement analyses. The final R and R_w values are 0.0815 and 0.0986. Full matrix least squares refinement with all unrestrained non-hydrogen atoms anisotropic gave reasonable convergence after 20 cycles of refinement using a damping factor of 10. Final electron density synthesis showed two peaks ~ 1 $e\text{Å}^{-3}$ these being associated with N(1) and N(3) ring protonation which is unclear, after which there was no feature $> 2/3$ e Å^{-3}. At convergence (Δ/σ) max = 0.0141 [C(22S) U23]. Atom C5 became non-positive definite and on examination this proved to be due to its U22 value; all U22's were significantly low in value. It is possible that this observation is due to absorptin effects, arising from the large crystal dimensions relative to μ, reducing the intensity of certain classes of reflections.

Table 3.7 Positional parameters (fractional coordinates * 10**4, or 10**3) with estimated standard deviations in parentheses.

ATOM	x/a	y/b	z/c	
N(1)	0993(2)	6949(9)	640(3)	
C(2)	0605(3)	621(1)	635(5)	
N(21)	0301(3)	751(1)	618(5)	
N(3)	0526(2)	4252(9)	631(4)	
C(4)	0828(3)	291(1)	631(4)	
O(41)	0756(2)	1079(9)	638(3)	
C(5)	1243(3)	366(1)	636(4)	
C(6)	1326(3)	567(1)	635(5)	
BR(51)	16741(3)	1810(1)	62599	I
C(1′)	1732(2)	656(1)	628(2)	
C(2′)	1886(2)	711(1)	444(2)	
C(3′)	2278(2)	793(1)	428(2)	
C(4′)	2515(2)	820(1)	597(2)	
C(5′)	2360(2)	766(1)	782(2)	
C(6′)	1969(2)	684(1)	797(2)	
C(1S)	107(1)	378(5)	130(6)	
O(1S)	1175(5)	529(3)	132(4)	
N(1S)	0837(5)	233(2)	143(5)	
C(21S)	094(1)	036(5)	135(6)	
C(22S)	0427(8)	273(6)	141(8)	
H(1)	035(3)	82(1)	64(2)	II
H(2)	-001(3)	70(1)	57(1)	II
H(4)	1703(2)	689(1)	313(2)	
H(5)	2397(2)	835(1)	286(2)	
H(6)	2817(2)	884(1)	586(2)	
H(7)	2544(2)	787(1)	913(2)	
H(8)	1850(2)	642(1)	940(2)	
H(9)	134(1)	303(5)	080(6)	
H(10)	126(1)	004(5)	128(6)	
H(11)	079(1)	-021(5)	005(6)	
H(12)	081(1)	-034(5)	265(6)	
H(13)	0366(8)	433(6)	147(8)	
H(14)	0301(8)	202(6)	271(8)	
H(15)	0287(8)	211(6)	011(8)	

I Bromine x/a * 10**5, z/c fixed during refinement.

II Hydrogens freely refining, no constraints.

All other hydrogens refining in rigid groups using AFIX option in SHELX.

Table 3.8 Anisotropic temperature factors, and isotropic temperature factors where shown; with standard deviations in parentheses.

ATOM	U11	U22	U33	U23	U13	U12
N(1)	.024(3)	.009(3)	.053(6)	-.003(9)	-.006(9)	-.003(3)
C(2)	.029(4)	.015(4)	.054(7)	.01(1)	.00(1)	-.002(3)
N(21)	.025(4)	.009(4)	.13(1)	.00(1)	-.03(1)	-.002(3)
N(3)	.026(4)	.009(3)	.052(6)	.00(1)	.00(1)	-.006(3)
C(4)	.038(5)	.011(4)	.041(7)	.01(1)	.02(1)	-.003(3)
O(41)	.038(4)	.010(3)	.069(6)	.007(9)	.00(1)	-.008(2)
C(5)	.030(4)	.010(4)	.064(8)	-.034(9)	-.01(1)	.001(3)
C(6)	.040(5)	.011(4)	.049(7)	.00(1)	.03(1)	.002(4)
BR(51)	.040(5)	.011(4)	.149(2)	-.005(2)	.012(2)	.002(4)
C(1S)	.36(2)	.26(2)	.026(9)	-.05(2)	.05(2)	-.22(1)
O(1S)	.23(1)	.13(1)	.10(1)	-.02(1)	-.04(2)	-.10(1)
N(1S)	.12(1)	.073(8)	.08(1)	-.02(1)	-.03(1)	-.037(8)
C(21S)	.46(2)	.17(2)	.08(1)	.03(2)	.06(2)	.07(2)
C(22S)	.14(2)	.50(2)	.07(1)	.04(2)	-.02(2)	-.03(2)

	U ISO
C(1′)	.039(1)
C(2′)	.039(1)
C(3′)	.039(1)
C(4′)	.039(1)
C(5′)	.039(1)
C(6′)	.039(1)
H(1)	.00(2)
H(2)	.00(2)
H(4)	.05(1)
H(5)	.05(1)
H(6)	.05(1)
H(7)	.05(1)
H(8)	.05(1)
H(9)	.17(2)
H(10)	.27(2)
H(11)	.27(2)
H(12)	.27(2)
H(13)	.27(2)
H(14)	.27(2)
H(15)	.27(2)

Table 3.9 Bond Lengths(A) with standard deviations

N(1) - C(2)	1.36(3)	C(4') -C(5')	1.396(12)
N(1) - C(6)	1.383(24)	C(4') - H(6)	1.077(12)
C(2) -N(21)	1.32(3)	C(5') -C(6')	1.395(12)
C(2) - N(3)	1.33(3)	C(5') - H(7)	1.081(12)
N(21) - H(1)	0.54(9)	C(6') -C(1')	1.395(12)
N(21) - H(2)	1.12(8)	C(6') - H(8)	1.079(12)
N(3) - C(4)	1.332(24)	C(1S) -O(1S)	1.06(4)
C(4) -O(41)	1.244(22)	C(1S) -N(1S)	1.23(4)
C(4) - C(5)	1.45(3)	C(1S) - H(9)	1.08(5)
C(5) - C(6)	1.37(3)	N(1S) -C(21S)	1.36(4)
C(5) -Br(51)	1.876(19)	N(1S) -C(22S)	1.37(5)
C(6) -C(1')	1.456(21)	C(21S)-H(10)	1.08(5)
C(2') -C(3')	1.398(12)	C(21S)-H(11)	1.08(5)
C(2') -C(1')	1.395(12)	C(21S)-H(12)	1.08(5)
C(2') - H(4)	1.079(12)	C(22S)-H(13)	1.08(6)
C(3') -C(4')	1.395(12)	C(22S)-H(14)	1.08(6)
C(3') - H(5)	1.080(12)	C(22S)-H(15)	1.08(6)

Table 3.10 Angles(degrees) with standard deviations

C(2) - N(1) - C(6)	120.7(16)	C(4') -C(5') - H(7)	119.8(9)
N(1) - C(2) -N(21)	117.7(19)	C(6') -C(5') - H(7)	120.0(9)
N(1) - C(2) - N(3)	122.5(18)	C(5') -C(6') -C(1')	120.0(8)
N(21) - C(2) - N(3)	119.6(19)	C(5') -C(6') - H(8)	120.0(9)
C(2) -N(21) - H(1)	111.1(98)	C(1') -C(6') - H(8)	120.0(9)
C(2) -N(21) - H(2)	122.2(42)	C(6) -C(1') -C(2')	117.6(10)
H(1) -N(21) - H(2)	126.3(**)	C(6) -C(1') -C(6')	122.3(10)
C(2) - N(3) - C(4)	120.9(17)	C(2') -C(1') -C(6')	120.0(8)
N(3) - C(4) -O(41)	121.2(17)	O(1S) -C(1S) -N(1S)	159.9(39)
N(3) - C(4) - C(5)	117.6(16)	O(1S) -C(1S) - H(9)	100.0(38)
O(41) - C(4) - C(5)	121.1(16)	N(1S) -C(1S) - H(9)	100.1(36)
C(4) - C(5) - C(6)	121.7(17)	C(1S) -N(1S) -C(21S)	126.6(29)
C(4) - C(5) -Br(51)	118.4(13)	C(1S) -N(1S) -C(22S)	116.8(29)
C(6) - C(5) -Br(51)	119.7(14)	C(21S)-N(1S) -C(22S)	116.0(28)
N(1) - C(6) - C(5)	116.5(17)	N(1S) -C(21S)-H(10)	116.4(37)
N(1) - C(6) -C(1')	117.9(15)	N(1S) -C(21S)-H(11)	105.0(35)
C(5) - C(6) -C(1')	125.6(16)	N(1S) -C(21S)-H(12)	106.7(36)
C(3') -C(2') -C(1')	120.0(8)	H(10) -C(21S)-H(11)	109.5(42)
C(3') -C(2') - H(4)	120.0(9)	H(10) -C(21S)-H(12)	109.5(42)
C(1') -C(2') - H(4)	120.1(9)	H(11) -C(21S)-H(12)	109.5(42)
C(2') -C(3') -C(4')	120.0(8)	N(1S) -C(22S)-H(13)	112.0(40)
C(2') -C(3') - H(5)	119.9(9)	N(1S) -C(22S)-H(14)	106.2(39)

C(4') -C(3') - H(5)	120.0(9)	N(1S) -C(22S)-H(15)	110.4(40)
C(3') -C(4') -C(5')	119.9(8)	H(13) -C(22S)-H(14)	109.4(47)
C(3') -C(4') - H(6)	120.0(9)	H(13) -C(22S)-H(15)	109.4(47)
C(5') -C(4') - H(6)	120.1(9)	H(14) -C(22S)-H(15)	109.4(47)
C(4') -C(5') -C(6')	120.1(8)		

Table 3.11 Torsion angles(degrees) with standard deviations

C(6) - N(1) - C(2) -N(21)	-172.9(18)	C(2') -C(3') -C(4') -C(5')	0.0(13)
C(6) - N(1) - C(2) - N(3)	2.0(29)	C(2') -C(3') -C(4') - H(6)	-179.9(9)
C(2) - N(1) - C(6) - C(5)	-3.3(26)	H(5) -C(3') -C(4') -C(5')	179.9(9)
C(2) - N(1) - C(6) -C(1')	176.2(15)	H(5) -C(3') -C(4') - H(6)	0.0(15)
N(1) - C(2) -N(21) - H(1)	-23.7(**)	C(3') -C(4') -C(5') -C(6')	0.1(13)
N(1) - C(2) -N(21) - H(2)	163.6(47)	C(3') -C(4') -C(5') - H(7)	179.9(9)
N(3) - C(2) -N(21) - H(1)	161.3(**)	H(6) -C(4') -C(5') -C(6')	180.0(9)
N(3) - C(2) -N(21) - H(2)	-11.4(54)	H(6) -C(4') -C(5') - H(7)	-0.1(15)
N(1) - C(2) - N(3) - C(4)	-0.4(30)	C(4') -C(5') -C(6') -C(1')	-0.1(13)
N(21) - C(2) - N(3) - C(4)	174.3(19)	C(4') -C(5') -C(6') - H(8)	179.9(9)
C(2) - N(3) - C(4) -O(41)	176.2(18)	H(7) -C(5') -C(6') -C(1')	-180.0(9)
C(2) - N(3) - C(4) - C(5)	0.3(27)	H(7) -C(5') -C(6') - H(8)	0.0(15)
N(3) - C(4) - C(5) - C(6)	-1.8(27)	C(5') -C(6') -C(1') - C(6)	178.5(11)
N(3) - C(4) - C(5) -Br(51)	-177.8(13)	C(5') -C(6') -C(1') -C(2')	0.0(13)
O(41) - C(4) - C(5) - C(6)	-177.7(18)	H(8) -C(6') -C(1') - C(6)	-1.5(16)
O(41) - C(4) - C(5) -Br(51)	6.3(24)	H(8) -C(6') -C(1') -C(2')	-180.0(9)
C(4) - C(5) - C(6) - N(1)	3.2(27)	O(1S) -C(1S) -N(1S) -C(21S)	-171.8(**)
C(4) - C(5) - C(6) -C(1')	-176.2(16)	O(1S) -C(1S) -N(1S) -C(22S)	17.1(**)
Br(51)- C(5) - C(6) - N(1)	179.2(13)	H(9) -C(1S) -N(1S) -C(21S)	11.7(49)
Br(51)- C(5) - C(6) -C(1')	-0.2(26)	H(9) -C(1S) -N(1S) -C(22S)	-159.5(34)
N(1) - C(6) -C(1') -C(2')	-84.8(17)	C(1S) -N(1S) -C(21S)-H(10)	5.1(57)
N(1) - C(6) -C(1') -C(6')	96.7(17)	C(1S) -N(1S) -C(21S)-H(11)	-116.2(41)
C(5) - C(6) -C(1') -C(2')	94.6(20)	C(1S) -N(1S) -C(21S)-H(12)	127.6(40)
C(5) - C(6) -C(1') -C(6')	-83.9(21)	C(22S)-N(1S) -C(21S)-H(10)	176.3(38)
C(1') -C(2') -C(3') -C(4')	-0.1(13)	C(22S)-N(1S) -C(21S)-H(11)	55.0(44)
C(1') -C(2') -C(3') - H(5)	-180.0(9)	C(22S)-N(1S) -C(21S)-H(12)	-61.1(44)
H(4) -C(2') -C(3') -C(4')	-180.0(9)	C(1S) -N(1S) -C(22S)-H(13)	-7.6(53)
H(4) -C(2') -C(3') - H(5)	0.1(15)	C(1S) -N(1S) -C(22S)-H(14)	-127.0(41)
C(3') -C(2') -C(1') - C(6)	-178.5(11)	C(1S) -N(1S) -C(22S)-H(15)	114.5(43)
C(3') -C(2') -C(1') -C(6')	0.1(13)	C(21S)-N(1S) -C(22S)-H(13)	-179.8(38)
H(4) -C(2') -C(1') - C(6)	1.4(16)	C(21S)-N(1S) -C(22S)-H(14)	60.9(46)
H(4) -C(2') -C(1') -C(6')	180.0(9)	C(21S)-N(1S) -C(22S)-H(15)	-57.6(48)

Table 3.12 Hydrogen bond contact distances

Hydrogen Bond	Donor-Acceptor Distance (Å)
$N(21)-H(1)_{I}...O(41)_{II}$	2.811
$N(1)-H$ $\text{predicted}_{I}...O(41)_{II}$	2.859
$N(21)-H(2)_{I}...N(3)_{III}$	2.949

The subscripts I, II and III refer to the equivalent positions X, Y, Z; X, 1+Y, Z; -X, 1-Y, Z.

Figure 3.3 ABPP.DMF Molecular plot and numbering scheme

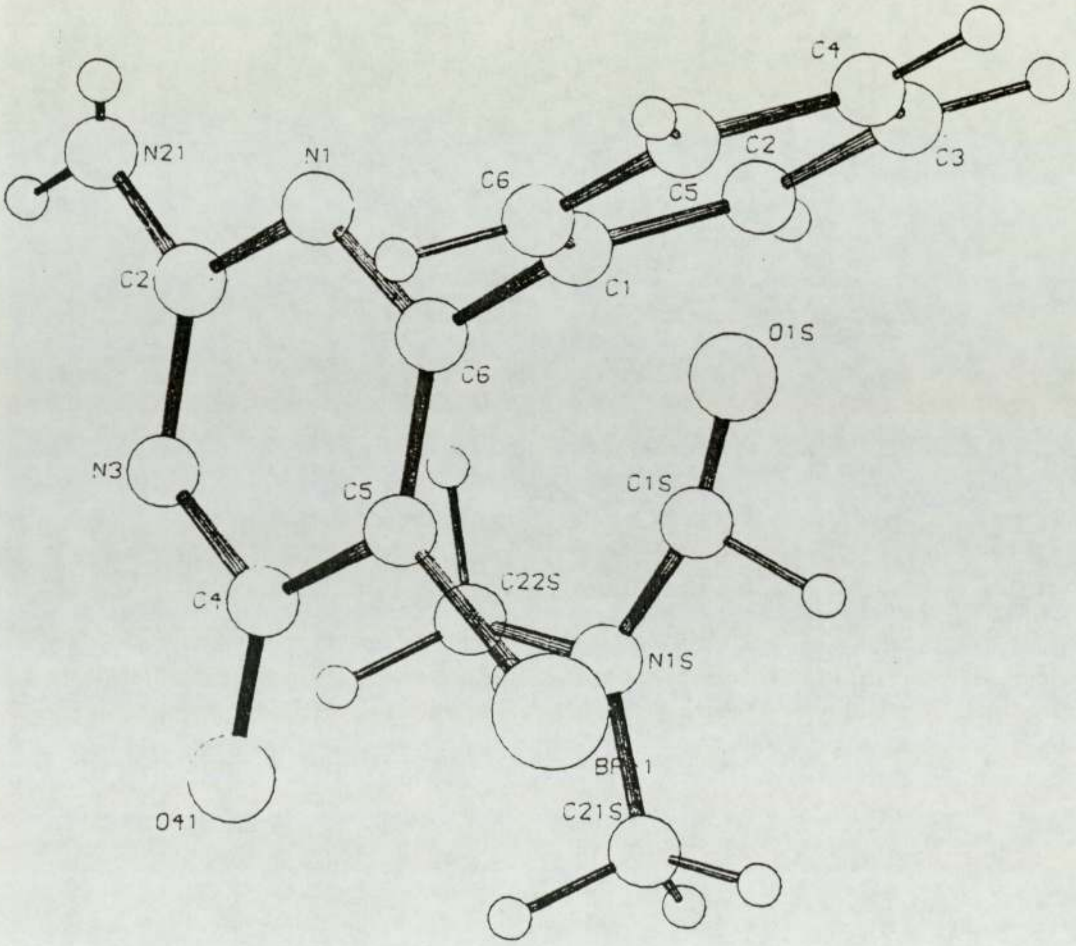
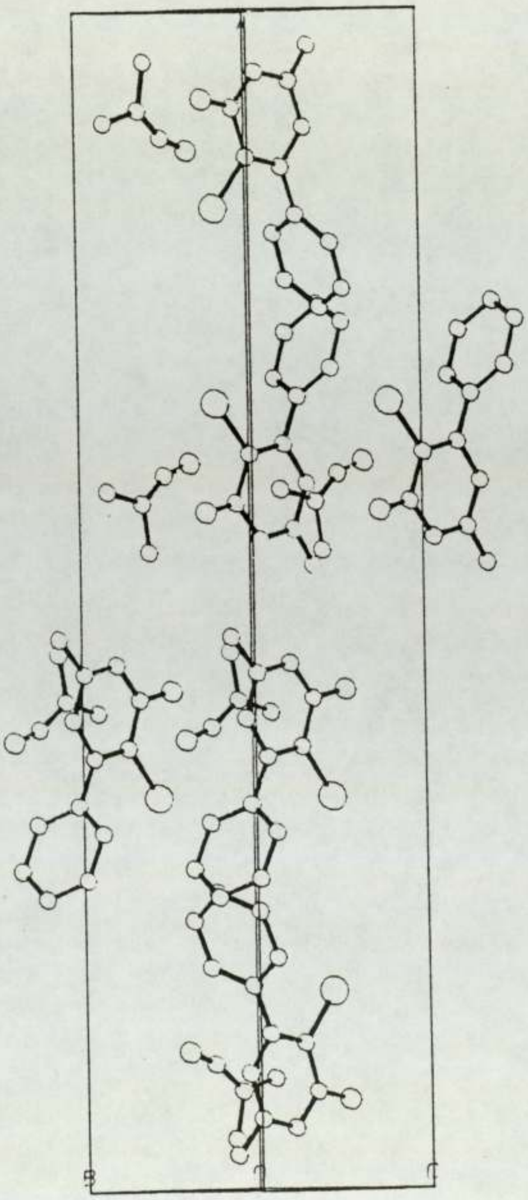


Figure 3.4 ABPP.DMF Unit cell packing diagram



3.4 Results and Discussion

The structure of ABPB.NMF provides much evidence for the existence of alternating 1H and 3H protonated forms. The hydrogen atoms in question were located in a difference electron density map. In molecule A the hydrogen atom uses up some of the valencey of N(3), and therefore on average the ring bonds to N(3) are longer than those to N(1) (Table 3.3); the reverse being the case in molecule B. Valance shell electron pair repulsion effects makes the internal ring angle smaller at an unprotonated nitrogen atom [N(1A) and N(3B)] compared with a protonated nitrogen atom where the relevant electrons are involved in bonding (Table 3.4). The pattern of tautomerism is consistent with the hydrogen bonding scheme (Table 3.6) showing triple base-pairing between 1H and 3H tautomers with the solvate molecule, NMF, bridging between base pairs, providing and extended hydrogen bonding pattern throughout the crystal.

Although hydrogen atoms are much less well observed in the structure of ABPP.DMF there is some evidence that suggests a 1H tautomer. The final difference map shows reasonable peaks near both N(1) and N(3), ring bond angles at N(1) and N(3) are essentially the same; but ring bonds to N(1) are significantly longer than those to N(3) (Table 3.9). The limited hydrogen bonding scheme (Table 3.12) provides the most conclusive proof of the tautomer. The vector from C(4) to O(41) lies close to the -b direction since the x and z values for the two atoms are nearly the same. Thus O(41) of the molecule translated by +b is able to accept hydrogen bonds from N(1) via the hydrogen atom it must bear, and from N(2) via H(1). The other H atom on the amino group, H(2), takes part in a hydrogen bond to N(3) of a nearby molecule. Unlike NMF the DMF molecule does not appear to make hydrogen bonds and this appears to be reflected in the quality of the structure obtained.

CHAPTER 4

CHAPTER 4

The Crystal Structures of two Azido-Substituted Antifolate Drugs: 2, 4-diamino-5 (4-chloro-3-azido) phenyl-6-ethyl pyrimidine ethanesulphonate salt (MZPES) and its 6-methyl analogue (MeMZPES).

4.1 Introduction

Lipophilic 2,4-diaminopyrimidines which inhibit the enzyme dihydrofolate reductase (DHFR) have found a limited clinical role in the treatment of methotrexate-resistant malignancies where resistance is mediated by modification of the active process which transports the polar drug into cells. Thus the lipophilic agents pyrimethamine, metoprin [2,4-diamino-5-(3,4-dichlorophenyl)-6-methyl pyrimidine] and etoprin achieve ingress to cells by passive diffusion and can accumulate in lipid compartments of the body, such as the brain. However, many have long biological half-lives, e.g. a plasma t $\frac{1}{2}$ of 216 h (85) for metoprin, resulting in slow clearance of any toxic effects which develop. The title compounds incorporate the lipophilic aromatic azido group which can be biotransformed metabolically to the corresponding polar aromatic amino group with the expectation of a relatively short biological half-life ($t\frac{1}{2}$). Thus these compounds should provide the desirable pharmacokinetic properties of lipophilic compounds whilst the biotransformability of the azide to the more polar amine may result in good cellular drug retention and also aid elimination.

4.2 The Crystal Structure of 2,4-diamino-5-(3-azido)-4-chlorophenyl)-6-ethylpyrimidine ethanesulphonate salt. (MZPES).

4.2.1 Experimental

A crystalline sample of the compound was obtained (76) and from it a suitable crystal was selected, the quality being estimated by its optical birefringent properties with a polarising microscope. The single crystal was then mounted onto a glass filament using epoxy adhesive, and the filament secured to a goniometer head stud by the hot wax method.

4.2.2 Crystal Data

$C_{12} H_{13} N_7 Cl.C_2 H_5 SO_3$, M = 399.861, F(000) = 1664. Monoclinic a = 9.233(3), b = 26.157(9), c = 18.872(5) Å, β = 125.51(5)°, V = 3710 $Å^3$ (by least squares refinement on diffractometer angles for 25 automatically centred reflections, λ = 0.71069 Å), space groups P 2_1/c, Z = 8, Dx = 1.432 gcm^{-3}, Dm = 1.44 gcm^{-3} (method of floatation in solvent mixture). White semi-opaque plates. Crystal dimensions, 0.6 x 0.4 x 0.1 mm (plane of plate 001), μ (Mo-Kα) = 2.99 cm^{-1}.

4.2.3 Data Collection and Processing

CAD4 diffractometer, w/2θ mode with w scan width = 1.00 + 0.35 tanθ, w scan speed 0.9 - 5.0 deg min^{-1}, graphite monochromated Mo-Kα radiation; 4868 reflections measured (2.0 $\geqslant \theta \leqslant$ 22.0° + h + k $\pm$ l), 4454 unique (merging R = 0.0261, no correction for absorption) giving 3220 with I $>$ 3 σ (I), number of parameters refined 237. Linear and approximately isotropic crystal decay ca. 10% (for 3 reference reflections, checked every 7200 s x-ray exposure time), corrected during processing.

4.2.4 Structure Determination and Refinement

The data were refined directly on coordinates obtained from a previous determination (78). Full matrix least squares refinement with all non-hydrogen atoms anisotropic did not give a converged structure. Difference Fourier analysis during isotropic refinement of non-hydrogen atoms located most of the hydrogen atom positions, which were included in anisotropic refinement of non-hydrogen atoms but not refined. The 3-azido group of the second molecule did not refine with acceptable bond lengths, C(3P')-N(1P')-N(2P')-N(3P'), so these were restrained, to the geometry found in the structure determinations of 6-methyl MZP (86), using the DFIX command, and refined isotropically. The weighting scheme $W = 5.3954/[\sigma^2 (F_o)$, with $\sigma(F_o)$ from counting statistics gave satisfactory agreement analyses. Convergence was achieved after 20 cycles of least squares refinement using a damping factor of 10, (Δ/σ) max in the final refinement cycle was 0.197 [N(3P') U11]. Final R and R_w values are 0.0972 and 0.1175. A final electron density synthesis showed features $\sim$1 e $Å^{-3}$ attributable to missing hydrogen atoms, after which there was no feature > 1/3 e $Å^{-3}$.

Table 4.1 Positional parameters (fractional coordinates * 10**4, x/a * 10**3) with estimated standard deviations in parentheses. Hydrogen positions unrefined.

ATOM	x/a	y/b	z/c
N(1)	355(1)	3762(3)	6460(5)
C(2)	221(1)	4067(4)	6298(7)
N(21)	184(1)	4061(4)	6867(6)
N(3)	134(1)	4367(3)	5631(5)
C(4)	176(1)	4356(4)	5059(7)
N(41)	080(1)	4644(4)	4351(6)
C(5)	326(1)	4060(4)	5221(7)
C(6)	411(1)	3763(4)	5931(7)
C(61)	568(2)	3427(5)	6188(7)
C(62)	514(2)	2913(6)	585(1)
C(1P)	385(1)	4110(4)	4628(6)
C(2P)	290(1)	3909(4)	3804(7)
C(3P)	350(1)	3973(4)	3279(6)
C(4P)	510(2)	4214(5)	3614(8)
C(5P)	605(2)	4399(5)	4418(9)
C(6P)	546(2)	4352(4)	4938(8)
CL(1)	587(5)	4297(2)	2985(3)
N(1P)	266(1)	3786(4)	2430(6)
N(2P)	135(2)	3515(5)	2158(7)
N(3P)	015(2)	3259(6)	1825(9)
S(1)	5760(4)	1692(1)	3714(2)
O(1)	711(1)	1365(3)	4351(5)
O(2)	584(1)	1754(3)	2989(5)
O(3)	403(1)	1522(3)	3464(5)
C(1E)	619(2)	2300(5)	4201(8)
C(2E)	489(2)	2685(5)	366(1)
N(1')	518(1)	4504(3)	0918(5)
C(2')	411(1)	4903(4)	0764(6)
N(21')	476(1)	5365(3)	0926(5)
N(3')	240(1)	4831(3)	0440(5)
C(4')	177(1)	4362(4)	0254(6)
N(41')	007(1)	4285(3)	-0031(5)
C(5')	282(1)	3918(4)	0347(6)
C(6')	458(1)	4016(4)	0715(6)
C(61')	591(1)	3615(4)	0927(8)
C(62')	686(2)	3452(5)	1891(8)
C(1P')	201(1)	3397(4)	0060(7)
C(2P')	138(1)	3224(5)	-0748(7)
C(3P')	063(2)	2741(5)	-0989(7)
C(4P')	052(2)	2444(4)	-044(1)
C(5P')	114(2)	2572(5)	0401(9)
C(6P')	190(1)	3068(5)	0607(7)
CL(1')	-0432(5)	1846(1)	-0817(3)
N(1P')	007(2)	2548(5)	-1806(8)
N(2P')	005(2)	2823(6)	-2349(9)
N(3P')	-000(3)	3005(8)	-291(1)
S(1')	0279(4)	0103(1)	2854(2)
O(1')	1132(9)	-0395(3)	3071(5)
O(2')	-068(1)	0218(3)	1931(4)
O(3')	1503(9)	0508(3)	3376(5)
C(1E')	-134(1)	0046(4)	3081(6)
C(2E')	-259(2)	0517(5)	2762(8)

Table 4.1 contd.

ATOM	x/a	y/b	z/c
H(1)	4243	3605	6948
H(2)	0961	4294	6828
H(3)	2306	3928	7376
H(4)	1004	4749	3978
H(5)	0146	4876	4382
H(6)	6327	3312	6825
H(11)	1628	3748	3580
H(12)	7226	4493	4519
H(13)	6218	4530	5526
H(14)	5955	2281	4667
H(16)	5494	2978	4032
H(1′)	6472	4581	1166
H(2′)	4073	5684	0773
H(3′)	6052	5403	1134
H(4′)	-0450	3942	- 0202
H(5′)	-0660	4620	- 0001
H(6′)	5261	3360	0396
H(7′)	6708	3592	0730
H(9′)	5865	3408	2003
H(10′)	7849	3240	1964
H(11′)	1885	3358	- 1056
H(13′)	2168	3164	1167
H(14′)	-1016	0068	3639

Table 4.2 Anisotropic temperature factors, and isotropic temperature factors where shown; with standard deviations in parentheses.

ATOM	U11	U22	U33	U23	U13	U12
N(1)	.042(5)	.070(6)	.041(5)	.013(5)	.026(5)	.017(5)
C(2)	.041(7)	.053(7)	.041(7)	.012(6)	.025(6)	.007(6)
N(21)	.062(6)	.094(8)	.055(6)	.028(5)	.041(5)	.035(6)
N(3)	.043(5)	.055(6)	.047(6)	.002(5)	.031(5)	.006(5)
C(4)	.036(6)	.048(7)	.044(6)	.003(5)	.021(6)	-.003(5)
N(41)	.057(6)	.068(6)	.060(6)	.013(5)	.044(5)	.018(5)
C(5)	.032(6)	.057(7)	.049(7)	-.008(6)	.026(6)	.000(5)
C(6)	.036(6)	.049(7)	.044(6)	.006(6)	.015(6)	.006(5)
C(61)	.055(7)	.071(8)	.049(7)	.017(6)	.027(6)	.026(6)
C(62)	.072(9)	.067(9)	.20(1)	-.053(9)	.04(1)	.011(8)
C(1P)	.045(7)	.050(7)	.034(6)	.002(5)	.019(6)	.013(6)
C(2P)	.044(7)	.063(8)	.058(7)	-.001(6)	.032(6)	.005(6)
C(3P)	.055(7)	.058(8)	.047(7)	-.009(6)	.032(6)	.008(6)
C(4P)	.060(8)	.079(9)	.054(7)	-.013(6)	.038(7)	-.001(7)
C(5P)	.058(8)	.078(9)	.095(9)	-.009(7)	.056(8)	-.006(7)
C(6P)	.055(8)	.061(8)	.068(8)	-.009(6)	.040(7)	-.008(6)
CL(1)	.101(3)	.151(4)	.106(3)	.002(3)	.084(3)	-.001(3)
N(1P)	.065(7)	.104(9)	.066(7)	-.022(6)	.037(6)	-.003(6)
N(2P)	.059(8)	.15(1)	.065(8)	-.022(7)	.039(7)	.005(8)
N(3P)	.074(9)	.23(1)	.13(1)	-.06(1)	.062(8)	-.030(9)
S(1)	.044(2)	.051(2)	.056(2)	-.008(2)	.024(2)	-.000(2)
O(1)	.051(5)	.074(6)	.105(7)	.008(5)	.031(5)	.019(5)
O(2)	.099(7)	.107(7)	.069(6)	-.028(5)	.054(5)	-.039(6)
O(3)	.043(5)	.086(6)	.079(6)	-.017(5)	.031(4)	-.019(4)
C(1E)	.082(9)	.055(8)	.099(9)	-.017(7)	.057(8)	-.005(7)
C(2E)	.12(1)	.038(9)	.22(1)	-.003(9)	.06(1)	.019(9)
N(1′)	.030(5)	.041(6)	.049(5)	-.005(4)	.021(4)	.000(4)
C(2′)	.038(7)	.041(7)	.040(6)	.002(5)	.020(5)	.005(6)
N(21′)	.030(5)	.046(6)	.070(6)	-.002(5)	.025(5)	-.003(4)
N(3′)	.028(5)	.038(5)	.045(5)	.001(4)	.018(4)	-.002(4)
C(4′)	.038(7)	.049(7)	.034(6)	.004(5)	.021(5)	.008(6)
N(41′)	.030(5)	.042(6)	.063(6)	-.002(4)	.024(4)	.003(4)
C(5′)	.038(6)	.043(6)	.035(6)	-.005(5)	.022(5)	-.000(5)
C(6′)	.039(7)	.054(7)	.042(6)	.004(5)	.025(5)	.011(6)
C(61′)	.047(7)	.059(8)	.081(8)	-.011(6)	.040(6)	.012(6)
C(62′)	.055(8)	.11(1)	.082(8)	.048(8)	.043(7)	.052(7)
C(1P′)	.028(6)	.045(7)	.057(7)	.002(6)	.025(5)	.010(5)
C(2P′)	.043(7)	.073(8)	.046(7)	-.022(6)	.025(6)	-.003(6)
C(3P′)	.052(7)	.078(9)	.051(7)	-.043(7)	.017(6)	-.004(7)
C(4P′)	.059(8)	.033(7)	.13(1)	.023(7)	.067(8)	.008(6)
C(5P′)	.048(8)	.10(1)	.087(9)	-.053(8)	.025(7)	.009(7)
C(6P′)	.050(7)	.068(8)	.052(7)	-.007(6)	.034(6)	.005(6)
CL(1′)	.079(3)	.060(2)	.140(3)	-.019(2)	.055(3)	-.015(2)

	U ISO
N(1P′)	.135(5)
N(2P′)	.150(6)
N(3P′)	.227(9)

S(1′)	.034(2)	.066(2)	.051(2)	-.006(2)	.027(2)	-.003(2)
O(1′)	.039(5)	.072(6)	.088(6)	-.008(5)	.027(4)	.007(4)

Table 4.2 contd.

ATOM	U11	U22	U33	U23	U13	U12
O(2′)	.066(5)	.098(6)	.043(4)	-.002(4)	.040(4)	-.002(5)
O(3′)	.051(5)	.071(5)	.072(5)	-.019(4)	.038(4)	-.018(4)
C(1E′)	.047(7)	.085(8)	.041(6)	.002(6)	.033(6)	.005(6)
C(2E′)	.056(8)	.094(9)	.072(8)	.023(7)	.036(7)	.041(5)

Table 4.3 Bond Lengths(A) with standard deviations

N(1) - C(2)	1.349(15)
N(1) - C(6)	1.361(15)
N(1) - H(1)	0.861(9)
C(2) -N(21)	1.303(17)
C(2) - N(3)	1.293(15)
N(21) - H(2)	0.984(11)
N(21) - H(3)	0.865(11)
N(3) - C(4)	1.341(15)
C(4) -N(41)	1.328(16)
C(4) - C(5)	1.457(17)
N(41) - H(4)	0.875(11)
N(41) - H(5)	0.879(11)
C(5) - C(6)	1.339(17)
C(5) -C(1P)	1.507(17)
C(6) -C(61)	1.519(18)
C(61) -C(62)	1.448(23)
C(61) - H(6)	1.029(13)
C(1P) -C(2P)	1.371(17)
C(1P) -C(6P)	1.395(18)
C(2P) -C(3P)	1.399(17)
C(2P) -H(11)	1.077(12)
C(3P) -C(4P)	1.376(19)
C(3P) -N(1P)	1.401(16)
C(4P) -C(5P)	1.326(21)
C(4P) -Cl(1)	1.716(15)
C(5P) -C(6P)	1.376(20)
C(5P) -H(12)	1.018(15)
C(6P) -H(13)	1.018(14)
N(1P) -N(2P)	1.233(18)
N(2P) -N(3P)	1.120(21)
S(1) - O(1)	1.411(10)
S(1) - O(2)	1.424(10)
S(1) - O(3)	1.448(10)
S(1) -C(1E)	1.763(15)
C(1E) -C(2E)	1.44(3)
C(1E) -H(14)	1.021(15)
C(2E) -H(16)	0.971(21)
N(1') -C(2')	1.348(15)
N(1') -C(6')	1.356(14)
N(1') -H(1')	1.014(9)
C(2') -N(21')	1.303(15)
C(2') -N(3')	1.335(15)
N(21')-H(2')	0.984(9)
N(21')-H(3')	1.023(9)
N(3') -C(4')	1.314(14)
C(4') -N(41')	1.354(15)
C(4') -C(5')	1.453(16)
N(41')-H(4')	0.978(9)
N(41')-H(5')	1.124(9)
C(5') -C(6')	1.370(16)
C(5') -C(1P')	1.495(16)
C(6') -C(61')	1.482(17)
C(61')-C(62')	1.551(19)
C(61')-H(6')	1.054(13)
C(61')-H(7')	1.004(13)
C(62')-H(9')	1.058(14)
C(62')-H(10')	1.010(14)
C(1P')-C(2P')	1.354(18)
C(1P')-C(6P')	1.393(18)
C(2P')-C(3P')	1.382(19)
C(2P')-H(11')	0.999(13)
C(3P')-C(4P')	1.351(20)
C(3P')-N(1P')	1.404(20)
C(4P')-C(5P')	1.375(21)
C(4P')-Cl(1')	1.733(16)
C(5P')-C(6P')	1.418(20)
C(6P')-H(13')	0.969(13)
N(1P')-N(2P')	1.245(24)
N(2P')-N(3P')	1.13(3)
S(1') -O(1')	1.453(9)
S(1') -O(2')	1.455(9)
S(1') -O(3')	1.441(9)
S(1') -C(1E')	1.786(12)
C(1E')-C(2E')	1.548(19)
C(1E')-H(14')	0.914(12)

Table 4.4 Angles(degrees) with standard deviations

C(2) - N(1) - C(6)	121.2(10)	C(2') -N(1') -C(6')	122.5(10)
C(2) - N(1) - H(1)	122.5(10)	C(2') -N(1') -H(1')	117.6(9)
C(6) - N(1) - H(1)	115.1(10)	C(6') -N(1') -H(1')	119.8(9)
N(1) - C(2) -N(21)	116.8(11)	N(1') -C(2') -N(21')	119.3(10)
N(1) - C(2) - N(3)	123.3(11)	N(1') -C(2') -N(3')	120.9(10)
N(21) - C(2) - N(3)	119.8(11)	N(21')-C(2') -N(3')	119.8(10)
C(2) -N(21) - H(2)	122.1(11)	C(2') -N(21')-H(2')	126.1(10)
C(2) -N(21) - H(3)	136.1(12)	C(2') -N(21')-H(3')	117.0(9)
H(2) -N(21) - H(3)	100.8(11)	H(2') -N(21')-H(3')	116.0(9)
C(2) - N(3) - C(4)	117.7(10)	C(2') -N(3') -C(4')	118.3(10)
N(3) - C(4) -N(41)	117.4(11)	N(3') -C(4') -N(41')	118.4(10)
N(3) - C(4) - C(5)	121.7(11)	N(3') -C(4') -C(5')	123.7(10)
N(41) - C(4) - C(5)	120.8(11)	N(41')-C(4') -C(5')	117.9(10)
C(4) -N(41) - H(4)	131.0(12)	C(4') -N(41')-H(4')	119.9(9)
C(4) -N(41) - H(5)	114.2(11)	C(4') -N(41')-H(5')	117.0(9)
H(4) -N(41) - H(5)	108.3(12)	H(4') -N(41')-H(5')	122.8(9)
C(4) - C(5) - C(6)	116.6(11)	C(4') -C(5') -C(6')	114.8(10)
C(4) - C(5) -C(1P)	120.5(10)	C(4') -C(5') -C(1P')	122.0(10)
C(6) - C(5) -C(1P)	122.8(11)	C(6') -C(5') -C(1P')	123.2(10)
N(1) - C(6) - C(5)	119.1(11)	N(1') -C(6') -C(5')	119.5(10)
N(1) - C(6) -C(61)	117.6(10)	N(1') -C(6') -C(61')	116.7(10)
C(5) - C(6) -C(61)	123.2(11)	C(5') -C(6') -C(61')	123.8(11)
C(6) -C(61) -C(62)	112.2(12)	C(6') -C(61')-C(62')	109.0(10)
C(6) -C(61) - H(6)	109.8(11)	C(6') -C(61')-H(6')	105.6(11)
C(62) -C(61) - H(6)	93.6(12)	C(6') -C(61')-H(7')	128.0(12)
C(5) -C(1P) -C(2P)	122.8(11)	C(62')-C(61')-H(6')	124.3(12)
C(5) -C(1P) -C(6P)	119.0(11)	C(62')-C(61')-H(7')	112.5(11)
C(2P) -C(1P) -C(6P)	118.2(11)	H(6') -C(61')-H(7')	75.0(9)
C(1P) -C(2P) -C(3P)	120.2(11)	C(61')-C(62')-H(9')	107.4(11)
C(1P) -C(2P) -H(11)	115.3(11)	C(61')-C(62')-H(10')	99.2(11)
C(3P) -C(2P) -H(11)	124.1(11)	H(9') -C(62')-H(10')	137.3(14)
C(2P) -C(3P) -C(4P)	119.4(11)	C(5') -C(1P')-C(2P')	120.8(11)
C(2P) -C(3P) -N(1P)	125.7(11)	C(5') -C(1P')-C(6P')	122.0(11)
C(4P) -C(3P) -N(1P)	114.8(11)	C(2P')-C(1P')-C(6P')	117.2(11)
C(3P) -C(4P) -C(5P)	120.8(13)	C(1P')-C(2P')-C(3P')	118.9(12)
C(3P) -C(4P) -Cl(1)	120.4(11)	C(1P')-C(2P')-H(11')	119.6(12)
C(5P) -C(4P) -Cl(1)	118.8(12)	C(3P')-C(2P')-H(11')	116.1(12)
C(4P) -C(5P) -C(6P)	120.8(14)	C(2P')-C(3P')-C(4P')	121.2(13)
C(4P) -C(5P) -H(12)	103.0(13)	C(2P')-C(3P')-N(1P')	119.1(13)
C(6P) -C(5P) -H(12)	135.1(15)	C(4P')-C(3P')-N(1P')	119.6(13)
C(1P) -C(6P) -C(5P)	120.6(13)	C(3P')-C(4P')-C(5P')	125.5(14)
C(1P) -C(6P) -H(13)	123.7(13)	C(3P')-C(4P')-Cl(1')	116.6(11)
C(5P) -C(6P) -H(13)	115.5(13)	C(5P')-C(4P')-Cl(1')	117.9(12)
C(3P) -N(1P) -N(2P)	115.7(12)	C(4P')-C(5P')-C(6P')	110.3(13)
N(1P) -N(2P) -N(3P)	171.5(16)	C(1P')-C(6P')-C(5P')	126.9(12)
O(1) - S(1) - O(2)	111.9(6)	C(1P')-C(6P')-H(13')	124.5(13)

O(1) - S(1) - O(3)	110.7(6)	C(5P')-C(6P')-H(13')	107.9(12)
O(1) - S(1) -C(1E)	106.5(6)	C(3P')-N(1P')-N(2P')	121.0(15)
O(2) - S(1) - O(3)	112.8(6)	N(1P')-N(2P')-N(3P')	169.5(22)
O(2) - S(1) -C(1E)	106.1(6)	O(1') -S(1') -O(2')	112.1(5)
O(3) - S(1) -C(1E)	108.5(6)	O(1') -S(1') -O(3')	112.6(5)
S(1) -C(1E) -C(2E)	114.6(12)	O(1') -S(1') -C(1E')	104.5(5)
S(1) -C(1E) -H(14)	108.0(11)	O(2') -S(1') -O(3')	110.9(5)
C(2E) -C(1E) -H(14)	98.3(14)	O(2') -S(1') -C(1E')	106.9(5)
C(1E) -C(2E) -H(16)	98.2(17)	O(3') -S(1') -C(1E')	109.3(5)
S(1') -C(1E')-C(2E')	112.1(9)	C(2E')-C(1E')-H(14')	94.1(10)
S(1') -C(1E')-H(14')	120.7(10)		

Table 4.5 Torsion angles(degrees) with standard deviations

C(6) - N(1) - C(2) -N(21)	-176.2(11)	C(3P) -C(4P) -C(5P) -H(12)	170.3(13)
C(6) - N(1) - C(2) - N(3)	1.8(18)	Cl(1) -C(4P) -C(5P) -C(6P)	178.4(11)
H(1) - N(1) - C(2) -N(21)	-9.3(18)	Cl(1) -C(4P) -C(5P) -H(12)	-11.6(17)
H(1) - N(1) - C(2) - N(3)	168.8(11)	C(4P) -C(5P) -C(6P) -C(1P)	-0.2(22)
C(2) - N(1) - C(6) - C(5)	-2.3(17)	C(4P) -C(5P) -C(6P) -H(13)	-175.2(14)
C(2) - N(1) - C(6) -C(61)	177.0(11)	H(12) -C(5P) -C(6P) -C(1P)	-166.3(16)
H(1) - N(1) - C(6) - C(5)	-170.2(11)	H(12) -C(5P) -C(6P) -H(13)	18.8(27)
H(1) - N(1) - C(6) -C(61)	9.1(16)	C(3P) -N(1P) -N(2P) -N(3P)	-173.7(**)
N(1) - C(2) -N(21) - H(2)	174.3(11)	O(1) - S(1) -C(1E) -C(2E)	-177.5(12)
N(1) - C(2) -N(21) - H(3)	7.4(23)	O(1) - S(1) -C(1E) -H(14)	-69.0(12)
N(3) - C(2) -N(21) - H(2)	-3.8(19)	O(2) - S(1) -C(1E) -C(2E)	63.2(14)
N(3) - C(2) -N(21) - H(3)	-170.7(14)	O(2) - S(1) -C(1E) -H(14)	171.6(10)
N(1) - C(2) - N(3) - C(4)	2.6(17)	O(3) - S(1) -C(1E) -C(2E)	-58.3(14)
N(21) - C(2) - N(3) - C(4)	-179.4(11)	O(3) - S(1) -C(1E) -H(14)	50.2(12)
C(2) - N(3) - C(4) -N(41)	176.4(11)	S(1) -C(1E) -C(2E) -H(16)	-173.1(12)
C(2) - N(3) - C(4) - C(5)	-6.4(17)	H(14) -C(1E) -C(2E) -H(16)	72.6(17)
N(3) - C(4) -N(41) - H(4)	162.7(13)	C(6') -N(1') -C(2') -N(21')	-175.8(10)
N(3) - C(4) -N(41) - H(5)	14.9(17)	C(6') -N(1') -C(2') -N(3')	3.4(17)
C(5) - C(4) -N(41) - H(4)	-14.6(22)	H(1') -N(1') -C(2') -N(21')	0.9(15)
C(5) - C(4) -N(41) - H(5)	-162.3(11)	H(1') -N(1') -C(2') -N(3')	-179.8(9)
N(3) - C(4) - C(5) - C(6)	5.8(17)	C(2') -N(1') -C(6') -C(5')	-0.7(16)
N(3) - C(4) - C(5) -C(1P)	-171.8(11)	C(2') -N(1') -C(6') -C(61')	-179.6(10)
N(41) - C(4) - C(5) - C(6)	-177.1(12)	H(1') -N(1') -C(6') -C(5')	-177.3(10)
N(41) - C(4) - C(5) -C(1P)	5.3(18)	H(1') -N(1') -C(6') -C(61')	3.8(15)
C(4) - C(5) - C(6) - N(1)	-1.3(17)	N(1') -C(2') -N(21')-H(2')	172.0(10)
C(4) - C(5) - C(6) -C(61)	179.4(11)	N(1') -C(2') -N(21')-H(3')	3.3(15)
C(1P) - C(5) - C(6) - N(1)	176.2(10)	N(3') -C(2') -N(21')-H(2')	-7.3(17)
C(1P) - C(5) - C(6) -C(61)	-3.1(19)	N(3') -C(2') -N(21')-H(3')	-175.9(9)
C(4) - C(5) -C(1P) -C(2P)	-73.0(16)	N(1') -C(2') -N(3') -C(4')	-1.7(16)
C(4) - C(5) -C(1P) -C(6P)	109.7(14)	N(21')-C(2') -N(3') -C(4')	177.6(10)
C(6) - C(5) -C(1P) -C(2P)	109.5(15)	C(2') -N(3') -C(4') -N(41')	176.6(10)

