THE STUDY OF BENTHIC INVERTEBRATE COMMUNITIES IN: -

(a) LOTIC EUTROPHIC WATERS;

(b) A RIVER IN STAGES OF RECOVERY FROM PAST POLLUTION.

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SUMMARY

A two year survey into the ecological recovery of what was once one of the most grossly polluted rivers in Britain is documented. The R. Cole was subjected to gross organic pollution as a result of two sewage effluents, one of which was also metallically toxic, these effluents being abated in 1971 and 1972. The effect of the changing water quality on the recovery of the macro-invertebrate structure of the benthic communities has been studied, noting changes in the community structure and membership.

Benthic macro-invertebrate communities living under different regimes of eutrophication in experimental streams and in natural rivers have been investigated with regard to numbers and diversity of species, productivity in terms of standing biomass, and the trophic dynamic aspect. The eutrophic conditions were simulated experimentally by mixing well oxidised sewage effluent with good quality river water, the resultant mixture being fed into three excavated experimental streams. Two rivers receiving well oxidised sewage effluent (R. Tean, Staffs., and R. Ray, Wilts.) were also utilised for comparative purposes.

One effect of eutrophication is to encourage huge growths of algae and mosses and in particular *Cladophora*, which may subsequently drastically affect the oxygen/carbon dioxide balance of a watercourse diurnally. An apparatus was developed capable of delivering and controlling fluctuating oxygen levels in both time and intensity while other parameters were kept constant. Using the apparatus, the reactions of the pollution sensitive amphipod *Gammarus pulex* was investigated.

The importance of the chironomid larvae in benthic communities has been emphasised throughout, and preliminary investigations into their respiratory rates at differing oxygen concentrations have been made using a respirometric chamber designed and developed for this purpose. An appendix containing comprehensive drawings of taxonomic features of the Chironomidae encountered has been included. The literature has been reviewed.

This work was carried out from October 1971 to July 1974 in the Department of Biological Sciences of the University of Aston in Birmingham. It has been done independently and has not been submitted for any other degree.

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CONTENTS.

Page

CHAPTER 1. INTRODUCTION 1

CHAPTER 2. LITERATURE REVIEW 11

2.1. CHEMICAL COMPOSITION OF NATURAL, EUTROPHIC, AND ORGANICALLY ENRICHED WATER. 12
- a) Nitrogen and Phosphorus 12
- b) Dissolved Gases 16
- c) Suspended Solids 18
- d) Poisons 19

2.2. SECONDARY EFFECTS OF EUTROPHICATION AND ORGANIC ENRICHMENT ON WATER CHEMISTRY 20

2.3. FACTORS INFLUENCING THE DISTRIBUTION OF BENTHIC INVERTEBRATES. 26
- a) Current Velocity 26
- b) Substratum 30
- c) Temperature 34
- d) Chemical nature of the water 36

2.4 THE EFFECTS OF ORGANIC AND NUTRIENT ENRICHMENT ON BENTHIC INVERTEBRATES 39

2.5 RECOVERY FROM ORGANIC POLLUTION 49

2.6 INTERPRETATION AND PRESENTATION OF DATA 55

2.7 EXPERIMENTAL RESPIRATORY STUDIES: - OXYGEN FLUCTUATIONS, TOXICITY AND ECOLOGY 61
- a) Temperature 64
- b) Velocity 65
- c) Oxygen reduction 66

CHAPTER 3. METHODS USED IN EXPERIMENTAL STREAMS AND RIVER SURVEYS. 74

3.1. DESCRIPTION OF HABITATS 75

3.1.1. The Experimental Streams 75

3.1.2. River surveys 81
- a) River Tean 82
- b) River Ray 86
- c) River Cole 88

3.2. ANALYTICAL METHODS 93

3.2.1. Chemistry 93
- a) pH 93
- b) Temperature 93
- c) Ammoniacal nitrogen and total oxidised nitrogen 93

Page

d) Total inorganic phosphorus 93
e) Alkalinity 93
f) Chloride 94
g) Permanganate value 94
h) Dissolved oxygen 94
i) Percentage saturation 94
j) Biochemical oxygen demand 94
k) Total suspended solids 94
l) Total hardness 94
3.2.2. Biological Sampling and Analysis 95
3.2.2.1. Quantitative sampling 95
a) Unit area of substratum 95
b) Drift 99
c) Insect Emergence 101
3.2.2.2. Qualitative sampling 103
3.2.2.3. Analysis of samples 103
a) Dry weight 103
b) Calorimetry 104
c) Taxonomy 105
d) Length/frequency analysis 106
CHAPTER 4. THE RECOVERY OF THE RIVER COLE FROM ORGANIC AND TOXIC POLLUTION 108
4.1. THE UPPER COLE 108
4.1.1. Chemical Data 108
a) Dissolved oxygen 109
b) Temperature 111
c) B.O.D. 113
d) Oxidised nitrogen and ammoniacal nitrogen 115
e) Suspended solids 118
f) Chloride 118
g) Total inorganic phosphorus 121
h) Alkalinity 121
i) pH 121
j) Permanganate value 121
k) Hardness 125
l) Metals 125
4.1.2. Biological Results and Inferences 126
4.1.3. Discussion 157
4.2. THE LOWER COLE 177

Page
4.2.1. Chemical Data 179
a) Dissolved oxygen 179
b) Temperature 181
c) B.O.D. 181
d) Ammoniacal nitrogen and oxidised nitrogen 181
e) pH 182
f) Metals 182
g) Phenol. 183
4.2.2. Biological Results and Inferences 184
4.2.3. Discussion 191
4.3. CONCLUSIONS 197
CHAPTER 5. STUDIES ON SPECIES COMPOSITION COMMUNITY STRUCTURE AND SECONDARY PRODUCTIVITY ON BENTHIC INVERTEBRATE COMMUNITIES IN RELATION TO NUTRIENT STATUS OF THE WATER. 223
5.1. INTRODUCTION 224
5.2. WATER CHEMISTRY OF THE EXPERIMENTAL STREAMS, R. TEAN AND R. RAY. 228
a) Temperature 228
b) Dissolved oxygen 235
c) Ammoniacal and oxidised nitrogen 236
d) Total inorganic phosphorus 237
e) pH 237
f) Alkalinity 238
g) Chloride 238
h) Permanganate value 239
i) Suspended solids 240
j) Hardness 241
k) Metals 241
5.2.1. Summary 242
5.3. OBSERVATIONS ON THE COLONISATION OF THE STREAMS 244
5.4. STANDING BIOMASS AND TROPHIC STRUCTURE 249
5.4.1. Discussion 264
5.5. BENTHIC MACRO-INVERTEBRATES AND EUTROPHICATION 271
a) Species distribution and the chemical effects of eutrophication. 271
b) Species distribution and the allochthonous and microbial effects of eutrophication 278

Page
c) Species distribution and filamentous algal and bryophytic growths 286
d) Later colonisation, interspecific competition and the effects of siltation 291
e) Emergence 296
5.6. CONCLUSIONS AND SUGGESTIONS FOR FURTHER WORK 301
CHAPTER 6. THE EFFECTS OF DIURNAL FLUCTUATIONS IN OXYGEN TENSIONS ON BENTHIC MACRO-INVERTEBRATES. 366
6.1. INTRODUCTION 367
6.2. DESIGN OF APPARATUS AND EXPERIMENTAL METHODS 371
6.2.1. Control of parameters 372
a) Control of oxygen tensions 372
b) Temporal fluctuations 374
c) Control of pH 374
d) Control of temperature 375
e) Re-oxygenation 376
f) Photoperiods 376
6.2.2. The experimental water 377
6.2.3. The experimental animals 377
6.3. PROCEDURE 377
6.3.1. The oxygen concentration/time fluctuations 378
6.3.2. Observation of mortality 380
6.3.3. Controls 381
6.4. RESULTS 381
6.4.1. Presentation 382
6.5. DISCUSSION 389
6.6. CONCLUSIONS 400
CHAPTER 7. RESPIROMETRIC STUDIES ON THE LARVAE OF SOME CHIRONOMIDAE (DIPTERA). 402
7.1. INTRODUCTION 403
7.2. RESPIROMETRIC APPARATUS 405
7.3. PROCEDURE 409
7.4. RESULTS 413
7.5. DISCUSSION 417
7.6. CONCLUSIONS 421

Page

CHAPTER 8. SOME OBSERVATIONS ON THE POPULATIONS OF ASELLUS AQUATICUS IN THE R. RAY (WILTSHIRE) AND EXPERIMENTAL STREAM 1. 423

8.1. INTRODUCTION 424

8.2. LIFE HISTORY 425

8.3. STANDING BIOMASS 432

8.4. CONCLUSIONS 439

APPENDIX THE TAXONOMIC FEATURES OF THE FRESHWATER LARVAL CHIRONOMIDAE IDENTIFIED FROM THE MIDLAND RIVERS AND EXPERIMENTAL STREAMS 441

REFERENCES 460

PLATES

Page

1. The Experimental Streams, mixing and sedimentation tank. 76
2. Diagrammatic representation of the Experimental Streams. 79
3. Sampling stations on the R. Tean. 84
4. Sampling station on the R. Ray. 87
5. Sampling stations on the Upper Cole. 89
6. Sampling stations on the Lower Cole. 91
7. The emergence trap, cylinder sampler and drift net specifications. 96
8. The emergence traps and drift nets in position. 102
9. Photograph of apparatus for the diurnal oxygen experiments. 369
10. Diagram of apparatus for controlling parameters during diurnal oxygen fluctuations 370
11. The Respirometer 407

12 - 24 447 - 459

TABLES

Tables 4.1 - 4.14 pages 202 - 222

Tables 5.1 - 5.23 pages 305 - 365

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CHAPTER 1.

INTRODUCTION.

In natural streams the concentration of materials needed for the maintenance of benthic communities may, at times, become limiting with consequences detrimental to autotrophic growth rates (Hawkes, 1962). Stream communities are therefore sensitive to change in nutritional status, the effects of the latter being felt at successive trophic levels.

The fertilisation of natural streams is a phenomenon mainly associated with pollution. Owens (1970) categorised the sources of enrichment into 'point' sources, i.e. sewage and industrial effluents and 'diffuse' sources, which include those like run-off, land drainage, bird droppings and leaf fall. Animal excreta from intensive farming is now considered as a 'point' source.

Owens <u>et</u> <u>al</u> (1972) placed these kinds of pollutants under three convenient headings in order to study the impact of waste discharges on the quality of the receiving water. The effects on the benthic communities of these types of pollutant can also be considered under these headings. They were: -

a) those which neither serve as plant nutrients nor are assimilated like oxygen demanding wastes, remaining unchanged or conservative, e.g. most minerals, chloride, bicarbonate, sulphate, etc.

b) nitrogen and phosphorus.

c) oxygen demanding pollutants.

This thesis is largely concerned with the last two.

Nitrogen and phosphorus are now considered to be the most significant nutrients limiting the growth of aquatic plants. In recent years the amounts of nitrogen and phosphorus in surface waters has increased in synchrony with escalating use of agrochemicals and cleansing agents. Also, and especially in lotic waters, the levels of nitrate nitrogen are increasing due to more efficient techniques in sewage purification resulting in highly nitrified effluents. Indirect evidence suggests that 50% of phosphorus in sewage effluents emanates from detergents (Owens & Wood, 1968) and it is estimated that during sewage purification only half the amount of phosphorus entering is retained. Increasing industrialisation and urbanisation has necessarily increased flow into treatment plants and some receiving rivers and streams have very high proportions of well oxidised effluent to natural water.

These nutrients encourage excess growths of algae and higher plants in natural waters which may induce changes in the chemistry, physico-chemistry, substratum, fauna and amenity value of the watercourse, these effects being enveloped by the term eutrophication. Photosynthesis and respiration from the enlarged growths can cause large diurnal fluctuations in oxygen, carbon dioxide and pH, and decomposing clumps of aquatic weeds can seriously reduce the dissolved oxygen content of a stream. The rate of oxygen utilisation of decaying weed has been estimated at half that of untreated sewage (Jewell, 1971). However, it must be realised that not all eutrophic rivers revert for example from 5% saturation with oxygen at night to 100% or more during the day. It may be that they only reach 50% saturation during daylight, (this being dependent upon numerous factors the most important of which are oxygen demand of pollutants, amount of re-aeration, solar radiation) so not only are different temporal fluctuations observed but different intensities in fluctuation also.

Although the effect of continuous low oxygen tensions on some benthic invertebrates are known, the consequences of large shifts in the diurnal oxygen/carbon dioxide balance are not documented. It was therefore decided to develop an apparatus and embark upon a preliminary examination into the effects of such fluctuations under laboratory conditions.

Large algal and macrophytic growths may modify the benthic faunal communities directly by replacing the normal food supply, and indirectly by altering the velocity and course of a current, encouraging or discouraging sedimentation thereby changing the nature of the substratum. Modifications to the fluviatile environment like these were first realised as being important ecologically by Percival and Whitehead (1929), Butcher et al (1937), Pentelow et al (1938), and Hawkes (1956).

The evaluation of eutrophication in running waters is necessary for efficient water management but methods and measures for appraisement of this are still at the developmental stage. Qualitative studies of the plankton and benthos with coincident water chemistry analysis, and more recently reinforcement with quantitative information on biomass, population dynamics and energetics of communities, are among those employed. To obtain a greater understanding of the consequence of differing nutrient levels in terms of benthic invertebrate ecology, it was felt necessary to approach the problem not only in rivers of dissimilar nutrient status but more closely under controlled field conditions. Two rivers (R. Ray, Wilts; R. Tean, Staffs.) were selected for scrutiny and the more controlled conditions were furnished by the excavation of three experimental streams in which differing proportions of nutrients were channelled. Using the above methods of study, riffle macroinvertebrate communities were investigated to realise the potential and pertinence of invertebrate ecology in eutrophication studies.

Some important consequential discoveries were anticipated especially concerning species and communities indicative of eutrophy. Chironomid larvae were given special attention because of the neglect they have received in the past due to problems with accurate identification. It was considered that some species or associations of these larvae would prove useful as "biological indicators", if not by design by serendipity. Furthermore it was expected that production measurements from the experimental streams might give an idea of the suitability of eutrophic waters for economic fish farming. One conception that did not manifest itself for reasons of time and money was to introduce fish into the Experimental Streams and monitor their growth rate and effect of grazing on the benthos.

The opportunity for investigation into the recovery of a river from pollution presented itself when two overloaded sewage works were closed down within a year of each other. The faunal communities had been subject in the past to insult by gross organic pollution and from one of the effluents, toxic contamination. The outcome of toxicity on stream fauna is usually in the form of reduction of species and numbers (diversity) according to their tolerance to the contaminants. Sometimes increases in numbers occur when a tolerant species has diminished competition for food and space. With increasing dilution downstream, species and numbers are expected to return in order of increasing sensitivity accompanied by an interspecific and intraspecific struggle to reach some form of stability.

Organic pollution is more complex in types and effects and categories b) and c) of Owens et al, shown above, demonstrate some of these types. Heavy organic pollution is typically accompanied by high suspended solids, saprobic organisms exploiting readily available high energy food and attributing to de-oxygenation of the river as a result of their respiration. It is also attended by poisons like ammonia and sulphides which tax the oxygen supplies of the river further, the synthesis of effects resulting in the familiar "oxygen sag" curve caused by high initial oxygen demand, with natural re-aeration, photosynthesis and dilution offsetting and finally satiating the demand. De-oxygenation imposes restrictions on the fauna according to their metabolic requirements, this dependence being the primum mobile of distribution of benthic organisms. Other imbalances in the water chemistry and changes in the substratum caused by deposition of suspended solids and secondary biotic factors all pressurise the ecological communities, sometimes causing well defined 'zones' of biological aggregations. Large heterotrophic increase in characteristic or organic pollution, and respiration will as a rule be greater than photosynthesis so that the ratio of respiration to photosynthesis is greater than one. Eutrophication, with its characteristically large autotrophic population is the inverse of this with photosynthesis being in excess of respiration by day. Usually at some stage of recovery from organic pollution by the stream (assuming it to be of sufficient length) the rate of respiration is compensated by photosynthesis and customarily is exceeded, in which case it may be said that the stream is now, in some respects, eutrophicated. This is quite often the end result of heavy organic enrichment, as the constituent nitrogen and phosphorus nutrients can only be utilised by autrophes when other limiting factors have been eliminated or reduced below their tolerance thresholds. In this respect it is often observed that faunal communities inhabiting mildly organic polluted waters are typical of eutrophic communities. The Asellus/Cladophora zone is a good example of this occurrence.

The biological recovery from organic pollution, as for any other type of contaminating pollutant following cessation of sources, will be, in part, a function of the resulting chemical, physical and biological characteristics of the stream. When organic pollution ceases, (as in the present case of a diverted effluent) the watercourse will be subject to an immediate drop in suspended solids, temperature (locally) and B.O.D. which illustrates three parameters likely to affect instantaneously the oxygen status of the river. The curtailment of toxic input will also immediately favour re-colonisation by less tolerant species but the release of residual toxic materials will ajourn the process by degrees. The nutrients nitrogen and phosphorus will now only be available from 'diffuse' sources unless there are 'point' sources upstream. However, the natural resumption of autotrophic density may be delayed a long time as phosphorus may be bound in bottom deposits after settling of suspended solids, and soluble phosphate ions may have combined chemically with metallic cations present to give precipitates like $FePO_4$, (Keup, 1968). Continual release of residual phosphorus from the river bed together with algal release on decomposition of nitrogen and phosphorus (Foree *et al*, 1971) could well support an increased algal population and give rise to a situation typical of mildly eutrophicated reaches. This stage of recovery involving the presence of eutrophication fauna and typically *Cladophora* may last a long time and in degrees relative to the volume and standard of the relinquished organic effluent(s). The biological characteristics of the stream, being inseparable parts of food webs, will determine at this point the presence or absence of other members, but in general an increase in algae (autotrophes) and less tolerant invertebrates (heterotrophes) are expected.

The severity and duration of the past stress will also determine how restoration of the status quo is achieved. Two broad predicaments of severity may be recognised; acute and chronic exposure. Acute exposure, which may come in the form of an industrial spill, temporary failure of oxidation processes or strike at a sewage treatment works, is very often sub-lethal to benthic communities. That is, because of the biological variability of response by individuals of a community to poisons, only a proportion of them may be killed off. These sub-lethal effects may slow down reproduction, growth, or feeding temporarily but the population may soon regain members to density dependent numbers. Generally, therefore, rapid recovery by the stream flora and fauna are expected but will vary with the severity of the exposure.

If this exposure is prolonged, gradually more of the members of the population will respond until the entire population is affected. They may be eliminated altogether by the toxic effect or by pressure exerted by more tolerant populations which eventually take over the new niche. This is chronic exposure. The ensuing changes in species complexity is not restricted to organic pollution. Typically, the number of species is reduced but the biomass of the populations may remain about the same, as more food and space and less competition are presented. Extremely severe organic contamination can reduce the number of macro-invertebrate species to one or two but numbers of animals per unit area will remain high. The recovery from chronic exposure is not expected to be so rapid due to the severity of it in the first place, and the residual toxicity in the second. Under these conditions, prolonged transitional communities may be formed. Speed of re-instatement of the natural communities will therefore be a function of the distance of the niche from the site of pollution also.

Recolonisation of the river would be foreseen to be related to the mobility of the species. Obviously fish would be the first to re-establish themselves but may be limited by the available food. The speed with which invertebrate communities could re-occupy will depend upon the existence and proximity of undamaged tributaries and downstream drift. Even so, a succession is expected emanating from interspecies differences in mobility. Insects have a distinct advantage here in being able to fly up or downstream to oviposit.

To discover the dynamics of recovery more closely, a two year ecological survey was undertaken on what was once one of the most grossly polluted rivers in Britain - the River Cole, a tributary of the River Tame, (Midlands). The severity of the pollution had been reduced by the early 1960's but it wasn't until 1971 and 2 that the remaining sources of contamination were eliminated. Armed with precursory information, an attempt has been made to ascertain the effect of the abated effluents on the recovery stages of the benthic invertebrate communities.

Hence the content of this thesis is essentially divided into two sections; one of eutrophication studies and the other of organic enrichment and attending recovery. Although the two sections overlap ecologically, they do not practically and methodologically. For this reason it was considered appropriate to write an account of the literature pertaining to both aspects combined into one chapter and the results and discussions of the investigations into separate chapters.

CHAPTER 2.

LITERATURE REVIEW.

2.1 CHEMICAL COMPOSITION OF NATURAL, EUTROPHIC, AND ORGANICALLY ENRICHED WATERS.

a) Nitrogen and Phosphorus.

Before the effects of eutrophication on benthic invertebrates may be described, the factors pertaining to the environment of which they are both a part, and by which they are affected, must be put in perspective.

Nitrogen and phosphorus in streams occur in small amounts naturally; rainfall, land drainage, run off and recycling from plants being the major contributary sources. They vary in concentration with the geography, topography, geology of the riparian land and with meteorological conditions. American Water Works Association figures, quoted by Lee (1970), for the usual concentration of nitrogen in rainfall, vary between 0.1 and 2.0 $mg.l^{-1}$ during discharge. Voight (1960) quotes rainstorm figures in Connecticut for nitrogen as between 0.05 and 0.07 $mg.l^{-1}$; and average phosphorus as 0.01 $mg.l^{-1}$. Variance in nitrogen and phosphorus from run off and drainage of land is, however, much greater. These amounts are functions of the type of land being drained; both geologically (e.g. phosphorus bearing rocks) and agriculturally. Water percolates very efficiently through chalk strata, the rate and subsequent concentrations being dependent on the volume of water and slope of the land, the greater the volume and slope the more nutrients are leached out. Mackenthun _et_ _al_ (1964) have demonstrated the slope effect:

Slope	N.Kg/ha/pa	P.Kg/ha/pa
20^{o}	34	1.6
8^{o}	16	0.45

(figures recalculated by Hynes, 1970).

The magnitude of nitrogen and phosphorus from agricultural run off obviously fluctuate with the amounts of fertiliser applied. Application commonly averages out at 125 Kg/ha/pa N. for cereals with variations of 50 Kg/ha to 500 Kg/ha per annum depending on localities (Green and Walker, 1969). According to the American Water Works Association (Lee, 1970), concentrations of nitrogen in run off, shown below are usual.

Rural Run Off	Usual concentration in discharge mg. $N.1^{-1}$
Agricultural land	1 - 70
Non-agricultural land	0.1 - 0.5
Urban Run Off	1 - 10

Phosphorus is present in streams as ionic orthophosphate and phosphate in both soluble and bound forms (Hynes, 1970) and eutrophication in terms of phosphorus is therefore measured as total phosphate or total inorganic phosphorus. Neel (1951), has demonstrated fluctuations of phosphate, coincident with changes in flow rates, between 0 - 8 mg. 1^{-1}. In this case the likely source was calcium diphosphate rock in upper layers of the soil which, although virtually insoluble in water, was eroded by carbonic acid following rainstorms. Phosphates, especially in ionic form, are quickly removed from the medium by plants (Hutchinson, 1957), and Neel (1951) has reported major consumptions of nutrient minerals occurring on riffles, so absolute figures are difficult to obtain, which elicits the conclusion that phosphorus is not the ideal measure of eutrophication. Nitrates, which is the fully oxidised form of nitrogen in rivers, is also taken up quickly by plants but nitrite and ammoniacal nitrogen can be utilised. In Sweden and other colder countries nitrous and ammoniacal nitrogen sources are common, as nitrification of ammonium compounds in the soil and later in the water is inhibited in winter because of the low temperatures (Brink and Widell, 1967).

In most cases though, the presence of ammonia, nitrite or a high level of nitrate is indicative of industrial or domestic pollution. Lee (1970), shows estimates of usual nitrogen concentration for domestic wastes as being between 18 and 20 mg. l^{-1} and from industrial wastes, anything from 1 - 10,000 mg. l^{-1}. These point sources of nitrogen and phosphorus are major contributors to the nitrogen and phosphorus balance in rivers; 'diffuse' sources only causing minor elevations in levels. Typical examples of the amounts by which sewage effluents can raise the nutrient concentrations in lotic waters are given by Pitcairn and Hawkes (1973): -

Table showing nutrient concentrations upstream and downstream of sewage effluents.

British Rivers	Annual $\bar{X}$ T.I.P. mg.l^{-1} P.	Annual $\bar{X}$ NO_3 mg.l^{-1} N.	Annual $\bar{X}$ NH_3 mg.l^{-1} N.
R. Arrow			
upstream	1.1	2.7	0.6
downstream	2.1	5.2	0.4
R. Cole			
upstream	0.7	4.9	0.4
downstream	1.8	4.3	2.6
R. Blythe			
upstream	1.5	4.3	0.9
downstream	2.9	6.7	0.9
R. Ray			
upstream	2.4	7.2	0.9
downstream	4.6	12.5	2.8
R. Darent *	1.0	4.8	0.05
R. Blithe *	0.8	3.5	0.4

* sewage free rivers.

It is impossible to select a quantity of nitrogen or phosphorus in a watercourse above which point the term eutrophicated may be applied. This of course is because of the numerous other parameters governing the rates of growth, each species very likely having specific requirements. However large growths of the green filamentous alga Cladophora are said to be particularly indicative of eutrophication, and in general, river water containing less than 1.0 $mg.l^{-1}$ total inorganic phosphorus produces only modest growths of it (Pitcairn and Hawkes, 1973). However, they could not correlate significantly the mean annual dry weight of Cladophora with nitrogen concentration ($r = 0.22$). That nitrogen can equally well be a minimum factor in autotrophic growth is apparent from an investigation of Brink (1965), who shows that the ratio N:P between directly consumable nitrogen and phosphorus compounds amounted in summer to 1:1 and less, against the normal 10:1 in the aquatic vegetation.

b) Dissolved Gases.

In natural running water the dissolved gases are approximately in equilibrium with the atmosphere. The nitrogen constituent remains relatively consistent (saturation) and the oxygen and carbon dioxide parts which vary inversely, will be more or less near saturation point depending upon the gradient and temperature of the stream. Mountain and turbulent streams will have oxygen saturations near 100% while lowland, and streams of more laminar flow tend to have greater variation. Owens et al (1964), have shown that the diffusion of oxygen through the surface of rivers is principally related to the water depth and velocity; so that rapid, shallow and turbulent streams will tend to be more oxygenated and display little variation compared with the deeper sluggish reaches. The solubility of gases varies inversely with temperature so that diurnal and seasonal fluctuation of a small nature would be expected, especially in shallow waters which are subject to short term changes in their temperature (Eckel, 1953). The diurnal variation of oxygen is usually only large when polluting sources or large amounts of algae or macrophytes are involved, but exceptions have been observed by Owens and Edwards (1964) in the River Yare showing differences of 10 $mg.l^{-1}$ O_2 between maxima and minima. Seasonal variation is to be expected with respect to light, temperature and growth. Diminution of oxygen tensions may be noticed in autumn (Hynes, 1966, 1970), as a result of leaf fall and decomposition. Conversely, the free carbon dioxide present in the open stream may increase or appear for the first time in the year during this period. Neel (1951) reported a bicarbonate maximum in a limestone stream during autumn as a result of the free carbon dioxide/pH drop.

The major cause of oxygen/carbon dioxide variation is that of pollution. The direct effect of organic enrichment is one of de-oxygenation, but secondary effects such as algal growth can amplify the oxygen status. (The latter effect is discussed in the next section). Many authors have reported the direct effects of organic pollution on oxygen balance: - Butcher et al (1927, 1937) in the River Lark and River Tees, Hawkes (1956, 1963a), Hawkes and Davies (1970) in some Midland rivers, demonstrate particularly severe cases : -

R. Lark. (just below effluent of sugar beet factory)
November. 8% saturation with oxygen.

R. Cole. (Midlands) 1 ppm O_2 over 24 hours with
temperature varying between 15° and 21°C.
(organic sewage effluent).

c) Suspended Solids.

Flowing waters always carry suspended solids with peaks occurring during 'spates' and 'troughs' during dry weather. The solids are usually inert, settling out gradually as a function of particle size, current velocity, temperature and thus viscosity of the water. Turbidity can reduce the penetration of light and therefore affect photosynthesis.

Solids in effluents are usually of two sorts; inert (mainly industrial sources) and organic(mainly domestic some industrial). Inert solids settle out as described above and form a layer on the substratum whereas organic solids are usually oxidisible and, although this is a slow process, the result is inevitably one of de-oxygenation.

The colloidal particles can stay in suspension much longer and act as stores of nutrients, heavy metals, and other toxic substances (Patrick, 1968). When these colloids settle out, they often form a soft flocculent mud in which bacteria flourish, and as a result, anaerobic conditions develop in the river bed. The relationship between oxygen consumption and other properties of settled solids and muds have been discussed by Edwards and Rolley (1965), and Rolley and Owens (1967).

d) Poisons.

Seepages of toxic materials have been occurring since the ice age and are not just the result of recent industrialisation and smelting. Habitats containing sulphides, heavy metals, high chlorides and high pH occur naturally, and one would expect that since the ice age, a number of tolerent species might have evolved. It is also unnecessary to think of the evolutionary time scale in millions of years because high selection coefficients are quite common: e.g., industrial melanism in moths, insect tolerance to insecticides - in less than two years house-flies and mosquitoes have become tolerant to D.D.T. However, the naturally occurring poisons are restricted in type and nature. Leakages (in small amounts) of Cu, Pb, Zn, Ni, Al, Fe, as a result of water passing through their ores would be expected, as well as anions like hydroxides and chlorides. The inorganic poisons found in sewage and industrial effluents are far greater in number and the pesticides, phenols, formaldehydes and other organic poisons, are of course contemporary problems.

The majority of literature available is concerned with the toxicity of substances to fish. Classical studies on fish toxicology are given by Doudoroff and Katz (1953) and Jones (1962).

2.2 SECONDARY EFFECTS OF EUTROPHICATION AND ORGANIC ENRICHMENT ON WATER CHEMISTRY.

Organic enrichment, together with increases in inorganic mineral levels that accompany sewage effluent discharges in rivers, will encourage growths of bacteria, fungi, protozoa and algae. These growths will cause indirect changes in the water chemistry, as opposed to the direct chemical modifications imposed by the effluent and the effect of these growths are particularly relevant to the oxygen/carbon dioxide/ph balance of the recipient stream.

The types and amounts of growth will be dependent upon the nature of the effluent. Organic degredation is accomplished primarily by bacteria, which occur in small numbers naturally in the river and in large numbers in the treatment plants. These saprophytes multiply in the river with organic enrichment often producing the 'sewage fungus' complex of bacteria, protozoa, fungi and trapped organic solids. Their growth is usually at its peak just below the outfall but may be more spread out if inhibitors, (e.g. heavy metals) are present in the effluent, (Southgate, 1969). Anaerobic bacteria which operate in the enriched mud were noted by Hawkes (1962) to evolve gas, an analysis of which was executed by Edwards(1957) showing high concentrations of methane: - 74.7% CH_4 by vol, 7.4 CO_2, 17.7 N_2, 0.2 O_2.

One physical effect noted by Edwards (1964) was the lifting of abandoned chironomid tubes by this gas production (spring temperature aiding the release of gas) releasing vast quantities of organic material into the river. Hawkes (1963a) recorded vernal sloughs of 'sewage fungus' in polluted streams.

Respiration of the aerobic bacteria causes the initial drop in the 'oxygen sag' curve; moreover the oxygen demand imposed during their decomposition makes itself felt further downstream reducing the recovery of the 'oxygen sag' curve.

Some physical effects were noted by Butcher _et al_ (1931). He submerged clean slides below the effluent of a sugar beet factory in the River Lark producing growths of _Sphaerotilus_ of 1 cm. in length after four days. It was at this length he noticed its ability to trap silt. Areas with growths of sewage fungus can trap vast amounts of silt blocking up the interstices between stones; (the biological significance of which will be discussed later) cause turbidity and reduce light penetration.

If, as a result, the de-oxygenation and reduction of light is severe, it will prevent the accumulation of autotrophic growths till some way downstream. With mild organic pollution, however, the growth of algae may be considerable just below the outfall. The chemical parameters of the water will progressively change downstream, firstly with the diminishing bacteria and secondly with algal peaks (controlled by nutrients, physical factors, grazing by animals, etc.).

Rooted plants may reduce the velocity and consequently increase the depth and rate of sedimentation. A reduction in velocity will also reduce the rate of re-aeration.

The pullulation of algae and macrophytes downstream with more suitable conditions, significantly enhances the oxygen resources of the river. Wide diurnal fluctuations of dissolved oxygen caused by them have been reported, and changes in pH and free carbon dioxide associated with it have been blamed for fish deaths (Westlake, 1959). Butcher _et al_ (1927) realised the limitations of spot samples and were the first to record dissolved oxygen (temperature, pH and ammonia) over 24 hours. They recorded not only hourly differences but differences due to light and algal intensity in the River Lark.

3 pm.	March 30th	Max.	157% sat. with O_2 (diatoms spring max growth)
5 pm.	April 27th	Max.	178% sat. with O_2
1 - 7 am.	March 30th	Min.	65% " "
4 - 7 am.	April 27th	Min.	57% " "

They suggested the high maxima were due to increased growths of algae as a result of past pollution. Their figures for the unpolluted River Itchen with less spring diatoms at the same period showed less diurnal variation.

2 pm.	April 17th.	Max.	112% sat. with O_2
5 am.	April 6th	Min.	80% " "

As the River Itchen was shallower and more rapid than the River Lark, the minimum and maximum oxygen content were expected to be nearer saturation.

The variations in more sluggish rivers may be even greater. Schroepfer (1942) has shown that variations from oxygen depletion to super-saturations can occur at any point during the day. He gives data on hourly, daily, cross-sectional, depth and tidal variation in level.

Diurnal min. and max. O_2 in mg.l^{-1}. Taken in summer.

		Mississippi River.	White River.
		Annual mean = 1.37	Annual mean = 3.13
Summer	7 am.	1.23	0.94
	3 pm.	4.93	13.77

Variation with depth. O_2 in mg.l^{-1}. Mississippi River. Sunny, between 1.40 and 2.10 pm.

Depth.	O_2
1'	12.55
3'	8.15
7'	1.65

The Mississippi River was organically polluted at the site of these determinations and shows that diurnal variations are caused by the planktonic algae, light not being able to penetrate through the turbidity to much effect past 3 feet. (However, Schroepfer does not mention the pollutional state of the river mud, and as Edwards (1968), Rolley and Owens (1967), have pointed out, the respiration of river muds can be high and may account for the low 7 ft. oxygen tension). Edwards (1968), shows the supply and demand of oxygen sources that influence the oxygen economy of the unpolluted River Ivel.

	Supply	Utilisation $(g/m^2/day)$
Mud	-	2.4
Rooted plants	7 - 16	5 - 12
Plankton	0.1	0.2
River surface	5	-

The respiration of polluted muds are usually much higher and in exceptional circumstances may be as high as $20g/m^2/day$ (Rolley and Owens, 1967).

The photosynthetic activities of large algal growths, together with high temperature and turbulent flow, will deplete the amount of free carbon dioxide and eventually the equilibrium carbon dioxide. This is usually coincident with a pH of approximately 8.3, the attainment of which precipitates $CaCO_3$, as the equilibrium below is pushed to the right.

$$ca(HCO_3)_2 \rightleftharpoons CaCO_3 + H_2O + CO_2$$

This deposition is a common feature of rivers with limestone origins, but more often seen in lentic waters.

Heterotrophic respiration and subsequent oxygen removal is usually small in degree but this is increased with organic enrichment (Edwards and Rolley, 1965). Those likely to cause the greatest effect are large populations of tubificids, chironomids and Asellus. Westlake (1959) reported the results of Meschkat who found tubificid populations of up to 1.7 million per m^2 in the River Elbe. This sized population was estimated as being capable of moving 50 tons of mud/acre/day. Release of organic solids accompanying this movement must inevitably make large demands on the river's oxygen resources. Edwards (1957) found up to 100,000 chironomids per m^2. in winter, spring and early summer in an effluent channel and estimated (1958a) that this sized population could alter the oxygen concentration of a stream by 1.5 ppm O_2 in one mile. Edwards and Learner (1960) showed the oxygen consumption of Asellus aquaticus to be the same as Chironomus plumosus (Edwards, 1958b) and therefore large populations of this isopod as often found in organically enriched situations, may have similar demands also.

Secondary effects, which are mostly shown as modifications to the oxygen resources of the river, are the type of manifestations of eutrophic and organic enrichment that lead authors like Fruh *et al* (1966) to believe that the best parameter for the measurement of eutrophication is that of diurnal variation in dissolved oxygen.

2.3. FACTORS INFLUENCING THE DISTRIBUTION OF BENTHIC INVERTEBRATES.

a) Current Velocity.

To understand the importance attached to current velocity in connection with distribution of benthic invertebrates it is necessary to realise that the effects are the results of a multiplicity of factors. Some species (rheostenic) are dependent upon the current for their respiratory requirements, being unable to obtain the necessary amount of oxygen when the current is slack. This aspect is dealt with in the chapter on respiratory studies and oxygen.

Other effects in the reticulation are ones of food, temperature and pollution. By far the most important factor related to current velocity is that it determines the types of substrata, which is intrinsically an important and inseparable feature of autecological distribution, but for practical reasons will be discussed more fully in the next section. The relationship as displayed by Butcher (1933) is however useful at this juncture as he has characterised the type of habitat together with current velocity and nature of the bed.

	Velocity ft/sec	cm/sec	Nature of bed.	Type of habitat
More than	4	121	rock	torrential
"	3	91	heavy shingle	"
"	2	60	light shingle	non-silted
"	1	30	gravel	partly silted
"	0.67	20	sand	" "
"	0.42	12	silt	silted
Less than	0.42	12	mud	pond like

Nielsen (1950) presented a list showing the relationship between stream velocity and the size of the mineral particles which the current could support but these lists are only of use as background value. (For instance, current is inconsistent.) During spates the rate of flow may be enlarged dramatically especially in 'flashy' rivers whose level may fluctuate rapidly even with the slightest rainstorm. Phillipson (1956) has shown how Simulium larvae can re-orientate their position on the substratum as the current in the river fluctuates. Drought, the other extreme, may reduce the current to such an extent that respiratory demands cannot be met by animals like Nemoura, or, some relatively immobile animals may actually become exposed. The effects of drought and spates on bethic invertebrates have been reviewed by Hynes (1970). Ideally the river flow should be constantly monitored but authors very infrequently quote averages when suggesting possible distributions in relation to current.

To demonstrate the relationship between current and individuals or biocoenoses, one is confronted with two problems. Firstly, since species react very differently to the current, some of them digging into the silt, crawling under or between stones, and others exposing themselves to its full force, data that is presented in the form of % occurrence against velocity in a stream or an area of stream bed is of little help. Secondly, it is extremely difficult if not impossible to determine the actual current to which they are exposed, so any numerical value attached to a species to designate its velocity preference must be treated with care.

The considerable reduction of current speed in the few millimetres above the surface of an object was demonstrated by Ambuhl (1959) using a flashing light and acetyl-cellulose particles. There was also a stagnant piece of water downstream of the object. The slack layer immediately above the smooth object and the dead water described are both distinct micro-biotopes.

Nevertheless, measurements made by various authors are useful in relating fauna and distribution. The derivation of these parameters are conducted either by taking measurements in the field of flow and benthic populations, or rheophilic animals are tested for their resistance, net building capacity (Philipson, 1954) or similar accomplishment, in experimental channels of a different gradient and the results related to their distribution in nature. Ambuhl (1959) recorded the following rheophilic conditions, having plotted flow (measured just above the bottom) against % catch at various speeds. Simulium maximum at 80 cm/sec, Hydropsyche augustipennis 60 cm/sec, Gammarus pulex 15 cm/sec, Baetis vernus 40 cm/sec, and Ephemerella ignita between 10 and 25 cm/sec. Scott (1958) plotted the number of larvae per square metre against the surface velocity, the peaks of which gave Hydropsyche fulvipes maximum numbers between 40 and 50 cm/sec, Rhyacophila dorsalis between 80 and 90 cm/sec and Stenophylax stellatus between 0 and 10 cm/sec. Dorier and Vaillant (1954) used a combination of the two methods for deriving their parameters. They measured the maximum and minimum speeds in which they found certain species, followed by measurements of the same species to maximum speed at which they would ascend, and maximum speed at which they were washed away. If the speed limits found in nature are plotted against the speed at which they lose grip there is an obvious relationship, with the exception of G. pulex whose maximum wash out speed is lower with respect to that found in nature, but may be related to the fact that it has no special appendages or adaptations bar its flattening.

A. priori, the decisive influence of speed upon zoocoenoses is realised and the emergence of rheostenic species like Baetis, Simulium, Hydropsyche, Rhyacophila and species finding their optimum at relatively low velocities (but not necessarily rheoeuryic), e.g. G. pulex, and Ephemerella ignita is clear also.

Finally, Jaag and Ambuhl (1963) mention some interesting observations pertaining to the effect of pollution. Spring water was pumped through three experimental channels in which all conditions were equal apart from the gradients. At the end of each year, for four years, the water quality was deteriorated artifically by the addition of waste water. The resulting biocoenoses in the channels were differentiated and in the case of half the species, the current exercised a stronger ecological effect than could be ascribed to the loading with polluted water. This effect draws the conclusion that the use of some biological indicator systems may be impaired. According to Zimmermann (1961, 1962) a high current velocity favours those organisms which have lower saprobic values whereas a low velocity favours biocoenoses which are higher up the saprobien scale. This aspect is discussed again under 'The Interpretation of Biological Data'.

b) Substratum. It has been shown that algae characteristically grow on certain substrata (Butcher, 1933), these substrata inevitably being associated with the current regime. Similarly it is also known that benthic invertebrates are distributed according to the type of substratum, some authors asserting that it is the major controlling factor, (Moon, 1939; Hynes, 1960; Macan, 1963). Cummins (1962) advocates the measurements of the physical constitution of the sediment as an integral part of all benthic investigations, because substrate particle size, as he instructs us, can serve as a common denominator in benthic ecology. He tabulated eleven categories ranging from boulders to clay with equivalent particle and sieve sizes as guide lines for adoption. However, this tabulation takes no account of the nature of the objects, i.e. whether the stones are regular or irregular. It is known that irregular stones offer more shelter and hence harbour higher densities than regular ones.

Other investigators have attempted to analyse the substrate conditions with regard to their effects upon the organisms as indicated by the type and density of the population. The classical papers by Percival and Whitehead (1929, 1930) have shown these relationships. They classified types of stream bed (1929) as a result of their work on West Riding (Yorks) streams, finding that well defined groups were evident. The groups agreed with the following divisions:

1) Loose rounded stones of more than 5 cm. diameter, forming an unstable substratum, no visible vegetation, approximately 4 cm/sec current and an assortment of rounded pebbles of diameter down to 5 mm.

2) As above plus diatomaceous growths, and stones to 9.5 mm. diameter. Stable.

3) Mixed clean stones 2.5 cm. - 0.5 mm. Unstable.

4) Rounded or flattened stones 30 cm. - 2.5 cm. with growth of Cladophora. Stable.

5) Large stones with growths of moss but little consequent accumulation of detritus.

6) Thick carpets of moss and accumulation of fine detritus.

7) As 4) but with Potamogeton perfoliatus growths.

These examples are obviously characteristic of clean eroding reaches with the possible exception of 6) and 7), also common in minnow reaches. Their faunal lists suggest, in general, that Ephemeroptera are most abundant in the loose stones - 33.2%, (the average % compositions of associated fauna) as are Plecoptera - 5.14%. Trichoptera favour stable substrata, 39.95%, as do gastropods 14.9%, on cemented stones, while Diptera preferred loose moss and thick moss on stones: - 65.34% and 42.0% respectively. Naids form a large proportion of fauna in thick moss, 26.4%, and in Cladophora 12.5%, however tubificids were at their greatest percentage in Potamogeton on stones - 14.66%.

Moon (1939) pointed out how the substratum changes according to the evolutionary stage of the river, young rivers being primarily 'eroding' becoming more depositing with age. Eroding substrata, rather like 1) to 5) of Percival and Whitehead's above classification, offer mechanical support to benthic organisms, shelter from turbulent water and predators, and in particular connection with endopterygote insects, a quiet place to pupate. The major food source of these steeper gradients is algae and therefore the fauna is dependent on the algal food chain. With lesser gradient the erosion is not so great and deposition of silt commences. This means the fauna is emancipated from algal dominated food chains as the space for algal growth is diminishing. Still lower downstream the siltation is way in excess of erosion with silt loving forms, worms, Sialis, Ephemera danica, chironomids and detrital feeders like Asellus becoming more numerous. Depositing substrata have therefore less variety of food to offer, this being accompanied by a consequent reduction in diversity of fauna. Moon (1939) quotes Tyrrells Ford, where a section, within a few yards, passes from a clear stony chalk stream to a turbid stagnant lagoon of mud. The density of insects per square foot in the stony reach was between 111 and 225 containing 12 and 11 species respectively, and in the lagoon section 1000 in number and 7 species, most of which were burrowing chironomids. Eggilshaw (1964) took note of the amount of detritus during benthic sampling and found the relationship of increasing numbers of animals with increasing detritus, but of course this relationship does not apply to carnivores of filter feeders, and confuses species diversity.

As a result of all these factors, the fauna of clean stony runs is richer than that of silted reaches and pools both in number and total biomass. Hynes (1970) has reviewed the literature pertaining to distribution with substratum and concludes that, in general, the more complex the substratum, the more diverse is the fauna.

It will be appreciated that enrichment with organic matter or nutrients will modify the biotopes physically. The organic solids will deposit between the stones, and smothering eggs and diatomaceous growths. (Hawkes, 1962, has estimated the organic solids from a sewage works at about 10 tons/dry wt./year). Animals relying on bare stones for adherence, *Ancylus*, *Lymnaea* and leeches will lose their habitat. Increases of *Cladophora*, *Stigeoclonium*, *Vaucheria* and bryophytes will become evident with fertilisation, trapping more silt and thus altering the substratum drastically, to the exclusion of a number of species. The effects of mild organic pollution can thus be likened to those of depositing substrata, and those classifications of Percival and Whitehead (1929) - thick moss, loose moss and *Cladophora*, much amplified.

c) Temperature.

The life histories of benthic invertebrates are largely determined by the temperature of the water, their growth rate being a function of it. The natural temperature gradients of a stream may influence the distribution of fauna so that one finds in headstreams stenothermic species like the much quoted *Crenobia alpina*. The actual temperature will depend upon the distance from the source, the gradient, volume of water, solar radiation and turbulence. Warm discharges have substantial ecological consequences on streams. The increased winter temperatures affect the life cycles directly by causing species to develop and emerge too soon, and consequently finding themselves without food or mates. Naylor (1958, 1965) has described the appearance of exotic tropical species as a result of power station effluents discharged into dock and estuaries, and likewise Mann (1958) reported the appearance of Branchiura sowerbyi in the Thames below a power station outlet.

The indirect effects of temperature are perhaps more important, especially in an already polluted river. The tolerance to various poisons is lowered, including the resistance to low oxygen. The B.O.D. is increased effectively, as the time for satiation of oxygen demand is reduced. The synthesis of effects of elevated metabolism, reduced solubility of oxygen, and local de-oxygenation due to increased bacterial activity can have lethal effects on fish and invertebrates.

However, the temperature increments encountered in the present investigations are very small (approx. $\pm$ 0.1°C) and for more detailed accounts the reader is referred to Langford (1970, 1971), Cairns (1956, 1958, 1969, 1970, 1971), Horvath and Brent (1972) and reviews by Macan (1963), Hynes (1966, 1970) and Hawkes (1969).

d) Chemical nature of the water.

Most authors agree with Moon (1939), that chemical and physical factors of water and distribution of species is only apparent in a general way, although Jaag and Ambuhl (1963) point out that the chemistry exerts a great influence on which organisms have a chance of occurring at all. This is particularly relevant to pollution chemistry where large variations in ions and organic matter are involved.

One ion that does vary naturally and has been shown to limit the range of a number of species (and whole taxa) is calcium. Macan (1963) has described a range of below 1 mg/l in soft water to over 100 mg/l in hard water. Moon found no evidence of chemical or physical factors being responsible for the distributions of A. aquaticus and A. meridianus in Windermere (1957a) or for the absence of Asellus from eleven out of fifteen other Lake District waters (1957b). He expected to find a correlation with Ca^{++} concentration but finding none, suggested the distribution was a mixture of geological and human factors. On the contrary, Reynoldson (1961a) after studying 65 lakes in northern Britain (although Asellus was not the central focus of the study) concluded that there was a relationship between Asellus and Ca^{++}, recognising occurrence within three ranges. In waters containing less than 5ppm Ca, Asellus was absent 22 out of 24 cases. Between 7.0 ppm and 12.5 ppm Ca there was an equal chance of occurrence (5 out of 11 cases), while in water containing more than 12.5 ppm

Asellus was present in 26 out of 30 cases. He also found a correlation between total dissolved matter and the Asellus distribution, the significance of which may be an elevation of the Ca^{++} threshold in situations where the total dissolved matter is high, (Tucker, 1958). This is of obvious importance with respect to organic enrichment.

Hynes (1954) was unable to show that Ca^{++} was a limiting factor in the distribution of G. pulex and G. duebeni in the Isle of Man, a relationship that some authors (Wundsch, 1922; Titcomb, 1927) have thought restricting. Sutcliffe and Carrick (1973) have also made observations on the distribution of Gammarus in connection with pH and Ca^{++}.

The freshwater Mollusca were studied by Boycott (1936) who, after many years observations, and from imperfect data available, concluded that a number of Mollusca do not live in water with less than about 20 mgl^{-1} Ca^{++}.

Hirudinean distribution showed that in hard water ($>$ 24 mgl^{-1} Ca) H. stagnalis was the most common of 12 species (Mann, 1955 a,b). Hemiclepsis marginata was not taken in soft water ($<$ 7 mgl^{-1} Ca) and Erpobdella octoculata was most abundant amongst the species taken in it. After a survey of 58 pieces of water during which the above observations were made, Mann concluded that in addition to hardness, a large volume of water appeared to be favourable also.

Reynoldson (1958 a,b) demonstrated for triclads the existence of characteristic Ca^{++} ranges and increased numbers of species with increasing eutrophication (hence increased Ca^{++} levels).

Unproductive Productive
0.4 2.5 12.5 63.1 (log)
Phagocata vitta
Polycelis ---------------------- nigra
4.0 Polycelis hepta ----------
4.0 Polycelis --------- tenuis
10.0 Dugesia lugubris
Dendrocoelum lacteum

C. alpina and *P. felina* were found to occupy a complete range of Ca^{++} from 2.0 - 120 ppm (Wright, 1974) and in agreement with Reynoldson shown in the confinement of *Ph. vitta* to streams of low Ca^{++} levels.

It appears therefore, that some animals are limited in range by their dependence on Ca^{++} and in general, numbers of species increase with increasing hardness. Hardness is also a factor in pollution ecology; calcium being a well established synergist and known to influence osmo-regulation and respiratory rates (Hynes, 1970).

2.4 THE EFFECTS OF ORGANIC AND NUTRIENT ENRICHMENT ON BENTHIC INVERTEBRATES.

Under the title of "Factors Affecting Benthic Invertebrates" above were discussed direct and indirect factors that may or may not be responsible for the distribution and range of benthic invertebrates. It was pointed out how all these factors; dissolved, suspended solids, temperature, etc., and their balance can be modified by the effects of pollution. Thus there is no way of allocating cause with much precision in such a complex situation. It is rarely possible to distinguish between certain types of organic pollution biologically, but, in general, there are notable similarities in the effects of organic enrichment. Such resemblances were realised by early workers like Kolkwitz and Marsson (1908, 1909), and ever since, enterprising authors have been modifying it or developing new biological yardsticks of pollution. A discussion of methods of interpretation of biological data and its measurement in connection with water quality follows later.

The biological effects of organic and nutrient enrichment will be outlined together, for nearly all organic contamination carries high levels of nutrients, and the differences between the two effects may be just a question of degree and time. After the bacterial activity has reduced the amount of solids, and the dissolved oxygen has risen enough to permit the growth of increasingly sensitive biota, the release of nutrients can now cause conditions of eutrophication.

A typical biological succession following the introduction of heavy organic effluent has been described by Butcher (1955) as one dominated by tubificids and chironomids in the foulest zone just below the outfall, gradually being succeeded with distance by Asellus, leeches and molluscs until a fairly repurified zone is reached consisting of Gammarus, Trichoptera and ephemerids. He then shows how discharges of gas liquor and sewage into the R. Trent displays successions similar to those of strong domestic pollution: - all organisms become scarce near the source, due to the poisoning, then, lower downstream a few tubificids, chironomids and Sphaerotilus appear, increasing in density with more dilution and purification until they are very abundant. Then, Asellus and Stigeoclonium tenue become competitive giving way further downstream to communities containing molluscs, leeches and Nitzchia palea and finally Gammarus with Hydropsyche.

Corresponding successions have been noted in other geographical regions although the very same species may not have been concerned. In the Krom Stream (South Africa) at Stellenbosch, (Harrison, 1958) organic pollution from storm water, irrigation furrows and distillery waste discharges, increased the oligochaete and chironomid abundance and decreased the ephemerid populations. An extract from his work shows the % composition of the fauna as the pollution eliminates the more tolerant species.

TABLE SHOWING THE EFFECTS OF THE WASTES, WHICH INCREASE IN SEVERITY FROM OCTOBER ONWARDS, ON THE % COMPOSITION AND TOTAL DENSITIES OF THE MAJOR FAUNA.

(from Harrison, 1958)

	Oct.	Nov.	Dec.	Jan.	Feb.	Mar.	Apr.
Baetis harrisoni	30	10	12	15	-	-	-
Chironomids	10	15	25	20	-	-	-
Naids	2	60	60	10	-	-	-
Lunbriculus	32	-	-	10	-	-	-
Lumbricidae	22	-	2	15	15	40	3
Tubificids	5	-	-	-	8	15	75
Total density	-	48,800	11,700	6,700	8,350	8,050	3,200

Finally Psychoda alterna and C. plumosus became established and remained virtually unaffected. Quantitative work like this is able to demonstrate the effect on the density of individuals as well as the diversity of species. The decrease in total density at Stellenbosch as the pollution increases from October is clearly exhibited. Harrison's work also reveals the characteristic decrease in diversity but this is shown to more effect in the Middle Illinois River as a result of Richardson's investigations (1921, 1929) when, between 1915 and 1920, the numbers of species he found dropped from 91 to 15 with the incidence of sewage pollution. In one section there was a reduction in the sensitive Sphaeridae from 1,709 to 46 per $yard^2$ and in Gastropods (total) 496 to 20 per $yard^2$, while the tolerant chironomids and tubificids increased in average abundance from 3 to 587, and 1.4 to 46 per $yard^2$ respectively. Other declines were noticed in the Hydropsychidae, leeches, Odonata, planarians, amphipods and ephemerids.

So, briefly recapitulating, explosions in numbers of tolerant species occurs together with reduction in numbers of more sensitive species (sometimes called Thienemann's second biocoenotical rule) - shown graphically by Bartsch (1948). There are only a relatively small number of tolerant species so the diversity is reduced also.

With subsequent dilution, re-aeration and other autecological factors changing downstream, the less sensitive species are able to compete for food and space with the dominant foul forms, so the picture of diminishing abundance in numbers of the tubificids, chironomids with increase in Asellus and leeches is realised.