C(6) - C(5) -C(1P) -C(6P)	-67.8(16)
N(1) - C(6) -C(61) -C(62)	87.9(15)
N(1) - C(6) -C(61) - H(6)	-14.8(16)
C(5) - C(6) -C(61) -C(62)	-92.8(16)
C(5) - C(6) -C(61) - H(6)	164.5(12)
C(5) -C(1P) -C(2P) -C(3P)	179.2(11)
C(5) -C(1P) -C(2P) -H(11)	6.5(18)
C(6P) -C(1P) -C(2P) -C(3P)	-3.5(18)
C(6P) -C(1P) -C(2P) -H(11)	-176.2(11)
C(5) -C(1P) -C(6P) -C(5P)	179.2(12)
C(5) -C(1P) -C(6P) -H(13)	-6.3(20)
C(2P) -C(1P) -C(6P) -C(5P)	1.8(19)
C(2P) -C(1P) -C(6P) -H(13)	176.3(13)
C(1P) -C(2P) -C(3P) -C(4P)	3.6(18)
C(1P) -C(2P) -C(3P) -N(1P)	179.8(12)
H(11) -C(2P) -C(3P) -C(4P)	175.7(12)
H(11) -C(2P) -C(3P) -N(1P)	-8.1(20)
C(2P) -C(3P) -C(4P) -C(5P)	-2.0(20)
C(2P) -C(3P) -C(4P) -Cl(1)	179.9(10)
N(1P) -C(3P) -C(4P) -C(5P)	-178.6(13)
N(1P) -C(3P) -C(4P) -Cl(1)	3.3(17)
C(2P) -C(3P) -N(1P) -N(2P)	-5.0(19)
C(4P) -C(3P) -N(1P) -N(2P)	171.4(13)
C(3P) -C(4P) -C(5P) -C(6P)	0.3(23)
C(6’) -C(61’)-C(62’)-H(10’)	-169.3(11)
H(6’) -C(61’)-C(62’)-H(9’)	-80.9(16)
H(6’) -C(61’)-C(62’)-H(10’)	65.3(16)
H(7’) -C(61’)-C(62’)-H(9’)	-167.6(11)
H(7’) -C(61’)-C(62’)-H(10’)	-21.4(15)
C(5’) -C(1P’)-C(2P’)-C(3P’)	-179.7(11)
C(5’) -C(1P’)-C(2P’)-H(11’)	27.1(19)
C(6P’)-C(1P’)-C(2P’)-C(3P’)	1.0(18)
C(6P’)-C(1P’)-C(2P’)-H(11’)	-152.2(12)
C(5’) -C(1P’)-C(6P’)-C(5P’)	178.7(13)
C(5’) -C(1P’)-C(6P’)-H(13’)	9.0(21)
C(2P’)-C(1P’)-C(6P’)-C(5P’)	-1.9(20)
C(2P’)-C(1P’)-C(6P’)-H(13’)	-171.6(13)
C(1P’)-C(2P’)-C(3P’)-C(4P’)	0.6(20)
C(1P’)-C(2P’)-C(3P’)-N(1P’)	-176.2(13)
H(11’)-C(2P’)-C(3P’)-C(4P’)	154.8(13)
H(11’)-C(2P’)-C(3P’)-N(1P’)	-22.1(20)
C(2’) -N(3’) -C(4’) -C(5’)	-2.6(16)
N(3’) -C(4’) -N(41’)-H(4’)	176.9(9)
N(3’) -C(4’) -N(41’)-H(5’)	-8.5(15)
C(5’) -C(4’) -N(41’)-H(4’)	-3.9(16)
C(5’) -C(4’) -N(41’)-H(5’)	170.7(9)
N(3’) -C(4’) -C(5’) -C(6’)	5.1(17)
N(3’) -C(4’) -C(5’) -C(1P’)	-174.9(11)
N(41’)-C(4’) -C(5’) -C(6’)	-174.1(10)
N(41’)-C(4’) -C(5’) -C(1P’)	5.9(16)
C(4’) -C(5’) -C(6’) -N(1’)	-3.2(16)
C(4’) -C(5’) -C(6’) -C(61’)	175.6(11)
C(1P’)-C(5’) -C(6’) -N(1’)	176.8(10)
C(1P’)-C(5’) -C(6’) -C(61’)	-4.4(18)
C(4’) -C(5’) -C(1P’)-C(2P’)	92.9(15)
C(4’) -C(5’) -C(1P’)-C(6P’)	-87.9(15)
C(6’) -C(5’) -C(1P’)-C(2P’)	-87.2(15)
C(6’) -C(5’) -C(1P’)-C(6P’)	92.1(15)
N(1’) -C(6’) -C(61’)-C(62’)	84.7(13)
N(1’) -C(6’) -C(61’)-H(6’)	-139.7(11)
N(1’) -C(6’) -C(61’)-H(7’)	-56.7(18)
C(5’) -C(6’) -C(61’)-C(62’)	-94.2(14)
C(5’) -C(6’) -C(61’)-H(6’)	41.5(16)
C(5’) -C(6’) -C(61’)-H(7’)	124.4(15)
C(6’) -C(61’)-C(62’)-H(9’)	44.5(14)
C(2P’)-C(3P’)-C(4P’)-C(5P’)	-1.6(24)
C(2P’)-C(3P’)-C(4P’)-Cl(1’)	-179.9(11)
N(1P’)-C(3P’)-C(4P’)-C(5P’)	175.2(15)
N(1P’)-C(3P’)-C(4P’)-Cl(1’)	-3.1(19)
C(2P’)-C(3P’)-N(1P’)-N(2P’)	-7.6(23)
C(4P’)-C(3P’)-N(1P’)-N(2P’)	175.5(16)
C(3P’)-C(4P’)-C(5P’)-C(6P’)	0.8(22)
Cl(1’)-C(4P’)-C(5P’)-C(6P’)	179.0(10)
C(4P’)-C(5P’)-C(6P’)-C(1P’)	1.0(21)
C(4P’)-C(5P’)-C(6P’)-H(13’)	172.1(13)
C(3P’)-N(1P’)-N(2P’)-N(3P’)	168.9(**)
O(1’) -S(1’) -C(1E’)-C(2E’)	-171.6(9)
O(1’) -S(1’) -C(1E’)-H(14’)	79.1(12)
O(2’) -S(1’) -C(1E’)-C(2E’)	-52.6(10)
O(2’) -S(1’) -C(1E’)-H(14’)	-161.9(10)
O(3’) -S(1’) -C(1E’)-C(2E’)	67.6(10)
O(3’) -S(1’) -C(1E’)-H(14’)	-41.7(12)

Table 4.6 Hydrogen bond contact distances.

Hydrogen Bond	Donor-Acceptor Distance (Å)
N(41')-H(5')$_{I}$...N(3')$_{II}$	3.014
N(1)-H(1)$_{I}$...O(2)$_{III}$	2.745
N(1')-H(1')$_{I}$...O(1')$_{IV}$	2.783
N(41)-H(4)$_{I}$...O(2')$_{V}$	2.797
N(41')-H(4')$_{I}$...O(1)$_{VI}$	2.827
N(21)-H(3)$_{I}$...O(3)$_{III}$	2.901
N(21')-H(3')$_{I}$...O(3')$_{IV}$	2.917
N(21')-H(2')$_{I}$...O(1)$_{IV}$	3.008
N(21)-H(2)$_{I}$...O(2')$_{III}$	3.054

The subscripts I, II, $_{III}$, IV, V and VI refer to the equivalent positions

X, Y, Z.
-X, 1-Y, -Z.
X, ½-Y, ½+Y, ½+Z.
1-X, ½+Y, ½-Z.
-X, ½+Y, ½-Z.
-1+X, ½-Y, -½+Z.

Figure 4.1 MZPES Molecular plot and numbering scheme

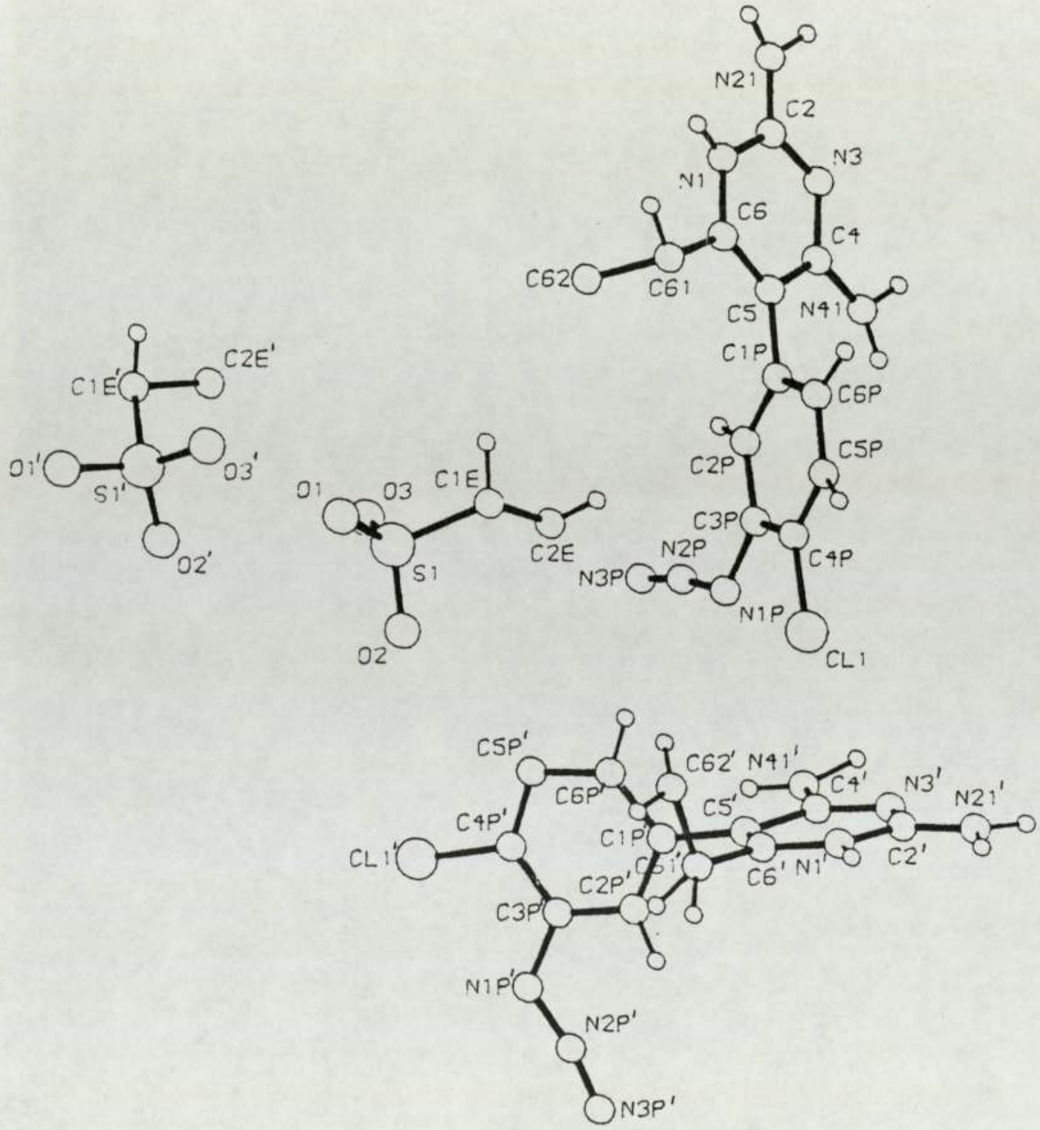
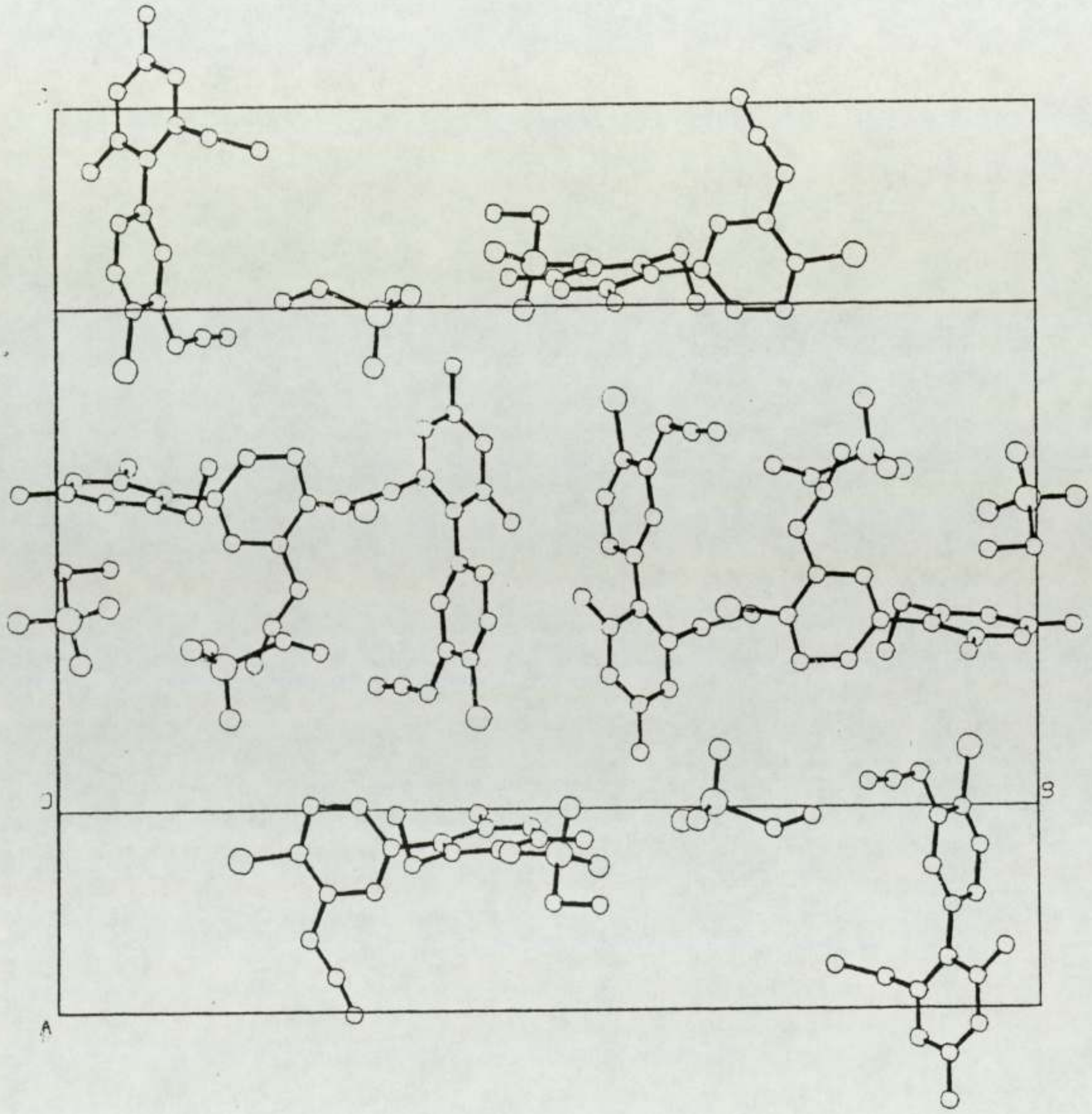


Figure 4.2 MZPES Unit cell packing diagram



4.3 The Crystal Structure of 2,4-diamino-5-(3-azido)-4-chlorophenyl)-6-methyl pyrimidine ethane sulphonate salt (6-MeMZPES).

4.3.1 Experimental

A crystalline sample of the compound was obtained (76) and from it suitable crystals were selected, their quality being estimated by optical bi-refringent properties, revealed by examination with a polarising microscope. Crystals were mounted on glass filaments using epoxy adhesive; the filament then mounted on goniometer-head studs by the hot wax technique. Crystal decay due to x-ray exposure neccessitated the use of two crystals to collect the appropriate data.

4.3.2 Crystal Data

$C_{11}\,H_{11}\,N_7\,Cl.\;C_2\,H_5\,SO_3$, M = 385.834, F(000) = 400, Triclinic a = 7.422(2), b = 10.569(2), c = 12.808(4) Å, α = 67.04(2), β = 80.75(2), γ = 76.49(2)° V = 897 $Å^3$ (by least squares refinement on diffractometer angles for 25 automatically centred reflections, λ = 0.71069 Å), space group $P\bar{1}$, Z = 2, D_x = 1.429 gcm^{-3}, Dm = 1.429 gcm^{-3} (method of flotation in solvent mixture). White rhombic plates, crystal (I) 0.80 x 0.44 x 0.04 mm, crystal (II) 0.45 x 0.35 x 0.08 mm, μ (Mo-Kα) = 3.08 cm^{-1}.

4.3.3 Data Collection and Processing

CAD4 diffractometer, w/2θ mode with w scan width = 1.35 + 0.35 tanθ (I), 1.20 = 0.35 tanθ (II), w scan speed 1 - 33°

min^{-1} (I and II), graphite monochromated Mo-Kα radiation; 2929 (I) and 3166 (II) reflections measured ($2.0 \leqslant \Theta \leqslant 25°$ + h $\pm$ k $\pm$ l), 3180 unique (merging R = 0.0860, no correction for absorption) after deriving and applying batch scale factors, for two sets of data, using common reflections and the linear least-squares method of Rae and Blake (87), giving 2215 with $I > 3\sigma(I)$, number of parameters refined 290. Linear and approximately isotropic crystal decay ca. 40% (I) - Ca.12% (II) (for 3 reference reflections, checked every 7200 s x-ray exposure time), corrected during processing.

4.3.4 Structure Determination and Refinement

Data reduction followed by direct methods (SHELX76) and electron density synthesis. All hydrogens located by difference Fourier analysis during isotropic least squares refinement of non-hydrogen atoms. Full matrix least squares refinement with all non-hydrogen atoms anisotropic gave convergence in 10 cycles. The weighting scheme $W = 1.4978/[\sigma^2(F_o) + 0.001\ F_o^{\ 2}]$, with $\sigma(F_o)$ from counting statistics, gave satisfactory agreement analyses. Final R and R_w values are 0.0619 and 0.0730. A final electron density synthesis showed a number of peaks ~ 0.3 e $Å^{-3}$ all found to be associated with either S(1), Cl (4') or oxygens of the ethane sulphonate counter ion, as ascribed to anisotropy in these regions or lone pairs; after these no feature in the map exceeded 0.3 $eÅ^{-3}$. At convergence (Δ/σ) max = 0.014 [H(7) Z/C].

Table 4.7 Positional parameters (fractional coordinates * 10**4, hydrogens * 10**3) with estimated standard deviations in parentheses.

ATOM	x/a	y/b	z/c
N(1)	5220(4)	1745(3)	3821(3)
C(2)	5977(5)	2279(4)	4410(3)
N(21)	7529(5)	1525(4)	4904(4)
N(3)	5231(4)	3489(3)	4518(3)
C(4)	3684(4)	4217(4)	3987(3)
N(41)	2998(5)	5439(4)	4083(3)
C(5)	2874(5)	3728(4)	3310(3)
C(6)	3668(5)	2474(4)	3254(3)
C(61)	2994(7)	1765(5)	2632(5)
C(1′)	1225(5)	4621(4)	2665(3)
C(2′)	1464(5)	5293(4)	1511(3)
C(3′)	-0031(5)	6121(4)	0877(3)
N(3′1)	0128(6)	6836(5)	-0315(3)
N(3′2)	1690(7)	6703(5)	-0789(4)
N(3′3)	3023(9)	6679(7)	-1318(5)
C(4′)	-1772(5)	6254(4)	1443(4)
CL(4′)	-3666(2)	7263(2)	0685(1)
C(5′)	-2000(6)	5611(5)	2594(5)
C(6′)	-0514(5)	4796(4)	3210(4)
S(1)	8786(1)	-1046(1)	3369(1)
O(1)	6795(4)	-0874(3)	3610(3)
O(2)	9493(4)	0035(4)	3529(3)
O(3)	9794(4)	-2423(3)	3964(3)
C(21)	9256(9)	-0754(7)	1903(5)
C(22)	1128(1)	-088(1)	1533(8)
H(1)	577(6)	097(5)	371(4)
H(2)	804(5)	091(4)	470(3)
H(3)	797(6)	191(5)	526(4)
H(4)	201(7)	585(5)	395(4)
H(5)	336(6)	569(4)	451(4)
H(6)	275(9)	101(7)	306(6)
H(7)	27(1)	232(8)	190(7)
H(8)	41(1)	147(7)	204(6)
H(9)	263(6)	512(4)	111(3)
H(10)	-307(8)	578(5)	297(5)
H(11)	-070(5)	436(4)	396(4)
H(12)	85(1)	-138(7)	195(6)
H(13)	855(9)	011(8)	157(6)
H(14)	1157(9)	-072(7)	077(7)
H(15)	119(1)	-18(1)	187(8)
H(16)	117(2)	-01(1)	16(1)

Table 4.8 Anisotropic temperature factors, and isotropic temperature factors where shown; with standard deviations in parentheses.

ATOM	U11	U22	U33	U23	U13	U12
N(1)	.031(2)	.035(2)	.048(2)	-.021(2)	-.014(1)	.005(1)
C(2)	.028(2)	.040(2)	.041(2)	-.017(2)	-.013(2)	.004(2)
N(21)	.048(2)	.051(2)	.078(3)	-.037(2)	-.035(2)	.018(2)
N(3)	.028(2)	.037(2)	.047(2)	-.020(1)	-.015(1)	.003(1)
C(4)	.025(2)	.035(2)	.035(2)	-.013(2)	-.007(1)	.001(1)
N(41)	.034(2)	.045(2)	.060(2)	-.030(2)	-.022(2)	.010(2)
C(5)	.025(2)	.036(2)	.034(2)	-.011(2)	-.008(1)	-.002(1)
C(6)	.028(2)	.038(2)	.034(2)	-.012(2)	-.007(1)	-.004(2)
C(61)	.053(3)	.045(3)	.057(3)	-.020(2)	-.023(2)	-.007(2)
C(1′)	.025(2)	.034(2)	.047(2)	-.013(2)	-.012(2)	-.003(1)
C(2′)	.029(2)	.041(2)	.043(2)	-.015(2)	-.009(2)	-.003(2)
C(3′)	.041(2)	.043(2)	.045(2)	-.010(2)	-.016(2)	-.007(2)
N(3′1)	.065(3)	.074(3)	.044(2)	-.007(2)	-.019(2)	-.004(2)
N(3′2)	.076(3)	.074(3)	.044(2)	-.005(2)	-.006(2)	-.014(2)
N(3′3)	.111(5)	.134(5)	.067(4)	-.008(3)	.020(3)	-.026(4)
C(4′)	.031(2)	.050(3)	.066(3)	-.012(2)	-.023(2)	.002(2)
CL(4′)	.045(1)	.110(1)	.097(1)	-.007(1)	-.041(1)	.011(1)
C(5′)	.024(2)	.071(3)	.067(3)	-.012(3)	-.004(2)	-.000(2)
C(6′)	.033(2)	.058(3)	.041(3)	-.004(2)	-.004(2)	-.004(2)
S(1)	.0334(5)	.0427(6)	.0550(7)	-.0209(5)	-.0163(4)	.0090(4)
O(1)	.031(2)	.055(2)	.010(3)	-.029(2)	-.010(2)	.002(1)
O(2)	.046(2)	.074(2)	.098(3)	-.054(2)	-.013(2)	-.008(2)
O(3)	.059(2)	.051(2)	.065(2)	-.015(2)	-.017(2)	.022(2)
C(21)	.088(4)	.063(4)	.067(4)	-.024(3)	-.025(3)	-.004(3)
C(22)	.110(6)	.095(6)	.084(5)	-.038(5)	.034(4)	-.016(5)

	U ISO
H(1)	.05(1)
H(2)	.03(1)
H(3)	.05(1)
H(4)	.05(1)
H(5)	.04(1)
H(6)	.10(2)
H(7)	.12(2)
H(8)	.12(2)
H(9)	.05(1)
H(10)	.08(2)
H(11)	.04(1)
H(12)	.11(3)
H(13)	.12(2)
H(14)	.11(2)
H(15)	.17(4)
H(16)	.21(5)

Table 4.9 Bond Lengths(A) with standard deviations

N(1) - C(2)	1.349(5)	C(2') - H(9)	0.95(4)
N(1) - C(6)	1.366(5)	C(3') -N(3'1)	1.414(6)
N(1) - H(1)	0.88(5)	C(3') -C(4')	1.382(6)
C(2) -N(21)	1.332(6)	N(3'1)-N(3'2)	1.223(7)
C(2) - N(3)	1.321(5)	N(3'2)-N(3'3)	1.107(9)
N(21) - H(2)	0.79(4)	C(4') -Cl(4')	1.728(5)
N(21) - H(3)	0.85(5)	C(4') -C(5')	1.362(7)
N(3) - C(4)	1.343(5)	C(5') -C(6')	1.373(7)
C(4) -N(41)	1.319(5)	C(5') -H(10)	0.88(6)
C(4) - C(5)	1.429(5)	C(6') -H(11)	0.89(4)
N(41) - H(4)	0.77(5)	S(1) - O(1)	1.443(4)
N(41) - H(5)	0.80(5)	S(1) - O(2)	1.456(4)
C(5) - C(6)	1.343(6)	S(1) - O(3)	1.438(4)
C(5) -C(1')	1.499(6)	S(1) -C(21)	1.769(7)
C(6) -C(61)	1.488(7)	C(21) -C(22)	1.494(12)
C(61) - H(6)	0.82(7)	C(21) -H(12)	0.92(7)
C(61) - H(7)	0.92(8)	C(21) -H(13)	0.91(8)
C(61) - H(8)	1.09(7)	C(22) -H(14)	0.92(8)
C(1') -C(2')	1.370(6)	C(22) -H(15)	0.94(11)
C(1') -C(6')	1.374(6)	C(22) -H(16)	0.91(13)
C(2') -C(3')	1.392(6)		

Table 4.10 Angles(degrees) with standard deviations

C(2) - N(1) - C(6)	121.0(3)	C(3') -C(2') - H(9)	117.8(27)
C(2) - N(1) - H(1)	122.4(32)	C(2') -C(3') -N(3'1)	123.8(4)
C(6) - N(1) - H(1)	116.1(32)	C(2') -C(3') -C(4')	118.2(4)
N(1) - C(2) -N(21)	117.3(4)	N(3'1)-C(3') -C(4')	118.0(4)
N(1) - C(2) - N(3)	122.7(4)	C(3') -N(3'1)-N(3'2)	116.3(4)
N(21) - C(2) - N(3)	120.0(4)	N(3'1)-N(3'2)-N(3'3)	172.7(7)
C(2) -N(21) - H(2)	115.8(30)	C(3') -C(4') -Cl(4')	119.6(3)
C(2) -N(21) - H(3)	114.1(33)	C(3') -C(4') -C(5')	120.5(4)
H(2) -N(21) - H(3)	127.9(44)	Cl(4')-C(4') -C(5')	119.9(4)
C(2) - N(3) - C(4)	117.6(3)	C(4') -C(5') -C(6')	120.7(5)
N(3) - C(4) -N(41)	116.3(4)	C(4') -C(5') -H(10)	120.9(37)
N(3) - C(4) - C(5)	121.9(3)	C(6') -C(5') -H(10)	118.0(37)
N(41) - C(4) - C(5)	121.7(4)	C(1') -C(6') -C(5')	120.0(4)
C(4) -N(41) - H(4)	124.5(37)	C(1') -C(6') -H(11)	120.8(28)
C(4) -N(41) - H(5)	122.8(33)	C(5') -C(6') -H(11)	119.2(28)
H(4) -N(41) - H(5)	108.4(50)	O(1) - S(1) - O(2)	110.82(21)
C(4) - C(5) - C(6)	117.8(4)	O(1) - S(1) - O(3)	114.65(20)
C(4) - C(5) -C(1')	120.2(3)	O(1) - S(1) -C(21)	107.4(3)

C(6) - C(5) -C(1')	122.0(4)	O(2) - S(1) - O(3)	111.89(21)
N(1) - C(6) - C(5)	118.9(4)	O(2) - S(1) -C(21)	105.2(3)
N(1) - C(6) -C(61)	115.3(4)	O(3) - S(1) -C(21)	106.2(3)
C(5) - C(6) -C(61)	125.8(4)	S(1) -C(21) -C(22)	112.9(6)
C(6) -C(61) - H(6)	111.2(52)	S(1) -C(21) -H(12)	92.0(47)
C(6) -C(61) - H(7)	115.6(51)	S(1) -C(21) -H(13)	102.6(49)
C(6) -C(61) - H(8)	112.9(39)	C(22) -C(21) -H(12)	125.2(47)
H(6) -C(61) - H(7)	131.7(73)	C(22) -C(21) -H(13)	115.0(49)
H(6) -C(61) - H(8)	101.8(65)	H(12) -C(21) -H(13)	104.9(68)
H(7) -C(61) - H(8)	70.8(64)	C(21) -C(22) -H(14)	114.6(48)
C(5) -C(1') -C(2')	119.3(4)	C(21) -C(22) -H(15)	110.0(66)
C(5) -C(1') -C(6')	121.3(4)	C(21) -C(22) -H(16)	108.0(82)
C(2') -C(1') -C(6')	119.4(4)	H(14) -C(22) -H(15)	101.3(81)
C(1') -C(2') -C(3')	121.2(4)	H(14) -C(22) -H(16)	98.8(95)
C(1') -C(2') - H(9)	120.6(27)	H(15) -C(22) -H(16)	123.7(**)

Table 4.11 Torsion angles(degrees) with standard deviations

C(6) - N(1) - C(2) -N(21)	-177.6(4)	C(5) -C(1') -C(6') -C(5')	-179.4(4)
C(6) - N(1) - C(2) - N(3)	3.2(6)	C(5) -C(1') -C(6') -H(11)	-1.9(33)
H(1) - N(1) - C(2) -N(21)	-5.2(38)	C(2') -C(1') -C(6') -C(5')	1.6(7)
H(1) - N(1) - C(2) - N(3)	175.5(38)	C(2') -C(1') -C(6') -H(11)	179.2(32)
C(2) - N(1) - C(6) - C(5)	-1.4(6)	C(1') -C(2') -C(3') -N(3'1)	-179.8(4)
C(2) - N(1) - C(6) -C(61)	179.5(4)	C(1') -C(2') -C(3') -C(4')	0.2(6)
H(1) - N(1) - C(6) - C(5)	-174.2(35)	H(9) -C(2') -C(3') -N(3'1)	-6.8(31)
H(1) - N(1) - C(6) -C(61)	6.7(36)	H(9) -C(2') -C(3') -C(4')	173.3(30)
N(1) - C(2) -N(21) - H(2)	14.5(33)	C(2') -C(3') -N(3'1)-N(3'2)	-0.4(7)
N(1) - C(2) -N(21) - H(3)	179.1(37)	C(4') -C(3') -N(3'1)-N(3'2)	179.6(5)
N(3) - C(2) -N(21) - H(2)	-166.3(33)	C(2') -C(3') -C(4') -Cl(4')	-179.6(3)
N(3) - C(2) -N(21) - H(3)	-1.6(37)	C(2') -C(3') -C(4') -C(5')	1.3(6)
N(1) - C(2) - N(3) - C(4)	-1.8(6)	N(3'1)-C(3') -C(4') -Cl(4')	0.5(6)
N(21) - C(2) - N(3) - C(4)	178.9(4)	N(3'1)-C(3') -C(4') -C(5')	-178.7(4)
C(2) - N(3) - C(4) -N(41)	-178.2(4)	C(3') -N(3'1)-N(3'2)-N(3'3)	-175.7(50)
C(2) - N(3) - C(4) - C(5)	-1.1(6)	C(3') -C(4') -C(5') -C(6')	-1.4(7)
N(3) - C(4) -N(41) - H(4)	-164.0(45)	C(3') -C(4') -C(5') -H(10)	171.8(43)
N(3) - C(4) -N(41) - H(5)	-10.1(40)	Cl(4')-C(4') -C(5') -C(6')	179.5(4)
C(5) - C(4) -N(41) - H(4)	18.9(46)	Cl(4')-C(4') -C(5') -H(10)	-7.3(44)
C(5) - C(4) -N(41) - H(5)	172.8(39)	C(4') -C(5') -C(6') -C(1')	-0.1(8)
N(3) - C(4) - C(5) - C(6)	2.7(6)	C(4') -C(5') -C(6') -H(11)	-177.7(32)
N(3) - C(4) - C(5) -C(1')	-175.6(4)	H(10) -C(5') -C(6') -C(1')	-173.5(42)
N(41) - C(4) - C(5) - C(6)	179.6(4)	H(10) -C(5') -C(6') -H(11)	8.9(53)
N(41) - C(4) - C(5) -C(1')	1.3(6)	O(1) - S(1) -C(21) -C(22)	179.0(5)
C(4) - C(5) - C(6) - N(1)	-1.4(6)	O(1) - S(1) -C(21) -H(12)	-51.1(47)
C(4) - C(5) - C(6) -C(61)	177.6(4)	O(1) - S(1) -C(21) -H(13)	54.7(50)
C(1') - C(5) - C(6) - N(1)	176.9(4)	O(2) - S(1) -C(21) -C(22)	60.9(6)

C(1′) - C(5) - C(6) -C(61)	-4.0(7)	O(2) - S(1) -C(21) -H(12)	-169.3(46)
C(4) - C(5) -C(1′) -C(2′)	105.0(5)	O(2) - S(1) -C(21) -H(13)	-63.5(50)
C(4) - C(5) -C(1′) -C(6′)	-74.0(5)	O(3) - S(1) -C(21) -C(22)	-57.9(6)
C(6) - C(5) -C(1′) -C(2′)	-73.3(5)	O(3) - S(1) -C(21) -H(12)	72.0(47)
C(6) - C(5) -C(1′) -C(6′)	107.7(5)	O(3) - S(1) -C(21) -H(13)	177.8(50)
N(1) - C(6) -C(61) - H(6)	58.4(55)	S(1) -C(21) -C(22) -H(14)	-179.5(52)
N(1) - C(6) -C(61) - H(7)	-134.0(57)	S(1) -C(21) -C(22) -H(15)	67.1(70)
N(1) - C(6) -C(61) - H(8)	-55.2(43)	S(1) -C(21) -C(22) -H(16)	-70.5(86)
C(5) - C(6) -C(61) - H(6)	-120.6(55)	H(12) -C(21) -C(22) -H(14)	70.4(78)
C(5) - C(6) -C(61) - H(7)	46.9(57)	H(12) -C(21) -C(22) -H(15)	-43.0(91)
C(5) - C(6) -C(61) - H(8)	125.7(43)	H(12) -C(21) -C(22) -H(16)	179.3(**)
C(5) -C(1′) -C(2′) -C(3′)	179.3(4)	H(13) -C(21) -C(22) -H(14)	-62.2(76)
C(5) -C(1′) -C(2′) - H(9)	6.5(32)	H(13) -C(21) -C(22) -H(15)	-175.6(89)
C(6′) -C(1′) -C(2′) -C(3′)	-1.7(6)	H(13) -C(21) -C(22) -H(16)	46.7(**)
C(6′) -C(1′) -C(2′) - H(9)	-174.5(31)		

Table 4.12 Hydrogen bond contact distance

Hydrogen Bond	Donor-Acceptor Distance (Å)
$N(1)-H(1)_{I}...O(1)_{I}$	2.841
$N(21)-H(2)_{I}...O(2)_{I}$	2.823
$N(41)-H(5)_{I}...N(3)_{II}$	3.043
$N(41)-H(4)_{I}...O(3)_{III}$	2.950
$N(21)-H(3)_{I}...O(2)_{IV}$	2.962
$N(21)-H(3)_{I}...)(3)_{IV}$	3.135

The subscripts I, II, III and IV refer to the equivalent positions X, Y, Z; 1-X, 1-Y, 1-Z; -1 + X, 1+Y, Z; 2-X, -Y, 1-Z.

Figure 4.3 6-MeMZPES Molecular plot and numbering scheme

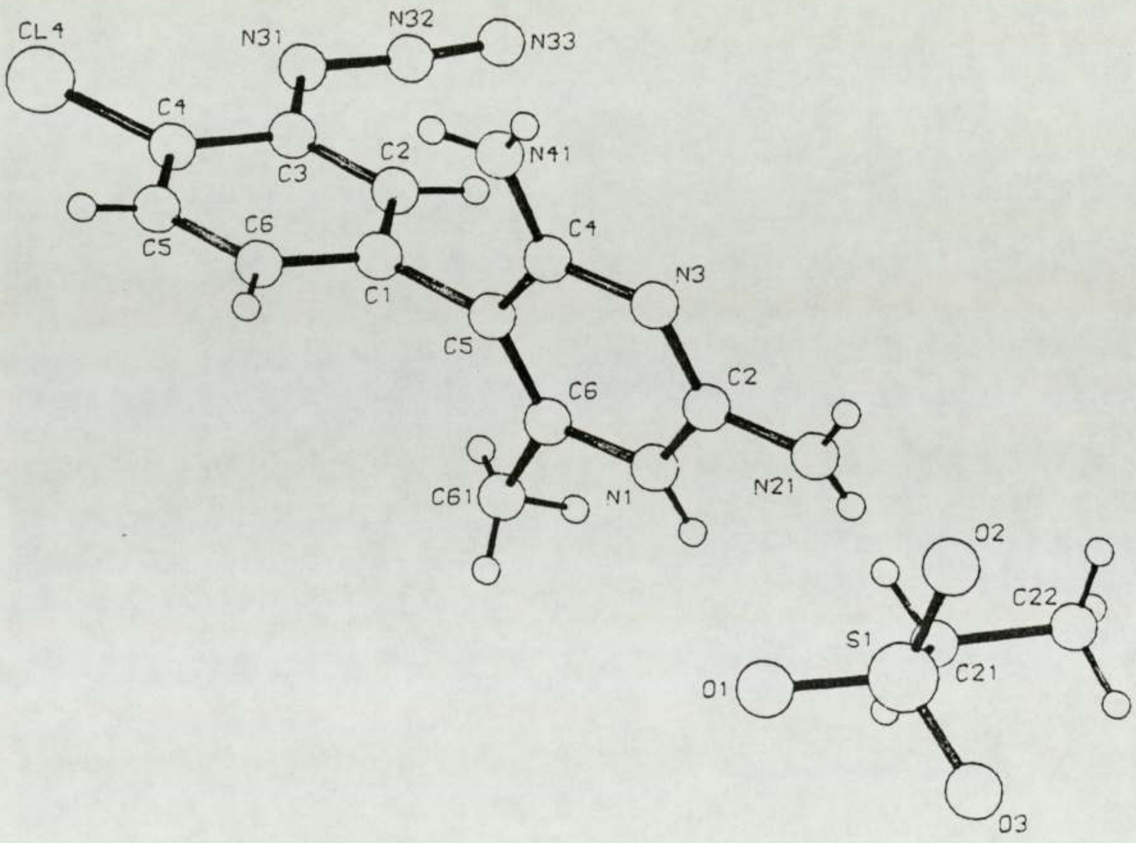
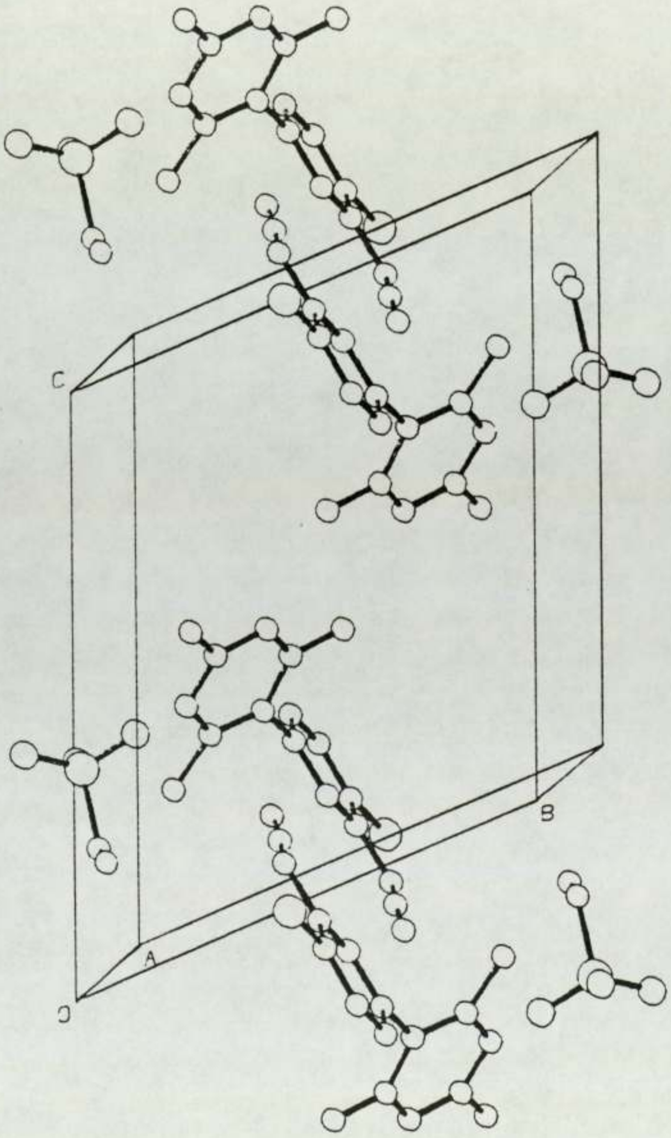


Figure 4.4 6-MeMZPES Unit cell packing diagram



4.4 Results and Discussion

From the crystal structurs of MZPES, I (two independent molecules) and 6-MeMZPES II, common features of molecular geometry were compared with values for the related compound metroprine (DDMP) (61) III. Figure 4.5 shows the common arylazido moiety in which R represents an attached or fused ring.

Figure 4.5

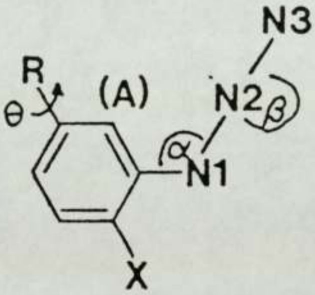


Table 4.13 Geometry of azido group attachment and ring junction

Mol.	C-N1	N1-N2	N2-N3 (Å)	C_A-C-N1	C_X-C-N1	α	β(°)
Ia	1.38	1.24	1.16	125	117	117	171
Ib	1.36	1.35	1.16	126	114	116	168
II	1.401	1.218	1.111	123.0	119.1	117.4	173.2
III	1.416	1.255	1.136	124.0	115.9	115.6	172.5

Table 4.13 (continued)

Mol.	C_A-C-N1-N2	C_X-C-N1-N2	C-N1-N2-N3	(°)
Ia	-8	169	-179	112
Ib	-6	175	173	-86
II	-0.4	179.9	-177.4	-73.4
III	-3.7	175.8	-178.6	N.A.

The N_3 group of Ib makes only distant intermolecular contacts; hence it suffers from high temperature factors and imprecise geometrical parameters. In all the compounds the N_3 unit is almost linear ($\beta \lesssim 170°$) and N2-N3 is shorter than N1-N2. The rather sharp bend at N1 ($\alpha < 120°$) brings N2 into contact with the adjacent hydrogen atom at position A. This contact is relieved by opening the C_A-C-N1 angle at the expense of C_X-C-N1, even when X is as bulky as chlorine. Strain relief via out-of-plane twisting about C-N1 is only of minor importance.

Pyrimidine geometry in II correspnds closely to that in the similarly substituted metoprine (61). There is a large twist between pyrimidine and phenyl rings as measured by Θ , the C6-C5-C1'-C2' torsion angle, which is 110° in metoprine and matches this value to within a twfold rotation in II, whle the change to a 6-ethyl substituent in I pemits but does not require an even greater twist.

The diaminopyrimidine moieties in all three crystal structures follow the general pattern (29) by forming N4-H...N3 hydrogen bonds around a centre of inversion to make a base pair. The protonated ring N1 and the 2-amino group direct protons towards anions to make further hydrogen bonds (Tables 4.6 and 4.12). The azido groups enter into van der Waals contacts only.

Inhibition data against rat liver DHFR show that it is possible to improve an inhibitor by introducing an azido substituent where it may contact a hydrophobic region of DHFR. Thus K_i values with 95% confidence limits are 1.6 ± 0.4 nM for I compared with 2.6 ± 0.3 for pyrimethamine lacking the azido group, 2.6 ± 0.8 for II, and 0.12 ± 0.04 for metoprine. Similarly, III inhibits mouse L1210 and <u>E. coli</u> RT500 (form I) DHFR more strongly than the rather feeble inhibitors 2,4-diamino-quinazoline and its 6-nitro- and 6-amino-congeners. Even so, two determinations of K_i for III against L1210 DHFR gave 51 ± 6 and 46 ± 3 nM.

Determinations of pK_a and log P (octanol-water) for I yielded 7.19 and 2.94 respectively, compared with 7.54 and 1.25 for the more basic and polar amine product of reduction. In antitumour screening tests I proved to be active. This compound is presently in Phase I clinical trial as an antitumour agent. Preliminary results indicate that t ½ for elimination is 32-43h. As intended, this is shorter than it is for many other lipophilic antifolates (88).

CHAPTER 5

CHAPTER 5

The Crystal Structurs of three related antifolate compounds. Benzoprim, Isobenzoprim and Butaprim.

5.1 The Crystal Structure of 2,4-diamino-5-(3-nitro-4-benzylamino)phenyl-6-ethylpyrimidine (Benzoprim).

5.1.1 Experimetal

A crystalline sample of the compound was obtained (76) and from it a suitable crystal was selected. The quality of the specimen crystal was confirmed by examining its optical birefringent properties with a polarising microscope. The single crystal was then mounted onto a glass filament secured to a goniometer head stud by the hot wax method.

5.1.2 Crystal Data

$C_{19}H_{20}N_6O_2$, M = 364.409, F(000) = 1536.
Monoclinic a = 12.411(3), b = 16.060(2), c = 18.462(5) Å, β = 97.19(2)°, V = 3651 Å^3 (by least squares refinement on diffractometer angles for 25 automatically centred reflections, λ = 0.71069 Å), space group C2/c, Z = 8, Dx = 1.326 gcm^{-3}, Dm = 1.325 gcm^{-3} (method of flotation in solvent mixture). Bright orange diamond lozenges. Crystal dimensions, 0.36 x 0.20 x 0.16 mm, μ (Mo-K) = 0.54 cm^{-1}.

5.1.3 Data Collection and Processing

CAD4 diffractometer, w/2θ mode with w scan width = 0.96 +

0.35 tanθ , w scan speed 0.6 - 3.4 deg min^{-1}, graphite monochromated Mo-Kα radiation; 3433 reflections measured ($2 \leqslant \theta \leqslant 25°$ $\underline{+}$ h + k + l, reflections absence due to space group symmetry ignored usingCAD4 option ABSENT = H + KN2), 3201 unique (merging R = 0.0269, no correction for absorption), giving 1173 with $I > 3\sigma(I)$, number of parameters refined 324 . Linear and approximately isotropic crystal decay ca. 1.8% (for 3 standard reflections checked every 7200 s x-ray exposure time), corrected during processing.

5.1.4 Structure Determination and Refinement

Data reduction was followed by direct methods (SHELX76) and electron density synthesis. All hydrogen atoms were located by subsequent difference Fourier analysis during isotropic least squares refinement of non-hydrogen atoms. Convergence was reached after 5 cycles of full matrix least squares refinement, the weighting scheme $W = 1.0293/[\sigma^2 (F_o) + 0.001 F_o^2]$, with $\sigma(F_o)$ from counting statistics, giving satisfactory agreement analyses. Final R and R_w values are 0.0567 and 0.0610. A final electron density synthesis showed no feature > 0.25 e Å^{-3}; (Δ/σ) max = .167 [H(11) U11] in the final refinement cycle but typically, Δ/σ was observed to be in the region of 0.003 and not normally exceeding 0.01. It should be noted that ratio of data to number of refinable parameters, ca. 4:1, is somewhat lower than desirable when describing a structure of this quality.

Table 5.1 Positional parameters (fractional coordinates * 10**4, hydrogens * 10**3) with estimated standard deviations in parentheses.

ATOM	x/a	y/b	z/c
N(1)	1038(3)	1666(3)	6768(3)
C(2)	0246(4)	1232(4)	6375(3)
N(21)	-0738(4)	1222(4)	6629(3)
N(3)	0315(3)	0806(3)	5757(3)
C(4)	1287(4)	0822(4)	5518(3)
N(41)	1370(4)	0377(4)	4912(3)
C(5)	2177(4)	1264(3)	5875(3)
C(6)	1992(4)	1690(3)	6489(3)
C(61)	2860(5)	2194(5)	6927(4)
C(62)	3375(7)	1781(7)	7591(5)
C(1′)	3237(4)	1253(4)	5569(3)
C(2′)	4124(4)	0863(4)	5910(4)
C(3′)	5116(4)	0847(4)	5630(3)
C(4′)	5236(4)	1245(3)	4960(3)
C(5′)	4302(5)	1634(4)	4611(4)
C(6′)	3354(5)	1625(4)	4899(4)
N(31′)	5985(4)	0396(3)	6038(3)
O(31′)	6919(3)	0449(3)	5854(2)
O(32′)	5813(3)	-0019(3)	6567(3)
N(1")	6172(4)	1244(3)	4646(3)
C(11")	6263(7)	1601(7)	3936(5)
C(1")	7386(5)	1421(4)	3735(4)
C(2")	7521(7)	0884(5)	3171(5)
C(3")	8604(9)	0717(6)	3028(6)
C(4")	9449(8)	1088(7)	3432(6)
C(5")	9324(7)	1628(6)	3984(5)
C(6")	8272(6)	1774(6)	4127(5)
H(1)	-086(5)	149(4)	710(4)
H(2)	-120(4)	089(3)	645(3)
H(3)	088(4)	002(4)	474(3)
H(4)	197(5)	038(4)	478(3)
H(5)	342(4)	244(3)	661(3)
H(6)	256(4)	278(3)	701(3)
H(7)	360(6)	117(5)	747(4)
H(8)	288(6)	162(4)	797(4)
H(9)	393(7)	204(5)	792(5)
H(10)	407(4)	064(3)	633(3)
H(11)	436(3)	190(2)	420(2)
H(12)	276(5)	195(4)	464(3)
H(13)	682(5)	101(4)	505(4)
H(14)	615(5)	215(4)	395(3)
H(15)	571(8)	141(7)	359(5)
H(16)	688(5)	063(4)	282(3)
H(17)	844(6)	044(4)	263(4)
H(18)	1024(7)	091(5)	327(5)
H(19)	1010(8)	186(6)	431(6)
H(20)	817(4)	216(4)	448(3)

Table 5.2 Anisotropic temperature factors, and isotropic temperature factors where shown; with standard deviations in parentheses.

ATOM	U11	U22	U33	U23	U13	U12
N(1)	.036(3)	.063(4)	.037(3)	-.002(3)	.013(3)	-.004(3)
C(2)	.030(3)	.046(4)	.044(4)	.008(3)	.010(3)	.010(3)
N(21)	.034(3)	.085(5)	.053(4)	-.015(4)	.023(3)	-.011(3)
N(3)	.026(3)	.054(3)	.039(3)	-.002(3)	.011(2)	.001(2)
C(4)	.031(3)	.046(4)	.034(4)	-.006(3)	.006(3)	.007(3)
N(41)	.029(3)	.075(5)	.065(4)	-.029(3)	.016(3)	-.013(3)
C(5)	.026(3)	.043(4)	.044(4)	-.005(3)	.010(3)	.004(3)
C(6)	.029(3)	.038(4)	.043(4)	.001(3)	.011(3)	.003(3)
C(61)	.042(4)	.065(5)	.049(5)	-.022(4)	.016(3)	-.011(4)
C(62)	.062(5)	.115(8)	.047(5)	-.020(5)	-.003(4)	-.009(5)
C(1′)	.030(3)	.037(4)	.041(4)	.004(3)	.010(3)	.004(3)
C(2′)	.037(3)	.048(4)	.036(4)	-.001(3)	.006(3)	-.003(3)
C(3′)	.032(3)	.038(4)	.051(4)	-.005(3)	.013(3)	.007(3)
C(4′)	.037(3)	.037(4)	.049(4)	-.012(3)	.017(3)	-.002(3)
C(5′)	.048(4)	.049(5)	.057(5)	.017(4)	.029(4)	.004(3)
C(6′)	.041(4)	.047(4)	.052(5)	.006(3)	.010(3)	.003(3)
N(31′)	.039(3)	.057(4)	.054(3)	.001(3)	.016(3)	.010(3)
O(31′)	.031(2)	.093(4)	.067(3)	-.002(3)	.012(2)	.009(2)
O(32′)	.053(3)	.096(4)	.063(3)	.024(3)	.012(2)	.017(3)
N(1")	.038(3)	.066(4)	.046(4)	.013(3)	.016(3)	.009(3)
C(11")	.067(6)	.073(7)	.088(7)	.028(5)	.046(5)	.012(5)
C(1")	.057(4)	.054(5)	.068(5)	.012(4)	.034(4)	-.003(4)
C(2")	.099(7)	.059(5)	.077(7)	.008(5)	.043(6)	-.001(5)
C(3")	.136(9)	.066(6)	.075(8)	.007(6)	.061(7)	.017(6)
C(4")	.081(6)	.102(8)	.106(8)	.032(7)	.051(6)	.031(6)
C(5")	.063(5)	.108(8)	.091(7)	.022(6)	.036(5)	.011(5)
C(6")	.071(6)	.088(6)	.073(6)	-.006(5)	.041(5)	-.003(5)

	U ISO
H(1)	.08(2)
H(2)	.04(2)
H(3)	.04(2)
H(4)	.07(2)
H(5)	.03(1)
H(6)	.04(2)
H(7)	.12(3)
H(8)	.09(2)
H(9)	.14(4)
H(10)	.04(2)
H(11)	.00(1)
H(12)	.07(2)
H(13)	.08(2)
H(14)	.06(3)
H(15)	.19(6)
H(16)	.08(3)
H(17)	.08(3)
H(18)	.14(3)
H(19)	.19(5)
H(20)	.06(2)

Table 5.3 Bond Lengths(A) with standard deviations

N(1) - C(2)	1.342(7)	C(3') -N(31')	1.432(8)
N(1) - C(6)	1.350(7)	C(4') -C(5')	1.401(8)
C(2) -N(21)	1.362(8)	C(4') -N(1")	1.362(7)
C(2) - N(3)	1.342(7)	C(5') -C(6')	1.350(10)
N(21) - H(1)	1.00(6)	C(5') -H(11)	0.88(4)
N(21) - H(2)	0.83(5)	C(6') -H(12)	0.97(6)
N(3) - C(4)	1.336(7)	N(31')-O(31')	1.251(7)
C(4) -N(41)	1.342(8)	N(31')-O(32')	1.223(7)
C(4) - C(5)	1.406(8)	N(1") -C(11")	1.448(11)
N(41) - H(3)	0.87(5)	N(1") -H(13)	1.09(6)
N(41) - H(4)	0.81(6)	C(11")-C(1")	1.514(12)
C(5) - C(6)	1.368(7)	C(11")-H(14)	0.89(6)
C(5) -C(1')	1.495(8)	C(11")-H(15)	0.93(10)
C(6) -C(61)	1.500(9)	C(1") -C(2")	1.378(11)
C(61) -C(62)	1.468(12)	C(1") -C(6")	1.362(11)
C(61) - H(5)	1.04(5)	C(2") -C(3")	1.427(14)
C(61) - H(6)	1.03(5)	C(2") -H(16)	1.04(6)
C(62) - H(7)	1.05(8)	C(3") -C(4")	1.347(16)
C(62) - H(8)	1.03(7)	C(3") -H(17)	0.86(7)
C(62) - H(9)	0.94(9)	C(4") -C(5")	1.362(14)
C(1') -C(2')	1.352(9)	C(4") -H(18)	1.10(8)
C(1') -C(6')	1.398(9)	C(5") -C(6")	1.384(13)
C(2') -C(3')	1.394(9)	C(5") -H(19)	1.13(10)
C(2') -H(10)	0.87(5)	C(6") -H(20)	0.92(6)
C(3') -C(4')	1.417(8)		

Table 5.4 Angles(degrees) with standard deviations

C(2) - N(1) - C(6)	115.3(5)	C(4') -C(3') -N(31')	122.2(5)
N(1) - C(2) -N(21)	116.6(5)	C(3') -C(4') -C(5')	115.6(5)
N(1) - C(2) - N(3)	127.2(5)	C(3') -C(4') -N(1")	124.0(5)
N(21) - C(2) - N(3)	116.2(5)	C(5') -C(4') -N(1")	120.4(5)
C(2) -N(21) - H(1)	122.4(36)	C(4') -C(5') -C(6')	121.7(6)
C(2) -N(21) - H(2)	118.6(37)	C(4') -C(5') -H(11)	117.3(26)
H(1) -N(21) - H(2)	117.4(52)	C(6') -C(5') -H(11)	120.9(26)
C(2) - N(3) - C(4)	115.3(5)	C(1') -C(6') -C(5')	123.0(6)
N(3) - C(4) -N(41)	115.6(5)	C(1') -C(6') -H(12)	120.6(35)
N(3) - C(4) - C(5)	123.0(5)	C(5') -C(6') -H(12)	116.0(35)
N(41) - C(4) - C(5)	121.5(5)	C(3') -N(31')-O(31')	119.0(5)
C(4) -N(41) - H(3)	122.7(37)	C(3') -N(31')-O(32')	120.0(5)
C(4) -N(41) - H(4)	115.2(44)	O(31')-N(31')-O(32')	120.9(5)
H(3) -N(41) - H(4)	120.6(57)	C(4') -N(1") -C(11")	123.5(6)

C(4) - C(5) - C(6)	115.9(5)	C(4') -N(1") -H(13)	107.3(34)
C(4) - C(5) -C(1')	119.6(5)	C(11")-N(1") -H(13)	128.6(34)
C(6) - C(5) -C(1')	124.5(5)	N(1") -C(11")-C(1")	108.9(7)
N(1) - C(6) - C(5)	123.3(5)	N(1") -C(11")-H(14)	110.0(42)
N(1) - C(6) -C(61)	114.5(5)	N(1") -C(11")-H(15)	110.8(64)
C(5) - C(6) -C(61)	122.2(5)	C(1") -C(11")-H(14)	110.5(42)
C(6) -C(61) -C(62)	114.5(6)	C(1") -C(11")-H(15)	112.9(64)
C(6) -C(61) - H(5)	112.6(26)	H(14) -C(11")-H(15)	103.6(76)
C(6) -C(61) - H(6)	108.6(28)	C(11")-C(1") -C(2")	120.5(7)
C(62) -C(61) - H(5)	113.1(26)	C(11")-C(1") -C(6")	119.8(7)
C(62) -C(61) - H(6)	114.9(28)	C(2") -C(1") -C(6")	119.7(7)
H(5) -C(61) - H(6)	90.9(37)	C(1") -C(2") -C(3")	117.6(8)
C(61) -C(62) - H(7)	110.0(43)	C(1") -C(2") -H(16)	123.9(36)
C(61) -C(62) - H(8)	116.7(39)	C(3") -C(2") -H(16)	118.3(36)
C(61) -C(62) - H(9)	122.8(54)	C(2") -C(3") -C(4")	120.0(10)
H(7) -C(62) - H(8)	96.3(57)	C(2") -C(3") -H(17)	97.6(48)
H(7) -C(62) - H(9)	110.6(68)	C(4") -C(3") -H(17)	141.5(49)
H(8) -C(62) - H(9)	96.8(66)	C(3") -C(4") -C(5")	122.8(10)
C(5) -C(1') -C(2')	122.2(5)	C(3") -C(4") -H(18)	113.4(44)
C(5) -C(1') -C(6')	121.5(5)	C(5") -C(4") -H(18)	123.8(44)
C(2') -C(1') -C(6')	116.2(6)	C(4") -C(5") -C(6")	116.7(9)
C(1') -C(2') -C(3')	122.8(6)	C(4") -C(5") -H(19)	116.2(54)
C(1') -C(2') -H(10)	117.2(34)	C(6") -C(5") -H(19)	126.8(54)
C(3') -C(2') -H(10)	119.9(34)	C(1") -C(6") -C(5")	123.1(8)
C(2') -C(3') -C(4')	120.6(5)	C(1") -C(6") -H(20)	118.7(37)
C(2') -C(3') -N(31')	117.1(5)	C(5") -C(6") -H(20)	118.0(37)

Table 5.5 Torsion angles(degrees) with standard deviations

C(6) - N(1) - C(2) -N(21)	-177.7(5)	C(2') -C(3') -C(4') -C(5')	-0.7(8)
C(6) - N(1) - C(2) - N(3)	2.2(8)	C(2') -C(3') -C(4') -N(1")	-178.8(5)
C(2) - N(1) - C(6) - C(5)	-4.0(8)	N(31')-C(3') -C(4') -C(5')	177.6(5)
C(2) - N(1) - C(6) -C(61)	178.7(5)	N(31')-C(3') -C(4') -N(1")	-0.5(9)
N(1) - C(2) -N(21) - H(1)	-3.6(43)	C(2') -C(3') -N(31')-O(31')	-171.5(5)
N(1) - C(2) -N(21) - H(2)	-168.5(43)	C(2') -C(3') -N(31')-O(32')	7.0(8)
N(3) - C(2) -N(21) - H(1)	176.5(43)	C(4') -C(3') -N(31')-O(31')	10.1(8)
N(3) - C(2) -N(21) - H(2)	11.6(43)	C(4') -C(3') -N(31')-O(32')	-171.4(5)
N(1) - C(2) - N(3) - C(4)	0.2(8)	C(3') -C(4') -C(5') -C(6')	-0.1(9)
N(21) - C(2) - N(3) - C(4)	-179.9(5)	C(3') -C(4') -C(5') -H(11)	178.3(29)
C(2) - N(3) - C(4) -N(41)	178.1(5)	N(1") -C(4') -C(5') -C(6')	178.1(6)
C(2) - N(3) - C(4) - C(5)	-1.2(8)	N(1") -C(4') -C(5') -H(11)	-3.5(30)
N(3) - C(4) -N(41) - H(3)	-12.8(44)	C(3') -C(4') -N(1") -C(11")	175.4(6)
N(3) - C(4) -N(41) - H(4)	-178.5(49)	C(3') -C(4') -N(1") -H(13)	-12.2(36)
C(5) - C(4) -N(41) - H(3)	166.5(43)	C(5') -C(4') -N(1") -C(11")	-2.7(9)
C(5) - C(4) -N(41) - H(4)	0.8(49)	C(5') -C(4') -N(1") -H(13)	169.7(35)

N(3) - C(4) - C(5) - C(6)	-0.4(8)	C(4′) -C(5′) -C(6′) -C(1′)	1.9(10)
N(3) - C(4) - C(5) -C(1′)	-179.7(5)	C(4′) -C(5′) -C(6′) -H(12)	175.6(39)
N(41) - C(4) - C(5) - C(6)	-179.7(5)	H(11) -C(5′) -C(6′) -C(1′)	-176.5(30)
N(41) - C(4) - C(5) -C(1′)	1.1(8)	H(11) -C(5′) -C(6′) -H(12)	-2.7(50)
C(4) - C(5) - C(6) - N(1)	3.2(8)	C(4′) -N(1") -C(11")-C(1")	-175.1(6)
C(4) - C(5) - C(6) -C(61)	-179.7(5)	C(4′) -N(1") -C(11")-H(14)	63.7(45)
C(1′) - C(5) - C(6) - N(1)	-177.6(5)	C(4′) -N(1") -C(11")-H(15)	-50.3(69)
C(1′) - C(5) - C(6) -C(61)	-0.5(8)	H(13) -N(1") -C(11")-C(1")	14.2(44)
C(4) - C(5) -C(1′) -C(2′)	-111.3(7)	H(13) -N(1") -C(11")-H(14)	-107.0(62)
C(4) - C(5) -C(1′) -C(6′)	66.6(8)	H(13) -N(1") -C(11")-H(15)	139.0(81)
C(6) - C(5) -C(1′) -C(2′)	69.5(8)	N(1") -C(11")-C(1") -C(2")	111.8(8)
C(6) - C(5) -C(1′) -C(6′)	-112.6(7)	N(1") -C(11")-C(1") -C(6")	-66.4(10)
N(1) - C(6) -C(61) -C(62)	76.6(8)	H(14) -C(11")-C(1") -C(2")	-127.3(45)
N(1) - C(6) -C(61) - H(5)	-152.4(28)	H(14) -C(11")-C(1") -C(6")	54.5(45)
N(1) - C(6) -C(61) - H(6)	-53.3(29)	H(15) -C(11")-C(1") -C(2")	-11.8(70)
C(5) - C(6) -C(61) -C(62)	-100.8(8)	H(15) -C(11")-C(1") -C(6")	170.0(69)
C(5) - C(6) -C(61) - H(5)	30.2(28)	C(11")-C(1") -C(2") -C(3")	-177.7(8)
C(5) - C(6) -C(61) - H(6)	129.4(29)	C(11")-C(1") -C(2") -H(16)	7.4(44)
C(6) -C(61) -C(62) - H(7)	48.0(46)	C(6") -C(1") -C(2") -C(3")	0.5(12)
C(6) -C(61) -C(62) - H(8)	-60.3(44)	C(6") -C(1") -C(2") -H(16)	-174.4(43)
C(6) -C(61) -C(62) - H(9)	-179.1(64)	C(11")-C(1") -C(6") -C(5")	178.9(8)
H(5) -C(61) -C(62) - H(7)	-82.8(53)	C(11")-C(1") -C(6") -H(20)	-6.8(43)
H(5) -C(61) -C(62) - H(8)	168.9(51)	C(2") -C(1") -C(6") -C(5")	0.7(13)
H(5) -C(61) -C(62) - H(9)	50.1(70)	C(2") -C(1") -C(6") -H(20)	175.1(41)
H(6) -C(61) -C(62) - H(7)	174.7(54)	C(1") -C(2") -C(3") -C(4")	-0.9(14)
H(6) -C(61) -C(62) - H(8)	66.3(53)	C(1") -C(2") -C(3") -H(17)	-172.4(48)
H(6) -C(61) -C(62) - H(9)	-52.4(71)	H(16) -C(2") -C(3") -C(4")	174.4(41)
C(5) -C(1′) -C(2′) -C(3′)	179.9(5)	H(16) -C(2") -C(3") -H(17)	2.8(63)
C(5) -C(1′) -C(2′) -H(10)	-3.5(39)	C(2") -C(3") -C(4") -C(5")	0.0(17)
C(6′) -C(1′) -C(2′) -C(3′)	1.9(9)	C(2") -C(3") -C(4") -H(18)	-179.7(47)
C(6′) -C(1′) -C(2′) -H(10)	178.5(38)	H(17) -C(3") -C(4") -C(5")	166.5(77)
C(5) -C(1′) -C(6′) -C(5′)	179.3(6)	H(17) -C(3") -C(4") -H(18)	-13.3(92)
C(5) -C(1′) -C(6′) -H(12)	5.8(41)	C(3") -C(4") -C(5") -C(6")	1.1(15)
C(2′) -C(1′) -C(6′) -C(5′)	-2.7(10)	C(3") -C(4") -C(5") -H(19)	175.7(60)
C(2′) -C(1′) -C(6′) -H(12)	-176.2(41)	H(18) -C(4") -C(5") -C(6")	-179.2(52)
C(1′) -C(2′) -C(3′) -C(4′)	-0.3(9)	H(18) -C(4") -C(5") -H(19)	-4.6(79)
C(1′) -C(2′) -C(3′) -N(31′)	-178.7(6)	C(4") -C(5") -C(6") -C(1")	-1.5(14)
H(10) -C(2′) -C(3′) -C(4′)	-176.8(39)	C(4") -C(5") -C(6") -H(20)	-175.9(41)
H(10) -C(2′) -C(3′) -N(31′)	4.8(40)	H(19) -C(5") -C(6") -C(1")	-175.4(67)

Table 5.6 Hydrogen bond contact distances

Hydrogen Bond	Donor-Acceptor Distance (Å)
$N(21)-H(1)_{I}\ldots N(1)_{II}$	3.111
$N(41)-H(3)_{I}\ldots N(3)_{III}$	2.977
$N(41)-H(4)_{I}\ldots O(31')_{IV}$	3.003

The subscripts I, II, III and IV refer to the equivalent positons X, Y, Z; -X, Y, 1½-Z; -X, -Y, 1-Z; 1-X, -Y, 1-Z.

Figure 5.1 Benzoprim Molecular plot and numbering scheme

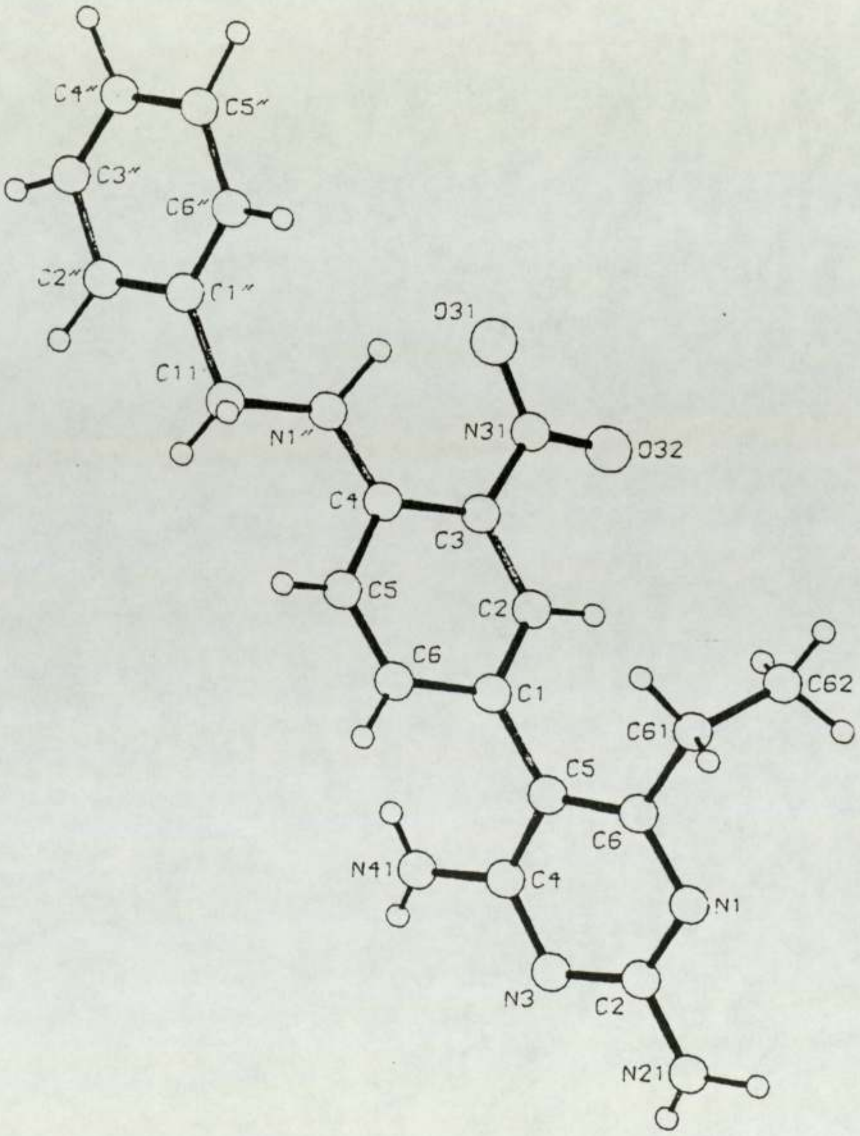
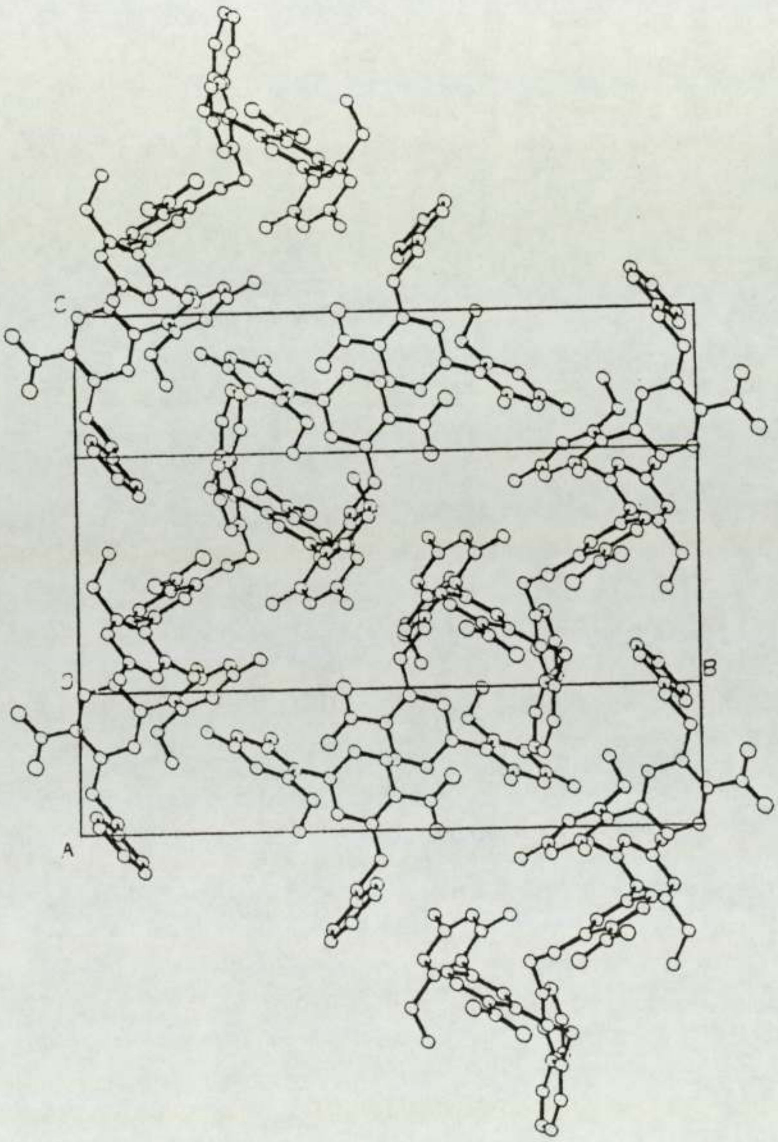


Figure 5.2 Benzoprim Unit cell packing diagram



5.2 The Crystal Structure of 2,4-diamino-5-(3-benzyl amino-4-nitro)phenyl-ethyl pyrimidine (Isobenzoprim)

5.2.1 Experimental

A crystalline sample of the compound was obtained (76) and from it a suitable crystal was selected. The quality of the specimen crystal was confirmed by examining its optical birefringent properties with a polarising microscope. The single crystal was then mounted onto a glass filament secured to a goniometer head stud by the hot wax method.

5.2.2 Crystal Data

$C_{19} H_{20} N_6 O_2$, M = 364.409, F (000) = 1536. Monoclinic a = 12.822(3), b = 22.846(7), c = 13.888(3) Å, β = 113.93(2)°, V = 3718 $Å^3$ (by least squares refinement on diffractometer angles for 25 automatically centred reflections, λ = 0.71069 Å), space group P $2_1/a$, Z = 8, Dx = 1.302 gcm^{-3}, Dm = 1.297 gcm^{-3} (method of flotation in solvent mixture). Orange needles. Crystal dimensions 0.45 x 0.20 x 0.20 mm, μ (Mo-Kα) = 0.53 cm^{-1}.

5.2.3 Data Collection and Processing

CAD4 diffractometer, w/2θ mode with w scan width = 0.8 + 0.35 tanθ, w scan speed 0.25 - 2.2 deg min^{-1}, graphite monochromated Mo-Kα radiation; 5253 reflections measured ($2 \leq \theta \leq$ 22.5° + h ± k + 1), 4839 unique (merging R = 0.0266, no correction for absorption), giving 1524 with I > 2 σ(I), number of parameters refined 210. No evidence of crystal decay (for three standard reflections checked every 7200 s x-ray exposure time).

5.2.4 Structure Determination and Refinement

Data reduction was followed by direct methods (SHELX76) and electron density synthesis. Due to the weak intensity of the data a 2σ cutoff was operated and consequenctly hydrogen positions were poorly located by difference Fourier analysis. In view of this only amino hydrogens were found crystallographically and refined under a common temperature factor with no refinement of positional parameters. Other hydrogen positions were calculated using a variety of AFIX options (SHELX76) these being allowed to refine with a common temperature factor and fixed geometry. In an effort to make best use of the data available the terminal ring of the benzylamino moiety was refined as a rigid group with fixed geometry. All other non-hydrogen atoms were refined anisotropically. Convergence of full matrix least squares refinement was achieved after 20 cycles. The weighting scheme $W = 1.3216/[\sigma^2 (F_o) + 0.000981 F_o^2]$ gave satisfactory agreement analyses with $\sigma (F_o)$ from counting statistics. Final R and R_w values are 0.0853 and 0.0859. A final electron density synthesis showed 4 features $\sim$0.25 e $Å^{-3}$, this density was located around the 'fixed' ring systems.

Table 5.7 Positional parameters (fractional coordinates * 10**4, or 10**3) with estimated standard deviations in parentheses.

ATOM	x/a	y/b	z/c
N(1A)	4601(8)	2155(5)	1577(7)
C(2A)	402(1)	2499(6)	1938(9)
N(21A)	4568(8)	2941(5)	2534(8)
N(3A)	2914(9)	2453(4)	1779(8)
C(4A)	238(1)	1973(7)	126(1)
N(41A)	1274(8)	1914(4)	1076(7)
C(5A)	291(1)	1535(5)	0884(9)
C(6A)	406(1)	1650(5)	108(1)
C(61A)	482(1)	1253(6)	077(1)
C(62A)	537(2)	1485(7)	016(2)
C(1'A)	234(1)	0986(5)	041(1)
C(2'A)	213(1)	0871(6)	-062(1)
C(3'A)	163(1)	0321(7)	-108(1)
C(4'A)	137(1)	-0064(6)	-045(1)
C(5'A)	152(1)	0073(5)	056(1)
C(6'A)	199(1)	0579(7)	098(1)
N(3SA)	149(1)	0182(5)	-209(1)
C(3SA)	176(1)	0580(7)	-279(1)
C(1"A)	2995(9)	0591(4)	-2624(7)
C(2"A)	3821(9)	0253(4)	-1853(7)
C(3"A)	4962(9)	0295(4)	-1700(7)
C(4"A)	5279(9)	0677(4)	-2320(7)
C(5"A)	4453(9)	1015(4)	-3092(7)
C(6"A)	3312(9)	0972(4)	-3244(7)
N(4'A)	083(1)	-0630(7)	-081(1)
O(1'A)	075(1)	-0988(5)	-0185(9)
O(2'A)	042(1)	-0728(4)	-1756(9)
N(1B)	3767(8)	4524(4)	4864(7)
C(2B)	411(1)	4185(6)	423(1)
N(21B)	5144(8)	4339(4)	4202(7)
N(3B)	3565(8)	3742(4)	3631(7)
C(4B)	257(1)	3606(5)	3666(9)
N(41B)	2020(8)	3145(4)	3043(7)
C(5B)	2092(9)	3920(5)	4275(9)
C(6B)	275(1)	4374(6)	487(1)
C(61B)	245(1)	4741(6)	563(1)
C(62B)	302(2)	449(1)	673(2)
C(1'B)	097(1)	3762(6)	4247(9)
C(2'B)	081(1)	3269(5)	4665(9)
C(3'B)	-027(1)	3082(7)	4633(9)
C(4'B)	-113(1)	3506(7)	414(1)
C(5'B)	-096(1)	4011(7)	371(1)
C(6'B)	007(1)	4151(6)	379(1)
N(3SB)	-0418(9)	2568(5)	5051(9)
C(3SB)	050(1)	2179(6)	561(1)
C(1"B)	1211(7)	2360(5)	672(1)
C(2"B)	0706(7)	2382(5)	744(1)
C(3"B)	1351(7)	2548(5)	849(1)
C(4"B)	2500(7)	2693(5)	881(1)
C(5"B)	3004(7)	2671(5)	808(1)
C(6"B)	2360(7)	2505(5)	705(1)
N(4'B)	-233(1)	3409(6)	402(1)

Table 5.7 contd.

ATOM	x/a	y/b	z/c	
O(1'B)	-2486(8)	2965(5)	4488(9)	
O(2'B)	-3050(9)	3753(5)	3555(9)	
H(1A)	5279	2888	2649	I
H(2A)	4240	3278	2711	I
H(3A)	0953	2285	1347	I
H(4A)	0814	1557	0753	I
H(5A)	433(1)	0867(6)	040(1)	
H(6A)	553(1)	1131(6)	148(1)	
H(7A)	599(2)	1249(7)	-004(2)	
H(8A)	561(2)	1942(7)	028(2)	
H(9A)	454(2)	1444(7)	-047(2)	
H(10A)	237(1)	1187(6)	-108(1)	
H(11A)	127(1)	-0232(5)	102(1)	
H(12A)	211(1)	0683(7)	177(1)	
H(13A)	1076	-0148	-2452	I
H(14A)	153(1)	1020(7)	-268(1)	
H(15A)	127(1)	0447(7)	-360(1)	
H(16A)	3576(9)	-0042(4)	-1373(7)	
H(17A)	5602(9)	0034(4)	-1103(7)	
H(18A)	6163(9)	0710(4)	-2202(7)	
H(19A)	4698(9)	1310(4)	-3572(7)	
H(20A)	2673(9)	1234(4)	-3842(7)	
H(1B)	5470	4701	4509	I
H(2B)	5500	4019	3800	I
H(4B)	1345	2914	3026	I
H(5B)	154(1)	4719(6)	540(1)	
H(6B)	270(1)	5191(6)	561(1)	
H(7B)	270(2)	475(1)	719(2)	
H(8B)	391(2)	459(1)	695(2)	
H(9B)	292(2)	403(1)	686(2)	
H(10B)	154(1)	2987(5)	5055(9)	
H(11B)	-166(1)	4311(7)	332(1)	
H(12B)	021(1)	4563(6)	348(1)	
H(13B)	-1117	2531	5086	I
H(14B)	018(1)	1737(6)	556(1)	
H(15B)	109(1)	2197(6)	523(1)	
H(16B)	-0183(7)	2270(5)	720(1)	
H(17B)	0960(7)	2564(5)	904(1)	
H(18B)	2999(7)	2821(5)	961(1)	
H(19B)	3894(7)	2783(5)	833(1)	
H(20B)	2750(7)	2489(5)	649(1)	

I Hydrogens refining common temperature factor only.

All other hydrogens refining in rigid groups using AFIX option in SHELX.

Table 5.8 Anisotropic temperature factors, and isotropic temperature factors where shown; with standard deviations in parentheses.

ATOM	U11	U22	U33	U23	U13	U12
N(1A)	.038(7)	.047(8)	.058(8)	-.029(6)	.025(6)	-.006(6)
C(2A)	.021(8)	.047(8)	.038(9)	-.008(7)	.002(7)	-.012(7)
N(21A)	.038(7)	.062(8)	.094(9)	-.034(7)	.035(7)	-.023(6)
N(3A)	.048(7)	.014(6)	.060(8)	-.010(6)	.017(7)	-.021(6)
C(4A)	.033(9)	.07(1)	.05(1)	.007(9)	.028(8)	.002(9)
N(41A)	.035(7)	.042(7)	.068(8)	-.029(6)	.029(6)	-.012(5)
C(5A)	.026(8)	.031(8)	.032(8)	.005(7)	.009(7)	.013(7)
C(6A)	.041(9)	.023(9)	.06(1)	-.007(7)	.008(8)	-.002(7)
C(61A)	.07(1)	.03(1)	.14(2)	-.05(1)	.04(1)	-.012(9)
C(62A)	.22(2)	.06(1)	.27(3)	-.07(2)	.22(2)	-.07(1)
C(1’A)	.07(1)	.017(8)	.06(1)	-.029(8)	.035(9)	-.017(8)
C(2’A)	.06(1)	.05(1)	.05(1)	-.013(9)	.022(9)	-.025(8)
C(3’A)	.06(1)	.06(1)	.023(9)	.001(9)	.000(9)	.010(9)
C(4’A)	.10(1)	.028(9)	.05(1)	-.031(9)	.05(1)	-.032(9)
C(5’A)	.12(1)	.014(8)	.07(1)	-.022(8)	.05(1)	-.032(9)
C(6’A)	.08(1)	.08(1)	.04(1)	-.01(1)	.014(9)	-.03(1)
N(3SA)	.12(1)	.08(1)	.057(9)	-.020(8)	.029(9)	-.036(8)
C(3SA)	.14(2)	.09(1)	.03(1)	-.020(9)	.04(1)	-.03(1)
N(4’A)	.09(1)	.08(1)	.06(1)	-.01(1)	.03(1)	.00(1)
O(1’A)	.14(1)	.065(9)	.10(1)	-.016(7)	.051(9)	-.038(8)
O(2’A)	.17(1)	.074(9)	.084(9)	-.025(7)	.04(1)	-.070(8)
N(1B)	.049(8)	.058(8)	.041(7)	-.024(6)	.032(6)	-.011(6)
C(2B)	.029(8)	.05(1)	.034(9)	.014(7)	.019(7)	-.008(7)
N(21B)	.041(7)	.058(7)	.048(7)	-.018(6)	.026(6)	-.015(6)
N(3B)	.032(7)	.020(6)	.057(8)	-.014(6)	.027(6)	-.013(6)
C(4B)	.018(8)	.024(9)	.032(8)	.007(6)	.002(7)	.005(6)
N(41B)	.034(7)	.054(8)	.058(8)	-.027(6)	.024(6)	-.025(6)
C(5B)	.016(8)	.051(9)	.041(9)	-.014(7)	.018(7)	-.005(7)
C(6B)	.029(8)	.06(1)	.05(1)	-.004(8)	.029(8)	-.007(8)
C(61B)	.06(1)	.10(1)	.13(2)	-.10(1)	.08(1)	-.061(9)
C(62B)	.19(2)	.23(3)	.07(2)	-.05(2)	.09(2)	-.02(2)
C(1’B)	.029(9)	.05(1)	.027(9)	-.012(7)	.004(7)	.000(8)
C(2’B)	.05(1)	.039(9)	.047(9)	.004(7)	.039(8)	.007(8)
C(3’B)	.025(9)	.07(1)	.036(9)	-.020(8)	.016(8)	-.011(8)
C(4’B)	.018(9)	.07(1)	.06(1)	-.018(9)	.028(8)	-.009(8)
C(5’B)	.04(1)	.07(1)	.04(1)	-.005(8)	.007(8)	.001(9)
C(6’B)	.036(9)	.06(1)	.07(1)	-.013(9)	.033(9)	-.017(9)
N(3SB)	.050(8)	.038(8)	.087(9)	.005(7)	.047(7)	-.011(7)
C(3SB)	.05(1)	.07(1)	.05(1)	.004(8)	.022(9)	.011(9)
N(4’B)	.10(1)	.04(1)	.09(1)	-.018(8)	.06(1)	.000(9)
O(1’B)	.049(7)	.11(1)	.13(1)	-.005(8)	.057(7)	-.015(7)
O(2’B)	.062(8)	.075(9)	.13(1)	.002(8)	.053(8)	.018(7)

	U ISO
C(1"A)	.083(5)
C(2"A)	.076(5)
C(3"A)	.092(5)
C(4"A)	.108(6)
C(5"A)	.118(6)
C(6"A)	.113(6)
C(1"B)	.063(4)
C(2"B)	.120(6)

Table 5.8 contd.