TABLE SHOWING ANNUAL MEANS OF SOME SPECIES PER 0.1m^2 AND 5 BELOW AN ORGANIC SOURCE OF POLLUTION. Hawkes and Davies (1970).

	Above effluent	Below effluent					
	1	2	3	4	5	6	Stations downstream →
Tubificidae	162	407	879	1049	1159	468	
Enchytraeidae	4	48	515	117	17	4	
Lumbriculidae	73	12	10	41	9	9	
Baetis	131	2	6	6	3	2.5	
Asellus	8	0.4	0.6	3	182	46	
C.riparius	0	104	331	187	7	4	
C. bicinctus	6	0.2	7	51	421	165	

This pattern occurs in successions as more species are able to compete. This means that species show, at some point, a peak in abundance which diminishes as another species reaches its peak. These peaks can be seen in the above data for tubificids, Asellus and C. bicinctus at station 5, with C. riparius and enchytraeids at station 3.

When de-oxygenation is severe as a result of very heavy organic pollution discharges, only those species physiologically adapted to surviving in near anoxic conditions or those that can breathe air directly are found. Gaufin and Tarzwell (1952, 1956) related the presence of air breathing Coleoptera, Diptera (*Culex* and *Eristalis*) and Mollusca (*Physa integra*) in what he called the 'septic zone' (having less than 1 mg.1^{-1} D.O.) in Lytle Creek. There were 8 species in this zone altogether, the % constitution being; Diptera 40%, Coleoptera 20%, Annelids 20%, Hemiptera 10% and Mollusca 10%. Further downstream in the 'recovery zone' they characteristically found more species but less numbers, with Trichoptera and *Asellus* absent for 6 miles.

Aston (1973) reviewed the occurrence of tubificids with water quality noting that *Limnodrilus hoffmeisteri* and *Tubifex tubifex* predominated over other species which were eliminated in accordance with the degree of pollution. Learner *et al* (1971) found the principal effects of organic enrichment in the R. Cynon were increased proportions of *L. hoffmeisteri* to the other oligochaetes, increasing representations of tubificids and the enchytraeid *Lumbricillus rivalis* and a decline in chironomid numbers and diversity.

It is evident, then, that not only are species restricted in distribution, but whole taxa may be, especially if the pollution is severe. When de-oxygenation is less severe, in accordance with self-purification or only mild pollution, the freshwater shrimp *G. pulex* often appears, and may do so in conjunction with *A. aquaticus* however they seem to do so in a ratio that suggests they are mutually exclusive. The fact that they are both detrital feeders may be an explanation. Berg (1948) records the occurrence of both in the R. Susaa above and below a polluted reach.

	Asellus	Gammarus
Susaa at Teestrup Bro. (clean)	rare	common
pollution ——		
Susaa at Mollebro (highly polluted)	rather common	absent
" at Nymollebro (highly polluted)	common	"
" at Hjelmsolille (apparently clean)	v. common	v. rare
" at Vetterslef (apparently clean)	"	absent

Although Berg describes the mud as being black and stinking of H_2S at Mollebro and Nymollebro, the condition of 'highly polluted' would perhaps be an overstatement compared with those preceeding examples, however it illustrates the principle of the Asellus/Gammarus relationship.

The effects of mild organic pollution often display the characteristics of natural eutrophication. There is an increase in productivity of algae which in turn raises the size of the primary consumer population and consequently the secondary consumers and so on. However, there are still factors affecting the nature of the substratum, and the effect of slight silting and encrustacions on stones may suppress the number of lithophilic and thigmotactic species. Butcher *et al* (1937) reported on the invertebrate fauna below a well treated effluent on the R. Tees. Some 300 yds. below, the mayfly populations were still represented, *Baetis* and *Ephemerella* being virtually unaffected and the more sensitive *Ecdyonurus* and *Rithrogena* showing reductions between one third and one eighth. Only *Leuctra* among the sensitive stoneflies were reduced significantly while there were increases in the chironomids, *Ancylastrum* together with the appearance of *Hydropsyche.* No doubt the mayflies suffered from the siltation of the substrata whereas the burrowing choronomids benefited, the overall effect being a 6 fold increase in total numbers per unit area. A result of eutrophication, as we have seen, is to enhance the growths of *Cladophora* and to a lesser extent, mosses. The refuge that these plants allow to some invertebrates is considerable, the work of Percival and Whitehead (1929, 1930) on density in mild growths alone, revealing the enormity. When the *Cladophora* reaches lengths of up to 10 metres the so called '*Cladophora*/*Asellus*' zone and mosses up to 1 metre in length, the area and protection they give to species like chironomids, naids and *Asellus* allows inordinate numbers to develop. This zone is analagous with parts of the recovery zone in organic enriched waters, so much so, that the same terminology is applicable in both. On other parts of the substratum, enlarged populations of filter feeders like *Hydropsyche* and *Simulium* abound; in the stones *E. testacea* giving way to *E. octoculata*; *Baetis* and Trichopteran populations increasing also.

Pentelow *et al* (1938) investigating the effects of milk wastes on the fauna of the Bristol Avon noticed increases in Tubificidae, *E. octoculata*, *Helobdella stagnalis*, *Asellus*, *Gammarus*, and pulmonate molluscs although *Simulium* uncharacteristically vanished which was definately attributable to the churn washings. *A. fluviatilis*, *Hydropsyche*, *Polycentropus*, *Bythnia tentaculata*, Ephemeroptera and chironomids also increased.

Hawkes (1962) has pointed out that the effects of mild pollution on the fauna is to eliminate or suppress those taxonomic groups typical of the mountain streams or upper zones of rivers. In general, eutrophication has a similar effect.

The overall macro-invertebrate productivity accompanying organic pollution is very similar to those described for plankton in the River Lieve (DeSmet and Evens, 1972). In unpolluted water the production was seen to be relatively high, depending on the type of water, and more or less constant throughout the year, with numerous different species in equilibrium. This total production decreases with increasing enrichment, with decreasing numbers of different species, some of which present high production maxima. With heavy pollution very low total production values occur throughout the year.

Hawkes (1962) summarises the changes in riffle fauna co-incident with organic pollution as three major trends: -

"1) the progressive reduction in numbers and eventual elimination of the non tolerant species in succession according to their degree of intolerance.

e.g. Rithrogena - Ephemerella - Gammarus.

II) The initial increase in numbers of those species which at first are able to tolerate the adverse effects of organic pollution and benefit from the increased food supply and reduced competition. As the degree of pollution increases these are also successively reduced in numbers and are eventually eliminated according to their degree of tolerance.

e.g. B. rhodani - S. ornatum - Hydropsyche - L. pereger - Erpobdella sp.

III) The invasion of the habitat by species which under natural conditions are not members of the community either because the environment is not suitable under natural or because they are not able to compete successfully with members of the normal communities. Such organisms are naturally members of the silted communities in sluggish stretches or ponds. As the degree of pollution increases even these are successfully eliminated;

e.g. Nais - Asellus - Sialis - C. riparius - Tubifex.

Thus, in the rapids both quantitative and qualitative changes occur in the nature of the community with different degrees of pollution".

In the case of mild organic pollution and eutrophication, the quantitative and qualitative changes may be very small, but nevertheless detectable, when the water chemistry reveals nothing.

The described similarities of biocoenoces produced under organic stress only scratch the surface of the actual situation in any one river, each river having unique conditions and successions of its own.

2.5. RECOVERY FROM ORGANIC POLLUTION.

Some of the factors influencing the rate of recolonisation of rivers following abatement of pollution were mentioned in the introduction. The relative mobility of species resulting in speed of colonisation is of obvious application to fish, and both Hynes (1966) and Brinkhurst (1965) have made reference to examples. Hynes quotes the successional appearance of a number of taxa following the cessation of an organic effluent to a small stream which was previously dominated by tubificids. Six months after cessation, triclads, *Asellus*, *Gammarus*, *Baetis*, *Lymnaea* and chironomids all appeared. As insufficient time had elapsed for a new generation to have colonised, it was concluded that the species had moved in from clean tributaries. *Baetis*, *Gammarus* and *Asellus* are fairly easily dislodged, and winter spates probably caused drifting of the better adapted. Some animals present in the tributaries hadn't emigrated into the main streams, these invertebrates having effective holdfast mechanisms:-

stoneflies (unaffected under stones) Rithrogena, Ecdyonurus and Ancylus. The relative immobility of Mollusca was demonstrated following a cyanide spill removing L. pereger for 16 miles downstream. Two years later they had only recolonised this distance by half a mile from above the spill, and from below upwards towards the spill, to mile 8½. However, Hawkes (1963a) recorded the swift re-introduction of Lymnaea pereger in a stretch of the R. Cole as a result of drift wood and debris, upon which egg masses were attached, being carried downstream during high flow and deposited on shallow riffles.

Drifts constitute a major part of the recruitment to damaged areas. The time of year that more favourable conditions are restored may affect the rate of recovery in two ways. The proximity of the reproductive cycle together with an associated increase in temperature precipitates drifting. Other subsiduary factors affecting the rate of drift, e.g. riffle zones producing more than pools, and inadvertent effects like children playing in streams (Waters, 1962), will to some extent determine the velocity of recovery. It must be remembered that it is not analagous to recolonisation of an area cleared of fauna deliberately (Waters, 1964) because existing organisms (competition) residual toxities and substratum all reduce the potential rehabilitation.

Hawkes (1963) records the change in the biota of a Midland stream (Langley Brook) between 1949 and 1955 following the overloading of a sewage works discharging into it. In 1952 the works was overloaded resulting in a large reduction of *Gammarus* and *Baetis* at two stations 100 yds. and 0.4 miles below the effluent. *Ecdyonurus*, the Plecoptera and *Ancylus* were likewise reduced while *Asellus* and *C. riparius* increased in number just below the effluent; *Cladophora* was eliminated during the greatest period of organic loading, being replaced in the riffles by *Stigeoclonium*, but in 1955 when a new works had been completed it returned to its original abundance although *Stigeoclonium* remained in small amounts. Similarly the *Gammarus* and *Baetis* populations regained strength some way downstream at first, but by 1956 it was almost back to original proportions 100 yds. below. *Ecdyonurus* remained rare as did *Ancylus* but increases in leeches and *Simulium* were noted just below the effluent, which in the case of *Simulium* remained at the same abundance to at least 1.5 miles below. As expected, the *C. riparius* population disappeared altogether within the first 6 months of 1956 but *Asellus* retained high numbers which were only reduced by late 1957 and 1958 when interspecific competition from *Gammarus* was suspected as the cause.

Pentelow *et al* (1938) also observed an increase in the *Gammarus*/*Asellus* ratio following the diversion of milk churn and condenser washings to a sewer. There had been an enlargement in the *Asellus*, leech and pulmonate mollusc populations below the effluent whereas these were rare above it. The *Gammarus* population was also slightly enlarged as a result of the organic enrichment and ratios of Gammarus to Asellus below the waste were: -

	Gammarus		Asellus	waste
↓	1	:	7.9	
	1	:	3.3	
downstream	1	:	32.6	←
	1	:	0.85	sewage effluent

After the diversion of the effluent into the sewer the ratio of Gammarus increased.

↓	1	:	0.3	
	1	:	1.6	← sewage effluent
	1	:	10.5	
	1	:	0.05	

He demonstrated how colonisation of enriched sediments was fast when the water quality allowed more sensitive species to reappear.

A number of authors have noted transient stages with similar community structures to the stages in self purification of rivers. Nearly all report those invertebrates (and algae) which are typical of the Asellus/Cladophora community as remaining stable for a long duration. This is quite often explained by the water quality not returning to its natural state but being improved due to better methods of treatment, and still subject to intermittent doses of high organic loads. Very little literature is available that demonstrates the effects of the return of natural chemical and physical water parameters and those that do relate to poisonous effluents or spills.

The River Derwent was seriously deoxygenated by sewage sludge deposits, industrial and power station effluents prior to 1957 when a new works was built (Derby S.D.W.) which accommodated the industrial effluents, and new cooling towers with recirculation techniques being built at the power station also. Brinkhurst (1965) has shown how the number of taxonomic entities varied from his first survey in 1958 and his seventh in 1962 which were comparable with the pre-recovery survey of Hynes in 1951. He points out that the number of taxa is restricted to one type of habitat only and these exclude the enchytraeids and lumbriculids. Above all major sources of pollution the mean number of taxa was 11.6. The mean of 5 stations above the Derby and Spondon S.D.W.'s was 6.2 comparing favourably with the 1951 situation of 3.3 (results of Hynes). Below the works (station 8) the mean was 2.6 (1.0 in 1951), showing gradual increase in numbers downstream to 5.2 (1.2 in 1951). Although the variation between surveys and between stations is quite considerable, a distinct improvement is noticed, station 8 remaining consistently worse.

Most of the species returning were typical of those associated with the Asellus/Cladophora community. In 1951 the only species present at and below station 8 were tubificids, chironomids (Orthocladiinae) and sewage fungus. In April 1958 (first survey) *Asellus*, *Gammarus* and two species of leech, *Trocheta* and *E. octoculata*, more chironomids (Chironominae, Tanypodinae) had appeared with a concomitant reduction of tubificids. By September 1958 *Ephemerella ignita*, *Caenis rivulorum*, *Sialis lutaria*, *L. pereger*, *Ancylus* and *Crangonyx* together with a number of Orthocladiinae had established themselves. April 1959 saw the disappearance of *Ephemerella ignita* and the appearance of *B. rhodani* although this was never recorded downstream again throughout the survey to September 1961. The lack of any significant improvement in the September 1958 condition was attributed to the intermittent pollution and lack of clean tributaties to dilute the effects of it. Occasional accumulations of sludge were washed downstream also. He does not state which species were available in the tributaries for ecolonisation but presumably as they are described as 'not clean' the fauna was restricted.

The above examples have shown relatively swift colonisation by a number of species, the least sensitive appearing first and these later diminishing in numbers as competition from more sensitive species for food and space becomes apparent. This effect is similar to those noted for spills of toxic materials (Cairns *et al* 1972 for toxic chemicals and acid mine drainage, and Hynes and Roberts 1962, for detergents).

The speed of recovery seems therefore to be a function of the amount of drift, number of clean tributaries, mobility of invertebrates, and duration and complexity of life cycles. Insects have a distinct advantage here in being able to fly upstream (or down) to oviposit. Another speed dependent is the time of year when contamination is abated. Butcher _et_ _al_ (1931) showed the repeated return of _Sialis_, _Gammarus_, leeches and snails by the spring following killings during the winters when a sugar beet effluent was particularly severe. This could not have been due to reproduction but immigration. Cessation in spring would bring massive recruitment to the population.

The type of recovery in terms of taxa will be a function of the history of the river, fauna of clean tributaries, type and severity of previous contamination, residual toxicity and distance from the abated outfall.

2.6 INTERPRETATION AND PRESENTATION OF DATA.

The relative immobility of benthic invertebrates together with the affiliations noticed between their density/diversity and the degree of organic enrichment were inevitably utilised in packages of biological indicators of water quality. A number of the systems of presenting data were developed for the non-biologist and, although admirably motivated, they are inelastic, prone to misuse and raw data was irretrievable. These systems essentially fall into two types: those of a qualitative nature and those of a quantitative nature.

To some extent history has determined these present day systems, the reasons for this being discussed by Hynes (1960, 1964). The value of recording species of the benthos as present or absent is not in dispute but is greatly enhanced when numbers are available.

The system of Kolkwitz and Marsson (1908, 1909) was the first of its type with the idea that, equipped with a list of species, it was possible to allocate the locality (or indeed the whole) of a river to a saprobic zone - which was a stage in a river undergoing microbial decomposition of organic matter. There were four zones corresponding to degrees of pollution which are still used loosely to define such phases: Polysaprobic, alpha and beta mesosaprobic and oligosaprobic, in order of increasing recovery and self purification. As the name implies, the system (which was based on microscopic organisms alone) applies to organically polluted watercourses but was soon misappropriated for use with other types of pollution. Later, Kolkwitz and Marsson (1950) modified and reinforced the system augmenting it with macro-invertebrates. Liebmann (1951), realising the importance of the community, introduced the idea of quantifying the data but it still remained predominately microscopic in nature. After much criticism of its rigidity, Sladacek elaborated further on the system, introducing extended terminology and resolving more zones (9) plus an extra two zones for toxic and radioactive wastes (1966, 1969). This method of presentation is still used extensively in Europe, together with other saprobien system based methods reviewed by Bick (1963) who concludes that they are all too rigid, showing a lack in knowledge of the complexity of the situation. Other critics (Hynes, 1959, 1964) point out that organisms can be found in a wide range of conditions and may occur in restricted zones for reasons other than water quality anyway. He also observes in the Sladacek system (allocating ranges of oxygen, B.O.D., S_2 and bacteriological assays) that B.O.D. and oxygen are not necessarily related and that which applies in summer probably will not in winter. Sladacek (1973a) gives a generous 33 point critique of saprobic systems. In practice Caspers and Schulz (1960, 1962) using the system in stagnant Hamburg canals reported how absurd results could arise comparing naturally productive and artificially enriched springs, and Zimmermann (1961, 1962) showed that different ratings could be allocated if the speed of the current differed, although the levels of pollution are actually the same. Pennak (1971) suggests ignoring taxonomic data for the classification of lotic water because of its tediousness, and then goes on to describe a long list of physical and chemical parameters to determine.

Schmitz (1970) tells us that the use of the Kolkwitz and Marsson system is restricted to central Europe and not used at all in the States. In America there seems to be a simple division of streams into zones of pollution together with allocation of characteristic biota. The number of zones vary: - e.g. 3 (septic, degredation and recovery) of Gaufin and Tarzwell (1952, 1956), but they do warn against the use of individual species for measuring pollution as did Richardson (1929). Campbell (1939) went so far as to say that certain common and readily recognisable biota offer accurate indices of oxygen content which therefore measures the intensity of pollution. He divided streams into four zones: - recent pollution, active decomposition, recovery, and cleaner water, and quotes species indicative of them. Biglane and Lafleur (1954), Butcher (1946) and Tumpling (1969) also recognised four zones for convenience.

The biotic indices predominantly used in Britain are, as Sladacek point out (1973b), merely an extension of the Saprobien system. However, they have been developed with the idea that modifications can be made according to the discretion of the biologist using it; i.e. subjectively, according to the river's characteristics. Chandler (1970) has reviewed the Trent Biotic Index (Woodiwiss, 1964), and the Lothians Biotic Index (Graham, 1965) which are both qualitative, and introduces a new 'score' system, based on the others, but quantitative only in that it takes account of 5 degrees of abundance.

Bick (1963) reviews two simple European systems; one using a biological index and the other a 'species deficit' product. The Gabriel system (1946) makes use of the initially high density of reducing bacteria (R) and subsequent prevailing consumer populations (C) and finally autotrophic produces (P). The index is represented by: $I = \frac{2P}{R+C}$ but owing to the differences in volume and activity of the components, variable results have limited its use.

The 'species deficit' system of Kothe relies upon the reduction in numbers of species following the introduction of an effluent comparing the number of species occurring above (A_1) and below (A_x) and expressing the species deficit as a percentage of A_1 $\frac{A_1 - A_x}{A_1} \times 100$

the results are then graphically displayed.

Cooke et al (1970) suggests a method using control charts on the differences in the numbers of animals above and below the outfall, as opposed to the number of species.

Other systems relating the reduction in numbers of species to pollution categories are in operation in America. Patrick (1950, 1951) used what she calls the 'biodynamic cycle approach' as a basis to the method. She compiled what she considered to be a balanced community of a healthy stream and compressed the findings into seven categories. The survey data is made into seven corresponding histograms and the relative heights are compared with those drawn for the healthy stream. Although it is primarily intended as a visual system it is obviously weak for the compression of the reference (healthy) into 7 types and the balance of what she calls representative of a clean stream is not the same for all other streams.

Wurtz (1955) and Wurtz and Dolan (1960) decided that 4 'life forms' would likewise be representative of the fluviatile biotopes. These forms, burrowing, sessile, foraging, and pelagic, are represented in histogram form as a frequency index and plotted either side of a neutral line above which sensitive species are drawn and below which tolerant species are drawn. The interpretation relies upon the relationship between the two variables. A river is considered clean when non tolerant species represent more than 50% of the population. The weakness of this system lies in the false premise that there are species which are adapted to life in polluted water, and that they can be divided into pollution tolerant and pollution sensitive; i.e. no gradations.

The advantages inherent in the various 'diversity index' systems include simplicity (assuming access to a desk computer), and an expression that is unaffected by geographical differences in species and environment. The 'biotic indices' also discerned diversity but in terms of taxonomic entities or species only, whereas the diversity indices use the ratio of species to numbers whose relationship with organic enrichment has been shown to be important (Thienemann's second biocoenotic rule). Assuming the index to be independent of sample area (which is closely associated with numbers of individuals) the maximum diversity will be obtained when each individual belongs to a different species. Gleason (1922) considered the relationship between the no. of individuals and the log. of the area to be linear, whilst Margalef (1956) considered the latter to be directly proportional to the number of individuals. The synthesis is $d = S - 1/\ln N$. where, s = species number, and N = number of individuals. Wilhm (1967) compared a number of diversity indices applicable to macro-invertebrates and organic wastes and found the above formula to be the most sensitive, i.e. the index had the highest r value with no. of species (0.99) and most effectively distinguished between stations.

It is logical that any presentation of data should be as informative as possible and possess enough raw material to allow re-working by other investigators. Thus the use of simple tables are claimed to be the most illuminating by some workers (Hynes, 1964; Brinkhurst, 1965).

2.7 EXPERIMENTAL RESPIRATORY STUDIES: - OXYGEN FLUCTUATIONS, TOXICITY AND ECOLOGY.

Investigations into the oxygen consumptions of benthic and necktonic organisms are usually performed with their ecological distributions in mind. Straightforward studies of uptake in well oxygenated conditions may be related to a number of variables, and many authors have tried correlating rates with temperature, body size, season, sex and velocity of current. Hynes (1970) tells us that oxygen is rarely a factor in the autecology of invertebrates inhabiting clean rivers, as the oxygen saturation of the water is always near 100%. However, Macan (1963) says that it is only very swift streams that can maintain oxygen pressures near saturation and these are not of frequent occurrence. Moreover, many authors have demonstrated the effect of natural organic debris on the oxygen balance in autumn, a reduction that could determine the range of some species. More significantly, the effects of organic pollution and eutrophication on the oxygen balance have led numerous authors to try and demonstrate the relationships between the respiratory uptake and reductions in the oxygen pressure of the milieu.

Investigators have used different methods to measure the respiration and Teal (1971) has drawn a table listing the various electrode methods. Manometric methods are rarely used now but many figures have been determined using the Barcroft modification. The method utilised will to some extent be dependent upon the mode of expression of results desired: i.e. if simple uptake of oxygen over 1 hour at a certain oxygen pressure is required, manometric or sealed bottle techniques are adequate, whereas electrodes or polarographic techniques are necessary when long periods of exposure to varying oxygen pressures (for solution of lethal time or dose effect curves) are used.

It must be remembered that there is always variability in biological processes especially when dealing with whole organisms, and Teal (1971) estimates respiratory variation, to the order of 5-10%, as inherent.

It is a prerequisite that respiratory rates should be measured with the organisms in conditions close to those of their natural environment. Therefore light, substratum and temperatures should be controlled.

Seasonal variation and physiological condition.

Before the significance of depressed oxygen concentration in streams can be interpreted it is necessary to know something about fluctuation in metabolic demand. Lang (1951) demonstrated periodicity in consumption of _A. aquaticus_ (using Drastich's micro-respirometer) through the year. There was not only a difference in rate between May/June and October/December, but differences between the sexes in May and June also. He also revealed a diurnal variation with minimum consumption occurring between 11 am. and 1 pm, and maximum between the hours of 5 pm. and 6 pm. Berg _et al_ (1958) demonstrated a peak of activity in the limpet _A.fluviatilis_ kept at 11°C during April, May and June. Berg and Ockelmann (1959) found a 1.5 factor increase in the same limpet in spring and summer than in autumn and winter and suggested the very likely expression of sexual activity. This most likely explains Lang's results, as the two breeding seasons of _Asellus aquaticus_ coincides with the increase, but why the male uses more than the female he does not comment upon. Another physiological condition that increases respiratory activity is ecdysis. Macan (1963) cites an example of ecdysis between nymphal stages and points out that during the formation of the flying insect inside the skin of the last instar, the transfer of oxygen may be difficult and with the increased demand may restrict the range of the species to waters of suitable saturation.

a) Temperature.

Temperature can be important autecologically for reasons of physical and biological phenomena. The amount of oxygen available in solution will be reduced minimally with increasing temperature, but under natural and thermally polluted regimes, when considered with the increase in activity and metabolism in accordance with van't Hoff's rule, the combined effects may constitute detrimental conditions. Berg and Ockelmann (1959) noticed a 65% increase in oxygen consumption in the snails *L. pereger*, *Physa fontinalis*, *Bithynia leachi*, *B. tentaculata* and *Myxas glutinosa*, when the temperature was raised from 11°C to 18°C. The respiratory rates of *A. fluviatilis* and *Acroloxus lacustris* rose similarly with temperatures of 32 - 34°C and 31°C respectively (Berg, 1952) and then fell off quickly, much to his surprise, as *A. fluviatilis* would have been expected to have a lower temperature maximum, it being a resident of swift streams. However, Macan (1963) suggests that *A. fluviatilis* was more widely distributed than Berg was aware. Certainly Walshe (1948) showed that the stenothermic conditions of streams were reflected in the lower thermal resistance of chironomids taken from streams compared with those from still water. It is also manifest from Edwards (1958b) that the third and fourth instars of C. riparius use oxygen 2.6 times as fast as 20°C as at 10°C.

b) Velocity.

Streams also present the correlation of oxygen consumption with water velocity, those species living in the headstream relying on the natural current for changing the oxygen supply to their respiratory surfaces, e.g. Baetis rhodani. Species living in the lowland stretch may have the ability to draw a current of water over themselves. Fox and Simmonds (1933) demonstrated this ability by enclosing B. rhodani and Clöeon dipterum in a jar, Baetis dying first, it being suggested that the ability of Clöeon to move its gills and supply itself with new water being the causal reason. Fox and Johnson (1934) noted the increased pleopod activity of A. aquaticus in low oxygen concentrations and similar behavioural effects in amphipod pleopods, which draw water over the branchiae. Edwards and Learner (1960) observed cessation of activity with low dissolved oxygen, as well as increased pleopod activity, in both A. aquaticus and A. meridianus, the former observation being an example of regulatory behaviour that reduces metabolism.

c) Oxygen Reduction.

It is generally accepted that oxygen consumption of benthic invertebrates is related to the oxygen pressure of the water in which they live, and in practice, when the oxygen consumption of an aquatic animal is measured at various oxygen concentrations of the water, it is found that some animals have an oxygen consumption which is nearly proportional to it and others are able to maintain constant oxygen consumptions over a considerable range of oxygen concentrations. Animals which respond in the manner of the latter are often referred to as being 'independent' while the others are called 'dependent' types. Thus when oxygen consumption is plotted against oxygen concentration for dependent species, the resultant line will be more or less straight, but this is not so with independent species. Here, at a point in oxygen concentration the animal regulates its metabolic activity so that the rate of oxygen consumption may depreciate. This point has been called the 'critical point' (Berg *et al*, 1962) or the'incipient limiting point' (Fry, 1947). This is an important parameter ecologically because it is the point at which consumption is reduced due to the availability of oxygen.

Mann (1956) showed that *Piscicola*, *E. octoculata* and *E. testacea* have dependent patterns of oxygen consumption, while *Helobdella stagnalis* and *Glossiphonia complanata* showed a degree of independence in winter (Mann, 1961). *Piscicola* respiration was several times higher than the others, which is in accordance with its fast flowing stream habitat and accessory respiratory organs. He repeated the experiments (Mann, 1961) with the leeches acclimated to the oxygen concentration of the experiment for eighteen hours before use. He found that all the species showed the same relationships but E. testacea now showed a high degree of independence during summer, the incipient limiting point being at approximately 2.2 $mg.l^{-1}$ O_2 concentration. This suggests that E. testacea is better adapted to life in low oxygen concentrations. Typical habitats revealed the mean oxygen concentrations: -

Locality	Leech present	Mean O_2 mgl^{-1}
Foundry Brook	E. octoculata	6.1
Whiteknights Lake	"	4.3
Farley Pond	E. testacea	1.8
Colman's South	"	2.9

The point at which respiratory regulation is initiated is not always at low concentrations of oxygen as Berg and Ocklemann (1959) demonstrated with L. pereger. In this case, immediately after a decrease in oxygen content the oxygen consumption was reduced by the animal and a further marked decrease is manifest below 8% O_2 saturation. Subsequent investigations on L.auricularia, M. glutinosa, P. fontinalis, B. leachi and Valvata piscinalis show a dependent relationship up to the critical point of oxygen supply. There now appears to be a dilemma. Whereas it is reasonable to assume that any reduction of the oxygen content below the level where the active metabolic rate begins to be restricted is presumably unfavourable to the species concerned, (for reasons of growth, reproduction and activity) it is favourable as the suppression of respiration with dropping oxygen concentration may mean survival, when, to a wholly dependent or non-regulatory species, it may mean death. A synthesis of both positions, it seems to me, is that which can happen in polluted rivers, with, for example, E. testacea succeeding E. octoculata, whose energy is being expended trying to maintain its oxygen consumption so that feeding and reproductive activities are reduced, allowing E. testscea an environmental coup, despite its 2.2 $mg.l^{-1}$ O_2 incipient limiting point.

Animals which live in anaerobic conditions or conditions approaching anaerobiosis would be expected to be of the independent type or be in possession of another mechanism favourable to survival. A. aquaticus, common in alpha-mesosaprobic conditions, suppressed its oxygen consumption by 15 - 20% when the oxygen concentration was reduced from 8.3 - 1.5 ppm O_2, an identical result being obtained with A. meridianus: - unknown in polluted conditions (Edwards and Learner, 1960). Allee, (1929) observed A. communis living in huge natural aggregations and their de-oxygenating effect on the surrounding water.

In the midst of the aggregation the dissolved oxygen was only 2.61 ppm, but those in the stream away from the main bunch (but still in thousands) were surrounded by water containing 6.37 ppm O_2. He subsequently demonstrated that those living in the aggregations used less oxygen than those scattered away from it, suggesting that some form of acclimation may have taken place.

Fox and Taylor (1955) corroborated the expectation that C. riparius would survive best in very low oxygen concentration in consideration to its habitat, (polysaprobic). It lived longest and grew best at 4% saturation. Anatopynia (not a tube dweller) also survived better in low saturation, together with Heterocypris incongruens and Tubifex. The haemoglobin content was exhibited to enlarge in C. plumosus, Anatopynia, Daphnia and Planorbis but not in Tubifex. C. riparius and Tubifex were shown to be poisoned at high oxygen levels, the suggestion being that the functioning of essential reducing enzymes was impaired by the oxygen. However, Heterocypris and Planorbis were tolerant of high oxygen. Physa and Pisidium they surprisingly found equally tolerant of 100%, 21% and 4% saturations, but Lymnaea lived longer in full aerated water. A. fluviatilis was seen to die faster than A. lacustris in anaerobic conditions (Berg, 1952), the greater sensitivity of the stream species being in good accordance with its distribution in clean waters, however Berg did suggest that there was an overriding factor explaining their distribution.

Augenfeld (1967) suggested that the ability of many invertebrates including C. plumosus to withstand anoxia was correlated with their possession of substantial stores of glycogen. This may be converted to lactic acid anaerobically using lactic dehydrogenase and a glycolytic enzyme system but this respiration yields only 10% of the energy resulting from complete oxidation. Large amounts of glycogen would be needed to make this worthwhile. C. plumosus and C. riparius have 13 - 14% of their body weights as glycogen which helps to explain their tolerance to pollution, whereas Tanytarsus, which is intolerant to de-oxygenation only has 2% glycogen by body weight. C. plumosus accumulates lactic acid for several days even when slight aerobic conditions are restored, and then the rate of assimilation is reduced, i.e. oxygen 'debts' are possible.

The larvae of Tanytarsus brunnipes and Anatopynia nebulosa common in streams show a linear relationship between oxygen consumption and oxygen content whereas two closely related species, native of stagnant water, maintain constant utilisation until the oxygen is very low (Walshe, 1948).

(C. longistylus I.L.P. 1.5-2.5ppmO_2)
(A. varia I.L.P. 2.5-3.5ppmO_2)

This shows how species that are used to de-oxygenated conditions have adapted to their environment, in this case augmented with presence of haemoglobin.

Having shown examples where oxygen consumptions of benthic invertebrates are related to the oxygen content of the water and that these observations, with particular reference to the useful incipient limiting points, can reflect and possibly explain some autecological distributions, it is desirous to show how one can be misled. Nagell (1973), apart from disclosing the importance of acclimation to light, water and food, measured the oxygen consumptions of a number of Ephemeroptera and Plecoptera. He found, as expected with fluviatile members of these orders, a high critical point (5.7 mgO_2/l at 8°C) in *Nemoura cinerea*, which is not normally exposed to low oxygen in nature, which suggests that it is particularly sensitive to low oxygen compared with *Clöeon* (pond species exposed to low oxygen occasionally; critical point 2.0 - 2.5 mgO_2/l at 8°C). However, $L.D._{50}$ extrapolations from mortality curves showed 1.9 mgO_2/l for *Clöeon* and 0.4 mgO_2/l for *Nemoura*. Thus despite the absence of a respiratory regulation mechanism, *Nemoura* survives low oxygen better than *Clöeon*. Counteracting this, *Clöeon* is more active up to its death whereas *Nemoura* loses movement after a short time which clearly gives *Clöeon* a better chance to leave areas of low oxygen.

Also using time/effect curves, the 24 hr. $L.T._{50}$ of *A. intermedius* was 0.03 mg/l (20°C), which shows it to be of similar resistance to low oxygen as the European *A. aquaticus* (Sprague, 1963). Two amphipods, *Gammarus* pseudolimnaeus and G. fasciatus, were also tested showing 24 hr. L.T.$_{50}$ of 2.2 mg/l and 4.3 mg/l at 20°C respectively. This is in good accordance with the european G. pulex and with observations of tolerance to oxygen concentrations.

Davies (1971) established a table demonstrating degrees of tolerance of benthic invertebrates to different water quality parameters. The following table gives his results for oxygen depletion in order of increasing tolerance: -

Rhyacophila dorsalis
Ecdyonurus dispar
Brillia longifurca
Hydropsyche angustipennis
Prodiamesa olivacea
A. aquaticus
Helobdella stagnalis
E. octoculata
E. testacea
C. riparius

This corresponded with his field observations of tolerance to organic pollution.

The ameliorative effects of current to tolerance to low oxygen tensions has been demonstrated by Jaag and Ambuhl (1963): -

TABLE SHOWING THE EFFECT OF CURRENT VELOCITY ON THE LETHAL CONCENTRATION OF O_2 IN mgl^{-1} OF SOME INVERTEBRATE SPECIES.

	1 cm/sec	6 cm/sec	current velocity
Ecdyonurus venosus	0.8	0.4	
Ecdyonurus ignita	1.2	1.2	
Rithrogena semicolorata	4.6	2.4	
Rhyacophila nubila	1.2	0.4	
Hydropsyche angustipennis	0.9	0.6	

This evidence shows how low oxygen conditions can be tolerated if the current velocity is fast, and is fundamental to the knowledge that metabolism depends upon the velocity of the current. They have summarised the various components, whose interplay finally determines the oxygen content with which the animal satiates its needs, into three influences: -

1) oxygen consumption by the animal (breathing)
2) oxygen content of the surrounding water
3) the rate at which the boundary layer is renewed, which the velocity of the current plays a decisive role in determining. The latter also helps to determine the thickness of the boundary layer, which is significant for the diffusive component of the gas exchange.

There is not, to my knowledge, any literature pertinent to tolerance of invertebrates subjected to fluctuating oxygen concentration in either time or intensity. This is surprising considering the large fluctuations described that can occur. It also motivated the present investigations.

CHAPTER 3.

METHODS USED IN EXPERIMENTAL STREAMS AND RIVER SURVEYS.

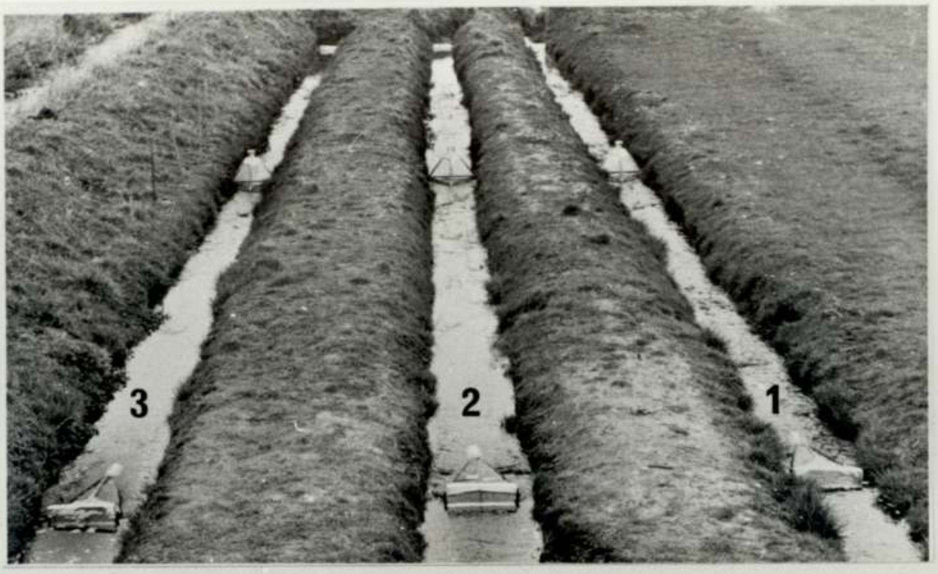
3.1. DESCRIPTION OF HABITATS.

3.1.1. The Experimental Streams

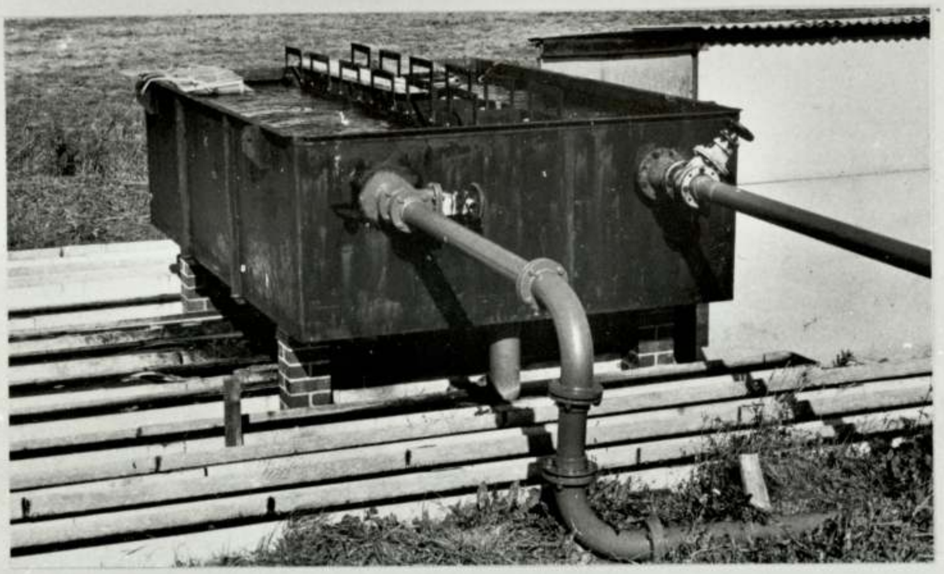
These artificial streams were excavated as part of a research project having financial support from the Nuffield Foundation. They are situated on the Blithe Valley Sewage Disposal Works site at Checkley in Staffordshire. This site was chosen because of the availability of high quality river water and well oxidised sewage effluent in sufficient quantities. Water from the trout stream was abstracted above the effluent by means of an existing hydram chamber feed operating by change of head. This feed was directed into an 11.2 m^3 sedimentation tank (plate 1) where some of the suspended solids were removed before its being pumped to the mixing tank (plate 1). The efficiency of the sedimentation chamber is demonstrated in Fig. 3.1 which shows its effect at high river flows. As there was no regulation of flow to the sedimentation tank, its efficiency was reduced i.e. when the river flow was high; so, consequently, was the input to the tank causing increased turbulence.

The mixing tank is divided into two chambers. One chamber receives pumped river water from the sedimentation tank, the other pumped final effluent from the sewage treatment works. Both chambers are provided with six adjustable weirs, over which the river water and final effluent flow to be mixed in the proportions determined by the weir setting and finally to be distri-

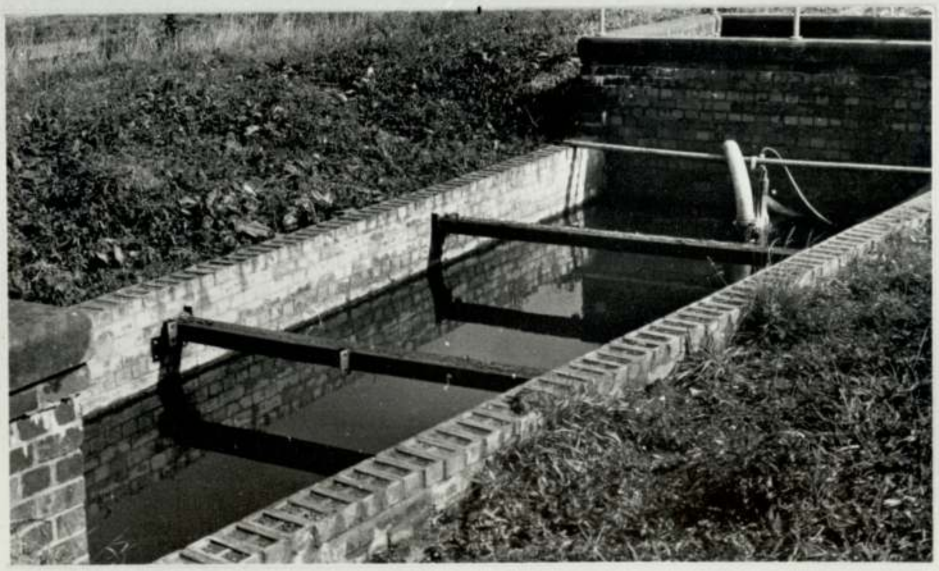
plate 1



EXPERIMENTAL STREAMS

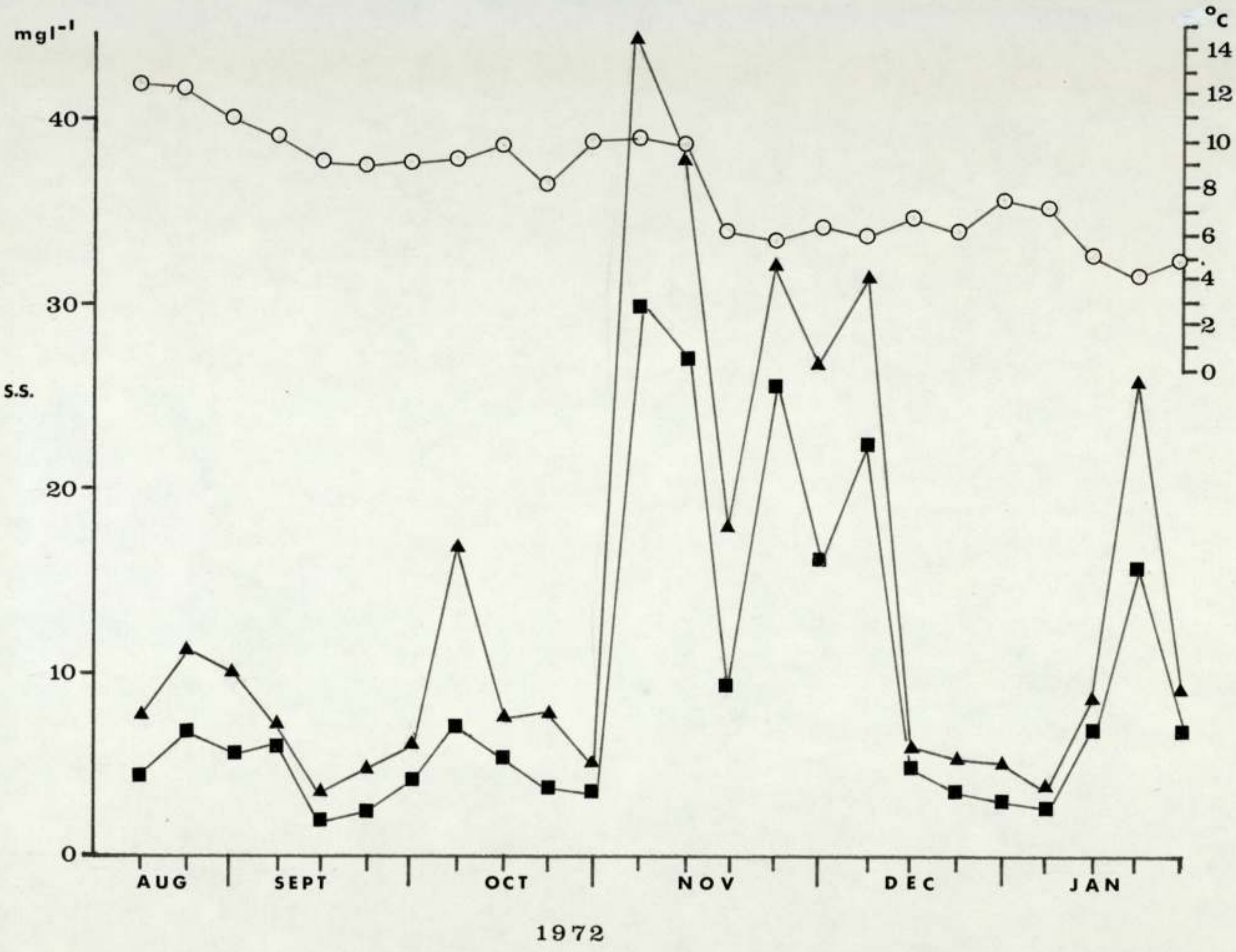


MIXING TANK



SEDIMENTATION TANK

FIG.3.1. showing the effect of the sedimentation tank over a period of six months; ▲ denotes the concentration of influent suspended solids; ■ denotes the concentration of suspended solids being pumped from the tank. ○ denotes temperature.



buted into six asbestos channels. These asbestos channels are not part of the present study except for the fact that they are the vehicle of water transport to the three excavated streams (plate 2) - the dilutions of stream water to effluent are also shown. The water from the six asbestos channels flows into the three excavated streams, the reduction being accomplished by pairing of the six flows into three, giving mixtures of sewage effluent to river water in the region of 1:3, 7:1, and a control stream receiving 100% river water. Actual percentages of mixing have been solved using (i) the levels in mgl^{-1} of total inorganic phosphorus (ii) the levels of chloride in mgl^{-1} and the equation: -

$$P_1X + P_2Y = Z$$

X = sewage effluent in mgl^{-1}

Y = river water in mgl^{-1}

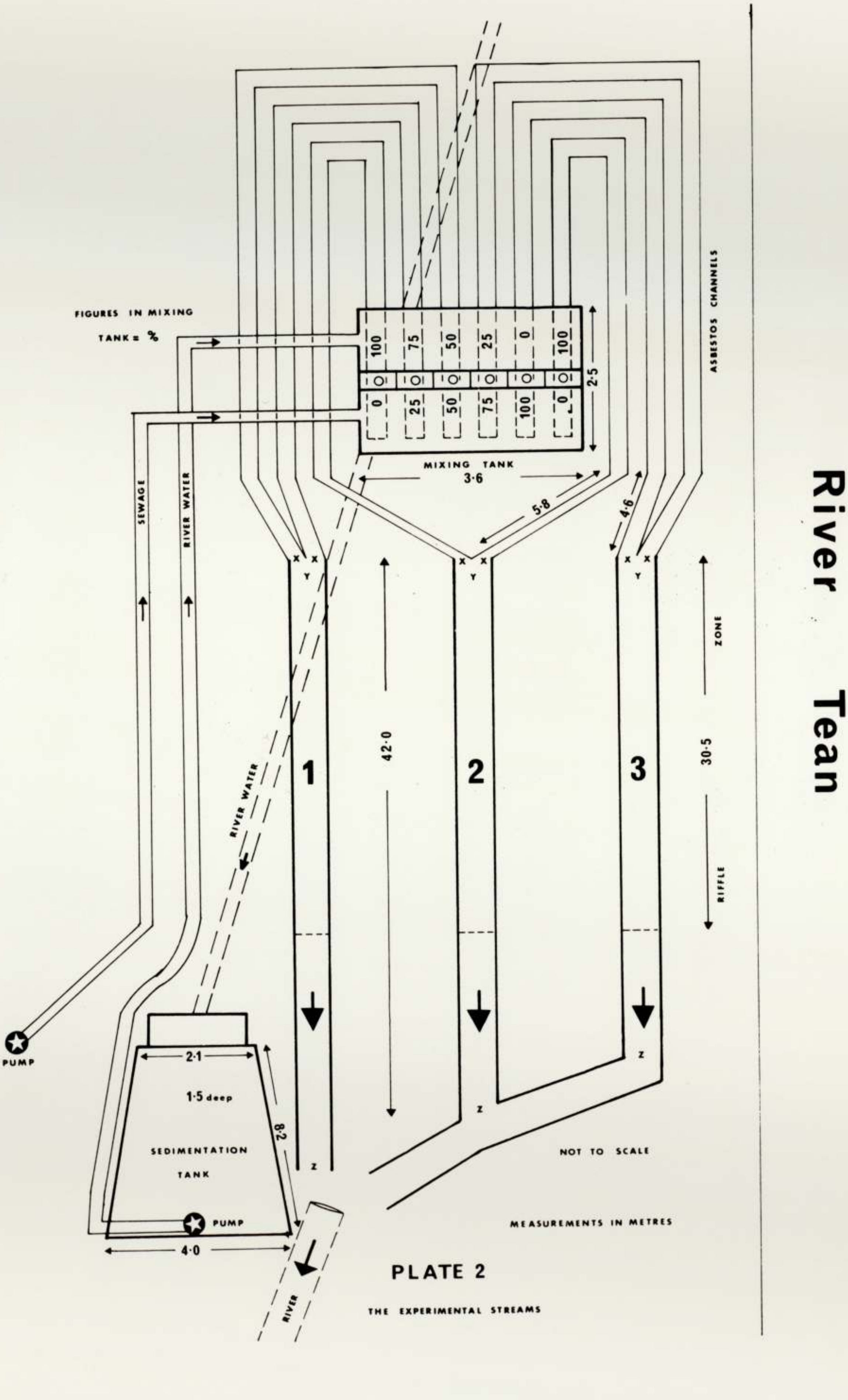
Z = resultant for stream 1 or 3 in mgl^{-1}

P_1 and P_2 = proportions of units in respective channels.

From this the actual percentages are shown to be: -

In stream 1	37.5 sewage effluent, 62.5 river water
In stream 2	100% river water
In stream 3	87.5 sewage effluent, 12.5 river water.

The experimental streams were excavated with a J.C.B. mechanical digger to a length 138 ft. (42m), with the exception of stream 3 which was only 126 ft. (38.4m). The width of all streams was 2.2 ft. (0.67m)



NOT TO SCALE

MEASUREMENTS IN METRES

PLATE 2

THE EXPERIMENTAL STREAMS

An equal downward gradient was given to all the streams so that a riffle effect should be obtained throughout their length.

It was decided that the substrata should be from local sources and as close to the substratum of the riffle areas of the river from which the water was being abstracted (for later biological comparison). As no stones could be obtained commercially which were of a suitable smoothness and diversity in size and colour, they had to be removed by hand from riffle sections of the river. These stones (between 1 in. and 4 in.) were then distributed similarly between the experimental streams to an average depth of 4 in. (10.2 cm) and to a length of 90 ft., i.e. 70% of the stream length. Therefore the first 90 ft. (27.5m) of all the streams were provided with a stoned riffle section.

The sides of the streams were bevelled, especially where the gradient was steepest, to allow more light to fall into the streams but not so much that a measure of protection could not be taken from direct sunlight. The streams run parallel to the R. Tean in a NW-SE position.

The flow rates down each stream should in theory have been equal but no doubt minute fluctuation did occur owing to voltage reductions, differences in surface tensions, and siltation rates. The outflow from each channel was combined and passed through a 12 in. pipe to the river.

Samples for chemical analysis were taken Monday to Friday at approximately 9.0 a.m. with the exception of dissolved oxygen executed at midday. A 24 hr. survey was made of all chemical parameters between 5 - 6th April 1974.

3.1.2. River Surveys.

Two rivers (the R. Ray and R. Tean) were chosen whose chemical and biological status were complementary to the experimental stream project. The choice of the R. Tean, whose water supplies the experimental streams, was obvious for use in two respects:

(i) to see how well the benthic macro-invertebrate communities established themselves in the artificial stream situation compared with those under similar chemical and physico-chemical conditions in the R. Tean, and

(ii) as a separate entity to illustrate the outcome of the contemporary problem of discharging highly oxidised effluents, i.e. the study of a eutrophicated lotic system.

A second river, the R. Ray (Wiltshire) was chosen because of its highly eutrophicated state resulting from the discharge of a highly nitrified effluent. Also, as a result of the above, huge blankets of the alga Cladophora would seasonally appear together with large populations of the isopod Asellus aquaticus. The latter was thought useful for a production study, especially as its results could be compared with those populations expected in the as yet uncompleted artificial streams.

The other major part of the thesis concerns the ecological recovery of a river from gross organic pollution. The river under observation was the R. Cole, a tributary of the R. Tame (Midlands). Historical ecological data was available from the 1950's while the river was subjected to the regime of two ineffective sewage treatment works whose effluents continued to discharge until the works were closed within a year of one another in 1971 and 1972.

The major discharge from the Yardley works was a highly organic and metallically toxic effluent into the River Cole about half way along its total course. For this reason and for convenience of illustration, the River Cole was divided into two reaches, the Upper Cole and the Lower Cole. The Upper Cole was the recipient in its upper reaches of a small but badly oxidised S.D.W. effluent. However, although small and metallically innocuous, the volume of the River Cole at this point was approximately doubled. It was in this upper reach that the recovery was expected to be most dramatic.