	U ISO
C(3"B)	.142(7)
C(4"B)	.121(7)
C(5"B)	.156(8)
C(6"B)	.129(7)
H(1A)	.08(1)
H(2A)	.08(1)
H(3A)	.08(1)
H(4A)	.08(1)
H(5A)	.08(2)
H(6A)	.08(2)
H(7A)	.14(4)
H(8A)	.14(4)
H(9A)	.14(4)
H(10A)	.08(2)
H(11A)	.08(2)
H(12A)	.08(2)
H(13A)	.08(1)
H(14A)	.17(2)
H(15A)	.17(2)
H(16A)	.17(2)
H(17A)	.17(2)
H(18A)	.17(2)
H(19A)	.17(2)
H(20A)	.17(2)
H(1B)	.10(1)
H(2B)	.10(1)
H(4B)	.10(1)
H(5B)	.10(2)
H(6B)	.10(2)
H(7B)	.19(5)
H(8B)	.19(5)
H(9B)	.19(5)
H(10B)	.10(2)
H(11B)	.10(2)
H(12B)	.10(2)
H(13B)	.10(1)
H(14B)	.44(7)
H(15B)	.44(7)
H(16B)	.44(7)
H(17B)	.44(7)
H(18B)	.44(7)
H(19B)	.44(7)
H(20B)	.44(7)

Table 5.9 Bond Lengths(A) with standard deviations

N(1A) -C(2A)	1.314(17)	N(1B) -C(2B)	1.369(17)
N(1A) -C(6A)	1.377(17)	N(1B) -C(6B)	1.347(17)
C(2A) -N(21A)	1.314(17)	C(2B) -N(21B)	1.388(17)
C(2A) -N(3A)	1.348(17)	C(2B) -N(3B)	1.319(17)
N(21A)-H(1A)	0.868(11)	N(21B)-H(1B)	0.947(10)
N(21A)-H(2A)	0.956(11)	N(21B)-H(2B)	1.123(10)
N(3A) -C(4A)	1.335(18)	N(3B) -C(4B)	1.332(16)
C(4A) -N(41A)	1.343(18)	C(4B) -N(41B)	1.364(16)
C(4A) -C(5A)	1.428(19)	C(4B) -C(5B)	1.424(17)
N(41A)-H(3A)	1.075(10)	N(41B)-H(4B)	1.006(10)
N(41A)-H(4A)	1.000(10)	C(5B) -C(6B)	1.379(18)
C(5A) -C(6A)	1.411(18)	C(5B) -C(1’B)	1.466(18)
C(5A) -C(1’A)	1.469(18)	C(6B) -C(61B)	1.513(21)
C(6A) -C(61A)	1.512(21)	C(61B)-C(62B)	1.52(3)
C(61A)-C(62A)	1.41(3)	C(61B)-H(5B)	1.080(22)
C(61A)-H(5A)	1.079(22)	C(61B)-H(6B)	1.080(22)
C(61A)-H(6A)	1.080(22)	C(62B)-H(7B)	1.08(3)
C(62A)-H(7A)	1.08(3)	C(62B)-H(8B)	1.08(3)
C(62A)-H(8A)	1.08(3)	C(62B)-H(9B)	1.08(3)
C(62A)-H(9A)	1.08(3)	C(1’B)-C(2’B)	1.321(18)
C(1’A)-C(2’A)	1.368(20)	C(1’B)-C(6’B)	1.392(19)
C(1’A)-C(6’A)	1.402(21)	C(2’B)-C(3’B)	1.430(19)
C(2’A)-C(3’A)	1.439(21)	C(2’B)-H(10B)	1.081(17)
C(2’A)-H(10A)	1.081(20)	C(3’B)-C(4’B)	1.420(20)
C(3’A)-C(4’A)	1.373(22)	C(3’B)-N(3SB)	1.358(18)
C(3’A)-N(3SA)	1.383(20)	C(4’B)-C(5’B)	1.355(21)
C(4’A)-C(5’A)	1.367(21)	C(4’B)-N(4’B)	1.488(21)
C(4’A)-N(4’A)	1.457(22)	C(5’B)-C(6’B)	1.327(21)
C(5’A)-C(6’A)	1.326(21)	C(5’B)-H(11B)	1.080(21)
C(5’A)-H(11A)	1.080(20)	C(6’B)-H(12B)	1.078(20)
C(6’A)-H(12A)	1.080(22)	N(3SB)-C(3SB)	1.423(18)
N(3SA)-C(3SA)	1.476(20)	N(3SB)-H(13B)	0.921(12)
N(3SA)-H(13A)	0.939(13)	C(3SB)-C(1"B)	1.507(19)
C(3SA)-C(1"A)	1.499(19)	C(3SB)-H(14B)	1.080(20)
C(3SA)-H(14A)	1.081(22)	C(3SB)-H(15B)	1.081(20)
C(3SA)-H(15A)	1.081(22)	C(2"B)-C(3"B)	1.396(17)
C(2"A)-C(3"A)	1.394(14)	C(2"B)-C(1"B)	1.395(17)
C(2"A)-C(1"A)	1.394(14)	C(2"B)-H(16B)	1.079(17)
C(2"A)-H(16A)	1.080(14)	C(3"B)-C(4"B)	1.395(17)
C(3"A)-C(4"A)	1.397(14)	C(3"B)-H(17B)	1.080(17)
C(3"A)-H(17A)	1.079(14)	C(4"B)-C(5"B)	1.394(17)
C(4"A)-C(5"A)	1.395(14)	C(4"B)-H(18B)	1.080(17)
C(4"A)-H(18A)	1.080(14)	C(5"B)-C(6"B)	1.394(17)
C(5"A)-C(6"A)	1.394(14)	C(5"B)-H(19B)	1.081(17)
C(5"A)-H(19A)	1.080(14)	C(6"B)-C(1"B)	1.395(17)
C(6"A)-C(1"A)	1.396(14)	C(6"B)-H(20B)	1.080(17)

C(6"A)-H(20A)	1.080(14)	N(4'B)-O(1'B)	1.267(19)
N(4'A)-O(1'A)	1.229(20)	N(4'B)-O(2'B)	1.188(19)
N(4'A)-O(2'A)	1.219(20)		

Table 5.10 Angles(degrees) with standard deviations

C(2A) -N(1A) -C(6A)	116.5(11)	N(3SA)-C(3SA)-H(15A)	107.9(15)
N(1A) -C(2A) -N(21A)	117.3(12)	C(1"A)-C(3SA)-H(14A)	107.6(15)
N(1A) -C(2A) -N(3A)	127.8(12)	C(1"A)-C(3SA)-H(15A)	108.0(15)
N(21A)-C(2A) -N(3A)	114.9(12)	H(14A)-C(3SA)-H(15A)	109.4(18)
C(2A) -N(21A)-H(1A)	105.2(11)	C(3"A)-C(2"A)-C(1"A)	120.2(9)
C(2A) -N(21A)-H(2A)	127.0(12)	C(3"A)-C(2"A)-H(16A)	119.9(11)
H(1A) -N(21A)-H(2A)	127.2(12)	C(1"A)-C(2"A)-H(16A)	119.9(11)
C(2A) -N(3A) -C(4A)	115.3(11)	C(2"A)-C(3"A)-C(4"A)	119.9(9)
N(3A) -C(4A) -N(41A)	117.2(12)	C(2"A)-C(3"A)-H(17A)	120.2(11)
N(3A) -C(4A) -C(5A)	123.8(13)	C(4"A)-C(3"A)-H(17A)	119.9(11)
N(41A)-C(4A) -C(5A)	118.9(12)	C(3"A)-C(4"A)-C(5"A)	120.0(9)
C(4A) -N(41A)-H(3A)	112.2(10)	C(3"A)-C(4"A)-H(18A)	120.0(11)
C(4A) -N(41A)-H(4A)	123.7(11)	C(5"A)-C(4"A)-H(18A)	120.1(11)
H(3A) -N(41A)-H(4A)	124.1(10)	C(4"A)-C(5"A)-C(6"A)	120.0(9)
C(4A) -C(5A) -C(6A)	114.6(12)	C(4"A)-C(5"A)-H(19A)	120.0(11)
C(4A) -C(5A) -C(1'A)	122.4(12)	C(6"A)-C(5"A)-H(19A)	120.0(11)
C(6A) -C(5A) -C(1'A)	122.7(11)	C(5"A)-C(6"A)-C(1"A)	120.1(9)
N(1A) -C(6A) -C(5A)	121.6(12)	C(5"A)-C(6"A)-H(20A)	120.0(11)
N(1A) -C(6A) -C(61A)	113.5(11)	C(1"A)-C(6"A)-H(20A)	120.0(11)
C(5A) -C(6A) -C(61A)	124.8(12)	C(3SA)-C(1"A)-C(2"A)	122.3(10)
C(6A) -C(61A)-C(62A)	119.0(14)	C(3SA)-C(1"A)-C(6"A)	117.8(10)
C(6A) -C(61A)-H(5A)	108.6(15)	C(2"A)-C(1"A)-C(6"A)	119.9(9)
C(6A) -C(61A)-H(6A)	106.6(15)	C(4'A)-N(4'A)-O(1'A)	120.6(15)
C(62A)-C(61A)-H(5A)	110.8(17)	C(4'A)-N(4'A)-O(2'A)	118.3(14)
C(62A)-C(61A)-H(6A)	101.8(16)	O(1'A)-N(4'A)-O(2'A)	121.0(15)
H(5A) -C(61A)-H(6A)	109.5(18)	C(2B) -N(1B) -C(6B)	114.8(11)
C(61A)-C(62A)-H(7A)	124.4(21)	N(1B) -C(2B) -N(21B)	115.9(11)
C(61A)-C(62A)-H(8A)	116.9(20)	N(1B) -C(2B) -N(3B)	127.9(12)
C(61A)-C(62A)-H(9A)	82.9(18)	N(21B)-C(2B) -N(3B)	116.2(11)
H(7A) -C(62A)-H(8A)	109.4(23)	C(2B) -N(21B)-H(1B)	117.4(10)
H(7A) -C(62A)-H(9A)	109.5(23)	C(2B) -N(21B)-H(2B)	115.9(9)
H(8A) -C(62A)-H(9A)	109.4(23)	H(1B) -N(21B)-H(2B)	126.8(10)
C(5A) -C(1'A)-C(2'A)	118.8(12)	C(2B) -N(3B) -C(4B)	115.2(11)
C(5A) -C(1'A)-C(6'A)	121.9(12)	N(3B) -C(4B) -N(41B)	114.5(11)
C(2'A)-C(1'A)-C(6'A)	119.3(13)	N(3B) -C(4B) -C(5B)	123.6(11)
C(1'A)-C(2'A)-C(3'A)	119.3(13)	N(41B)-C(4B) -C(5B)	121.9(11)
C(1'A)-C(2'A)-H(10A)	120.3(15)	C(4B) -N(41B)-H(4B)	129.7(10)
C(3'A)-C(2'A)-H(10A)	120.4(15)	C(4B) -C(5B) -C(6B)	115.1(11)
C(2'A)-C(3'A)-C(4'A)	117.4(14)	C(4B) -C(5B) -C(1'B)	120.9(11)

C(2'A)-C(3'A)-N(3SA)	120.0(13)	C(6B) -C(5B) -C(1'B)	124.0(11)
C(4'A)-C(3'A)-N(3SA)	122.4(14)	N(1B) -C(6B) -C(5B)	123.4(12)
C(3'A)-C(4'A)-C(5'A)	122.2(14)	N(1B) -C(6B) -C(61B)	112.2(12)
C(3'A)-C(4'A)-N(4'A)	123.5(14)	C(5B) -C(6B) -C(61B)	124.3(12)
C(5'A)-C(4'A)-N(4'A)	114.1(14)	C(6B) -C(61B)-C(62B)	109.8(14)
C(4'A)-C(5'A)-C(6'A)	119.9(14)	C(6B) -C(61B)-H(5B)	108.6(15)
C(4'A)-C(5'A)-H(11A)	120.3(15)	C(6B) -C(61B)-H(6B)	110.5(15)
C(6'A)-C(5'A)-H(11A)	119.8(16)	C(62B)-C(61B)-H(5B)	107.3(16)
C(1'A)-C(6'A)-C(5'A)	121.6(14)	C(62B)-C(61B)-H(6B)	111.2(17)
C(1'A)-C(6'A)-H(12A)	119.1(16)	H(5B) -C(61B)-H(6B)	109.4(18)
C(5'A)-C(6'A)-H(12A)	119.4(17)	C(61B)-C(62B)-H(7B)	104.6(21)
C(3'A)-N(3SA)-C(3SA)	124.2(13)	C(61B)-C(62B)-H(8B)	103.0(21)
C(3'A)-N(3SA)-H(13A)	122.7(13)	C(61B)-C(62B)-H(9B)	120.2(22)
C(3SA)-N(3SA)-H(13A)	112.1(13)	H(7B) -C(62B)-H(8B)	109.4(26)
N(3SA)-C(3SA)-C(1"A)	115.3(12)	H(7B) -C(62B)-H(9B)	109.6(26)
N(3SA)-C(3SA)-H(14A)	108.5(15)	H(8B) -C(62B)-H(9B)	109.5(26)
C(5B) -C(1'B)-C(2'B)	121.6(12)	C(1"B)-C(3SB)-H(14B)	111.7(14)
C(5B) -C(1'B)-C(6'B)	118.5(12)	C(1"B)-C(3SB)-H(15B)	103.3(13)
C(2'B)-C(1'B)-C(6'B)	119.9(13)	H(14B)-C(3SB)-H(15B)	109.3(16)
C(1'B)-C(2'B)-C(3'B)	124.2(12)	C(3"B)-C(2"B)-C(1"B)	120.0(11)
C(1'B)-C(2'B)-H(10B)	118.1(13)	C(3"B)-C(2"B)-H(16B)	120.0(13)
C(3'B)-C(2'B)-H(10B)	117.6(13)	C(1"B)-C(2"B)-H(16B)	120.0(13)
C(2'B)-C(3'B)-C(4'B)	111.5(12)	C(2"B)-C(3"B)-C(4"B)	120.0(11)
C(2'B)-C(3'B)-N(3SB)	123.3(12)	C(2"B)-C(3"B)-H(17B)	119.9(13)
C(4'B)-C(3'B)-N(3SB)	125.2(13)	C(4"B)-C(3"B)-H(17B)	120.1(13)
C(3'B)-C(4'B)-C(5'B)	124.2(14)	C(3"B)-C(4"B)-C(5"B)	120.0(11)
C(3'B)-C(4'B)-N(4'B)	121.2(13)	C(3"B)-C(4"B)-H(18B)	120.0(13)
C(5'B)-C(4'B)-N(4'B)	114.5(13)	C(5"B)-C(4"B)-H(18B)	120.1(13)
C(4'B)-C(5'B)-C(6'B)	120.1(14)	C(4"B)-C(5"B)-C(6"B)	120.0(11)
C(4'B)-C(5'B)-H(11B)	121.0(16)	C(4"B)-C(5"B)-H(19B)	120.0(13)
C(6'B)-C(5'B)-H(11B)	118.9(16)	C(6"B)-C(5"B)-H(19B)	120.0(13)
C(1'B)-C(6'B)-C(5'B)	119.9(13)	C(5"B)-C(6"B)-C(1"B)	120.0(11)
C(1'B)-C(6'B)-H(12B)	119.9(15)	C(5"B)-C(6"B)-H(20B)	120.0(13)
C(5'B)-C(6'B)-H(12B)	120.2(15)	C(1"B)-C(6"B)-H(20B)	119.9(13)
C(3'B)-N(3SB)-C(3SB)	122.8(12)	C(3SB)-C(1"B)-C(2"B)	118.9(11)
C(3'B)-N(3SB)-H(13B)	114.4(12)	C(3SB)-C(1"B)-C(6"B)	121.1(11)
C(3SB)-N(3SB)-H(13B)	121.3(12)	C(2"B)-C(1"B)-C(6"B)	120.0(11)
N(3SB)-C(3SB)-C(1"B)	114.2(11)	C(4'B)-N(4'B)-O(1'B)	115.8(13)
N(3SB)-C(3SB)-H(14B)	110.3(14)	C(4'B)-N(4'B)-O(2'B)	120.4(14)
N(3SB)-C(3SB)-H(15B)	107.7(14)	O(1'B)-N(4'B)-O(2'B)	123.6(15)

Table 5.11 Torsion angles(degrees) with standard deviations

C(6A) -N(1A) -C(2A) -N(21A)	171.1(12)	C(2'A)-C(3'A)-C(4'A)-C(5'A)	3.5(23)
C(6A) -N(1A) -C(2A) -N(3A)	-8.3(20)	C(2'A)-C(3'A)-C(4'A)-N(4'A)	178.5(14)

C(2A) -N(1A) -C(6A) -C(5A)	5.7(18)
C(2A) -N(1A) -C(6A) -C(61A)	-175.3(12)
N(1A) -C(2A) -N(21A)-H(1A)	-7.9(16)
N(1A) -C(2A) -N(21A)-H(2A)	163.5(12)
N(3A) -C(2A) -N(21A)-H(1A)	171.6(11)
N(3A) -C(2A) -N(21A)-H(2A)	-17.0(20)
N(1A) -C(2A) -N(3A) -C(4A)	5.9(20)
N(21A)-C(2A) -N(3A) -C(4A)	-173.5(12)
C(2A) -N(3A) -C(4A) -N(41A)	-179.0(12)
C(2A) -N(3A) -C(4A) -C(5A)	-1.0(20)
N(3A) -C(4A) -N(41A)-H(3A)	2.5(17)
N(3A) -C(4A) -N(41A)-H(4A)	-174.6(11)
C(5A) -C(4A) -N(41A)-H(3A)	-175.6(11)
C(5A) -C(4A) -N(41A)-H(4A)	7.3(19)
N(3A) -C(4A) -C(5A) -C(6A)	-0.7(20)
N(3A) -C(4A) -C(5A) -C(1'A)	174.2(13)
N(41A)-C(4A) -C(5A) -C(6A)	177.3(12)
N(41A)-C(4A) -C(5A) -C(1'A)	-7.8(20)
C(4A) -C(5A) -C(6A) -N(1A)	-1.7(18)
C(4A) -C(5A) -C(6A) -C(61A)	179.5(13)
C(1'A)-C(5A) -C(6A) -N(1A)	-176.6(12)
C(1'A)-C(5A) -C(6A) -C(61A)	4.6(21)
C(4A) -C(5A) -C(1'A)-C(2'A)	117.8(15)
C(4A) -C(5A) -C(1'A)-C(6'A)	-62.2(19)
C(6A) -C(5A) -C(1'A)-C(2'A)	-67.7(18)
C(6A) -C(5A) -C(1'A)-C(6'A)	112.3(16)
N(1A) -C(6A) -C(61A)-C(62A)	-53.0(19)
N(1A) -C(6A) -C(61A)-H(5A)	179.1(15)
N(1A) -C(6A) -C(61A)-H(6A)	61.2(18)
C(5A) -C(6A) -C(61A)-C(62A)	126.0(17)
C(5A) -C(6A) -C(61A)-H(5A)	-2.0(22)
C(5A) -C(6A) -C(61A)-H(6A)	-119.9(17)
C(6A) -C(61A)-C(62A)-H(7A)	173.8(21)
C(6A) -C(61A)-C(62A)-H(8A)	30.8(28)
C(6A) -C(61A)-C(62A)-H(9A)	-77.3(20)
H(5A) -C(61A)-C(62A)-H(7A)	-59.2(29)
H(5A) -C(61A)-C(62A)-H(8A)	157.8(22)
H(5A) -C(61A)-C(62A)-H(9A)	49.6(22)
H(6A) -C(61A)-C(62A)-H(7A)	57.2(28)
H(6A) -C(61A)-C(62A)-H(8A)	-85.9(25)
H(6A) -C(61A)-C(62A)-H(9A)	166.0(19)
C(5A) -C(1'A)-C(2'A)-C(3'A)	176.6(12)
C(5A) -C(1'A)-C(2'A)-H(10A)	-2.2(22)
C(6'A)-C(1'A)-C(2'A)-C(3'A)	-3.4(21)
C(6'A)-C(1'A)-C(2'A)-H(10A)	177.8(16)
C(5A) -C(1'A)-C(6'A)-C(5'A)	-177.1(14)
C(5A) -C(1'A)-C(6'A)-H(12A)	3.5(24)
C(2'A)-C(1'A)-C(6'A)-C(5'A)	2.9(22)
C(2'A)-C(1'A)-C(6'A)-H(12A)	-176.4(16)
N(3SA)-C(3'A)-C(4'A)-C(5'A)	179.4(14)
N(3SA)-C(3'A)-C(4'A)-N(4'A)	-5.6(24)
C(2'A)-C(3'A)-N(3SA)-C(3SA)	-3.7(22)
C(2'A)-C(3'A)-N(3SA)-H(13A)	-171.8(14)
C(4'A)-C(3'A)-N(3SA)-C(3SA)	-179.5(14)
C(4'A)-C(3'A)-N(3SA)-H(13A)	12.4(24)
C(3'A)-C(4'A)-C(5'A)-C(6'A)	-4.1(23)
C(3'A)-C(4'A)-C(5'A)-H(11A)	176.9(16)
N(4'A)-C(4'A)-C(5'A)-C(6'A)	-179.5(14)
N(4'A)-C(4'A)-C(5'A)-H(11A)	1.5(23)
C(3'A)-C(4'A)-N(4'A)-O(1'A)	170.6(15)
C(3'A)-C(4'A)-N(4'A)-O(2'A)	-12.7(24)
C(5'A)-C(4'A)-N(4'A)-O(1'A)	-14.1(22)
C(5'A)-C(4'A)-N(4'A)-O(2'A)	162.7(15)
C(4'A)-C(5'A)-C(6'A)-C(1'A)	0.8(23)
C(4'A)-C(5'A)-C(6'A)-H(12A)	-179.8(17)
H(11A)-C(5'A)-C(6'A)-C(1'A)	179.8(16)
H(11A)-C(5'A)-C(6'A)-H(12A)	-0.8(26)
C(3'A)-N(3SA)-C(3SA)-C(1"A)	82.5(18)
C(3'A)-N(3SA)-C(3SA)-H(14A)	-38.2(22)
C(3'A)-N(3SA)-C(3SA)-H(15A)	-156.7(16)
H(13A)-N(3SA)-C(3SA)-C(1"A)	-108.3(14)
H(13A)-N(3SA)-C(3SA)-H(14A)	131.0(16)
H(13A)-N(3SA)-C(3SA)-H(15A)	12.5(20)
N(3SA)-C(3SA)-C(1"A)-C(2"A)	-0.6(18)
N(3SA)-C(3SA)-C(1"A)-C(6"A)	-178.2(11)
H(14A)-C(3SA)-C(1"A)-C(2"A)	120.6(15)
H(14A)-C(3SA)-C(1"A)-C(6"A)	-57.0(18)
H(15A)-C(3SA)-C(1"A)-C(2"A)	-121.4(15)
H(15A)-C(3SA)-C(1"A)-C(6"A)	61.0(18)
C(1"A)-C(2"A)-C(3"A)-C(4"A)	0.0(15)
C(1"A)-C(2"A)-C(3"A)-H(17A)	179.9(11)
H(16A)-C(2"A)-C(3"A)-C(4"A)	-179.9(11)
H(16A)-C(2"A)-C(3"A)-H(17A)	0.0(18)
C(3"A)-C(2"A)-C(1"A)-C(3SA)	-177.5(11)
C(3"A)-C(2"A)-C(1"A)-C(6"A)	0.0(15)
H(16A)-C(2"A)-C(1"A)-C(3SA)	2.4(18)
H(16A)-C(2"A)-C(1"A)-C(6"A)	-180.0(11)
C(2"A)-C(3"A)-C(4"A)-C(5"A)	-0.1(15)
C(2"A)-C(3"A)-C(4"A)-H(18A)	180.0(11)
H(17A)-C(3"A)-C(4"A)-C(5"A)	-180.0(11)
H(17A)-C(3"A)-C(4"A)-H(18A)	0.1(18)
C(3"A)-C(4"A)-C(5"A)-C(6"A)	0.1(15)
C(3"A)-C(4"A)-C(5"A)-H(19A)	-179.9(11)
H(18A)-C(4"A)-C(5"A)-C(6"A)	180.0(11)
H(18A)-C(4"A)-C(5"A)-H(19A)	0.0(18)
C(4"A)-C(5"A)-C(6"A)-C(1"A)	0.0(15)
C(4"A)-C(5"A)-C(6"A)-H(20A)	180.0(11)
H(19A)-C(5"A)-C(6"A)-C(1"A)	-180.0(11)

C(1'A)-C(2'A)-C(3'A)-C(4'A)	0.3(21)
C(1'A)-C(2'A)-C(3'A)-N(3SA)	-175.7(13)
H(10A)-C(2'A)-C(3'A)-C(4'A)	179.1(16)
H(10A)-C(2'A)-C(3'A)-N(3SA)	3.1(23)
H(20A)-C(6"A)-C(1"A)-C(2"A)	180.0(11)
C(6B) -N(1B) -C(2B) -N(21B)	178.7(11)
C(6B) -N(1B) -C(2B) -N(3B)	0.9(19)
C(2B) -N(1B) -C(6B) -C(5B)	-0.7(18)
C(2B) -N(1B) -C(6B) -C(61B)	176.6(11)
N(1B) -C(2B) -N(21B)-H(1B)	-10.8(17)
N(1B) -C(2B) -N(21B)-H(2B)	170.0(10)
N(3B) -C(2B) -N(21B)-H(1B)	167.2(11)
N(3B) -C(2B) -N(21B)-H(2B)	-12.0(16)
N(1B) -C(2B) -N(3B) -C(4B)	-1.6(19)
N(21B)-C(2B) -N(3B) -C(4B)	-179.3(11)
C(2B) -N(3B) -C(4B) -N(41B)	-179.6(11)
C(2B) -N(3B) -C(4B) -C(5B)	2.0(17)
N(3B) -C(4B) -N(41B)-H(4B)	167.8(11)
C(5B) -C(4B) -N(41B)-H(4B)	-13.9(19)
N(3B) -C(4B) -C(5B) -C(6B)	-1.9(18)
N(3B) -C(4B) -C(5B) -C(1'B)	177.6(12)
N(41B)-C(4B) -C(5B) -C(6B)	179.9(11)
N(41B)-C(4B) -C(5B) -C(1'B)	-0.6(18)
C(4B) -C(5B) -C(6B) -N(1B)	1.2(19)
C(4B) -C(5B) -C(6B) -C(61B)	-175.8(12)
C(1'B)-C(5B) -C(6B) -N(1B)	-178.3(12)
C(1'B)-C(5B) -C(6B) -C(61B)	4.7(21)
C(4B) -C(5B) -C(1'B)-C(2'B)	69.5(17)
C(4B) -C(5B) -C(1'B)-C(6'B)	-111.8(14)
C(6B) -C(5B) -C(1'B)-C(2'B)	-111.1(16)
C(6B) -C(5B) -C(1'B)-C(6'B)	67.6(18)
N(1B) -C(6B) -C(61B)-C(62B)	-83.6(16)
N(1B) -C(6B) -C(61B)-H(5B)	159.4(14)
N(1B) -C(6B) -C(61B)-H(6B)	39.4(19)
C(5B) -C(6B) -C(61B)-C(62B)	93.7(17)
C(5B) -C(6B) -C(61B)-H(5B)	-23.3(21)
C(5B) -C(6B) -C(61B)-H(6B)	-143.3(16)
C(6B) -C(61B)-C(62B)-H(7B)	-174.9(19)
C(6B) -C(61B)-C(62B)-H(8B)	70.7(22)
C(6B) -C(61B)-C(62B)-H(9B)	-51.4(28)
H(5B) -C(61B)-C(62B)-H(7B)	-57.0(25)
H(5B) -C(61B)-C(62B)-H(8B)	-171.4(21)
H(5B) -C(61B)-C(62B)-H(9B)	66.5(28)
H(6B) -C(61B)-C(62B)-H(7B)	62.6(25)
H(6B) -C(61B)-C(62B)-H(8B)	-51.8(25)
H(6B) -C(61B)-C(62B)-H(9B)	-173.9(24)
C(5B) -C(1'B)-C(2'B)-C(3'B)	-177.9(12)
C(5B) -C(1'B)-C(2'B)-H(10B)	1.9(21)
C(6'B)-C(1'B)-C(2'B)-C(3'B)	3.4(21)
H(19A)-C(5"A)-C(6"A)-H(20A)	0.0(18)
C(5"A)-C(6"A)-C(1"A)-C(3SA)	177.6(11)
C(5"A)-C(6"A)-C(1"A)-C(2"A)	-0.1(15)
H(20A)-C(6"A)-C(1"A)-C(3SA)	-2.4(17)
C(2'B)-C(3'B)-C(4'B)-N(4'B)	-179.8(13)
N(3SB)-C(3'B)-C(4'B)-C(5'B)	-178.7(14)
N(3SB)-C(3'B)-C(4'B)-N(4'B)	-1.5(22)
C(2'B)-C(3'B)-N(3SB)-C(3SB)	3.0(21)
C(2'B)-C(3'B)-N(3SB)-H(13B)	169.4(12)
C(4'B)-C(3'B)-N(3SB)-C(3SB)	-175.1(13)
C(4'B)-C(3'B)-N(3SB)-H(13B)	-8.8(21)
C(3'B)-C(4'B)-C(5'B)-C(6'B)	-3.8(24)
C(3'B)-C(4'B)-C(5'B)-H(11B)	177.9(16)
N(4'B)-C(4'B)-C(5'B)-C(6'B)	178.8(14)
N(4'B)-C(4'B)-C(5'B)-H(11B)	0.5(23)
C(3'B)-C(4'B)-N(4'B)-O(1'B)	8.2(21)
C(3'B)-C(4'B)-N(4'B)-O(2'B)	-175.9(14)
C(5'B)-C(4'B)-N(4'B)-O(1'B)	-174.4(14)
C(5'B)-C(4'B)-N(4'B)-O(2'B)	1.6(22)
C(4'B)-C(5'B)-C(6'B)-C(1'B)	4.0(22)
C(4'B)-C(5'B)-C(6'B)-H(12B)	-176.9(16)
H(11B)-C(5'B)-C(6'B)-C(1'B)	-177.6(16)
H(11B)-C(5'B)-C(6'B)-H(12B)	1.5(26)
C(3'B)-N(3SB)-C(3SB)-C(1"B)	78.6(16)
C(3'B)-N(3SB)-C(3SB)-H(14B)	-154.6(15)
C(3'B)-N(3SB)-C(3SB)-H(15B)	-35.5(19)
H(13B)-N(3SB)-C(3SB)-C(1"B)	-86.8(16)
H(13B)-N(3SB)-C(3SB)-H(14B)	40.0(20)
H(13B)-N(3SB)-C(3SB)-H(15B)	159.2(14)
N(3SB)-C(3SB)-C(1"B)-C(2"B)	63.8(16)
N(3SB)-C(3SB)-C(1"B)-C(6"B)	-116.2(14)
H(14B)-C(3SB)-C(1"B)-C(2"B)	-62.2(18)
H(14B)-C(3SB)-C(1"B)-C(6"B)	117.8(16)
H(15B)-C(3SB)-C(1"B)-C(2"B)	-179.6(13)
H(15B)-C(3SB)-C(1"B)-C(6"B)	0.5(18)
C(1"B)-C(2"B)-C(3"B)-C(4"B)	0.1(18)
C(1"B)-C(2"B)-C(3"B)-H(17B)	-179.9(13)
H(16B)-C(2"B)-C(3"B)-C(4"B)	-180.0(13)
H(16B)-C(2"B)-C(3"B)-H(17B)	0.0(21)
C(3"B)-C(2"B)-C(1"B)-C(3SB)	179.9(11)
C(3"B)-C(2"B)-C(1"B)-C(6"B)	-0.1(18)
H(16B)-C(2"B)-C(1"B)-C(3SB)	0.0(20)
H(16B)-C(2"B)-C(1"B)-C(6"B)	180.0(13)
C(2"B)-C(3"B)-C(4"B)-C(5"B)	-0.1(18)
C(2"B)-C(3"B)-C(4"B)-H(18B)	-179.9(13)
H(17B)-C(3"B)-C(4"B)-C(5"B)	179.9(13)
H(17B)-C(3"B)-C(4"B)-H(18B)	0.1(21)
C(3"B)-C(4"B)-C(5"B)-C(6"B)	0.1(18)
C(3"B)-C(4"B)-C(5"B)-H(19B)	-180.0(13)

C(6'B)-C(1'B)-C(2'B)-H(10B)	-176.7(14)	H(18B)-C(4"B)-C(5"B)-C(6"B)	179.9(13)
C(5B) -C(1'B)-C(6'B)-C(5'B)	177.4(13)	H(18B)-C(4"B)-C(5"B)-H(19B)	-0.1(21)
C(5B) -C(1'B)-C(6'B)-H(12B)	-1.7(22)	C(4"B)-C(5"B)-C(6"B)-C(1"B)	-0.1(18)
C(2'B)-C(1'B)-C(6'B)-C(5'B)	-3.9(21)	C(4"B)-C(5"B)-C(6"B)-H(20B)	179.9(13)
C(2'B)-C(1'B)-C(6'B)-H(12B)	177.0(15)	H(19B)-C(5"B)-C(6"B)-C(1"B)	180.0(13)
C(1'B)-C(2'B)-C(3'B)-C(4'B)	-2.9(19)	H(19B)-C(5"B)-C(6"B)-H(20B)	-0.1(21)
C(1'B)-C(2'B)-C(3'B)-N(3SB)	178.7(13)	C(5"B)-C(6"B)-C(1"B)-C(3SB)	-180.0(12)
H(10B)-C(2'B)-C(3'B)-C(4'B)	177.3(14)	C(5"B)-C(6"B)-C(1"B)-C(2"B)	0.1(18)
H(10B)-C(2'B)-C(3'B)-N(3SB)	-1.1(21)	H(20B)-C(6"B)-C(1"B)-C(3SB)	0.1(20)
C(2'B)-C(3'B)-C(4'B)-C(5'B)	3.0(21)	H(20B)-C(6"B)-C(1"B)-C(2"B)	-179.8(13)

Table 5.12 Hydrogen bond contact distances

Hydrogen Bond	Donor-Acceptor Distance (Å)
$N(41B)-H(3B)_{I}\ldots N(3A)_{I}$	2.920
$N(21A)-H(2A)_{I}\ldots N(3B)_{I}$	2.986
$N(21B)-H(1B)_{I}\ldots N(1B)_{II}$	2.985
$N(41B)-H(4B)_{I}\ldots N(1A)_{III}$	3.019
$N(21B)-H(2B)_{I}\ldots O(2'B)_{IV}$	3.107
$N(41B)-H(4B)_{I}\ldots O(1'B)_{III}$	3.135
$N(41A)-H(4A)_{I}\ldots O(1'A)_{V}$	3.184

The subscripts I, II, III, IV and V refer to the equivalent positions X, Y, Z; 1-X, 1-Y, 1-Z; $-\frac{1}{2}$+X, $\frac{1}{2}$-Y, Z; 1+X, Y, Z; -X, -Y, -Z.

Figure 5.3 Isobenzoprim Molecular plot and numbering scheme

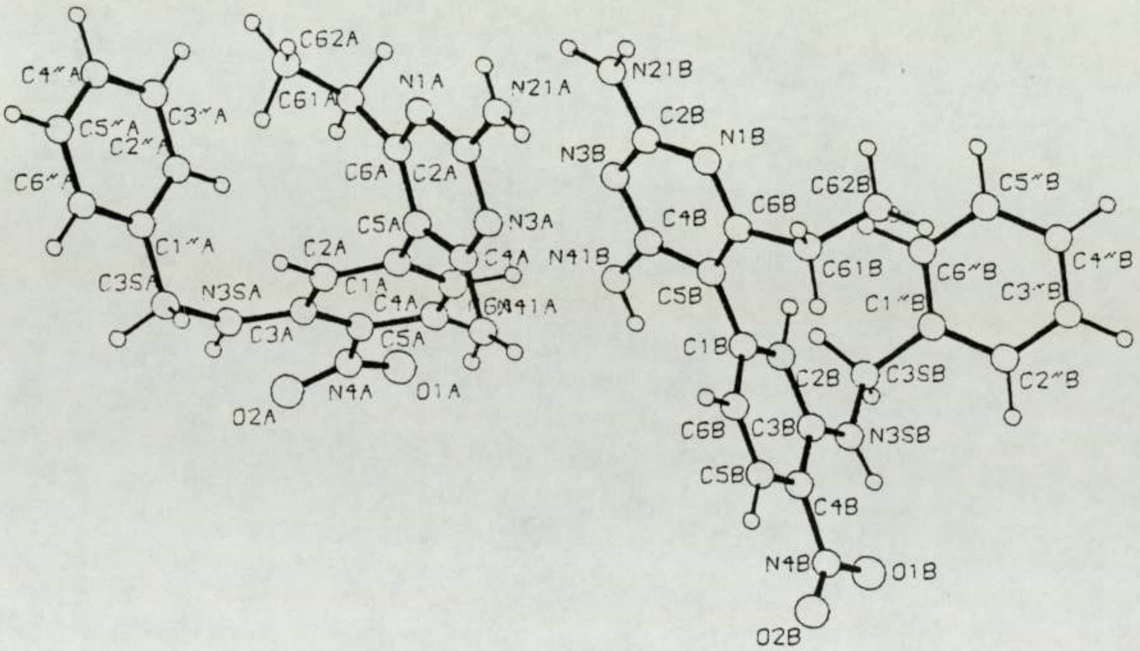
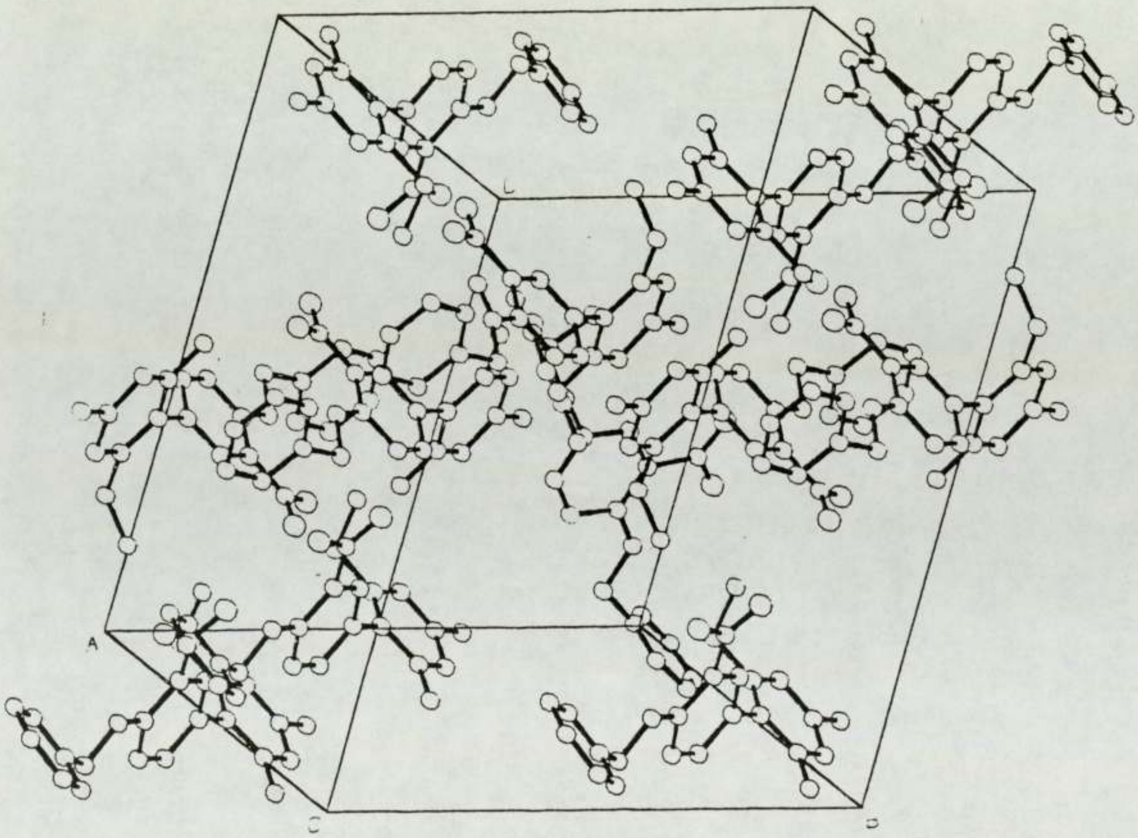


Figure 5.4 Isobenzoprim Unit cell packing diagram



5.3 The Crystal Structure of 2,4-diamino-5-(3-nitro-4-butylamino)phenyl-6-ethyl pyrimidine ethanesulphonate salt. monohydrate (Butaprim).

5.3.1 Experimental

A crystalline sample of the compound was obtained (76) and from it a suitable crystal was selected. The quality of the specimen crystal was confirmed by examining its optical birefringent properties with a polarising microscope. The single crystal was then mounted onto a glass filament secured to a goniometer head stud by the hot wax method.

5.3.2 Crystal Data

$C_{16}H_{23}N_6O_2 . C_2H_5SO_3 . H_2O$, M = 458.538, F(000) = 936. Monoclinic, a = 15.518(6), b = 8.482(4), c = 18.432(7) Å, β = 103.64(3)°, V = 2358 $Å^3$ (by least squares refinement on diffractometer angles for 25 automatically centred reflections, λ = 0.71069 Å), space group $P2_1/n$, Z = 4, Dx = 1.292 gcm^{-3}, Dm = 1.287 gcm^{-3} (method of flotation in solvent mixture). Orange diamonds. Crystal dimensions 0.32 x 0.28 x 0.20 mm, μ (Mo-Kα) = 1.37 cm^{-1}.

5.3.3 Data Collection and Processing

CAD4 diffractometer, w/2θ mode with w scan width = 0.9 + 0.35 tanθ, w scan speed 0.4 - 2.6 deg min^{-1}, graphite monochromated Mo-Kα radiation; 4603 reflections measured ($2° \leq \theta \leq 25°$ + h + k ± l), 4118 unique (merging R = 0.0238, no correction for absorption), giving 1741 with I $> 2\sigma(I)$, number of parameters refined 337. Linear and approximately isotropic crystal decay ca. 2.3% (for three standard reflections checked every 7200 s x-ray exposure time).

5.3.4 Structure Determination and Refinement

Data reduction was followed by direct methods (SHELX76) and electron density synthesis. All hydrogen atoms were located by difference Fourier analysis during isotropic least squares refinement, but further refinement with all non-hydrogen atoms anisotropic gave poor convergence and unacceptable geometry where hydrogen atoms were involved. All hydrogen atoms, except those of the 2 and 4-amino, ring proton and a water were thereafter constrained to refine under idealised geometry (AFIX-SHELX76) and common temperature factors for groups. Bond lengths for C(6)-C(61)-C(62) were found to deviate considerably from those observed in other related structures having this 6-ethyl substituent. An attempt was made to refine this moiety while restraining the bond lengths to average values. This was not greatly successful and it is interesting to note that other workers have found this region of the ethyl-pyrimidine moiety to be ill defined and displaying large thermal motion (29) (75).

Convergence of full matrix least squares refinement, with all non-hydrogen atoms anisotropic, was reached after 20 cycles using a damping factor of 10; the weighting scheme W = $1.8554/[\sigma^2 (F_o)]$, with $\sigma(F_o)$ from counting statistics, gave satisfactory agreement analyses. Final R and R_W values are 0.0647 and 0.0564. A final electron density showed 2 peaks of residual density at ~ 0.3 e A^{-3}, and these were located close to the 6-ethyl group, after which there was no feature > 0.25 e A^{-3}. At convergence (Δ/σ) max = -.568 [C(62) AFIX C rotation], more commonly $\Delta/\sigma = 0.01$.

Table 5.13 Positional parameters (fractional coordinates * 10**4, hydrogens * 10**3) with estimated standard deviations in parentheses.

ATOM	x/a	y/b	z/c	
N(1)	-1091(4)	-0468(6)	0267(3)	
C(2)	-1006(4)	-1639(8)	-0206(3)	
N(21)	-1355(4)	-1400(7)	-0931(3)	
N(3)	-0588(3)	-2982(6)	0022(2)	
C(4)	-0266(4)	-3162(7)	0763(3)	
N(41)	0139(4)	-4508(7)	0979(3)	
C(5)	-0383(4)	-2011(7)	1296(3)	
C(6)	-0801(5)	-0635(8)	1033(3)	
C(61)	-1278(8)	0614(9)	1533(4)	
C(62)	-0703(7)	173(2)	1490(6)	
C(1′)	-0057(4)	-2334(8)	2114(3)	
C(2′)	-0549(4)	-3175(8)	2491(3)	
C(3′)	-0259(5)	-3467(8)	3258(3)	
C(4′)	0563(5)	-2886(7)	3673(3)	
C(5′)	1073(5)	-2045(8)	3266(3)	
C(6′)	0785(5)	-1782(8)	2515(3)	
N(3′)	-0838(4)	-4396(9)	3588(3)	
O(31′)	-0656(4)	-4584(7)	4270(3)	
O(32′)	-1492(4)	-5005(9)	3191(3)	
N(1")	0875(4)	-3116(8)	4415(3)	
C(1")	1701(5)	-253(1)	4879(4)	
C(2")	1454(7)	-130(1)	5417(6)	
C(3")	2133(9)	-072(2)	5944(5)	
C(4")	1909(8)	044(1)	6442(6)	
S(1E)	2442(1)	2516(3)	4020(1)	
O(1E)	3051(4)	2765(6)	4733(2)	
O(2E)	2548(3)	3736(6)	3499(2)	
O(3E)	2471(3)	0938(6)	3750(3)	
C(1E)	1369(7)	274(1)	4184(5)	
C(2E)	0653(6)	246(1)	3500(6)	
O(1W)	1607(4)	-5979(6)	1999(3)	
H(1)	-143(4)	048(8)	004(4)	
H(2)	-175(4)	-057(6)	-108(3)	
H(3)	-136(3)	-213(5)	-123(3)	
H(4)	034(4)	-524(7)	057(3)	
H(5)	041(4)	-465(6)	136(3)	
H(6)	-1245	0217	2096	*
H(7)	-1954	0913	1265	*
H(8)	-0960	2601	1810	*
H(9)	-0711	2192	0943	*
H(10)	-0031	1432	1774	*
H(11)	-1182	-3636	2193	*
H(12)	1712	-1591	3555	*
H(13)	1205	-1148	2223	*
H(14)	047(3)	-345(5)	461(3)	
H(15)	2073	-3486	5193	*
H(16)	2093	-1981	4536	*
H(17)	0996	-1854	5697	*
H(18)	1131	-0326	5085	*
H(19)	2452	-1691	6279	*
H(20)	2594	-0174	5664	*
H(21)	2479	0863	6849	*

Table 5.13 contd.

ATOM	x/a	y/b	z/c	
H(22)	1481	-0208	6721	*
H(23)	1550	1421	6144	*
H(24)	1305	3925	4379	*
H(25)	1294	1908	4608	*
H(26)	0000	2588	3603	*
H(27)	0725	1282	3297	*
H(28)	0741	3310	3088	*
H(29)	-186(4)	-422(7)	-239(3)	
H(30)	-193(4)	-472(6)	-181(3)	

* Hydrogens unrefined; constrained by AFIX in SHELX least squares refinement.

Table 5.14 Anisotropic temperature factors, and isotropic temperature factors where shown; with standard deviations in parentheses.

ATOM	U11	U22	U33	U23	U13	U12
N(1)	.103(5)	.042(4)	.042(3)	.002(3)	.000(3)	.027(3)
C(2)	.059(4)	.055(4)	.037(4)	.000(4)	.005(3)	.006(4)
N(21)	.094(5)	.059(4)	.039(3)	.000(3)	.005(3)	.035(4)
N(3)	.068(4)	.054(4)	.031(3)	.004(3)	.003(3)	.018(3)
C(4)	.056(4)	.046(4)	.040(3)	.005(3)	.008(3)	.009(4)
N(41)	.100(5)	.066(4)	.035(3)	.006(3)	.005(3)	.042(4)
C(5)	.061(4)	.036(4)	.042(3)	-.003(3)	.008(3)	.003(3)
C(6)	.110(6)	.041(4)	.047(4)	-.004(3)	.004(4)	.015(4)
C(61)	.33(1)	.055(5)	.058(5)	.007(4)	-.049(7)	-.032(7)
C(62)	.177(9)	.30(1)	.101(7)	-.028(8)	.014(7)	.006(9)
C(1′)	.066(5)	.047(4)	.036(3)	.001(3)	.011(3)	.005(4)
C(2′)	.049(4)	.071(5)	.041(3)	-.001(3)	.002(3)	.005(4)
C(3′)	.057(5)	.069(5)	.044(4)	.011(4)	.016(4)	.002(4)
C(4′)	.068(5)	.049(5)	.037(3)	-.002(3)	.009(3)	.012(4)
C(5′)	.075(5)	.067(5)	.046(4)	.004(3)	-.001(4)	-.020(4)
C(6′)	.077(5)	.062(4)	.052(4)	.010(4)	.016(4)	-.015(4)
N(3′)	.071(5)	.117(5)	.067(4)	.027(4)	.018(4)	-.002(4)
O(31′)	.105(4)	.168(5)	.060(3)	.038(4)	.028(3)	-.009(4)
O(32′)	.089(4)	.231(7)	.102(4)	.052(5)	-.004(4)	-.076(5)
N(1")	.080(5)	.109(5)	.039(3)	.003(3)	.014(3)	.001(4)
C(1")	.100(6)	.143(7)	.046(4)	-.020(5)	-.019(4)	-.011(6)
C(2")	.191(8)	.120(7)	.136(7)	-.031(6)	-.027(7)	-.072(7)
C(3")	.248(9)	.170(9)	.106(7)	-.017(7)	.014(7)	-.021(9)
C(4")	.247(9)	.202(9)	.170(8)	-.098(8)	.005(8)	.028(9)
S(1E)	.103(2)	.055(1)	.056(1)	-.015(1)	.012(1)	-.023(1)
O(1E)	.223(6)	.074(4)	.070(3)	-.004(3)	-.042(4)	-.064(4)
O(2E)	.121(4)	.104(4)	.046(3)	.004(3)	.004(3)	-.056(3)
O(3E)	.080(4)	.069(4)	.135(4)	-.053(3)	.019(3)	-.013(3)
C(1E)	.179(8)	.091(6)	.130(6)	-.005(6)	.097(6)	.015(7)
C(2E)	.106(7)	.124(7)	.218(8)	-.007(8)	.073(7)	.011(7)
O(1W)	.083(4)	.078(4)	.046(3)	.004(3)	.001(3)	-.010(3)

	U ISO
H(1)	.13(1)
H(2)	.07(1)
H(3)	.04(1)
H(4)	.13(1)
H(5)	.06(1)
H(6)	.14(1)
H(7)	.06(1)
H(8)	.39(1)
H(9)	.39(1)
H(10)	.39(1)
H(11)	.05(1)
H(12)	.11(1)
H(13)	.09(1)
H(14)	.03(1)
H(15)	.23(1)
H(16)	.17(1)
H(17)	.25(1)
H(18)	.31(1)
H(19)	.35(1)

Table 5.14 contd.