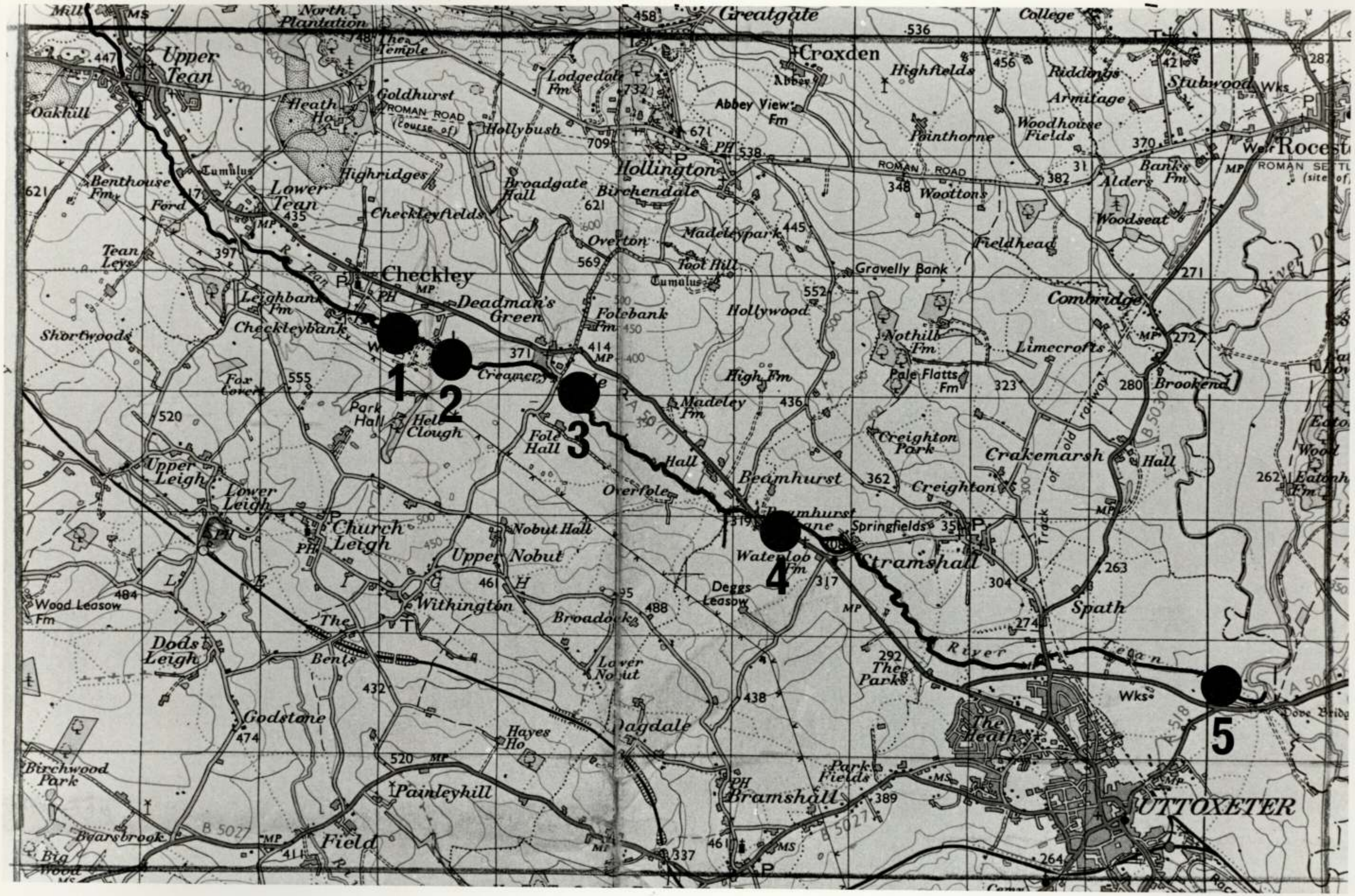
a) River Tean. The river starts at Huntley (SK 006415) about 14.5 km. E of Stoke-on-Trent after the union of two streams draining farmland NW and NE of Cheadle (Staffs) and flows for about 16 km. SE to its confluence with the River Dove near Uttoxeter. The only sources of contamination, apart from the two described below under Station descriptions, are agricultural run-off. The survey was carried out between November 1972 and November 1973. A map of the area is shown in plate 3.

Station 1 SK 028377. 9.3 Km from confluence. Approximate Current 45 cm/sec. This station is about 273m. above the outfall of the Blithe Valley S.D.W. The river is wide, shallow and shaded here with frequent small growths of the moss Leptodyctium riparium, Fontinalis antipyretica, and small tufts of Cladophora and Vaucharia may be seen all the year round, but with peaks occurring between March and July. There is never much more than 30% cover though some of the tufts may reach 5 - 7 cm.

Station 2 SK 037373. 8.7 Km from confluence. Approximate Current 77 cm/sec. This station is about 182 m. downstream of the effluent and only rarely is any Cladophora observed here, but some 'sewage fungus' grows on the stones in winter. This riffle area was displaced a little downstream by a detour of flow caused by a fallen tree in June 1973.

Station 3 SK 046371. 7.5 Km from confluence. Approximate Current 60 cm/sec. This riffle is situated just below a Creamery. The Creamery effluent, although good, may give rise to a little 'sewage fungus' seen on

plate 3



R. Tean

the sides of the riffle in winter but this never lasts long. Some patches of grassy macrophytes about one metre in length are found here - Sparganium, Ranunculus and Potamogeton, in particular which tend to accumulate grey silty deposits. Anglers sometimes complain that the river turns grey on occasions and blame it on the Creamery.

Station 4 SK 068357. 4.6 Km from the confluence. Approximate Current 66 cm/sec. The riffle here is wide and shallow (10 cm. on average) and the largest quantities of Cladophora are encountered here, especially in the slower flowing edges. There may be up to 80% cover in July and August. Some farm effluent enters the side of the riffle and causes small 'sewage fungus' growths during the colder months, this area being avoided during sampling. The water level may be controlled here by flood gates upstream and often the depth of water in summer was only 5 - 8 cm.

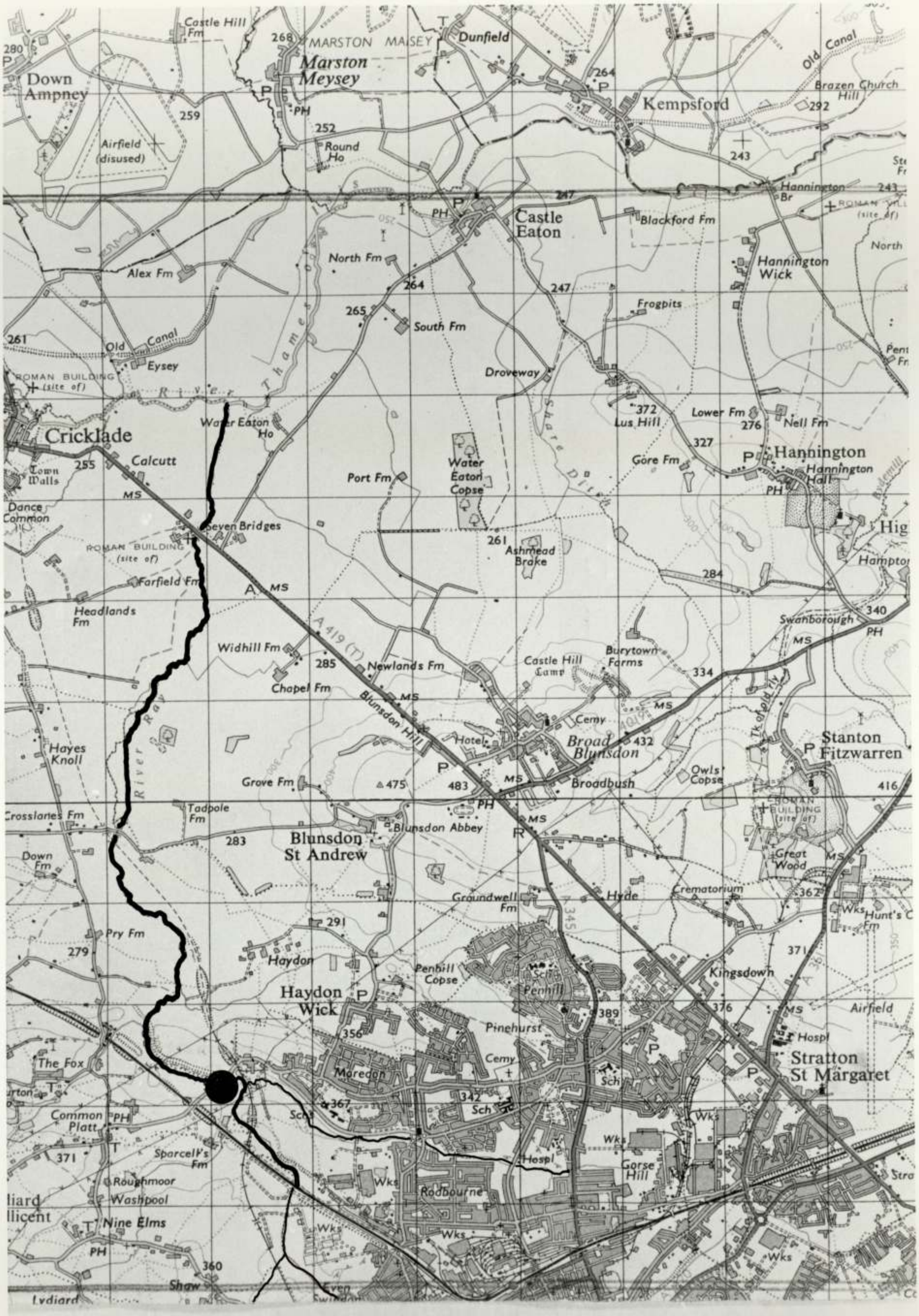
Station 5 SK 102346. 0.25 Km from the confluence. Approximate Current 70 cm/sec. The river here is swift flowing with a gravelly bottom to the riffle and consequently has a shifting substratum with very little macrophytic of algal growth. Some Cladophora grows in small tufts in summer.

Chemical and biological samples were taken monthly at all Stations during the period November 1972 - November 1973.

b) River Ray. The River Ray drains Kimmeridge clay and the chalky Marlborough Downs. It joins the River Swinbourne at Mannington which is made up of mostly effluent from the Wroughton S.D.W. (1978 m^3.d. d.w.f.) 1.3 Km downstream Rodbourne S.D.W. effluent enters (285×10^3 m^3d. d.w.f.), which constitutes a major part of the Ray's flow and almost all of it in summer. The River Ray then flows through agricultural land for a distance of 12.2 Km to its confluence with the River Thames at Water Eaton. The depth of water in the River Ray may vary greatly within short time periods, i.e. it is a flashy river. After a preliminary survey of three stations, one was selected for study and this was surveyed monthly from December 1971 until April 1973.

Station SU 121871 Elborough Bridge Moredon.
See Plate 4. This site was 2.1 Km downstream of the Rodbourne outfall and just downstream of a C.E.G.B. generating station. (The cooling water from the station was cooled in ponds and re-circulated). The river here, which was 6.3 m. wide, was rapid flowing (41 cm/sec) with a bed of stones and shingle on a clay base. The average water level was between 0.5 and 1.m. Huge growths of Cladophora have been recorded here and, although present all the year round, peaks occur in July (111.2 g/m^2 1971 and 210 g/m^2 1972 - Urquhart 1972) and streamer lengths may reach 6 m. at these times. The moss Fontinalis antipyretica peaks around April and May but in much smaller quantities (between 50 and 80 g/m^2 - Urquhart

plate 4


Castle Hill Fm
Marston Meysey
Dunfield
Down Ampney
Kempsford
Old Canal
Brazen Church Hill
Airfield (disused)
Round Ho
Castle Eaton
Blackford Fm
Hannington Br
North Fm
South Fm
Hannington Wick
Alex Fm
Frogpits
Old Canal
Eysey
ROMAN BUILDING (site of)
River Thames
Droveway
Water Eaton Ho
Lus Hill
Lower Fm
Nell Fm
Cricklade
Calcutt
Town Walls
Port Fm
Water Eaton Copse
Gore Fm
Hannington
Hannington Hall
Dance Common
Seven Bridges
Ashmead Brake
Farfield Fm
Headlands Fm
Widhill Fm
Chapel Fm
Newlands Fm
Castle Hill Camp
Burytown Farms
Swanborough
Blunsdon Hill
River Ray
Hayes Knoll
Hotel
Broad Blunsdon
Stanton Fitzwarren
Grove Fm
Broadbush
Owls Copse
Crosslanes Fm
Tadpole Fm
Blunsdon St Andrew
Blunsdon Abbey
Down Fm
Great Wood
Groundwell Fm
Hyde
Crematorium
Pry Fm
Haydon
Haydon Wick
Penhill Copse
Penhill
Kingsdown
Pinehurst
Stratton St Margaret
The Fox
Moredon
Common Platt
Sparcells Fm
Rodbourne
Gorse Hill
Roughmoor
Washpool
Nine Elms
Shaw
Lydiard Millicent

R. Ray

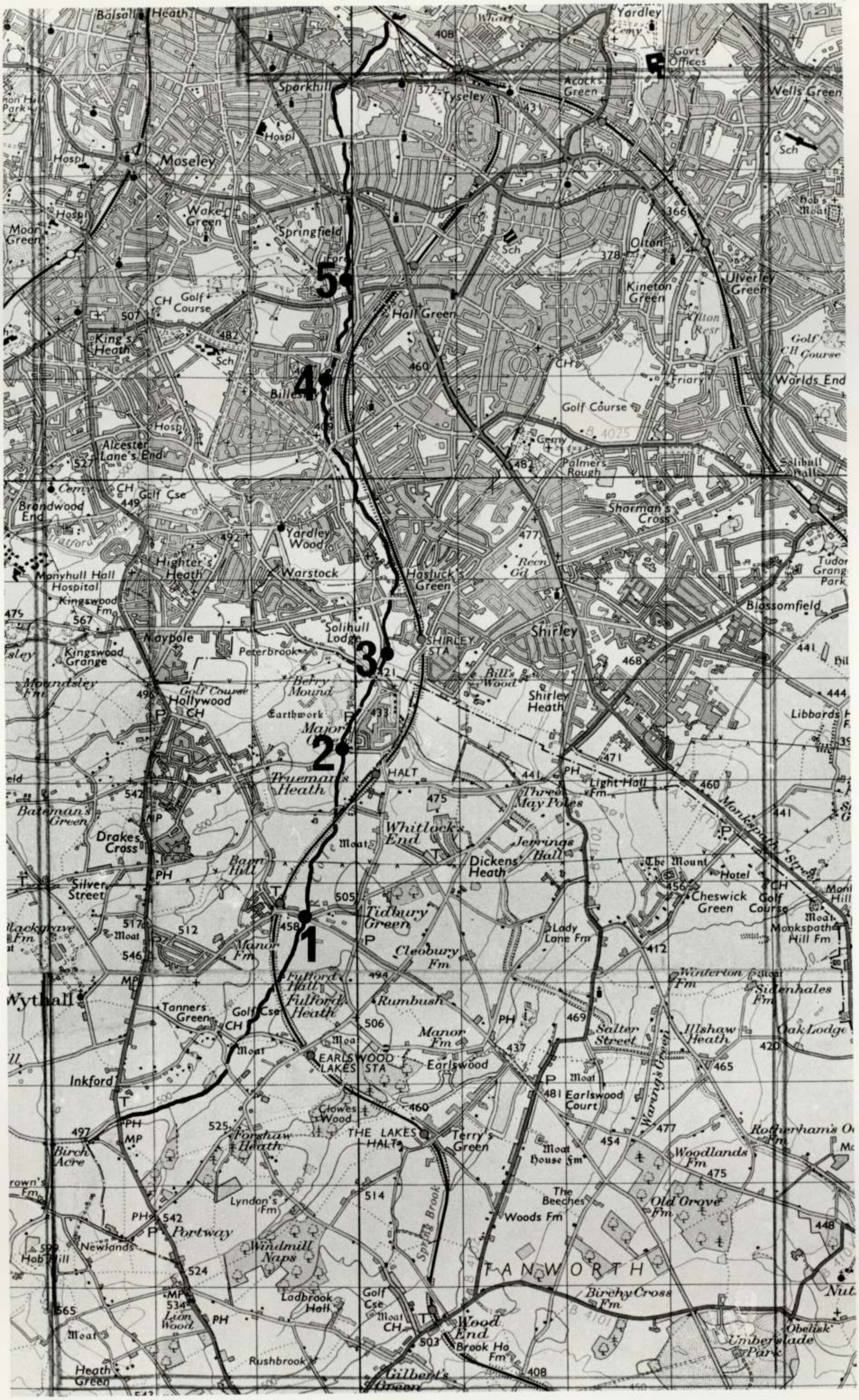
1972) and may reach up to 1 m. in length. Vaucheria and Oscillatoria were also observed in small quantities.

c) River Cole. It rises about 24 Km south of Birmingham passing through its suburbs and flowing east towards its confluence with the River Blythe, a total distance of about 40 Km. The survey was undertaken from January 1972 until September 1973, which takes in a period of six months before the closure of Houndsfield Lane S.D.W., and fifteen months afterwards for studying the ecological recovery. The Yardley S.D.W. closed in June 1971 and information pertaining to pre-closure history was obtained from Hawkes (1964), Hawkes and Davies (1971), and Martin (1973). Maps of the areas are shown on Plates 5 and 6.

Station 1 SP 095755. Lowbrook Bridge. 28.8 Km. from confluence. This station is 1.3 Km upstream of the Houndsfield Lane S.D.W. and consists of a riffle 1.5m wide and about 10 cm. deep. In summer, short tufts of Cladophora are seen attached to the stones and gravel of the substratum.

Station 2 SP 099744. Truemans Heath. 27.1 Km. from confluence. The river here was made up of about half effluent from the Houndsfield Lane S.D.W. prior to its closure and consequently, there was almost complete cover with 'sewage fungus' in winter.

plate 5



Upper Cole

Station 3 SP 103784. Aquaduct Road. 26 Km. from confluence. The riffle here is about 3 m. wide, shaded, and often tufts of Cladophora and Stigeoclonium are seen growing. The bottom sometimes becomes silty and frequently rubbish is dumped into it.

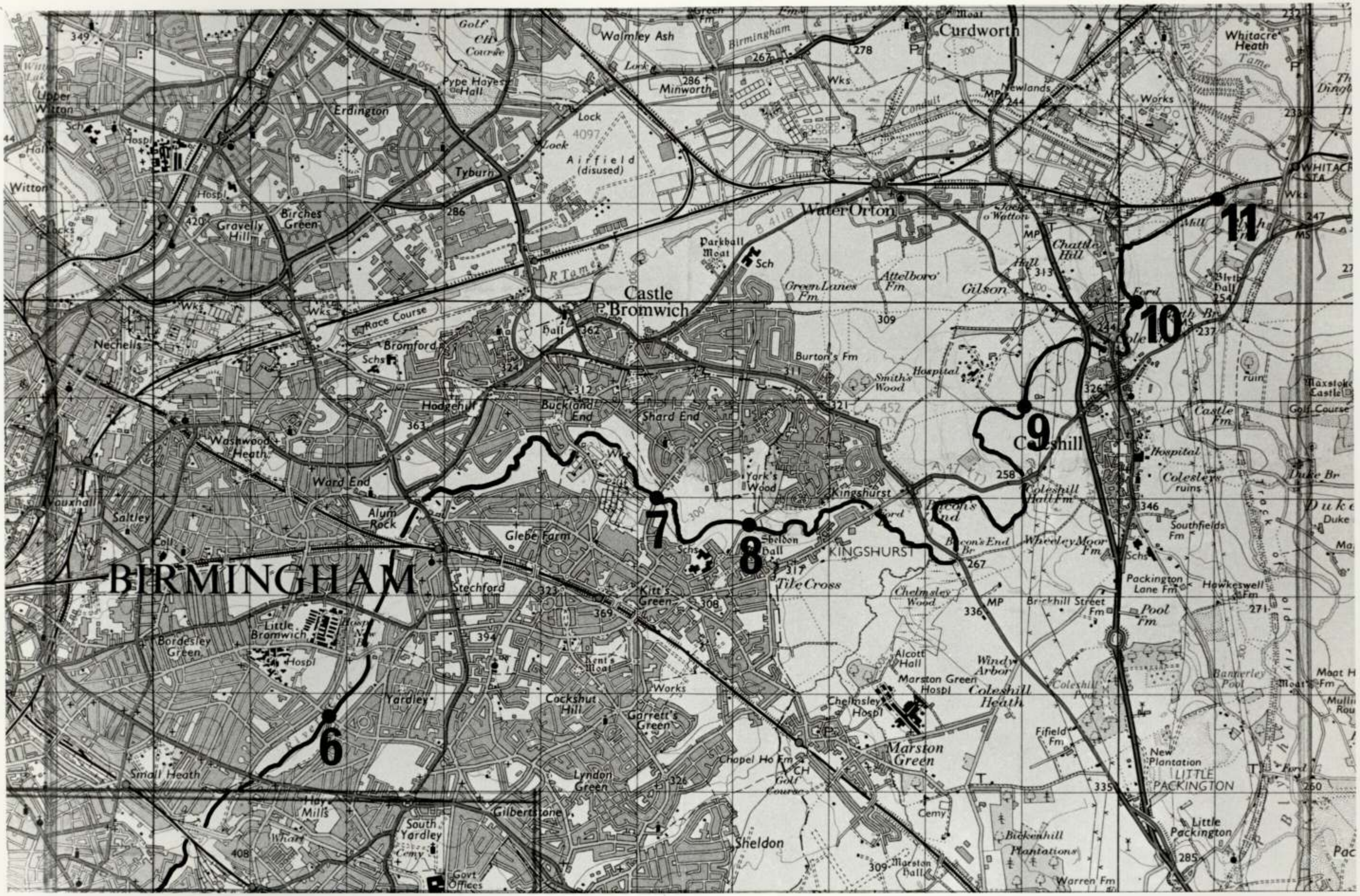
Station 4 SP 098812. Brook Lane. 23.3 Km. from confluence. The flow is increased here by a tributary, Chinn Brook. Small patches of Cladophora occur all the year round with peaks in summer (up to 1.5 m.). The riffle is approximately 3 m. wide and 25 cm. deep.

Station 5 SP 099832. Sarehole Mill. 22.5 Km. from confluence. Long growths of Cladophora up to 3 m. in autumn occur on the very stony and slightly silted bed.

Station 6 SP 122865. Bordesley Green. 17.9 Km. from confluence. The riffle is almost always covered by junk/debris thrown in by children and adults. The stoney bottom is often silted and some macrophytic growths of Ranunculus and Potamogeton occur, while in winter 'sewage fungus' is prevalent. The river here is about 6 m. wide with large and irregular shaped boulders not being uncommon.

Station 7 SP 144885. Below Yardley. 12.2 Km. from confluence. This station is situated about 100 m. downstream of the disused Yardley S.D.W. There are only a few large stones here, the bottom being mostly pebbles and gravel. In winter there is frequent cover with Stigeoclonium.

plate 6



Lower Cole

Station 8 SP 162877. York Wood. 10.7 Km. from confluence. The riffle here is about 6 m. wide and 20 cm. deep which supports small tufts of Cladophora and Stigeoclonium.

Station 9 SP 190889. Coleshill Hospital. 4.3 Km. from confluence. A wide and long riffle here with some algal cover of the stone and gravel substratum. There is occasionally disturbance of the riffle by cattle drinking.

Station 10 SP 200898. Wheatsheaf.
More sluggish here with extensive cover of filamentous algae in summer. Some silt amongst stones.

Station 11 SP 210911. Shustoke.
Stony with gravel and some silt. Some streamers of Cladophora up to 3 ft. in summer and autumn. Riffle about 6 m. wide and 30 cm. deep.

Chemical and biology samples were taken monthly at Stations 1 - 5 and every two months at Stations 6 - 11, between January 1972 until September 1973.

3.2. ANALYTICAL METHODS.

The following analytical methods were used for all the river and experimental stream surveys.

3.2.1. Chemistry

a) pH. This was measured on a Pye taut band pH meter with temperature compensation, Model 78. pH was read to 0.1 of a unit.

b) Temperature was read to 0.5°C. with a -5°C to 50°C. mercury thermometer for accuracy in the determination of % saturation of water samples with oxygen.

c) Ammoniacal nitrogen and total oxidised nitrogen was determined on a Technicon Auto-analyser Mk II using the methods and flow diagrams of Chapman, Cooke and Whitehead (1967). Results were expressed in mgl^{-1} N.

d) Total inorganic phosphorus was likewise determined on the Auto-analyser utilising the methodology of Technicon (Industrial method 3-68W), but with a modification of 30 samples per hour and standards recalculated for phosphorus. Results were expressed in mgl^{-1} total inorganic phosphorus.

The following analyses were all determined using the methods described in "Analysis of Raw, Potable and Waste Waters", Dept. of Environment, HMSO 1972.

e) Alkalinity - the titrimetric method using n/10 HCL screened methyl orange as indicator (1g methyl orange and 1.4 g xylene cyanol F.F. in 500 ml 50% C_2H_5OH). All results were expressed as total alkalinity as mgl^{-1} $CaCO_3$.

f) Chloride - $AgNO_3$ method. All results were expressed as mgl^{-1} chloride as Cl^-.

g) Permanganate value. The 4-hour P.V. at 27^oC procedure was adopted. All results were expressed as mgl^{-1}.

h) Dissolved oxygen. The Alsterberg azide modification described was used. Also 500g NaOH and 150g KI were used in synthesising the alkaline iodide solution instead of the 400g NaOH and 900g NaI per litre). Results were expressed in mgl^{-1} N/40 $Na_2S_2O_3.5H_2O$ was used.

i) Percentage saturation. The dissolved oxygen was converted into percentage saturation utilising the O_2 solubility tables given, so that results of Winkler determinations would be in a form more acceptable to some biologists.

j) Biochemical oxygen demand. The dilution method was used and nitrification not suppressed. The alkaline iodide reagent was similar to that described for dissolved oxygen. Results were expressed in mgl^{-1}.

k) Total suspended solids. Expressed as mgl^{-1}.

l) Total hardness. Expressed as mgl^{-1} $CaCO_3$.

Meteorological data was supplied weekly for the Birmingham area, and monthly for the Newport (Staffs) area (nearest to the experimental streams and R. Tean) by Birmingham University Observatory.

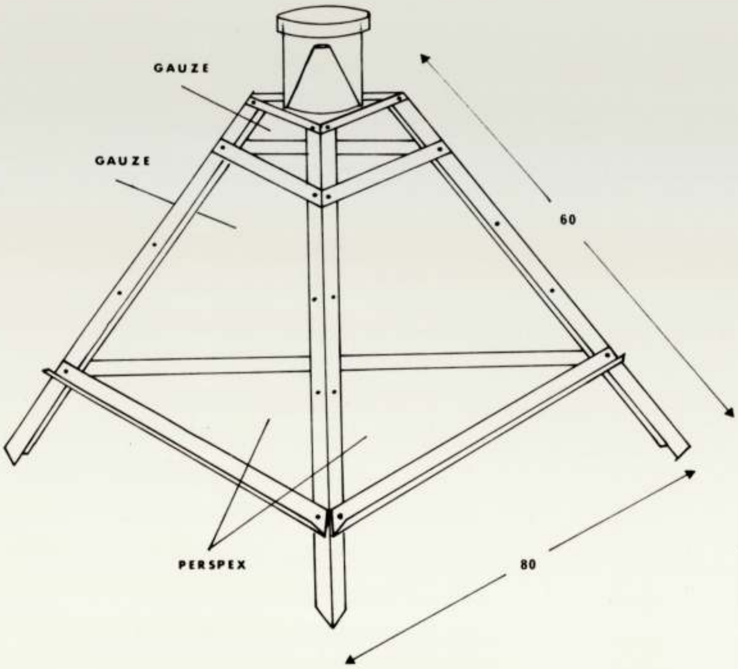
All samples taken for chemical analysis were stored in all glass 'Winchesters', no polythene was used.

3.2.2. Biological Sampling and Analysis.

3.2.2.1. Quantitative Sampling.

a) Unit area of substratum. Quantitative results of benthic macroinvertebrates were required from the Rivers Tean and Ray and from the experimental channels. All were sampled in riffle areas having typically a loose and thin stone substratum all resting on a clay bed. Methods for sampling such stony streams have been discussed by Macan (1958) and Schwoerbel (1970). The choice of a cylindrical sampler was made on considerations of the type of substratum speed, and depth of flow. One was made in the Department workshops after a pattern of Neill (1938) but without the two sliding doors. This type of sampler was later recommended by Hynes (1971) for stony substrata, although the plastic skirt was absent in the model utilised.

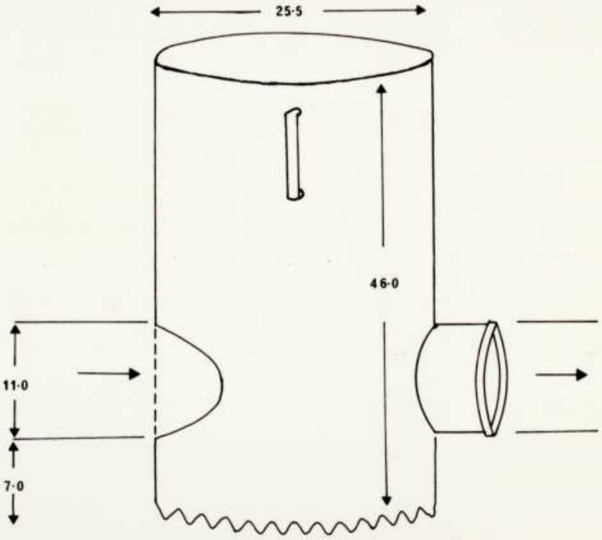
The sampler used (plate 7) was made of stainless steel for strength and possessed a serrated bottom edge which was driven into the substratum to a depth of approximately 5 cm (hyporheic fauna not required). Water passed through the meshed entrance (facing upstream) and washed into a net those animals displaced within the $0.05m^2$ area enclosed. The large stones were



EMERGENCE TRAP

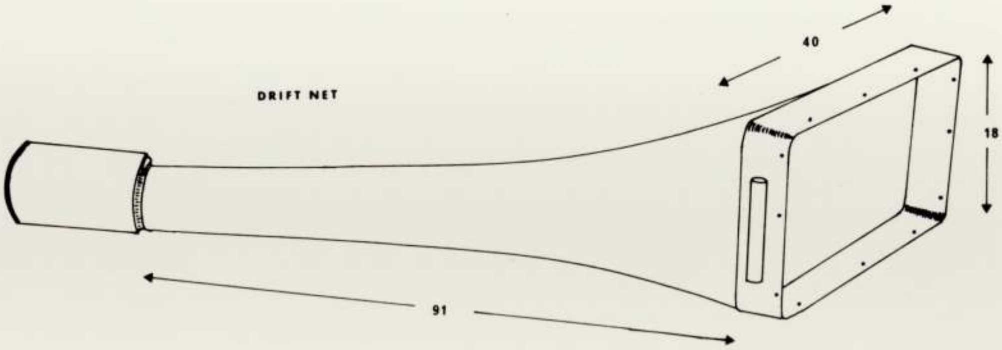
PLATE 7

MEASUREMENTS IN CM.



CYLINDER SAMPLER

DRIFT NET



first lifted and attached algae and animals removed (by means of a brush if necessary) and collected. The remaining smaller stones and silt were stirred with the fingers for about 30 seconds. It was supposed that most of the invertebrates had been dislodged by this time. The cylinder was then removed and the contents of the net tipped into a one-litre plastic bottle containing river water. Most animals would thus remain alive until they reached the laboratory for sorting. It was assumed that the net of 400 μ mesh apertures would hold all but the smallest stages of the invertebrates sought.

In the River Tean, Ray, and in the experimental streams, three cylinder samples were taken monthly at the various stations. Although three samples may be considered by some workers to be too few, the restriction on the number of samples was determined by the following:

(i) the impracticability of coping with the extra work involved in analysing a greater number of samples each month;

(ii) the animals would need to be sorted before autolysis - for accurate determination of calorific value (freeze drying before identification and counting is impossible).

(iii) careful selection of the three sample areas to obtain a representative sample could be practised.

The areas of the riffle zone in which the three samples were to be taken were chosen by eye to give as far as is possible a satisfactory representation of the whole. The proportion of bare and algal covered stones in the riffle (everything else being essentially equal) was visually assessed and samples placed for representative distribution of these or any other outstanding factors.

The artificial streams were sampled in a similar manner but, owing to the relatively large area of sample taken compared with the total area of riffle, markers were employed so that the same area would not be sampled more than once in six months.

The treatment of these quantitative samples in the laboratory was the tedious but most accurate one of sorting by eye. Flotation and screening procedures were abandoned, for more often than not the samples contained clumps of burrowed invertebrates. No substitute was found for the sorting of these animals out of filamentous algae by hand. Sorting of large samples was aided by elutriation overnight in a perspex trough when many animals were washed during dark into the collecting sieves. Individual samples were then narcotised for five minutes by a stream of CO_2 gas and tipped into a nest of three sieves of decreasing mesh width. A top sieve caught debris and the largest invertebrates. Successive sieves of 500 microns and 250 microns trapped the required invertebrates permitting silt to be displaced. A jet of water for approximately five minutes accomplished satisfactory differentiation. The contents of each sieve were in turn washed on to a white sorting tray divided into six equal parts. From three of these parts (always the same areas) were extracted all the fauna for identification, counting and weighing. When impractical numbers for extraction of one or more species were encountered, the sample was agitated by large air bubbles from a diffuser (to avoid bias of stirring etc.) in a beaker containing two litres of water. During "aeration" an unknown volume was quickly poured out on to the sorting trays and the extractions made. The volume was then measured and appropriate proportionation calculated.

Further treatment (dry weight, identification) is discussed in succeeding subsections.

b) Drift. It was decided that a knowledge of the diel invertebrate drift into and out of the three experimental streams would be useful for a number of reasons. Nets were designed after consideration of those parameters that would be encountered and those that needed to be measured. The problems of sampling relationships (Elliot, 1970) were reduced to simple volume - number and number - time determinations as the current velocities were constant and the same between each stream. The nets (plate 7) were held in position by brass bars driven into the experimental stream bed - plate 8. The mesh size was 42g.g.n., i.e. equivalent to 400 μ. mesh apertures, the size used for the cylinder sampler. It was assumed that this would withhold the invertebrates but let diatoms, other algae, and plankton escape.

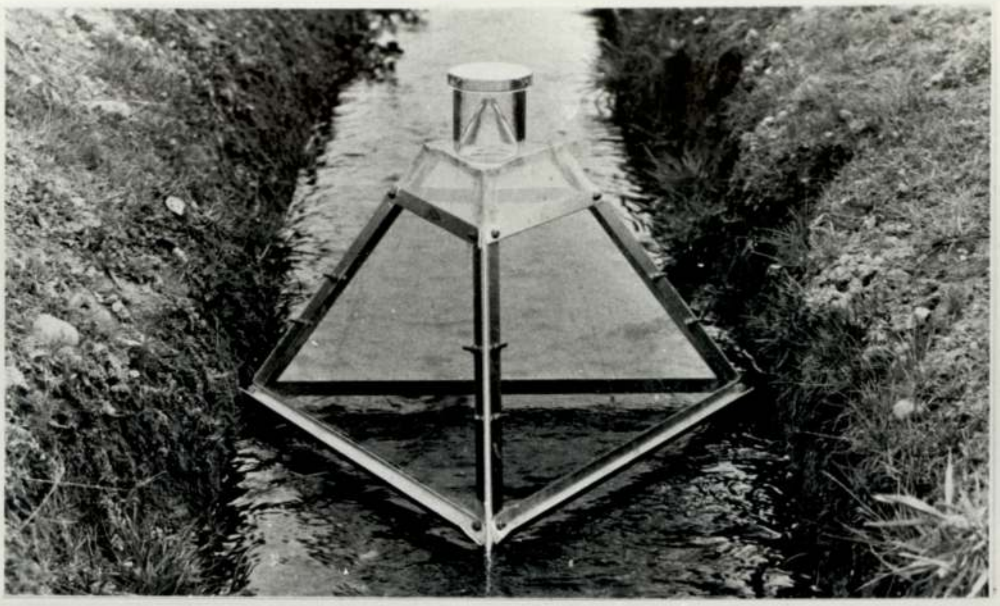
Nets were placed over the inflows to the streams (position x on plate 2) to catch all incoming invertebrates. They were also placed for 10 minutes at the beginning of the streams (y) to check the efficiency of the nets at (x) and at the end of the streams (z) to show the amount of drift from the stream alone. Sampling was executed on a 24-hour basis - each channel being netted every three hours. In the spring and summer there were huge algal blooms and especially blanket growths of diatoms in the upstream asbestos channels. The O_2 bubbles produced within the blankets would break off large amounts of algae during the day and would continue to lift off lumps during the night with diminished frequency. These were so large that within ten minutes of netting, the meshes were completely clogged and collection useless. To have used wider mesh would have ameliorated the problem but would have allowed a number of chironomid larvae and other invertebrates to escape. The diel drifts were abandoned after this spring attempt of 6 sample periods were made. The drift nettings that were undertaken (9/5/73) were sorted in a similar fashion as described for the benthic samples. No drift nets were used in the rivers.

c) Insect Emergence. Three identical emergence traps were built to suit the conditions and dimensions of the experimental streams. The streams were particularly suited to study of emergence because the water flow was constant, there being no fear of the trap being washed away. The design is after the pyramidal trap of Mundie (1964) but with a different specification (plates 7 & 8). Macan (1964) concluded that pyramidal traps were the most successful in stony streams. The frame was made from 3 mm. thick aluminium, two sides from 3 mm. perspex and the third and downstream side of nylon gauze (42 g.g.n.) The collecting vessel was made in one piece from glass and contained about 10 ml 4% HCHO. A petri dish served as a lid.

The bottom spars of the trap could be positioned within millimetres of the water surface thereby reducing the likelihood of catching those aerial insects that are carried in the air current set up by the running water. This meant that the imagines caught could be related to the area under its base. As the dimensions of the base area overlapped to some extent the bank edge, some of these Plecoptera and Neuroptera which crawl to the bank to emerge would have been caught. The area of substratum that the trap covered was 1.8×10^3 cm^2.

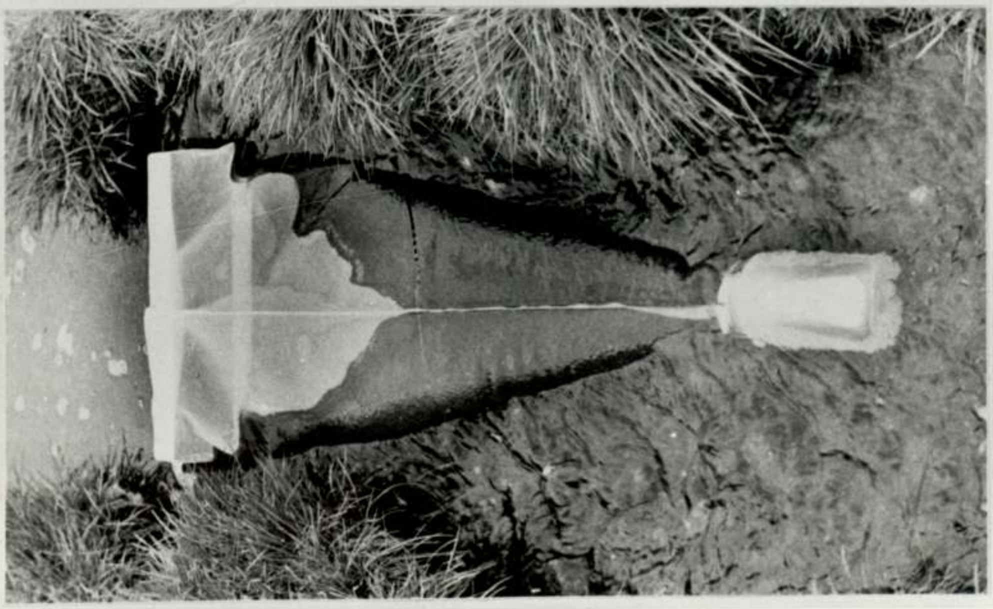
The traps were positioned in the three channels as shown in the photograph (plate 1). They were used both weekly and monthly (during emergence periods) for a period of 24 hours and then emptied. They were not used

plate 8



EMERGENCE TRAP

^
DRIFT NETS
v



when the drift nets were in operation (shown in conjunction in the photograph (plate 1) for convenience).

The identification of the imagines was for the most part taken to the family only. Numbers and biomass (dry weight) were determined. No emergence determinations were carried out in the rivers.

3.2.2.2. Qualitative Sampling.

Qualitative data (R. Cole surveys) was collected in terms of ranked data of species; the ranks being abundant, frequent, occasional, and rare. The method of trampling the substratum (heel sampling) upstream of a net for a fixed period of time was used. Surfaces of stones were examined and any fauna removed into a collecting jar. Three such heel samples were taken at every station, each lasting about 30 seconds. The contents of the net were emptied into a jar to give one composite sample for each station. No preservative was used unless the samples could not be assessed on return to the laboratory. Sorting was facilitated and assessment of ranks made on white trays.

3.2.2.3. Analysis of samples.

a) Dry weight. Samples were blotted on filter paper before transfer into pre-weighed and desiccated pieces of aluminium foil. These were then placed in a drying oven at $60^{\circ}C$ for 24 hours followed by cooling in a desiccator before re-weighing. This temperature was chosen to prevent volatilization or decomposition of fats thus avoiding erroneously low weights and calorific values. Reiners (1971) observed that the difference between various samples of tissue dried at 55°C and 100°C was only 2% and it was not certain if this error was caused by water or volatile organic weight loss.

Ash weights were determined in a muffle furnace at 500°C in pre-weighed nickel dishes. Samples were cooled in evacuated desiccators. Smaller samples for use in calorific estimations of ash free dry wt. were made in platinum pans.

b) Calorimetry. Calorific values were determined on most of the invertebrates encountered. There was not time to do more than three determinations per species or values seasonally.

Calorific measurements were made utilising a Phillipson type microbomb calorimeter (Gentry & Wiegert Instruments). Benzoic acid (certified by the National Physical Laboratory) was used as the thermochemical standard (26455.0 joules/g). It was found that the bomb needed recalibrating approximately every 20 determinations.

The samples were pelleted after drying at 60°C for 24 hours, Slobodkin and Richman (1960), in a pellet press following grinding in an agate pestle and mortar. Three pellet sizes were used per species - approximately

2, 5, and 10 mg. Weighings on tared platinum pans of 0.001 inch platinum foil were made on a Mettler balance to the nearest 0.01 mg. Some later weighings using a Cahn Electrobalance were made. Platinum fuse wire (0.0035" dia.) was used for ignition under 425 p.s.i. of O_2. The increase in heat was recorded on a lmv f.s.d. Leeds & Northrup recorder at 30"/hr. Pre and post heat change values were used in correction but no acid or electrical input correction factor was used. A number of runs using only fuse wire showed no such electrical correction was necessary.

Results were expressed as Calories/g ash free dry weight after muffle furnace estimation of % ash at 500°C to avoid volatilization of salts, Paine (1964) Reiners (1971), Richman (1971), and Grove (1961). This temperature and the same position in the muffle was always used.

When evidence of incombustion was detected, the determination was discarded and repeated. It was not necessary to use Benzoic acid/sample amalgams.

c) Taxonomy. The following keys were used in the identification of invertebrates: -

Plecoptera	- Hynes 1958.
Ephemeroptera	- Macan 1961, Kimmins 1954.
Trichoptera	- Hickin 1967.
Neuroptera	- Kimmins 1962.
Simulidae	- Davies 1968.
Crustacea	- Hynes *et al*, 1960.

Tricladida - Reynoldson 1967.
Oligochaeta - Brinkhurst 1963.
Gastropoda - Macan 1960.
Hirudinea - Mann 1964.
Chironomidae - Bryce 1960, Bryce and Hobart 1972, Chernovskii 1949, Mason 1968, and Thienemann 1954.
General - Macan 1959.

d) *Length/frequency analysis.* Analysis of this type was performed on populations of *Asellus aquaticus* from the River Ray at Moredon. The method is similar to that of Steel (1961) with ranking in terms of millimetre differences in lengths plotted against frequency. The dry weights, numbers and calorific values of these analyses were also determined.

Methods pertinent to the laboratory diurnal fluctuations and respirometric work are given later within the relevant chapters.

CHAPTER 4.

THE RECOVERY OF THE RIVER COLE FROM ORGANIC AND TOXIC POLLUTION.

It is convenient to discuss the recovery of the R. Cole with it sub-divided into upper and lower courses, the upper (Plate 5) involving the recovery from the Houndsfield Lane S.D.W. effluent and the lower (Plate 6), the effects of cessation of the toxic effluent from Yardley S.D.W.

4.1 THE UPPER COLE

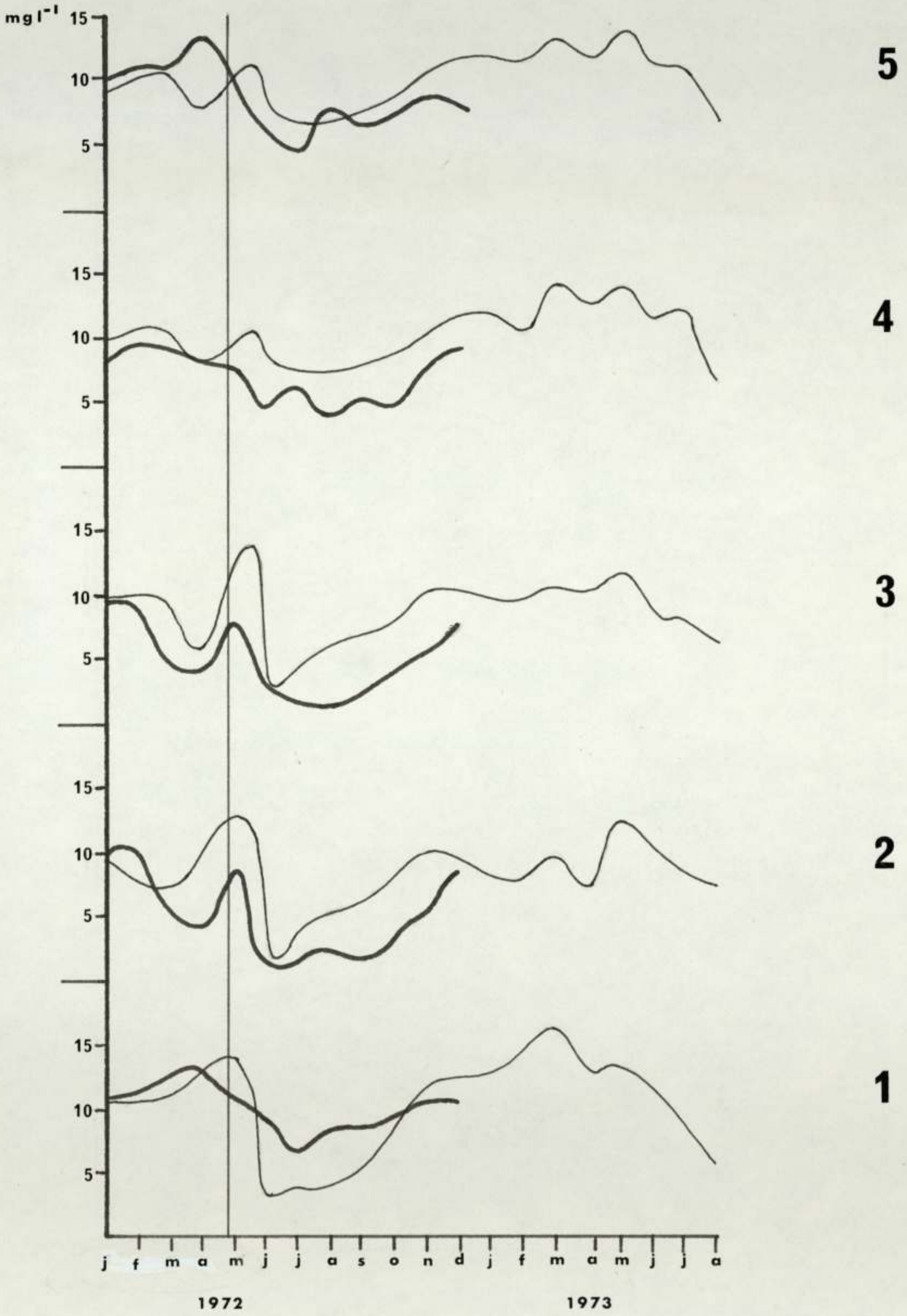
4.1.1. Chemical Data.

The results have been presented in two ways. Firstly as monthly data over the whole survey period, i.e. January 1972 - September 1973, which illustrates the seasonal fluctuations in water quality at each sampling station (Figs. 4.1 - 4.7). Superimposed over the period January - December 1972 are the monthly figures for the period January - December 1967 taken from Davies (1971) in an attempt to show how the water chemistry over the two periods has varied, not only with contamination prior to the closure of the sewage works in May 1972 but seasonally owing to meteorological factors. The vertical line represents the closure of the Works. Secondly, annual means calculated from data relevant to post-closure chemistry only has been displayed in histogram form (+ standard errors) against distance in kilometres from the confluence (Figs. 4.8 - 4.10). The data of Davies, although less comprehensive, has been re-calculated to give annual means and standard errors for the year 1967 and subsequently presented on the same histogram. These spatial graphs clearly demonstrate the changes in water chemistry downstream that have occurred as a result of the abatement of the effluent. The author's pre-closure data only covered a period of five months and, while this is seen to be similar to Davies's data for the same months, his data provided annual means. Both sets of data were gathered at the same sampling stations. Full tabulated chemical analyses are shown in Tables 4.8 - 4.12.

a) <u>Dissolved Oxygen</u> (Fig. 4.1) Inspection of the seasonal data shows the effect of the spring outbursts of diatoms and other algae at all stations. However, following this increase in oxygen saturation there is marked reduction between the months May and July in all years, none more so than at the 'control' Station 1 in 1972, the effects of which were manifest downstream to Station 5. This exceptional deoxygenation was anomalous, but nevertheless a trend of slighter proportions is shown every year during these months above and below the sewage effluent (cessated or not). The unfortunate pollution of the river above the Works in 1972 hindered the chemical and biological recovery for a number of weeks and was presumed to be the result of silage and general tip drainage above Station 1. Very high suspended solids were recorded at this time and the B.O.D. showed these solids to be organic in nature (Fig. 4.3).

At Station 2, before the closure of the S.D.W., both 1967 and 1972 figures show that vernal sloughing of 'sewage fungus' between January and May (at a peak during

FIG.4.1. The concentrations of dissolved oxygen at Stations 1-5 of the Upper Cole during 1972 and 1973 (thin line), and Jan. - Dec.1967 (thick line - from Davies,1971).



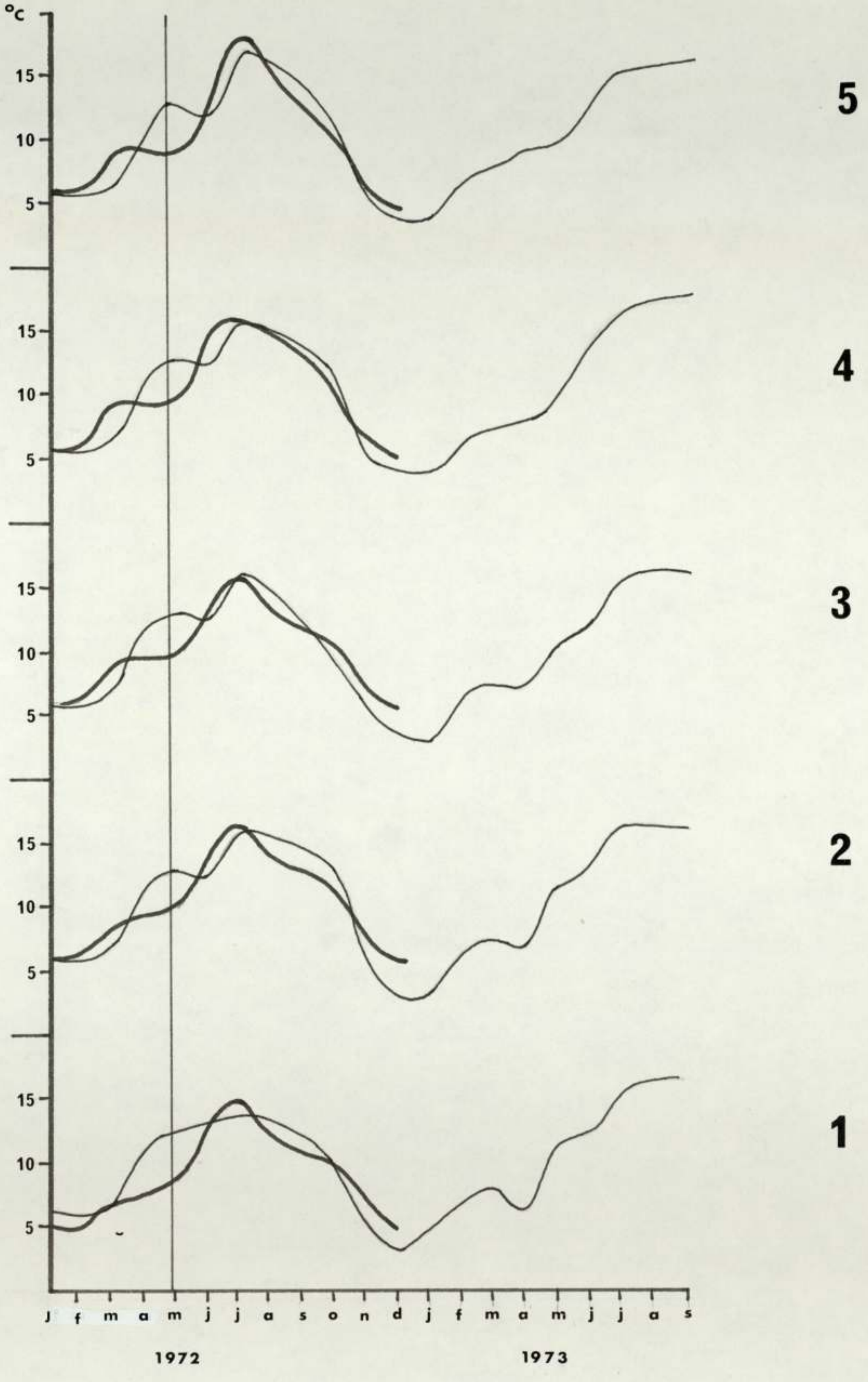
April, (Hawkes 1963a) like the suspended solids figures for 1972) keep the dissolved oxygen low despite well saturated water from algal blooms upstream. After the abatement of the effluent in May 1972, vast quantities of decaying 'sewage fungus' were noticed piled up against obstacles in the river, with B.O.D.'s and suspended solids remaining high until July/August. Increasing temperatures, causing gas release and chironomid emergence, loosened organic debris and resulted in diminished oxygen resources.

Similar seasonal patterns were seen at subsequent stations downstream but through dilution, self-purification and re-oxygenation were never as severe.

The spatial graph (Fig. 4.9) demonstrates however that, in general, the dissolved oxygen content at Station 1 is always very high and, although a 'sag' is evident even in 1973 means, the high oxygen content is maintained downstream. The 1967 'oxygen sag curve' shows the situation before closure of the S.D.W., very low levels being demonstrated at Stations 2 and 3.

b) Temperature (Fig. 4.2). Seasonal fluctuations are encountered with meteorological conditions accounting for the discrepancies between years. The warmer sewage effluent which is responsible for about half the flow at Station 2 is responsible for elevated temperatures there. The spatial graph (Fig. 4.9) shows this effect for 1967 when the river is maintained at slightly higher temperatures than post-closure averages (although meteorological conditions are probably partly responsible). The greater flow during 1967 would

FIG.4.2. The temperature at Stations 1-5 of the Upper Cole during 1972 and 1973 (thin line) and Jan.-Dec.1967 (thick line - from Davies, 1971).



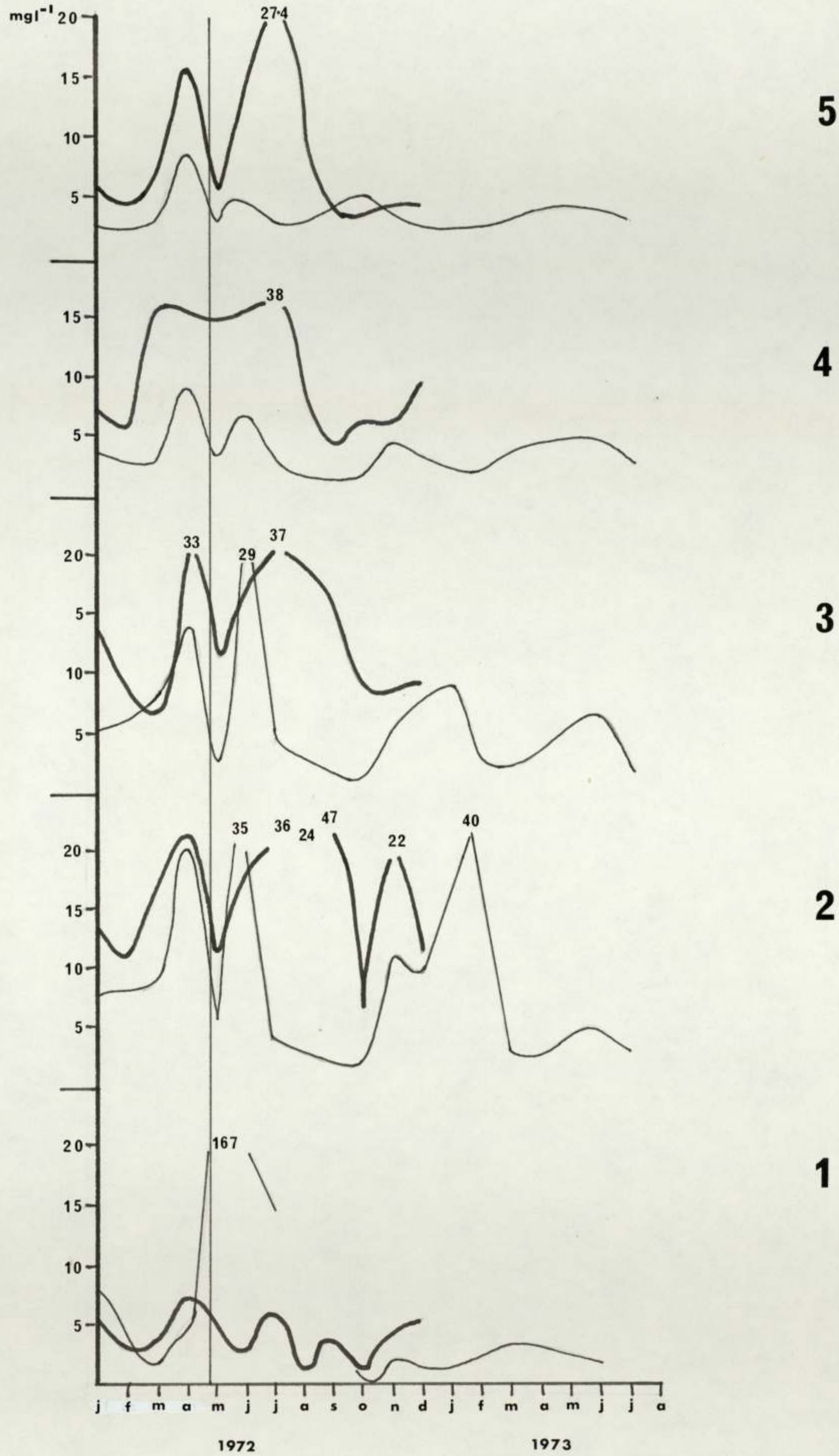
have tended to keep the temperature more stable, a theory substantiated by the larger standard errors at Stations 2 and 3 for 1973. The error bars are much the same at Stations 4 and 5 where the flow is similar to the pre-closure days owing to the supply of water from Peterbrook and Chinn Brook.

The inverse relationships shown between dissolved oxygen and temperature are smoothed out in a percentage saturation curve, a parameter that Hynes (1966) for some reason distinguishes as being more ecologically significant.

c) Biochemical Oxygen Demand. (Fig. 4.3). Comparing the pre-closure 1972 B.O.D.'s with those of 1967 exemplifies the similarity of organic loadings at Station 1 (between 1.5 and 8) which is low; Station 2 (7.5-23 mgl^{-1} B.O.D.) which is high; whereas Stations 3, 4 and 5 show considerably higher 1967 loadings. However subsequent de-oxygenation was only serious at Stations 2 and 3 where the flow is relatively small. In May 1972 the B.O.D. dropped right off only to increase sharply a few weeks later owing to fungal sloughing and heavy loading above Station 1 (167.5). After recovery in July/August the B.O.D.'s were comparatively low at all stations, with one exception of February 1973 at Station 2 ($>$ 40).

The spatial graph (Fig. 4.9) displays an inverse type of relationship with dissolved oxygen, with the effect of the effluent roughly doubling the B.O.D. at Stations 3, 4 and 5. Even in 1973 the B.O.D. at Station 2 is rather high (15.0) which may be attributable to the Houndsfield

FIG.4.3. The B.O.D. at Stations 1-5 of the Upper Cole during 1972 and 1973 (thin line) and Jan.-Dec.1967 (thick line - from Davies 1971)



Lane tributary (abundant in C. riparius) and the grazing of cows and horses on riparian farmland. After Station 2 the river enters urban areas and dilution of the B.O.D. between Stations 2 and 3 comes in the form of Peterbrook tributary and between Stations 3 and 4 from Chinn Brook.

d) Oxidised Nitrogen and Ammoniacal Nitrogen. (Figs. 4.4 & 4.5). Nitrates are a natural feature of most waters, with elevated levels being caused by agricultural run-off and sewage effluents. The data of 1967 is an analysis of nitrate content whereas the author's data is of oxidised nitrogen, the discrepancy between the seasonal plots for the two sets therefore being primarily one of nitrite. Nitrites are unusual in clean rivers, being intermediate products of oxidation of ammonia or reduction of nitrate. The lower levels of nitrates in 1967 at Station 2 and continuing downstream indicate incomplete oxidation of the very high ammonia levels in the effluent and no doubt much of the nitrogen is in the nitrite form. Only one set of nitrite figures is available for 1967: -

Station	10 a.m.	2 p.m.	6 p.m.
1	4.5	4.7	4.0
2	2.1	2.7	2.3
3	1.8	2.2	1.5
4	1.8	2.1	1.9
5	3.3	3.7	2.9

The low nitrates of March 1967 are presumably the result of infrequent functioning of oxidation processes at the S.D.W. (see March 1967 ammoniacal nitrogen).

FIG.4.4. The concentrations of oxidised nitrogen (thin line) at Stations 1-5 of the Upper Cole during 1972 and 1973, and nitrate (thick line-from Davies,1971) from Jan. to Dec.1967.

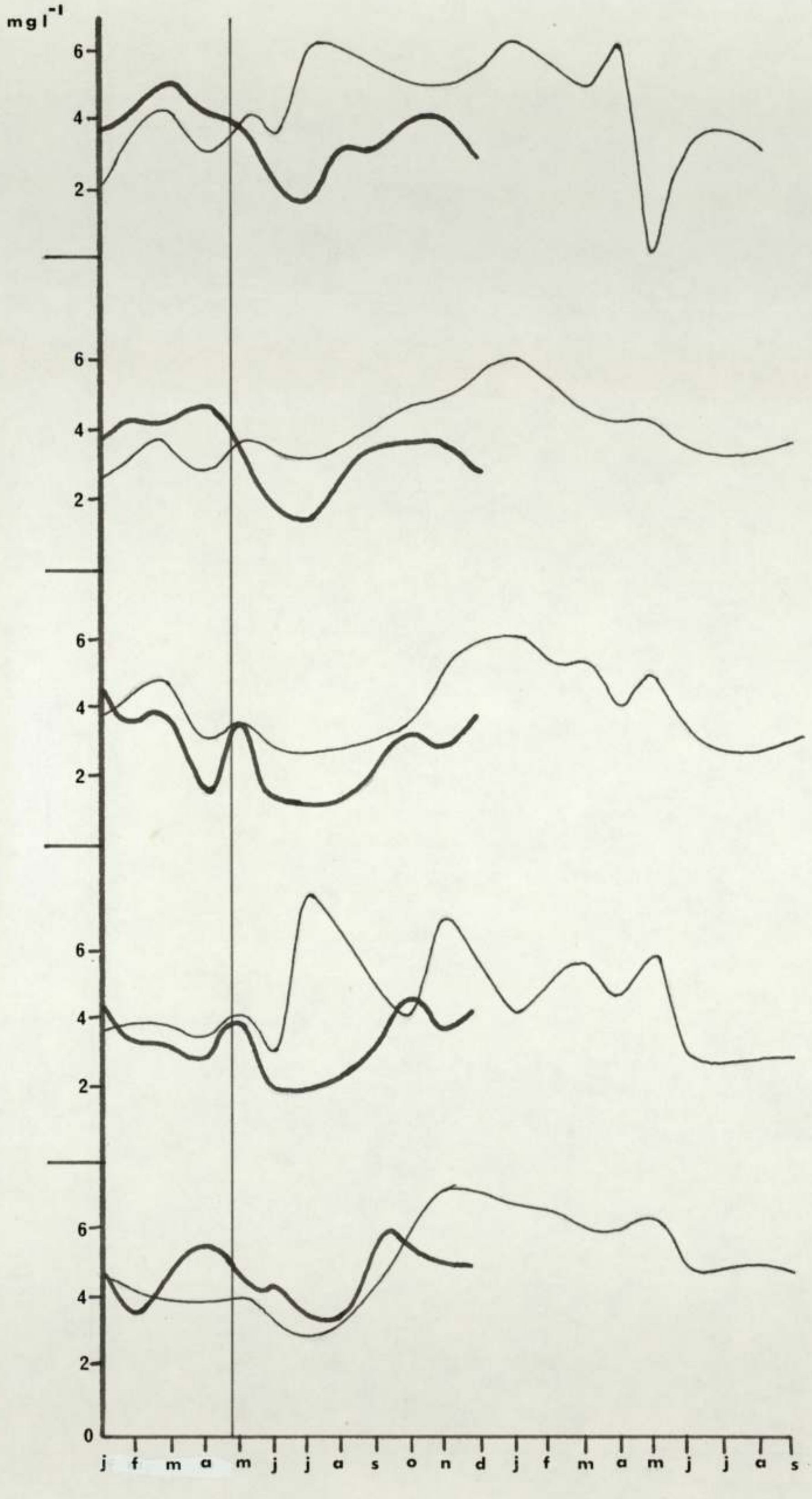
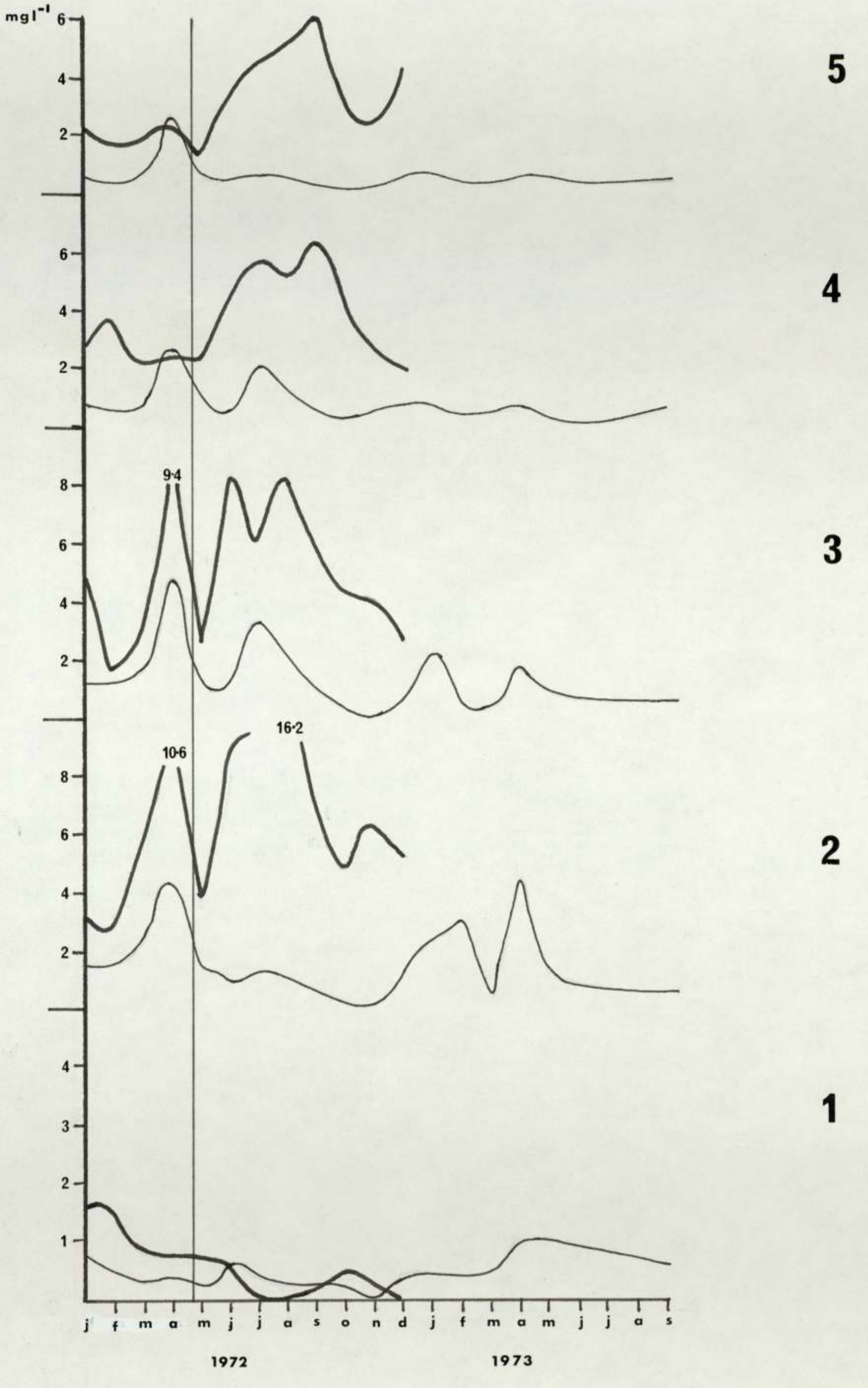


FIG.4.5. The concentrations of ammoniacal nitrogen at Stations 1-5 of the Upper Cole during 1972 and 1973 (thin line) and Jan.-Dec.1967 (thick line - from Davies, 1971).



In general, ammoniacal nitrogen varies inversely with the oxidised nitrogen figures, and after closure of the treatment plant the ammonia at all stations was low. The infrequent peaks in the early part of 1973 has been blamed on the tributary at Houndsfield Lane. The 1967 figures demonstrate how very toxic the levels were for most of the time - actual toxicity of course depending upon synergistic factors.

The spatial graphs (Figs. 4.9 & 4.10) visualise the low post-closure figures for ammonia and low nitrate for 1967, the latter being a reflection of poor nitrification of the effluent as are the high ammonia figures for the same period.

e) Suspended Solids.(Figs. 4.6 & 4.10). These are of two kinds in the Upper Cole. At Station 2 they are mainly organic (see B.O.D. and P.V.) and below Station 3 mainly inert as a result of many fords and river and sewer diversion work undertaken during the whole period. In particular, average solids figures are not very reliable when samples are only taken monthly.

f) Chlorides. (Figs. 4.7 & 4.10). No figures are available for 1967 and the considerable fluctuations could be attributable to a multiplicity of factors. The extremely high peaks in November and April 1973 are the result of road salting. Station 1 typical of spring water concentration and Station 5 similar to that of weak sewage.

FIG.4.6. The suspended solid concentrations at Stations 1-5 of the Upper Cole during 1972 and 1973.

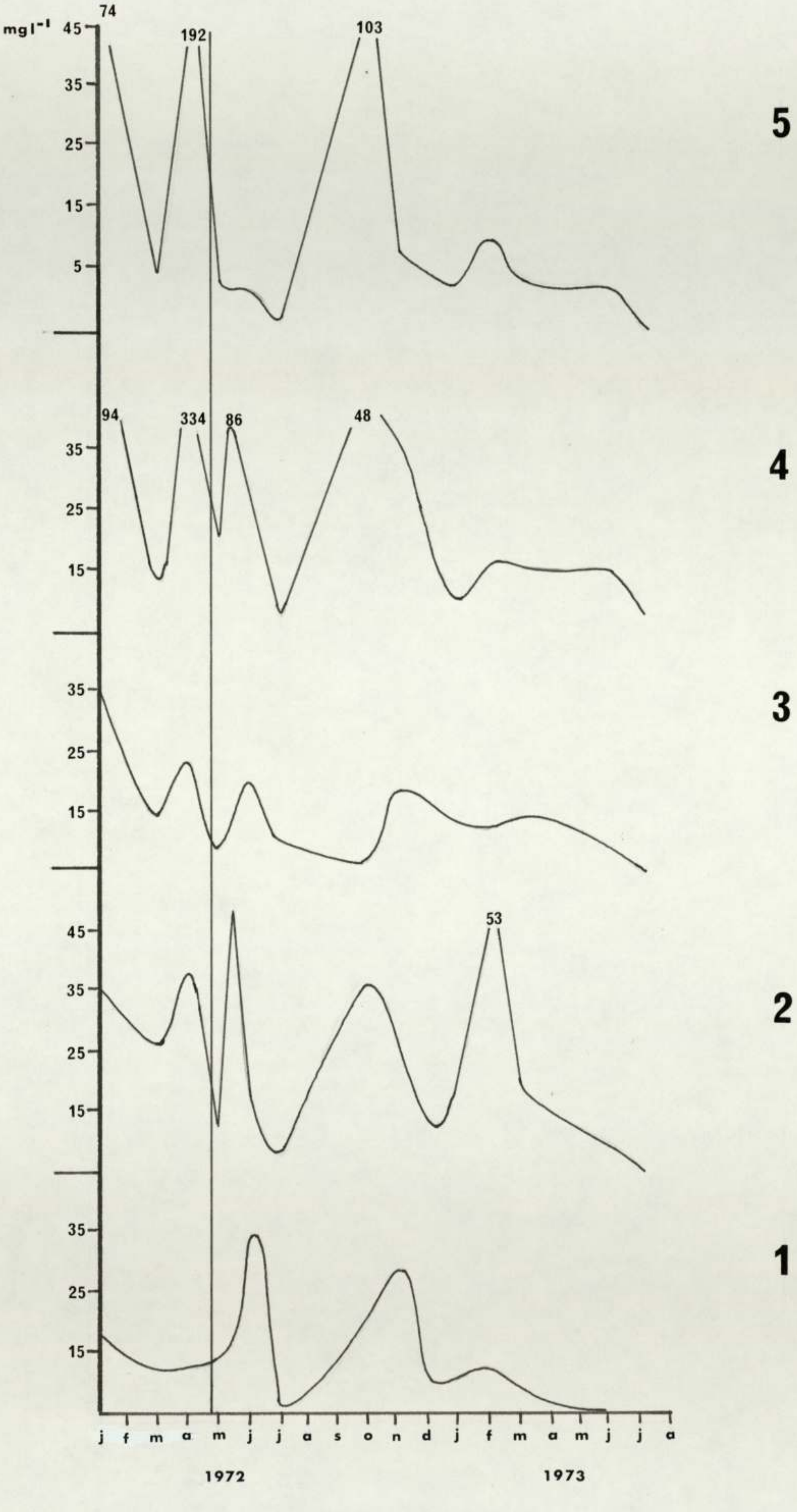
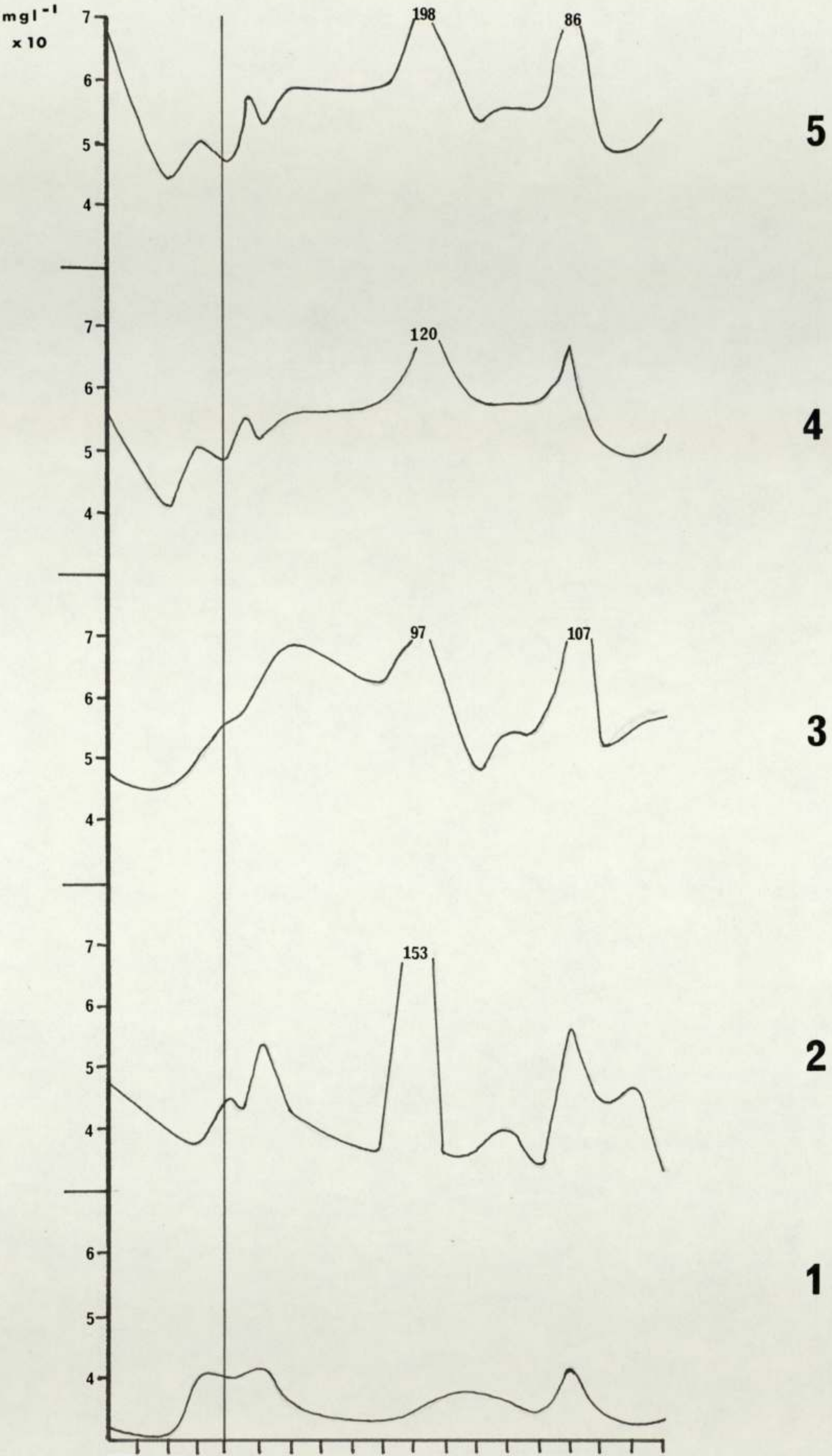


FIG.4.7. The concentration of chloride at Stations 1-5 of the Upper Cole during 1972 and 1973.



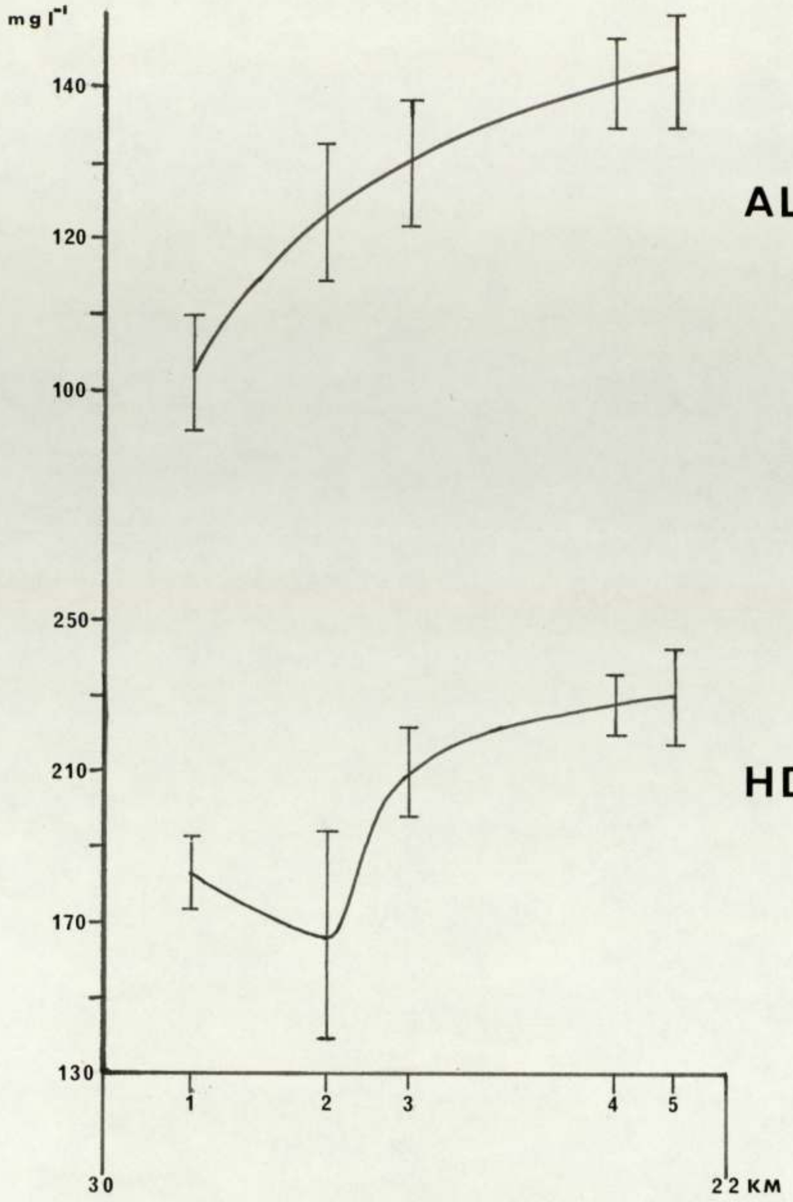
g) Total Inorganic Phosphorus. The spatial graph (Fig.4.10) shows high levels of T.I.P. even at Station 1, with agricultural run-off and silage escapes perhaps being responsible. Residual phosphorus releasing slowly from sediments below the old outfall may account for elevated values at Station 2. Cladophora appears below at Stations 3, 4 and 5 in increasing amounts and may be responsible for the uptake of some of the element.

h) Alkalinity. (Fig. 4.8) Again, no figures are available for the year 1967 but presumably the quantity of alkali present for this year at Station 1 may have been slightly higher than the present averages, knowing that oxidation of the effluent was poor. The spatial graph demonstrating increases downstream cannot successfully be pinpointed to any one factor like CO_2 utilisation or bicarbonate-carbonate shifts due to green plants.

i) pH. (Fig. 4.9) Slight pH rises occur downstream and may be associated with an increase in alkalinity and temperature. The water is very slightly alkaline making little or no difference to the toxicity of ammonia.

j) Permanganate Value (Fig. 4.10) Inspection of the spatial graphs for B.O.D. (Fig. 4.9) and P.V. suggests that the major source of pollution is organic as they are very similar in shape and probably possess a similar ratio downstream. The 1973 figures show how repurification and dilution reduces the P.V. from Station 2 downstream.

FIG.4.8.



FIGS.4.8.-4.10.

Spatial graphs of annual mean chemical and physical parameters of Stations 1-5 of the Upper Cole plotted against distance in km from the confluence. Standard error bars inserted. Heavy black lines (FIG.4.9) and nitrate (FIG.4.10.) indicate data of Davies (1971) for 1967.

ABBREVIATIONS:

ALK=alkalinity; HDNS=hardness; °C=temperature;
D.O.=dissolved oxygen; B.O.D.=biochemical oxygen demand;
NH_3=ammoniacal nitrogen; Cl^-=chloride;
S.S.=suspended solids; T.I.P.=total inorganic phosphorus.
NO_3=nitrate; P.V.=permanganate value.

FIG.4.9. See FIG.4.8. for details.

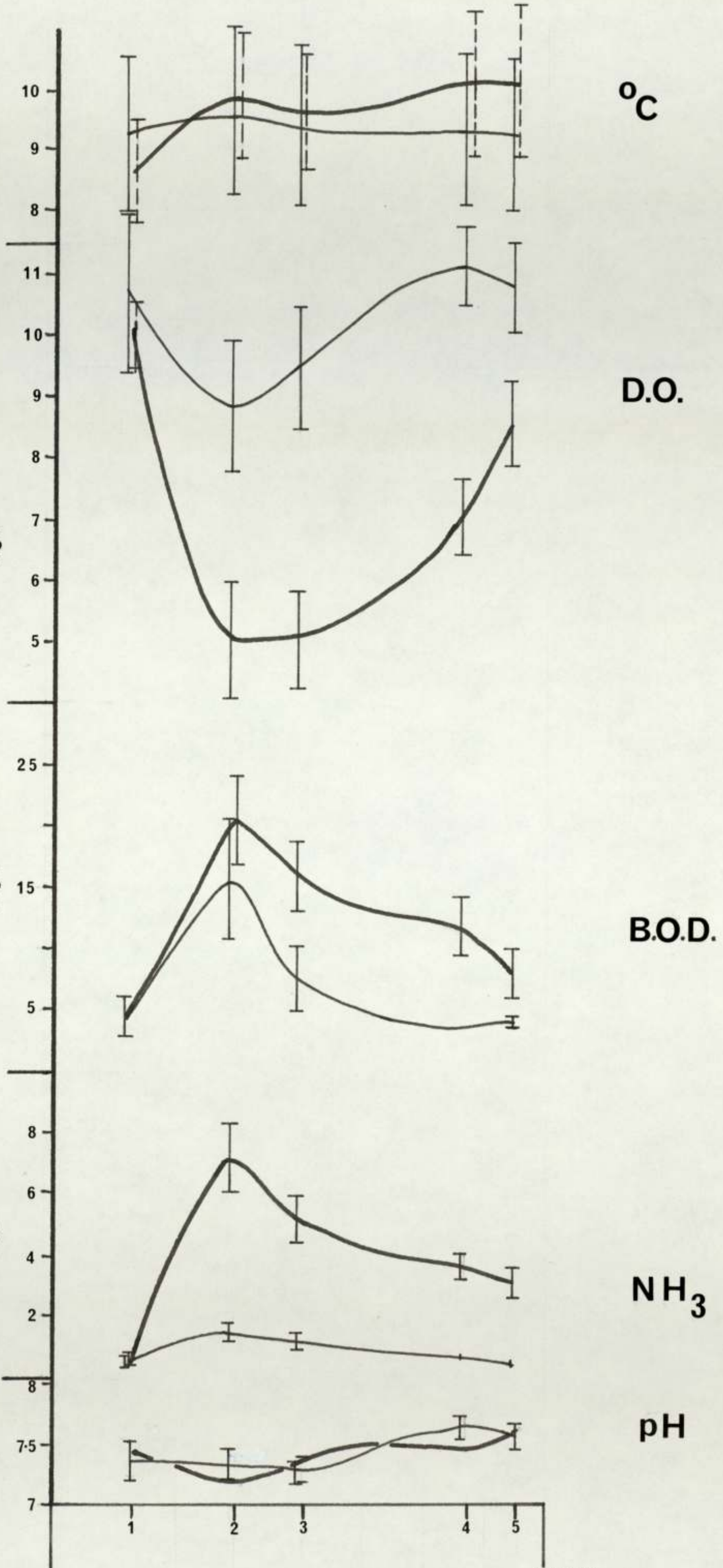
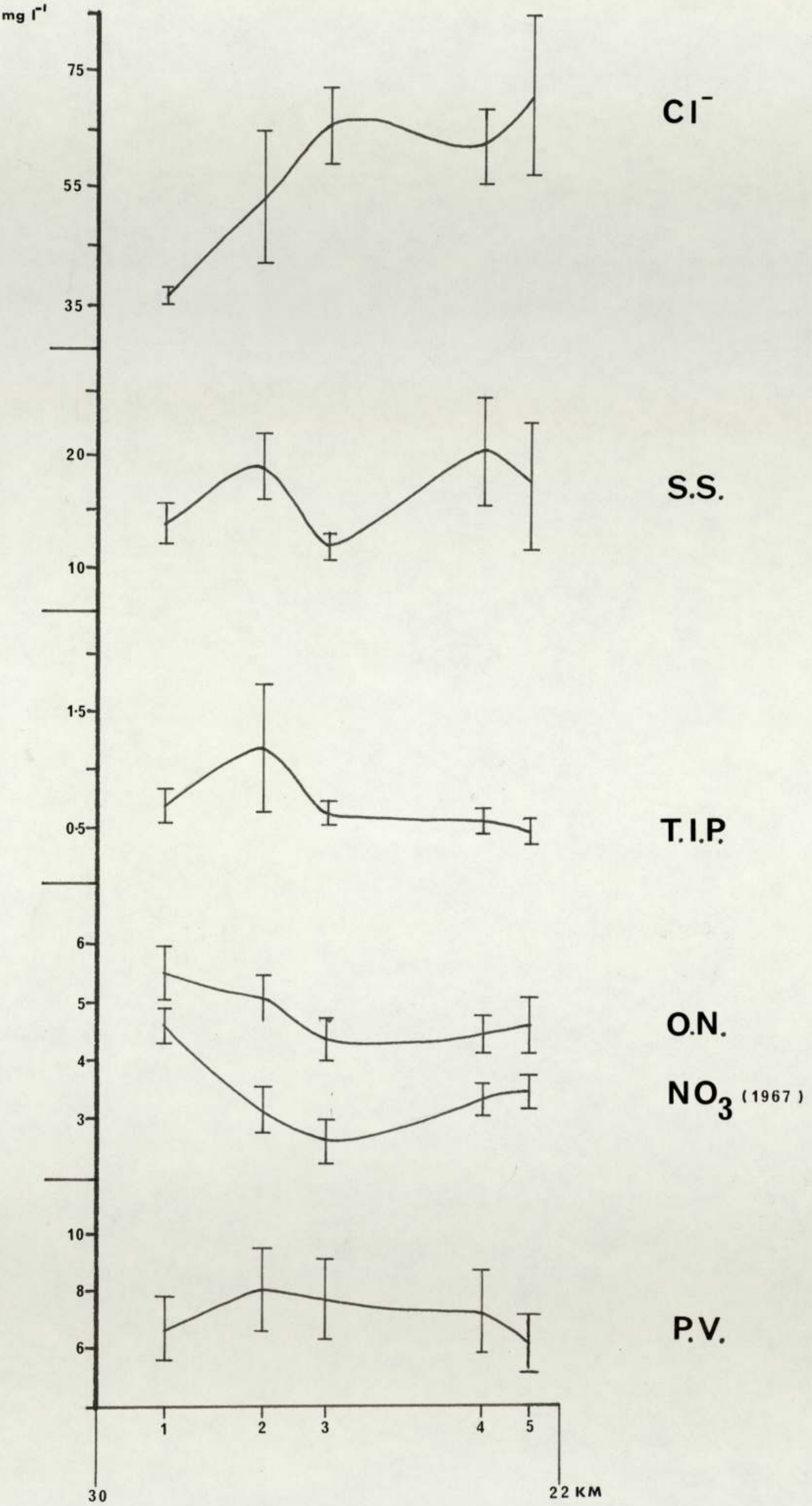


FIG.4.10. See FIG.4.8. for details.



k) Hardness (Fig. 4.8) The spatial graph illustrating hardness downstream is an exception in that it represents the period January - July 1972. Consequently the effects of the sewage effluent are measured. The R. Cole water can be considered to be moderately 'hard', which would be of considerable significance if metals like lead and zinc had been present in the effluent.

l) Metals. No lead or cadmium in a total or soluble form was ever shown to be present in the Upper Cole. Very small amounts of the metals chromium, copper, nickel and zinc were detected, typical examples of which are shown in the table below: -

Six Determinations of 4 metals taken in 1973 expressed as Total Metal in ppm.

	Cr	Cu	Ni	Zn
Station 1	0.03	0.03	0.04	0.07
	0.03	0.03	0.04	0.1
	0.03	0.03	0.04	0.06
	0.03	0.03	0.02	0.02
	0.05	0.02	0.04	0.04
	0.03	0.04	0.01	0.15
Station 2	0.02	0.01	0.01	0.02
	0.03	0.02	0.04	0.01
	0.04	0.04	0.03	0.09
	0.03	0.02	0.02	0.02
	0.03	0.02	0.05	0.04
	0.03	0.08	0.03	0.07
Station 5	0.01	0.02	0.01	0.03
	0.03	0.03	0.04	0.07
	0.02	0.02	0.02	0.03
	0.03	0.03	0.02	0.04
	0.04	0.03	0.04	0.04
	0.04	0.04	0.01	0.27

4.1.2. BIOLOGICAL RESULTS AND INFERENCES.

Brief History.

The biological history of the Upper Cole from 1950 onwards is fairly well documented, mainly as a result of the work of Hawkes (1956, 1963a) and also Hawkes and Davies (1970), together with some unpublished data collected by students of the former author, in particular Davies's work for the year 1967-1968.

Since 1950 a rich and varied fauna has always been reported at Station 1 although mild organic contamination as a result of small piggeries, a caravan site, and possibly silage escape have persistently prevented large representations from the Plecoptera, Ephemeroptera, and Trichoptera and maintained small numbers of Asellus and tubificids. The major source of organic enrichment (whose abatement was the primum mobile for the present work) was the S.D.W. at Houndsfield Lane which discharged into the R. Cole between Stations 1 and 2. The effluent in 1950 was well oxidised, so much so that "there was little chemical evidence of the cause of the milder biotic imbalance" which was reported (Hawkes 1963a) at Station 2 and whose condition was constant for the succeeding decade. However, in the early 1960's housing developments increased the load to the S.D.W. with the consequence that by the late 1960's the oxidative processes were overloaded and the ammonia concentration, suspended solids and B.O.D. of the effluent was high. Quantitative estimates of these parameters are reflected in the figures of Davies (1967) for Station 2 during 1967 (above).

The invertebrate fauna below the outfall was seriously altered, the effects being evident for some 7 km downstream, whereas between 1950 and 1959 the fauna was very stable and similar over this distance. The worsening effluent had by 1968 eliminated Gammarus pulex and suppressed the abundance of Erpobdella octoculata, Glossiphonia complanata, and Baetis rhodani to a few animals per m^2 over the 7 km to Station 5. Typically, conditions were favourable for compensatory increases in biomass of the Enchytraeidae, Tubificidae and many Chironomidae.

These early surveys echoed those findings of Butcher and Pentelow (1931), Percival and Whitehead (1930) and Gaufin and Tarzwell (1956) for organically enriched streams. May 1972 saw the diversion of sewage from Houndsfield Lane to Minworth S.D.W. and the subsequent biological recovery has been assessed over the same stretch of river.

PRESENTATION. The oligochaete systematics in the early documentation at most proceeded to the family and similar definition has been allocated to this freshwater class during the present study. Many chironomid larvae were identified to species level in 1967 following the work of Davies. Special attention has been given to the identification of Chironomid larvae because they were expected to play a major role in recovery, colonisation of the Experimental Streams (Chapter 5) and because of their potential as 'biotic indicators' of organic enrichment.

Those results that demonstrate some of the clearer patterns of recovery and change have been drawn in histogram form in an attempt to capture their nature quickly, but it must be pointed out that the time axis is in accordance with sampling dates and not necessarily linear (Figs. 4.11 - 4.26).

After the closure of Houndsfield Lane S.D.W. in May 1972, three samples were taken within one week of each other at all stations below but not at the control Station 1 above, where normal monthly sampling was maintained. As a result, Station 1 histograms are blank for the consecutive samples 24th May and 30th May 1972, which at first sight illude to the absence of species. These histograms (Figs. 4.11 - 4.26) are supplemented on their left hand side (stippled) by a summary of the situation in 1967 as reported by Hawkes and Davies (1970) and Davies (1971). Tables 4.1 - 4.5. give a full list of species and sampling dates.

RESULTS.

June 1972.

The biological recovery since May 21st, 1972 is somewhat complicated by the acute but disastrous organic contamination that occurred in late May and June above the control station and which affected all stations downstream. As a result the 'control fauna' at Station 1 were also suppressed, rendering changes due to seasonal factors harder to decipher. The 'background' Asellus population here was eliminated for three months along with Gammarus pulex, (Figs.4.11, 4.12), Baetis rhodani (Fig. 4.13) and the more sensitive leeches Erpobdella octoculata (Fig. 4.15) and Glossiphonia complanata (Fig. 4.16). This pattern was

Histograms showing the relative abundances of two species at 5 Stations of the Upper Cole. The horizontal axis represents months of sampling. Stippling = mean abundance for 1967-from Davies, 1971.
Fig.4.11. <u>Asellus aquaticus</u>
Fig.4.12. <u>Gammarus pulex</u>

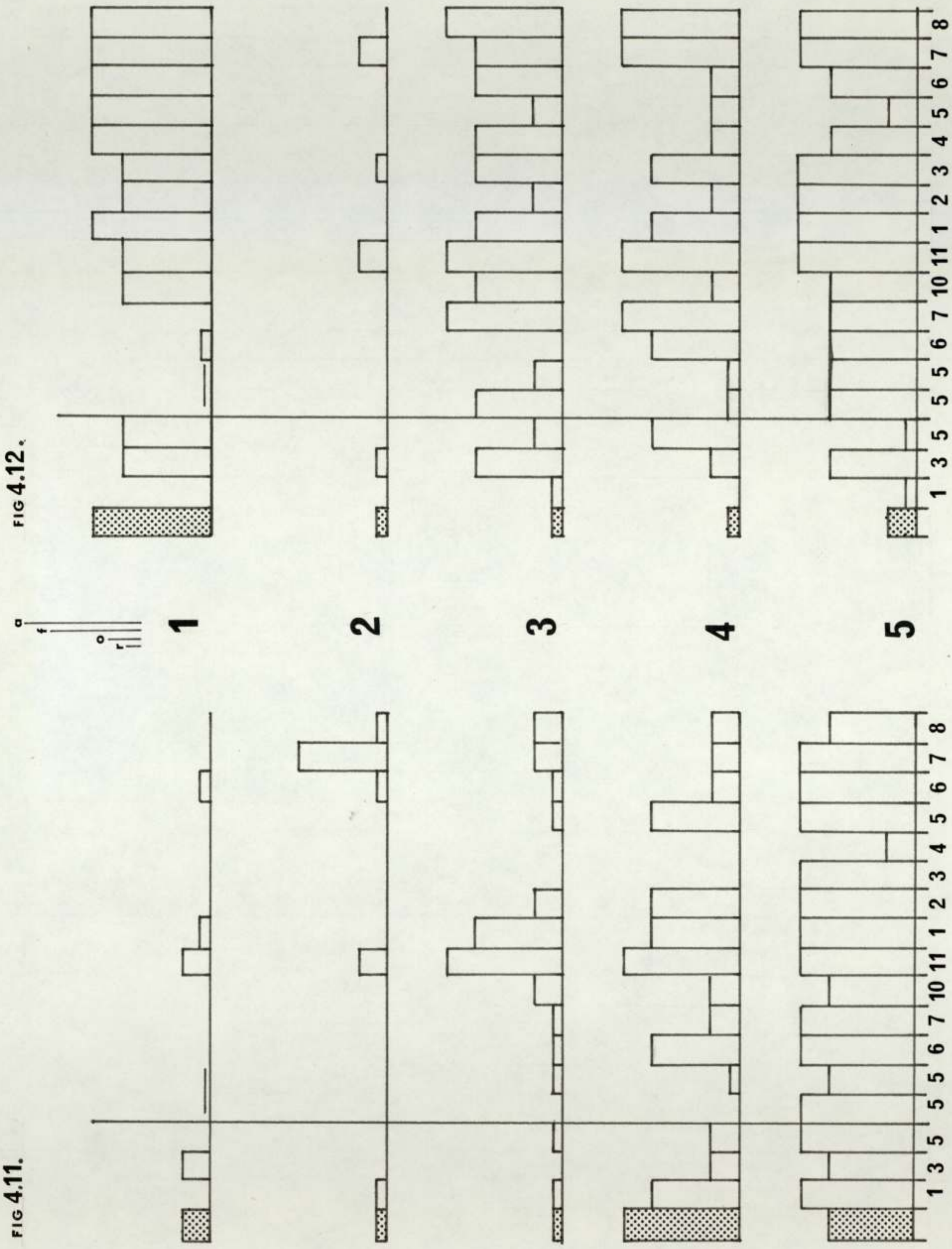


FIG 4.12.

FIG 4.11.

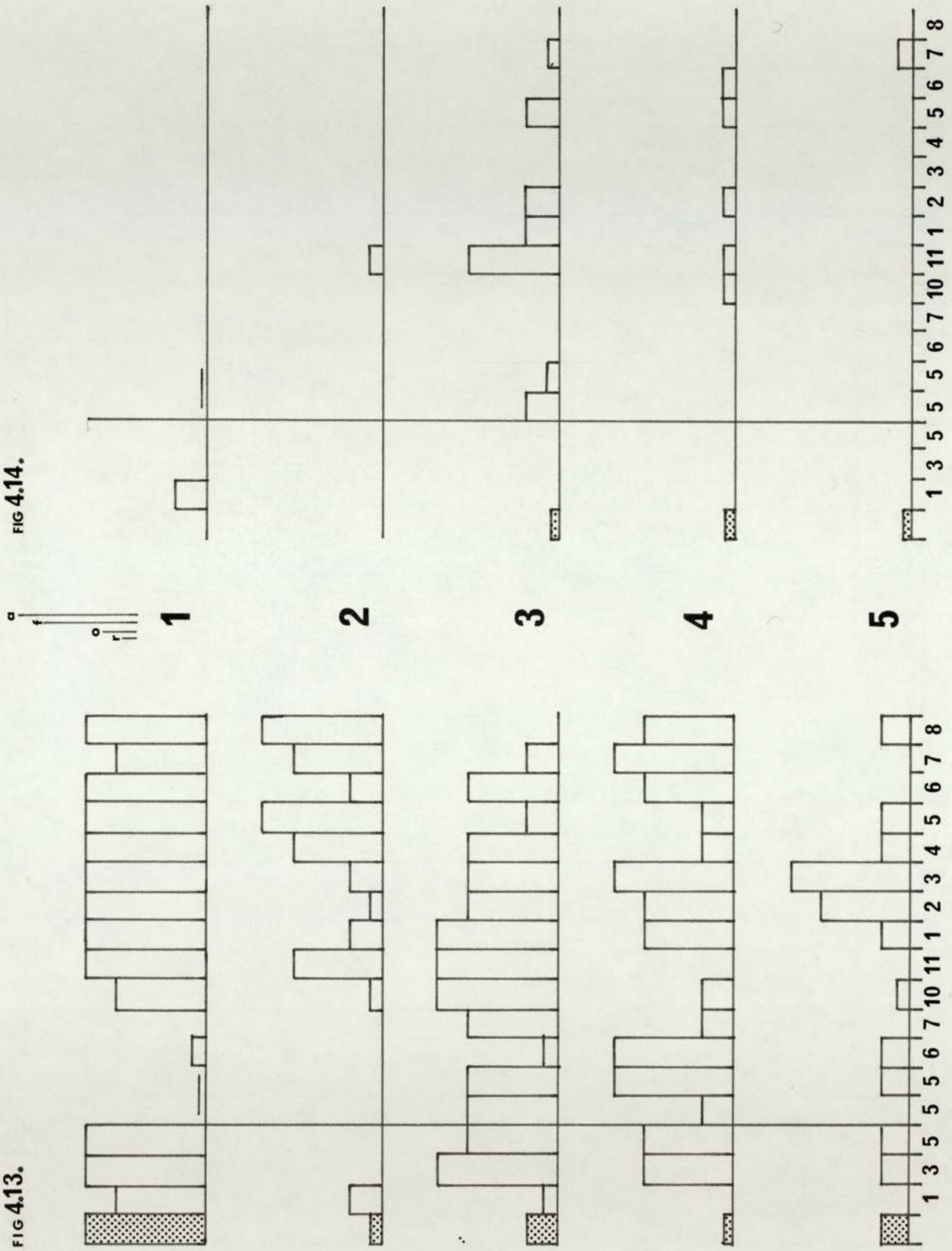
Histograms showing the relative abundances of two species at 5 Stations of the Upper Cole. The horizontal axis represents months of sampling. Stippling = mean abundance for 1967-from Davies, 1971.

Fig.4.13. Baetis rhodani

Fig.4.14. Lymnaea pereger

FIG 4.13.

FIG 4.14.



Histogram showing the relative abundances of two species at 5 Stations of the Upper Cole. The horizontal axis represents months of sampling. Stippling = mean abundance for 1967 - from Davies, 1971.
Fig. 4.15. Erpobdella octoculata
Fig. 4.16. Glossiphonia complanata

FIG 4.16.

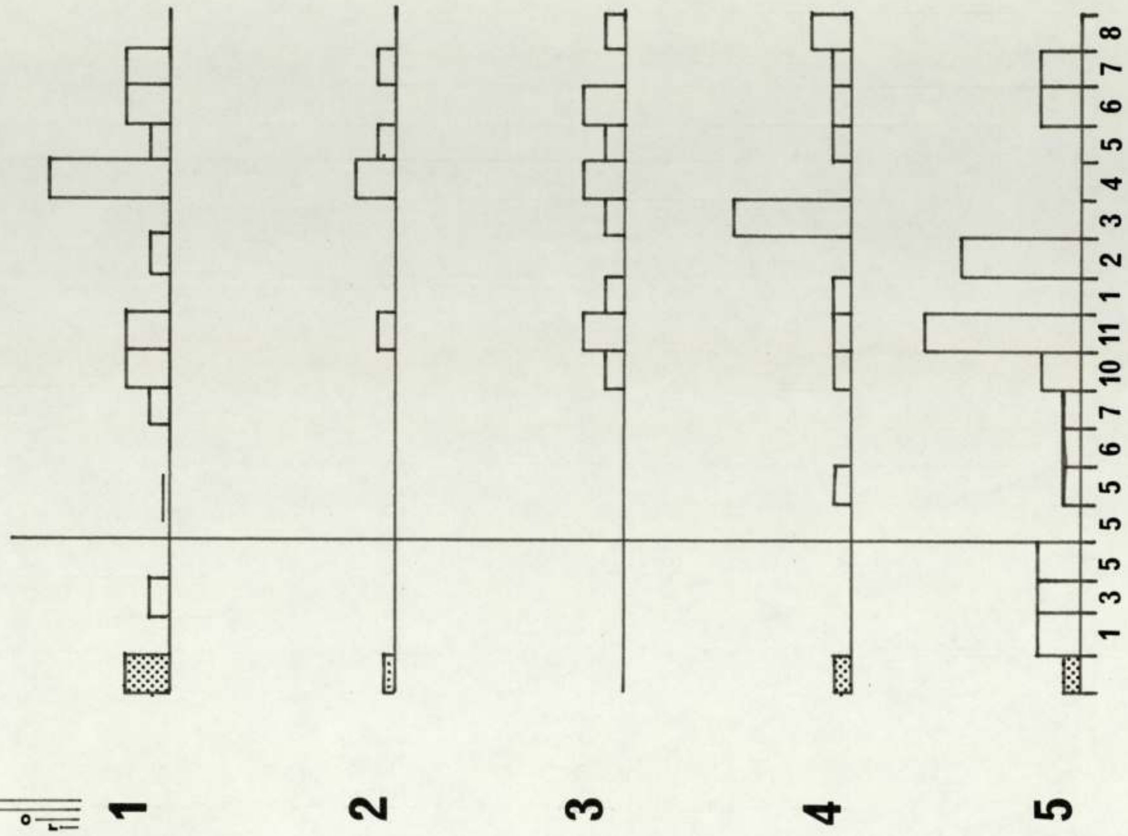
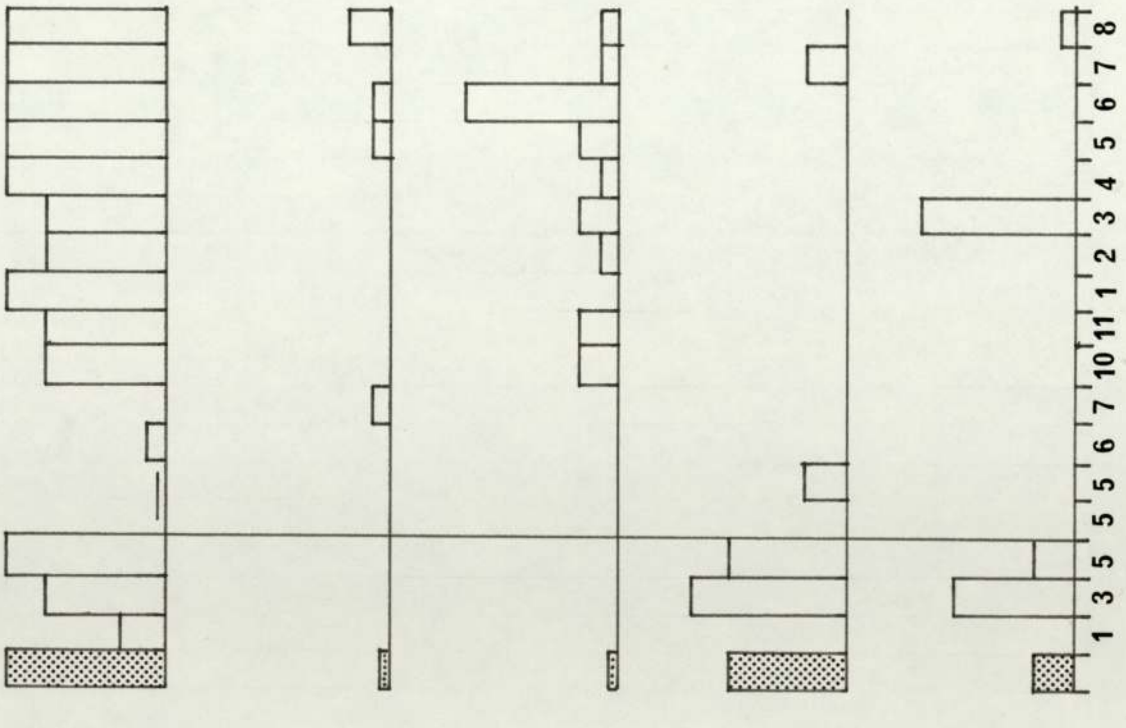


FIG 4.15.



Histograms showing the relative abundances of two species at 5 Stations of the Upper Cole. The horizontal axis represents months of sampling. Stippling = mean abundance for 1967 - from Davies, 1971.
Fig.4.17. Tubificidae
Fig.4.18. *Erpobdella testacea*

FIG 4.18.