	U ISO
H(20)	.26(1)
H(21)	.39(1)
H(22)	.39(1)
H(23)	.39(1)
H(24)	.24(1)
H(25)	.22(1)
H(26)	.19(1)
H(27)	.19(1)
H(28)	.19(1)
H(29)	.09(1)
H(30)	.07(1)

Table 5.15 Bond Lengths(A) with standard deviations

N(1) - C(2)	1.348(8)	C(5') -H(12)	1.080(10)
N(1) - C(6)	1.384(9)	C(6') -H(13)	1.081(10)
N(1) - H(1)	1.00(7)	N(3') -O(31')	1.232(9)
C(2) -N(21)	1.334(9)	N(3') -O(32')	1.217(9)
C(2) - N(3)	1.329(8)	N(1") -C(1")	1.451(10)
N(21) - H(2)	0.93(5)	N(1") -H(14)	0.85(5)
N(21) - H(3)	0.83(5)	C(1") -C(2")	1.547(13)
N(3) - C(4)	1.348(7)	C(1") -H(15)	1.082(12)
C(4) -N(41)	1.319(8)	C(1") -H(16)	1.080(12)
C(4) - C(5)	1.427(8)	C(2") -C(3")	1.348(16)
N(41) - H(4)	1.08(6)	C(2") -H(17)	1.079(15)
N(41) - H(5)	0.74(5)	C(2") -H(18)	1.079(15)
C(5) - C(6)	1.367(9)	C(3") -C(4")	1.438(17)
C(5) -C(1')	1.498(9)	C(3") -H(19)	1.080(17)
C(61) -C(62)	1.314(15)	C(3") -H(20)	1.079(17)
C(61) - H(6)	1.081(13)	C(4") -H(21)	1.079(17)
C(61) - H(7)	1.079(13)	C(4") -H(22)	1.079(17)
C(62) - H(8)	1.081(17)	C(4") -H(23)	1.080(17)
C(62) - H(9)	1.080(17)	S(1E) -O(1E)	1.442(6)
C(62) -H(10)	1.080(17)	S(1E) -O(2E)	1.447(5)
C(1') -C(2')	1.351(9)	S(1E) -O(3E)	1.432(6)
C(1') -C(6')	1.420(9)	S(1E) -C(1E)	1.771(10)
C(2') -C(3')	1.401(9)	C(1E) -C(2E)	1.490(14)
C(2') -H(11)	1.079(9)	C(1E) -H(24)	1.080(14)
C(3') -C(4')	1.412(9)	C(1E) -H(25)	1.079(14)
C(3') -N(3')	1.434(9)	C(2E) -H(26)	1.080(14)
C(4') -C(5')	1.406(9)	C(2E) -H(27)	1.080(14)
C(4') -N(1")	1.353(9)	C(2E) -H(28)	1.081(14)
C(5') -C(6')	1.369(10)		

Table 5.16 Angles(degrees) with standard deviations

C(2) - N(1) - C(6)	121.7(6)	C(3') -N(3') -O(32')	119.7(6)
C(2) - N(1) - H(1)	116.3(38)	O(31')-N(3') -O(32')	121.1(7)
C(6) - N(1) - H(1)	121.6(38)	C(4') -N(1") -C(1")	127.0(6)
N(1) - C(2) -N(21)	117.2(6)	C(4') -N(1") -H(14)	110.5(32)
N(1) - C(2) - N(3)	122.8(6)	C(1") -N(1") -H(14)	120.7(32)
N(21) - C(2) - N(3)	120.0(6)	N(1") -C(1") -C(2")	106.9(7)
C(2) -N(21) - H(2)	119.9(33)	N(1") -C(1") -H(15)	110.1(8)
C(2) -N(21) - H(3)	119.4(34)	N(1") -C(1") -H(16)	110.1(8)
H(2) -N(21) - H(3)	117.6(47)	C(2") -C(1") -H(15)	110.1(8)
C(2) - N(3) - C(4)	117.0(5)	C(2") -C(1") -H(16)	110.1(8)

N(3) - C(4) -N(41)	116.1(5)	H(15) -C(1") -H(16)	109.4(9)
N(3) - C(4) - C(5)	122.9(5)	C(1") -C(2") -C(3")	116.1(9)
N(41) - C(4) - C(5)	120.9(6)	C(1") -C(2") -H(17)	107.7(10)
C(4) -N(41) - H(4)	119.2(33)	C(1") -C(2") -H(18)	107.7(10)
C(4) -N(41) - H(5)	123.6(42)	C(3") -C(2") -H(17)	107.8(11)
H(4) -N(41) - H(5)	111.6(53)	C(3") -C(2") -H(18)	107.8(11)
C(4) - C(5) - C(6)	117.8(6)	H(17) -C(2") -H(18)	109.5(12)
C(4) - C(5) -C(1')	120.0(5)	C(2") -C(3") -C(4")	116.4(11)
C(6) - C(5) -C(1')	122.2(6)	C(2") -C(3") -H(19)	107.8(12)
N(1) - C(6) - C(5)	117.5(6)	C(2") -C(3") -H(20)	107.8(12)
C(62) -C(61) - H(6)	113.7(11)	C(4") -C(3") -H(19)	107.6(12)
C(62) -C(61) - H(7)	113.8(11)	C(4") -C(3") -H(20)	107.6(12)
H(6) -C(61) - H(7)	109.5(11)	H(19) -C(3") -H(20)	109.5(14)
C(61) -C(62) - H(8)	96.7(11)	C(3") -C(4") -H(21)	113.0(12)
C(61) -C(62) - H(9)	117.4(12)	C(3") -C(4") -H(22)	103.2(12)
C(61) -C(62) -H(10)	113.3(12)	C(3") -C(4") -H(23)	112.0(12)
H(8) -C(62) - H(9)	109.5(14)	H(21) -C(4") -H(22)	109.5(14)
H(8) -C(62) -H(10)	109.4(14)	H(21) -C(4") -H(23)	109.5(14)
H(9) -C(62) -H(10)	109.5(14)	H(22) -C(4") -H(23)	109.4(14)
C(5) -C(1') -C(2')	121.1(6)	O(1E) -S(1E) -O(2E)	110.3(3)
C(5) -C(1') -C(6')	120.9(6)	O(1E) -S(1E) -O(3E)	112.7(3)
C(2') -C(1') -C(6')	117.9(6)	O(1E) -S(1E) -C(1E)	105.6(4)
C(1') -C(2') -C(3')	121.8(6)	O(2E) -S(1E) -O(3E)	114.8(3)
C(1') -C(2') -H(11)	119.1(7)	O(2E) -S(1E) -C(1E)	107.2(4)
C(3') -C(2') -H(11)	119.1(7)	O(3E) -S(1E) -C(1E)	105.5(4)
C(2') -C(3') -C(4')	121.3(6)	S(1E) -C(1E) -C(2E)	112.4(7)
C(2') -C(3') -N(3')	116.2(6)	S(1E) -C(1E) -H(24)	108.7(9)
C(4') -C(3') -N(3')	122.6(6)	S(1E) -C(1E) -H(25)	108.6(9)
C(3') -C(4') -C(5')	116.0(6)	C(2E) -C(1E) -H(24)	108.8(10)
C(3') -C(4') -N(1")	124.0(6)	C(2E) -C(1E) -H(25)	108.7(10)
C(5') -C(4') -N(1")	120.0(6)	H(24) -C(1E) -H(25)	109.5(11)
C(4') -C(5') -C(6')	122.1(6)	C(1E) -C(2E) -H(26)	112.2(10)
C(4') -C(5') -H(12)	119.0(7)	C(1E) -C(2E) -H(27)	108.9(10)
C(6') -C(5') -H(12)	118.9(7)	C(1E) -C(2E) -H(28)	107.1(10)
C(1') -C(6') -C(5')	120.9(6)	H(26) -C(2E) -H(27)	109.5(11)
C(1') -C(6') -H(13)	119.6(7)	H(26) -C(2E) -H(28)	109.5(11)
C(5') -C(6') -H(13)	119.5(7)	H(27) -C(2E) -H(28)	109.5(11)
C(3') -N(3') -O(31')	119.2(6)		

Table 5.17 Torsion angles (degrees) with standard deviations

C(6) - N(1) - C(2) -N(21)	176.0(6)	C(3') -C(4') -C(5') -H(12)	-179.0(7)
C(6) - N(1) - C(2) - N(3)	-4.7(10)	N(1") -C(4') -C(5') -C(6')	-180.0(6)
H(1) - N(1) - C(2) -N(21)	2.1(43)	N(1") -C(4') -C(5') -H(12)	0.0(11)
H(1) - N(1) - C(2) - N(3)	-178.5(43)	C(3') -C(4') -N(1") -C(1")	-178.6(7)

C(2) - N(1) - C(6) - C(5)	3.1(9)	C(3') -C(4') -N(1") -H(14)	-13.9(35)
H(1) - N(1) - C(6) - C(5)	176.6(45)	C(5') -C(4') -N(1") -C(1")	2.5(11)
N(1) - C(2) -N(21) - H(2)	-14.3(39)	C(5') -C(4') -N(1") -H(14)	167.2(34)
N(1) - C(2) -N(21) - H(3)	-174.0(39)	C(4') -C(5') -C(6') -C(1')	1.0(10)
N(3) - C(2) -N(21) - H(2)	166.4(38)	C(4') -C(5') -C(6') -H(13)	-178.9(7)
N(3) - C(2) -N(21) - H(3)	6.6(39)	H(12) -C(5') -C(6') -C(1')	-178.9(7)
N(1) - C(2) - N(3) - C(4)	2.0(9)	H(12) -C(5') -C(6') -H(13)	1.2(12)
N(21) - C(2) - N(3) - C(4)	-178.7(6)	C(4') -N(1") -C(1") -C(2")	112.5(8)
C(2) - N(3) - C(4) -N(41)	179.4(5)	C(4') -N(1") -C(1") -H(15)	-127.9(9)
C(2) - N(3) - C(4) - C(5)	2.1(8)	C(4') -N(1") -C(1") -H(16)	-7.2(12)
N(3) - C(4) -N(41) - H(4)	15.8(38)	H(14) -N(1") -C(1") -C(2")	-50.9(38)
N(3) - C(4) -N(41) - H(5)	167.3(50)	H(14) -N(1") -C(1") -H(15)	68.8(38)
C(5) - C(4) -N(41) - H(4)	-166.9(38)	H(14) -N(1") -C(1") -H(16)	-170.5(38)
C(5) - C(4) -N(41) - H(5)	-15.4(51)	N(1") -C(1") -C(2") -C(3")	174.2(9)
N(3) - C(4) - C(5) - C(6)	-3.5(9)	N(1") -C(1") -C(2") -H(17)	53.2(11)
N(3) - C(4) - C(5) -C(1')	176.1(5)	N(1") -C(1") -C(2") -H(18)	-64.8(11)
N(41) - C(4) - C(5) - C(6)	179.4(6)	H(15) -C(1") -C(2") -C(3")	54.6(13)
N(41) - C(4) - C(5) -C(1')	-1.0(9)	H(15) -C(1") -C(2") -H(17)	-66.4(12)
C(4) - C(5) - C(6) - N(1)	0.8(9)	H(15) -C(1") -C(2") -H(18)	175.6(10)
C(1') - C(5) - C(6) - N(1)	-178.8(6)	H(16) -C(1") -C(2") -C(3")	-66.2(12)
C(4) - C(5) -C(1') -C(2')	-83.3(8)	H(16) -C(1") -C(2") -H(17)	172.8(11)
C(4) - C(5) -C(1') -C(6')	95.9(7)	H(16) -C(1") -C(2") -H(18)	54.8(13)
C(6) - C(5) -C(1') -C(2')	96.3(8)	C(1") -C(2") -C(3") -C(4")	179.5(9)
C(6) - C(5) -C(1') -C(6')	-84.5(8)	C(1") -C(2") -C(3") -H(19)	-59.6(14)
H(6) -C(61) -C(62) - H(8)	-62.7(14)	C(1") -C(2") -C(3") -H(20)	58.5(15)
H(6) -C(61) -C(62) - H(9)	-178.8(13)	H(17) -C(2") -C(3") -C(4")	-59.6(15)
H(6) -C(61) -C(62) -H(10)	51.9(16)	H(17) -C(2") -C(3") -H(19)	61.3(15)
H(7) -C(61) -C(62) - H(8)	63.7(14)	H(17) -C(2") -C(3") -H(20)	179.4(13)
H(7) -C(61) -C(62) - H(9)	-52.4(17)	H(18) -C(2") -C(3") -C(4")	58.6(15)
H(7) -C(61) -C(62) -H(10)	178.3(12)	H(18) -C(2") -C(3") -H(19)	179.5(13)
C(5) -C(1') -C(2') -C(3')	-179.0(6)	H(18) -C(2") -C(3") -H(20)	-62.4(15)
C(5) -C(1') -C(2') -H(11)	1.1(10)	C(2") -C(3") -C(4") -H(21)	-179.8(12)
C(6') -C(1') -C(2') -C(3')	1.8(10)	C(2") -C(3") -C(4") -H(22)	62.0(15)
C(6') -C(1') -C(2') -H(11)	-178.1(7)	C(2") -C(3") -C(4") -H(23)	-55.5(16)
C(5) -C(1') -C(6') -C(5')	178.4(6)	H(19) -C(3") -C(4") -H(21)	59.3(17)
C(5) -C(1') -C(6') -H(13)	-1.7(10)	H(19) -C(3") -C(4") -H(22)	-58.9(15)
C(2') -C(1') -C(6') -C(5')	-2.4(10)	H(19) -C(3") -C(4") -H(23)	-176.5(14)
C(2') -C(1') -C(6') -H(13)	177.4(7)	H(20) -C(3") -C(4") -H(21)	-58.7(17)
C(1') -C(2') -C(3') -C(4')	0.3(10)	H(20) -C(3") -C(4") -H(22)	-176.9(13)
C(1') -C(2') -C(3') -N(3')	-179.1(6)	H(20) -C(3") -C(4") -H(23)	65.5(16)
H(11) -C(2') -C(3') -C(4')	-179.8(7)	O(1E) -S(1E) -C(1E) -C(2E)	-177.9(7)
H(11) -C(2') -C(3') -N(3')	0.8(10)	O(1E) -S(1E) -C(1E) -H(24)	61.6(9)
C(2') -C(3') -C(4') -C(5')	-1.7(10)	O(1E) -S(1E) -C(1E) -H(25)	-57.5(9)
C(2') -C(3') -C(4') -N(1")	179.4(6)	O(2E) -S(1E) -C(1E) -C(2E)	64.5(8)
N(3') -C(3') -C(4') -C(5')	177.6(6)	O(2E) -S(1E) -C(1E) -H(24)	-56.0(9)
N(3') -C(3') -C(4') -N(1")	-1.4(11)	O(2E) -S(1E) -C(1E) -H(25)	-175.1(8)
C(2') -C(3') -N(3') -O(31')	-174.0(6)	O(3E) -S(1E) -C(1E) -C(2E)	-58.3(8)
C(2') -C(3') -N(3') -O(32')	7.1(10)	O(3E) -S(1E) -C(1E) -H(24)	-178.8(8)
C(4') -C(3') -N(3') -O(31')	6.7(10)	O(3E) -S(1E) -C(1E) -H(25)	62.1(9)

C(4')	-C(3')	-N(3')	-O(32')	-172.2(7)	S(1E)	-C(1E)	-C(2E)	-H(26)	179.7(9)
C(3')	-C(4')	-C(5')	-C(6')	1.0(10)	S(1E)	-C(1E)	-C(2E)	-H(27)	58.2(11)
S(1E)	-C(1E)	-C(2E)	-H(28)	-60.1(11)	H(25)	-C(1E)	-C(2E)	-H(26)	59.3(14)
H(24)	-C(1E)	-C(2E)	-H(26)	-59.9(14)	H(25)	-C(1E)	-C(2E)	-H(27)	-62.1(13)
H(24)	-C(1E)	-C(2E)	-H(27)	178.7(11)	H(25)	-C(1E)	-C(2E)	-H(28)	179.5(11)
H(24)	-C(1E)	-C(2E)	-H(28)	60.4(13)					

Table 5.18 Hydrogen bond contact distance

Hydrogen Bond	Donor-Acceptor Distance (Å)
$N(41)-H(4)_{I}..N(3)_{II}$	3.005
$N(1)-H(1)_{I}...O(1E)_{III}$	2.717
$N(21)-H(2)_{I}...O(2E)_{III}$	2.875
$N(41)-H(5)_{I}...O(1W)_{I}$	2.874
$N(21)-H(3)_{I}...O(1W)_{II}$	2.934
$O(1W)-H(30)_{I}...O(3E)_{IV}$	2.744
$O(1W)-H(29)_{I}...O(2E)_{V}$	2.816

The subscripts I, II, III, IV and V refer to the equivalent positions:

X, Y, Z.

-X, -1-Y, -Z.

$-\frac{1}{2}$+X, $\frac{1}{2}$-Y, $-\frac{1}{2}$+Z.

$\frac{1}{2}$-X, $\frac{1}{2}$+Y, $\frac{1}{2}$-Z.

X, 1+Y, Z.

Figure 5.5 Butaprim Molecular plot and numbering scheme

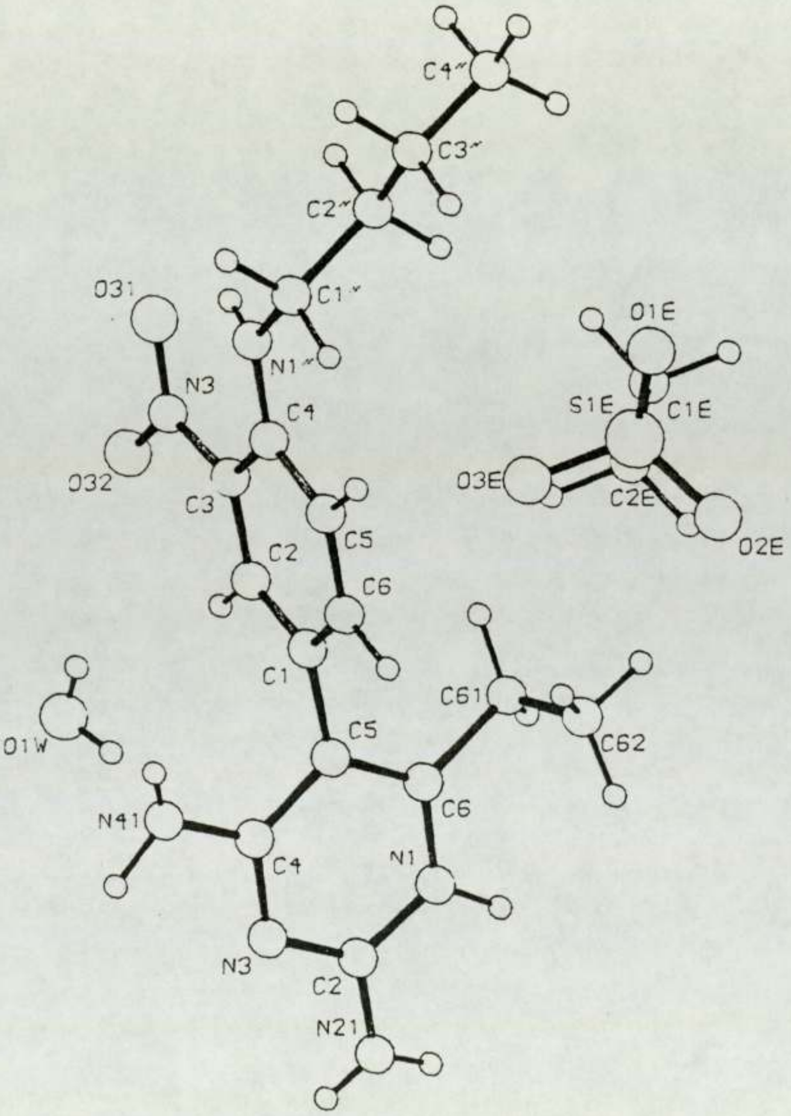
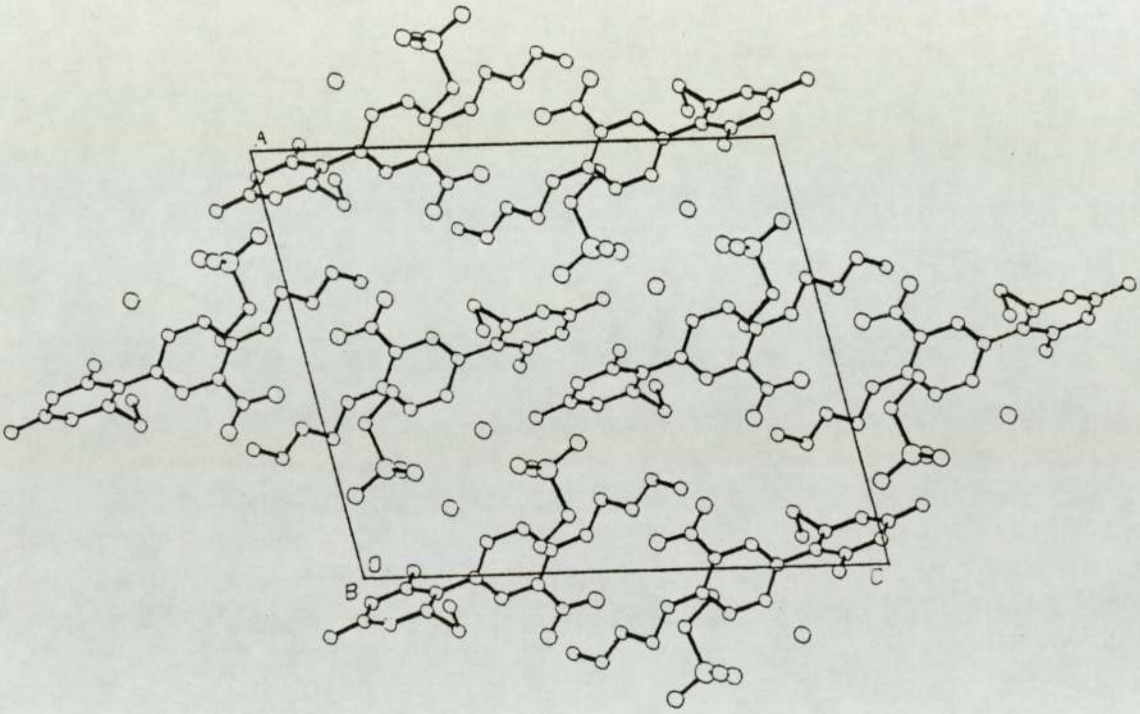


Figure 5.6 Butaprim Unit cell packing diagram



5.4 Results and Discussion

Similar hydrogen bonding patterns are observed in both Benzoprim and Isobenzoprim (Tables 5.6 and 5.12) the former forming an extended pattern with N(21) and N(41) amino hydrogens to N(1) and N(3) of the pyrimidine ring; whilst the latter forms a dimer through N(41B)--H(3B)...N(3A) and N(21A)-H(2A)...N(3B) between the two distinct molecules in the assymetric unit, which is then linked to adjacent dimer units via the remaining amino hydrogens and the N(1) positions of the pyrimidine ring. Hydrogen bonds are also observed to nitro group oxygens but these seem to be fortuituous and secondary to packing requirements.

The above pattern is somewhat modified in the structure of Butaprim due to the presence of the ethanesulphonate counter ion and a water. A dimer is observed between adjacent molecules [N(41)-H(4)...N(3)] but, as observed in all ethanesulphonate salts studied, the preferred interaction at N(1) (the site of protonation) is ionic and hydrogen bonding with the anion. The oxygen of water appears to act as both donor and acceptor linking N(41) (in the same assymetric unit) and N(21) of an adjacent molecule; the hydrogens of the water forming bonds to two ethane sulphonate molecles.

It is clear that a hydrophilic substituent at the 4 position of the phenyl ring is an essential requirement for tight binding. Enzyme binding studies (92) have shown a 1000 fold decrease for Isobenzoprim, where this substituent is at the 3 position, compared with Benzoprim.

CHAPTER 6

CHAPTER 6

The Crystal Structure of 2,4-diamino-5-(4-chlorophenyl)-6-ethyl pyrimidine. 2 N-methyl formamide [Pyrimethamine. NMF. solvate], with implications for Inhibitor Binding to Dihydrofolate Reductase (DHFR).

6.1 Experimental

Crystals were grown from a solution of pyrimethamine in NMF. Crystals which appeared, after leaving a solution in a refrigerator at ~ 5°C for 3 days, were examined for quality under a polarising microscope. Good birefringence was observed while the crystals remained in contact with their supernatant; however on removal from solution and blotting on filter paper, reinspection revealed microcracks and a coating of white powder on the crystal surface. It was assumed that this phenomenon was due to drying out of the NMF from the crystal, and subsequent x-ray analysis showed this to be likely as a single crystal, as described, showed no diffraction. In an attempt to deal with the fugitive nature of the solvate molecule, crystal was mounted in a Lindemann glass capillary between columns of mother liquor. The capillary was sealed and mounted on a goniometer head stud with hot wax.

6.2 Crystal Data

$C_{12}H_{13}N_4Cl.2[C_2H_5NO]$, M = 366.853, F(000) = 388, Triclinic a = 8.674(4), b = 10.519(3), c = 11.618(2), Å, α = 89.45(2), β = 86.36(3), γ = 68.46(3)°, V = 984 Å^3 (by least squares refinement on diffractometer angles for 25 automatically centred reflections, λ = 0.71069 Å), space group $P\bar{1}$, Z = 2, Dx = 1.238 gcm^{-3}, Dm - 1.25 gcm^{-3} (method of floatation in solvent mixture). White monoclinic plates. Crystal dimensions, 1.40 x 0.32 x 0.08 mm, μ (Mo-Kα) = 1.76 cm^{-1}.

6.3 Data Collection and Processing

CAD4 diffractometer, w/2θ mode with w scan width = 0.9 + 0.35 tanθ , w scan speed 0.8 - 3.4°min^{-1}, graphite-monochromated Mo-Kα radiation; 4156 reflections measured ($2.0 \leq \theta \leq 26°$ + h, ± k, ± l) 3886 unique (merging R = 0.0172, no correction for absorption) giving 1994 with $I > 3\ \sigma(I)$, number of parameters refined 248. Linear and approximately isotropic crystals decay ca. 1.6% (for 3 standard reflections, checked every 7200 s, x-ray exposure time), corrected during processing.

6.4 Structure Determination an Refinement.

Data reduction followed by direct methods (MULTAN80) and electron density synthesis. All hydrogen atoms were located by difference Fourier analysis during isotropic least squares refinement of non-hydrogen atoms. Poor geometry of the hydrogen atoms on all methyl groups required the use of AFIX (SHELX76) to constrain these positions to ideal tetrahedral geometry during refinement; these atoms were also refined with a common temperature factor. Density corresponding to expected hydrogen atoms was found around C(2"B); however on calculating positions and attempting to refine as a rigid group, convergence was poor and the R factor worsened. These hydrogen atoms were omitted entirely and convergence was reached after 10 cycles of full matrix least squares refinement with all non-hydrogen atoms anisotropic, refined U_{iso} only for hydrogen atoms and using the weighting scheme $W = 1.6276/[\sigma^2 (F_o]$, satisfactory agreement analyses were obtained. Final R and R_w values are 0.0566 and 0.0586; (Δ/σ) max = 0.010 [C(2"B) X rotation refinement of pivot atom in rigid group]. A final electron density synthesis showed no peak > 0.25 e Å^{-3}.

Table 6.1 Positional parameters (fractional coordinates * 10**4) with estimated standard deviations in parentheses.

ATOM	x/a	y/b	z/c
N(1)	2091(4)	4308(3)	5649(2)
C(2)	2305(5)	3132(4)	5115(3)
N(21)	1096(4)	3134(3)	4446(3)
N(3)	3589(4)	1941(3)	5188(2)
C(4)	4790(4)	1953(3)	5853(3)
N(41)	6053(4)	0752(3)	5956(3)
C(5)	4720(4)	3160(3)	6417(3)
C(6)	3331(5)	4298(3)	6290(3)
C(61)	3085(6)	5621(4)	6894(4)
C(62)	2275(8)	5734(5)	8041(5)
C(1’)	6140(4)	3131(3)	7101(3)
C(2’)	7388(5)	3527(4)	6613(3)
C(3’)	8698(6)	3484(5)	7235(4)
C(4’)	8782(5)	3033(4)	8338(4)
C(5’)	7572(5)	2640(4)	8862(3)
C(6’)	6260(5)	2690(4)	8216(3)
Cl(1)	10477(2)	2924(2)	9112(1)
C(1"A)	0281(6)	0737(4)	1990(4)
N(2"A)	-1024(5)	0424(4)	2275(4)
C(2"A)	-1500(8)	0191(6)	3442(6)
O(1"A)	1224(4)	0877(3)	2665(3)
C(1"B)	503(1)	0828(8)	11279(6)
N(2"B)	3677(7)	1819(6)	11568(5)
C(2"B)	3458(5)	3221(5)	11366(4)
O(1"B)	6317(5)	0914(5)	10806(4)
H(1)	0267(5)	3958(5)	4357(4)
H(2)	1233(5)	2427(5)	4032(4)
H(3)	6091(5)	-0027(5)	5618(4)
H(4)	6828(5)	0640(5)	6408(4)
H(5)	2664(5)	6532(5)	6363(4)
H(6)	4024(5)	6069(5)	6663(4)
H(7)	3047(8)	4870(5)	8514(5)
H(8)	1101(8)	5632(5)	7920(5)
H(9)	2061(8)	6665(5)	8519(5)
H(10)	7346(5)	3889(5)	5763(4)
H(11)	9548(5)	3683(5)	6877(4)
H(12)	7694(5)	2178(5)	9713(4)
H(13)	5438(5)	2417(5)	8615(4)
H(14)	0482(5)	0905(5)	1105(4)
H(15)	-1683(5)	0485(5)	1712(4)
H(16)	-1030(8)	0742(6)	4019(6)
H(17)	-1035(8)	-0880(6)	3655(6)
H(18)	-2840(8)	0590(6)	3531(6)
H(19)	5192(5)	-0087(5)	11450(4)
H(20)	2977(5)	1620(5)	11980(4)

All hydrogens constrained to ride on attached non-hydrogen atoms during SHELX least squares refinement.

Table 6.2 Anisotropic temperature factors, and isotropic temperature factors where shown; with standard deviations in parentheses.

ATOM	U11	U22	U33	U23	U13	U12
N(1)	.055(2)	.045(2)	.061(2)	-.004(2)	-.018(2)	-.014(2)
C(2)	.051(2)	.045(2)	.050(2)	-.002(2)	-.011(2)	-.015(2)
N(21)	.060(2)	.049(2)	.077(2)	-.017(2)	-.027(2)	-.005(2)
N(3)	.048(2)	.047(2)	.056(2)	-.004(2)	-.013(2)	-.016(2)
C(4)	.050(2)	.044(2)	.051(2)	-.001(2)	-.006(2)	-.015(2)
N(41)	.061(2)	.045(2)	.089(2)	-.010(2)	-.034(2)	-.008(2)
C(5)	.054(2)	.043(2)	.046(2)	-.003(2)	-.005(2)	-.019(2)
C(6)	.060(3)	.040(2)	.055(2)	-.004(2)	-.008(2)	-.017(2)
C(61)	.090(3)	.046(3)	.076(3)	-.012(2)	-.018(3)	-.021(2)
C(62)	.179(7)	.084(4)	.098(4)	-.028(3)	-.014(5)	-.024(4)
C(1')	.052(2)	.045(2)	.049(2)	-.002(2)	-.008(2)	-.019(2)
C(2')	.073(3)	.093(3)	.058(3)	.006(2)	-.007(2)	-.046(3)
C(3')	.071(3)	.114(4)	.081(3)	-.003(3)	-.006(3)	-.061(3)
C(4')	.068(3)	.086(3)	.072(3)	-.008(3)	-.023(3)	-.038(3)
C(5')	.077(3)	.082(3)	.066(3)	.008(2)	-.023(3)	-.043(3)
C(6')	.066(3)	.070(3)	.056(3)	.006(2)	-.007(2)	-.040(2)
CL(1)	.093(1)	.161(1)	.123(1)	.001(1)	.007(2)	-.071(1)
C(1"A)	.070(3)	.065(3)	.082(4)	.007(2)	-.011(3)	-.022(3)
N(2"A)	.074(3)	.086(3)	.110(4)	.005(2)	-.012(3)	-.039(2)
C(2"A)	.162(6)	.156(6)	.122(5)	.009(4)	.030(5)	-.092(5)
O(1"A)	.070(2)	.086(2)	.109(3)	.002(2)	-.034(2)	-.032(2)
C(1"B)	.141(6)	.135(6)	.131(6)	.046(5)	-.043(5)	-.106(6)
N(2"B)	.081(4)	.175(6)	.119(4)	-.011(4)	.000(3)	-.068(4)
C(2"B)	.162(7)	.099(5)	.237(8)	-.012(5)	.011(6)	-.065(5)
O(1"B)	.097(3)	.152(4)	.142(3)	.045(3)	-.023(3)	-.063(3)

	U ISO
H(1)	.07(1)
H(2)	.05(1)
H(3)	.07(1)
H(4)	.09(1)
H(5)	.09(1)
H(6)	.08(1)
H(7)	.19(2)
H(8)	.19(2)
H(9)	.19(2)
H(10)	.09(1)
H(11)	.09(2)
H(12)	.11(1)
H(13)	.06(1)
H(14)	.09(1)
H(15)	.17(3)
H(16)	.33(3)
H(17)	.33(3)
H(18)	.33(3)
H(19)	.12(2)
H(20)	.16(3)

Table 6.3 Bond Lengths(A) with standard deviations

N(1) - C(2)	1.335(5)	C(2') -C(3')	1.372(7)
N(1) - C(6)	1.343(5)	C(2') -H(10)	1.053(6)
C(2) -N(21)	1.344(5)	C(3') -C(4')	1.359(7)
C(2) - N(3)	1.343(5)	C(3') -H(11)	0.913(7)
N(21) - H(1)	0.911(6)	C(4') -C(5')	1.370(6)
N(21) - H(2)	0.856(6)	C(4') -Cl(1)	1.739(5)
N(3) - C(4)	1.339(5)	C(5') -C(6')	1.387(6)
C(4) -N(41)	1.345(5)	C(5') -H(12)	1.091(6)
C(4) - C(5)	1.414(5)	C(6') -H(13)	0.955(6)
N(41) - H(3)	0.901(6)	C(1"A)-N(2"A)	1.313(7)
N(41) - H(4)	0.853(6)	C(1"A)-O(1"A)	1.216(6)
C(5) - C(6)	1.366(5)	C(1"A)-H(14)	1.056(7)
C(5) -C(1')	1.499(5)	N(2"A)-C(2"A)	1.437(8)
C(6) -C(61)	1.502(6)	N(2"A)-H(15)	0.882(7)
C(61) -C(62)	1.451(8)	C(2"A)-H(16)	1.080(10)
C(61) - H(5)	1.092(7)	C(2"A)-H(17)	1.080(10)
C(61) - H(6)	1.100(7)	C(2"A)-H(18)	1.079(10)
C(62) - H(7)	1.080(9)	C(1"B)-N(2"B)	1.281(10)
C(62) - H(8)	1.080(9)	C(1"B)-O(1"B)	1.243(9)
C(62) - H(9)	1.080(9)	C(1"B)-H(19)	0.942(9)
C(1') -C(2')	1.384(5)	N(2"B)-C(2"B)	1.435(8)
C(1') -C(6')	1.369(5)	N(2"B)-H(20)	0.834(8)

Table 6.4 Angles(degrees) with standard deviations

C(2) - N(1) - C(6)	115.8(3)	C(2') -C(1') -C(6')	118.0(3)
N(1) - C(2) -N(21)	116.6(4)	C(1') -C(2') -C(3')	120.7(4)
N(1) - C(2) - N(3)	126.9(4)	C(1') -C(2') -H(10)	121.1(4)
N(21) - C(2) - N(3)	116.5(4)	C(3') -C(2') -H(10)	118.2(5)
C(2) -N(21) - H(1)	116.1(5)	C(2') -C(3') -C(4')	119.7(4)
C(2) -N(21) - H(2)	119.4(5)	C(2') -C(3') -H(11)	119.1(6)
H(1) -N(21) - H(2)	123.3(6)	C(4') -C(3') -H(11)	121.0(6)
C(2) - N(3) - C(4)	116.0(3)	C(3') -C(4') -C(5')	121.8(4)
N(3) - C(4) -N(41)	116.1(3)	C(3') -C(4') -Cl(1)	119.7(4)
N(3) - C(4) - C(5)	121.7(3)	C(5') -C(4') -Cl(1)	118.4(3)
N(41) - C(4) - C(5)	122.3(3)	C(4') -C(5') -C(6')	117.5(4)
C(4) -N(41) - H(3)	122.8(5)	C(4') -C(5') -H(12)	122.1(5)
C(4) -N(41) - H(4)	123.0(5)	C(6') -C(5') -H(12)	119.9(4)
H(3) -N(41) - H(4)	113.8(6)	C(1') -C(6') -C(5')	122.3(4)
C(4) - C(5) - C(6)	116.6(3)	C(1') -C(6') -H(13)	122.8(5)
C(4) - C(5) -C(1')	119.4(3)	C(5') -C(6') -H(13)	114.9(5)
C(6) - C(5) -C(1')	124.0(3)	N(2"A)-C(1"A)-O(1"A)	125.0(5)

N(1) - C(6) - C(5)	123.0(3)
N(1) - C(6) -C(61)	115.7(3)
C(5) - C(6) -C(61)	121.2(3)
C(6) -C(61) -C(62)	113.0(4)
C(6) -C(61) - H(5)	115.5(5)
C(6) -C(61) - H(6)	115.5(5)
C(62) -C(61) - H(5)	116.5(5)
C(62) -C(61) - H(6)	123.2(5)
H(5) -C(61) - H(6)	64.8(4)
C(61) -C(62) - H(7)	106.2(6)
C(61) -C(62) - H(8)	105.4(6)
C(61) -C(62) - H(9)	116.5(6)
H(7) -C(62) - H(8)	109.5(7)
H(7) -C(62) - H(9)	109.5(7)
H(8) -C(62) - H(9)	109.5(7)
C(5) -C(1') -C(2')	120.7(3)
C(5) -C(1') -C(6')	121.2(3)
N(2"A)-C(1"A)-H(14)	116.1(5)
O(1"A)-C(1"A)-H(14)	118.9(5)
C(1"A)-N(2"A)-C(2"A)	123.4(5)
C(1"A)-N(2"A)-H(15)	115.0(6)
C(2"A)-N(2"A)-H(15)	121.1(6)
N(2"A)-C(2"A)-H(16)	108.7(6)
N(2"A)-C(2"A)-H(17)	112.4(7)
N(2"A)-C(2"A)-H(18)	107.3(6)
H(16) -C(2"A)-H(17)	109.4(8)
H(16) -C(2"A)-H(18)	109.5(8)
H(17) -C(2"A)-H(18)	109.5(8)
N(2"B)-C(1"B)-O(1"B)	126.8(7)
N(2"B)-C(1"B)-H(19)	122.2(8)
O(1"B)-C(1"B)-H(19)	110.9(8)
C(1"B)-N(2"B)-C(2"B)	122.5(6)
C(1"B)-N(2"B)-H(20)	116.1(8)
C(2"B)-N(2"B)-H(20)	120.6(7)

Table 6.5 Torsion angles(degrees) with standard deviations

C(6) - N(1) - C(2) -N(21)	-178.3(3)
C(6) - N(1) - C(2) - N(3)	2.7(6)
C(2) - N(1) - C(6) - C(5)	-1.2(5)
C(2) - N(1) - C(6) -C(61)	-179.8(3)
N(1) - C(2) -N(21) - H(1)	5.3(6)
N(1) - C(2) -N(21) - H(2)	173.4(5)
N(3) - C(2) -N(21) - H(1)	-175.6(5)
N(3) - C(2) -N(21) - H(2)	-7.5(7)
N(1) - C(2) - N(3) - C(4)	-1.3(6)
N(21) - C(2) - N(3) - C(4)	179.7(3)
C(2) - N(3) - C(4) -N(41)	178.0(3)
C(2) - N(3) - C(4) - C(5)	-1.6(5)
N(3) - C(4) -N(41) - H(3)	-1.6(7)
N(3) - C(4) -N(41) - H(4)	-174.2(5)
C(5) - C(4) -N(41) - H(3)	178.0(5)
C(5) - C(4) -N(41) - H(4)	5.4(7)
N(3) - C(4) - C(5) - C(6)	2.8(5)
N(3) - C(4) - C(5) -C(1')	-176.9(3)
N(41) - C(4) - C(5) - C(6)	-176.8(3)
N(41) - C(4) - C(5) -C(1')	3.6(5)
C(4) - C(5) - C(6) - N(1)	-1.3(5)
C(4) - C(5) - C(6) -C(61)	177.2(3)
C(1') - C(5) - C(6) - N(1)	178.3(3)
C(1') - C(5) - C(6) -C(61)	-3.2(6)
C(4) - C(5) -C(1') -C(2')	95.4(4)
H(6) -C(61) -C(62) - H(8)	154.9(6)
H(6) -C(61) -C(62) - H(9)	33.3(9)
C(5) -C(1') -C(2') -C(3')	-179.2(4)
C(5) -C(1') -C(2') -H(10)	2.2(7)
C(6') -C(1') -C(2') -C(3')	-0.4(6)
C(6') -C(1') -C(2') -H(10)	-179.0(5)
C(5) -C(1') -C(6') -C(5')	179.3(4)
C(5) -C(1') -C(6') -H(13)	-2.4(7)
C(2') -C(1') -C(6') -C(5')	0.5(6)
C(2') -C(1') -C(6') -H(13)	178.8(5)
C(1') -C(2') -C(3') -C(4')	0.8(7)
C(1') -C(2') -C(3') -H(11)	175.0(6)
H(10) -C(2') -C(3') -C(4')	179.4(5)
H(10) -C(2') -C(3') -H(11)	-6.3(9)
C(2') -C(3') -C(4') -C(5')	-1.3(7)
C(2') -C(3') -C(4') -Cl(1)	177.9(4)
H(11) -C(3') -C(4') -C(5')	-175.4(6)
H(11) -C(3') -C(4') -Cl(1)	3.7(8)
C(3') -C(4') -C(5') -C(6')	1.3(7)
C(3') -C(4') -C(5') -H(12)	173.5(5)
Cl(1) -C(4') -C(5') -C(6')	-177.8(3)
Cl(1) -C(4') -C(5') -H(12)	-5.7(7)
C(4') -C(5') -C(6') -C(1')	-0.9(6)
C(4') -C(5') -C(6') -H(13)	-179.4(5)
H(12) -C(5') -C(6') -C(1')	-173.3(5)

C(4) - C(5) -C(1′) -C(6′)	-83.3(4)	H(12) -C(5′) -C(6′) -H(13)	8.3(7)
C(6) - C(5) -C(1′) -C(2′)	-84.2(5)	O(1"A)-C(1"A)-N(2"A)-C(2"A)	-0.6(8)
C(6) - C(5) -C(1′) -C(6′)	97.0(5)	O(1"A)-C(1"A)-N(2"A)-H(15)	170.7(6)
N(1) - C(6) -C(61) -C(62)	90.5(5)	H(14) -C(1"A)-N(2"A)-C(2"A)	-178.7(6)
N(1) - C(6) -C(61) - H(5)	-47.2(6)	H(14) -C(1"A)-N(2"A)-H(15)	-7.4(8)
N(1) - C(6) -C(61) - H(6)	-120.0(5)	C(1"A)-N(2"A)-C(2"A)-H(16)	25.8(9)
C(5) - C(6) -C(61) -C(62)	-88.1(5)	C(1"A)-N(2"A)-C(2"A)-H(17)	-95.5(8)
C(5) - C(6) -C(61) - H(5)	134.2(5)	C(1"A)-N(2"A)-C(2"A)-H(18)	144.1(6)
C(5) - C(6) -C(61) - H(6)	61.4(6)	H(15) -N(2"A)-C(2"A)-H(16)	-145.0(7)
C(6) -C(61) -C(62) - H(7)	57.9(7)	H(15) -N(2"A)-C(2"A)-H(17)	93.8(9)
C(6) -C(61) -C(62) - H(8)	-58.3(7)	H(15) -N(2"A)-C(2"A)-H(18)	-26.7(9)
C(6) -C(61) -C(62) - H(9)	-179.9(6)	O(1"B)-C(1"B)-N(2"B)-C(2"B)	0.3(12)
H(5) -C(61) -C(62) - H(7)	-164.9(6)	O(1"B)-C(1"B)-N(2"B)-H(20)	-169.8(8)
H(5) -C(61) -C(62) - H(8)	79.0(7)	H(19) -C(1"B)-N(2"B)-C(2"B)	176.6(8)
H(5) -C(61) -C(62) - H(9)	-42.6(9)	H(19) -C(1"B)-N(2"B)-H(20)	6.5(13)
H(6) -C(61) -C(62) - H(7)	-89.0(8)		

Table 6.6 Hydrogen bond contact distances

Hydrogen Bond	Donor-Acceptor Distance (Å)
$N(21)-H(3)_I \ldots O(1'')_I$	2.883
$N(1)-H(1)_I \ldots O(2'')_I$	2.846
$N(41)-H(4)_I \ldots N(3)_{II}$	3.274
$N(41)-H(5)_I \ldots O(3'')_{III}$	2.969
$N(21)-H(2)_I \ldots O(1'')_{IV}$	2.788

The subscripts I, II, III and IV refer to the equivalent positions X, Y, Z; 1-X, 1-Y, 1-Z; -1+X, 1+Y, Z; 2-X, -Y, 1-Z.