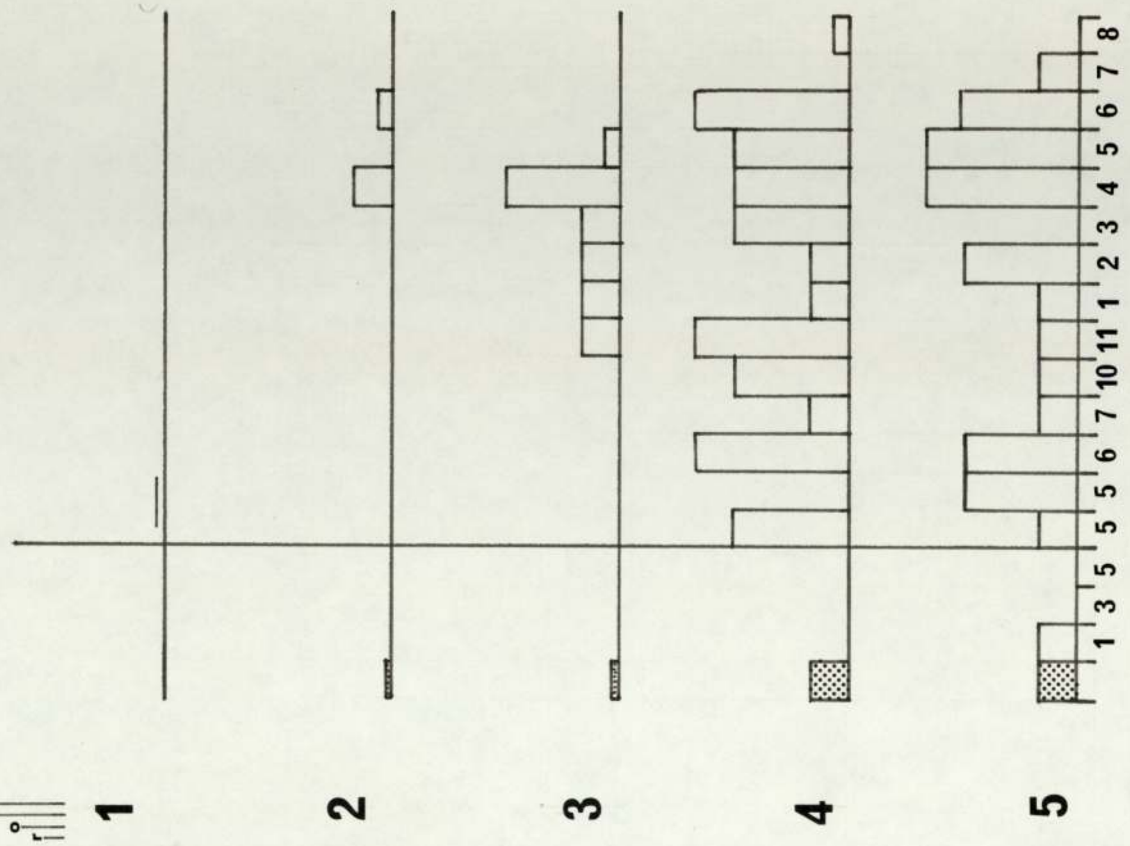
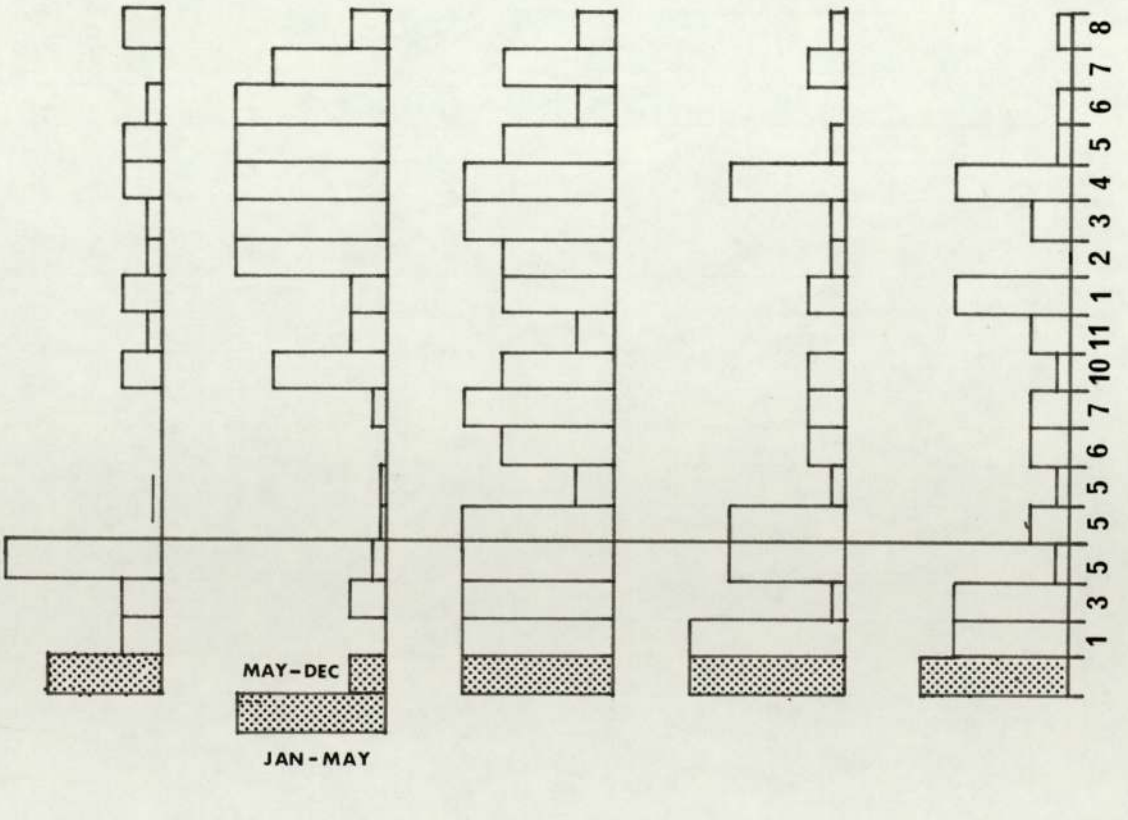


FIG 4.17.



Histograms showing the relative abundances of two species at 5 Stations of the Upper Cole. The horizontal axis represents months of sampling. Stippling = mean abundance for 1967 - from Davies, 1971.
Fig.4.19. <u>Chironomus riparius</u>
Fig.4.20. <u>Prodiamesa olivacea</u>

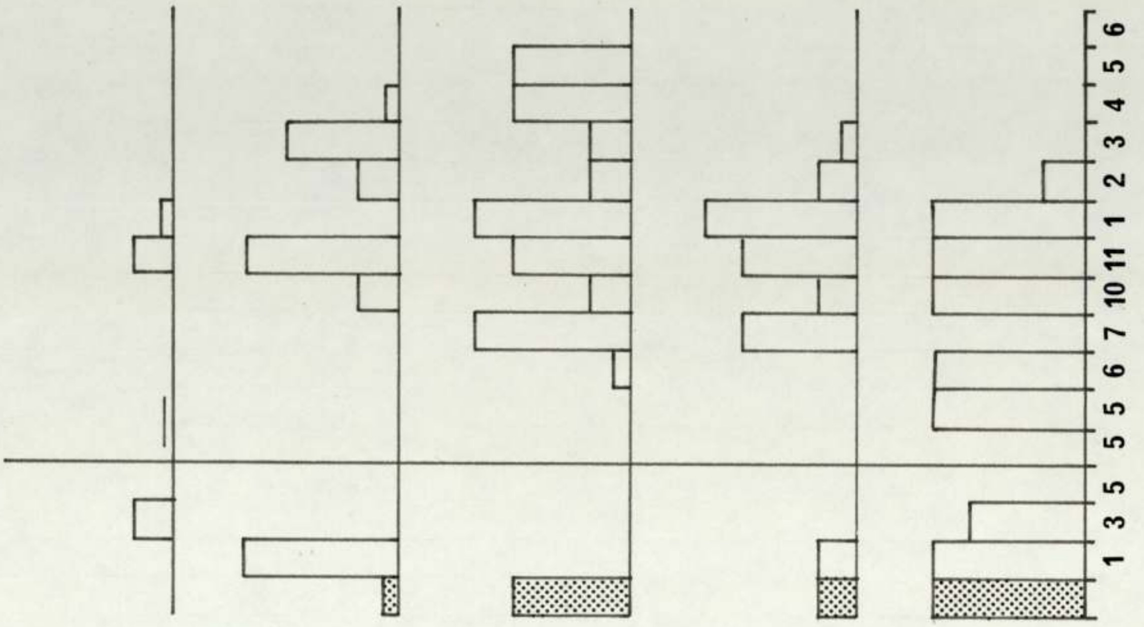


FIG 4.20.

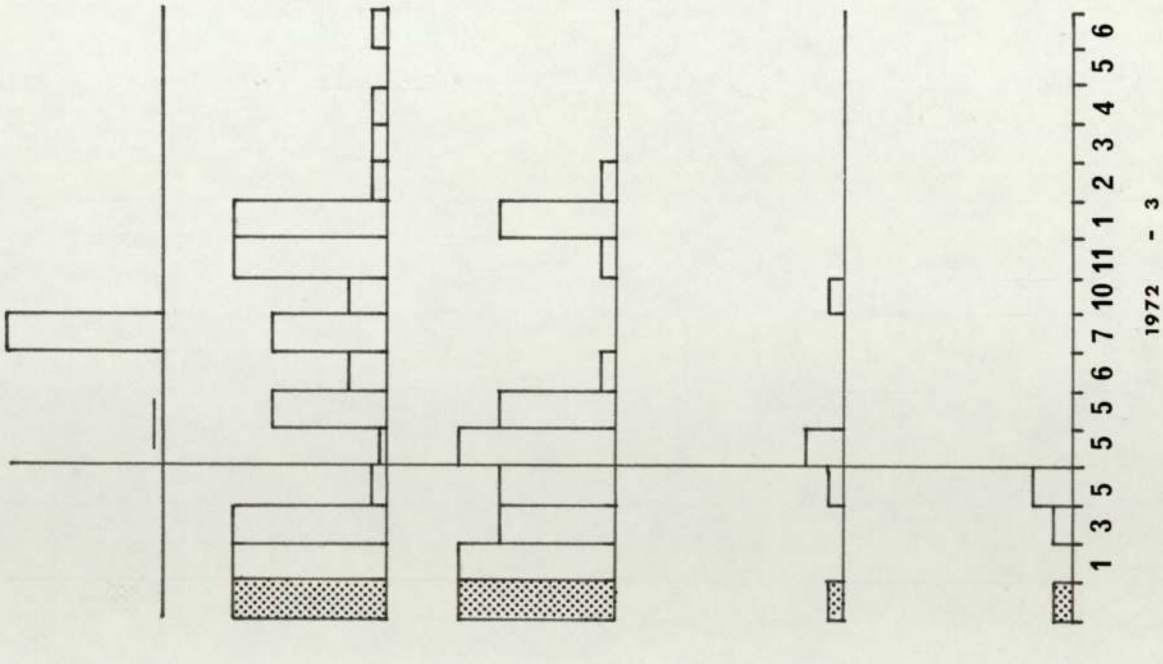


FIG 4.19.

Histograms showing the relative abundances of two species at 5 Stations of the Upper Cole. The horizontal axis represents months of sampling. Stippling = mean abundance for 1967 - from Davies, 1971.
Fig.4.21. Eukiefferiella hospita
Fig.4.22. Cricotopus (Cricotopus) ex.gr.bicinctus

FIG 4.21.

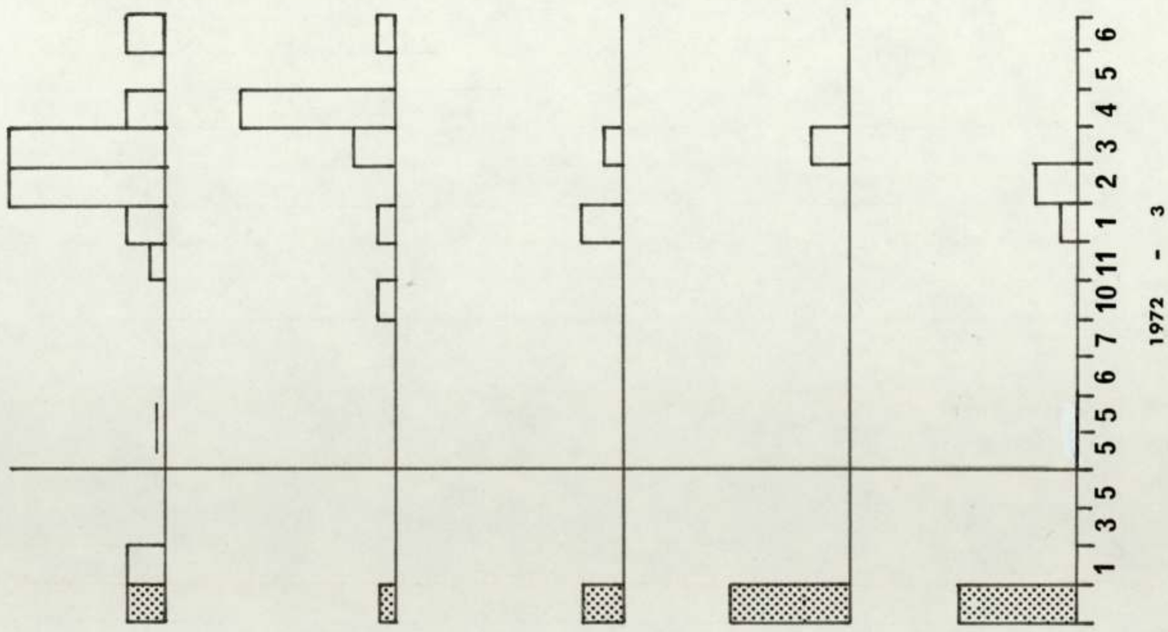
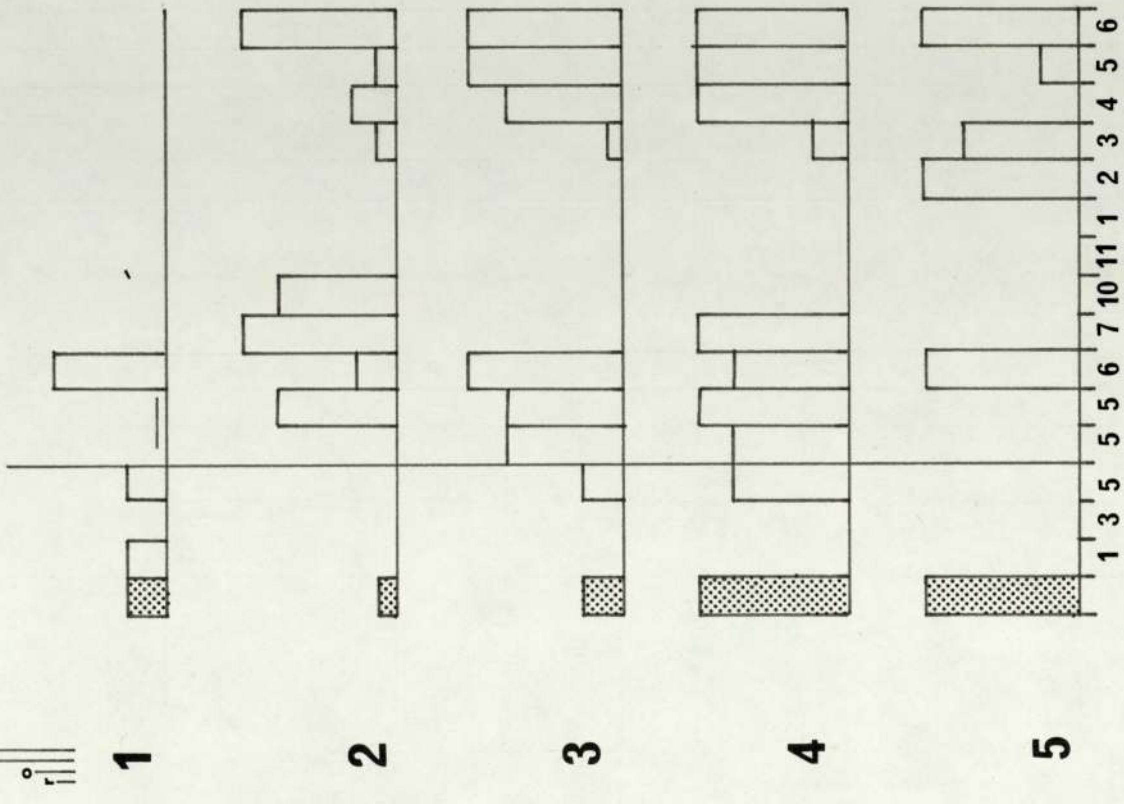


FIG 4.22.

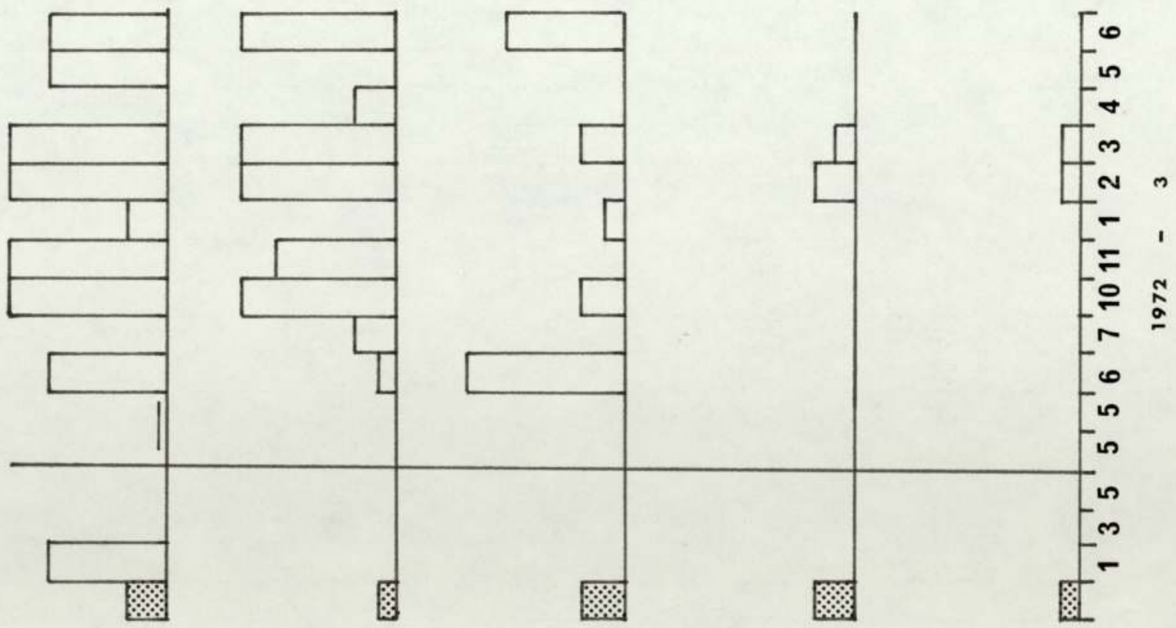


Histograms showing the relative abundances of two species at 5 Stations of the Upper Cole. The horizontal axis represents months of sampling. Stippling = mean abundance for 1967 - from Davies, 1971.

Fig.4.23. Micropsectra atrofasciatus

Fig.4.24. Cricotopus (Cricotopus) sp. albeforceps?

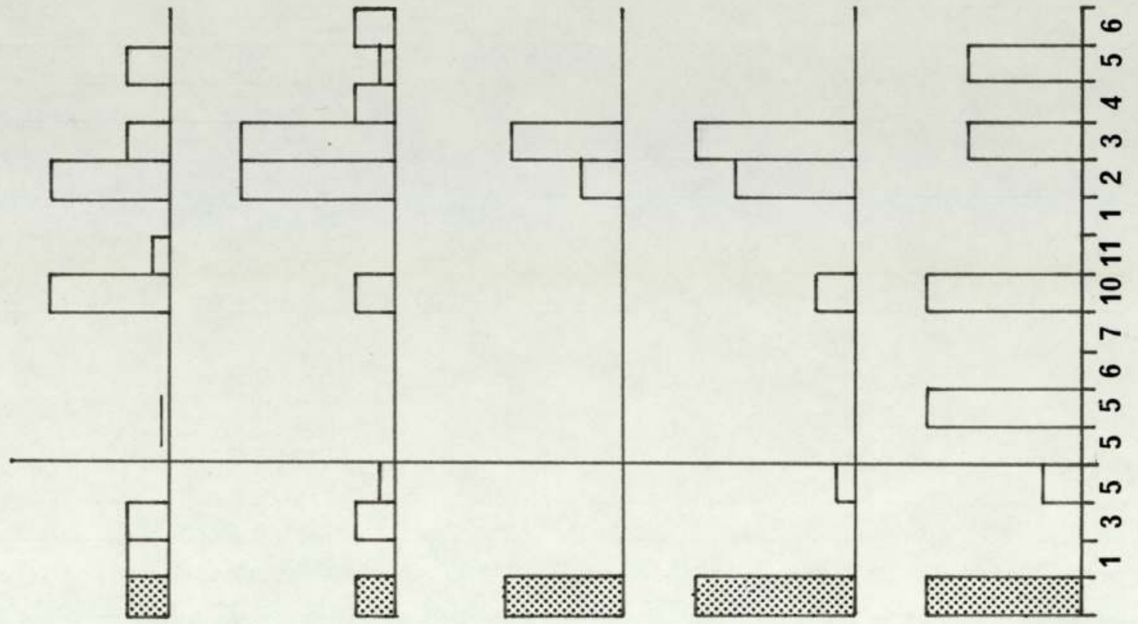
FIG 4.23.



a f o r

1 2 3 4 5

FIG 4.24.



Histograms showing the relative abundances of two species at 5 Stations of the Upper Cole. The horizontal axis represents months of sampling. Stippling = mean abundance for 1967 - from Davies, 1971.
Fig.4.25. <u>Brillia longifurca</u>
Fig.4.26. <u>Thienemannimyia ex.gr.lentiginosa</u>

FIG 4.26.

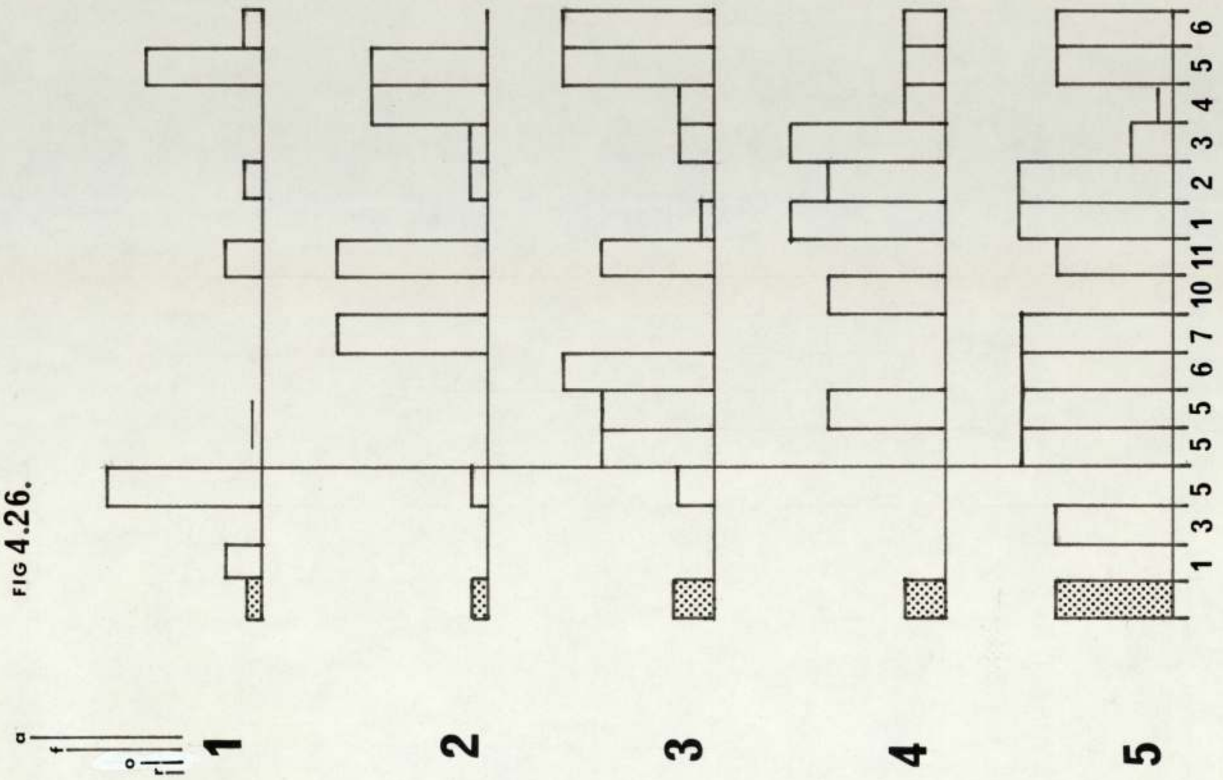
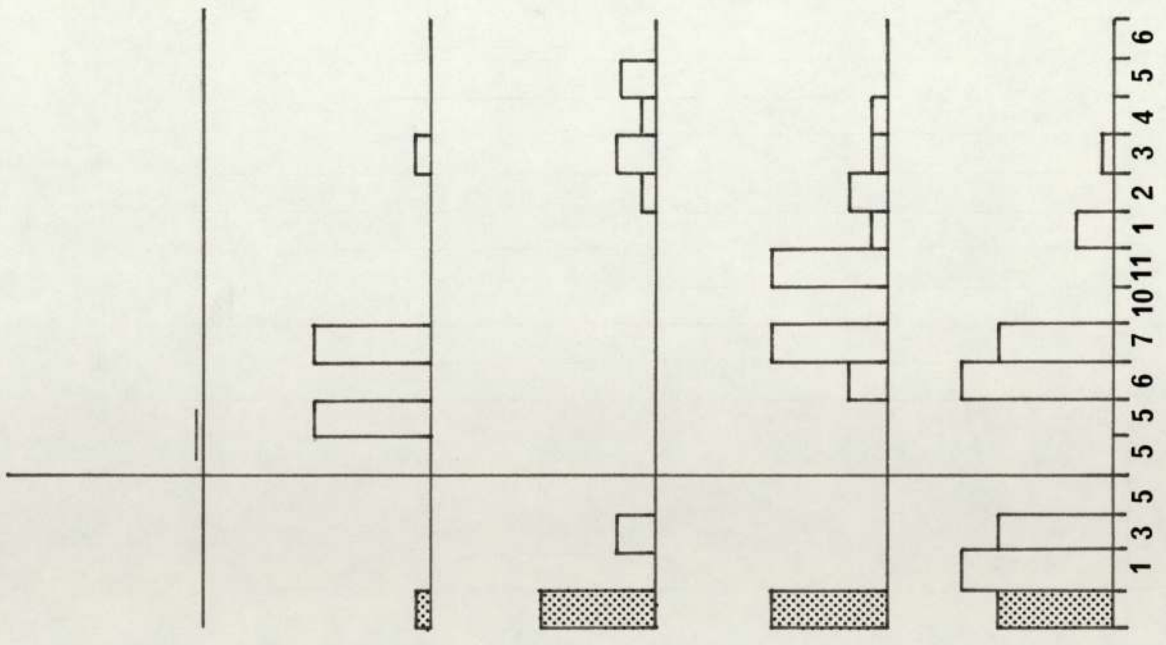


FIG 4.25.



duplicated at Station 2 (and 3 in the case of the Hirudinea) with reductions in numbers at Station 3 for at least two months and absence for one month. With the oxygen demand being very high at Station 1 during this acute pollution (B.O.D. > 167) even the tubificid population was removed and the increased loading encouraged large numbers of Chironomus riparius in an area from which they were previously absent (Fig. 4.19). However, the pollution demonstrates the short term effects of an 'acute' discharge on the benthic ecology with factors other than chemical ones prolonging the recovery. For example, the 'sewage fungus' cover was enormous at Stations 1 and 2 and thigmotactic and crevice living fauna would have been physically prevented from recolonisation until its breakdown. This breakdown caused severe daytime de-oxygenation and levels must have been even lower at night. Huge clumps of rotting 'sewage fungus' were seen at Stations 1 and 2 after July, large quantities already having sloughed off at Station 2 after May 21st but the acute discharge causing new fungal and bacterial growths in mid June. These growths receded and disappeared during August.

Amphinemoura sulcicollis suffered particularly badly, not returning until February 1973 (Table 4.1) although its seasonal absence appears to be in the autumn anyway. Hydropsyche angustipennis and Sphaerium corneum, although they have been described as fairly tolerant of organic pollution, were found surprisingly in small numbers in June and July. *Pisidium* was also rare in June and, as observed for milk wastes (Pentelow et al, 1938) decreased with the discharge. The chironomid larvae, where the pollution from this discharge was diluted at Stations 2 and 3 (B.O.D. 35 and 29), increased in species and in the case of *Prodiamesa olivacea* (Fig. 4.20), *Cricotopus bicinctus* (Fig. 4.22), *Brillia longifurca* (Fig. 4.25) and *Thienemannimyia* (Fig. 4.26) increased in number. A daytime determination in June showed the dissolved oxygen in the order of $1.5 mgl^{-1}$ at Station 2, and assuming the night time levels to be approaching zero it is not surprising to observe almost all the species eliminated, even Tubificidae, but with very small numbers of *C. riparius*, *Procladius sp.*, *Microsectra atrofasciatus* (Fig. 4.23) and *C. bicinctus* present. It is probably significant that the first three possess respiratory pigments.

Those sampling points (Stations 4 and 5) which were 5 and 6 km. downstream from the discharge were virtually unaffected, with the possible exception of *G. pulex* whose numbers should have been high following the spring brood.

The resumption of the status quo at Station 1 by the end of August demonstrates the speed of recovery from acute organic pollution and, having indicated some of the enigmas complicating and marring the main issue, the recovery from the chronic organic pollution will now be dealt with.

Station 1. The mild pollution evident upstream as noted in the historical review still exerted a selective pressure sufficient to prevent sensitive and varied representatives of the Plecoptera, Ephemeroptera, and Trichoptera from living and breeding. In this state it is reminiscent of a fauna in advanced stages of recovery from mild enrichment, and often short tufts of Cladophora are encountered during the summer. In the absence of the sensitive species of the above Orders typical of clean water faunas, the invertebrates were dominated by Gammarus, Baetis, Hydropsyche, Similium ornatum and leeches. E. octoculata was the dominant leech but G. complanata was well represented, having Sphaerium and Pisidium to feed upon. Helobdella stagnalis and E. testacea were never recorded.

The tolerant ephemeropteran B. rhodani has always abundant with the exception of the June 1972 pollution (Fig. 4.13) from which it recovered in force by the end of August. The tubificid population is likewise a feature of this Station as it was in 1967, but its abundance, although persistent, has always been of a low level. The isopod Asellus aquaticus is an infrequent visitor but a small yet ephemeral population developed from the June pollution only to disappear by February 1973 (Fig. 4.11).

It is suggested that the fine silt settling on the stones here prevents the presence of Ancylus fluviatilis, partly indirectly through its food source and directly as it is very sensitive to organic pollution. (Berg 1952 demonstrated its dependent oxygen relationship). The pulmonate Lymnaea pereger (Fig. 4.14) was altogether absent from January 1972 onwards but was never recorded previously, the unsettled nature of the current here not being ideal to its suggested requirements. (Percival and Whitehead, 1929).

C. riparius was collected once in July as a result of the June discharge, but otherwise all the known tolerant species were absent. The carnivore *Thienemannimyia* was persistent but *Micropsectra atrofasciatus* was the most abundant and dominant larva, together with *Eukiefferiella hospita* (Fig. 4.21) and to a lesser and more seasonal extent *Cricotopus* (*Cricotopus*) *albeforceps*?(Fig. 4.24). These three were all present in 1967 also. Meagre and seasonal populations of *Rheocricotopus*, *Diamesa campestris*, and *C. bicinctus* were conspicuous.

Station 2. From the changes indicated in the quality of the water by the chemistry, with particular reference to dissolved oxygen and ammonia, fairly profound and rapid biological transformations were to be expected in late 1972 and in 1973.

The abundance of *B. rhodani* by August/September 1972 well exceeded the 1967 population but the progress was hindered by the annual increase in B.O.D. during January 1973 (40, NH_3 3.1) causing small growths of 'sewage fungus'. Such growths cause Baetis to lose its habitat and they themselves become objects for growth which must be disadvantageous to survival. Consequently numbers were greatly diminished, and again in similar circumstances in June 1973 after a swift recovery of numbers (Fig. 4.13). Oxygen has been plentiful since August 1972 so it is the effect of toxic ammonia levels or the physical effect of 'fungal' growths. Lymnaea neither obtained a hold here after May 21st not had it ever been recorded beforehand (Fig. 4.14). The absence at Station 1 precluded the possibility of immigration even if the time span had been long enough for such a sluggish animal, and it has not been recorded at present in the nearer Houndsfield Lane tributary. Given time, it may be possible to invade Station 2 from the substantial population of Station 3 but this is regarded as unlikely owing to the annual growths of 'fungus' and instability of flow here.

In early October, three months after the closure of Houndsfield Lane S.D.W., a collection indicated a diversity more than double that of June with the appearance of Rheocricotopus, C. (cricotopus) albeforceps?, E. hospita and Prodiamesa olivacea in large numbers accounting for just under half the increase. One month later another six species (none chironomids) i.e. Sialis, Sphaerium, Lymnaea, Gammarus, Asellus and Glossiphonia, made an ephemeral attempt to colonise but the severity of the winter, high B.O.D. and growth of 'sewage fungus' prohibited any establishment until spring. Oddly enough, Simulium ornatum managed to retain a hold after its first appearance in October but its reduced numbers over winter and spring (Table 4.2) probably reflect the unsuitability of the substratum owing to 'fungal' growth, and S.ornatum is said to be most abundant in middle reaches of rivers being absent from extreme upper and lower reaches (Maitland and Penney, 1967). The river at Station 2 since the closure of the S.D.W. may be described as being in late upper reach. In Spring the chironomid larvae increased to exceed the number of species of the control station. Amphinemoura sulcicollis, a slightly less sensitive plecopteran, made an appearance for the first time along with Gammarus again. Although winter had caused increases in abundance of C. riparius they were recorded as rare, when present at all, from spring 1973 onwards. This is very different from the pre-closure situation, and a theme which was duplicated at Station 3 until March, from when it was completely absent (Fig. 4.19).

Asellus began to establish itself only after one year and recent observations (June 1974) by the author confirms their continued abundant presence. However, Gammarus only appeared intermittently in small numbers and disappeared altogether in winter. The conditions of winter, despite its relative mildness, caused growths of 'sewage fungus' on Gammarus and no doubt physical difficulties with the substratum decreased its chances of survival. Both Gammarus and Asellus were virtually at the same meagre levels of population as those that occurred intermittently during 1967-72. Gammarus no doubt was able to endeavour in its attempts of re-colonisation because of its greater mobility and presence upstream whereas Asellus is less mercurial notwithstanding its paucity at Station laand absence in the Houndsfield Lane tibutary.

The Tubificidae (Fig. 4.17) do not seem to have altered their pattern of abundance when compared with the pre-closure data suggesting that something here is favourable to their way of life but not those things that usually favour C. riparius and tubificids together, as the former is rare for most of 1973 whereas Tubificidae are abundant. Tubificidae are usually considered to be more tolerant to organic pollution than C. riparius so the relationship is strange. This is not to say that tubificids do not occur in natural waters, but such abundance would be very rare.

The remarkable numbers of chironomid species that have appeared at this station compared with those months before closure and with those of all the other stations (see histogram Fig. 4.27) must reflect the aerial advantage and physiologically adapted variety of this group as well as the potential of a virtually vacant biotope that Station 2 presented after May 1972.

Station 3. Despite similarly low dissolved oxygen tensions recorded for both Station 2 and 3, the latter has been subjected to less intense or saltatory movements with respect to suspended solids, B.O.D. and ammonia levels. This, coupled with greater dilution and a much increased hardness both as a result of the Peterbrook tributary, most likely explains the greater stability in number of taxa and biomass. Recovery in terms of diversity and biomass would therefore be expected to show less dramatic changes following effluent cessation.

Asellus and *Gammarus* were confidently established before the Houndsfield Lane effluent deteriorated but even between 1967 and 1972 were present, albeit tenuously. Both were removed with the acute pollution of June 1972 only to return, and in the case of *Gammarus* in vast numbers the following month (Fig. 4.12). *Asellus* recovered less rapidly but, presumably as a result of the larger autumn broods, were abundant by November 1972. Why the isopod should have failed to have contained a firm grip at the station is a mystery not accountable for by pollution as *Gammarus* was not affected, but may be as a result of the *Gammarus*/*Asellus* relationship which has often proved (for unsubstantiated reasons) to be one of mutual inhibition.

The way in which *Gammarus* has managed to establish itself in speed is probably a result of its mobility and presence upstream at Station 1 and in Peterbrook, migration from the latter possibly enhancing re-establishment in July 1972 when no *Gammarus* could have come from Station 1 or 2. Comparison with the 1967 situation shows how successful re-colonisation has been (Fig. 4.12).

B. rhodani shows an almost identical and parallel recovery pattern to that of Gammarus, this nymph having similar powers of movement and location in large numbers in Peterbrook. Eggs laid at the end of May would fortuitously have escapted the effects of the effluent and developed in October whereas those laid in October would not have developed until the following August, so the abundance of Baetis may be due in part to its pattern of reproduction.

Lymnaea pereger was never recorded at Station 1 except in January 1972 although it was a regular inhabitant of the three lowest stations up to 1959 (Hawkes 1964) until its elimination in the 1960's. It appears to have gained a foothold, though rather staccato, at Station 3 for the first time since its long absence. Physical conditions here are rather extraordinary with numerous large objects dumped in the bed, i.e. old bicycles and pushchairs, large bricks and pieces of piping, etc., a habitat which is attractive to Lymnaea in slightly silty conditions. The amount of silt cannot favour growths of epiphytic algae for Lymnaea to browse, although Mann (1953) has reported molluscs feeding on the cocoons of Erpobdella octoculata.

The attraction that lay in the slightly sandy and silted bottom before May 1972 was disclosed for tubificids when their abundance dropped and appeared patchy some months afterwards. It is conjectured that the decrease of organic matter washing downstream and being trapped between the stones caused the change, and Fox and

Taylor (1955) have reported Tubifex susceptible to oxygen poisoning in highly aerated water.

The most dramatic changes are demonstrated within the Hirudinea (Figs. 4.15, 16 & 18). Erpobdella testacea, the most tolerant of the erpobdellids, has inhabited this station where it is in competition with E. octoculata; the only Station where the two occur together in reasonable numbers. It seems as though the average organic loading during the recovery period (B.O.D. mean 7.5) is sufficient to keep the numbers of E. octoculata down with E. testacea then being able to compete more successfully and finally appropriating the habitat at Stations 4 and 5 (Fig. 4.18) with almost complete eviction of E. octoculata. The presence of large growths of Cladophora at the two downstream stations (rarely more than short tufts at Station 3) increases the populations of Asellus and chironomid larvae (orthocladinids) allowing the leech population food for vast explansion. Remarkable installation of E. testacea is demonstrated at the lower Stations downstream where, before the closure of Houndsfield Lane, the low oxygen and high ammonia no doubt precluded its establishment. Its population explosion at Stations 4 and 5 may be related to its breeding in March (Mann, 1961) and emerging from the cocoon in June onwards. With a now diminutive ammonia concentration, to which they are fairly tolerant, the free living leeches could establish themselves with no relapses in growth owing to the previously recurrent effluent or the acute discharge problems, whereas E. octoculata, having a two-year cycle anyway, does not begin breeding until June and July (Mann, 1953). The more sensitive *E. octoculata* were not reported until the end of August/ beginning of September and probably few managed to mate. Any offspring, which is unlikely, would then have substantial competition from the more numerous and by this time larger *E. testacea.* It is improbable that food was limiting *E. octoculata* here as it has the same diet as *E. testacea* (Mann, 1955), but Mann has reported a preference of *E. octoculata* for soft water in lakes and although we are dealing with lotic water here, Station 1, which supports a large population of *E. octoculata*, is soft and dystrophic whereas Station 3 shows a much increased hardness (refer to spatial graph, Fig. 4.8) and a very small population of *E. octoculata*.

The presence of *Glossiphonia complanata* is indicative of molluscan inhabitation. As far as is known, *G. complanata* has appeared for the first time at Station 3 since the mid 1960's. It has been recorded as scarce at Stations 2, 4 and 5 later than this in 1967. There has always been a small population of *G. complanata* at Station 1, probably maintained on *Ancylus* and bivalves, *Lymnaea* being absent, but at Station 3 when *Lymnaea* attains reasonable numbers and with occasional *Sphaerium* and *Pisidium* communities, the population of *Glossiphonia* is seen to increase. Small numbers of *Lymnaea* and *Ancylus* occur at Stations 4 and 5 where the *Glossiphonia* colony is well established. The increase in the numbers of molluscs at all stations since May 1972 has probably furthered the development of *Glossiphonia* communities.

A clear pattern of regression in *C. riparius* (Fig. 4.19) after years of abundance under the polluted regime is detected and, despite the furtherance of input upstream during June, conditions here are too good, even perhaps toxic (Fox and Taylor, 1955) causing its absence from July until October 1972 when it returns for the winter in small numbers. *Prodiamesa olivacea* does not share its exceptional tolerance to such severe organic enrichment and low oxygen tensions and can therefore thrive at this Station, the slightly silty nature of the substratum being particularly favourable. Nevertheless, *P. olivacea* is fairly tolerant to organic pollution and the population has always been described as frequent even in 1967. Its absence between January and May 1972 is thought not to be seasonal considering its presence at all the other stations (Fig. 4.20).

Brillia longifurca (Fig. 4.25) behaved in a similar manner to *C. riparius* and shares, to a lesser extent, its enthusiasm for high solids coupled with low oxygen tensions. Consequently, where once abundant at Station 3 before closure, its occurrence is now reduced to peaks of sparse numbers in February to May 1973 when vernal sloughs of 'fungus' and mild contamination produced B.O.D.'s of greater than 40 and elevated ammonia levels upstream (Figs. 4.3., 4.5).

Thienemannimyia, *C. bicinctus*, *Micropsectra* atrofasciatus, Polypedilum arundineti and Procladius spp. (choreus. Meigen?) have all made appearances of a substantial nature since May 1972 when, prior to the organic pollution, they were scarce or absent.

C. bicinctus occurs in two very distinctive seasonal peaks (Fig. 4.22) and in vast numbers at all stations below Station 1 during May and June. Observations made before May 1972 show only great scarcity and data of Davies echoes this even during May and June. Its rarity at Station 1, coupled with its seasonal abundance after improvement in the water quality and affluence at lower stations (4 and 5) before the latter, is suggestive of a mildly tolerant species typical of the 'recovery zone'.

Brief and inconspicuous appearances have been made by Rheocricoptopus (previously unrecorded), Eukiefferiella hospita, Brillia modesta, Cricotopus (isocladius)? spp. and C. (cricotopus) sp. albeforceps?

Station 4. This station is situated far enough downstream from the effluent(s) to permit it to enjoy the water quality of a river in advanced stages of self purification and recovery with respect to the invertebrate fauna. Chinn Brook enters the R. Cole just above this station affording dilution with water of fair quality. Even in 1967 the 'oxygen sag' had almost fully recovered, certainly to respectable proportions (mean 7.08 ppm, S.E. ± 0.68) and for the year 1973 the river had on average a higher dissolved oxygen concentration than Station 1, with a percentage saturation only 5% less than the control station as a result of the latter's consistently lower temperature.

The state of recovery reached was typified by the marked increase in Cladophora and Asellus. The Cladophora, both before and after closure, was typically present in small tufts throughout the winter and blooming in June until October in lengths up to 2 - 3 ft. but never reaching the proportions of percentage cover attained at Station 5 below. Cover at Station 4 could be as much as 60% in summer. Alkalinity and total hardness had reached 140 mgl^{-1} $CaCO_3$ and 227 mgl^{-1} $CaCO_3$ both becoming asymptotic; ammonia concentrations and B.O.D. were much reduced both in 1967 and 1973 although suspended solids (inert) reach their highest here in 1973 (Figs. 4.3, 4.5, 4.6).

The Asellus population has always been large and is probably the result of the Cladophora association. The absence of the first breeding population in 1973 is a mystery but thought not to be caused by water quality. The riffle is subject to a lot of disturbance at certain times of the year as children play in the surrounding recreational area.

The increased stability in both numbers and species expected with the more complex ecosystems encountered in recovery zones is demonstrated. The numbers of chironomid larvae have increased with cessation of the effluent but fewer species have appeared compared with Stations upstream. Although a natural ecosystem contains more species than an unnatural one, the severe competition for the depleted sites upstream (Station 2 in particular) by the Chironomidae, with their aerial advantage, causes a large number of species within this family to be expected at first. The same is true for many of the more mobile invertebrates not belonging to Chironomidae or Diptera.

Improvement of water quality has rectified the 1967 barren status of the sensitive Gammarus population to one of considerable proportions. The tubificid numbers have been decimated with recovery but with one slight increase coincident with a rise in ammonia, B.O.D. and slight drop in oxygen during April 1973. The causal factor of the decrease could, apart from chemical factors, be change in substratum, since in 1967 and prior to June 1972 the 'sewage fungus' encouraged large amounts of sand and silt to accumulate which completely disappeared with betterment of the water quality.

Large populations of E. testacea abounded here with consistent representation from G. complanata also. The similarly small populations of Lymnaea probably sufficed as food in a predatory/prey relationship with Glossiphonia of the type Solomon (1969) describes as 'reciprocal' oscillations. The high suspended solids of an inert nature caused by fords and, in 1972-3 river divertion work, are not conducive to the creation of large populations of molluscs (Boycott, 1936) and may explain their poor colonisation. Too much emphasis must not be placed on the *Glossiphonia*/*Lymnaea* relationship, as Station 5 is almost devoid of *Lymnaea* and unless this accounts for the increased population of *Glossiphonia* here, it would not be in the interests of the leech to eliminate them completely. The rarity of *Glossiphonia* at Station 5 between February and June 1972, may be the result of over predation but during their absence between February and June 1973, *Lymnaea* does not reappear. It is quite likely, then, that the solids or flow regime keep *Lymnaea* and *Ancylus fluviatilis* absent from Station 4.

Physa acuta appeared in November 1972 in small numbers only (Table 4.4), but for the first time in the history of the Upper Cole. The following section on the Lower Cole will show how *Physa* has been steadily moving upstream from the confluence with the River Blythe.

Erpobdella testacea show a rapid increase in numbers since May 1972. Previously, only very low numbers were recorded here which is a reversal of roles with *E. octoculata.* This leech is now very scarce here, when, prior to May 1972, it was present in fairly large numbers. (The hypothesis expounded to explain this is given in the previous section).

B. rhodani has managed to colonise this Station well although its large populations are subject to large oscillations in spring and autumn. Emergence, for which *Baetis* moves downstream (Macan, 1964) and *Cladophora* cover of stones, may account for such fluctuations as the habit of *Baetis* is to cling onto the sides of stones and only move up when the current is slack (Percival and Whitehead, 1929). *Cladophora* greatly reduces the current locally and would prevent such behaviour. The relative ease with which *Baetis* managed to colonise this and Station 3 after May 1972, becomes credible when considered in the light of 'sewage fungus' free stones now with epiphytic growths, improved oxygen tensions, oviposition in May and October (from Harris 1952) and drift from upstream Stations and tributaries.

The asexually reproducing Naididae appear typically associated with the seasons of *Cladophora* and with the exclusion of the Chironomidae, there is a generally poor representation from other taxa. The bivalves *Sphaerium* and *Pisidium* are absent, which may be correlated with the lack of sand, silt or mud (Pentelow *et al*, 1936).

C. riparius have been absent since the elevated B.O.D.'s and suspended solids of October 1972, but nevertheless have always been scarce here, even in 1967. *Brillia longifurca* and *Prodiamesa olivacea* were recorded as being frequent during October 1972 also, but they subsided afterwards. *Thienemannimyia* has established a large population (Fig. 4.26), and visitations from *Brillia modesta*, which was as scarce as the former in 1967, have been made. No change in the seasonal abundance of *Cricotopus bicinctus* has been observed since May 1972 but the proportion of *Eukiefferiella hospita*, which was always fairly large, was only represented once - in early 1973 (Fig. 4.22). Three species, *Rheocricotopus*, *Diplocladius cultriger*, and *Micropsectra atrofasciatus* were rare and spasmodic, the two former making their debut at this Station. Most notable of all is the complete absence of *Cricotopus sylvestris* which, in 1967-8, averaged $86/0.1m^2$ with a monthly maximum of $590/0.1m^2$, and *Cricotopus* (*Cricotopus*) *albeforceps*? which averaged $140/0.1m^2$ with a monthly maximum of $822/0.1m^2$. (Davies, 1971, and Hawkes and Davies, 1970, although they refer to the latter species as *Trichocladius rufiventris*. Meigen).

Station 5. Even prior to May 1972 the communities at this Station would be described as distinctly improved as a result of self-purification, and this final Station to be investigated on the Upper Cole exhibits the chemistry of a river in very advanced stages of recovery. Post-closure chemistry shows a drop in B.O.D. and P.V. compared with Station 4, although the mean dissolved oxygen for 1973 is slightly lower, however, a moderately low temperature keeps the % saturation higher. The suspended solids have settled out since Station 4 but ammonia levels are much the same. Inspection of the 'spatial' graphs shows at a glance how the chemical parameters have improved with the abatement of the effluent.

The high levels of plant nutrients together with other conditions like flow, become ideal for the growth of *Cladophora* so that during summer and autumn vast blankets of this weed occur up to 15 ft. long and over 80% cover at times. With the alga in these proportions and presumably as they were in the late 1960's, it is not surprising to find *Asellus aquaticus* abundant throughout the year - a situation also very similar to the late 1960's. The *Gammarus* population is also very large, much more so than its pre-closure status. Both the isopod and amphipod populations are large and show less susceptibility to fluctuation than at the other stations (with the exclusion of Station 1 for *Gammarus*), but the slight reductions in April and May respectively are not understood. Reference made to the population dynamics of *Asellus* in the R. Ray (Figs. 8.1 and 8.2, Chapter 8) shows large percentages and numbers of adults during these months.

Despite the improvement in water quality over the previous station, *Baetis* is less abundant and of little greater relative abundance than the population in 1967 - May 1972. It is therefore suggested that the percentage cover of *Cladophora* is responsible for maintaining depressed numbers (as at Station 4), *Baetis* showing maximum abundance during February and March 1973 before the *Cladophora* growth escalates.

The leeches follow the trend of Station 4 with *Glossiphonia complanata* increasing slightly in number after the cessation of the outfall but being relatively rare the following year. Erpobdella octoculata only appears to any degree once during 1973, E. testacea remaining the dominant leech in the habitat. Both E. testacea and G. complanata populations are vastly increased in size compared with the 1967 - 1972 situation.

The increased silt, sand, and free gravel that Cladophora traps beneath it would favour the bivalve Sphaerium corneum but it is never abundant. This substratum also permits small populations of tubificids to live but they do so in greatly diminished numbers to pre-May 1972. Naids are common in the Cladophora and mosses. Ancylus fluviatilis persist at this station though they are rare, whereas Lymnaea pereger has never taken a hold here.

Thienemannimyia dominates the chironomid population with Prodiamesa olivacea, Cricotopus bicinctus and Cricotopus (Cricotopus) albeforceps? sub-dominant. These have always managed to survive well here as has Brillia longifurca but this species became increasingly scarce in 1973. C. riparius has never appeared since May 1972 and Micropsectra atrofasciatus and Eukiefferiella hospita only exceptionally, but with the more frequent occurrence of E. hospita in 1967 it would seem that the water quality has never suited C. riparius. Metriocnemius atratulus, E. hospita, Micropsectra, Polypedilum scalaenum, and Rheocricotopus have made small and successive appearances.

4.1.3 DISCUSSION.

The most characteristic observations made in studies of organic enrichment have been the suppression of the natural clean water riffle fauna and replacement with one consisting of animals that are more representative of depositing substrata or stagnant pools. These are, in varying degrees, already adapted to silted or muddy bottoms, high and low oxygen concentrations, and plenty of detritus but they find the conditions in organically enriched riffles particularly favourable for large populations, there being little or reduced competition for space and food. Before May 1972 all the clean water fauna had been eliminated or suppressed throughout the Upper Cole with the exception of Station 1. Just downstream of the effluent, the riffles were dominated by Tubificidae, Enchytraeidae, and C. riparius which, downstream and after some self-purification and dilution of the river, were progressively suppressed (with the exception of Tubificidae) as Asellus, leeches, chironomids, and small populations of other more tolerant invertebrates competed for food and space. This is recovery with self-purification. Brinkhurst (1965) has suggested that one might expect biological changes in time, after the removal of the source of pollution, to follow the same pattern and stages of recovery as a river undergoing this self-purification. This is an assumption that probably only holds true very infrequently, for example, when the type of pollution has been toxic (in the sense of directly poisonous) and is slowly leaching out of the substratum (i.e. residual toxicity) so that dilutions are approximate, in time, to those encountered downstream before the effluent was relinquished. The 'stages' are only feebly definable in a river undergoing self-purification and can only be typical of a particular river as a result of every river having its own unique combinations of biological, geological, and chemical characteristics. With regard to his expectation of biological changes <u>in time</u> after the removal of an effluent going through the same stages of recovery demonstrated during self-purification, the speed with which this will be achieved (if at all) will be dependent upon permutations of some river characteristics amply exemplified in the Upper Cole.

The Upper Cole has been subjected to an effluent which is non-toxic, but in the absence of residual toxicity it would be naive to think that invertebrates should be able to inhabit the severely depleted area below it with great ease. The situation differs from the colonisation of the experimental streams (Chapter 5) which are, in effect, just areas of artificially denuded substratum. The differences lie in the river at Station 2, for example, not being without its own fauna altogether as the experimental streams were but containing a large biomass of a few species that newcomers, with the cessation of the effluent, would find themselves initially in competition. Residual physical factors like 'sewage fungus' and siltation would take time to deteriorate and be washed away with 'spates', thereby making way for those who require clean stones and crevices. The clearance of these 'residual physical' factors has been hampered as the cessation of the effluent did not see the end of organic enrichment in the upper stretch and, as the chemical data has shown, intermittent low levels of pollution were occurring above Station 1 with slightly worse relapses during winter. It has been shown how this 'background' organic pollution has impeded the establishment of a number of species at the higher stations, particularly the more sensitive ones like *Baetis*, *Gammarus*, and *Ancylus*. Yet despite this hindrance, there has not been a time at Station 2 when the recovery stage typified by *Asellus* and leeches has presented itself despite this clear stage being manifest at Station 4 and 5 both before and after the closure of Houndsfield Lane S.D.W. Moreover, both *Baetis rhodani* and *Micropsectra atrofasciatus* have inhabited Station 2 in abundance beginning only four months after the closure. The exclusion of the '*Asellus* stage' and the hastening of the cleaner water fauna is most likely a reflection of two factors which affect the time of recovery, i.e. voltinism and mobility.