Figure 6.1 Pyrimethamine.NMF Molecular plot and numbering scheme

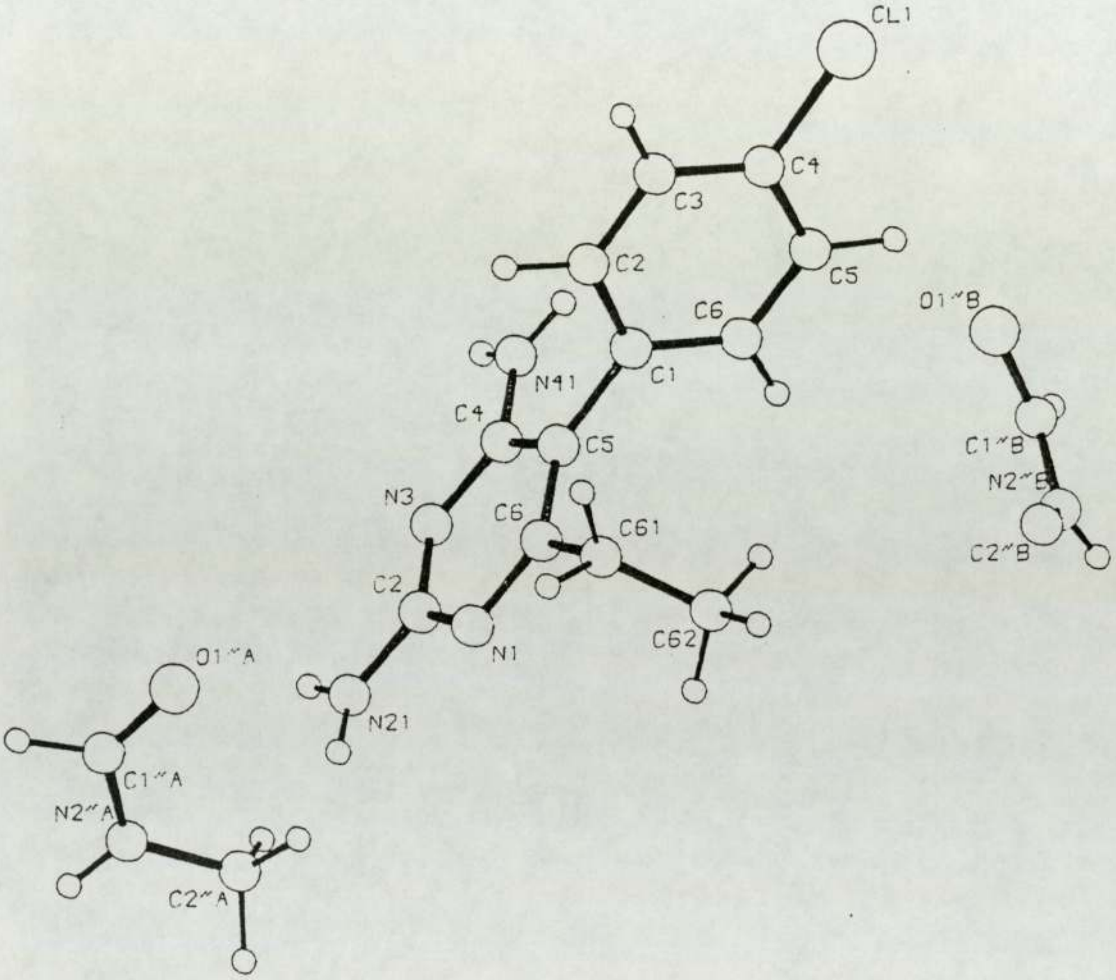
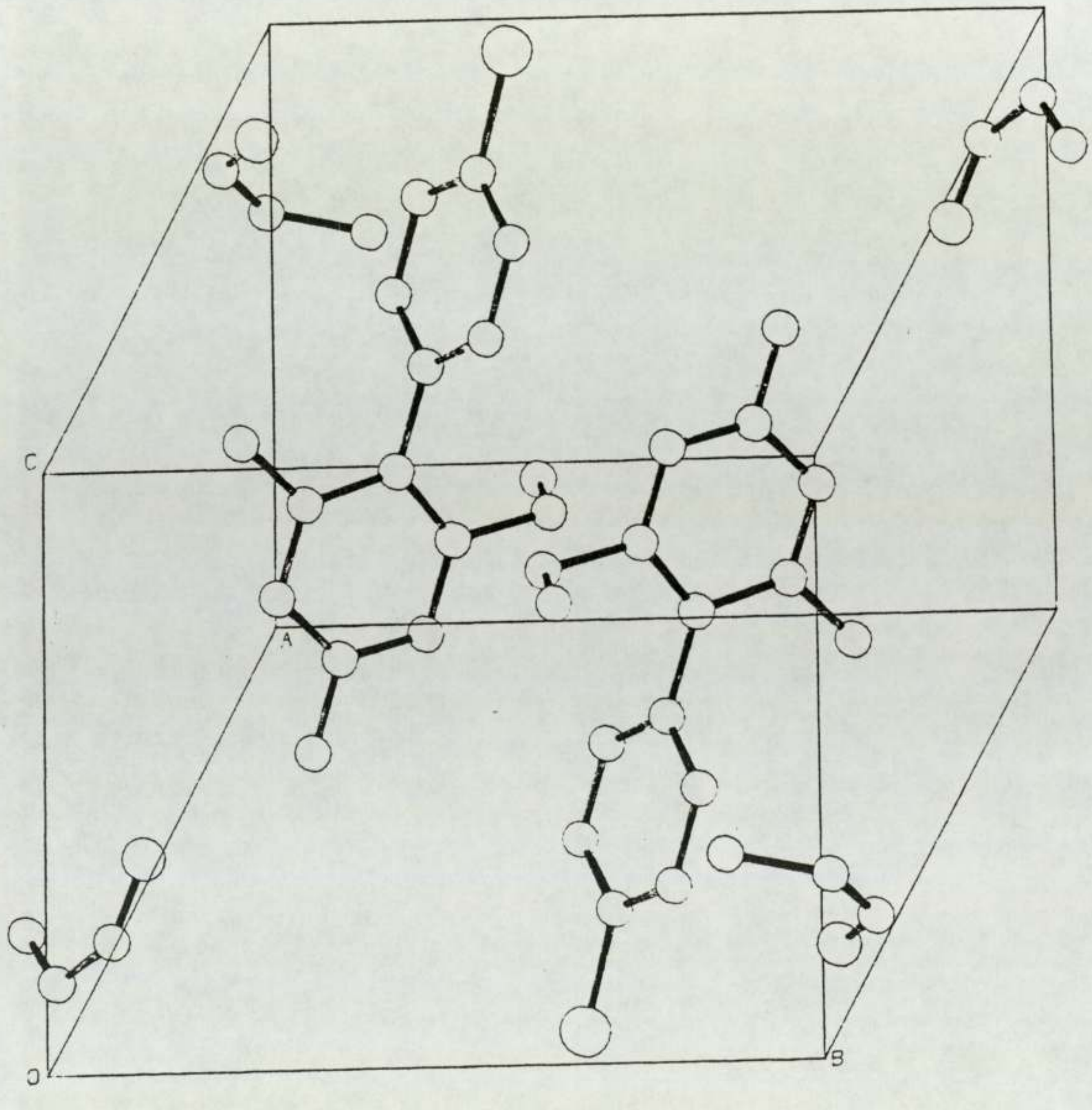


Figure 6.2 Pyrimethamine.NMF Unit cell packing diagram



6.5 Results and Discussion

Work by Villafranca et al (79) has shown that strong inhibitor binding can persist even when the known ionic interaction between the side chain carboxylate of the enzyme (ASP27) and a protonated 2,4-diaminopyrimidine moiety is precluded. It is suggested that the major component of binding is therefore hydrogen bonding. The title compound was selected as a model for this interaction as to observe this in the crystal the NMF would be require to adopt a non-preferred conformation (E) thus incurring a calculated energy penalty of + 1.3 $KJmol^{-1}$ (89).

A comparison of this structure with Pyrimethamine free base (71) shows great similarity in hydrogen bonding pattern and ring-ring-6-ethyl geometries, as may be expected. Molecules hydrogen bond pairwise around a centre of symmetry (Table 6.6). This is not observed in Pyrimethamine free base but both substance crystallise in space group P1, form paired N-H...N hydrogen bonds, and exhibit similar torsion angles: T ring-ring 83.8, and 74.4 and 82.4; T ethyl group -90.1, and 98.0 and 99.0 (Pyrimethamine.NMF and two independent molecules of Pyrimethamine respectively). As no close contacts are observed between phenyl ring and 6-ethyl side chain, in either compound, it is not possible to explain the difference in torsion angle from the structural data. It is clear however that the Pyrimethamine moiety in Pyrimethamine.NMF mimics the hydrogen bonding observed in Pyrimethamine rather than incurring the energy penalty of flipping the NMF molecule into a conformation facilitating paired hydrogen bonds.

One NMF molecule (A) finds proton donors at the amino groups N(21) and N(41) of Pyrimethamine and N(2"B) of a second NMF molecule; the latter NMF molecule (B) acts as a proton acceptor for N(2"A). Both NMF molecules retain their preferred cis geometry. The suggestion is that the enzyme substrate hydrogen bonding in DHFR is not sufficiently strong to account for the tight binding in themutant strain (79), and it appears that the binding mode of 2,4-diamino heterocycles may be more sophisticated than first believed.

CHAPTER 7

CHAPTER 7

The Crystal Structure of 2,4-diamino-5-(3-nitro-4-chlorophenyl)-6-ethylpyrimidine Ethane sulphonate salt (Nitropyrimethamine)

7.1 Experimental

A crystalline sample of the compound was obtained (76), and from it suitable crystals were selected, their quality being estimated by optical birefringent properties, revealed by examination with a polarising microscope. A crystal was mounted on a glass filament using epoxy adhesive; the filament was then secured to a goniometer-head study by the hot wax method.

7.2 Crystal Data

$C_{12}H_{13}N_5O_2Cl.C_2H_5SO_3$, M = 403.845, F(000) = 420, Triclinic a = 8.208(1), b = 11.105(2) c = 11.751(2) Å, α = 73.20(2), β = 71.56(1) γ = 70.64(2)° V = 938 $Å^3$ (by least squares refinement on diffractometer angles for 25 automatically centred reflections, λ = 0.71069 Å), space group $P\bar{1}$, Z = 2, Dx = 1.429 gcm^{-3}, Dm = 1.426 gcm^{-3} (method of floatation in solvent mixture). White plate, 0.6 x 0.20 x 0.04 mm, μ (Mo-Kα) = 3.00 cm^{-1}.

7.3 Data Collection and Processing

CAD4 diffractometer, w/2θ mode with w scan width = 0.85 + 0.35 tanθ, w scan speed 0.4 - 2.9 deg min^{-1} graphite monochromated Mo-Kα radiation; 5409 reflections measured ($2° \leqslant \theta \leqslant$

25° ± h ± k = 1), 3215 unique (merging R = 0.1289, no correction for absorption, giving 1780 with $I > 3\sigma(I)$, number of parameters refined 284. Linear and approximately isotropic crystal decay ca, 2% (for 3 reference reflections checked every 7200 s x-ray exposure time), corrected during processing.

7.4 Structure Determination and Refinement

Data Reduction followed by direct methods (SHELX76) electron density synthesis. All hydrogens attached to the cation were located by difference Fourier analysis during isotropic least squares refinement of non-hydrogen atoms. Hydrogens attached to the ethane-sulphonate molecule were not unambiguously located; therefore they were generated in fixed ideal geometries using the AFIX option (SHELX76) and refined with common temperature factors. It was also found to be necessary to constrain the terminal methyl group of the 6-ethyl substituent, in a similar way., during refinement. Convergence was reached after 10 cycles of full matrix least squares refinement with all non-hydrogen atoms anisotropic. The weighting scheme $W = 12.7230/[\sigma^2 (F_o) + 0.001 F_o^2]$, with $\sigma(F_o)$ from counting statistics, gave satisfactory agreement analyses. Final R and R_w values are 0.0732 and 0.0926. Examination of the final electron density synthesis showed no peak > 0.5 e Å^{-3}; (Δ/σ) max = 0.174 [C(2") Y rotation refinement of pivot atom of rigid group] commonly $\Delta/\sigma = 0.01$.

Table 7.1 Positional parameters (fractional coordinates * 10**4, some * 10**3; hydrogens * 10**3, some * 10**2) with estimated standard deviations in parentheses.

ATOM	x/a	y/b	z/c
N(1)	5454(7)	1580(6)	3747(5)
C(2)	6108(8)	2157(7)	4304(6)
N(21)	7513(9)	1444(8)	4772(7)
N(3)	5399(7)	3408(5)	4392(5)
C(4)	3982(8)	4086(6)	3888(5)
N(41)	326(1)	5325(7)	3986(7)
C(5)	3285(8)	3519(6)	3251(6)
C(6)	4045(8)	2240(6)	3212(6)
C(61)	348(1)	148(1)	260(1)
C(62)	490(1)	114(1)	1458(8)
C(1′)	1812(8)	4339(6)	2620(6)
C(2′)	219(1)	5088(6)	1478(6)
C(3′)	0865(9)	5878(6)	0902(6)
N(31′)	146(1)	6660(7)	-0338(6)
O(31′)	293(1)	6322(9)	-0912(7)
O(32′)	042(1)	760(1)	-0719(8)
C(4′)	-093(1)	5898(7)	1466(7)
C(L4′)	-2721(3)	6881(3)	0821(3)
C(5′)	-134(1)	5117(8)	2585(8)
C(6′)	002(1)	4356(8)	3151(8)
S(1")	9048(2)	-1574(2)	3502(2)
O(1")	9601(9)	-1094(7)	4289(6)
O(2")	7109(7)	-1096(5)	3657(5)
O(3")	9727(8)	-2951(5)	3671(6)
C(1")	999(1)	-093(1)	1990(9)
C(2")	1203(2)	-124(2)	162(1)
H(1)	588(8)	080(6)	374(5)
H(2)	792(9)	178(6)	519(6)
H(3)	81(1)	065(8)	473(6)
H(4)	37(1)	559(8)	408(8)
H(5)	22(1)	588(7)	372(6)
H(6)	25(1)	22(1)	212(9)
H(7)	32(1)	10(1)	296(9)
H(8)	493(1)	211(1)	0958(8)
H(9)	443(1)	068(1)	0965(8)
H(10)	622(1)	057(1)	1554(8)
H(11)	322(9)	507(7)	124(6)
H(12)	-27(1)	527(8)	295(8)
H(13)	-03(1)	395(8)	379(7)
H(14)	946(1)	011(1)	1830(9)
H(15)	960(1)	-132(1)	1409(9)
H(16)	1270(2)	-082(2)	072(1)
H(17)	1265(2)	-228(2)	183(1)
H(18)	1213(2)	-079(2)	229(1)

Table 7.2 Anisotropic temperature factors, and isotropic temperature factors where shown; with standard deviations in parentheses.

ATOM	U11	U22	U33	U23	U13	U12
N(1)	.041(3)	.031(4)	.054(4)	-.013(3)	-.020(3)	.007(3)
C(2)	.034(4)	.048(4)	.045(4)	-.003(3)	-.015(3)	.000(3)
N(21)	.059(4)	.058(5)	.079(5)	-.017(4)	-.046(4)	.010(4)
N(3)	.038(3)	.042(4)	.045(3)	-.010(3)	-.018(3)	-.002(3)
C(4)	.037(3)	.034(4)	.032(4)	-.006(3)	-.007(3)	-.005(3)
N(41)	.052(5)	.042(4)	.061(5)	-.018(3)	-.023(4)	-.001(4)
C(5)	.031(3)	.027(3)	.040(4)	-.006(3)	-.012(3)	-.002(3)
C(6)	.033(4)	.041(4)	.042(4)	-.009(3)	-.007(3)	-.006(3)
C(61)	.070(6)	.041(5)	.072(6)	-.016(5)	-.030(5)	-.007(5)
C(62)	.076(6)	.109(8)	.067(6)	-.043(6)	-.027(5)	.001(5)
C(1′)	.037(4)	.031(4)	.039(4)	-.005(3)	-.013(3)	-.004(3)
C(2′)	.040(4)	.039(4)	.047(4)	-.008(3)	-.010(3)	-.013(3)
C(3′)	.057(5)	.037(4)	.048(4)	-.004(3)	-.023(4)	-.011(3)
N(31′)	.091(6)	.069(5)	.050(5)	.008(4)	-.031(4)	-.010(4)
O(31′)	.105(6)	.145(8)	.080(5)	.032(5)	-.006(5)	-.030(6)
O(32′)	.161(9)	.20(1)	.113(7)	.098(7)	-.032(7)	.008(8)
C(4′)	.056(5)	.049(5)	.067(5)	-.013(4)	-.037(4)	.003(4)
CL(4′)	.081(2)	.095(2)	.119(2)	.001(2)	-.072(2)	.007(1)
C(5′)	.033(4)	.072(6)	.069(6)	.001(5)	-.014(4)	-.002(4)
C(6′)	.041(4)	.059(5)	.050(5)	.002(4)	-.007(4)	-.009(4)
S(1")	.046(1)	.043(1)	.047(1)	.002(4)	-.007(4)	-.009(4)
O(1")	.084(5)	.127(6)	.111(5)	-.065(5)	-.049(4)	.007(4)
O(2")	.056(3)	.054(3)	.090(4)	-.009(3)	-.031(3)	.006(3)
O(3")	.086(4)	.050(4)	.099(5)	.000(3)	-.016(4)	.025(3)
C(1")	.094(7)	.085(7)	.064(6)	.000(5)	-.002(5)	-.025(6)
C(2")	.096(9)	.18(1)	.14(1)	-.05(1)	.024(8)	-.067(9)

	U ISO
H(1)	.03(2)
H(2)	.04(2)
H(3)	.05(2)
H(4)	.04(3)
H(5)	.06(2)
H(6)	.12(4)
H(7)	.09(4)
H(8)	.12(2)
H(9)	.12(2)
H(10)	.12(2)
H(11)	.05(2)
H(12)	.09(3)
H(13)	.06(3)
H(14)	.16(4)
H(15)	.16(4)
H(16)	.87(9)
H(17)	.87(9)
H(18)	.87(9)

Table 7.3 Bond Lengths(A) with standard deviations

N(1) - C(2)	1.349(9)	C(1') -C(6')	1.404(11)
N(1) - C(6)	1.379(9)	C(2') -C(3')	1.380(10)
N(1) - H(1)	0.82(7)	C(2') -H(11)	0.79(7)
C(2) -N(21)	1.343(11)	C(3') -N(31')	1.488(11)
C(2) - N(3)	1.337(9)	C(3') -C(4')	1.409(11)
N(21) - H(2)	0.89(7)	N(31')-O(31')	1.177(13)
N(21) - H(3)	0.86(8)	N(31')-O(32')	1.183(14)
N(3) - C(4)	1.367(9)	C(4') -Cl(4')	1.756(9)
C(4) -N(41)	1.329(11)	C(4') -C(5')	1.360(12)
C(4) - C(5)	1.437(9)	C(5') -C(6')	1.384(13)
N(41) - H(4)	0.62(9)	C(5') -H(12)	1.03(9)
N(41) - H(5)	0.98(8)	C(6') -H(13)	0.77(8)
C(5) - C(6)	1.357(10)	S(1") -O(1")	1.434(8)
C(5) -C(1')	1.513(10)	S(1") -O(2")	1.470(6)
C(6) -C(61)	1.504(13)	S(1") -O(3")	1.421(7)
C(61) -C(62)	1.522(15)	S(1") -C(1")	1.741(11)
C(61) - H(6)	1.13(11)	C(1") -C(2")	1.531(19)
C(61) - H(7)	0.66(11)	C(1") -H(14)	1.079(16)
C(62) - H(8)	1.080(14)	C(1") -H(15)	1.080(16)
C(62) - H(9)	1.080(14)	C(2") -H(16)	1.081(22)
C(62) -H(10)	1.080(14)	C(2") -H(17)	1.080(22)
C(1') -C(2')	1.361(10)	C(2") -H(18)	1.079(22)

Table 7.4 Angles(degrees) with standard deviations

C(2) - N(1) - C(6)	122.6(6)	C(1') -C(2') -C(3')	121.5(7)
C(2) - N(1) - H(1)	119.4(46)	C(1') -C(2') -H(11)	112.4(54)
C(6) - N(1) - H(1)	118.0(46)	C(3') -C(2') -H(11)	125.8(54)
N(1) - C(2) -N(21)	118.3(7)	C(2') -C(3') -N(31')	116.2(7)
N(1) - C(2) - N(3)	121.7(6)	C(2') -C(3') -C(4')	120.7(7)
N(21) - C(2) - N(3)	120.0(7)	N(31')-C(3') -C(4')	123.1(7)
C(2) -N(21) - H(2)	120.2(46)	C(3') -N(31')-O(31')	119.4(8)
C(2) -N(21) - H(3)	127.4(54)	C(3') -N(31')-O(32')	118.6(9)
H(2) -N(21) - H(3)	112.4(71)	O(31')-N(31')-O(32')	122.1(10)
C(2) - N(3) - C(4)	117.2(6)	C(3') -C(4') -Cl(4')	124.3(6)
N(3) - C(4) -N(41)	117.4(6)	C(3') -C(4') -C(5')	118.9(7)
N(3) - C(4) - C(5)	122.7(6)	Cl(4')-C(4') -C(5')	116.8(6)
N(41) - C(4) - C(5)	119.9(6)	C(4') -C(5') -C(6')	119.0(8)
C(4) -N(41) - H(4)	115.6(81)	C(4') -C(5') -H(12)	112.5(52)
C(4) -N(41) - H(5)	123.9(47)	C(6') -C(5') -H(12)	128.0(52)
H(4) -N(41) - H(5)	118.8(93)	C(1') -C(6') -C(5')	123.1(8)
C(4) - C(5) - C(6)	117.1(6)	C(1') -C(6') -H(13)	120.3(63)

C(4) - C(5) -C(1')	120.7(6)
C(6) - C(5) -C(1')	122.2(6)
N(1) - C(6) - C(5)	118.6(6)
N(1) - C(6) -C(61)	116.7(7)
C(5) - C(6) -C(61)	124.7(7)
C(6) -C(61) -C(62)	111.0(8)
C(6) -C(61) - H(6)	105.2(56)
C(6) -C(61) - H(7)	115.5(97)
C(62) -C(61) - H(6)	97.0(56)
C(62) -C(61) - H(7)	112.2(97)
H(6) -C(61) - H(7)	114.3(**)
C(61) -C(62) - H(8)	98.4(10)
C(61) -C(62) - H(9)	111.1(10)
C(61) -C(62) -H(10)	118.2(10)
H(8) -C(62) - H(9)	109.5(12)
H(8) -C(62) -H(10)	109.5(12)
H(9) -C(62) -H(10)	109.4(12)
C(5) -C(1') -C(2')	120.5(6)
C(5) -C(1') -C(6')	122.9(6)
C(2') -C(1') -C(6')	116.7(7)
C(5') -C(6') -H(13)	116.6(63)
O(1") -S(1") -O(2")	110.6(4)
O(1") -S(1") -O(3")	110.0(4)
O(1") -S(1") -C(1")	108.1(5)
O(2") -S(1") -O(3")	114.8(4)
O(2") -S(1") -C(1")	106.3(4)
O(3") -S(1") -C(1")	106.7(5)
S(1") -C(1") -C(2")	115.9(9)
S(1") -C(1") -H(14)	108.1(10)
S(1") -C(1") -H(15)	107.3(10)
C(2") -C(1") -H(14)	108.5(12)
C(2") -C(1") -H(15)	107.5(12)
H(14) -C(1") -H(15)	109.5(13)
C(1") -C(2") -H(16)	119.8(15)
C(1") -C(2") -H(17)	112.2(15)
C(1") -C(2") -H(18)	95.3(14)
H(16) -C(2") -H(17)	109.5(18)
H(16) -C(2") -H(18)	109.4(18)
H(17) -C(2") -H(18)	109.5(18)

Table 7.5 Torsion angles(degrees) with standard deviations

C(6) - N(1) - C(2) -N(21)	-178.3(7)
C(6) - N(1) - C(2) - N(3)	1.4(11)
H(1) - N(1) - C(2) -N(21)	3.4(54)
H(1) - N(1) - C(2) - N(3)	-176.9(53)
C(2) - N(1) - C(6) - C(5)	-0.2(10)
C(2) - N(1) - C(6) -C(61)	178.8(7)
H(1) - N(1) - C(6) - C(5)	178.1(52)
H(1) - N(1) - C(6) -C(61)	-2.9(53)
N(1) - C(2) -N(21) - H(2)	-175.5(53)
N(1) - C(2) -N(21) - H(3)	3.4(69)
N(3) - C(2) -N(21) - H(2)	4.8(54)
N(3) - C(2) -N(21) - H(3)	-176.3(68)
N(1) - C(2) - N(3) - C(4)	-0.3(10)
N(21) - C(2) - N(3) - C(4)	179.4(7)
C(2) - N(3) - C(4) -N(41)	179.3(7)
C(2) - N(3) - C(4) - C(5)	-1.9(10)
N(3) - C(4) -N(41) - H(4)	20.8(90)
N(3) - C(4) -N(41) - H(5)	-174.4(56)
C(5) - C(4) -N(41) - H(4)	-158.1(90)
C(5) - C(4) -N(41) - H(5)	6.7(57)
N(3) - C(4) - C(5) - C(6)	3.0(10)
N(3) - C(4) - C(5) -C(1')	-175.0(6)
C(5) -C(1') -C(2') -C(3')	-177.8(7)
C(5) -C(1') -C(2') -H(11)	-4.1(59)
C(6') -C(1') -C(2') -C(3')	3.4(11)
C(6') -C(1') -C(2') -H(11)	177.1(58)
C(5) -C(1') -C(6') -C(5')	179.1(8)
C(5) -C(1') -C(6') -H(13)	0.9(73)
C(2') -C(1') -C(6') -C(5')	-2.1(12)
C(2') -C(1') -C(6') -H(13)	179.7(73)
C(1') -C(2') -C(3') -N(31')	179.3(7)
C(1') -C(2') -C(3') -C(4')	-2.2(11)
H(11) -C(2') -C(3') -N(31')	6.5(67)
H(11) -C(2') -C(3') -C(4')	-175.0(66)
C(2') -C(3') -N(31')-O(31')	22.1(12)
C(2') -C(3') -N(31')-O(32')	-158.5(9)
C(4') -C(3') -N(31')-O(31')	-156.4(9)
C(4') -C(3') -N(31')-O(32')	23.0(13)
C(2') -C(3') -C(4') -Cl(4')	179.2(6)
C(2') -C(3') -C(4') -C(5')	-0.6(12)
N(31')-C(3') -C(4') -Cl(4')	-2.5(11)
N(31')-C(3') -C(4') -C(5')	177.8(8)
C(3') -C(4') -C(5') -C(6')	1.9(12)
C(3') -C(4') -C(5') -H(12)	174.9(56)

N(41)	- C(4)	- C(5)	- C(6)	-178.2(7)	Cl(4')	-C(4')	-C(5')	-C(6')	-177.9(7)
N(41)	- C(4)	- C(5)	-C(1')	3.8(10)	Cl(4')	-C(4')	-C(5')	-H(12)	-4.8(57)
C(4)	- C(5)	- C(6)	- N(1)	-1.8(10)	C(4')	-C(5')	-C(6')	-C(1')	-0.6(13)
C(4)	- C(5)	- C(6)	-C(61)	179.2(7)	C(4')	-C(5')	-C(6')	-H(13)	177.7(70)
C(1')	- C(5)	- C(6)	- N(1)	176.1(6)	H(12)	-C(5')	-C(6')	-C(1')	-172.4(66)
C(1')	- C(5)	- C(6)	-C(61)	-2.9(11)	H(12)	-C(5')	-C(6')	-H(13)	5.9(97)
C(4)	- C(5)	-C(1')	-C(2')	82.3(9)	O(1")	-S(1")	-C(1")	-C(2")	58.3(10)
C(4)	- C(5)	-C(1')	-C(6')	-99.0(9)	O(1")	-S(1")	-C(1")	-H(14)	-63.6(11)
C(6)	- C(5)	-C(1')	-C(2')	-95.6(9)	O(1")	-S(1")	-C(1")	-H(15)	178.4(9)
C(6)	- C(5)	-C(1')	-C(6')	83.2(9)	O(2")	-S(1")	-C(1")	-C(2")	177.1(9)
N(1)	- C(6)	-C(61)	-C(62)	-68.6(10)	O(2")	-S(1")	-C(1")	-H(14)	55.1(10)
N(1)	- C(6)	-C(61)	- H(6)	-172.4(58)	O(2")	-S(1")	-C(1")	-H(15)	-62.8(10)
N(1)	- C(6)	-C(61)	- H(7)	60.6(**)	O(3")	-S(1")	-C(1")	-C(2")	-60.0(10)
C(5)	- C(6)	-C(61)	-C(62)	110.4(9)	O(3")	-S(1")	-C(1")	-H(14)	178.1(9)
C(5)	- C(6)	-C(61)	- H(6)	6.6(59)	O(3")	-S(1")	-C(1")	-H(15)	60.1(10)
C(5)	- C(6)	-C(61)	- H(7)	-120.5(**)	S(1")	-C(1")	-C(2")	-H(16)	-175.7(14)
C(6)	-C(61)	-C(62)	- H(8)	-58.9(11)	S(1")	-C(1")	-C(2")	-H(17)	53.8(17)
C(6)	-C(61)	-C(62)	- H(9)	-173.6(10)	S(1")	-C(1")	-C(2")	-H(18)	-59.7(15)
C(6)	-C(61)	-C(62)	-H(10)	58.6(14)	H(14)	-C(1")	-C(2")	-H(16)	-54.0(20)
H(6)	-C(61)	-C(62)	- H(8)	50.4(57)	H(14)	-C(1")	-C(2")	-H(17)	175.6(15)
H(6)	-C(61)	-C(62)	- H(9)	-64.4(57)	H(14)	-C(1")	-C(2")	-H(18)	62.0(16)
H(6)	-C(61)	-C(62)	-H(10)	167.9(57)	H(15)	-C(1")	-C(2")	-H(16)	64.3(20)
H(7)	-C(61)	-C(62)	- H(8)	170.2(**)	H(15)	-C(1")	-C(2")	-H(17)	-66.2(18)
H(7)	-C(61)	-C(62)	- H(9)	55.5(**)	H(15)	-C(1")	-C(2")	-H(18)	-179.7(14)
H(7)	-C(61)	-C(62)	-H(10)	-72.2(**)					

Table 7.6 Hydrogen bond contact distances

Hydrogen Bond	Donor-Acceptor Distance (Å)
N(21)-H(1)$_{I}$...N(1)$_{II}$	3.077
N(41)-H(3)$_{I}$...N(3)$_{III}$	3.043
N(41)-H(4)$_{I}$...O(1"A)$_{III}$	2.922
N(21)-H(2)$_{I}$...O(1"A)$_{I}$	3.131
N(2"A)-H(15)$_{I}$...O(1"B)$_{IV}$	2.847
N(2"B)-H(20)$_{I}$...O(1"A)$_{V}$	2.883

The subscripts I, II, III, IV and V refer to the equivalent positions:

X, Y, Z.

-X, 1-Y, 1-Z.

1-X, -Y, 1-Z.

-1+X, Y, -1+Z.

X, Y, 1+Z.

Figure 7.1 Nitropyrimethamine Molecular plot and numbering scheme

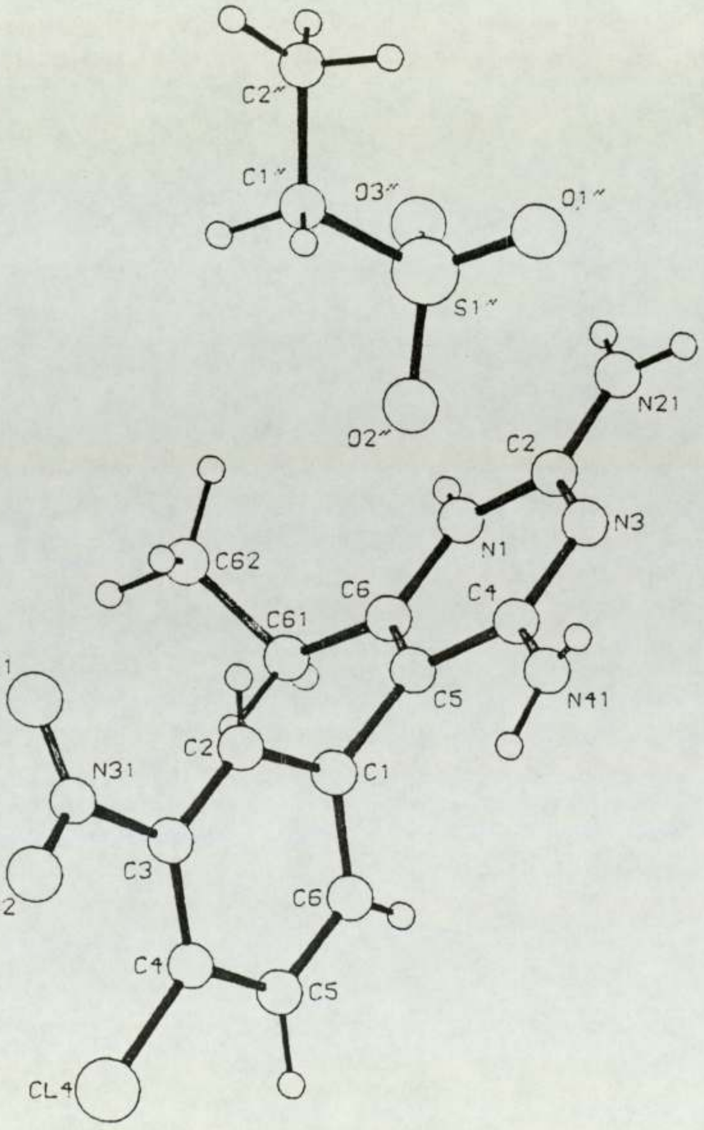
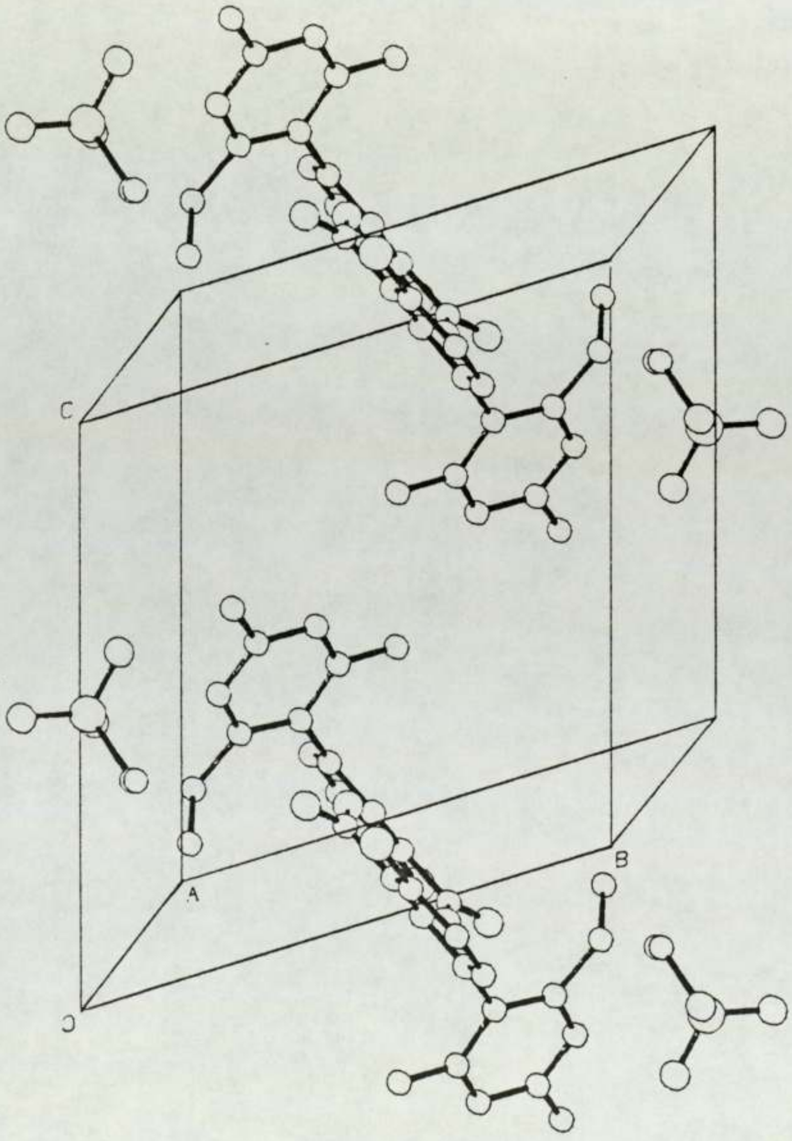


Figure 7.2 Nitropyrimethamine Unit cell packing diagram



7.5 Results and Discussion

Nitropyrimethamine (90), a potent inhibitor of DHFR from L1210 cells (91) also exhibits a high level of activity against rat liver enzyme as the ethanesulphonate salt (92), (I_{50} = 7.5 x 10^{-8}). The presence of the nitro group offers the possibility of enhanced hydrogen bonding in the enzyme active site, although no formal hydrogen bonds involving the oxygens of this moiety are observed in the crystal. There are however contacts at hydrogen bond distances between these oxygens and phenyl substituent ring hydrogens which may cause the observed out of plane twist of the nitro group of 22.1° (Table 7.5). The rotational freedom observed here in response to crystal packing forces would facilitate hydrogen bond formation in the active site.

As would be expected hydrogen bonding (Table 7.6) is observed between N(21) amino and N(1) ring hydrogens and oxygens of the ethanesulphonate counter ion, modelling the strong ionic and hydrogen bond interaction found in the substrate-bound enzyme. Dimerisation, involving N(41) amino hydrogen donation to N(3) of the pyrimidine ring, occurs across a centre of symmetry and other amino hydrogens complete an extended hydrogen bonding pattern throughout the crystal, bonding to the remaining oxygen O(3") of the ethanesulphonate molecules of adjacent pairs.

CHAPTER 8

CHAPTER 8

The Crystal Structure of 2,4-diamino-5-(4-dibenzylamino-3-nitrophenyl)-6-ethylpyrimidine (Dibenzyl).

8.1 Experimental

A crystalline sample of the compound was obtained (76) and from it a suitable crystal was selected. The quality of the specimen crystal was confirmed by examining its optical birefringent properties with a polarising microscope. The single crystal was then mounted onto a glass filament secured to a goniometer head stud by the hot wax method.

8.2 Crystal Data

$C_{26} H_{26} N_6 O_2$, M = 454.274, F(000) = 480 Triclinic a = 10.336(3), b = 10.545(4), c = 11.636(3) Å, α = 80.88(3), β = 88.49(2), γ = 72.43(3)°, V = 1193 $Å^3$ (by least squares refinement on diffractometer angles for 25 automatically centred reflections, λ = 0.71069Å), space group $P\bar{1}$, Z = 2, Dx = 1.264 gcm^{-3}, Dm = 1.270 g cm^{-3} (method of floatation in solvent mixture). Bright orange monoclinic crystals. Crystal dimensions 0.6 x 0.3 x 0.1 mm, μ (Mo-Kα) = 0.48 cm^{-1}.

8.3 Data Collection and Processing

CAD4 diffractometer, W/2θ mode with w scan width = 0.95 + 0.35 tanθ, w scan speed 0.5 - 3.6 deg min^{-1}, graphite monochromated Mo-Kα radiation; 4444 reflections measured ($2.0 \leqslant \theta \leqslant 25°$ + h $\pm$ k $\pm$ l), 4189 unique (merging R = 0.0470, no correction for absorption), giving 2528 with $I > 3\sigma(I)$, number of parameters refined 307. Linear and approximately isotropic crystal decay ca. 1% (for 3 reference reflections checked every 7200 s x-ray exposure time), corrected during processing.

8.4 Structure Determination and Refinement

Data reduction followed by direct methods (SHELX76) and electron density synthesis. All hydrogens located by difference fourier analysis during isotropic least squares refinement of non-hydrogen atoms. Full matrix least squares refinement with all non-hydrogen atoms anisotropic achieved convergence in 5 cycles, and the weighting scheme $W = 2.1420/[\sigma^2(F_o)]$ gave satisfactory agreement analyses. Final R and R_w values are 0.0455 and 0.0453, (Δ/σ) max in the final refinement cycle = 0.012 [H(3) Z/C].

Table 8.1 Positional parameters (fractional coordinates * 10**4, hydrogens * 10**3) with estimated standard deviations in parentheses.

ATOM	x/a	y/b	z/c
N(1)	4118(2)	4084(2)	9114(2)
C(2)	3099(3)	4513(3)	9823(2)
N(21)	3357(2)	5117(3)	10692(2)
N((3))	1841(2)	4422(2)	9750(2)
C(4)	1558(2)	3916(3)	8839(2)
N(41)	0286(2)	3830(2)	8770(2)
C(5)	2538(2)	3490(3)	7997(2)
C(6)	3818(3)	3567(3)	8204(2)
C(61)	5007(3)	3032(3)	7444(3)
C(62)	5363(4)	4135(4)	6669(3)
C(1′)	2172(2)	3062(3)	6924(2)
C(2′)	2015(2)	3917(2)	5878(2)
C(3′)	1699(2)	3548(2)	4858(2)
C(4′)	1442(2)	2322(2)	4819(2)
C(5′)	1629(3)	1453(3)	5895(2)
C(6′)	1982(3)	1818(3)	6897(2)
N(3′)	1754(2)	4477(2)	3790(2)
O(3′1)	2276(2)	4006(2)	2937(2)
O(3′2)	1306(2)	5695(2)	3805(2)
N(1")	1015(2)	1983(2)	3832(2)
C("A)	-0012(3)	2946(3)	3029(2)
C(1"A)	-1338(3)	2624(3)	3036(2)
C(2"A)	-2031(3)	2457(3)	4056(3)
C(3"A)	-3293(4)	2257(3)	4034(3)
C(4"A)	-3868(4)	2232(4)	2990(4)
C(5"A)	-3203(4)	2395(4)	1976(4)
C(6"A)	-1941(3)	2582(3)	1994(3)
C("B)	1347(3)	0574(3)	3679(2)
C(1"B)	1805(3)	0368(3)	2465(2)
C(2"B)	1298(4)	-0420(4)	1877(3)
C(3"B)	1738(7)	-0640(7)	0769(5)
C(4"B)	265(1)	-0032(7)	0251(4)
C(5"B)	3142(7)	0718(4)	0811(5)
C(6"B)	2735(5)	0936(4)	1931(3)
H(1)	414(3)	529(3)	1076(2)
H(2)	286(3)	515(2)	1126(2)
H(3)	-030(3)	428(2)	925(2)
H(4)	004(2)	359(2)	817(2)
H(5)	491(2)	235(2)	699(2)
H(6)	594(3)	259(2)	783(2)
H(7)	554(4)	484(4)	721(3)
H(8)	642(5)	358(5)	633(4)
H(9)	458(4)	452(4)	602(3)
H(10)	216(2)	472(2)	582(2)
H(11)	149(2)	061(2)	593(2)
H(12)	205(2)	120(2)	761(2)
H(13)	039(2)	300(2)	221(2)
H(14)	-018(2)	379(2)	328(2)
H(15)	-162(2)	250(2)	479(2)
H(16)	-378(3)	213(3)	477(2)
H(17)	-476(3)	211(3)	301(2)
H(18)	-361(3)	243(3)	120(2)

Table 8.1 contd.

ATOM	x/a	y/b	z/c
H(19)	-139(2)	267(2)	128(2)
H(20)	056(2)	021(2)	387(2)
H(21)	209(2)	008(2)	422(2)
H(22)	060(3)	-062(3)	217(2)
H(23)	123(4)	-134(4)	042(3)
H(24)	289(5)	-018(4)	-057(3)
H(25)	362(4)	120(3)	048(3)
H(26)	308(3)	156(3)	227(3)

Table 8.2 Anisotropic temperature factors, and isotropic temperature factors where shown; with standard deviations in parentheses.

ATOM	U11	U22	U33	U23	U13	U12
N(1)	.035(1)	.084(2)	.037(1)	-.023(1)	.005(1)	-.026(1)
C(2)	.038(2)	.073(2)	.030(1)	-.014(1)	.004(1)	-.027(1)
N(21)	.051(2)	.136(3)	.037(1)	-.044(2)	.017(1)	-.054(2)
N((3))	.035(1)	.069(2)	.029(1)	-.017(1)	.017(1)	-.025(1)
C(4)	.035(2)	.051(2)	.030(1)	-.005(1)	-.001(1)	-.019(1)
N(41)	.038(1)	.077(2)	.040(1)	-.023(1)	.003(1)	-.025(1)
C(5)	.035(2)	.050(2)	.034(1)	-.012(1)	.002(1)	-.016(1)
C(6)	.040(2)	.064(2)	.038(2)	-.016(1)	.006(1)	-.018(1)
C(61)	.046(2)	.096(3)	.058(2)	-.035(2)	.012(1)	-.028(2)
C(62)	.075(2)	.114(3)	.066(2)	-.028(2)	.017(2)	-.043(2)
C(1′)	.036(2)	.048(2)	.032(1)	-.012(1)	.003(1)	-.015(1)
C(2′)	.037(2)	.040(2)	.041(1)	-.015(1)	.007(1)	-.018(1)
C(3′)	.035(1)	.043(2)	.028(1)	-.008(1)	.007(1)	-.015(1)
C(4′)	.036(1)	.041(2)	.035(1)	-.014(1)	.004(1)	-.012(1)
C(5′)	.060(2)	.044(2)	.043(2)	-.012(1)	.003(1)	-.025(1)
C(6′)	.060(2)	.050(2)	.034(1)	-.005(1)	-.001(1)	-.021(2)
N(3′)	.044(1)	.057(2)	.035(1)	-.008(1)	.002(1)	-.025(1)
O(3′1)	.081(2)	.077(2)	.038(1)	-.017(1)	.022(1)	-.037(1)
O(3′2)	.067(1)	.046(1)	.056(1)	-.002(1)	-.002(1)	-.020(1)
N(1")	.055(2)	.040(1)	.041(1)	-.015(1)	-.009(1)	-.011(1)
C("A)	.054(2)	.044(2)	.042(2)	-.012(1)	-.004(1)	-.015(1)
C(1"A)	.053(2)	.037(2)	.050(2)	-.012(1)	-.007(1)	-.012(1)
C(2"A)	.057(2)	.044(2)	.062(2)	-.010(2)	.001(2)	-.016(2)
C(3"A)	.060(2)	.054(2)	.104(3)	-.011(2)	.014(2)	-.019(2)
C(4"A)	.053(2)	.070(3)	.149(4)	-.030(3)	-.014(3)	-.019(2)
C(5"A)	.071(3)	.081(3)	.103(3)	-.033(2)	-.028(2)	-.022(2)
C(6"A)	.065(2)	.064(2)	.067(2)	-.022(2)	-.017(2)	-.016(2)
C("B)	.058(2)	.045(2)	.046(2)	-.017(1)	-.001(1)	-.015(1)
C(1"B)	.064(2)	.051(2)	.048(2)	-.020(2)	-.004(2)	-.001(2)
C(2"B)	.074(3)	.124(3)	.100(3)	-.077(3)	-.021(2)	.005(2)
C(3"B)	.166(6)	.149(6)	.102(4)	-.091(4)	-.058(4)	.044(4)
C(4"B)	.27(1)	.111(5)	.058(3)	-.009(3)	.003(4)	.073(6)
C(5"B)	.255(7)	.068(3)	.113(4)	-.008(3)	.107(5)	.003(4)
C(6"B)	.136(4)	.069(3)	.091(3)	-.031(2)	.052(3)	-.025(3)

	U ISO
H(1)	.081(9)
H(2)	.066(8)
H(3)	.070(8)
H(4)	.053(7)
H(5)	.060(7)
H(6)	.067(8)
H(7)	.13(1)
H(8)	.20(2)
H(9)	.16(1)
H(10)	.025(5)
H(11)	.056(7)
H(12)	.049(6)
H(13)	.056(7)
H(14)	.047(6)
H(15)	.054(7)
H(16)	.081(9)

Table 8.2 contd.

	U ISO
H(17)	.10(1)
H(18)	.10(1)
H(19)	.065(8)
H(20)	.058(7)
H(21)	.054(7)
H(22)	.11(1)
H(23)	.18(2)
H(24)	.19(2)
H(25)	.14(1)
H(26)	.11(1)

Table 8.3 Bond Lengths(A) with standard deviations

N(1) - C(2)	1.334(4)	N(3') -O(3'2)	1.230(3)
N(1) - C(6)	1.348(4)	N(1") -C("A)	1.460(4)
C(2) -N(21)	1.349(4)	N(1") -C("B)	1.459(4)
C(2) - N(3)	1.338(4)	C("A) -C(1"A)	1.508(4)
N(21) - H(1)	0.88(3)	C("A) -H(13)	1.027(23)
N(21) - H(2)	0.83(3)	C("A) -H(14)	0.941(22)
N(3) - C(4)	1.336(3)	C(1"A)-C(2"A)	1.385(4)
C(4) -N(41)	1.350(3)	C(1"A)-C(6"A)	1.391(4)
C(4) - C(5)	1.414(4)	C(2"A)-C(3"A)	1.384(5)
N(41) - H(3)	0.90(3)	C(2"A)-H(15)	0.980(23)
N(41) - H(4)	0.850(23)	C(3"A)-C(4"A)	1.374(6)
C(5) - C(6)	1.379(4)	C(3"A)-H(16)	0.99(3)
C(5) -C(1')	1.487(4)	C(4"A)-C(5"A)	1.363(6)
C(6) -C(61)	1.514(4)	C(4"A)-H(17)	0.97(3)
C(61) -C(62)	1.487(5)	C(5"A)-C(6"A)	1.378(5)
C(61) - H(5)	0.991(24)	C(5"A)-H(18)	1.00(3)
C(61) - H(6)	1.01(3)	C(6"A)-H(19)	1.001(24)
C(62) - H(7)	1.10(4)	C("B) -C(1"B)	1.505(4)
C(62) - H(8)	1.17(5)	C("B) -H(20)	1.003(23)
C(62) - H(9)	1.06(4)	C("B) -H(21)	0.970(23)
C(1') -C(2')	1.377(3)	C(1"B)-C(2"B)	1.377(5)
C(1') -C(6')	1.389(4)	C(1"B)-C(6"B)	1.366(5)
C(2') -C(3')	1.382(3)	C(2"B)-C(3"B)	1.387(8)
C(2') -H(10)	0.899(19)	C(2"B)-H(22)	0.86(3)
C(3') -C(4')	1.404(3)	C(3"B)-C(4"B)	1.371(10)
C(3') -N(3')	1.467(3)	C(3"B)-H(23)	1.15(4)
C(4') -C(5')	1.410(4)	C(4"B)-C(5"B)	1.315(10)
C(4') -N(1")	1.376(3)	C(4"B)-H(24)	1.01(4)
C(5') -C(6')	1.375(4)	C(5"B)-C(6"B)	1.391(7)
C(5') -H(11)	0.941(23)	C(5"B)-H(25)	0.86(4)
C(6') -H(12)	0.956(22)	C(6"B)-H(26)	0.97(3)
N(3') -O(3'1)	1.220(3)		

Table 8.4 Angles(degrees) with standard deviations

C(2) - N(1) - C(6)	115.96(24)	O(3'1)-N(3') -O(3'2)	123.01(23)
N(1) - C(2) -N(21)	116.7(3)	C(4') -N(1") -C("A)	122.14(21)
N(1) - C(2) - N(3)	126.6(3)	C(4') -N(1") -C("B)	120.43(21)
N(21) - C(2) - N(3)	116.7(3)	C("A) -N(1") -C("B)	116.11(22)
C(2) -N(21) - H(1)	122.1(17)	N(1") -C("A) -C(1"A)	113.45(23)
C(2) -N(21) - H(2)	118.0(17)	N(1") -C("A) -H(13)	108.4(13)
H(1) -N(21) - H(2)	118.1(25)	N(1") -C("A) -H(14)	106.4(14)

C(2) - N(3) - C(4)	116.58(23)	C(1"A)-C("A) -H(13)	111.7(13)
N(3) - C(4) -N(41)	116.24(23)	C(1"A)-C("A) -H(14)	107.8(14)
N(3) - C(4) - C(5)	121.82(23)	H(13) -C("A) -H(14)	108.9(19)
N(41) - C(4) - C(5)	121.94(23)	C("A) -C(1"A)-C(2"A)	121.5(3)
C(4) -N(41) - H(3)	114.3(16)	C("A) -C(1"A)-C(6"A)	120.1(3)
C(4) -N(41) - H(4)	119.1(16)	C(2"A)-C(1"A)-C(6"A)	118.3(3)
H(3) -N(41) - H(4)	123.6(23)	C(1"A)-C(2"A)-C(3"A)	120.6(3)
C(4) - C(5) - C(6)	116.01(24)	C(1"A)-C(2"A)-H(15)	119.0(14)
C(4) - C(5) -C(1')	121.26(23)	C(3"A)-C(2"A)-H(15)	120.4(14)
C(6) - C(5) -C(1')	122.64(24)	C(2"A)-C(3"A)-C(4"A)	119.7(3)
N(1) - C(6) - C(5)	122.8(3)	C(2"A)-C(3"A)-H(16)	120.1(16)
N(1) - C(6) -C(61)	114.84(25)	C(4"A)-C(3"A)-H(16)	120.2(16)
C(5) - C(6) -C(61)	122.3(3)	C(3"A)-C(4"A)-C(5"A)	120.6(4)
C(6) -C(61) -C(62)	112.0(3)	C(3"A)-C(4"A)-H(17)	117.4(18)
C(6) -C(61) - H(5)	115.1(14)	C(5"A)-C(4"A)-H(17)	122.0(18)
C(6) -C(61) - H(6)	118.6(14)	C(4"A)-C(5"A)-C(6"A)	120.0(4)
C(62) -C(61) - H(5)	110.4(14)	C(4"A)-C(5"A)-H(18)	122.5(18)
C(62) -C(61) - H(6)	96.7(14)	C(6"A)-C(5"A)-H(18)	117.4(18)
H(5) -C(61) - H(6)	102.3(20)	C(1"A)-C(6"A)-C(5"A)	120.8(3)
C(61) -C(62) - H(7)	108.6(19)	C(1"A)-C(6"A)-H(19)	115.8(14)
C(61) -C(62) - H(8)	102.7(24)	C(5"A)-C(6"A)-H(19)	123.3(14)
C(61) -C(62) - H(9)	106.1(22)	N(1") -C("B) -C(1"B)	112.07(23)
H(7) -C(62) - H(8)	106.0(31)	N(1") -C("B) -H(20)	111.8(13)
H(7) -C(62) - H(9)	117.5(29)	N(1") -C("B) -H(21)	106.2(14)
H(8) -C(62) - H(9)	114.8(32)	C(1"B)-C("B) -H(20)	109.6(13)
C(5) -C(1') -C(2')	119.86(22)	C(1"B)-C("B) -H(21)	108.4(14)
C(5) -C(1') -C(6')	123.85(24)	H(20) -C("B) -H(21)	108.6(19)
C(2') -C(1') -C(6')	116.29(23)	C("B) -C(1"B)-C(2"B)	120.0(3)
C(1') -C(2') -C(3')	121.63(21)	C("B) -C(1"B)-C(6"B)	121.3(3)
C(1') -C(2') -H(10)	121.6(12)	C(2"B)-C(1"B)-C(6"B)	118.7(3)
C(3') -C(2') -H(10)	116.7(12)	C(1"B)-C(2"B)-C(3"B)	120.3(4)
C(2') -C(3') -C(4')	123.01(20)	C(1"B)-C(2"B)-H(22)	116.4(21)
C(2') -C(3') -N(3')	115.51(19)	C(3"B)-C(2"B)-H(22)	122.1(21)
C(4') -C(3') -N(3')	121.31(20)	C(2"B)-C(3"B)-C(4"B)	119.3(6)
C(3') -C(4') -C(5')	114.35(21)	C(2"B)-C(3"B)-H(23)	110.7(22)
C(3') -C(4') -N(1")	123.95(20)	C(4"B)-C(3"B)-H(23)	129.9(22)
C(5') -C(4') -N(1")	121.69(22)	C(3"B)-C(4"B)-C(5"B)	120.5(7)
C(4') -C(5') -C(6')	121.9(3)	C(3"B)-C(4"B)-H(24)	115.9(26)
C(4') -C(5') -H(11)	119.2(14)	C(5"B)-C(4"B)-H(24)	123.5(26)
C(6') -C(5') -H(11)	118.9(14)	C(4"B)-C(5"B)-C(6"B)	121.3(6)
C(1') -C(6') -C(5')	122.7(3)	C(4"B)-C(5"B)-H(25)	122.2(25)
C(1') -C(6') -H(12)	119.4(13)	C(6"B)-C(5"B)-H(25)	115.8(25)
C(5') -C(6') -H(12)	117.9(13)	C(1"B)-C(6"B)-C(5"B)	119.8(4)
C(3') -N(3') -O(3'1)	118.46(21)	C(1"B)-C(6"B)-H(26)	122.9(19)
C(3') -N(3') -O(3'2)	118.49(21)	C(5"B)-C(6"B)-H(26)	117.0(19)

Table 8.5 Torsion angles(degrees) with standard deviations

C(6) - N(1) - C(2) -N(21)	-175.6(3)	C(2') -C(3') -C(4') -C(5')	-4.4(3)
C(6) - N(1) - C(2) - N(3)	3.4(4)	C(2') -C(3') -C(4') -N(1")	174.31(22)
C(2) - N(1) - C(6) - C(5)	0.6(4)	N(3') -C(3') -C(4') -C(5')	170.65(22)
C(2) - N(1) - C(6) -C(61)	-177.4(3)	N(3') -C(3') -C(4') -N(1")	-10.7(3)
N(1) - C(2) -N(21) - H(1)	5.1(21)	C(2') -C(3') -N(3') -O(3'1)	137.13(23)
N(1) - C(2) -N(21) - H(2)	-159.3(20)	C(2') -C(3') -N(3') -O(3'2)	-40.9(3)
N(3) - C(2) -N(21) - H(1)	-173.9(21)	C(4') -C(3') -N(3') -O(3'1)	-38.2(3)
N(3) - C(2) -N(21) - H(2)	21.6(20)	C(4') -C(3') -N(3') -O(3'2)	143.73(23)
N(1) - C(2) - N(3) - C(4)	-3.9(4)	C(3') -C(4') -C(5') -C(6')	2.4(4)
N(21) - C(2) - N(3) - C(4)	175.13(25)	C(3') -C(4') -C(5') -H(11)	-179.1(16)
C(2) - N(3) - C(4) -N(41)	-179.86(24)	N(1") -C(4') -C(5') -C(6')	-176.3(3)
C(2) - N(3) - C(4) - C(5)	0.4(4)	N(1") -C(4') -C(5') -H(11)	2.2(16)
N(3) - C(4) -N(41) - H(3)	12.6(18)	C(3') -C(4') -N(1") -C("A)	-41.8(3)
N(3) - C(4) -N(41) - H(4)	173.8(18)	C(3') -C(4') -N(1") -C("B)	151.81(23)
C(5) - C(4) -N(41) - H(3)	-167.6(18)	C(5') -C(4') -N(1") -C("A)	136.8(3)
C(5) - C(4) -N(41) - H(4)	-6.4(18)	C(5') -C(4') -N(1") -C("B)	-29.6(3)
N(3) - C(4) - C(5) - C(6)	3.1(4)	C(4') -C(5') -C(6') -C(1')	0.3(4)
N(3) - C(4) - C(5) -C(1')	-173.58(24)	C(4') -C(5') -C(6') -H(12)	177.2(15)
N(41) - C(4) - C(5) - C(6)	-176.70(25)	H(11) -C(5') -C(6') -C(1')	-178.1(16)
N(41) - C(4) - C(5) -C(1')	6.7(4)	H(11) -C(5') -C(6') -H(12)	-1.3(22)
C(4) - C(5) - C(6) - N(1)	-3.6(4)	C(4') -N(1") -C("A) -C(1"A)	-110.8(3)
C(4) - C(5) - C(6) -C(61)	174.3(3)	C(4') -N(1") -C("A) -H(13)	124.5(14)
C(1') - C(5) - C(6) - N(1)	173.00(25)	C(4') -N(1") -C("A) -H(14)	7.5(14)
C(1') - C(5) - C(6) -C(61)	-9.1(4)	C("B) -N(1") -C("A) -C(1"A)	56.1(3)
C(4) - C(5) -C(1') -C(2')	103.5(3)	C("B) -N(1") -C("A) -H(13)	-68.6(14)
C(4) - C(5) -C(1') -C(6')	-77.1(4)	C("B) -N(1") -C("A) -H(14)	174.4(14)
C(6) - C(5) -C(1') -C(2')	-72.9(3)	C(4') -N(1") -C("B) -C(1"B)	-136.27(24)
C(6) - C(5) -C(1') -C(6')	106.5(3)	C(4') -N(1") -C("B) -H(20)	100.2(14)
N(1) - C(6) -C(61) -C(62)	-76.3(3)	C(4') -N(1") -C("B) -H(21)	-18.1(15)
N(1) - C(6) -C(61) - H(5)	156.5(15)	C("A) -N(1") -C("B) -C(1"B)	56.6(3)
N(1) - C(6) -C(61) - H(6)	34.9(17)	C("A) -N(1") -C("B) -H(20)	-66.9(15)
C(5) - C(6) -C(61) -C(62)	105.6(3)	C("A) -N(1") -C("B) -H(21)	174.8(14)
C(5) - C(6) -C(61) - H(5)	-21.6(16)	N(1") -C("A) -C(1"A)-C(2"A)	54.9(4)
C(5) - C(6) -C(61) - H(6)	-143.2(16)	N(1") -C("A) -C(1"A)-C(6"A)	-129.6(3)
C(6) -C(61) -C(62) - H(7)	54.7(20)	H(13) -C("A) -C(1"A)-C(2"A)	177.8(14)
C(6) -C(61) -C(62) - H(8)	166.6(25)	H(13) -C("A) -C(1"A)-C(6"A)	-6.8(14)
C(6) -C(61) -C(62) - H(9)	-72.5(23)	H(14) -C("A) -C(1"A)-C(2"A)	-62.6(15)
H(5) -C(61) -C(62) - H(7)	-175.7(25)	H(14) -C("A) -C(1"A)-C(6"A)	112.9(14)
H(5) -C(61) -C(62) - H(8)	-63.7(29)	C("A) -C(1"A)-C(2"A)-C(3"A)	175.4(3)
H(5) -C(61) -C(62) - H(9)	57.2(27)	C("A) -C(1"A)-C(2"A)-H(15)	-3.8(16)
H(6) -C(61) -C(62) - H(7)	-69.8(24)	C(6"A)-C(1"A)-C(2"A)-C(3"A)	-0.2(5)
H(6) -C(61) -C(62) - H(8)	42.1(29)	C(6"A)-C(1"A)-C(2"A)-H(15)	-179.3(15)
H(6) -C(61) -C(62) - H(9)	163.0(27)	C("A) -C(1"A)-C(6"A)-C(5"A)	-174.8(3)
C(5) -C(1') -C(2') -C(3')	178.84(22)	C("A) -C(1"A)-C(6"A)-H(19)	6.8(16)
C(5) -C(1') -C(2') -H(10)	2.3(15)	C(2"A)-C(1"A)-C(6"A)-C(5"A)	0.8(5)
C(6') -C(1') -C(2') -C(3')	-0.6(4)	C(2"A)-C(1"A)-C(6"A)-H(19)	-177.7(15)

C(6') -C(1') -C(2') -H(10)	-177.2(14)
C(5) -C(1') -C(6') -C(5')	179.3(3)
C(5) -C(1') -C(6') -H(12)	2.5(15)
C(2') -C(1') -C(6') -C(5')	-1.3(4)
C(2') -C(1') -C(6') -H(12)	-178.1(15)
C(1') -C(2') -C(3') -C(4')	3.6(4)
C(1') -C(2') -C(3') -N(3')	-171.64(21)
H(10) -C(2') -C(3') -C(4')	-179.6(14)
H(10) -C(2') -C(3') -N(3')	5.1(14)
C(3"A)-C(4"A)-C(5"A)-H(18)	-176.5(21)
H(17) -C(4"A)-C(5"A)-C(6"A)	179.2(21)
H(17) -C(4"A)-C(5"A)-H(18)	2.4(30)
C(4"A)-C(5"A)-C(6"A)-C(1"A)	-0.8(6)
C(4"A)-C(5"A)-C(6"A)-H(19)	177.5(17)
H(18) -C(5"A)-C(6"A)-C(1"A)	176.1(20)
H(18) -C(5"A)-C(6"A)-H(19)	-5.6(26)
N(1") -C("B) -C(1"B)-C(2"B)	-134.4(3)
N(1") -C("B) -C(1"B)-C(6"B)	46.4(4)
H(20) -C("B) -C(1"B)-C(2"B)	-9.7(15)
H(20) -C("B) -C(1"B)-C(6"B)	171.1(14)
H(21) -C("B) -C(1"B)-C(2"B)	108.6(15)
H(21) -C("B) -C(1"B)-C(6"B)	-70.5(15)
C("B) -C(1"B)-C(2"B)-C(3"B)	-178.5(4)
C("B) -C(1"B)-C(2"B)-H(22)	13.4(23)
C(6"B)-C(1"B)-C(2"B)-C(3"B)	0.7(6)
C(6"B)-C(1"B)-C(2"B)-H(22)	-167.4(23)
C("B) -C(1"B)-C(6"B)-C(5"B)	179.6(4)
C("B) -C(1"B)-C(6"B)-H(26)	-6.0(23)
C(1"A)-C(2"A)-C(3"A)-C(4"A)	-0.4(5)
C(1"A)-C(2"A)-C(3"A)-H(16)	179.2(18)
H(15) -C(2"A)-C(3"A)-C(4"A)	178.7(16)
H(15) -C(2"A)-C(3"A)-H(16)	-1.7(24)
C(2"A)-C(3"A)-C(4"A)-C(5"A)	0.4(6)
C(2"A)-C(3"A)-C(4"A)-H(17)	-178.6(20)
H(16) -C(3"A)-C(4"A)-C(5"A)	-179.2(18)
H(16) -C(3"A)-C(4"A)-H(17)	1.8(27)
C(3"A)-C(4"A)-C(5"A)-C(6"A)	0.2(6)
C(2"B)-C(1"B)-C(6"B)-C(5"B)	0.4(6)
C(2"B)-C(1"B)-C(6"B)-H(26)	174.8(22)
C(1"B)-C(2"B)-C(3"B)-C(4"B)	-2.0(9)
C(1"B)-C(2"B)-C(3"B)-H(23)	178.5(23)
H(22) -C(2"B)-C(3"B)-C(4"B)	165.4(25)
H(22) -C(2"B)-C(3"B)-H(23)	-14.1(34)
C(2"B)-C(3"B)-C(4"B)-C(5"B)	2.2(11)
C(2"B)-C(3"B)-C(4"B)-H(24)	-176.6(28)
H(23) -C(3"B)-C(4"B)-C(5"B)	-178.4(28)
H(23) -C(3"B)-C(4"B)-H(24)	2.8(41)
C(3"B)-C(4"B)-C(5"B)-C(6"B)	-1.1(11)
C(3"B)-C(4"B)-C(5"B)-H(25)	-171.0(29)
H(24) -C(4"B)-C(5"B)-C(6"B)	177.6(31)
H(24) -C(4"B)-C(5"B)-H(25)	7.8(43)
C(4"B)-C(5"B)-C(6"B)-C(1"B)	-0.2(8)
C(4"B)-C(5"B)-C(6"B)-H(26)	-174.9(21)
H(25) -C(5"B)-C(6"B)-C(1"B)	170.3(27)
H(25) -C(5"B)-C(6"B)-H(26)	-4.4(35)

Table 8.6 Hydrogen bond contact distances

Hydrogen Bond	Donor-Acceptor Distance (Å)
$N(21)\text{-}H(1)_{I}\ldots N(1)_{II}$	2.999
$N(41)\text{-}H(3)_{I}\ldots N(3)_{III}$	3.091
$N(21)\text{-}H(2)_{I}\ldots O(3'1)_{IV}$	3.018
$N(41)\text{-}H(4)_{I}\ldots O(3'2)_{V}$	3.350

The subscripts I, II, III, IV, and V refer to the equivalent positions:

X, Y, Z.
1-X, 1-Y, 2-Z.
-X, 1-Y, 2-Z.
X, Y, 1+Z.
-X, 1-Y, 1-Z.

Figure 8.1 Dibenzyl Molecular plot and numbering scheme

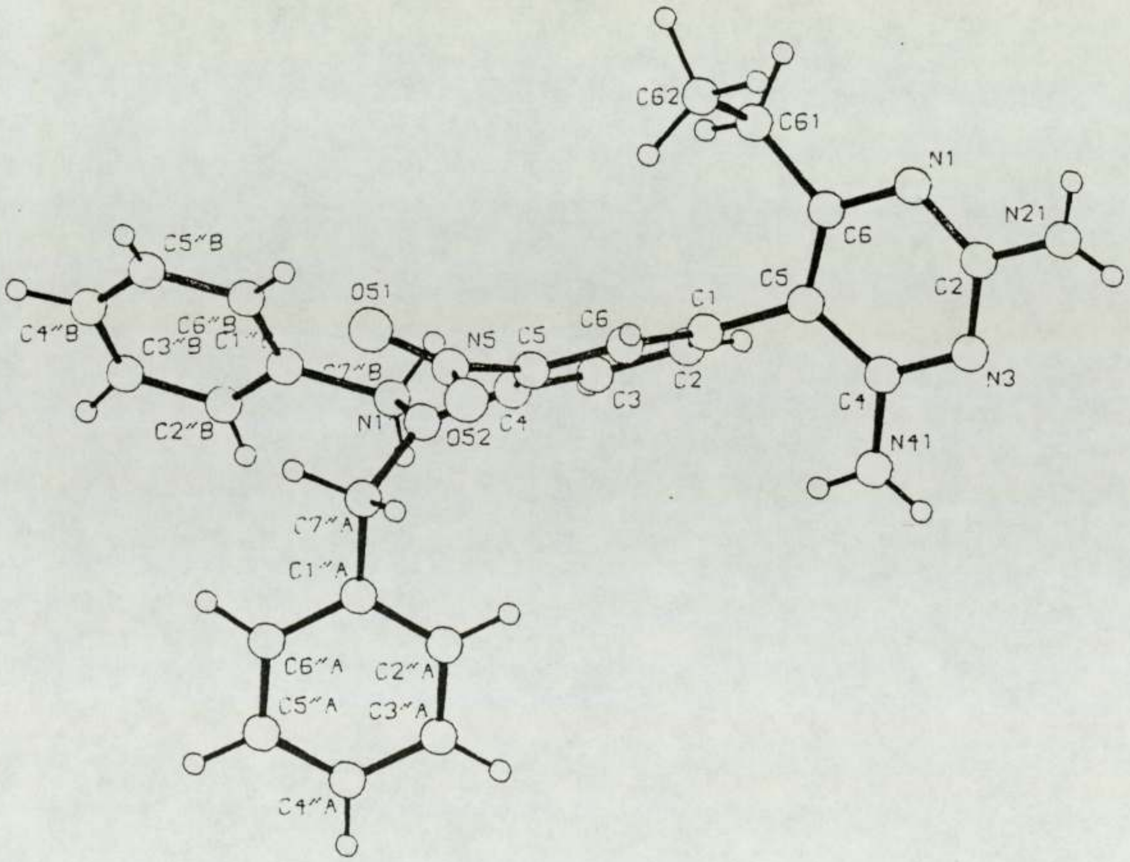
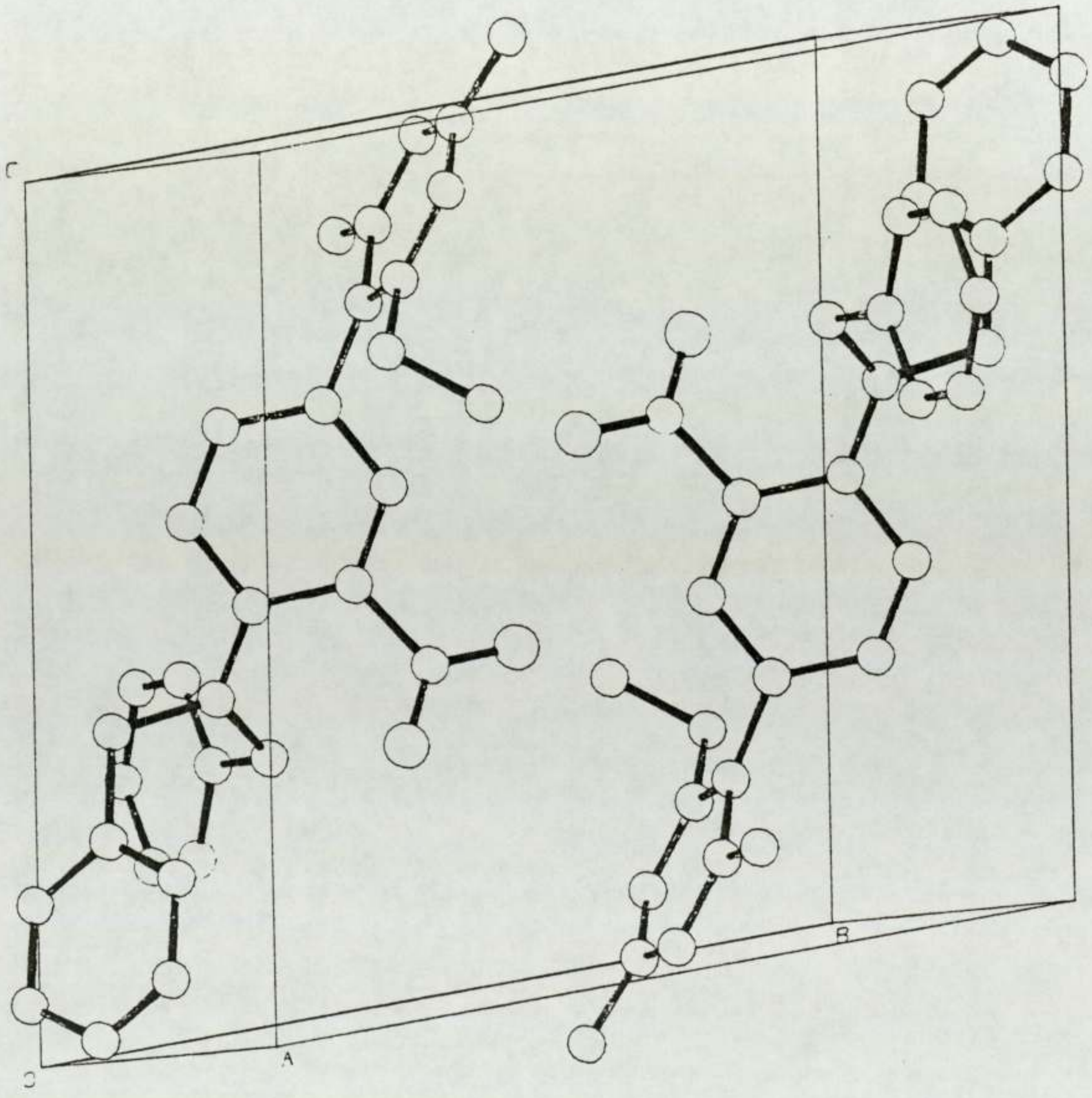


Figure 8.2 Dibenzyl Unit cell packing diagram



8.5 Results and Discussion

The compound displays an extended hydrogen bonding pattern (Table 8.6) mediated through N(21) and N(41) amino hydrogens to N(1) and N(3) of the pyrimidine ring respectively. Unlike nitropyrimethamine hydrogen bonding is observed to the nitro group oxygens of adjacent molecule by the remaining amino hydrogens. The bulky dibenzylamino substituent displays much conformational flexibility as evidenced by the high anisotropic temperature factors observed in this region. It does not appear to respond to ring stacking forces and makes only Van der Waals contacts with other molecules. This flexibility is desirable if the molecule is to make improved hydrophobic interactions in the hydrophobic pocket of the active site of DHFR.

CHAPTER 9

CHAPTER 9

A Modelling Study of an Important Enzyme-Inhibitor Interaction (97).

9.1 Introduction

The availability of crystallographic coordinates from the literature offers the possibility to construct experiments based on comparisons between common interactions found in a number of related compounds, in the crystalline state. Many interactions enable antifolate drugs to bind to the target enzyme, DHFR. Of these, hydrogen bonding and ionic attraction between the $H_2N-C=NH$ moiety of the protonated drug and carboxylate ion (ASP or GLU) from the enzyme appear to be the most important (79). Due to practical limitations protein crystallography is only able to provide a low resolution representation of the moieties in question. This study was undertaken in an attempt to refine this picture, using high resolution geometric data from X-ray studies of compounds modelling this interaction in the crystal.

It has been common to study antifolates as their ethanesulphonate ($EtSO_3^-$) salts as, with this anion, salts have tended to crystallise easily, giving crystals of suitable quality for X-ray experiments. Here in order to undertake a realistic study, a series of salts of the antimalarial antifolat drug pyrimethamine (19) with carboxylic acids (Table 9.1) are examined for structural regularities in this interaction, and compared with the known structure of Trimethoprim acetate (Fig. 9.1). Other geometric features of interest are also discussed.

Fig. 9.1 Structural Parameters Compared.

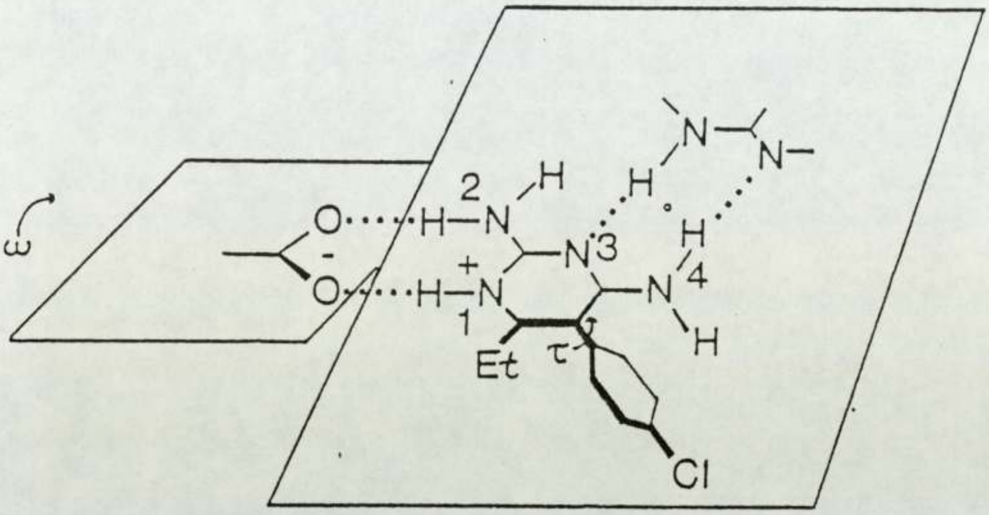


Table 9.1 Compounds Included in Study

COMPOUND	ABBREVIATION USED
Pyrimethamine hydrochloride (71)	Py
Pyrimethamine Acetate monohydrate (78)	$Py+OAc.H_2O$
Pyrimethamine Salicylate (78)	$Py+Sal^{-}A$
(two independent molecules)	$Py+Sal^{-}B$
Pyrimethamine Salicylate Isopropanol solvate (78)	$Py+Sal^{-}$ IPrOH
Trimethoprim Acetate (98)	$TMP^{+}OAc^{-}$
2,4-diamino-6-ethyl-5-(3-nitro-4-piperidino) pyrimidine HCl (73)	$NPP^{+}Cl^{-}$*

*Non carboxylate included for comparison, see (9.4).

For the carboxylate salts of pyrimethamine listed in Table 9.1 the X-ray diffraction data were collected by Joan Colby Hill (M.Phil Thesis, Aston University, 1987), who also solved and refined the structure. However, the convergence behaviour of these refinements was not entirely satisfactory. The present study involved an analysis of the problems affecting these data sets followed by repetition of the refinements with procedures designed to overcome the difficulties.

All available evidence suggested that the quality of the sample crystal of pyrimethamine acetate monohydrate was fairly good and the unit cell contents could be described by an ordered model. However, data collection had been interrupted twice for necessary alterations to the X-ray tube and diffractometer, and the three parts of the data set were not on the same scale. The two intensity monitor reflections were lower by 18.6% and 26.9% at the end of the data collection process than at the beginning. A correction factor linearly relating intensity to time was used within each block of data to compensate for any progressive degraadation of crystal or beam. In the procedure eventually used for final refinement a separate scale factor was refined for each portion of the data. Coordinates and anisotropic thermal parameters were refined for the non-hydrogen atoms. Coordinates and isotropic temperature factors were refined for hydrogen atoms except for those in the ethyl group, where the methylene hydrogen atoms were assumed to ride on the methylene carbon atom and the methyl group was treated as a rigid body. Reflections were weighted by $W = 1.5410/(\sigma^2(F) + .001079\ F^2$. Final discrepancy indices were R = 0.057, wR = 0.070 for 267 adjustable parameters and 1498 independent reflections with $F > 3\sigma$. In the final cycle no parameter shifted by more than 0.14 times its estimated standard deviation. No feature on a final difference electron density map exceeded + 0.35 and -0.22 e $\AA^{-3}$. Changes in molecular geometry from the values previously reported (J.C. Hill, 1987, Table 10.1) were negligible, never exceeding 1 unit in the last place.

For pyrimethamine salicylate the quantity and quality of data were satisfactory, but one chlorophenyl ring exhibited extremely large thermal parameters, and C-C bond distances as short as 1.34(2) Å. The problems this effect posed to the refinement process were exacerbated by the presence of two pyrimethamine salicylate ion pairs in the asymmetric unit. Even with the generous number of parameters allowed by the implementation of SHELX76 at the University of Manchester Regional Computer Centre it was necessary to refine each ion pair in a separate block. Two types of evidence suggested that the thermal parameters are indicative of genuine freedom of movement of the chlorophenyl group in question, C(1P) - C(6P), rather than some flaw in the data or the refinement model. The SHELX76 listing of intermolecular contacts contains several van der Waals interactions with atoms from the other chlorophenyl ring, C(1P) - C(6P), but none whatsoever for C(1P') - C(6P'). Also, the U_{11} values increase monotonically from the anchor point C(1P') through C(6P') and C(2P'), then C(5P') and C(3P') to C(4P') and achieve their maximum at Cl', the most distant atom. Coordinates and anisotropic thermal parameters were refined for all non-hydrogen atoms. All hydrogen atoms that could be found in difference Fourier syntheses were entered in fixed positions without refinement. Common isotropic temperature factors were refined for the following types of hydrogen atoms: N-H and NH_2 ethyl, unprimed chlorophenyl, primed chlorophenyl. With a weighting scheme of $W = 1.3550/(\sigma^2(F) + 004066\ F^2)$, where g converged to 0.004066, the final discrepancy indices were R = 0.069 and wR = 0.096 for 493 adjustable parameters and 2816 reflections with $F > 3\sigma$. Except for one of the common temperature factors, which shifted by 0.28 times its estimated standard deviation, no parameter changed by more than 0.12 e.s.d. in the last cycle in which it was refined. No feature on a diffence electron density map exceeded + 0.43 or -0.37 e $\AA^{-3}$. Changes in molecular geometry from that in Table 10.1 of J.C.H. once again never exceeded 1 in the last digit.

The problem with the third data set, pyrimethamine salicylate isopropanol solvate, was more serious. During data collection the intensity monitor reflections declined rapidly, and the experiment was prematurely terminated when the decline reached ca. 25%. It seems possible that isopropanol was being lost since other crystals in the same batch deteriorated on standing. All further attempts to grow crystals of the isopropanol solvate failed, crystals of the unsolvated form appearing instead. Therefore it was necessary to work with data that were few in number, unequally distributed in reciprocal space, and in need of drastic correction for intensity decline. It was considered that the refinements attempted by J.C.H. had been over-ambitious, and a more suitable goal would be to refine coordinates and anisotropic thermal parameters for the chlorine atom only, coordinates and isotropic temperature factors for other non-hydrogen atoms, and coordinates and common isotropic temperature factors only for those hydrogen atoms (NH, NH_2 and methylene) that were relatively distinct on a difference Fourier synthesis and of importance in describing the intermolecular interactions and conformation. Other hydrogen atoms were placed in calculated positions. The salicylate ring exhibited two-fold positional disorder about the bond from carboxyl group to ring. This had the effect of positioning the phenolic hydroxyl group on either side of the carboxylate ion, where intramolecular hydrogen bonding would be equally possible. A site occupancy factor of s for one position and 1 - s for the other was included in the refinement and converged to s = 0.3743. Reflections were weighted by $W = 4.6680/(\sigma^2(F) + .000434\ F^2)$ where g converged to 0.000434. The final discrepancy indices were R = 0.102, wR = 0.125 for 170 adjustable parameters and 955 observed reflections with $F > 3\sigma$. Parameter shifts did not exceed 0.10 e.s.d. in the final cycle. The largest positive and negative peaks on a difference electron density map were +0.35 and -0.29 e A^{-3}. Not surprisingly, molecular geometry was more strongly influenced by the refinement procedure for this structure than for the other ones, with differences of as much as 0.03 Å in bond distances from those in Table 10.1 of J.C.H. (1987).

9.2 Objectives

As the study proceeded, and based on the data available, a number of objectives became clearly defined. These can be enumerated in the following way.

To assess the constancy or variability of the following geometrical features in a series of Pyrimethamine salts as the anion is changed or the phenyl ring is substituted differently.

1) Pyrimidine ring geometry
2) Relationship between pyrimidine and phenyl ring planes
3) Nature of the interaction between pyrimidine and phenyl ring planes
4) Hydrogen bond geometry
5) For carboxylate salts; angle between planes through pyrimidine and carboyxlate ions.

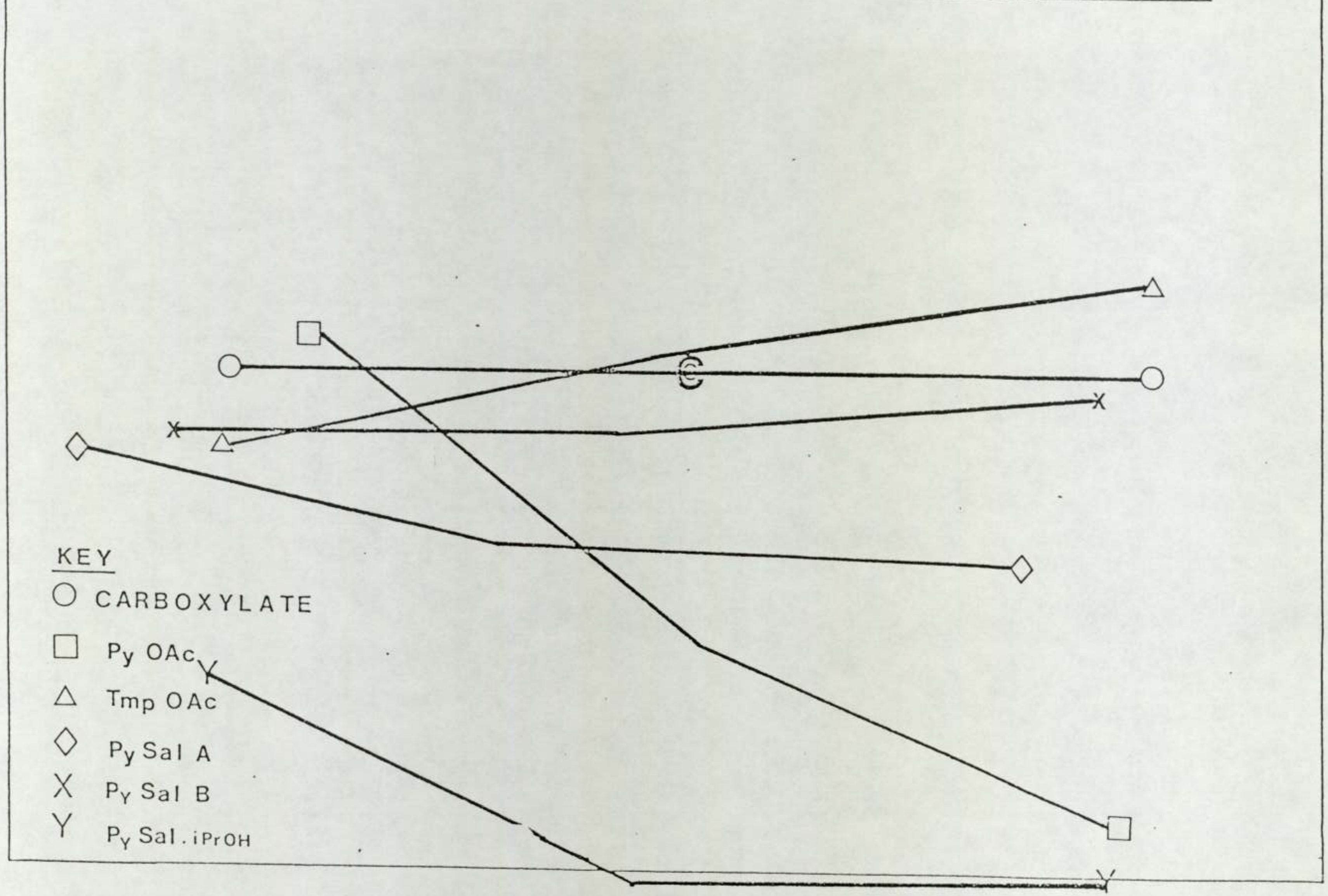
Table 9.2 H-bond distances, ring C-N amino bond lengths and dihedral angles

Compound	Py+AC- .H20	Py+Sal- A	Py+Sal- B	Py+Sal- .iPrOH	Tmp+ OAc-	NPP+Cl-
N1...O(Å)	2.670	2.707	2.688	2.73	2.60	2.806
N2...O(Å)	2.773	2.860	2.847	2.78	2.78	
N3...N(Å)	3.054	2.980	2.964	3.07	3.04	3.030
W(°)	38	12	7	27	11	
C2-N2(Å)	1.323	1.325	1.337	1.35	1.33	1.331
C4-N4(Å)	1.322	1.340	1.331	1.34	1.34	1.319
T(°)	76	70	76	79		57

Table 9.3 PYRIMIDINE RING GEOMETRY

Compound	Py+Cl-	Py+OAC-	Py+Sal-	Py+Sal-	Py+Sal-	NPP+Cl-
Distances (Å)						
N1-C2	1.369	1.356	1.306	1.339	1.33	1.345
C2-N3	1.334	1.340	1.344	1.326	1.32	1.320
N3-C4	1.344	1.347	1.359	1.344	1.38	1.340
C4-C5	1.431	1.427	1.423	1.421	1.37	1.431
C5-C6	1.346	1.359	1.349	1.350	1.35	1.352
C6-N1	1.365	1.375	1.373	1.360	1.38	1.362
C5-C1P	1.486	1.489	1.472	1.499	1.49	1.488
Interior Angles (°)						
C2-N1-C6	121.9	120.7	123.1	121.4	122	121.7
N3-C2-N1	120.9	121.9	122.1	122.7	123	121.6
C4-N3-C2	118.3	117.6	116.2	117.0	115	118.3
C5-C4-N3	112.2	122.9	123.6	122.4	125	122.3
C6-C5-C4	117.7	116.7	116.1	117.4	118	116.6
N1-C6-C5	119.0	119.7	118.9	118.9	118	119.4
Exterior Angles (°)						
N2-C2-N1	116.9	117.0	119.5	117.9	118	117.9
N2-C2-N3	112.3	121.1	118.4	119.4	120	120.4
N4-C4-N3	115.0	115.6	115.1	116.5	112	116.9
N4-C4-C5	122.8	121.6	121.3	121.1	123	120.9
C1P-C5-C4	120.7	121.0	120.2	119.0	119	121.2
C1P-C5-C6	121.6	122.2	123.6	123.6	123	122.2

DEVIATION FROM COPLANARITY BETWEEN O^{-}–C–O AND $\overset{+}{N}$=C–N PLANES



9.4 Results and Discussions

It can be seen from Table 9.3 that the changes of counter-ion of phenyl ring substituent have little effect on pyrimidine ring geometry. Another consideration would be the altered packing forces encountered in the differing environments; however the observed geometries do not vary sufficiently to demonstrate general sensitivity to these forces. The most sensitive region appears to be the N(1)-C(2) bond and, as would be expected, the external angles involving H-bonded amino groups. The pyrimidine and phenyl rings are far from coplanar (Table 9.2), this being due to steric factors and crystal packing forces.

Crystal structures of inhibitors bound to DHFR show that the protonated N(1) and the N(2) amino of the inhibitor form N-H...O H-bonds to a carboxylate ion from the enzyme. It was, in view of this, most interesting that all carboxylate salts of antifolates reported here formed a similar association, strongly suggesting that such association is spontaneous rather than constrained by the geometry of the enzyme, and implying that it should indeed make an important contribution to binding. The N-H...O distances are found to be relatively constant with mean ring N...O distance 2.58(5) Å and amino N...O 2.81(5)Å. These bond distances are about 0.1Å shorter than typical ones found in structures with ethanesulphonate counter ion thus emphasising the strength of the interaction with the carboxylate ion. This strength of interaction with carboxylate ions may be rationalised by considering electrostatic effects. In a carboxylate ion there are only two oxygen atoms to share the overall negative charge, while in the ethanesulphonate ion there are three. The N-H...N systems which produce base pairs are also found to be constant. By contrast the dihedral angle (W) between Pyrimidine ring and carboxylate ions planes (Table 9.2) shows much greater freedom and can be twisted and tilted away from coplanarity by as much as 38° (Figure 9.2); averaging 19(13)° it appears to alter easily in response to crystal packing forces.

The phenyl ring moiety of these compounds would be expected to enter into a hydrophobic interaction at the enzyme active site so an examination of this region in the crystal was indicated. This revealed that, in all the structures discussed, the 4-chlorophenyl portion of the pyrimethamine cation makes only Van der Waals contacts with adjacent molecules (as it would need to do in the active site). Temperature factors show this group to be almost unrestrained in one case. The inclusion of NPP^+Cl in the study, where this moiety is altered to 3-nitro-4-piperidino phenyl, provides a compound with enhanced polarity in this region. Protonation in the compound still occurs at N(1) of the pyrimidine ring, however in response to this substitution a water molecule adjoins the polar region and appears to form a bifurcated H-bond. It is clear from this that the environment of the phenyl ring responds to changes in polarity of substituents.

REFERENCES

1. Kepler (1611), Hooke (1663), Bergman (1773) Hauy (1782). Glusker, J.P. and Trueblood, K.N. Crystal Structure Analysis a Primer 2nd edition. Oxford University Press 1985 Chapter 2.