Multivoltine species are at an obvious advantage over univoltine or species with overlapping generations, not only in that they have greater fecundity per year but with the possession of more chances of oviposition or brooding near to the time of effluent abatement. It is suggested that the bivoline and possible multivoltine cycles of many of the Chironomidae were responsible, in part, for their swift success in colonisation of Station 2 and 3 in particular, after May 1972. However, consider the situation if the effluent had been abated in winter when all the Chironomidae were in the larval stage. No oviposition could have been made until the larvae emerged in spring (depending on temperature and voltinism) thus giving time for other fauna to invade the denuded habitat, and in particular those having brooded in September/ October like Asellus. The importance of the time of year that the effluent is removed can be put into perspective therefore, and mention of the role it played in E.testacea and E. octoculata colonisation has been made. Invertebrates capable of asexual reproduction like the Naididae and some Tubificidae will be at an advantage also, and the sudden appearance in large numbers of the former at Station 2 only one month after the severe June pollution may emphasise this. Provided food is available to sustain the newcomers, the utility of voltinism and other factors influencing speed of recovery is realised.

The prominence of mobility as a factor influencing the speed of recovery has been referred to in the results. Mobility, it seems, does not only refer to the physical capabilities of an animal like a triclad making its way slowly upstream but includes those phenomena associated with migration. Baetis is a poor swimmer and drifts downstream for reasons of dispersal and completion of its life cycle. Spates may dislodge those better swimmers and those better adapted to securing themselves, like Gammarus and Asellus, also. The drift factor must have been important in establishing the *Baetis* community at Station 2 so swiftly after May 1972, and in the supply of *Gammarus* to Station 3.

Upstream movement, exemplified best by fish moving up or downstream into improved quality water, is with their exception a slow process on the whole. It would have required considerable time for members of the large populations of *Asellus* at Stations 4 and 5 before the closure to crawl the 4.5 km. upstream to Station 2 (Plate 5). This mobility may have helped it to colonise Station 3 but the residual colony there was strong anyway and reproduction would have attained the large number seen by the winter of 1972/3. The small population at Station 1 would also have supplemented, by drift, the downstream populations. Re-instatement of a leech or molluscan population would be expected to be very slow and Hynes (1966) has quoted the wearisome progress of *Lymnaea pereger* in the R. Lee. However, Hawkes (1963a) noted swift re-instatement of *Lymnaea* in the Lower Cole from eggs and adults on drifting debris. The existence of fish populations at stations have not been documented for the period before May 1972 but occasional *Gasterosteus aculeatus* were taken in heel samples at both Stations 1 and 5 and presumed absent at all the other stations. A more recent electro-fishing survey executed in October 1972 revealed a number of small fish at all stations but nothing can be inferred about the speed with which they moved in.

Table showing the number of fish caught 50m up and downstream of the Sampling Station in October 1972.

	Stations				
	(1)	(2)	(3)	(4)	(5)
Gasterosteus aculeatus	100	12	130	400	300
Nemacheilus barbatulus	10	-	9	-	15

The relative numbers may, however, be indicative of the relative productivity in terms of invertebrate food available, as the water quality was fairly uniform at all stations after May 1972 and therefore not thought to be a limiting factor.

Also pertinent to mobility and to the success that B. rhodani and the Chironomidae demonstrated in recolonising Stations 2 and 3 is aerial dispersal. The bivoltine and possibly multivoltine rhythms of the Chironomidae favour this group over the univoltine B. rhodani giving more chance to the chironomid adult to oviposit soon after a source of pollution is relinquished.

The common denominator of all the modes of distribution and mobility is the existence of sources from which to recruit. Unaffected or recovered parts of the river are rich and obvious sites from which to recruit but some of these may be miles up or downstream of the injured area. The speed of recovery of the R. Cole was aided by the existence of fairly clean tributaries and the role of Houndsfield Lane tributary for a supply of Baetis; Peterbrook as a supply of Baetis and Gammarus, and Chinn Brook for sources of replenishment to Stations 4 and 5 are discussed in the previous section.

It was the intention of the three-week survey commencing one week after the closure of Houndsfield Lane S.D.W. to highlight any migrants colonising with exceptional speed. M. atrofasciatus gained a foothold at Station 2 during the third week and exploded in numbers soon afterwards. Large numbers also appeared at the same time at Stations 1 and 3 so, although the life history is as yet undetermined, it looks as if the adult population emerged for the first time since the over-wintering to gain a 'seasonal advantage' in relation to the effluent cessation. Prodiamesa olivacea appeared at Station 3 in the third week and was abundant by the next month. The population was already large downstream at Station 5, indicating the aerial advantage as it was absent at Station 3 for the first five months of 1972. The longer delay involved in inhabitation of Station 2 was put down to presence of still large amounts of 'sewage fungus' - this probably prevented Baetis and other chironomids (not C. bicinctus) getting a hold until later.

The appearances of G. complanata and E. testacea within the three weeks at Station 4 may have been due to the upstream movement of small populations from Station 5, and their success afterwards being the result of fortuitous mating time. The decline of C. riparius at Station 3 to nothing within the three weeks is equally spectacular and other drastic recessions by tubificids at Station 4 are also notable. A steady increase was seen in the number of species during the second and third weeks and it can be seen that not all are members of the Chironomidae.

Reference has been made to the existence of residual colonies of invertebrates which relates to those members of the community whose numbers have been suppressed by the prevailing water quality. A number of such animals may well have been undetected during sampling as a result of gregariousness and sparcity. The improvement of water quality very likely favoured their multiplication and this 'local' recruitment would therefore have been a factor influencing the speed of recovery. However, as the majority of these species would have had a univoltine rhythm or extended and overlapping generations, for example E. octoculata and Asellus, they would consequently have taken up to a year or more before increasing their numbers. The existence of residual colonies at Station 3, where conditions were less severe yet more unstable than at Stations 4 and 5, may have contributed substantially to the improvements in density of many species noticed after one year of good quality water.

The community structure and abundance attending recovery is therefore seen as being a function of a number of factors including mobility and breeding rhythms of individuals, with these in particular being responsible for the short term structure of recovery, the density independent factor of poor water quality having been removed.

The change from a highly organic enriched condition to one of singularly low status is often detected by differences in species type - the so-called 'biological indicators' and the species relative abundance. Pentelow *et al* (1938) and Berg (1948) remarked upon the reduction in numbers of *Asellus* together with the increase in numbers of *Gammarus* both after the removal of the source of organic enrichment and with the process of self-purification downstream from such a source. A change in the *Gammarus*/*Asellus* community before and after cessation of the effluent was observed in the Upper Cole also, and it is likely that the ratio or relative abundance of certain species will prove useful as indices of a river's condition. Only subjective estimates of the proportions of *Gammarus*/*Asellus* rather than quantitative ratios could be made with the 'heel' samples taken in the R. Cole.

Abundance of Gammarus and Asellus at the five stations of the Upper Cole before and after removal of the effluent.

	Before May 1972		Approx. 1 year after	
Station	Gammarus	Asellus	Gammarus	Asellus
1	abundant	rare	abundant	rare
2	rare	rare	rare	rare
3	rare	rare	abundant	occasional
4	rare	abundant	abundant	occasional
5	occasional	abundant	abundant	abundant

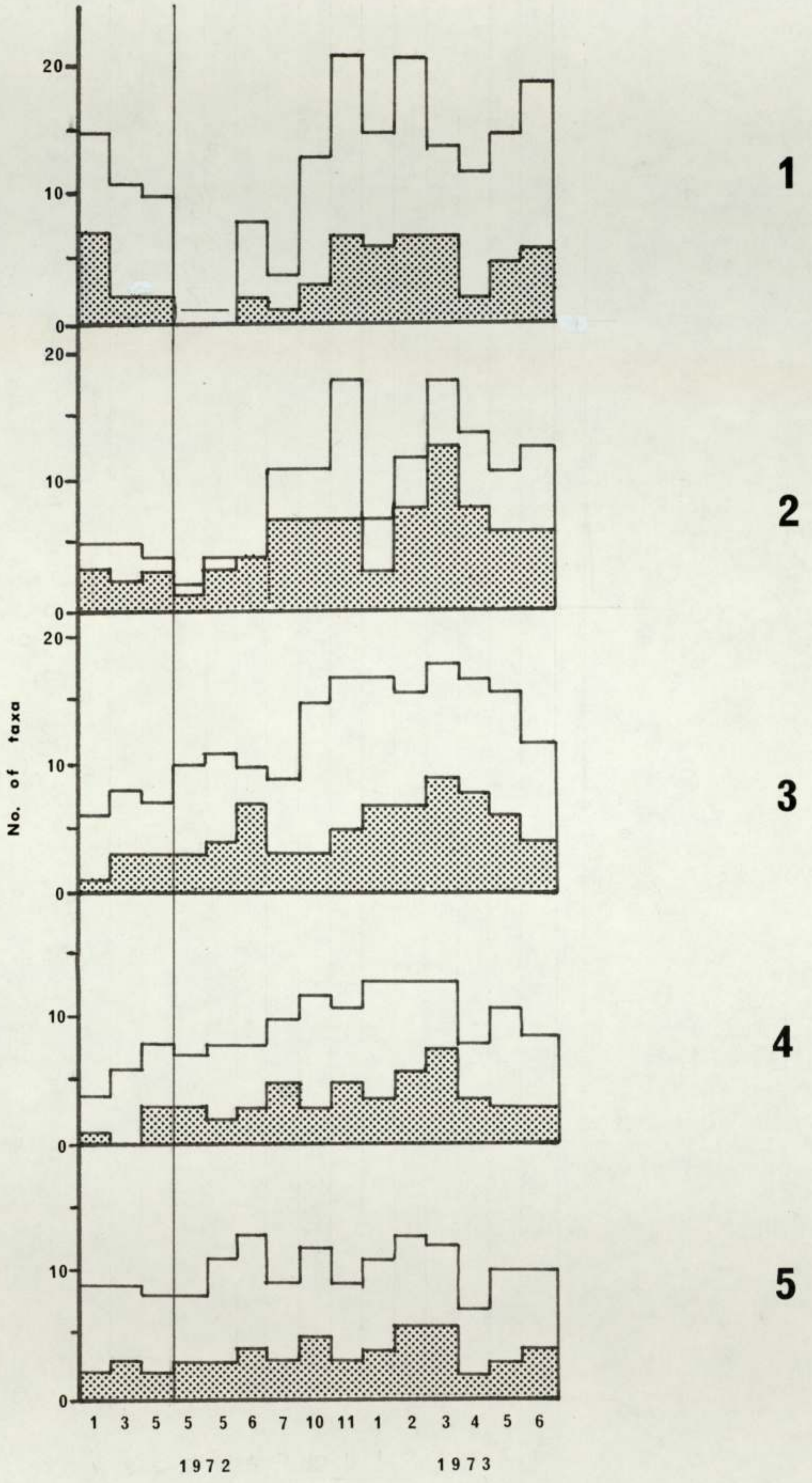
The relative abundances before May 1972 from Stations 2 - 5 show a typical relationship as self-purification proceeds downstream, and one year later some transient stages during recovery are seen. Something, perhaps reproductive power or lack of food, has prevented the establishment of large populations of either species at Station 2 although recent observations (June 1974) has shown *Asellus* to be 'abundant' and *Gammarus* 'occasional'. Stations 3 and 4 show a tendency towards the normal relationship as described by the control Station 1, while Station 5 shows a stable abundance of both, which may be attributable to the two distinct habitats of bare stones and stones and *Cladophora* here. Changing numerical relationships have been described above between *E. octoculata* and *E. testacea* and quantitative work might reveal ratios of use to the river biologist. Relationships probably also exist between some species of the Chironomidae but their unstable numbers caused by seasonal cycles and irregular decimation from areas by spates would make them unsuitable.

Other structural changes in the community were expected with regard to the balance of trophic levels, i.e. the relative numbers of producers, consumers, and scavengers. Organically enriched biotopes are dominated by the detrital feeders or scavengers typified by tubificids, *Asellus*, *C. riparius*, *P. olivacea*, etc. and recovery of the Upper Cole has seen a reduction in numbers of these as well as an increase in the number of herbivore consumers like *Baetis rhodani*, *Cricotopus bicinctus*, *Cricotopus*(*Cricotopus*) *albeforceps*?, some molluscs together with carnivores like the hirudinean representatives and *Thienemannimyia*. An increase in primary production, although not measured, was expected as the levels of T.I.P., NO_3 and NH_3 remained much the same whereas the heavy solids had been removed and the abundance of herbivores like Baetis rhodani and some of the Orthocladiinae confirmed these suspicions.

The stability of the ecosystem is increased with its complexity so that as successive recovery 'stages' downstream attain greater numbers of species it follows that the ecosystem, although still simplified, is achieving greater stability. Ecosystem complexity has sometimes been measured using the criteria of diversity, species numbers or number of taxonomic entities, etc., and there is some value in such measurements when one considers it with regard to Thienemann's Second Biocoenotical Rule. Nethertheless it is only of value when certain limitations are allotted to it, and the example of Patrick (1961) who tried to compare numbers of species not only between rivers but between rivers in different geographical areas demonstrates its misguided use. By defining the frames of reference in which the number of taxa are to be compared is to increase the validity of such comparisons.

The number of taxa, restricted to the riffle sections within the Upper Cole, at Stations 1 - 5 have been compared to demonstrate the improvement in stability of the ecosystem with changing water quality (histograms Fig. 4.27). Station 2 showed a marked increase in numbers of species ensuing after the effluent and the acute pollution in June had subsided, and closer inspection

FIG.4.27. Total number of species at the five Stations on the Upper Cole before and after the closure of Houndsfield Lane (thin line). Shaded areas denote chironomid spp. only.



revealed this to be in the first instance the result of chironomid colonisation (38% chironomid larvae in November 1972, 72% in March 1973) and attempts by Glossiphonia complanata, Lymnaea pereger, and Sphaerium to become established. Both the chironomid and other macro-invertebrate populations showed gradual increases in numbers after the closure of Houndsfield Lane S.D.W. but, if the mean numbers during the months until June 1972 (when the chemistry shows all gross pollution to have abated) were compared with similar months for 1973, an increase from 7.5 to 19.2 species is observed. A larger proportion of chironomids (66%) made up the totals when the water quality was bad whilst during recovery a reduction in the percentage was noticed (57%) and, although an increased number of chironomids are shown with recovery, the slightly reduced percentage indicated an equal improvement in the other benthic invertebrate populations.

Observations made at the Control Station 1 showed the chironomid populations to be relatively stable, varying in degrees with emergence and times of reproduction and only showing a reduction during the June 1972 pollution emanating from upstream. The other taxa showed peaks in November 1972 and February 1973 as a result of brief appearances from Asellus, Sphaerium, Pisidium, Amphinemoura, and dipteran larvae.

The histogram for Station 3 (Fig. 4.27) shows, even before closure, a greater proportion of species and greater stability when compared with the station upstream and also a greater proportion of macro-invertebrates other than chironomids during recovery. The more stable structure of Station 3 comprising Asellus, Gammarus, Baetis, Lymnaea and Tubificidae was detectable even before the closure of the S.D.W. The acute poisoning in June 1972 eliminated Gammarus and Lymnaea and allowed the numbers of chironomid taxa to increase here more than at any other Station, and it is suggested that the higher chironomid populations of Station 3 as opposed to Stations 4 and 5 may represent the relative instability of this Station. The improvement since cessation is very noticeable; firstly in the numbers of taxonomic entities, and secondly in the maintenance of these numbers - a feature not yet wholly detectable in the Station upstream. The mean number of species over the period up to June 1972 was 11.2 and for the same months of 1973 was 22.2, so the number of species has roughly doubled itself in one year of recovery. The percentage of chironomids parallelled the pattern of Station 2 in that it fell slightly with recovery, i.e. 45% to 41% over the respective months relating to these figures. These reductions may be seen as increasing pressure on the chironomid population as other invertebrates compete for the same food and space. However, with such small differences and numbers of the former being dependent upon the prevailing weather, it is difficult to assess with any certainty.

Recovery has increased the mean numbers of taxa at Station 4 from 6.5 to 11.5 over the equivalent pre-and post-closure months. The equivalent percentage of chironomids before closure was 26% and afterwards 39%, Stations 4 and 5 being the only stations to show a substantial increase in the chironomid percentage with recovery. The 17% drop in total numbers of species attending the recovery months over the upstream Station 3 is anomalous but it may be a feature of more unstable recovering habitats like Station 3 to have a greater number of species attempting to colonise as a result of the undetermined and less secure nature of the habitat.

The increased relative stability and quality of the water at Station 5 prior to closure was expected to favour a more stable and diverse community. The numbers of taxonomic entities has certainly remained fairly stable and only a slight increase has been noted since the removal of all contamination (in June 1972). Equivalent means of taxa for months before and after cessation of the contamination are 9.8 and 10.8 which suggests, as the water chemistry does, that the water quality has not changed appreciably and the community reorganisation has apparently affected the biocoenoses very little. The percentage composition of chironomid species has, like Station 4, increased from 28.5 to 39.5 over the same periods before and after closure. It may be significant therefore that those stations which were shown to have similar or near similar chemistries, both before and after the closure of Houndsfield Lane, should demonstrate overall increases in the percentage of chironomid composition, i.e. Stations 1, 4 and 5, while those with vastly differing chemistries for the same periods show decreases in the percentage of chironomid composition, i.e. Stations 2 and 3.

Mean percentage chironomid composition for the same months before and after closure of Houndsfield Lane. Station 1 percentage is also for the same months.

	Station	Before	After	
	1	29	38	
effluent →	2	66	57	
	3	45	41	downstream ↓
	4	26	39	
	5	28.5	39.5	

The following points emerge from the Table above: -

1. The percentage composition of Chironomidae is highest below the effluent at Station 2 and diminishes with distance and self-purification downstream.
2. The percentage composition of Chironomidae is also highest at Station 2 up to one year after the abatement of the effluent. This percentage also diminishes with distance although self-purification is virtually negligible.
3. The percentage composition of Chironomidae at Station 4 and 5 before and after the cessation of the effluent shows a striking resemblance to that

of the Control Station 1. The similarity between these percentage compositions of Chironomidae would suggest that both Stations 4 and 5 were in an advanced state of recovery before the closure as the chemistry has indicated. The stability of the communities at these stations are much the same as they were before as after cessation of the effluent and at least as stable as the Control Station, the variation being inherent in all these.

4. The percentage composition of chironomids at Stations 2 and 3 drops over the two periods described, whereas it increases over the same period at Stations 1, 4 and 5.

It is difficult to generalise about the distributions of chironomid larvae with respect to water pollution, especially when the field data shows infrequent occurrences and considering the notoriously labile nature of chironomid populations anyway. If domicile were more or less permanent, like <u>C. riparius</u> in very badly polluted waters, speculation would be easier. However, colonisation with recovery from organic pollution is an area that chironomids would be expected to excel in. Their mobility in the adult stage and voltine rhythms explains their ability to explode in numbers from a residual population of very few. Bryce (1972) recorded almost complete removal of chironomid populations with flooding in the R. Hodder (Yorks.) yet from minute populations sheltering on the downstream sides of boulders and in moss they could regain their numbers in summer to around 60,000 per m^2 as long as scouring had not occurred for 5 - 6 weeks beforehand. Large quantities are often reported in Cladophora and bryophytes, and Percival and Whitehead (1929) observed up to 97% of midges of Orthocladiinae inhabiting these plants. Those that build their fine tubes on stones, like Micropsectra, were observed in such quantity at Stations 1 and 2 after the June 1972 pollution that the stones appeared to be covered in silt and sand. A majority of others make their tubes in sandy substrates (e.g. Prodiamesa olivacea) and in silt and detritus which makes organically polluted rivers particularly suitable when considering their method of feeding and food supply, and some references have been made in the previous section concerning the distributions of some larvae with substratum.

The use of chironomid larvae as 'biological indicators' of water quality immediately recalls the Chironomus 'thummi' and 'plumosus' groups to mind, but of course no 'indicators' are obligatory of any type of water quality so that discretion must be exercised in their use. Naturally the possession of haemoglobin in solution is advantageous to living in near anoxic conditions and Procladius sp. may be misleading as the very young forms are phytophagous (Chernovskii, 1949) and would necessarily inhabit different biototes to the adult carnivores. Micropsectra also contains haemoglobin but, from factors which emerge from this study, does not share the enthusiasm of C. riparius or adult Procladius for severe organic pollution.

A valuable 'indicator' will conform to the pattern of appearing in large numbers with worsening conditions and receding with improving conditions again, but there are probably also some that are indicative of degrees of pollution when they are in small numbers or in certain association with each other. However, quantitative data would be needed to decipher these and even then the relationships would only tenuously hold true for the river and habitat described.

Those chironomid larvae that appear indicative of severe pollution in the R. Cole are Chironomus riparius, Brillia longifurca and Procladius spp. Cricotopus (Cricotopus) albeforceps? and Eukiefferiella hospita are tolerant to a lesser extent and also appear in large numbers in moderately polluted areas, e.g. Stations 4 and 5 both before and after closure of the S.D.W. Prodiamesa olivacea, Micropsectra atrofasciatus, Cricotopus bicinctus and Thienemannimyia are all variably tolerant of gross pollution but when in large numbers seem indicative of mild pollution. These are the species that were so successful at Station 5 before May 1972 and afterwards at Stations 2 and 3 typifying the moderately polluted conditions found there.

It has already been noted that the abundance of chironomid larvae is a function of the previous weather - emerging in good weather and being delayed in cold weather.

Some of the changes described in the chironomid populations may have been influenced, after closure, indirectly by the change in flow rate and volume. The smaller volume renders the water temperature more susceptible to fluctuations with air temperature as reflected by the greater variation of the standard errors of the 1973 means. The length of development and times of emergence have surely been affected by this greater fluctuating temperature.

The Chironomidae are among the greatest producers of animal food with the herbivorous and carnivorous forms representing a large part of the primary and secondary consumer organisms. The diet of most fish include this family, the contents of some fish stomachs being 80% chironomids. Aspects of the ecology of the Chironomidae are therefore important not only to the pollution biologist but to other aspects of water management.

4.2. THE LOWER COLE.

An investigation into the nature of the changes in benthic communities following the removal of a toxic sewage effluent was subordinate to the study of the Upper Cole communities. The source of the toxic effluent was Yardley S.D.W. which was closed down in June 1971, i.e. before investigations were possible by the author. The most comprehensive account of this stretch of the river was an early one by Hawkes (1963a) who examined the benthic ecology between 1950 and 1960. Also, a number of changes were seen in the quality and types of effluent entering upstream of the Yardley S.D.W. between 1960 and 1971 but the amount of chemical and ecological data available and pertinent to this time was tenuous although biological samples did show that very little change in the benthic community structures had taken place.

Between Station 5 (of the Upper Cole) and Station 6 of the Lower Cole was a distance of 4.6 km which flowed through an industrialised part of the city and received occasional toxic factory discharges and sewage overflows. This 'middle' part of the R. Cole was not examined. The outfall from Yardley S.D.W. entered the river just above Station 7 and it was the effect of the cessation of this major effluent that was being measured in terms of the macro-invertebrate fauna. The enquiry was less intensive than that on the Upper Cole as it was not expected to recover as fast, and this, together with flimsy prior documentation, led to its being sampled approximately every 2 months over a period of eighteen months beginning November 1971 to May 1973. This meant that the first sample was taken some five months after the closure of the Works. Plate 6 shows a map with the course of the Lower Cole.

Station 6 was used as a control Station although its condition was only a little better than those below the Works due to the industrial discharges in the middle stretch and a sewage overflow discharging for several hours a day. In 1957 a new sewer was constructed which relieved the discharges above Station 6 but overloaded the S.D.W. at Yardley. The resulting changes in the faunal communities noted by Hawkes were very slight which led him to believe that it was the toxic part of the discharges that accounted for the restricted communities. Likewise, very slow changes were expected when Yardley S.D.W. closed in 1971.

The nature of the crude sewage entering Yardley S.D.W. was typically industrial, with metals, phenols and cyanides from metal finishing and plastics industries making oxidation inefficient. This factor together with the overloading meant that the standard of the effluent was poor and about 1 million gallons/day of settled sewage went straight into the river. The following analyses from Devey et al (1972) shows the average chemical composition of the tripartite effluent from April 1970 - March 1971. Figures in mgl^{-1}.

Sample	B.O.D.	S.S.	NH_3	Oxid. N.
settled sewage	137.0	173	19.1	2.6
effluent of rectangular humus tank	21.8	43	9.3	18.1
effluent of circular humus tank	30.9	48	14.4	10.4

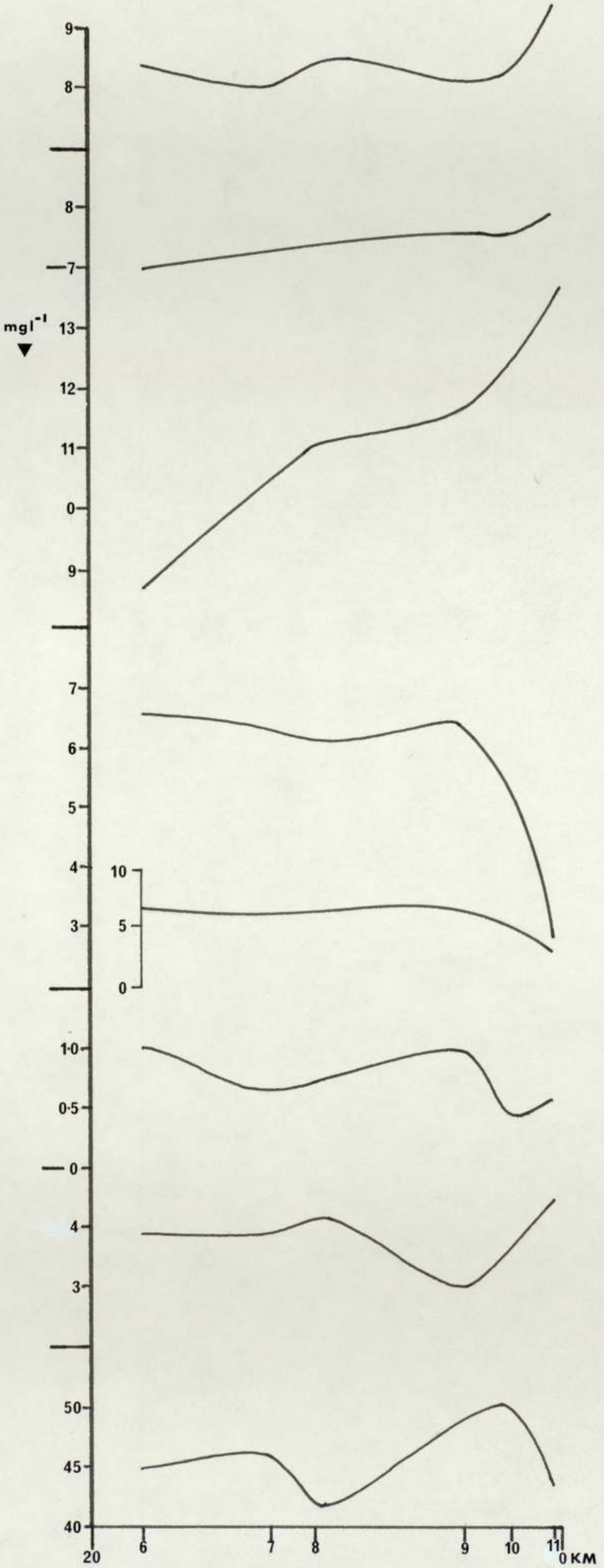
In June 1971 Yardley S.D.W. was closed and the sewage diverted to Minworth S.D.W.

4.2.1. Chemical Data.

Owing to the variability of spot samples taken at roughly two monthly intervals, graphs illustrating seasonal fluctuations in water quality have not been drawn and incomplete data for sampling Stations before and around the time of Yardley's closure were likewise left in tabular form. However, the means of the samples from November 1972 to May 1973 were plotted for each Station against their distance in km. from the confluence with the River Blythe. These spatial graphs (Fig. 4.28) indicate the chemical trends over the sampling periods as the river flowed downstream. The full chemical analyses are shown in Tables 4.13 and 4.14.

a) Dissolved oxygen. Since the closure of the Works substantial increments in the amount of dissolved oxygen downstream have been made. The concentration at Station 6 remained about the same and the average figures for Stations 7, 8 and 9 demonstrate that only those species very sensitive to oxygen levels would have been affected

FIG.4.28. Spatial graphs of mean annual chemical and physical parameters at Stations 6-11 of the Lower Cole plotted against distance in km from the confluence. See FIG.4.8. for abbreviations.



as the minimum (at Station 8) was only 41.9% saturated. The spatial graph (Fig. 4.28) demonstrates the daytime increases due to self-purification and photosynthesis which were markedly amplified from Station 9 onwards where Cladophora grew well; so much so that a supersaturation of 169% was recorded in spring 1973 as a result of the vast 'blankets' at Station 11.

b) Temperature. The figures available for the periods prior to the closure of Yardley indicate the variability inherent in the annual means, not only between different stations but at the same stations. Post closure figures for equivalent stations demonstrate the effect the effluent had on the river, i.e. even taking into consideration the variance present beforehand. The variations between the means of the same stations are regarded as the result of local discharges.

c) Biochemical Oxygen Demand. The organic loading in 1969, 1970 and 1971 was increased by the input from the effluent below Station 6 and the mean figures show how self-purification relieved the B.O.D. downstream to half of its value over the 8 km. to Station 9. The spatial graph shows the amount of organic material since the closure to be of a much reduced level, with some reduction taking place in the latter stretches.

d) Ammoniacal and Oxidised Nitrogen. The ammoniacal nitrogen dropped to a fraction of its original level after June 1971, and subsequently varied in only small degrees -

not exceeding 1 mgl^{-1} on average. Considering the pH range of 7.0 - 7.6, the hardness and amounts of dissolved oxygen in the water, the ammonia levels were fairly innocuous. The oxidised nitrogen varied inversely with the ammoniacal nitrogen levels, the two providing ample nitrogen for plant growth.

e) pH. During the daytimes the pH increased steadily downstream as the carbon dioxide utilisation during photosynthesis became greater. This was in accordance with the greater oxygen levels and amounts of algae encountered as the confluence was approached.

f) Metals (Table 4.13.) Results for Stations 6 and 7 were only available in the form of combined total metals up to 1971. Nevertheless, the increase in input from the S.D.W. was dramatic and declined with dilution downstream. Individual total metal for copper, zinc, nickel and chromium are shown for Stations 8 and 9 before closure and for Stations 7, 10 and 11 after closure. These metals were typical of the plating industries discharging into the sewer and small amounts of lead have been detected in some spot samples also - (0.17, 0.14 and 0.25 mgl^{-1} Pb exceptionally). The heavy-metal figures show a greater then 10 fold decrease in concentration when compared with the yearly averages for the period before the effluent was diverted to Minworth S.D.W. Unfortunately these samples, which are only monthly spot samples, often miss higher concentrations resulting from 'spills', either accidental or deliberate and Hawkes (1963a) has recorded 4.8 mgl^{-1} Cu and greater than 2 mgl^{-1} Ni in some samples, and data from the City of Birmingham, Public Works Department, have shown .2 mgl^{-1} Zn (7.1.70), 0.75 mgl^{-1} Pb (13.8.70), all of which were discharged above the outfall of Yardley S.D.W. Therefore, even though most of the toxicity of the Lower Cole was removed with the closure of Yardley S.D.W., intermittent discharges by factories quite likely occurred.

g) Phenol. Other toxic substances like phenols have been recorded and although figures are only available for below Sparkbrook, i.e. above Station 6, total levels were around 0.8 mgl^{-1}. A survey in July 1972 revealed total phenols in the Lower Cole and it may be assumed that as phenol is mostly untreatable, levels were higher before the Works closed down.

Table showing levels of Total Phenol during spot sampling in July 1972.

Station	mgl^{-1}
6	0.7
7	1.05
9	0.7
10	0.7
11	0.7

4.2.2. BIOLOGICAL RESULTS AND INFERENCES.

Brief History.

Hawkes (1963) has documented the macro-invertebrate communities along with the occurrence of *Cladophora*, 'sewage fungus' and the resistant alga *Stigeoclonium* over the decade 1950 - 1960.

He found that the benthic communities at Station 6 were severely restricted in variety as a result of the industrial contamination and sewage overflows being discharged just upstream. The toxic nature of the discharges also accounted for the absence of saprobic micro-organisms with the exception of small amounts of 'sewage fungus'. The riffle was covered with growths of *Stigeoclonium* and the fauna was restricted to a few chironomid larvae, tubificids and *Lymnaea pereger*. Some *Cladophora* appeared in 1959 when a newly contructed sewer stopped the daily overflow upstream and industrial wastes were put into the sewer also.

Before the construction of this new sewer which took wastes to an already inefficient Yardley S.D.W., all stations below the latter's outfall were devoid of life except for *C. riparius*, tubificids and other species of Chironomidae. *Lymnaea pereger* had been eliminated and only a little 'sewage fungus' and *Stigeoclonium* appeared nearer to the confluence where the dilution was greater. It was 1957 when Yardley S.D.W. started receiving the contents of the new sewer. The very high ammonia concentrations and B.O.D.s that followed as a result of overloading caused oxygen saturations of between 5 and 10% over the 11 km. to Station 11, where the saturation was then doubled. *C. riparius* was eliminated and tubificids were sparce until some self-purification and dilution had occurred by Station 10 when *C. riparius*, *Stigeoclonium*, and 'sewage fungus' appeared and tubificids increased in number.

The situation remained much the same until June 1971 when Yardley S.D.W. was closed although some improvements were seen above at Station 6 by 1970 - tubificidae and *Lymnaea* were abundant, *E. testacea* rare, *Asellus* rare, and increased numbers of chironomid species.

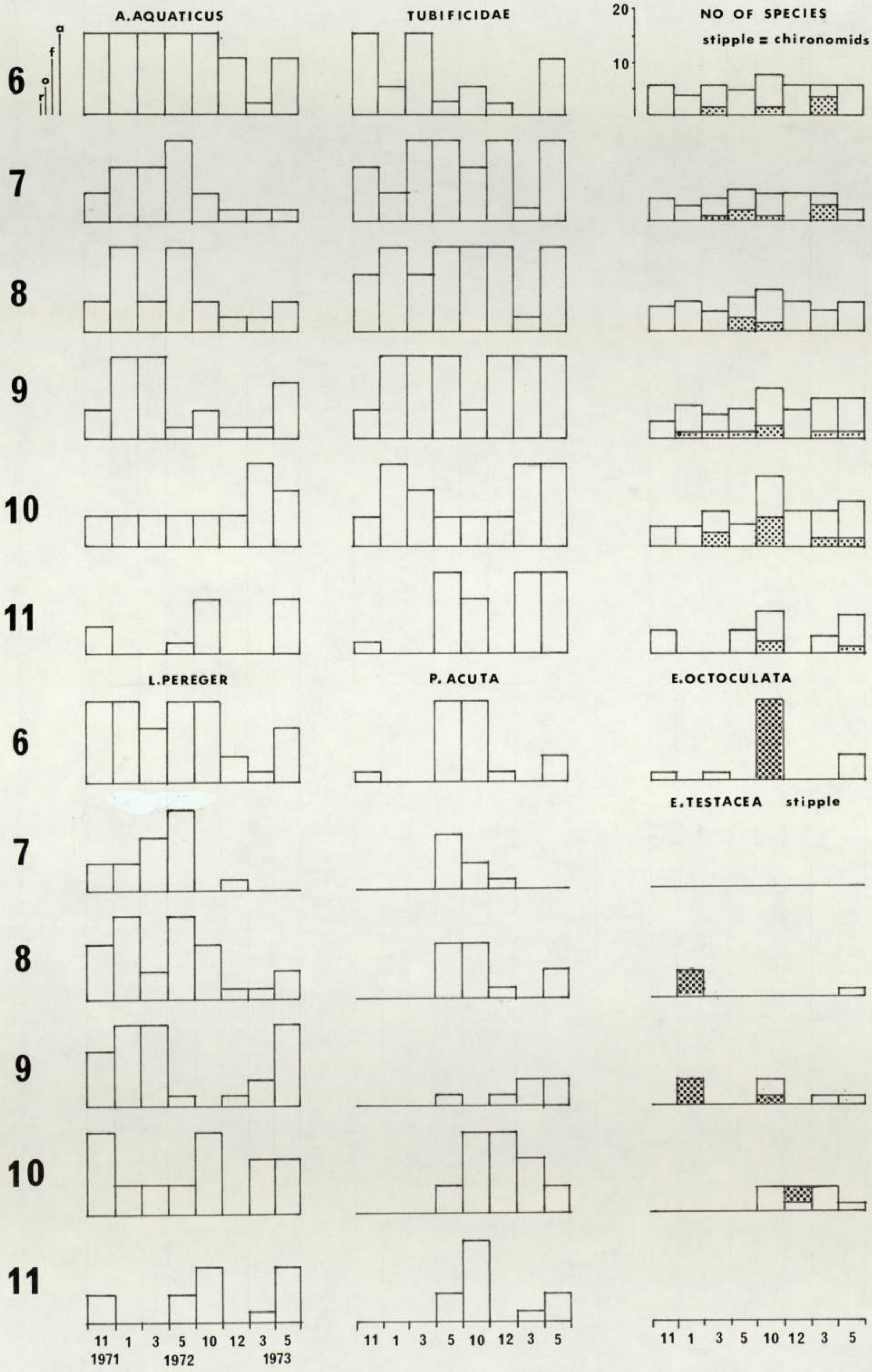
Presentation.

The presentation is essentially the same as that for the Upper Cole. Histograms (Fig. 4.29) have been drawn against the time of sampling for the dominant fauna (only 6 species). The remainder of the benthos together with the dominant algae and macrophytes remain in tabulated form (Tables 4.7 and 4.8). Station 11 was only accessible after driving across the River Blythe just above their confluence, and on three occasions when the R. Blythe was flooded no samples were obtained. The gaps in the histograms corresponding to the dates 19.1.72, 1.3.72, and 11.12.72. do not therefore indicate absence of species.

November 1971 onwards.

Station 6. Only five invertebrate species were detected since the previous survey of 1970 and only two of these were new arrivals - *Erpobdella octoculata* and *Physa acuta*.

FIG.4.29. Histograms showing total numbers of species and the relative abundances of 6 species at Stations 6-11 of the Lower Cole. The horizontal axis represents months of sampling.



These were only sparce to begin with, however, the former only being taken three times in the following 17 months. Physa acuta, on the other hand, became a dominant member of the scavenging population and proved to be, in 1972, a very formidable competitor with Lymnaea pereger and Asellus aquaticus (Fig. 4.29). The riffle population in 1970 was dominated by L. pereger and tubificids, while Asellus was only recorded as rare. Within the span of about one year the Asellus population had expanded enormously and it is seen from the histograms to be very stable and dominant along with L. pereger. Only towards the end of 1972 did the population of L. pereger look like declining from this competition but numbers increased again after the reduction of Asellus, which was thought to be the result of an industrial spill. The tubificid population also appeared to have declined over 1972 despite conditions conducive to their presence like the regular growths of 'sewage fungus' and Cladophora. Most notable at this Station was the presence in late 1972 onwards, of large growths of macrophytes and the absence of Gammarus. The restricted number of species was undoubtedly the result of intermittent spills of toxic wastes.

Station 7. Despite the fact that five months had elapsed since the closure of Yardley S.D.W., only four species (one of which was an unidentified dipteran larva) were present. The dominant species were Asellus, Lymnaea and Tubificidae which probably reflects the effect of residual toxicity upon the fauna: although the intermittent spills described above Station 6 may have played a part also. Cladophora, 'sewage fungus' and chironomid larvae were rarely recorded over the recovery study and leeches were absent. The appearance in considerable numbers of Physa acuta in May 1972 apparently reduced the populations of Lymnaea at all the Stations through competition, but most seriously at this one. The chironomid larvae Brillia longifurca, Thienemannimyia, Cricotopus (Cricotopus) albeforceps?, Eukiefferiella hospita and Cricotopus bicinctus all made unsuccessful attempts at colonisation but it is notable that they are all species that were tolerant to organic enrichment and were present in large numbers in the Upper Cole following the cessation of Houndsfield Lane effluent. It is therefore suggested that, despite the reduction in the concentration of metals by a factor of ten, the toxicity of the metals were responsible.

Previously unrecorded quantities of Stigeoclonium were seen covering the riffle in early 1973, the residual toxicity having prevented its appearance for nearly two years after the closure of the Works.

Station 8. This Station shows a similar pattern to Station 7 with Asellus, Lymnaea and tubificidae established as the dominant benthos but with increasing competition from Physa acuta which appears to have colonised this Station more substantially. E. testacea made an unsuccessful early attempt to colonise in 1972 and E. octoculata, in May 1973 also tried, and recent observations by the author (June 1974) have shown it to have been successful. Again the numbers of chironomid larvae are very small and collections made after heavy rain failed to show any. The pattern of algal growths are also similar to those of Station 7.

Station 9. Very small amounts of zinc, nickel and copper have been detected in Kingshurst Brook which flows into the Cole between this Station and Station 8. This tributary also contained fairly large numbers of *Gammarus* and *Baetis*, some Chironomidae and *Asellus* but no *C. riparius* and the dilution it afforded probably accounted for the slightly greater number of species found here. However the dominant fauna are still *Asellus*, *Lymnaea* and tubificidae despite the frequent drifting of *Baetis* and *Gammarus* that must occur. *E. octoculata* appears to have attained a background position with time, and *Baetis* and *Simulium* were taken in the first half of 1973 - nearly two years after the removal of the toxicity. The amount of *Cladophora* and number of chironomid larvae also increased during this final period.

Station 10. This Station is 9.9 km. below the Yardley S.D.W. and consequently self-purification and dilution allowed small growths of 'sewage fungus' and *Stigeoclonium* before June 1971 but the concentration of heavy metals was still sufficient to prevent all invertebrate life except for tubificids and a few *C. riparius*. After the removal of the effluent *C. riparius* disappeared and *Asellus* and *Lymnaea* again became dominant forms of invertebrate life together with the tubificidae. However, the swiftness of the current here may be responsible for the lower densities of the animals and scarcity of chironomid larvae compared with the other Stations above. *Physa acuta* achieved its highest numbers downstream of the relinquished effluent here, and *E. octoculata* established itself securely during 1973 which may have been responsible for only temporary populations of *E. testacea.* Large growths of *Cladophora* are now frequently found here.

Station 11. Before the cessation of the effluent, this Station supported large growths of *Stigeoclonium* and 'sewage fungus', tubificids and occasional chironomid larvae including *C. riparius*. Now, the apparance of 'sewage fungus' is a rarity although *Stigeoclonium* in winter and large blankets of *Cladophora* in summer are usual. The swiftness of the current obviously breaks off large clumps of *Cladophora* in which live many Naididae and Chironomidae who find it difficult to remain lodged between the stones. *E. octoculata* is thought to be sparce here because of the scouring effect of the current when the river is high. Despite the three occasions when sampling was impossible, it was obvious that the dominant fauna was that now characteristic to the Lower Cole, i.e. *Asellus*, *Lymnaea*, *Physa*, and Tubificidae.

4.2.3. Discussion.

The most explicit theme that emerges from the observations on the benthos of the Lower Cole is that of the reduced number of species, i.e. diversity. The chemistry has shown the water quality downstream of the redundant outfall to be of a reasonable standard with B.O.D.'s and ammonia concentrations low and dissolved oxygen ranging between 10.5 and 13.9 mgl^{-1}. The combination of reduced diversity and the chemistry described above precipitates the conclusion that it is the residual toxicity of the heavy metals that is responsible for the paucity of species. This residual toxicity may not only be suppressing species directly through poisoning, but secondarily as a result of reduced food supplies as algae and macrophytes are suppressed also. It appears, therefore, that those species that are successful under these conditions are those that are tolerant to the poisons and independent of algal and macrophytic growths for food supply.

The work on the speed of recovery of the Upper Cole communities has shown how new species can move into areas depleted of fauna quickly, providing the conditions are favourable and there is a supply of these species for recruitment. Areas for recruitment to the Lower Cole are present in the form of the Upper Cole, Kingshurst Brook, and the River Blythe where it confluences with the River Cole. <u>Lymnaea</u>, <u>Asellus</u> and tubificids have managed to move in relatively quickly to those areas depleted of life downstream of the outfall, whereas species like Gammarus, Baetis, many Chironomidae and whole taxa have not accomplished this although they too are present in the recruitment areas mentioned. Jones (1937) has shown the toxicity of many dissolved metallic salts to Gammarus (Amphipoda) and to the triclad Polycelis nigra - two species and taxonomic entities which are absent. He also determined the lethal toxicity thresholds of many other species that could explain the void of some species in the rivers Rheidol and Ystwyth (1940, 1949, 1958) and in particular the complete absence of Crustacea, Hirudinea, Oligochaeta and Mollusca. The levels of zinc he encountered in the Ystwyth varied between 0.7 and 1.2 mgl^{-1} (1940) and between 0.2 and 0.7 mgl^{-1} (1958) (depending upon the water level) and have only been matched in the Lower Cole before June 1971 and on average have been under 0.6 mgl^{-1}. However the effect of occasional spills, sometimes of greater magnitude than 1.2 mgl^{-1} Zn no doubt had profound effects on even the more tolerant fauna, preventing the establishment of populations for a considerable time. The absence of Gammarus is not therefore surprising even after removal of the toxic effluent as 0.4 mgl^{-1} Zn was taken in a spot sample in July 1972 and other higher levels may have been occurring.

The presence of the Mollusca is anomalous when considered in the light of Jones's work but the synergy of various low levels of metals and the hardness of the Cole water must have had an ameliorative effect, permitting

Lymnaea and Physa acuta to establish themselves. The hirudineans, which are sensitive to selective metal ions too, were shown as present at the lower Stations but only after one year elapsed from the time of effluent termination. It is difficult to know whether the residual and perhaps intermittent toxicity caused their absence over this long period but it is very likely their long life cycle coupled with relative immobility that prolonged the colonisation.

A similar explanation cannot be true for the distribution of Baetis rhodani which has only appeared infrequently at the bottom three Stations since the closure of Yardley Works. Its drifting and aerial mobility together with its presence in the neighbouring R. Blythe and Kingshurst Brook suggests that it is sensitive to the synergistic effects of the metals, or to intermittent spills of phenol that are known to have occurred. Jones (1958) found it abundantly in the Ystwyth where it was subjected to between 0.2 and 0.7 mgl^{-1} Zn. However, the dilution at Stations 9, 10 and 11 was great enought to allow it to inhabit the riffles there.

Other Insecta including Simulium sp. and the Chironomidae also showed a reluctance towards inhabitation despite the fact that Jones (1940) found that they were not restricted by zinc or traces of lead (0.05 mgl^{-1}). Again, the combined effects of the poisons may have been responsible although some Simulium spp., e.g. S. ornatum are rarely found in the lower reaches anyway (Maitland and

Penney, 1967), and chironomid larvae are very susceptible to being washed out of the substrate after heavy rain. However, it does appear that many chironomid larvae may be sensitive to metallic pollution and where the effects were palliated by dilution at the lower Stations, the numbers were reduced because of the wash out.

Whereas the dominant animal in the 'recovery zone' of an organically polluted river is invariably Asellus with Lymnaea pereger playing a subordinate scavenging role, the situation was reversed at Station 6 after the regular overflow of the sewer upstream (till 1957) had been stopped, so that only intermittent spills from factories were encountered. The tubificids and Lymnaea were then abundant and the Asellus population was recorded as rare, which was not altogether surprising as Jones (1937, 1940) demonstrated the sensitivity of malacostracan Crustacea to metals. However by November 1971 the Asellus population was again prolific but only sharing this dominance with Lymnaea pereger, so whether this second reversal was due to factories putting all their effluent down the new sewer thus removing the toxicant for Asellus, or whether some other cause is responsible is unknown. But below the redundant outfall the residual toxicity has not been great enough to prevent large densities of Asellus forming together with Lymnaea and later on, Physa acuta, the Mediterranean species only encountered in the Cole since the closure of Yardley Works. It is presumed that the population of Asellus only decreased at Stations 10 and 11, while *Lymnaea* retained more or less the same density as Stations above, because of its greater susceptibility to being dislodged from the substratum in high waters than the broad-footed *Lymnaea*.

Fish were known to be absent from Station 7 to the confluence under the regime of Yardley S.D.W. and the earliest comprehensive record (apart from occasional *Gasterosteus aculeatus* taken six months after closure) made in October 1972 by an undergraduate shows the variety of fish that are obviously unaffected by the low toxic level present.

Table showing the number of fish caught 50m either side of the Station. October 1972.

Station	*Gasterosteus aculeatus*	*Nemachilus barbatulus*	*Phoxinus*	*Pygosteus*	*Cottus gobio*	*Perca fluviatilis*	*Cyprinus*
7	11						
8	300	20	1	3			
9	100	8	100				
10	230						
11*	10	10			2	1	1

* only 50 m electrofished.

It would seem that the more sensitive fish are only present at Station 11 and probably eminate from the well stocked Blythe 100 yds. downstream at their confluence. It is fairly certain that Gasterosteus moved in almost immediately after June 1971 and its numbers reflect its tolerance to toxic materials. These fish, as Macan (1962) has suggested for Cottus, may keep Gammarus from inhabiting the lower reaches of a river.

In character with toxic pollution, the diversity and numbers of individual species are very low and constitute a measure of the residual toxicity and perhaps unknown intermittent discharges from isolated areas. The restricted increase in species from Stations 7 - 11 following the closure of Yardley Works would seem to be the result of the lightened toxic loading in the river, but the residual toxicity, and to an unknown extent the low level of contamination emanating from above Station 6 keeps the numbers down. Termination of the effluent then, together with dilution downstream allowed a fairly rapid but slight increase in the number of species at the Stations, and increases with time, due to immigration and reproduction following the drop below the toxicity threshold value of some species, are seen, - however the increase is very slow especially when compared with the recovery of the Upper Cole which demonstrates the totally different effects of a toxic effluent on the nature and speed of recovery.

4.3 CONCLUSIONS.

The suppression of the natural riffle faunas as a result of organic pollution and its replacement with a specialised community has been observed in the Upper Cole. Changes in the latter community have been studied over a period of seventeen months following the termination of the pollution source.

The suppression of fauna due to toxic discharges has been demonstrated in the Lower Cole together with its recovery observed over an eighteen month period following the termination of the major source of pollution six months before the study was commenced.

A number of factors influence the nature of the faunal recovery, many of which were common to both stretches of the river and are in all probability applicable to other rivers.

1) The rate of recovery is influenced by: -
    a) residual physical factors; e.g. 'sewage fungus' cover, silt etc.
    b) residual biological factors; e.g. existing high density populations of a few species.
    c) the background level of pollution (if any) after cessation of the effluent.
    d) the time of year that the effluent is abated.
    e) the residual colony, no matter how small, which may expand rapidly.
    f) available sources for the recruitment of species.

- g) the decrease in flow and consequent temperature fluctuations in the river following effluent cessation if the latter forms a considerable part of the flow.
- h) the biological characteristics of the species;
  - i) voltinism
  - ii) mobility
  - iii) sexual or asexual reproduction.

2) Population and depopulation with recovery. *Micropsectra atrofasciatus* and *Prodiamesa olivacea* are particularly fast colonisers. Those species like *B. rhodani* which are hampered by residual physical factors are slower. Equally fast is the decline of the very tolerant populations like *Chironomus riparius* and Tubificidae.

3) Changes in the balance of trophic levels occur; there is:

- a) increase in herbivores with recovery.
- b) increase with carnivores with recovery.
- c) reduction in detrital feeders with recovery.
- d) increase in primary producers as measured by the increase in herbivors like *Baetis rhodani* and some of the Orthocladiinae, accepting that a small proportion of their diet may be of allochthonous origin.

4) Stability of the ecosystem; there is an increase in the diversity of species with recovery and the relative stability of the riffle benthos may possibly be measured

by the % composition of chironomid larvae.

5) Biological indicators: -

- a) the following relationships, if measured quantitatively, may be useful as indicators of organic pollution: - Asellus/Gammarus, E. octoculata/E. testacea, chironomid 'Species A'/chironomid 'Species B'.
- b) the following chironomid larvae may be useful indicators of organic pollution: -
  - i) gross: C. riparius, B. longifurca and Procladius spp.
  - ii) less severe: C. (Cricotopus) albeforceps? and E. hospita.
  - iii) mild: when in large numbers, P. olivacea, M. atrofasciatus, C. bicinctus and T. lentiginosa, but all are tolerant to the harsher conditions of ii).

Recovery with particular relevance to toxic (metallic) pollution as seen in the River Cole: -

6) Successful species are those tolerant of low level metal pollution whose food supply is independent of primary producers, i.e. mainly scavengers - Asellus, Lymnaea, Physa and tubificids.

7) The effects of occasional spills of greater magnitude than the background can have profound effects upon even the more tolerant fauna preventing establishment of populations for a considerable time.

8) Physa acuta has proved itself to be tolerant to low levels of many metal ions. The occurrence of this Mediterranean species is rather exceptional in that it is usually confined to warmer waters (Macan 1960).

9) It appears that many chironomid larvae are sensitive to low level metal contamination.

10) A reversal of the Asellus/Lymnaea association with toxic contamination leaving Lymnaea dominant, has been demonstrated, and on reduction of the toxic level Asellus has returned to dominance or co-dominance.

11) In character with toxic pollution the diversity and numbers of individual species are both low.

12) The recovery from toxic effluents is markedly slower than recovery from organic effluents due to the residual toxicity in the bottom deposits.

Table illustrating the major changes in benthic invertebrate communities as a result of the discontinuation of Houndsfield Lane effluent.

Dominant fauna

	BEFORE	AFTER May 1972 - Sept. 1973
Station 2.	Tubificidae and C. riparius	B. rhodani, Tubificidae, M. atrofasciatus, C. bicinctus, Thienemannimyia, P. olivacea, C. (Cricotopus) albeforceps?.
	Small populations of: B. longifurca, P.olivacea, E. hospita, C. (Cricotopus) albeforceps?.	Small populations of: Gammarus Asellus and Leeches.
Station 3.	Tubificidae, C. riparius, C. (Cricotopus) albeforceps?, B. longifurca, Enchytraeidae	B. rhodani, Tubificidae, Gammarus, Asellus, M. atrofasciatus, Thienemannimyia, P. olivacea, C. bicinctus.
	Small populations of: B. rhodani, E. hospita, M. atrofasciatus, P. olivacea.	Small populations of: E. octoculata, E. testacea, G. complanata, L. pereger.
Station 4.	Tubificidae, Asellus, E. octoculata, H. stagnalis, E. hospita, B. longifurca, C. (Cricotopus) albeforceps?.	B. rhodani, Tubificidae, Asellus, Gammarus, P. olivacea, C. bicinctus, Thienemannimyia, G. complanata, E. testacea.
	Small populations of: M. atrofasciatus.	Small populations of: B. longifurca, L. pereger.
Station 5.	Tubificids, E. octoculata, Asellus, B. longifurca, Thienemannimyia, C. bicinctus, E. hospita, P. olivacea, C.(Cricotopus) albeforceps?	B. rhodani, Tubificidae, Asellus, Gammarus, E. testacea, G. complanata, C. bicinctus, Thienemannimyia, P. olivacea, C.(Cricotopus) albeforceps?
	Small population of: B.rhodani.	

TABLE 4.1.

Biological results for Station 1 of the Upper Cole.

a = abundant f = frequent o = occasional r = rare.

	1972									1973							
Date	26.1	8.3	3.5	24.5	30.5	8.6	21.7	3.10	16.11	12.1	21.2	19.3	11.4	7.5	4.6	9.7	3.9
A. aquaticus		o							o	r					r		
G. pulex		f	f			r		f	f	a	f	f	a	a	a	a	a
G. complanata		r				r	o	o		r		f	r	o	o		
E. octoculata	o	f	a			r	r	f	o	o	r	o	f	a	f	f	o
L. pereger	o																
S. corneum	a	o	r			r		o		o		r	o	o			
Pisidium			r			r					f				r	r	
Tubificidae	o	o	a					o	r	o	r	r	o	o	r		o
Naididae									f	o	f	r	o	f	r		
B. rhodani	f	a	a			r		f	a	a	a	a	a	a	a	f	a
A. sulcicollis	o		r								r	o	o	o			
Sialis		r							r								
H. angustipennis	r	o	o			r		a	f		r		r	o	o		o
A. fluviatile			o							o					r	o	
Tipulidae	r								o	o	o					o	
S. ornatum						r		f	o	f	f	o	f		f	o	
Coleoptera l.								r									
Corixa sp.								r									
Limnephilus sp.								r	r					r			

continued ...

TABLE 4.1.

(continued)

				1972									1973				
Date	26.1	8.3	3.5	24.5	30.5	8.6	21.7	3.10	16.11	12.1	21.2	19.3	11.4	7.5	4.6	9.7	3.9
Hydracarina									o		r				o	o	
Ephydridae											r						
C. riparius							a										
P. olivacea		o							o	r							
B. modesta	o								o	r		r					
P. arundineti								f							o		
T. lentiginosa	o		f						o		r			f	r		
M. atrofasciatus	f					f		a	a	o	a	a		f	f		
E. hospita	o								r	o	a	a	o		o		
C. albeforceps?	o	o						f	r		f	o		o			
C. bicinctus	o		o			f									r		
Rheocricotopus	o										a	a		r	r		
Eukieff.sp.C2.									r								
D. campestris										r	o	r					
D. cultriger										r							
C.(isocladius)sp.											o	f	a	f			

TABLE 4.2.

Biological results for Station 2 of the Upper Cole.

a = abundant f = frequent o = occasional r = rare

vr = very rare

	1972								1973							
Date	26.1	8.3	3.5	24.5	30.5	8.6	21.7	3.10	16.11	12.1	21.2	19.3	11.4	7.5	4.6	9.7	3.9
A. aquaticus	r								o						r	f	r
G. pulex		r							o			r				o	
E. octoculata							r							r	r		o
E. testacea													o		r		
G. complanata									r				o	r		r	
L. pereger									r								
S. corneum									r							r	
Tubificidae		o	r	vr	vr		r	f	o	o	a	a	a	a	a	f	o
Naididae							f		a	o			o	o	a		
Lumbricillus		r					a										
B. rhodani	o							r	f	o	r	o	f	a	o	f	a
A. sulcicollis												r					
Sialis									r								
Tipulidae								r	r		r					r	
S. ornatum								o		o		o	o		o		f
Coleoptera l.									r								
P. tenuis											r						
H. stagnalis																a	

continued ...

TABLE 4.2

(continued)

	1972									1973							
Date	26.1	8.3	3.5	24.5	30.5	8.6	21.7	3.10	16.11	12.1	21.2	19.3	11.4	7.5	4.6	9.7	3.9
C. riparius	a	a	r	vr	f	o	f	o	a	a	r	r	r		r		
P. olivacea	a							o	a		o	f	r				
B. longifurca					f		f					r					
B. modesta											r	r		r			
P. arundineti	a								r				o	f			
T. lentiginosa			r				a		a		r	r	f	f			
Procladius sp.						o	r		a			r					
M. atrofasciatus						r	o	a	f		a	a	o		a		
E. hospita								r		r		o	a		r		
C. albeforceps?		o	r					o			a	a	o	r	o		
C. bicinctus					f	o	a	f				r	o	r	a		
C. fuscus							o										
Rheocricotopus								o	r		r						
Eukieff. sp. C2												r					
D. campestris												r					
D. cultriger											r						
P. scalaenum										r							
C. (isocladius) sp.												r		r			
Orthocladius sp. C10															o		

TABLE 4.3

Biological results for Station 3 of the Upper Cole.

a = abundant f = frequent o = occasional r = rate.

					1972								1973				
Date	26.1	8.3	3.5	24.5	30.5	8.6	21.7	3.10	16.11	12.1	21.2	19.3	11.4	7.5	4.6	9.7	3.9
A. aquaticus	r		r		r	r	r	o	a	f	o			r	r	o	o
G. pulex	r	f	o	f	o		a	f	a	f	o	f	f	o	f	f	a
E. octoculata								o	o		r	o	r	o	a	r	r
E. testacea									o	o	o	o	f	r			
G. complanata								r	o	r		r	o	r	o		r
H. stagnalis																r	
L. pereger				o	r				f	o	o			o		r	
S. corneum				r					o		r		r		r		r
Pisidium								r		r							
Tubificidae	a	a	a	a	o	f	a	f	o	f	f	a	a	f	o	f	o
Naididae		r		a	o			r		o	o	f	f	f	o		
B. rhodani	r	a	f	f	f	r	f	a	a	a	f	f	f	o	f	o	
A. fluviatilis																	r
Tipulidae	r								r								
S. ornatum		f					o	a	o	o		o	o				
Coleoptera l.																	r
Limnephilus sp.								r									r
Hydracarina				f	o												
Ephydridae																r	

continued ...

TABLE 4.3

(continued)

	1972									1973							
Date	26.1	8.3	3.5	24.5	30.5	8.6	21.7	3.10	16.11	12.1	21.2	19.3	11.4	7.5	4.6	9.7	3.9
P. tenuis									r						r	r	
H. jenkinsii								r									
Lumbriculidae							r	r				o	o				
C. riparius	a	f	f	a	f	r			r	f	r						
P. olivacea						r	a	o	f	a	o	o	f	f			
B. longifurca		o									r	o	r	o			
B. modesta		r											r	r			
P. arundineti					f		a	o	o	f	f	r		o	f		
T. letiginosa			o	f	f	a			f	r		o	o	a	a		
Procladius sp.						o	f		o	r	r						
M. atrofasciatus						a		o		r		o			f		
E. hospita											o		r				
C. albeforceps?												o	f				
C. bicinctus			o	f	f	a							r	f	a	a	
Rheocricotopus														o			
D. cultriger						r											
C. (isocladius) sp.											r	r					
Orthocladiinae sp. C10													o				
Orthocladiinae sp. C11													f				

TABLE 4.4

Biological results for Station 4 of the Upper Cole.

a = abundant f = frequent o = occasional r = rare vr = very rare

	1972									1973							
Date	26.1	8.3	3.5	24.5	30.5	8.6	21.7	3.10	16.11	12.1	21.2	19.3	11.4	7.5	4.6	9.7	3.9
A. aquaticus	f	o	o		r	f	o	o	a	f	f			f	o	o	o
G. pulex	vr	o	f	r	r	f	a	o	a	f	o	f	o	o	o	a	a
E. octoculata		a	f		o											o	
E. testacea				f		a	o	f	a	o	o	f	f	f	a		r
G. complanata					r			r	r	r		f		r	r	r	o
L. pereger								r	r		r			r	r		
P. acuta									o							r	
Tubificidae	a	r	f	f	r	o	o	o		o	r	r	f	r		o	r
Naididae		o								o	r	r		r			
B. rhodani		f	f	o	a	a	o	o		f	f	a	o	o	f	a	f
Limnephilus sp.								r									
Hydracarina										r							
Lumbriculidae										r							
C. riparius			r	o				r									
P. olivacea	o						f	o	f	a	o	r					
B. longifurca						o	f		f	r	o	r	r				

continued ...

TABLE 4.4.

(continued)

					1972								1973				
Date	26.1	8.3	3.5	24.5	30.5	8.6	21.7	3.10	16.11	12.1	21.2	19.3	11.4	7.5	4.6	9.7	3.9
B. modesta					a				r		o						
P. arundineti						o		o	r					r			
T. lentiginosa				f			f		a	f	a	o	o	o			
Procladius sp.							f										
M. atrofasciatus											o	r					
E. hospita												o					
C. albeforceps?			r					o			f	a					
C. bicinctus			f	f	a	f	a					o	a	a	a		
Rheocricotopus												f			o		
D. cultriger										r			r				
Orthocladiinae sp. C10															r		

<u>TABLE 4.5</u>

<u>Biological results for Station 5 of the Upper Cole</u>

a = abundant f = frequent o = occasional r = rare.

					1972									1973			
Date	26.1	8.3	3.5	24.5	30.5	8.6	21.7	3.10	16.11	12.1	21.2	19.3	11.4	7.5	4.6	9.7	3.9
A. aquaticus	a	f	a	a	f	a	a	f	a	a	a	a	o	a	a	a	f
G. pulex	o	f	o	f	f	f	f	f	a	a	a	a	f	o	f	a	a
E. octoculata		f	o									a					r
E. testacea	o			o	f	f	o	o	o	o	f		a	a	f	o	
H. sanguisuga													r				
G. complanata	o	o	o		r	r	r	o	a		f				o	o	
S. corneum											r			r		o	
Tubificidae	f	f	r	o	r	o	o	r	o	f		o	f	r	r		r
Naididae	f			f	r	o	r	r	r	o		o	o	r	r		
Lumbriculidae	o					o											
B. rhodani		o	o		o	o		r		o	f	a	o	o			o
A. fluviatile					r	r						r				r	r
Tipulidae										r							
Coleoptera 1											r						
L. pereger																r	
G. aculeatus																	f
C. riparius			r	o													

continued ...

TABLE 4.5
(continued)

	1972									1973							
Date	26.1	8.3	3.5	24.5	30.5	8.6	21.7	3.10	16.11	12.1	21.2	19.3	11.4	7.5	4.6	9.7	3.9
B. longifurca	a	f				a	f			o		r					
P. olivacea	a	f			a	a		a	a	a	o						
T. lentiginosa		f		a	a	a	a		f	a	a	o	r	f	f		
C. albeforceps?			o		a			a				f		f			
B. modesta				o													
C. bicinctus					a					a	f		o	a			
Procladius sp.							f	r	f								
C. fuscus								o						o			
M. atratulus								r									
E. hospita										r	o						
M. atrofasciatus											r	r					
P. scalaenum											r						
Rheocricotopus												r	r		r		

TABLE 4.6

Biological results for Stations 6, 7, 8 and 9 of the Lower Cole

a = abundant f = frequent o = occasional r = rare.

	STATION 6								STATION 7							
	1971			1972			1973		1971			1972			1973	
Date	10.11	19.1	1.3	10.5	18.10	11.12	21.3	25.5	10.11	19.1	1.3	10.5	18.10	11.12	21.3	25.5
A. aquaticus	a	a	a	a	a	f	r	f	o	f	f	a	o	r	r	r
E. octoculata	r		r					o								
E. testacea					a											
L. pereger	a	a	f	a	a	o	r	f	o	o	f	a		r		
Tubificidae	a	o	a	r	o	r		f	f	o	a	a	f	a	r	a
Naididae	f					o	r						f	f		
P. acuta	r			a	a	r		o				f	o	r		
C. albeforceps			r				r								r	
B. longifurca											r					
P. olivacea												r				
T. lentiginosa												r				
C. fuscus					o		r						r			
E. hospita							r								r	
C. bicinctus															r	
Cladophora	f			r	r	r			r							
'sewage fungus'	f		a			f	a				f					
Stigeoclonium				r		a									a	
Ranunculus					f			f								
Potamogeton					f	r		f								

continued ...

TABLE 4.6

(continued)

	STATION 8								STATION 9							
	1971			1972			1973		1971			1972			1973	
Date	10.11	19.1	1.3	10.5	18.10	11.12	21.3	25.5	11.10	19.1	1.3	10.5	18.10	11.12	21.3	25.5
A. aquaticus	o	a	o	a	o	r	r	o	o	a	a	r	o	r	r	f
G. pulex		r								r						
E. octoculata								r					o		r	r
E. testacea		o								o			r			
L. pereger	f	a	o	a	f	r	r	o	f	a	a	r		r	o	a
Tubificidae	f	a	f	a	a	a	r	a	o	a	a	a	o	a	a	a
Naididae	r				o	o								o	o	
Tabanidae			r													
P. acuta				f	f	r		o				r		r	o	o
Elmidae					r											
Hydracarina													r			
B. rhodani													f			r
S. ornatum										o	r					
P. olivacea				r									r			
T. lentiginosa				r												
C. albeforceps?												r	a			
C. fuscus					r											
C. bicinctus															f	f

TABLE 4.7

Biological results for Stations 10 and 11 of the Lower Cole

a = abundant f = frequent o = occasional r = rare.

	STATION 10								STATION 11							
	1971			1972			1973		1971			1972			1973	
Date	10.11	19.1	1.3	10.5	18.10	11.12	21.3	25.5	10.11	19.1	1.3	10.5	18.10	11.12	21.3	25.5
A. aquaticus	o	o	o	o	o	o	a	f	o			r	f			f
G. pulex									r							
E. octoculata					o	r	o	r								r
E. testacea						o										
L. pereger	a	o	o	o	a		f	f	o			o	f		r	f
P. acuta				o	a	a	f	o				o	a		r	o
Tubificidae	o	a	f	o	o	o	a	a	r			a	f		a	a
Naididae					o	o							r			
B. rhodani					r			o		no sample	no sample		o	no sample		o
P. olivacea			o		f								o			
T. lentiginosa			o			r										
C. albeforceps					o											
C. fuscus					o								r			
C. trifascia					r											
Procladius sp.					r											
C. bicinctus								f								f
Hydracarina					o											
Cladophora	f				f			f	o			a				f
Stigeoclonium	f						a		o						a	
Culcidae			r													

TABLE 4.8

Physical and chemical data for Station 1 of the Upper Cole. Means and standard errors for the year 30.5.72 - 7.5.73.

Date	pH	^{o}C	S.S.	D.O.	% sat.	Cl^{-}	Alk.	NH_3	T.I.P.	O.N.	P.V.	B.O.D.	HDNS.
1972													
26th Jan.	7.0	6.0	18	10.2	81.7	32	415	0.75	0.7	4.7	10.8	8.0	146
8th Mar.	6.95	6.0	11.5	11.0	88.1	29	55	0.3	0.2	3.8	6.6	1.5	162
3rd May	7.6	11.0	12.0	14.8	134.5	41	65	0.4	0.3	3.8	4.8	5.0	200
30th "	8.1	13.0	15	11.7	110.4	40	100	0.25	0.25	4.1	5.8	5.7	186
8th June	6.6	13.0	35	3.4	32.0	42	130	0.65	1.15	3.3	47.6	167.5	206
21st July	7.4	14.0	6	4.0	38.6	35	150	0.3	0.4	2.7	5.6	15.0	190
3rd Oct.	7.3	11.0	20	8.1	73.6	33	110	0.2	0.5	5.8	–	0.9	–
16th Nov.	7.1	5.0	30	11.8	92.4	34	95	ngl	0.5	7.4	–	2.1	–
1973													
12th Jan.	7.1	3.0	10	12.8	95.0	38	100	0.5	0.7	6.7		1.2	
21st Feb.	7.0	7.0	13	12.6	103.8	37	85	0.4	0.4	6.6		2.4	
19th Mar.	8.2	8.5	9	16.9	146.3	33	100	0.4	0.4	6.1		3.9	
11th Apr.	7.0	6.0		12.1	97.2	43	90	1.1	1.2	5.8			
7th May	8.1	12.0		13.9	129.0	34	60	–	1.7	6.5			
4th June		12.0	5	11.7	108.6	32	100	–	1.9	4.5		2.0	
9th July	7.0	16.0	–	8.6	87.3	33	–	–	–	4.8			
3rd Sept.		16.5		5.9	61.1			0.6	0.8	4.8			
Mean	7.39	9.25	17.25	10.73	91.8	36.9	102	0.42	0.72	5.5		24.8	
S.E.	0.16	1.22	3.67	1.35	11.4	1.2	7.75	0.1	0.14	0.48		20.4	

TABLE 4.9.

Physical and Chemical Data for Station 2 of the Upper Cole.

Means and standard errors for the year 30.5.72. - 7.5.73.

Date	pH	°C	S.S.	D.O.	% Sat.	Cl^-	Alk.	NH_3	T.I.P.	O.N.	P.V.	B.O.D.	HDNS
1972													
26 Jan.	7.0	6.0	36	9.7	77.7	48	375	1.5	1.0	3.7	10.8	7.5	160
8 Mar.	6.8	6.0	25	9.7	77.7	41	110	1.7	0.55	3.8	8.0	8.1	182
3 May	6.9	11.0	39	7.0	63.6	37	125	4.3	1.3	3.4	7.8	20.5	194
24 "	7.4	13.0	12	10.4	98.1	46	150	1.4	0.6	4.3	6.1	5.7	206
30 "	8.1	13.0	48	13.4	126.4	43	155	1.3	0.35	4.1	6.0	16.8	206
8 June	7.0	12.0	21	1.6	15.0	55	165	1.0	0.5	2.9	15.0	35.0	214
21 July	7.3	16.0	7	5.2	52.5	42	165	1.3	0.6	7.8	3.2	4.0	210
3 Oct.	7.3	13.0	37	8.9	83.9	36	110	0.2	0.5	3.8	-	2.1	-
16 Nov.	7.1	5.0	27	10.5	82.2	153	85	ngl	0.4	7.1	-	11.4	-
1973													
12 Jan.	7.0	3.0	12	9.5	70.6	36	110	1.4	6.3	4.1		9.6	
21 Feb.	7.2	7.0	53	8.9	73.4	41	120	3.2	0.6	4.8			
19 Mar.	7.5	8.0	18	10.5	88.6	33	105	0.3	0.4	5.8		3.3	
11 Apr.	7.0	6.5		7.0	57.7	57	125	4.5	0.7	4.7			
7 May	7.8	12.0		12.8	119.0	43	90	1.0	1.7	6.0			
4 June		13.0	11	11.0	104.5	47	150	-	1.9	2.9		5.1	
9 July	7.2	17.0	3	8.1	83.9	33	115	-	1.7	2.8		3.0	
3 Sept		16.5		7.5	77.7			0.7	0.8	2.9			
Mean	7.33	9.6	27.8	8.83	76.9	53.9	123.0	1.42	1.2	5.11		15.27	
S.E.	0,1	1.32	5.9	1.1	10.15	11.28	9.28	0.43	0.57	0.47		5.16	

TABLE 4.10

Physical and Chemical Data for Station 3 of the Upper Cole.

Means and standard errors for the year 30.5.72 - 7.5.73.

Date	pH	^{o}C	S.S.	D.O.	% Sat.	Cl^-	Alk.	NH_3	T.I.P.	O.N.	P.V.	B.O.D.	HDNS.
1972													
26 Jan.	7.1	6.0	37	9.9	79.3	48	350	1.1	0.7	3.6	10.4	5.0	170
8 Mar.	6.9	6.0	12.5	9.9	79.3	45	110	1.4	0.4	4.9	7.0	7.2	196
3 May	7.0	11.0	24	5.7	51.8	48	125	4.8	1.2	2.9	7.4	14.0	182
24 "	7.8	13.0	7	12.1	114.1	56	140	1.1	0.5	3.6	5.0	2.4	218
30 "	8.3	13.0	12	14.2	133.9	57	145	0.7	0.4	3.4	8.6	7.2	220
8 June	7.1	12.0	20	2.9	26.7	61	155	1.2	0.4	2.9	14.0	29.0	238
21 July	7.2	16.0	10	4.8	48.2	69	170	3.3	1.0	2.9	2.4	4.2	250
3 Oct.	7.3	12.0	6	8.1	74.7	62	130	0.2	0.5	3.5		0.9	
16 Nov.	7.1	5.0	19	10.6	83.0	97	90	ngl	0.4	5.4		5.1	
1973													
12 Jan.	7.1	3.0	13	10.0	74.3	47	120	2.3	0.6	6.1		9.0	
21 Feb.	7.2	7.0	12	9.9	81.6	55	130	0.4	0.4	5.1		2.1	
19 Mar.	7.5	7.5	14	11.3	95.4	54	135	0.3	0.5	5.4		2.1	
11 Apr.	7.0	7.0	–	10.4	85.7	107	125	1.7	0.5	3.8		–	
7 May	7.6	11.0	–	12.1	109.8	52	95	0.9	1.6	5.2		–	
4 June		12.0	9	8.2	76.1	55	140	–	2.0	3.1		6.9	
9 July	7.4	16.0	1	8.3	84.3	57	150	–	2.3	2.7		2.1	
3 Sept	–	16.0	–	6.6	67.0	–	–	0.6	0.6	3.0		–	
Mean	7.3	9.35	13.3	9.43	81.3	66.1	129.5	1.1	0.63	4.37		7.45	
S.E.	0.1	1.28	1.6	1.06	9.4	6.3	7.76	0.33	0.1	0.37		3.22	

TABLE 4.11.

Physical and chemical data for Station 4 of the Upper Cole.

Mean and standard errors for the year 30.5.72 - 7.5.73.

Date	pH	^oC	S.S.	D.O.	% Sat.	Cl^-	Alk.	NH_3	T.I.P.	O.N.	P.V.	B.O.D.	HDNS
1972													
26 Jan.	7.3	6.0	94	10.0	80.1	56	300	0.8	0.7	2.5	11.4	3.5	212
8 Mar.	7.2	6.0	12	10.6	84.9	40	115	0.5	0.2	3.9	6.0	2.4	200
3 May	7.3	11.0	334	8.1	73.6	51	150	2.7	0.9	2.7	13.4	9.2	224
24 "	7.4	13.0	20	9.4	88.6	48	180	0.7	0.4	3.9	7.2	3.0	220
30 "	8.2	13.0	86	11.2	105.6	56	145	0.4	0.3	3.9	5.2	6.3	236
8 June	7.5	12.0	30	8.3	76.6	52	160	0.4	0.3	3.4	6.6	6.9	230
21 July	7.6	16.0	6	7.4	74.4	57	160	2.1	0.7	3.1	3.2	2.4	264
3 Oct.	7.3	12.0	48	9.1	84.0	56	135	0.2	0.5	4.9		1.5	
16 Nov.	7.2	5.0	36	10.8	84.6	120	105	0.6	0.3	4.8		4.5	
1973													
12 Jan.	7.4	3.5	9	12.6	96.1	57	135	0.9	0.8	6.2		2.1	
21 Feb.	7.5	6.5	16	11.0	90.7	57	150	0.4	0.5	5.5		1.8	
19 Mar.	7.9	7.5	15	14.4	121.6	56	155	0.4	0.4	4.5		3.6	
11 Apr.	7.9	8.0	-	12.3	103.9	67	140	0.7	0.4	4.3		-	
7 May	8.1	10.0	-	14.4	127.6	50	110	0.3	1.6	4.5		-	
4 June	-	14.0	15	11.6	112.7	49	145	-	1.6	3.4		4.8	
9 July	8.1	17.0	8	12.8	132.6	51	155	-	1.9	3.2		3.0	
3 Sept.	-	17.0	-	6.9	70.5	-	-	0.6	0.4	3.7		-	
Mean	7.66	9.35	30.75	11.15	86.1	62.8	139.5	0.64	0.58	4.51		3.63	
S.E.	0.1	1.23	9.38	0.74	10.95	6.5	6.07	0.17	0.10	0.28		0.74	

TABLE 4.12

Physical and Chemical Data for Station 5 of the Upper Cole.

Means and standard errors for the year 30.5.72 - 7.5.73.

Date	pH	^{o}C	S.S.	D.O.	% Sat.	Cl^{-}	Alk.	NH$_3$	T.I.P.	O.N.	P.V.	B.O.D.	HDNS
1972													
26 Jan.	7.4	6.0	74	9.4	75.3	68	350	0.6	0.6	1.9	10.0	2.5	194
8 Mar.	7.3	6.0	14.5	10.6	84.9	43	110	0.4	0.4	4.3	5.6	2.4	210
3 May	7.3	11.0	192	7.5	68.1	50	165	2.6	1.3	2.9	9.4	8.7	206
24 "	7.6	13.0	11	10.5	99.0	46	155	0.6	0.5	3.5	4.2	2.4	208
30 "	8.0	12.0	11	11.6	107.1	57	150	0.3	0.4	4.2	6.4	4.5	256
8 June	7.5	11.5	11	7.8	71.0	52	160	0.4	0.2	3.4	5.4	4.8	246
21 July	7.3	17.0	4	6.8	69.8	58	175	0.6	0.7	6.2	2.8	2.7	280
3 Oct.	7.3	11.0	103	8.5	77.2	57	140	0.1	0.3	4.9		6.0	
16 Nov.	7.2	5.0	18	10.9	85.4	198	100	0.3	0.3	4.9		3.9	
1973													
12 Jan.	7.4	3.5	12	12.1	92.3	52	150	0.9	0.5	6.3		2.1	
21 Feb.	7.5	6.5	20	11.2	92.3	56	150	0.4	0.2	5.6		2.4	
19 Mar.	7.9	7.5	13	13.3	112.3	55	150	0.3	0.4	4.7		3.6	
11 Apr.	7.8	9.0		11.5	99.6	86	140	0.6	0.4	6.4		–	
7 May	8.1	9.5		14.5	141.0	48	105	0.3	1.6	ngl.			
4 June	–	13.0	12	11.0	104.5	48	150	–	1.7	2.9		4.2	
9 July	8.1	16.0	4	11.5	109.2	52	155	–	1.4	3.7		3.6	
3 Sept	–	16.0		6.8	69.0	–	–	0.5	0.4	3.2		–	
Mean	7.6	9.25	24.0	10.82	94.8	71.9	142.0	0.42	0.5	4.66		3.75	
S.E.	0.1	1.23	11.41	0.76	6.84	14.39	7.3	0	0.1	0.6		0.46	

TABLE 4.13

Lower Cole Chemistry.

Mean monthly figures between dates shown.

T^oC	D.O.	% Sat.	pH	Tot Metals	S.S.	P.V.	BOD	NH_3	ON	Fe	Cr	Cu	Ni	Zn	Date
STATION 6*															
13.1	9.9	98	7.5	0.38	12	3.8	4.7	2.0	5.4	0.85					Aug 70/Jly 71
10.9	7.4	68.7	7.5	0.36	15	11.3	11.5	3.1	4.3	0.98					1969/70
STATION 7*															
11.0	5.4	49.6	7.1	1.4	60	19.7	27.3	8.2	8.3	2.14					1969/70
14.5	7.7	79.5	7.5	1.4	30.5	10.6	20.0	7.0	11.7	1.22					Aug 70/Jly 71
										0.30	0.027	0.02	0.025	0.09	July 72.
STATION 8+															
13.8	4.32	41.9	7.3	1.7	-	16.6	23.3	7.1	7.3	5.65	0.24	0.36	0.60	0.5	Sept 68/Aug 70
STATION 9+															
12.8	6.1	57.9	7.4	1.01	-	10.5	11.7	4.9	6.1	2.41	0.13	0.20	0.37	0.31	Sept 68/Aug 70
				0.19							0.025	0.025	0.018	0.118	July 1972
STATION 10															
				0.62							0.095	0.07	0.05	0.04	
STATION 11															
				0.08							0.024	0.027	0.02	0.01	July 72
STATION 6															
											0.032	0.025	0.033	0.123	July 72

* = Data of City of Birmingham Public Works Dept.
\+ = Data from Martin 1973.

TABLE 4.14

Physical and Chemical Data for Stations 6-11 of the Lower Cole.

Date	pH	^{o}C	S.S.	D.O.	% Sat.	Cl^-	Alk.	NH_3	T.I.P.	O.N.	P.V.	B.O.D.	HDNS
STATION 6.													
10.11.71		5.0		10.1	78.9								
19.1.72	7.2	6.5	123	10.3	82.5	54	80	0.5	0.5	2.3		13.0	192
1.3.72	7.0	6.5	115	9.6	77.0	40	95	0.8	0.5	2.9	10.8	13.0	154
10.5.72	6.8	13.0	34	9.0	84.9	42	125	1.2	0.6	3.8	7.2	5.7	220
18.10.72	7.1	12.0	5	7.0	64.9	44	130	0.4	0.5	42		2.1	
11.12.72	7.0	4.5	48	10.7	83.8	40	60						
21.3.73	7.0	8.0	8	5.6	47.3	50	140	0.4	0.4	4.3		10.2	
25.5.73	7.0	12.0	48	7.0	64.9	48	135		1.7	6.2		6.3	
Mean	7.0	8.4	54.4	8.6	72.9	45	93.1	1.0	0.7	3.9	9.0	6.6	-
STATION 7.													
10.11.71		4.0		12.3	93.9								
19.1.72	7.2	6.5	123	9.5	76.1	59	105	0.3	0.4	2.4		6.0	200
1.3.72	6.9	6.5	105	9.5	76.0	48	85	0.8	0.4	2.6	11.2	13.0	160
10.5.72	7.4	13.0	33	9.8	92.5	38	125	1.0	0.4	4.1	7.0	5.4	196
18.10.72	7.5	11.0	5	10.4	94.4	43	130	0.4	0.5	4.1		2.7	
11.12.72	7.1	4.5	47	10.3	80.7	41	70						
21.3.73	7.9	7.0	28	156	128	49	150	0.4	0.4	4.4		7.2	
25.5.73	7.0	12.0	22	6.5	60.4	47	135		1.9	6.0		4.2	
Mean	7.3	8.0	52	10.5	87.8	46	114	0.6	0.7	3.9	9.1	6.4	-
STATION 8.													
10.11.71		5.0		14.0	109								
19.1.72	7.2	6.0	90	9.8	78.5	46	95	0.9	0.5	2.5		5.5	192
1.3.72	6.9	7.0	153	9.0	74.0	30	90	0.6	0.4	2.9	13.0	17.0	140
10.5.72	7.6	14.0	37	10.1	97.4	38	125	1.2	0.5	4.2	6.6	5.7	194
18.10.72	7.6	11.5	6	10.4	96.6	41	125	0.4	0.6	4.3		1.8	
11.12.72	7.1	4.5	44	10.3	80.6	40	75						
21.3.73	8.3	8.0	12	18.3	154.6	48	152	0.4	0.5	4.8		7.2	
25.5.73	7.2	12.0	27	6.9	64.1	46	135		1.9	6.5		4.5	
Mean	7.4	8.5	53	11.1	94.4	41	96	0.7	0.7	4.2	9.8	6.1	-

TABLE 4.14

(continued)

Date	pH	^{o}C	S.S.	D.O.	% Sat.	Cl^-	Alk.	NH_3	T.I.P.	O.N.	P.V.	B.O.D.	HDNS
						STATION 9.							
10.11.71.		3.5		13.0	99.0								
19.1.72.	7.2	6.0	103	9.4	75.3	79	110	0.3	0.5	1.5		6.5	222
1.3.72	7.0	7.0	152	9.0	74.0	38	95	0.6	0.4	2.9	12.2	16.0	184
10.5.72	7.8	14.0	29	11.7	112.8	37	165	1.1	0.3	4.1	5.8	6.0	222
18.10.72	7.8	10.5	4	12.0	108.9	41	140	1.0	0.6	3.6		2.1	
11.12.72	7.2	4.5	41	10.6	83.0	60	75						
21.3.73	8.4	7.5	9	17.8	150.3	49	150	0.4	0.5	4.3		3.9	
25.5.73	7.5	12.0	14.0	9.5	88.2	44	140		1.7	1.5		4.4	
Mean	7.6	8.1	50.2	11.6	96.9	50	121	1.0	0.7	2.9	99.0	6.5	-
						STATION 10.							
10.11.71		4.0		15.2	115.7								
19.1.72	7.2	6.0	105	9.4	75.3	75	125	0.3	0.5	2.3		6.0	216
1.3.72	7.0	7.0	145	8.8	72.0	39	100	0.6	0.3	2.9	11.2	14.0	186
10.5.72	7.8	14.0	32	11.7	112.8	38	160	1.7	0.4	4.1	5.0	5.4	214
18.10.72	7.8	10.5	4	11.8	107.0	40	145	0.1	0.7	3.8		1.8	
11.12.73	7.2	4.5	46	10.6	83.0	63	75						
21.3.73	8.5	7.5	110	19.9	168.1	54	160	0.4	0.5	4.4		3.6	
25.5.73	7.0	12.0	14	-	-	44	155		1.4	5.3		1.2	
Mean	7.5	8.3	32	12.4	104.8	50	131	0.4	0.6	3.8	8.1	5.3	-
						STATION 11.							
10.11.71		4.0		14.2	108.1								
19.1.72	Flooded												
1.3.72	"												
10.5.72	8.1	15.0	31	13.0	128.0	38	140	0.5	0.3	4.1	5.2	5.1	206
18.10.72	7.9	10.0	5	12.0	106.4	41	140	0.1	0.7	4.1		0.9	
11.12.73	Flooded												
21.3.73	8.6	6.5	13	20.5	169.0	51	160	1.2	0.5	4.4		3.3	
25.5.73	7.2	12.0	12	9.6	89.1	43	155		1.7	5.1		2.1	
Mean	7.9	9.5	15.2	13.9	120.1	43	149	0.6	0.8	4.4	5.2	2.8	

CHAPTER 5.

STUDIES ON SPECIES COMPOSITION, COMMUNITY STRUCTURE AND SECONDARY PRODUCTIVITY OF BENTHIC INVERTEBRATE COMMUNITIES IN RELATION TO NUTRIENT STATUS OF THE WATER.

5.1. INTRODUCTION.

An attempt to simulate rivers in different states of eutrophication or in a river whose nutrient status changes with distance downstream, was made using three excavated experimental streams whose chemical status differs with particular regard to nitrogen and phosphorus. Three conditions were achieved by mixing nitrified sewage effluent (containing considerable amounts of phosphorus) in certain proportions with water of good quality from the adjacent River Tean. After flowing down the experimental streams, these mixtures are returned to the R. Tean along with the greater proportion of final effluent discharged from the outfall of the Blithe Valley Sewage Treatment Works. As a result the R. Tean below this outfall is not only increased in volume by approximately 1/3 (d.w.f.) but in nutrient enrichment also, which is seen to diminish with dilution and utilisation downstream.

Experimental streams allow a number of variables to be controlled, a situation not possible in natural rivers: even when rivers of the required levels of fertilisation have been located, their geographical position, geology, flow rate, natural biota, etc. will have density independent effects upon the eutrophication fauna. The Experimental Streams present similar substrata, flow rates, geographical position, rainfall and photoperiods, together with the same natural cource of invertebrate supply. Moreover these Streams, as opposed to laboratory systems, do not suffer from the build up of metabolites and possess the singular stream characteristic of continual downstream movement rather than re-circulation. These Streams do not get any shading from trees or shrubs as small streams often do,nor do they get much natural allochthonous material and are thus more like headstreams in these respects. However, the orientation of the Streams in a NW-SE direction permits some shade at nearly all times of the day due to the depth of the banks. Solar radiation is one of the major energy sources in the Experimental Streams and large diatomaceous and filamentous growths of algae occur in all the Streams, with large bryophytic growths in Streams 1 (62.5% river water) and 2 (100% river water) in spring/summer. The major sources of energy in Streams 1 and 3 (12.5% river water) are the pre-formed organic solids from the sewage treatment processes.

Systems like these do suffer from the gradual build up of solids as there are no 'spates' to wash them away or indeed to dislodge members of the benthic communities that would naturally constitute drift and facilitate dispersal. Also, fish were not introduced into the Streams (although some fry did survive the journey through the pump and weirs) so that lack of predation on some species would not perhaps have given a realistic picture of the faunal densities and structure. The dangers involved with artificial streams lie in supposing the ecosystems to be operating like natural ones and deriving spurious inferences from the 'specialised' data. But if one is aware of the drawbacks and the results are interpreted within certain frames of reference, misinterpretations may be obviated. The problem of 'artificiality' has been tackled by the use of a 'control'

Stream (No. 2) which receives 100% river water from the R. Tean and whose community structure may be compared with that of the river near to the point of abstraction and above the outfall. Moreover, four riffles on the R. Tean below the outfall and one riffle on the R. Ray (Wiltshire) have been selected to demonstrate the 'natural' communities that are associated with varying degrees of fertilisation, organic input, and the other chemical parameters encountered downstream of a well oxidised sewage effluent. The River Ray receives highly polished effluent from Rodbourne Sewage Disposal Works at Swindon which accounts for about half the flow of the river and more than two-thirds of the flow in dry summer periods. One riffle was selected at Moredon because of its huge growths of *Cladophora* and biomass of *Asellus aquaticus*, an association indicative of highly eutrophic conditions. Length/frequency analyses, biomass, and calorific determinations were made on the *Asellus* populations here (Chapter 8) as a way of determining the relative production of this species in this river, the R. Tean, and the Experimental Streams with different states of eutrophication.

The aims of the experimental stream research essentially fell then into three main areas: -

1. to compare the production in terms of biomass (weight and calories) and numbers of invertebrates in the three experimental streams of dissimilar nutrient status, and to see if any ratios or associations were appropriate to the degree of fertilisation.

2. to examine the trophic structure of the streams by categorising the herbivores, carnivores, and scavengers and representing the size of the structures by weight and energy.

3. to see if there were any outstanding differences in species distribution with the degree of eutrophication associated with each Stream of the R. Tean.

In each Stream the artificiality and reality was measured as far as possible with the real situation of the R. Tean and, in the case of Asellus, the R. Ray. Secondary considerations included comment upon the artificiality of the Experimental Streams, the colonisation of the Streams from a habitat void of species, and an examination of the comparative chemistry.

5.2. WATER CHEMISTRY OF THE EXPERIMENTAL STREAMS, R. TEAN AND R. RAY.

Monthly means of daily analyses have been plotted for the majority of parameters covering the period August 1972 to April 1974. This data clearly demonstrates the seasonal variations encountered in the Experimental Streams (Figs. 5.1., 5.2). Annual means and standard errors for the year 1973 have been drawn in histogram form alongside those for the five stations of the R. Tean. (Figs 5.3 & 5.4). The changes in water quality of the R. Tean have been demonstrated by plotting some of the more important parameters (dissolved oxygen, suspended solids, ammonia, oxidised nitrogen, total inorganic phosphorus) of the five stations against the distance in km. from the confluence with the R. Dove (Fig. 5.5). A 24-hour survey was conducted on the Experimental Streams in April 1974 to show the diurnal fluctuations, and in particular those of dissolved oxygen as a result of the spring outburst of diatoms and other algae (Fig. 5.6). Tables 5.1 - 5.10 give comprehensive physical and chemical data relating to the Experimental Streams and Stations of the R. Tean.

(a) Temperature (Fig. 5.1).

The range of seasonal variations between the three Streams is about the same, with Stream 3 (87.5% sewage e effluent) being approximately 1°C higher than Stream 1, and the latter 1°C higher than the river water in Stream 2. The mean temperature in Stream 2 (100% river water) is a fraction higher than the river at Station 1 (where the water is abstracted) owing to the heating effect in the sedimentation

FIG.5.1. Graphs of mean monthly physical and chemical data in the Experimental Streams.

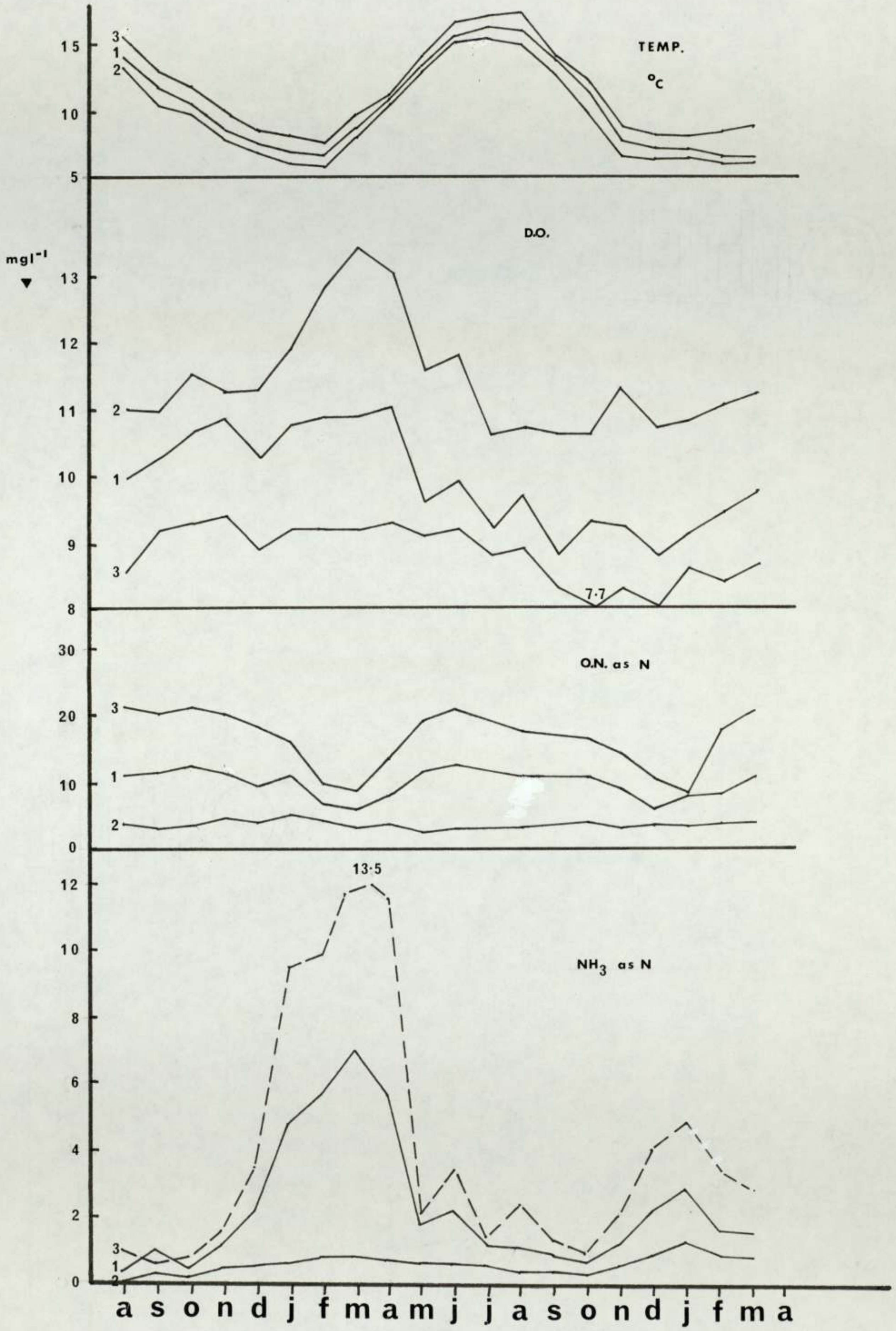


FIG.5.2. Graphs of mean monthly chemical data in the Experimental streams.

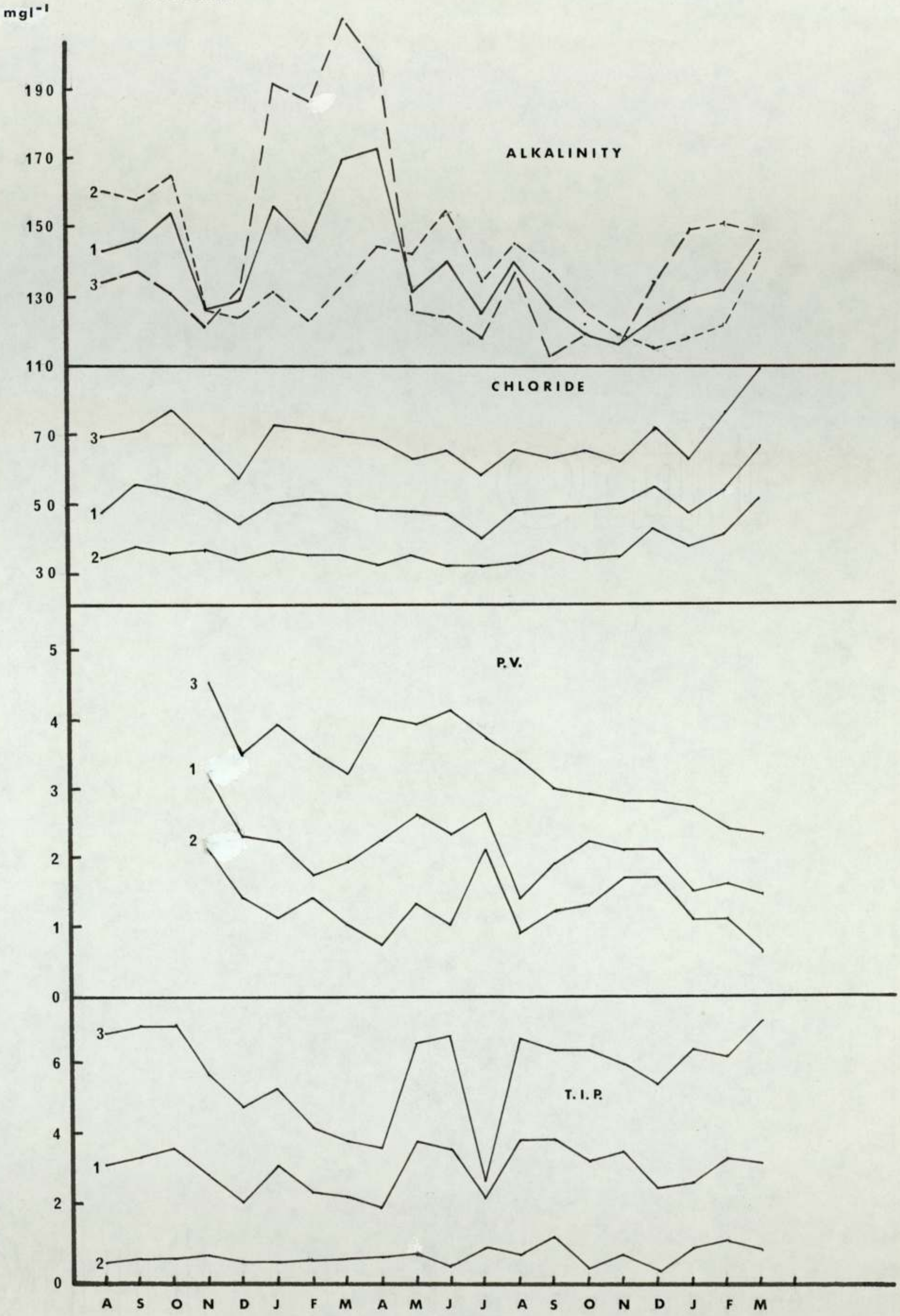


FIG.5.3

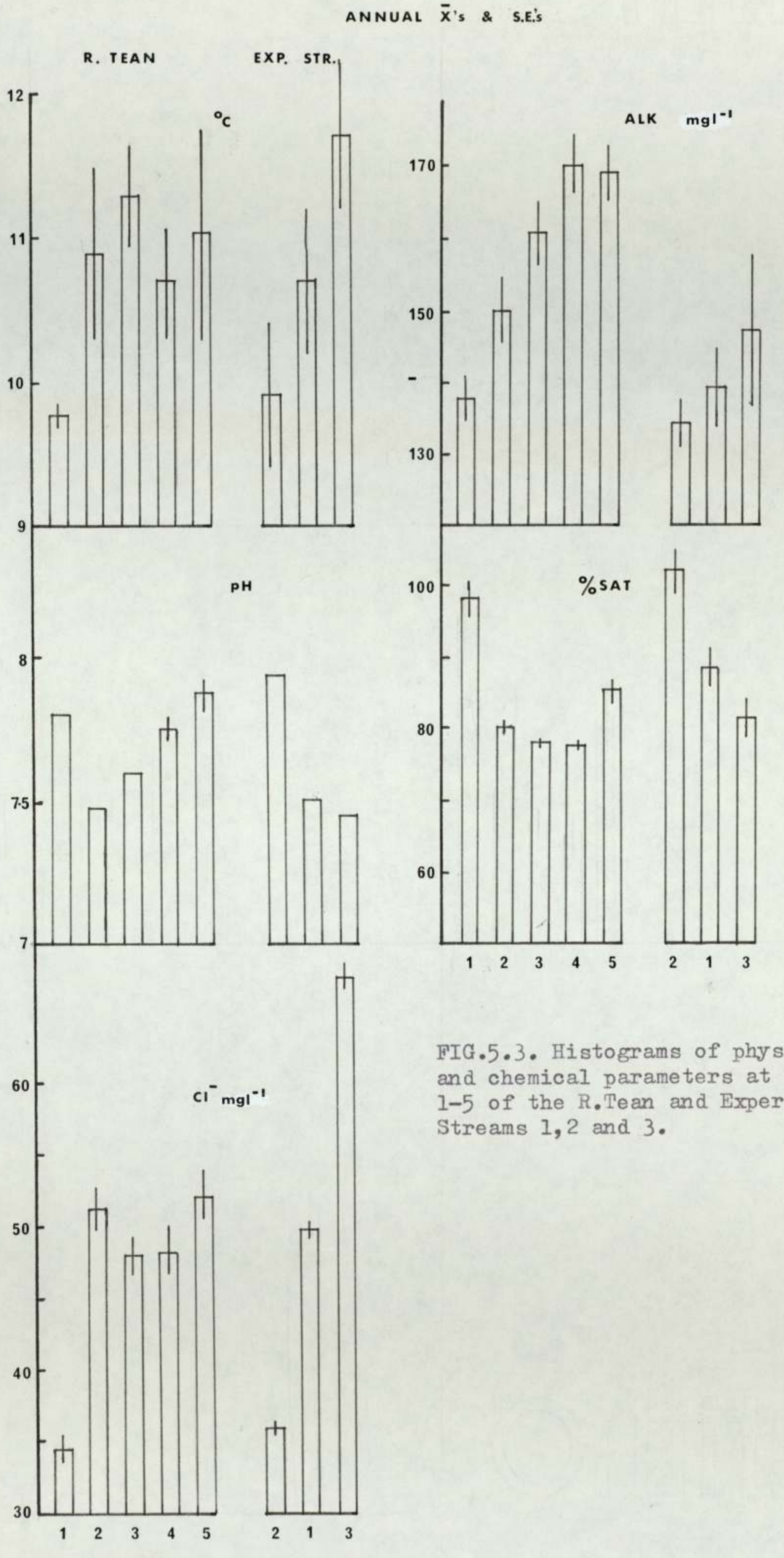


FIG.5.3. Histograms of physical and chemical parameters at Stations 1-5 of the R.Tean and Experimental Streams 1,2 and 3.

FIG.5.4. Histograms of chemical parameters at Stations 1-5 of the R.Tean and in Experimental Streams 1,2 and 3.