2. Von Laue (1912). W.H. and Bragg, W.L. X-rays and Crystal Structure, 4th ed. G. Bell and Sons, 1924. Chapter 1.

3. Friedrich W. and Knipping, P. Interferenz-Erscheinungen bei Rontgenstrahlen. Sitzungsber. Math.-Phys. Klasse der Koniglich Bayer, Akad. Wiss. Munchen, 303-322, 1912. reprinted in Naturwiss, 361-368, 1952.

4. Bragg, W.L. Proc. Roy. Soc. (London) (A) 89, 248-277, 1913.

5. Pratt, W.B. and Ruddon, R.W. The Anticancer Drugs. Oxford University Press, 1980.

6. Gilman and Philips. Science, 103, 409, 1946.

7. Seeger, D.R., Cosulich, D.B., Smith, J.M. (Jnr.), and Hultquist, M.E., J. Am. Chem. Soc., 1949, 71, 1753.

8. Farber, S., Diamond, L.K., Mercer, R.D., Slvester, R.J. (Jnr.) and Wolff, J.A., N. Engl. J. Med., 1948, 238, 787.

9. Seeger, D.R., Smith, J.M. (Jnr.) and Hultquist, M.E., J. Am. Chem. Soc., 1947, 69, 2567.

10. Blakely, R.L. The Biochemistry of Folic Acid and Related Pteridines, North Holland, Amsterdam, 1969.

11. Hitchings, G.H., Elion, G.B., Vanderwerf, H. and Falco, E.A. J. Biol. Chem., 174, 756, 1984.

12. Hitchings, G.H., Falco, E.A., Elion, G.B., Singer, S., Waring, G.B., Hutchinson, D.J. and Burchenal, J.H. Arch. Biochem., 40, 479, 1952.

13. Heidelberger, C., Griesbach, L. and Ghobar, A. Cancer Chemother. Rep., 6, 37, 1960.

14. Ferone, R. and Hitchings, G.H. J. Protozool., 13, 504, 1966.

15. Zakrzewski, S.F. and Nichol, C.A. J. Biol. Chem., 235, 2984, 1960.

16. Futterman, S. J. Biol. Chem. 228, 1301, 1957.

17. Rader, J.I. and Huennekens, F.M. (1973). Folate Coenzymediated Transfer of One-carbon Groups, in "The Enzymes, Vol. IX", Academic Press, New York.

18. Jukes, T.H. and Broquist, H.P. (1963). Metabolic Inhibitors, Hochester, R.M. and Quaztel, J.H. eds. Academic Press, New York, 481-534.

19. Futterman, S. J. Biol. Chem., 228, 1031, 1957.

20. Zakrzewski, S.F. J. Biol. Chem., 238, 1485-1490, 1963.

21. Perault, A.M. and Pullman, B. Biochem. Biophys. Acta, 52, 266, 1961.

22. Zakrzewski, S.F. and Nichol, C.A. J. Biol. Chem. 235, 2984, 1960.

23. Baker, B.R. Cancer Chemother. Rep., 4, 1-10, 1959.

24. Mathews, D.A., Alden, R.A., Bolin, J.T., Freer, S.T., Hamlin, R., Xuong, N., Kraut, J., Poe, M., Williams, M., and Hoogsteen, K. Science, 197, 452-455, 1977.

25. Mathews, D.A., Alden, R.A., Boli, J.T., Filman, D.T., Freer, S.T., Hamlin, R., Hol, W.G.T., Kisluik, R.L., Pastore, E.J., Plante, L.T., Xuong, N. and Kraut, J. J. Biol. Chem., 253, 6946-6954, 1978.

26. Hunt, W.E., Schwalbe, C.H., Bird, K. and Mallinson, P.D. J. Biochem., 187, 533-536, 1980.

27. Koetzle, T.F. and Williams, G.B. J. Amer. Chem. Soc. 98, 2074-2078, 1976.

28. Cody, V. and Zakrzewski, S.F. J. Med. Chem., 25, 427-430, 1982.

29. Schwalbe, C.H. and Cody, V. "Structural Studies on Antifolate Drugs" in "Chemistry and Biology of Pteridines". (1983). Blair, J.A., Ed., Walter de Gruyter and Co., Berlin, New York.

30. Hitchings, G.H., Elion, G.B., Vanderwerff, H. and Falco, E.A. J. Biol. Chem., 174, 765, 1948.

31. Murphy, L.M., Ellison, R.R., Karnofsky, D.A. and Burchenal, J.H. J. Clin. Invest., 33, 1388, 1954.

32. Roth, B. and Cheng, C.C. Prog. Med. Chem., 19, 270, 1982.

33. Hittings, G.H. and Burchall, J.J. Adv. Enzyme., 27. 416, 1965.

34. Hitchings, G.H. and Smith, S.L. Adv. Enz. Reg., 18, 349, 1980.

35. Geils, G.F., Scott, C.W. (Jnr.), Baugh, C.M. and Butterworth, C.E. (Jnr.). Blood, 28, 131, 1971.

36. Hill, B.T., Goldie, J.H. and Price, L.A. Br. J. Cancer, 28, 263, 1973.

37. Hill, B.T. and Price, L.A. Cancer Treat. Rev., 7, 95, 1980.

38. Hitchings, G.H. Trans. Roy. Trop. Soc. Hyg., 9, 467, 1952.

39. Baker, B.R. Cancer Chemother. Rep., 4, 1-10, 1959.

40. Baker, B.R., 'Design of Ative-Site-Directed Irreversible Enzyme Inhibitors', Wiley, New York, p. 192, 1967.

41. Bolin, J.T., Filman, D.J., Matthews, D.A., Hamlin, R.C and Kraut, J. J. Biol. Chem., 257, 13650, 1982.

42. Blakley, R.L., in 'Folates and Pterins', eds. Blakley, R.L. and Benkovic, S.J., Wiley, New York, p. 191, 1984.

43. Blakley, R.L. and Cocco, L. Biochem., 24, 4704, 1985.

44. Poe, M. J. Biol. Chem., 252, 3724, 1977.

45. Warren, R.D., Nichols, A.P. and Bender, R.A. Cancer Res., 38, 668, 1978.

46. Rosowsky, A., Beardsely, G.P., Ensminger, W.D., Lazarus, H. and Yu, C.H. J. Med. Chem., 21, 380, 1978.

47. Johns, D.G., Farquhar, D., Wolpert, M.K., Chabner, B.A. and Loo, T.L. Drug Metab. Disp., 1, 580, 1973.

48. Rosowsky, A., Lazarus, H., Yuan, G.C., Beltz., W.R., Mangini, L., Abelson, H.T., Mdest, E.J. and Frei III, E. Biochem. Pharmacol., 29, 648, 1980.

49. Rosowsky, A., Freisheim, J.H., Bader, H., Forsch., R.A., Susten, S.S., Cucchi, C.A. and Frei III, E. J. Med. Chem., 28, 660, 1985.

50. Shapiro, W.R., Young, D.F. and Mehta, B.M. N. Engl. J. Med. 293, 161, 1975.

51. Harper, M.E. and Kellems, R.E. Cancer Bull. 33, 43, 1981.

52. Bertino, J.R. Handb. Exp. Pharmakol., 72, 615, 1984.

53. Albrecht, A.M. and Biedler, J.L., in "Folate Antagonists as Therapeutic Agents", ed. Sirontak, F.M., Burchall, J.J., Ensminger, W.B. and Montogomery, J.A., Academic Press, London, vol. 1, p. 137, 1984.

54. Jonak, J.P., Zakrzewski, S.F. and Mead, L.H. J. Med. Chem. 14, 408, 1971.

55. Jonak, J.P., Zakrzewski, S.F., and Mead, L.H. J. Med. Chem. 15, 662, 1972.

56. Elslager, E.F., Johnson, J.L. and Werbel, L.M. J. Med. Chem. 26, 1753, 1983.

57. Bertino, J.R., Sawicki, W.L., Moroson, B.A., Cashmore, A.R. and Elslager, E.F. Biochem. Pharmacol., 28, 1983, 1979.

58. Ho, Y.K., Zakrzewski, S.F. and Mead, L.H. Bioche., 12, 1003, 1973.

59. Greco, W.R. and Hakala, M.T. J. Pharmacol. Exp. Ther., 212, 39, 1980.

60. Cody, V. and Zakrzewski, S.F. J. Med. Chem., 25, 427, 1982.

61. Cody, V. Cancer Biochem., Biophys., 6, 173, 1983.

62. Hill, B.T., Price, L.A., Harrison, S.I. and Goldie, J.H. Biochem. Pharmacol., 24, 535, 1975.

63. Cowan, J.H. and Jolivet, J. Clin. Res., 31, 508A, 1983.

64. Weinstein, G.D., McCullough, J.L., Eaglestein, W.H. and Golub, A. Arch. Dermatol., 117, 388, 1981.

65. Ashton, R.E., Millward-Sader, G.H. and White, J.E. J. Investig. Dermatol., 79, 229, 1982.

66. Wood, H.C.S. Observation at S.E.R.C. School 'Enzyme Chemistry Mechanisms, Inhibition, and Pharaceutical Importance', University of Strathclyde, 1984.

67. Hansch, C., Leo, A., Unger, S.H., Kim, K.H., Kaitani, D.N. and Lien, E.J. J. Med. Chem., 16, 1207, 1973.

68. Yoshimoto, M. and Hansch, C. J. Med. Chem., 19, 71, 1976.

69. Baker, B.R. "Design of Active-Site-Directed Irreversible Enzyme Inhibitors", Wiley, New York, 1967.

70. Hitchings, G.H. and Burchall, J.J. Advan. Enzymol. 17, 417, 1965.

71. Jenks, R.J. B.Sc. Project, Wolverhampton Polytechnic, 1983.

72. Haltiwanger, R.C. M.Sc. Thesis, University of Virginia, Charlottesville, U.S.A., 1971.

73. Lowe, P.R. Ph.D. Thesis, The University of Aston in Birmingham, 1984.

74. Schwalbe, C.H., Wong, K.P., Lower, P.R., Randall, A.J. and Williams, G.J.B. British Crystallographic Association Conference, Durham, U.K., 1982.

75. Cody, V. Cancer Biochem. Biophys., 6, pp. 173-177, 1983.

76. Gift from Dr. R.J. Griffin, Department of Pharmaceutical Sciences, University of Aston in Birmingham.

77. Jellinek, F. Acta Cryst 10, 277, 1957.

78. Hill J. Colby. M.Sc. Thesis, The University of Aston in Birmingham, 1987.

79. Villafranca, J.E. et al. Science, 222, 782-788, 1983.

80. Wierenga, W., Skulnick, H.I., Strungfellow, D.A., Weed, S.D., Renis, E. and Edison, E.E. J. Med. Chem., 1980, 23, 237-239.

81. LaManna, G. J. Mol. Struct. (Theochem), 152, 83-87, 1987.

82. Sharma, B.D. and McConnel, J.F. Acta Cryst., 19, 797-806, 1965.

83. Lowe, P.R., Schwalbe, C.H. and Williams, G.J.B. Acta Cryst., C43, 330-333, 1987.

84. Chidester, C.G., Duchamp, D.J., Wierenga, W. and Skulnick, H.I. Program and Abstracts, American Crystallographic Association Winter Meeting, vol. 10 p. 41, Gaithersburg, Maryland, U.S.A., 1982.

85. Cavallito, J.C., Nichol, C.A., Brenkman, Jnr., W.D., DeAngelis, R.L., Stickney, D.R., Simmons, W.S. and Sigel, C.W. Drug Metabolism and Disposition, 6, 173-177, 1978.

86. This Work.

87. Rae and Blake Acta Cryst., 20, 586, 1966.

88. Bryant, P.K., Wong, K.P., Colby, J., Schwalbe, C.H., Stevens, M.F.G., Griffin, R.J. and Bliss, E.A. Chemistry and Biology of Pteridines. Cooper, B.A. and Whitehead, V.M. Eds., Walter de Gruyter and Co., Berlin, 1986.

89. Radom, L. and Riggs, N.V. Aust. J. Chem. 35, 1071, 1982.

90. Nunberg, J.H., Kaufman, R.J., Schimke, R.T., Urlab, G. and Chasin, L.A. Proc. Nat. Acad. Sci. USA, 75, 5553, 1978.

91. Bliss, E.A. Ph.D. Thesis, The University of Aston in Birmingham, 1980.

92. Griffin, R.J. Ph.D. Thesis, The University of Aston in Birmingham, 1986.

97. Bryant, P.K., Colby, J., Jenbis, R.G., Lower, P.R. and Schwalbe, C.H. XIIIth International Congress of Crystallography, Hamburg, August 1984.

98. Bryan, R.F. and Haltiwanger, C. In Publication.

99. Gillespie, R.J. J. Am. Chem. Soc., 82, 5978, 1960.

100. Chatar Singh. Acta Cryst., 9, 861, 1965.

101. Coppens, P. Science, 158, 1577, 1967.

102. Coppers, P. and Vos, A. Acta Cryst. B27, 146, 1971.

103. Stout, G.H. and Jensen, L.H. X-ray Structure Determination, MacMillan, New York, 1968.

104. Wilson, A.J.C. Nature, 150, 151, 1942.

105. Karle, Hauptman and Christ. Acta Cryst., 11, 757, 1958.

APPENDIX I

Appendix I

X-ray Crystallography

I.1 Usefulness and limitations

The technique of study crystal x-ray crystallography provides 'a window on the nano-world', (structures of dimension in the order of 10^{-9}m). Well refined structures of organic molecules give unambiguous information on connectivity and here the technique is used to confirm, refute, or merely add additional information to structures proposed on the basis of synthetic pathways, or routine analytical procedures such as mass spectrometry, NMR, infrared and ultraviolet spectroscopy and CHNO microanalysis. Structural analysis from x-ray derived atomic positions in the crystal can provide much more information than a simple connectivity. These data may be considered to fall into two general categories as shown in Table I.1.

Table I.1 Information derived from x-ray crystallography

1) Intramolecular:	Architecture (3-D structure)
	Overall dimensions, Van der Waals Volume
	Interatomic vectors, bond lengths (10^{-3} Å), bond angles (10^{-2} °)
	Hydrogen bonding
	Thermal parameters
2) Intermolecular:	Crystal packing and stacking
	Hydrogen bonding

In addition to this, estimation of bond order and hybridisation may be adduced from bond length and bond angle. The fully refined structure of an organic molecule provides a model of a local minimum energy configuration. This is particularly useful when performing computational studies on biologically active molecules which are assumed to adopt a global energy minimum in vivo, at their site of action.

Crystallography provides much information at the molecular level which is unobtainable by other available analytical procedures. Using high-powered computing facilities, sophisticated software and precision instrumentation to mimic the action of a lens, an image of the structure is produced from which information detailed in Table I.1 can be obtained. It is well to remember that this computed image approximates to the true structure and the degree to which it is either basically correct in terms of connectivity or geometry, or precise dimensionally is reflected in the difference between the observed and calculated structure amplitudes which can be compared and quoted as a figure of merit, the R factor (Appendix II). With this in mind interpretation of bond lengths and angles should be done with due consideration of the precision and overall accuracy of the determination and the concept of absolute configuration should not be neglected (if appropriate) where geometry is concerned.

When applied to the investigation of small molecules of pharmacological interest the technique suffers from two major weaknesses. The first of these is that nitrogen atoms may not be unambiguously distinguishable from carbon atoms; an important feature in the study of heterocyclic systems where the position of the nitrogen atoms is essential to the activity of the molecule.

Here, both routine analytical procedures (if the information is available) and bond lengths may be of considerable aid in interpreting location. The order of atom-atom distances is $\mathrm{C} \overset{\cdots}{-} \mathrm{C} > \mathrm{C} \overset{\cdots}{-} \mathrm{N} > \mathrm{N} \overset{\cdots}{-} \mathrm{N}$, this inequality being dependent on approximately equal π-bonding contribution in all three cases; in heterocyclic systems this is unlikely, and bond distance may only be taken as a guide to the identification of nitrogen positions. During refinement, the insertion of all heterocyclic atoms as carbons should result in unusually low temperature factors at the nitrogen positions; unfortunately, this effect is often masked by the variation in thermal motion at different points in the heterocycle. Nevertheless comparison of isotropic temperature factors (U_{iso}) for atoms at similar positions should show difference of approximately 0.02 $Å^2$ between nitrogen and carbon atoms. A combination of these methods together with knowledge of the synthetic pathway usually gives a clear indication of the positions of the nitrogen atoms.

A second weakness in x-ray derived structure analysis is the question of hydrogen atom positions. Given high-quality intensity data hydrogen density can be expected to appear in a difference Fourier synthesis with intensities in the range 0.3 to 0.4 $eÅ^{-3}$ but may well be above or below these limits. Poor quality data or large thermal parameters or indeed a combination of the two may make hydrogen positions unobservable. This problem is important where the position of protonation is essential to the understanding of the biological activity of the molecule (Section I.2). The ideal solution to this problem, provided a suitable crystal can be grown and beam facilities are available, is to collect neutron diffraction intensity data, where because hydrogen has a scattering length approximately one half in magnitude to that of carbon, hydrogen atomic positions should be easily visible in Fourier synthesis. Failure to obtain either suiable crystals or a neutron beam facility would normally result in one placing unobservable hydrogens in calculated positions and then attempting to refine them isotropically. This would only be done if their existence was demonstrated by other analytical procedures.

It is well to remember that in structure analysis of compounds of biological importance the conformation obtained relates to material in the crystalline state. The conformation, or even the isomeic form, may not be the one the molecule adopts in its biologically active state. Some idea of the conformations available may, however, be obtained by varying torsions about freely rotatable bonds thus eliminating conformations giving impossible Van der Waals interactions. In the particular case of protonated species hydrogen bonding plays an important role in biological activity. Intra- and inter-molecular hydrogen bonding arrangements between a molecule and the active site of its receptor. In this way small molecule crysallographic investigations complement those of protein crystallography.

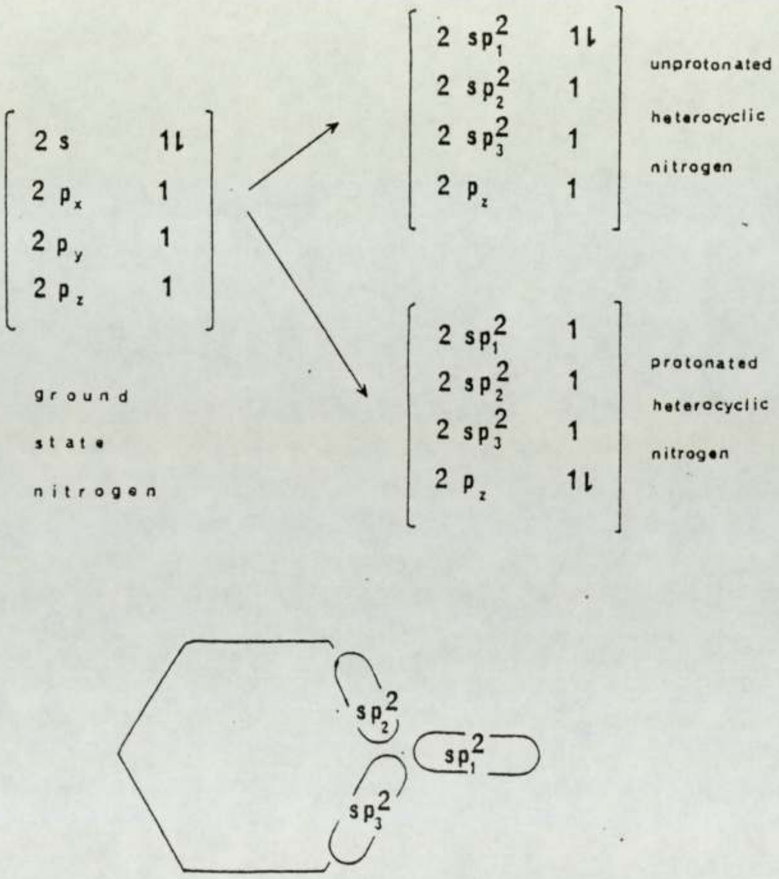
I.2 Protonation of ring nitrogens

A question of great interest in heterocyclic systems is that of protonation of ring nitrogens. N(1) or N(3) protonation is an essential element in current models of binding of 2, 4-diaminopyrimidines to the enzyme dihydrofolate reductase (see Chapter 1). Because of the difficulty of imaging hydrogen atoms from X-ray data and the scarcity of neutron facilities, the crystallographer must often rely on other evidence for the presence or absence of protonation.

In the case of the unprotonated heterocyclic nitrogen atom, the fully occupied Sp^2 hybridised lone-pair orbital leaves a single 2pz electron to be contributed to the π-system of the heterocycle, whereas the protonated nitrogen receives one electron from the hydrogen and contributes one electron to each of the Sp^2 orbitals. The remaining two electrons occupy 2pz and are non-bonding, as shown in Figure I.1. The bonds to the protonated nitrogen atoms are, therefore, formally single whereas the unprotonated species form one double bond. Allowing for delocalisation, it can therefore be expected that the bonds to the protonated nitrogen atom will be longer than corresponding bonds to the unprotonated one. Moreover, the lone-pair electrons can be expected to be more spread out than the corresponding bonding orbital and repulsion effects between the lone pair and the heterocyclic bonding orbitals can be expected to reduce the bond angle from 120° (Gillespie, 1960) (99). The crystallographic literature was searched by Chatar Singh (1965) (100) for 6-membered heterocyclic structures containing protonated and unprotonated nitrogen atoms. From his survey, the range of values 125 ± 3° (for C-NH-C) and 116 ± 3° (for C=N-C) was observed.

The above valence state electron pair repulsion theory has been investigated in detail crystallographically for simple

Fig. I.1


2 s 1↿⇂
2 p_x 1
2 p_y 1
2 p_z 1
ground state nitrogen
2 sp_1^2 1↿⇂
2 sp_2^2 1
2 sp_3^2 1
2 p_z 1
unprotonated heterocyclic nitrogen
2 sp_1^2 1
2 sp_2^2 1
2 sp_3^2 1
2 p_z 1↿⇂
protonated heterocyclic nitrogen
sp_2^2
sp_1^2
sp_3^2

systems. With use of low-temperature x-ray and neutron data from the unprotonated species S-triazine and the protonated cyanuric acid (Figure I.2) X-N synthesis were undertaken (Coppens, 1967; Coppens and Vos, 1971) (101,102). This technique is a variation of difference Fourier synthesis and, for centrosymmetric structures, is expressed as:

$$P\,(\underline{r}) = \frac{1}{V} \sum_{\underline{h}} \left[(Fo)_{\underline{h}} - (F_c)_{\underline{h}} \right] \qquad \cos 2\pi\, \underline{h}.\underline{r}$$

where the terms $(F_o)_{\underline{h}}$ refer to the x-ray data and the terms $(F_c)_{\underline{h}}$ refer to the structure factors calculated using x-ray scattering factors but atomic positions refined from neutron data. Since the neturon atomic positions are not influenced by the electron distribution, X-N synthesis reveals deviations from spherical symmetry of the electron cloud about the nuclei, and hence the bonding and lone pair orbitals. Both the lone pair orbitals of S-triazine and the bonding orbitals at the protonations sites of cyanuric acid were imaged and the slight additional spreading of the lone-pair orbitals observed. In addition, expected differences in bond distances and bond angles were found (Table I.1).

I.3 Some experimental considerations

I.3.1 Type of radiation

Although data collection on structures reported in this work used exclusively Mo-Kα(λ = 0.71069) on the CAD4 diffractometer (see Section I.4) at Aston, x-ray tubes with either molybdenum or copper targets are routinely available. Table I.3 compares the

Table I.2

Bond lengths (Å) Bond angles (°)	I	II
C(1) - N(2)	1.372	1.317
N(2) - C(2)	1.369	-
C(2) - N(1)	1.369	-
C(1) - N(2) - C(2)	124.6	114.8
C(2) - N(1) - C(2)	126.6	-

properties on which selection is usually made. The factor of 8 in the first line arises because the reflectivity of a sample is proportional to the cube of the wavelenth. This is also the case for the absorption coeffient for a single species (over an energy range where no absorption edges occur). A combination of these effects would result in observed intensities using Cu-Kαradiation which are larger than those using Mo-Kα by a factor of slightly less than 8 for the same crystal and flux of characteristic radiation.

The observed intensity is related to the structure factor by $I \propto F^2$. Lp and so the behaviour of the Lp term is reflected in the observed count rate. Whenever $2\theta_{Cu}$ is less than about 127°

Table I.3 A comparison of some properties of Cu-Kα and Mo-Kα x-rays

Property	Cu $\bar{\lambda}$ = 1.5418A	Mo $\bar{\lambda}$ = 0.7101A
Reflectivity	1	1/8
Absorption coefficent	1	1/8
Resolution (d) min)	0.77Å	0.35Å
Anomalous dispersion f" detectable	$Z \geqslant 8$	$Z \geqslant 16$
Fluroescence OK	Z < Ca, Z > Co	$Ca \leq Z \leq Ca$
Radiation damage	relative to absorption	
Detection efficieny NaI (Tl)	0.95	1.0
V crit	8.98 kV	20.0 kV

(ignoring partial polarisation of the incident beam) for a given set of diffracting planes the Lp term will favour data collected with Mo-K radiation. At higher angles however the Lp factor favours data collected with the longer wavelenth and this in fact will enhance weaker data compounds which diffract to very high angles will benefit from the increased resolution available with Mo-Kα; conversely the 'expanded' reciprocal lattice obtained by using Cu-Kα implies that the reflections will be better resolved. This may be a determining factor for studies involving large unit cells ($d > 20$Å) and is in fact the radiation of choice for protein crystallography where unit cells are commonly an order of magnitude greater than those of small molecules.

Using Cu-Kα radiation the effects of anomalous dispersion may be detectable for elements heavier than oxygen but with Mo-Kα this effect is small unless $Z \geqslant 16$. This parameter has implications for absolute configuation studies, data averaging procedures, potential coordinate errors and possible phasing techniques which have a bearing on wavelength selection. No use of anamolous dispersion has been made in this work and, as structures were in the main of CHNO, it was ignored for all practical purposes. Similarly no problems were encountered with fluroescence; however anamolous dispersion, absorption and fluroescence are intimately related so if an experiment should require an optimal $\Delta f''$ this may only be obtained at the expense of increases in the other two effects.

The most energetic radiation produced by an x-ray tube is given by $\lambda_{min} = 12.4/V$ (Kv). The voltage required to eject a K electron from a given target is obtained by substituting the wavelength of the K absorption edge into this expression. For Cu and Mo these critical voltages are given in Table I.3. The greatest intensity of the characteristic (Kα) line relative to the continuous spectrum occurs when V/V crit $\approx$ 4. Normal operating potentials are in the 40-50 kV range and so the spectrum emitted from a molybdenum tube is more contaminated with white radiation than is that from a copper tube.

It is generally assumed that the better absorbed radiation will produce the most damage to a specimen and this would favour Mo radiation. If Cu radiation is a prerequisite for an experiment the potentially greater data-collection rate may offset this problem.

I.3.2 Collimators

The simple purpose of these devices is to limit the amount of radiation emerging from the source to just that which allows all parts of the crystal to 'see' all parts of the source. To evaluate the parameters which must be set in order to satisfy this requirement the operator must consider the size of the source, the largest dimension of the crystal, the crystal to source distance as well as the diameters and positions of the collimator components. Standard collimators supplied by Enraf-Nonius with the CAD4 diffractometer are 0.4, 0.8, and 1.3 mm in diameter and in practice that collimator was selected which was slightly larger than the crystal.

I.3.2 Monochromation of primary beam

This is achieved by an appropirated oriented pyrolitic graphite crystal in the beam which will eliminate all except the desired radiation and will provide this characteristic with good efficiency. This monochromator is superior to filtering techniques (using thin metal foils of atomic number just less than that of the target material) but has two major disadvantages.

Firstly, the intensity profile of the beam is highly direction dependent. For the geometry of the CAD4 diffractometer (diffraction vector of monochromator in a plane perpendicular to the equatorial plane) the vertical direction of the beam will show a narrow profile. This fact gives an upper limit to the size of crystal which may be used. The monochromator crystal may be tuned by planar translation if significant fluctuations are noted over that portion of thebeam intercepted by a test crystal.

Secondly graphite crystals behave neither as if they were ideally perfect not as if they were ideally mosaic and this introduces an uncertainty in the form of the polarisation correction. For molybdenum radiation the error introduced by assuming the wrong form of the correction factor is about 1% (maximum at $2\theta=90°$) but this may be as high as 5% for Cu-K . This effect may be due to the fine structure of the monchromator and ideally the degree of perfection should be determined whenever the experimental arrangement changes significantly. In practice the assumption of something between zero and 50% perfection seems to be common.

I.3.4 Selection and mounting of crystal

The eventual outcome of the experiment is probably more sensitvely dependent upon this step than upon any other. The chosen crystal should be of appropriate size ($t_{opt} = 1/4\,\mu$, where μ = linear absorption coefficent), shape, physical perfection (no cracks, inclusions or vacancies) and singularity. All crystals selected for experiment in this work were microscopically examined for quality, and sharp extinction when rotated between cross-polars indicated singular nature. Absorption corrections may be required if the selected crystal has significantly different dimensions and/or large μ value. They were considered to be unnecessary for compounds studied here due to the lack of heavy atoms and consequently low μs.

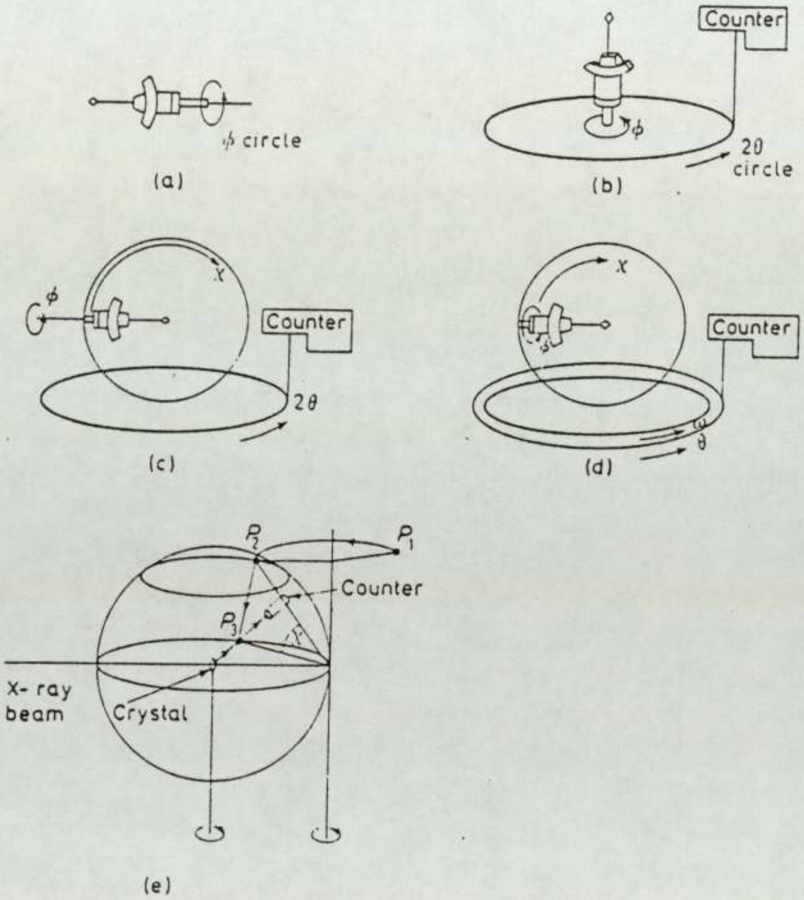
After selection suitable, air-stable, specimens may be mounted at the tip of a glass filament using a suitable adhesive i.e. one which will not attack the crystal, is x-ray transparent and has good mechanical stability. Araldite epoxy adhesive has been found to be good and, with practice, excellent mountings can be achieved where adhesive and filament are most unobtrusive.

I.4 CAD4 Diffractometry

The CAD4 automatic diffractometer collects data using moving crystal-moving counter techniques. Normally the omega/two-theta (W/2θ) scan where the detector moves through twice the angle traversed by the crystal is chosen in preference to the W-scan where the detector is stationary while the crystal rotates through an angle W (Fig. I.2). The four circles of rotation available, ϕ, θ, χ and W enable the geometry of crystal and counter to bring every set of crystal planes into diffracting position with respect to the primary beam. The unique feature of the CAD4 design is that the χ circle is replaced by functionally equivalent K axis, which is set at an oblique angle to the other axes. This design offers excellent mechanical stability and an uncluttered space above the crystal in which to position accessories, at the cost of some small restrictions on crystal positioning. In $W/2\theta$ mode all reflection data are measured with th counter revolving in a horizontal plane on the 2θ circle. If the crystal is mounted about a crystallographic axis (which it need not be), and if the crystal spindle axis is at right angles to this circle then the zero layer of reflection data of the rotation axis of the crystal will all lie in this circle and rotation of the counter through different angles 2θ will allow measuremnts to be made of all the zero layer data. In this arrangement ϕ and 2θ are concentric. Using the diffractometer in this way is limiting and the K axis (χ circle) is employed to enable any reflection which has been brought into coincidence with the reflecting sphere by means of the ϕ circle to be brought into the horizonal plane; the reflection may then be measured by setting the counter to the appropriate angle on the 2θ cirlce.

The χ circle is relted to the ϕ and 2θ circles as shown in Figure I.2, the ϕ circle (crystal spindle) mounted inside the χ circle. The $W/2\theta$ scan keeps the χ circle perpendicular to he 2θ circle. During measurement of a refection the counter moves through an angle 2θ, while the χ circle rotates about a vertical axis through an angle $W = \theta$. This is achieved by the W circle, which is concentric with the 2θ circle and on which the χ circle

Fig. I.2 Four circle Diffractometer



The four-circle diffractometer. (a) The ϕ-circle. (b) The ϕ and 2θ circles. (c) The ϕ, χ, and 2θ circles. (d) The ϕ, χ, ω and 2θ circles. (e) Rotation of the crystal through ϕ degrees moves the reciprocal lattice point from P_1 to P_2 on the reflecting sphere. Rotation of the χ circle then moves the point to P_3 when the diffracted ray lies in the equatorial plane (in which the counter moves) is mounted. The usual W setting is that where the plane of the circle bisects the incident and reflected beams.

Hence, the ϕ and χ circles bring a set of reflecting planes to that position where the reflected beam lies in a horizontal plane. The W and 2θ circles cause the crystal to rotate through a reflecting position and allow the reflected beam to fall on the counter. Automated search routines are employed to determine peak positions, from which cell constants are calculated, when values are computed for all reflections to be measured, on the basis of minimising goniometer movement.

I.5 The Temperature Factor

Atoms in a lattice may be classically regarded as point scatterers but in fact they have a defined spatial volume. This volume is a function of the atoms electron density, its envelope being defined by the thermal vibration of the atom about its equilibrium position. Restoring forces are linear but vary with direction, hence the mean-square displacement marks out an ellipsoid. The legnths and directions of its principal axes describe the average thermal motion of the atom. Combining the six principal axes U11, U22, U33 ... etc., the general form of the anisotropic tempature factor is:

$$T_{hke} = e^{-2} \quad {}^{2}(U_{11}h^{2}a^{*2}+U_{22}k^{2}b^{*2}+U_{33}({}^{2}C^{*2}+$$

$$2U_{12}hka^{*}v^{*}+2U_{23}klb^{*}c^{*}+2U_{31}lhc^{*}a^{*})$$

Non hydrogen atomic positions are commonly refined asuming fully anisotropic behaviour. Hydrogen atoms, only having on electron, are much less well observed and are therefore ascribed an isotropic thermal parameter (if they are refined at all), U_{iso}, in refinement. This parameter approximates the true thermal ellipsoid to a sphere.

APPENDIX II

APPENDIX II

Data Reduction Procedures and Crystallographic Software used in Structure Solving and Refinement. Other Associated Software.

II.1.1 Introduction

All of the crystallographic calculations reported in this work, with the exception of data reduction, were carried out using library programs stored on computer at the University of Manchester Regional Computer Centre (UMRCC).

Files containing reduced crystallographic data were initially stored at Aston on an ICL 1904S computer and then transmitted to Manchester using the UASWAN link. At Manchester they were queued on a second, front end, ICL 1904S before being submitted to a CDC 7600 computer system for processing. Replacement of Manchester's 1904S with an AMDAHL 470/V8 in April 1985 allowed direct access, via the Joint Academic Network (JANET), to the AMDAHL, thus greatly facilitating computational work and allowing data files to be stored at Manchester.

II.1.2 Data Reduction

Integrated X-ray intensities collected using a CAD4 four circle diffractometer form the raw data from which crystal structure is determined. These intensities represent all the information which is obtained by physical measurements made on

This was carried out at Aston on a HARRIS 500 computer system, using a program written by Dr. T.A. Hamor and modified by D.J. Stops. The program comprises the following routines:

i) Background subtraction (Stout and Jensen, 1968-P456) (103)

ii) Lorentz and polarisation corrections

iii) Simple linear correction of intensity as a function of irradiation time

iv) Attenuator factor correction

v) Output, in the form h, k, l, the relative structure factor (F_R), and its standard deviation $\sigma(F_R)$ calculated on the basis of counting statistics and including a correction for variation of the x-ray beam with time.

These proceses are now described in full.

II.1.3 Background Subtraction

The raw intensity data were corrected for bacground using the method for background subtraction, described in Stout and Jensen (1968-P456). This may be extended to generalised counting times for peak and background as follows:

$$N_{PK} = N_T - r\,(N_{BL} + N_{BR}) \quad 1.1$$

where N_T is the total count accumulated over the scan of the peak, N_{PK} is the peak count attributable to the reflection being measured. N_{BL} and N_{BR} are the background counts to the left and right of the peak respectively, each collected over a period of 1/r x peak count time.

II.1.4 Standard Deviation of Peak

The standard deviation in N_{PK}, due to counting statistics is given by the expression:

$$\sigma^2_{PK} = \sigma^2_{T} + \sigma^2_{BL} + \sigma^2_{BR} \qquad 1.2$$

where $\sigma^2_T = N_T$ and $\sigma_{BL} = r\sqrt{N_{BL}}$
The standard deviation of the peak must take account of any variation of the x-ray beam with time. Therefore the final standard deviation, on the peak σ^2_I, includes an expression as follows:

$$\sigma^2_{I} = \sigma^2_{PK} + (fN_{PK})^2 \qquad 1.3$$

where f is a parameter which depends upon the constancy of the x-ray beam, which in turn is dependent upon the x-ray generator and mains supply used.

II.1.5 Intensity Control and Orientation

During data collection of a small number of reflections are remeasured at regular intervals of x-ray exposure time. The observation of monotonic decrease in intensity of these reflections indicates deterioration of the crystal due to x-ray exposure; for a sudden drop in intensity a change in crystal alignment is indicated. If required a simple linear intensity correction may be applied to the measured data using the variation in intensity of these reflections over time.

Where data collection has proceeded without any overall trend in the intensities of the control reflections then the spread of measuremnts may be utilised to give an estimate of f (equation 1.3). This is achieved by attributing any part of the variation in intensity of the control reflections, not accounted for by counting statistics, to variation in intensity of the main beam. If N is the mean count for n measurments and σ^2 (N) the variance of the counts (a value expected on the basis of counting statistics to be $\bar{N}/fn-1$) then:

$$\sigma^2(N) = \frac{\bar{N}}{n-1} + (f\bar{N})^2 \qquad 1.4$$

rearranging equation 1.4 we get:

$$f = \frac{1}{\bar{N}}\left(\sigma^2(N) - \frac{\bar{N}}{n-1}\right)^{\frac{1}{2}} \qquad 1.5$$

From 1.5 it is possible to find an accurate value for f.

II.1.6 Lorentz and Polarisation Corrections

The relationship between the observed structure factor, F_o, and the square root of the intensity measured for each reflection, depends upon a number of geometrical factors which relate to the individual reflection and to the apparatus used to measure intensity. Two such factors are important, the Lorentz factor (L) and the polarisation factor (p). The Lorentz factor arises because the time required for a reciprocal lattice point to pass through the sphere of reflection is not constant, but varies with its position in reciprocal space and with the direction in which it approaches the sphere. The polarisation factor arises because of the nature of the x-ray beam and the manner in which its reflecting efficiency varies with the reflection angle, and is independent of data collection method.

The Lorentz factor depends on the precise measurment technique and is a function of 2θ. (International Tables for X-ray Crystallography, Vol. II, P266).

For the W-2 scan technique used here:

$$L = 1/\mathrm{Sin}2\theta \qquad 1.6$$

And in the presence of a graphite monochromator, the polarisation factor is given by:

$$p = \frac{\mathrm{Cos}^2\theta m + \mathrm{Cos}^2 2\theta}{2} \qquad 1.7$$

where θm is the monochromator angle.

Combining 1.6 and 1.7 we get:

$$Lp = \frac{\mathrm{Cos}^2\theta m + \mathrm{Cos}^2 2\theta}{2\mathrm{Sin}2\theta} \qquad 1.8$$

Output from the data reduction program, for each reflection, consists of the Milller indices, the relative structure factor (F_R) and $\sigma(F_R)$. The structure factor data are not on any fixed scale and must therefore be rescaled to convert into observed structure factors (F_o) for use by the Direct Methods routines (see section 1.2.5).

II.1.7 Normalised Structure Factors

The magnitude of a structure factor depends not only on the degree to which atoms scatter cooperatively but also on the scattering angles, since atoms scatter less strongly at high angles. From a theoretical point of view there is an advantage in producing a structure factor which is corrected for fall-off in scattering angle such that the numerical valueof the structure factor is independent of its position in reciprocal space. The principle method of achieving this is the Wilson Plot Method (104), where the Lp-corrected intensities have been averaged over a particular range in θ (mean value $<I_\theta>$), then:

$$\langle I_\theta \rangle = K\Sigma_\theta \exp(-2B\sin^2\theta/\lambda^2) \qquad 1.9$$

Where K is a scale factor and Σ_θ is the structure factor for randomly distributed set of atoms evaluated at the appropriate $\mathrm{Sin}\theta/\lambda$; B is an overall isotropic temperature factor.

$$\log_e \left[\frac{\langle I\theta \rangle}{\Sigma\theta} \right] = \log_e K - 2B\sin^2\theta/\lambda^2$$

a plot of the $\log_e$ of the ratio on the left against $\mathrm{Sin}2\theta/\lambda 2$

will have an intercept $\log_e K$ and a gradient $-2B$. It is essential that the averaging involved in the calculation of $\langle I_\theta \rangle$ is carried out over all points in the reciprocal lattice including those corresponding to systematic abscences.

The above procedures enable the scaling of the relative structure factors:-

i.e. $$|F_{hkl}| = KI_{hkl}^{\frac{1}{2}}$$

where $K = 1/\sqrt{K}$

and the placement of the structure factors on an absolute scale is

$$E_{hkl} = F'_{hkl}/\varepsilon\Sigma\theta$$

where ε is a factor correcting for systematic absences and the structure factors have been corrected for thermal vibration:

$$|F_{hkl}| = |F'_{hkl}| \exp(-B\sin^2\theta/\lambda)$$

These functions are incorporated in the program NORMAL. Additionally NORMAL provides the optin to calculate $\sum_\theta$ based on a randomly oriented known molecular fragment. This approach can help eliminate some of the irregularity resulting from the non-randomly nature of the structure.

SHELX76 provides an alternative routine for normalisation, selected by the MERG command. The method used is that of Karle et al (1958) (105).

Table II.1 Crystallographic Software

PROGRAM	USE
MULTAN 80 (106) (NORMAL)	Normalisation of structure factors and direct methods
EEES (SHELX76) (107)	Centrosymmetric direct methods
LS (SHELX76)	Least squares refinement
GEOM78 (108)	Least squares planes Torsion angles, bond angles and interatomic distances
PLUTO78 (109)	Generation of all molecular diagrams
BIBSER (110)	Bibliographic search program
CONNSER (110)	Connectivity search program
RETRIEVE (110)	Data retreival program
CDS (CSSR)	SERC Chemical databank system. Crystal structure search and retreival
Datred4 (111)	Data reduction
CALC (112)	As interactive molecular geometry program, including routines for the preparation of tables

II.2 Crystallographic Software

II.2.1 MULTAN80

MULTAN80 is a computer program for the largely aautomatic solution of crystal structurs having up to about 150 atoms in the asymmetric unit; it is applicable to both centrosymmetric and non-centrosymmetric structures. In its present form, structure solution involves the following stages.

i) Input reduced data and process to give normalised structure factors (E's) NORMAL

ii) Statistical analysis of E's to give space group information or to reveal abnormal features of the structure.

iii) Find the set of largest structure factors from which the structure may be determined. Typically up to about 350 structure factors for a structure containing 50 non-hydrogen atoms in the assymmetric unit.

iv) Record triplets of the type $\underline{h}\ \underline{k}\ \underline{l}$, $\underline{h}'\ \underline{k}'\ \underline{l}'$ and $\underline{h}-\underline{h}'\ \underline{k}-\underline{k}'\ \underline{l}-\underline{l}'$ the expected sign of the reflection E(h k l) being given for a centrosymmetric structure by

$$s\{E(hkl)\} \approx s\{E(h'k'l')\} \,.\, s\{E(h-h'\ k-k'\ l-l')\}$$

where S$\{\ \}$ indicates the sign of the quantity in curly brackets.

In the case of general phases for non-centrosymmetric structures:

$$\varphi h \approx \varphi h' + \varphi h\text{-}h'$$

where the vector index h is used to represent h k l. (SIGMA 2).

v) Find a good starting set for phase development from which other phases can be developed (CONVERGE)

vi) For each permuted starting point generate new phases and refine the complete set of phases via the tanget formulat (FASTAN)

vii) Calculate figures of merit (FORMs) for each phase set developed. All figures of merit are combined into a single combined figure of merit (CFOM)

viii) For the set of phases with the highest CFOM (or any other user selected set) calculate an electron density map with the Fast Fourier Transform program.

II.2.2 SHELX76 (EEEs)

An alternative, centrosymmetric, direct methods approach to structure solution is 'EEEs', the basis of the SHELX program. This method relies on starting with a very large number of permutations of signs (in the order of 2^{10} to 2^{20}), eliminating early on during sign expansion, when the process is discontinued and the set rejected.

E-maps are computed from surviving sets and figures of merit are calculated.

The choice of which direct methods package to use normally depended on whether the structure under investigation was centrosymmetric or not. SHELX was used whenever feasible, being the package of choice due to its relative ease of use. It must be said that both packages have their separate advantages. In SHELX the initial large number of sign permutatins computed, whilst in MULTAN, the normalising procedure and particularly the ability to use randomly oriented fragments in calculating E's prove to be a distinct advantage.

II.3 Least Squares Refinement

In all structures reported in this work, full matrix least squares refineent has been carried out using LS in SHELX.

The parameters refined are as follows:

i) The $|Fo|$ scale factor

ii) The atomic coordinates

iii) Either the individual isotropic temperature factor or the six components of the individual atomic anistropic vibration tensors Uij.

The version of SHELX currently available on the CDC 7600 at UMRCC allows for the refinement of up to 400 parameters simulateously with the option for blocking (BLOC) where more parameters are to be refined. Small variations to these are made on each cycle to produce a test agreement between observed and