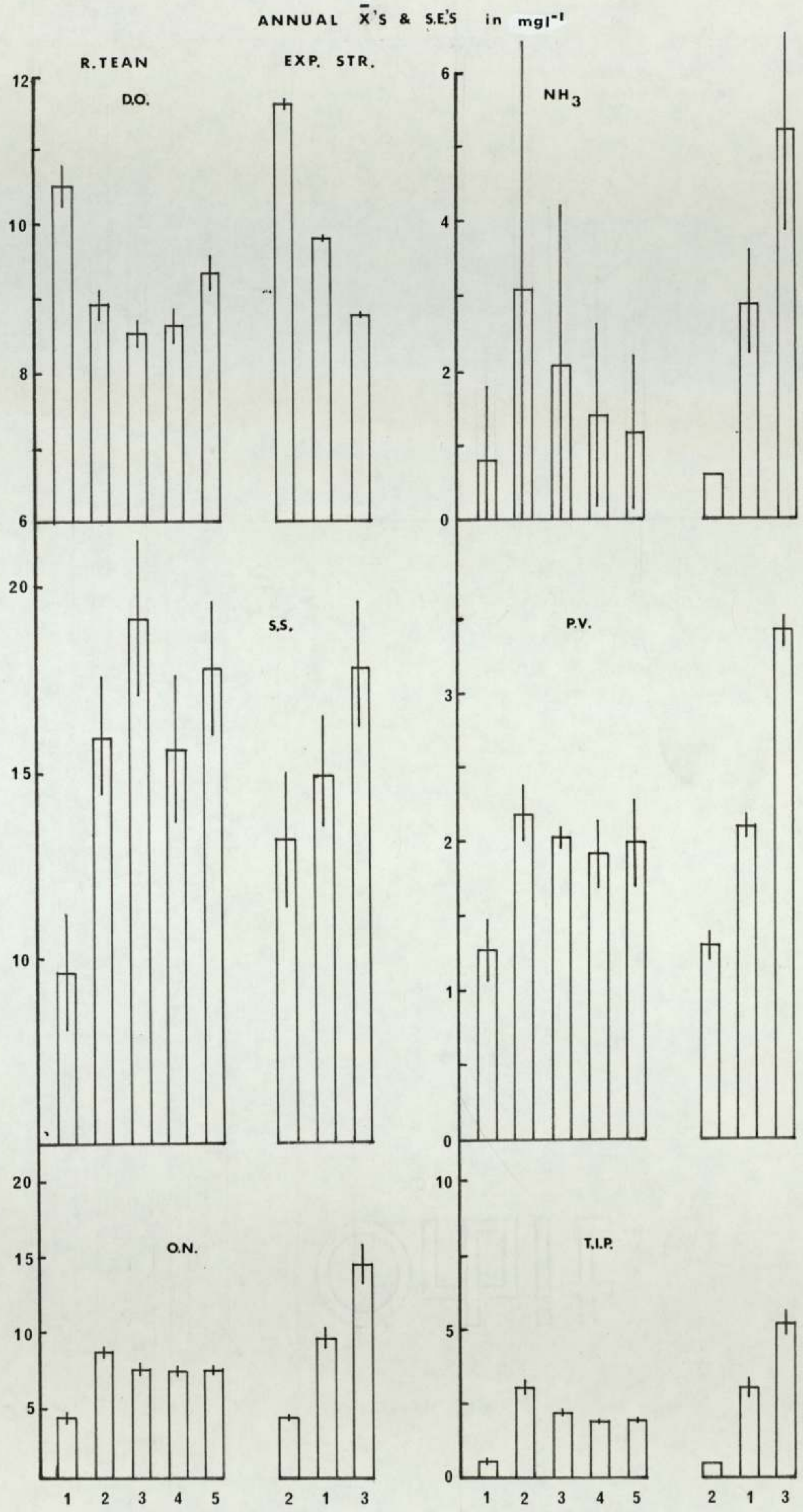
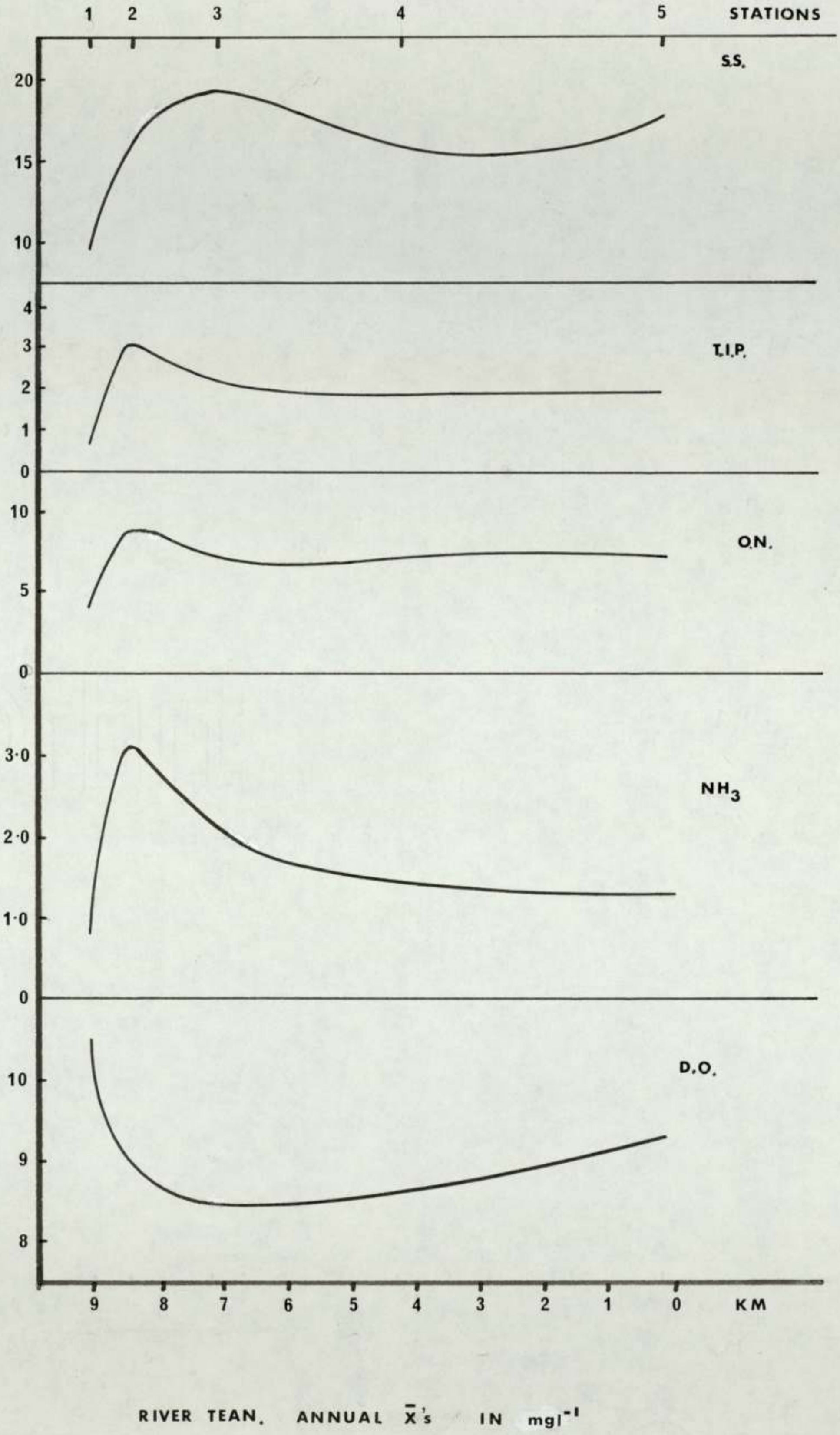
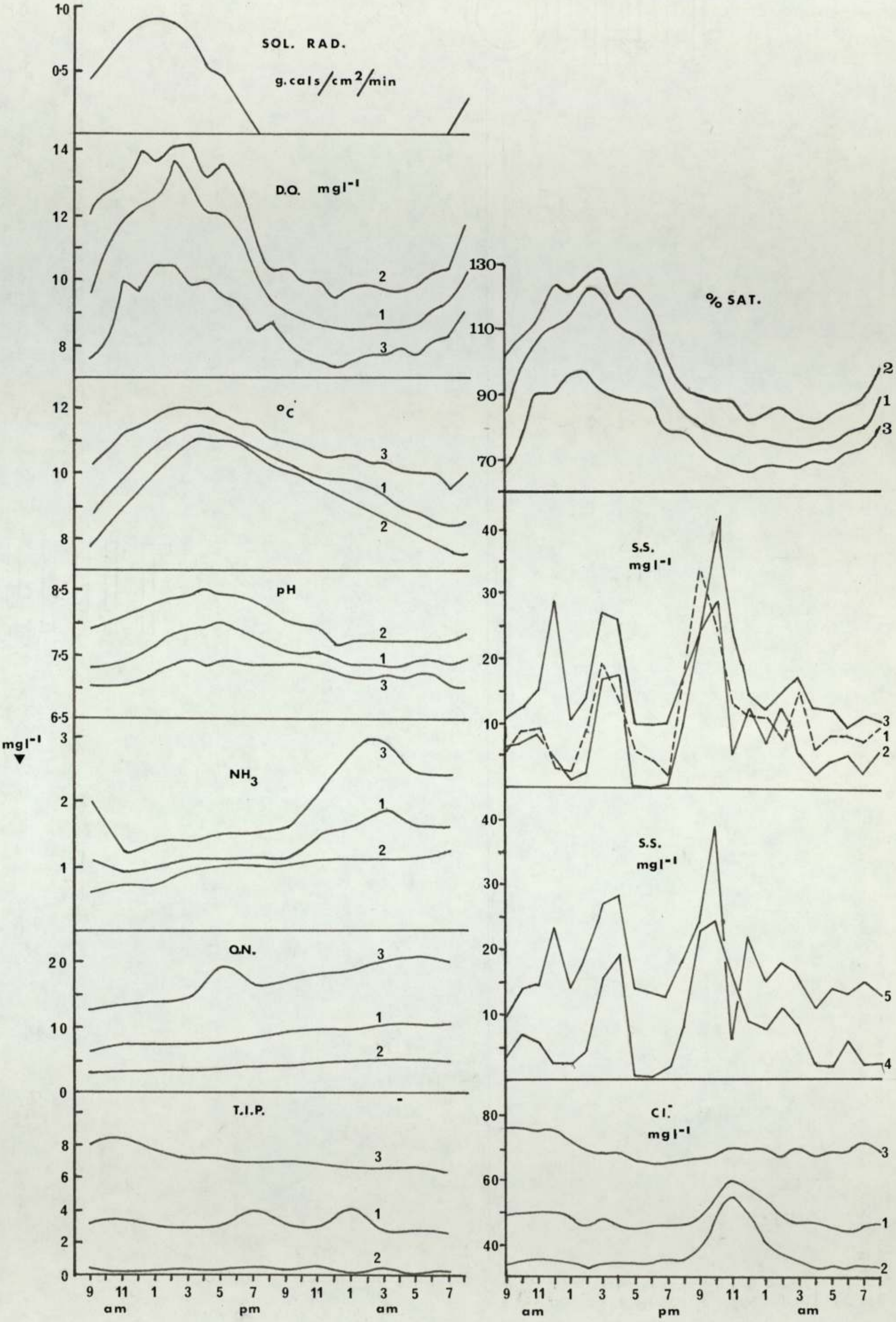


FIG.5.5. Graphs of mean annual chemical parameters at five Stations of the R.Tean against distance in km from the confluence.



RIVER TEAN. ANNUAL $\bar{X}$'s IN mgl^{-1}

FIG.5.6. 24hr. physical and chemical data from the Experimental Streams: 5-6.4.74.



tank and asbestos channels prior to its passing into the Experimental Streams (see Plate 2). Stream 1 (37.5% sewage effluent) is approximately the same temperature as Station 4 of the R. Tean whereas the heat supplied by the high percentage of sewage effluent to Stream 3 keeps it way above any part of the R. Tean. The river, after the entry of the outfall above Station 2, shows an average increase of 1°C, which rises slightly at Station 3 owing to the discharge of an effluent from a dairy.

(b) Dissolved Oxygen.

The seasonal graphs (Fig. 5.1) show the effect of the increased light, temperature, and growth rates on the algae in spring, which is most marked in Stream 2 followed by Stream 1. Stream 3, having only 12% river water, does not show a spring increase but nevertheless contains fairly high levels of dissolved oxygen all the year round. The annual means (Fig. 5.4) with very small standard errors demonstrate the differences between each of the three Streams, and between them and the Stations of the R. Tean. Station 1 has overall a lower level of dissolved oxygen than Stream 2 as the latter receives O_2 from the asbestos algal channels. Streams 1 and 3 differ marginally from the other Stations of the R. Tean, although in terms of percentage saturation they are most similar to Station 5. A classical 'oxygen sag' curve is demonstrated by the R. Tean data in its plot against km. from the confluence (Fig. 5.5). Self-purification from the sewage effluent is hampered slightly by the dairy effluent at Station 3 and animal excreta throughout the rest of the water course but, even at its lowest (Station 3) is still approximately 80% saturated.

The greatest diurnal fluctuations recorded were in Streams 1 and 2 with peaks at 2.0 p.m., and troughs at midnight where the differences in oxygen were in excess of 4 mgl^{-1}.

(c) Ammoniacal and Oxidised Nitrogen.

The exceptionally high 1973 annual means of ammoniacal nitrogen for Streams 1 and 3 are the result of the poor nutrification of the effluent over the 1972/73 winter (see Table 5.5 for effluent). The seasonal graphs (Fig. 5.1) show how averages for this period were in excess of 10 $mg.N.1^{-1}$ NH_3 in Stream 3 while in Stream 1 a maximum peak of 7 $mg.N.1^{-1}$ in March 1973 was reached. This is reflected by the parallel drop in oxidised nitrogen for Streams 1 and 3 whereas no drop occurs in Stream 2, ruling out the possibility of algal growths being responsible. Fortunately the biological effects of such high ammonias would have been ameliorated by the high oxygen input from the river and a pH range from only 7.3 to 7.6, thereby restraining the amount of un-ionised ammonia. Lower temperatures over this period would have also kept more oxygen in solution and reduced metabolic activity of the benthos. Ammonia and oxidised nitrogen levels in Stream 2 were consistently low, matching those of Station 1 in the Tean; and diurnal levels in the effluent showed their greatest effects in the Streams at 2.0 a.m. where for example the level of NH_3 was doubled over 14 hours in Stream 3. The low level daytime levels of NH_3 were thought not to be the result of nitrogen utilisation by the algae as no such pattern was discernible in Stream 2, which received water from the asbestos channels containing the most algae.

A typical dilution curve for NH_3 levels is seen in the R. Tean with distance downstream (Fig. 5.5), and oxidation of this ammonia is shown with the counteraction of dilution on the oxidised nitrogen concentrations. Mean levels of ammonia are fairly high at Station 2, being in the region of 3 $mg.N.l^{-1}$ which puts this station on a par with Stream 1. However, the standard errors of the Tean stations are in excess of the mean so that Station 3 is also similar to Stream 1 concentrations. The quantities of ammonia and oxidised nitrogen in Stream 3 are nowhere near met in the R. Tean or the R. Ray.

(d) Total Inorganic Phosphorus (T.I.P.)

The seasonal and diurnal levels of T.I.P. in the Experimental Streams are fairly constant, with the levels declining thus: Stream 3 > Stream 1 > Stream 2 in accordance with dilution of the sewage effluent. The mean levels in Stream 2 and in the river above the outfall are very low (0.4 ± 0.00) yet sufficient to support large growths of algae in the Experimental Stream. Below the outfall the concentration of phosphorus is increased six-fold but dilution and utilisation reduces it to approx. 2 mgl^{-1} for the remainder of its course.

(e) pH.

This rises both diurnally and seasonally with light intensity and day length as a primary result of photosynthesis and CO_2 utilisation. The higher annual mean of Stream 2 compared with Station 1 of the R. Tean is due to the greater density of algae in the asbestos channels whereas the lower pH's of Streams 1 and 3 are due to the almost neutral intensity of the sewage effluent.

The pH of the R. Tean is similarly lowered after the outfall above Station 2, but increasing dilution from water of high pH upstream and increasing eutrophication downstream elevates the levels to 7.9 by the time it confluences.

(f) Alkalinity. The quantity of alkali in the sewage effluent was greater than that in the river water above the outfall. Consequently the greatest alkalinity was in Stream 3 > 1 > 2. The alkalinity increased after the discharge into the R. Tean and continued increasing downstream with eutrophication. The concentrations as $CaCO_3$ in the Experimental Streams therefore were all within the range of concentrations occurring between Station 1 and 2 of the Tean. The seasonal highs in Streams 1 and 3 are due to the poor oxidation of ammonia (see Ammonia graph).

(g) Chloride (Cl^-) The amount of chloride in the R. Tean upstream of the outfall and consequently the amount in Stream 2 resembled that of spring water, i.e. low (about 35 mgl^{-1}, Fig. 5.3). Although these levels were not seen to vary much seasonally, a considerable input of chloride was detected in the river during a 24-hour survey around midnight, which inevitably elevated the levels in Stream 2 and, to a lesser extent, Stream 1. The level of chloride in Stream 3 is so high owing to the percentage of effluent that the midnight increase (Fig. 5.6), perhaps because of cattle upstream, went undetected.

The fractionally increased concentration of Cl^- in Stream 2 over Station 1 of the river would indicate a small leakage somewhere within the channel/Stream system. The dilution offered by the river water below the outfall causes the levels to drop downstream, although a further input of chloride is noticeable between Station 4 and 5 (Fig. 5.3) where large dairy cattle herds are grazed on the riparian land. The dilution of effluent in Stream 1 is seen as being similar to that of the river at Stations 2, 3 and 4. Exceptionally high values were obtained in winter following road salting (Tables 5.9., 5.10).

(h) Permanganate Value (P.V.)

Biological Oxygen Demand determinations were abandoned after finding the distilled water, i.e. blanks, to have a B.O.D. of between 1.0 and 2.0 mgl^{-1}. De-ionisation cured the problem but every two days the resin column required changing - an expense that could not be incurred. Four-hour permanganate values were therefore used as a measure of organic materials in both the Experimental Streams and the R. Tean. Naturally Stream 3 (87.5% sewage effluent) had the consistently highest P.V. followed by Stream 1 (37.5% sewage effluent) and, finally, Stream 2 with a very low value. No seasonal fluctuations were detected. The strikingly low values in all the Experimental Streams and in the R. Tean is indicative of the low levels of industrial wastes and efficiency of the Works in removing the carbonaceous matter. Most of the solids recorded in the Experimental Streams originating from the river are therefore mostly inert. The organic and inorganic loading of the

R. Tean at Stations 3 and 4 are very similar to that of Stream 1.

(i) Suspended Solids.

The fluctuations inherent within the Experimental Streams owing to solids from the effluent and the river made the seasonal graph visually undecipherable, and it has therefore been excluded. However, the 24-hour survey (Fig. 5.6) shows the influence both the river (marked 4) and the effluent (marked 5) have on the Streams. Stream 3 emulates, to a lesser degree, the solids figures of the effluent and, similarly, Stream 2 duplicates the river solids, with Stream 1 solids being of proportional magnitude between the two. Solids from both the river and the effluent are higher in winter, although the major effect of the river solids is one of scouring rather than oxygen demanding. Some of the river solids are settled out in the sedimentation tank (Fig. 3.1). The annual mean solids in Stream 2 is considerably higher than Station 1 of the river because of the large amount of 'algal drifts' emanating from the asbestos channels; when the growths were very dense, the oxygen bubbles produced during the day lifted off whole layers of epiphytic algae which then drifted into the Streams to be recorded.

The general pattern of solids proceeding downstream in the R. Tean is seen on the graph - Fig. 5.5. The process of precipitation starting after the outfall can be seen as well as more solids from the dairy discharged just above Station 3. Later these settle out but the increased flow nearer the confluence again increases the solid levels.

Nevertheless the mgl^{-1} scale is somewhat expanded and the solids figures in the Streams and in the river below the outfall are all very similar.

(j) Hardness.

This increases with the proportion of effluent in the streams as do the detergents which are of a very low level in the effluent initially (Tables 5.1 - 5.5).

(k) Metals.

On normal working days samples for metal analysis were taken around mid-day from the Experimental Streams, the final effluent, and the river at the point of abstraction. The Blithe Valley S.T.W. receives effluent from a predominently residential area of Stoke-on-Treant and consequently a limited concentration and number of metals from trade wastes were expected. This proved to be correct, the levels of total metal in the Experimental Streams 1 and 3 being low and, considering the hardness, dissolved oxygen and slightly alkaline pH were very likely rendered innocuous.

Table showing the amounts of total metals (in mgl^{-1}) taken during a spot sample 10.2.1973.

Sample	Cr	Cu	Zn	Ni	Pb	Cd	Fe
Stream 1	ngl	0.06	0.16	0.06	ngl	0.008	0.25
" 2	ngl	0.04	0.13	ngl	ngl	ngl	"
" 3	0.02	0.07	0.19	0.07	ngl	0.014	"
River above outfall (4)	ngl	0.02	0.07	ngl	ngl	ngl	"
Final Effluent (5)	0.04	0.08	0.27	0.11	ngl	0.018	"

River Ray Analysis

Yearly averages have been calculated for the period February 1972 - January 1973 and tabulated in Table 5.11. Examination of these means shows that they are similar to those of Experimental Stream 3 with respect to oxidised nitrogen, total inorganic phosphorus, dissolved oxygen, temperature, chloride and suspended solids. The organic loading in the R. Ray at the sampling station (2.1 km. downstream of Rodbourne S.D.W.) is twice as great as in the R. Tean and Stream 3 owing to differences in dilution of the effluent. The mean P.V. and B.O.D. of the Ray are 6.5 and 9.5 respectively and the highly nitrified and tertiary treated effluent only loads the river here with 2.7 $mg.N.l^{-1}$ NH_3, i.e. approximately half the concentration that Stream 3 receives. Also the R. Ray is 20% 'harder' in terms of $CaCO_3$ than Stream 3.

5.2.1. SUMMARY.

Stream 2, containing 100% river water abstracted above the outfall, is of course identical in chemical nature to Station 1 of the R. Tean, although small differences in the temperature, pH, and dissolved oxygen levels exist owing to the algal effects in the asbestos channels on the water.

Stream 1 is similar with respect to nutrient levels, organic loading, and dilution in terms of chloride values to all the Stations of the R. Tean below the outfall. Temperature and suspended solids are analagous to Station 4 in particular whilst only ± 1.3 mgl^{-1} exists in the differences in dissolved oxygen between this Stream and all Stations of the R. Tean.

Stream 3 is parallelled in the R. Tean with respect to dissolved oxygen, pH, suspended solids, and organic loading (within 1 mgl^{-1} permanganate value). However its nutrient status is far in excess of any R. Tean Station and resembles more those of the R. Ray at Moredon, i.e. with the exception of ammoniacal nitrogen which is considerably higher than in both rivers.

Because of the labile nature of rivers with particular reference to volume (= dilution), associated rate of flow (= speed) and, to a lesser extent, nature of the substratum (= siltation), it is not acceptable to use the Stations which display similar chemical trends to the Experimental Streams as rigid controls for comparison of benthic communities without reservations.

5.3 OBSERVATIONS ON THE COLONISATION OF THE STREAMS.

Water started to flow down the Experimental Streams during the last week of July 1972. There was slight concern over the recruitment of species to the Streams for, although there were ample supplies in the river where the water was being abstracted, the powerful pumps and weir tank would have to be negotiated before invertebrates could gain access to the Streams. Undoubtedly some species would have been placed into the Streams with the stones removed from the river. These would have been in either larval or egg form depending upon the species life history.

Patterns of invasion by benthic macro-invertebrates into excavated streams were the subject of study by Patrick (1959) and Leonard (1942), and into artificially denuded natural streams by Muller (1954) and Waters (1964). By investigating their results, the short term colonisation of the Experimental Streams could be compared for speed and species as no pumps were involved in any of the above workers' studies. All these authors expressed surprise at the rapidity of colonisation by a number of invertebrates. Leonard (1942) in mid-October found that within one week the new stream contained 2,000 Similium venustum per m^2; after two months eight species had colonised including Ephemerella, Baetis, Rhyacophila, Hydropsyche, and three species of chironomid larvae; after three months 19 species were established, and one month later 21 species were present. Patrick (1959), whose stream was free of eggs, also found similar patterns of invasion with Similium vittatum (Zett) being the first to colonise followed by Baetis, Chironomidae and, after six months, a firmly established population of Physa heterostropha.

The substrata of the Experimental Streams 1, 2, and 3 were not disturbed until October, i.e. after two months of operation, when the first quantitative sample was taken, but a visual inspection showed Simulium ornatum to be well established within a week, and after two weeks their numbers were estimated as 10,000 per $0.05m^2$ or $48g/m^2$. A few Baetis species were observed and no doubt chironomid larvae were inhabiting the riffles but no Gammarus were seen.

A similar pattern to those of the above authors was observed when quantitative results were examined over the remainder of a six-month period.

Table of mean numbers of individuals per $0.05m^2$ taken in the Experimental Streams in the first six months of flow.

	B. rhodani			G. pulex			S. ornatum			R. dorsalis		
Stream No.	(1)	(2)	(3)	(1)	(2)	(3)	(1)	(2)	(3)	(1)	(2)	(3)
Aug.	-	-	-	-	-	-	-	-	-	-	-	-
Sept.	-	-	-	-	-	-	-	-	-	-	-	-
Oct.	3	64	0	0	2	0	235	673	71	9	17	0
Nov.	27	141	0	0	3	0	339	32	255	3	4	0
Dec.	8	44	0	0	12	0	328	12	88	2	10	0
Jan.	20	128	0	2	6	0	98	0	0	2	8	6

The Streams were expected to have been colonised by Baetis and Gammarus quickly because of their high rates of drift in autumn, but something had prevented Gammarus from doing so for at least six months. As the very delicate

B. rhodani had managed to survive the pumping, the absence of the hardier *Gammarus* was presumed to be a biotic effect caused, perhaps, by the lack of food in such a new stream. Waters (1964) found that in a natural riffle section he had denuded of *Gammarus limnaeus* (Smith), the enormity of the autumn drift returned the large population back to normal in one day and, in winter, four days. The absence of these sensitive species and *Rhyacophila dorsalis* in Stream 3 was expected because of the water quality. Some of the trichopteran larvae must have drifted in, as relatively large numbers like these could not have remained alive during transfer of the stones. *Simulium* species, it would seem, are particularly suitable for invading habitats in large numbers because of their short life histories (many multivoltine) and capacity for expansion, with little or no competition being valuable assets. The amount of filterable material in the Streams and the clean stones for attachment allowed populations the size of these to develop.

Chironomid larvae were also naturally successful owing to their aerial dispersal, and those species which oviposited about August were particularly numerous by the time of the quantitative surveys. These were notably *Micropsectra atrofasciatus*, *Eukiefferiella hospita*, *Procladius sp.*, *Chironomus riparius*, and *Thienemannimyia lentiginosa* although a few other species were also recorded. The high percentages of chironomid larvae in the first six months are evident from the table below.

Table showing the total number of invertebrate species established per 0.05 m^2 in the Experimental Streams with numbers in brackets representing numbers of non-chironomid species. Station 1 of the River Tean for part of the same period is included.

	STREAMS 1	STREAMS 2	STREAMS 3	R. TEAN Station 1
August	(1)	(1)	(1)	-
September	(2)	(2)	(2)	-
October	7 (3)	10 (5)	8 (2)	-
November	15 (6)	13 (7)	10 (4)	10 (7)
December	12 (4)	14 (6)	9 (2)	11 (9)
January	14 (7)	10 (4)	6 (1)	10 (9)

This table also demonstrates how, after only three months of flow, 15, 13 and 10 species were present in Streams 1, 2, and 3 respectively, which is greater than the number of species supported in the river at Station 1 and showing the imbalance of the ecosystem that chironomid larvae can enjoy. A more strict comparison by inspect of Stream 2 (Tables 5.13 & 5.14) and Station 1 (Tables 5.16 & 5.17) illuminates the differences in species composition and the labile nature of the new stream with its declining numbers when the numbers and proportions of species in the river are stable. Remarkable is the absence of the Mollusca which do not appear as established until July 1973, i.e. after one year of operation (Table 5.13). Muller (1954) made similar observations, which are not surprising considering their immobility and indisposition to drifting in the adult form.

These preliminary examinations appear therefore to be in accordance with the results of other workers, and the effect of pumping would not seem to present a hazard to invading species.

5.4. STANDING BIOMASS AND TROPHIC STRUCTURE.

The importation of allochthonous material forms a supply of energy to a body of water. This is particularly true of lotic waters where detritus often forms the major source of energy with autotrophic production occupying a subordinate accompaniment. In natural streams the rithron algal production can be heavy compared with the production in the potamon but both receive organic material in the forms of leaf debris (which may have passed through guts already), animal remains, and, especially in the lower stretches, fine detritus from above, bacteria and algal remains. The importance of organic material to secondary productivity in natural streams has been pointed out by Hynes (1963) while Scott, D.C. (1958) showed the food value in organically enriched streams of organic material from sewage effluents.

The composition of final sewage effluent solids will be a function of the type of treatment, but in any case will represent a high organic proportion of solids containing bacteria, fungi, protozoa, algae and larger members of the grazing fauna if filter bed oxidation is being utilised. These saprozoic, saprophytic and holozoic forms constitute a high energy food source for what is usually termed the primary consumers, although those feeding on the holozoa and saprozoa are really secondary consumers. The importance of this in terms of production efficiencies is realised from the work of Nelson and Scott (1962) who found that the ratio of weight of detritus feeders to weight of detritus was higher than the ratio of weight of herbivores to weight of plant material. It appeared, therefore, that bacteria and fungi feeding on the detritus (some primary consumers) were of greater importance to the detrital feeders than was imagined, and that the latter were in effect secondary consumers with, consequently, higher production efficiencies.

The result, a priori, is that with an energy input like sewage effluent containing high dissolved and colloidal solids together with high energy primary consumers in the form of protozoa, bacteria and fungi, the gross production efficiency at the next trophic level will be increased and be of far greater importance compared with the role of energy from radiation and allochthonous production.

The vast amount of high energy organic input from the effluent into the Experimental Streams and into the river were seen to alter not only the amounts of production in terms of standing biomass, but affect the species density, diversity and therefore the trophic entities - in Lindemann's terms (1942). The stability of the trophic levels depend upon the numbers of food chains and amounts of food in terms of energy between trophic levels. This stability, which is likely to be upset by the introduction of high energy materials, may be measured in terms of numbers of species since each species has its own food web. The first concern would be to determine the various trophic levels within the Streams and the river but this being of impossible dimensions, considering the complexity of the food webs that would have been encountered and the determinations of them using gut analyses, radiotracers and precipitin reactions, a more simplistic scheme was drawn up. The species involved in the benthic communities were assigned, depending upon their known feeding habits, to three trophic entities vaguely described by detrital feeders or decomposers, grazers and carnivores. The detrital category included those species which were primarily known to feed upon decaying plant and animal material bacterial or fungal flora; grazers included those herbivores feeding upon algae and plant material while carnivores were primarily predacious. Difficulties were encountered in assigning species of chironomid larvae to grazer or decomposer entities as there was very little information on their feeding habits, however, on the basis of their mean annual numbers and species occurrence in the Experimental Streams in which it was found that most species and numbers occurred in those Streams with the enriched organic and nutrient supply (1 and 3), they were assigned to the decomposer category. The carnivorous chironomid species were obviously attached to the carnivore category along with Rhyacophila dorsalis, which although is known to eat filamentous algae (Percival and Whitehead, 1929) was considered primarily a predator of chironomid larvae, tubificid worms and B. rhodani. The greatest problem was in placing Lymnaea pereger, commonly known as a grazer of algae but also recorded as grazing bacterial slimes (Hawkes, 1963b, describes them on high rate re-circulation beds at Harrogate). Their occurrence downstream of the effluent at Station 3 of the R. Tean seemed indicative of their feeding on bacterial slimes or detritus - a phenomenon that also appeared to occur in the Lower Cole in the absence of most algae; but their dominance and abundance in Stream 2 reflected their algal grazing activities. They were eventually placed in the grazer category for the purposes of calculation of the standing biomass in terms of dry weight and energy.

The primary production and biomass of autotrophes were not measured but a representation of the amount was apparent from the standing crop of grazers, and, assuming food chain efficiency at this end of the trophic scale to be much less than 10%, (the figure used for gross ecological efficiency) the amount of energy supplied by the photosynthetic component must be at least 10 times the energy of the standing crop of grazers. The detritral component of the river and Stream 2 was only measured as suspended solids although allochthonous material in the Streams from leaf fall and decaying macrophytes was small. Suspended solids determinations in Streams 1 and 3 and Stations 2 - 5 of the R. Tean do give some measure of allochthonous input, and the microbial populations accompanying the solids is apparent in the permanganate value.

It was not the primary objective of the investigation to research into the energy flow through the Experimental Streams, but the summation of mean calories/ $0.05m^2$/yr within the trophic entities described does give an indication of the structure of the communities, their productivity and potential energy source as a result of eutrophication.

The members of the various trophic entities in the Experimental Streams and the R. Tean are shown in Tables 5.18 and 19. Mean dry weight and ash free calories/$0.05m^2$/yr summated for the trophic entities have been drawn in bar pyramid form (Figs. 5.7 & 8), and histograms of mean total macroinvertebrate standing biomass/0.05m^2/yr have been drawn (Fig. 5.10).

Results.

The pyramids representing mean standing biomass of the trophic entities in mg/0.05m^2/yr show how the entity structure varies with the degree of eutrophication. Stream 2 (Fig. 5.7) which is composed of 100% river water approximates best to the rithron condition and to the condition of the R. Tean at Station 1 (Tables 5.21) although differences, for reasons discussed later, do exist. Stream 2 demonstrates the largest proportion of its weight/0.05m^2/yr as grazers, followed fairly closely by detritus feeders and then by carnivores. Streams 1 and 3 however, which contain 37% and 87% sewage effluent respectively, demonstrate the dominance of the grazing population. The carnivorous component in these two streams also show a decline in terms of weight which is a result of not only less species and numbers but the lack of large species like Erpobdella and Rhyacophila. This incompatibility of comparison when large numbers of molluscs and carnivores are encountered is easily reconciled by reference to the bar pyramids of mean calories/0.05m^2/yr. (Fig. 5.8). Energy values in this form were calculated from mean dry wt/0.05m^2/yr and the calorific values (ash free) determined for the species (Table 5.20).

The trophic entity energy bars for Stream 2,show more or less the same pattern of trophic structure, except that it appears as though here too the detrital feeders are proportionally larger (in terms of calories) than the

FIG.5.7. Pyramids of three trophic entities as mean standing biomass in the Experimental Streams. Expressed as mg dry wt/$0.05m^2$/yr.

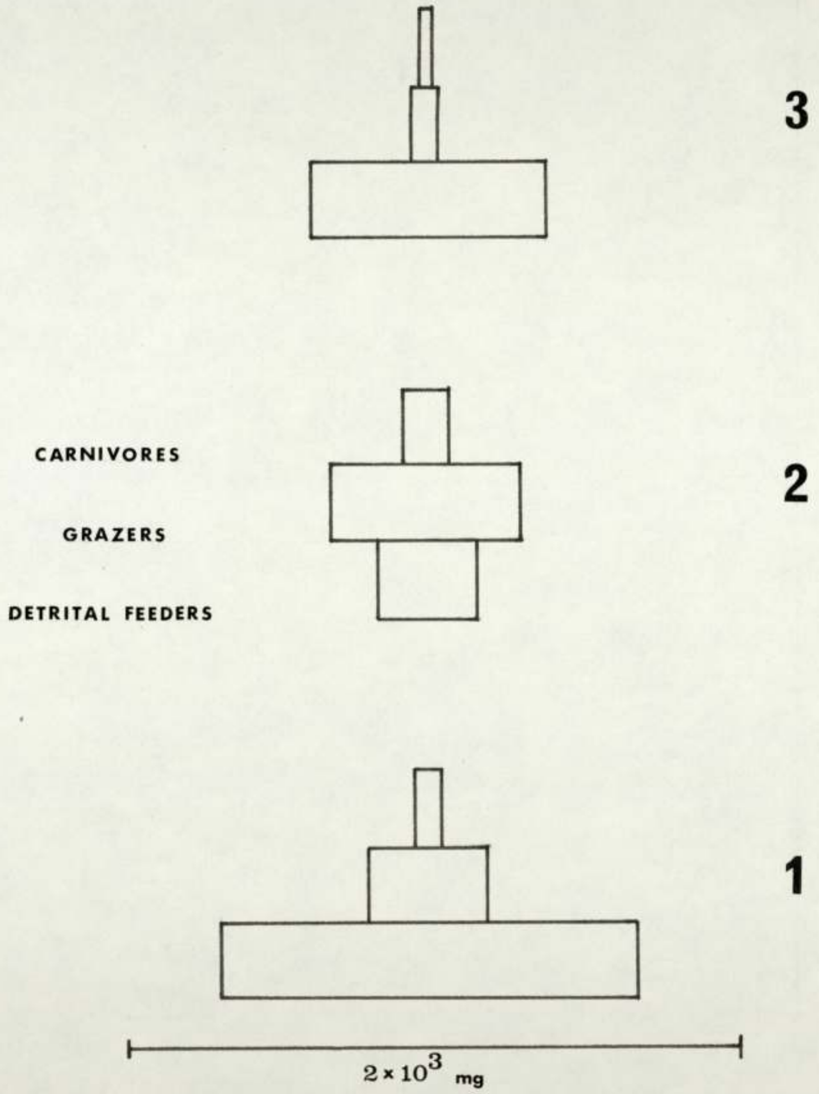
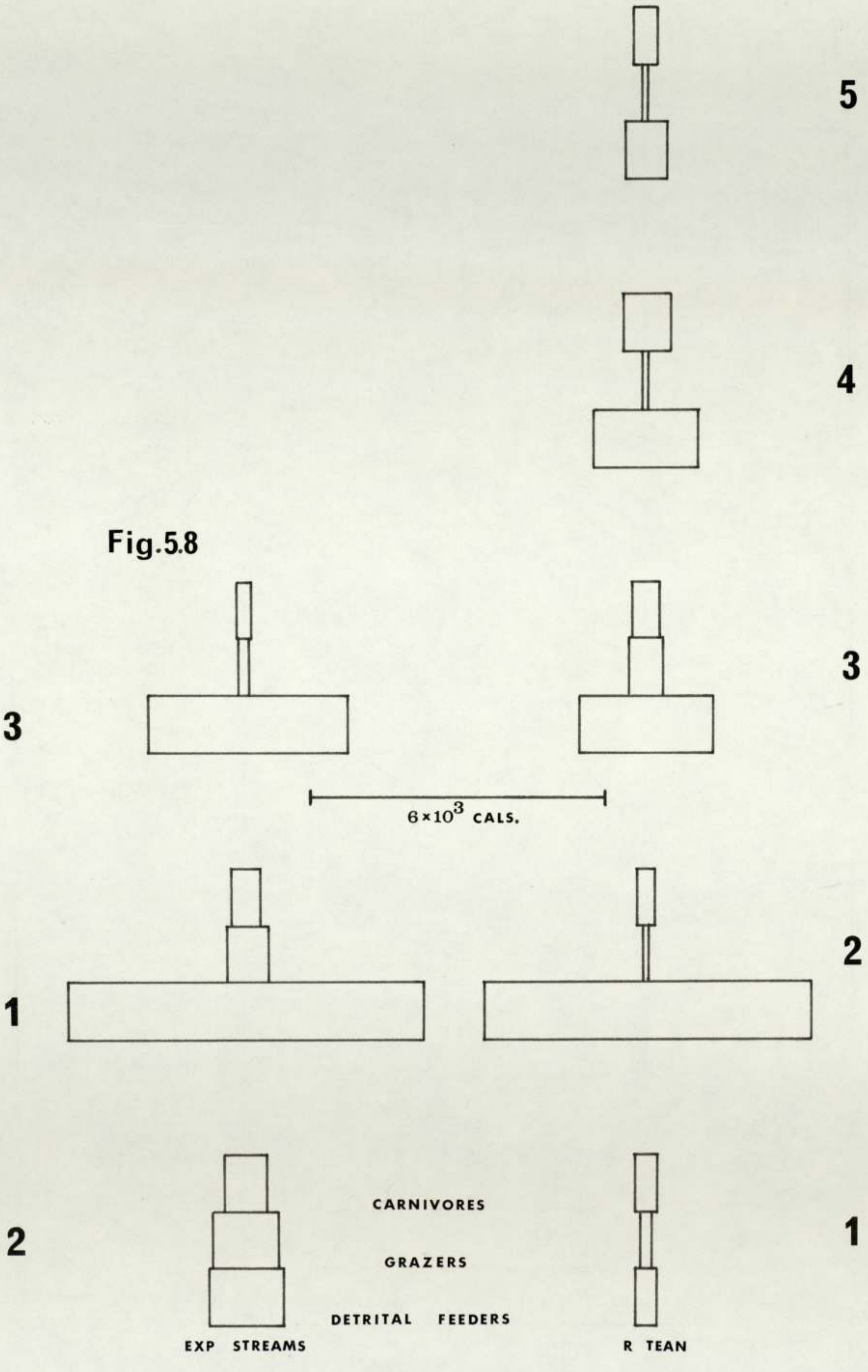


FIG.5.8. Pyramids of three trophic entities as mean standing biomass in cals/0.05m^2/yr. (ash free dry wt.), in the Experimental Streams and Stations 1-5 of the R.Tean.

Fig.5.8



grazing population which only appeared dominant before (Fig. 5.7) because of the Lymnaea shell weights. The structure of the Tean community at Station 1 is also very similar to that of Stream 2 but a difference is apparent in the amount of biomass. However, it must be remembered that there are neither fish to predate the Streams nor are there any spates to deplete the standing biomass. In both the river and the Stream the dominant carnivores are Rhyacophila and the leeches: - E. octoculata in the Stream and G. complanata in the river. Another notable difference exists in the grazing population where Lymnaea is by far the dominant grazer in the Stream (Table 5.19) but is virtually absent in the river (Table 5.18) where Ancylus and Baetis are the major herbivores. It is suggested that the artificiality of the Stream in terms of consistent flow and build up of Cladophora and Bryophytes favours Lymnaea apart from any lack of predation.

The proportion of grazers in Stream 1 is likewise diminished when observed in terms of calories, this again being due to the weights of the molluscan shells. The standing biomass of detrital feeders is by far the greatest of all the three Streams and of all the Stations of the Tean with the exception of Station 2 which has a very similar composition. This might be expected as Station 2 is the first Station below the outfall and is similar with respect to its water chemistry; the permanganate value, suspended solids, nitrogen and phosphorus components being particularly equatable. The standing biomass of detrital feeders in Stream 3 is not as great as that of Stream 1 but does exceed the equivalent trophic entities of the Tean Stations with the exception of Station 2. The structure differs slightly from that of Stream 1 with the percentage of detrital feeders accounting for 91% of the mean total calorific biomass as opposed to the 84% of Stream 1. The percentage proportion of grazers and secondary producers are essentially similar in both Streams 1 and 3 and the lack of some species and calorific biomass is seen as a result of the harsher chemical conditions, especially with respect to ammonia, oxygen and suspended solids.

The gradual reduction of the standing biomass of detrital feeders downstream of the outfall (Fig. 5.8) is indicative of the diminishing amount of organic solids that are being degraded by bacteria, eaten by decomposers and settling on the substratum as detritus. The graph of mean solids against distance on the R. Tean (Fig. 5.5) demonstrates this point and the rise encountered after Station 4, most likely caused by increased current and scouring, gives rise to a greater proportion of inert solids which would not be expected to increase the proportion of decomposers. Also, the shifting nature of the substratum at Station 5 together with the speed of the current is not conducive to large populations of Asellus, Gammarus or Simulium, and in fact 43% of the decomposer population is composed of tubificid worms, which are capable of withstanding these conditions.

The grazing population, which appears to be at its largest at Station 3, is similar in both structure and total energy to the equivalent trophic level of Stream 1. The greatest resemblance is in the molluscan populations and while Stream 1 carries a greater proportion of these, the *B. rhodani* population at Station 3 balances the total energy for the trophic level. The reasons for the magnitude of the grazing populations at this Station compared with all the other Stations of the Tean may well be related to the churn washings discharged from the dairy just above Station 3. Pentelow *et al* (1938) also noticed an increase in pulmonate moluscs below a dairy effluent although they recorded *Hydrobia* as depleted in numbers. Oddly enough, however, Station 3 of the Tean is the only Station at which *Hydrobia* was recorded. It is at this juncture that the ambivalence about the placing of *Lymnaea* in the grazing population becomes apparent. The exceptional density of this species, it is thought, may be due to its grazing the bacterial slimes that noticeably result from the milk wastes which, under the above definitions of trophic levels, would place it into the detrital category and reduce the biomass of the grazers substantially. However, it is presumed that epiphytic as well as the more obvious filamentous growths were occurring which, while supporting the large populations of *B. rhodani* could equally well have been maintaining the *Lymnaea* community.

The grazing populations at Stations 4 and 5 are nothing like the proportions of Station 3 yet they are not far removed from the control situation at Station 1. With the increasing amount of the filamentous alga *Cladophora* from Station 3 onwards downstream, reaching a peak at Station 4, a larger grazing component might have been expected especially as the water quality was good enough by this stage to support all but the very sensitive plecopterans, ephemeropterans and trichopterans. From observations in the R. Ray where huge blankets of Cladophora containing massive densities of Asellus occur, it had been noticed that those separated from the alga possessed bright green guts, indicative of them feeding off their supporting medium. Furthermore, the author kept a healthy population for over a month after which time they still appeared to be growing whilst their only food source had been that of Cladophora. Therefore, it appears that an Asellus population can, at least partially, survive on the diet of a herbivore which may well be the case at the lower Stations of the Tean. It is seen from the histograms of numbers of Asellus against distance downstream (Fig. 5.11) that Asellus is most abundant at Station 4 and reference to the table of numbers against sampling date (Table 5.13) demonstrates its occurrence with the Cladophora season - of course its largest broods also occur in March giving large juvenile recruitment in May onwards (Fig. 8.1., Chapter 8). The grazing component at Stations 3 and 4 in particular would be enlarged by the transference of obviously some of the Asellus that are feeding within the protection of the Cladophora blankets, and the detrital component would be functionally reduced.

Coincident with the decline of the detrital component in the Tean downstream of the outfall is the increase in the secondary consumers (a situation not matched in Stream 3). While the water quality allows E. octoculata to dominate the carnivore level at Station 4, and E. testacea at Station 3 (Fig. 5.12), the proportion of carnivores at Station 2 is low (Fig. 5.8), which cannot be from lack of prey but more likely the result of harsher chemical conditions.

The total mean standing biomass of macro-invertebrates depicted as histograms in the form of mg dry wt./ $0.05m^2$/yr and cals/ash free dry wt./$0.05m^2$/yr (Fig. 5.10) demonstrate the relative productivity in these terms between the Streams and also between the Stations of the R. Tean. 'Productivity' is being used in the sense of relative standing biomass of macro-invertebrates. By far the most productive of the Streams is Stream 1 which has a total standing biomass (energy) 57% higher than that of Stream 2 and 47% higher than that of Stream 3. The latter is 20% greater than Stream 2 which is seen as the least productive with reference to cals/$0.05m^2$/yr.

The productivity of the Tean Stations are seen to vary in a similar manner, i.e. Station 1, which has the same water chemistry as Stream 2, is the least productive, followed by Station 5 where dilution is at its greatest and nutrients at their lowest concentration. Most similar to Stream 1 is Station 2 which has 83% of the biomass (energy) that the artificial stream carries and by far the greatest production in the whole of the Tean. Stations 3 and 4 are progressively less productive although they do not lag far behind the crop for Stream 3.

The total invertebrate standing biomass for the Station on the R. Ray is undetermined, but the energy from the <u>Asellus</u> population alone is a considerable proportion

FIG.5.9. Histograms of annual mean numbers of chironomid larvae per 0.05m^2 in the Experimental Streams and Stations 1-5 of the R.Tean.

FIG.5.10. Histograms of mean standing biomass of macro-invertebrates in the Experimental Streams and Stations 1-5 of the R.Tean expressed as mg dry wt./0.05m^2/yr. and cals/ash free dry wt./0.05m^2/yr. (stipple).
The R.Ray histogram represents the total standing biomass of Asellus aquaticus only

Fig. 5.9

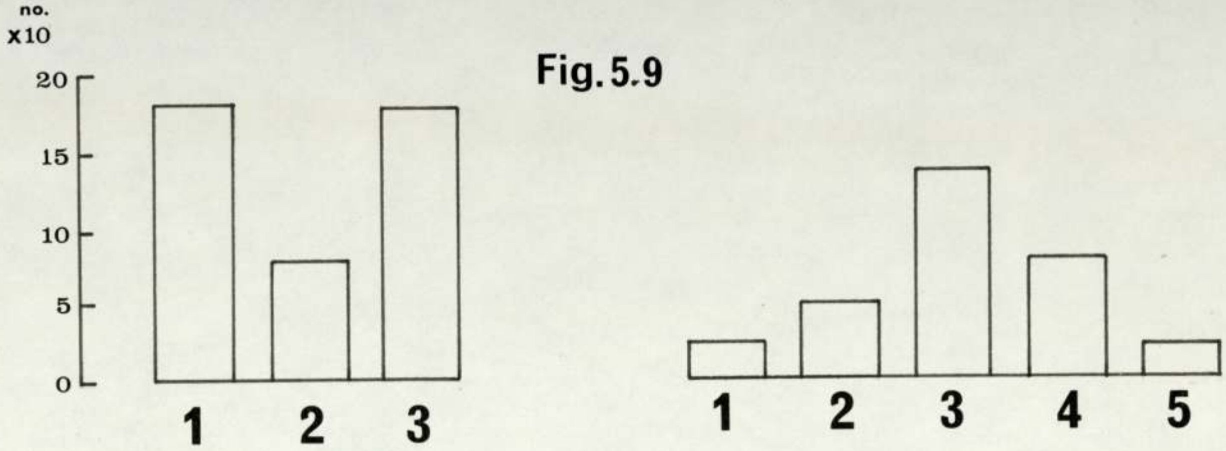
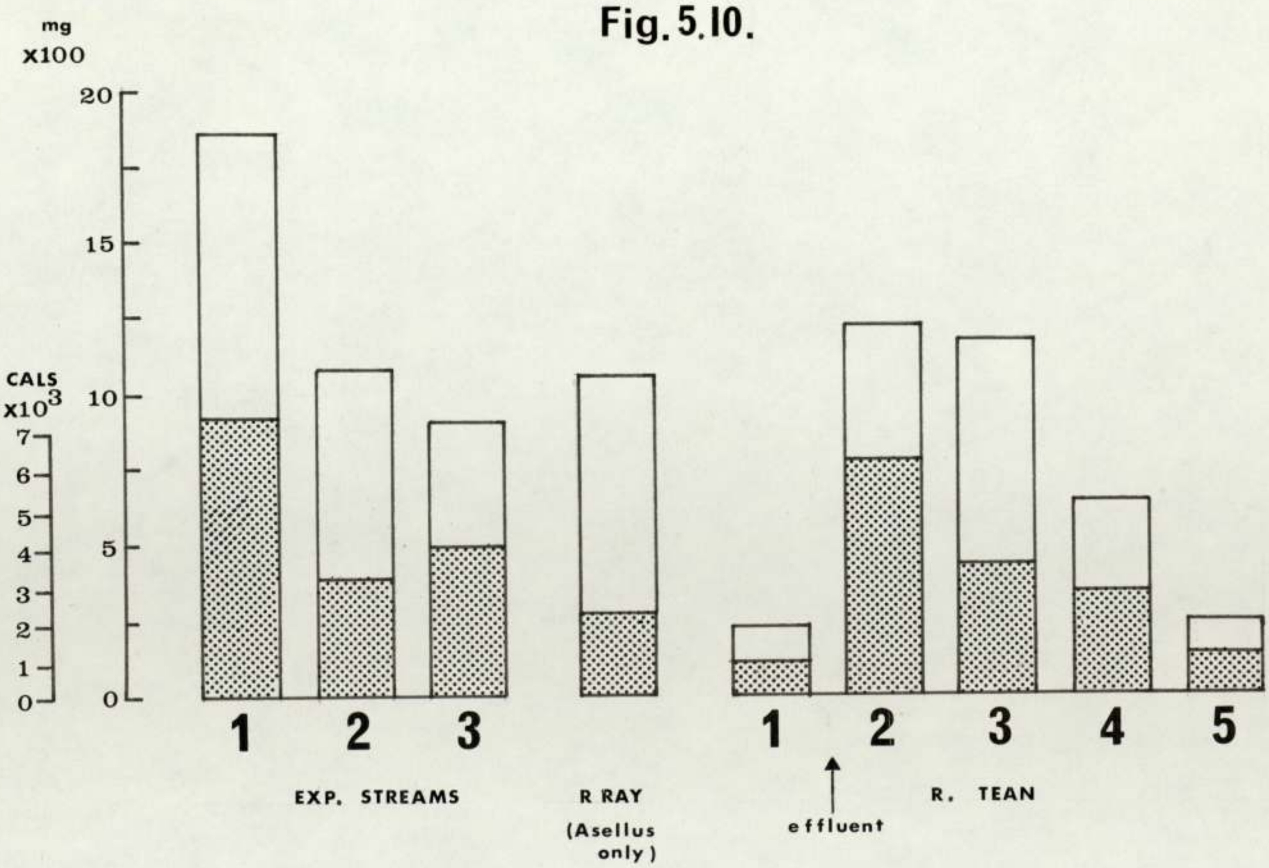


Fig. 5.10.



relative to the totals for the Streams and the R. Tean. In fact the standing biomass of Asellus in the R. Ray is only a little greater than the standing biomass of Asellus in Streams 1 and Station 3 of the Tean.

Standing crop of Asellus aquaticus. cals/0.05m^2/yr (ash free)

Stream 1	Station 3	R. Ray.
1854.67	1904.5	2094.05

Also of interest are the relative proportions of Simulium to the total standing biomass per year. Simulium, which is particularly favoured by a constant flow and a stable substratum (Muller, 1953) formed high percentages of the total biomass where they were in contact with a high concentration of bacteria, i.e. in Stream 3 (87% effluent) Stream 1 (37% effluent) and Station 2, below the outfall.

Simulium ornatum as a percentage of the total standing biomass cals/0.05m^2/yr (ash free).

Stream 1	Stream 3	Stream 2	Station 2	Station 3
55	63	1	87	3

The percentage of total biomass in Stream 2 highlights the role bacteria are playing in the production of Simulium by their absence. The dramatic drop in percentage at Station 3 is, however, thought not to be the result of bacterial diminution (the permanganate value clearly demonstrates high amounts of of oxidisable material still) so much as predation by fish and competition from other species which are able to live in the improved water quality by this distance downstream.

5.4.1. DISCUSSION.

It has been shown that the trophic structure of macro-invertebrates within both the Experimental Streams and the R. Tean varied with the degree of eutrophication. By eutrophication it is usually understood, as pointed out by the definition in the Introduction, that there occurs a fertilisation of natural waters that results in increased primary production. Moreover, and consequently, there are also increased in the productivity of the herbivorous, primary, secondary and perhaps tertiary consumer populations, i.e. there is a change in the community structures and productivities. The situation in the Experimental Streams, with the exception of Stream 2, is that they are receiving large amounts of organic matter in the form of detritus, fungi, and bacteria, as well as the high levels of plant nutrients which also encourage the growths of algae. In a eutrophicated river the enlarged algal growths inevitably soon become detritus due not only to death but as a result of passing through the guts of herbivores and, perhaps for a second time or more, likewise through scavengers. This rain of detritus attracts large numbers of bacteria and fungi, which increase in numbers owing to the nutrients present and degrade the detritus. So this situation is not so very far removed from that of the Experimental Streams or that below an outfall where large numbers of bacteria, fungi and protozoa emanate from the effluent. In this sense then it is feasible to define the Experimental Streams as being eutrophicated while, although they are certainly organically enriched as the recovery zones of the Upper Cole were, the difference lies in the degree of nitrification during treatment and oxygen demand subsequent to the effluents discharge, which permits or eliminates members of the benthic community. If this definition is acceptable, then the large percentages of Simulium larvae that comprise the benthic communities in Streams 1 and 3 and Station 2 of the R. Tean are the result of eutrophication rather than organic pollution and it is increasingly possible to see the similarity between the two types of pollution.

The reasons for the differing structures of the trophic entities between the Streams and the Stations of the Tean are manifestly concerned with the relative proportions of effluent under which the communities survive. However, other factors concerning these inequalities of structure and biomass, either directly or indirectly, can be discerned.

The effluent in Streams 1 and 3 and in the R. Tean below the outfall must be restricting a number of species through its chemistry, i.e. toxicity, despite the generally high quality of discharge. Noticeably the detrital entity in Streams 1 and 3 are dominated by those species which are fairly tolerant to the high ammonia levels occasionally encountered under a lowered oxygen regime, which is a way of saying that the herbivore component that is usually associated with well-oxygenated conditions of the rithron is reduced in the Experimental Streams and the Tean directly because of the water quality. The distribution of the three Ephemeroptera and Ancylus in the Streams is a case in point, and even the Lymnaea population in Stream 3 is sparse (Table 5.19). A similar position is evident in in the distribution of carnivores, where Rhyacophila dorsalis and Erpobdella octoculata are greatly reduced in number in Streams 1 and 3 when compared with the population of Stream 2 (Table 5.13). The more tolerant species E. testacea has managed to colonise Stream 1 scantily and its absence from Stream 2 (from competition with E. octoculata?) and Stream 3 bears witness to the chemical effect.

The absence of fish as the top carnivores in the Experimental Streams is a result of design rather than water quality, although at Station 2 on the Tean the massive proportions of the Simulium population is indicative of reduced predation by fish whose activities may be restricted by the proximity of the outfall. In any case, the lack of predation by fish in the streams must have had profound effects upon the trophic structure and standing biomass of the benthic communities. Predation of this kind is seen as being primarily responsible for the differences in standing biomass between Station 1 of the Tean and Stream 2 and constitutes the major attribute of artificiality of the Experimental Stream ecosystem. By Station 3 of the Tean and also at the downstream Stations, where fish are known to be numerous once again, the biomass is similarly reduced as opposed to that of Station 2, and it has been pointed out that the Simulium populations in relation to bacterial activity cannot be solely responsible for the reduction of their numbers.

Another facet of the artificiality of the Streams is the constant flow or, more significantly, a lack of spates which in the R. Tean must account for a substantial reduction in the standing biomass through drift and scouring action. A 24-hour drift net collection made in May 1973 (Table 5.22) demonstrates the virtual non-existence of the drift in the Streams, with the exception of chironomid adults (carried along on the surface before flying off) which are not included in the benthic biomass anyway. Although drift in the R. Tean has not been measured, it is certain that it must be of the same magnitude as other rivers and during times of high water the amount of biomass dislodged is no doubt substantial.

Natural variation in biomass owing to the effects of drift and spares is a phenomenon that is not encountered therefore in the Experimental Streams. This, together with the total lack of fish predation is probably responsible for the larger standing crops of invertebrates demonstrated in the mean total biomass figures as opposed to those determined for the R. Tean (Fig. 5.10). If time had allowed the intended introduction of fish into the Streams, this conclusion might have been ratified. It is fairly clear then that the 'productivity' of the Streams in terms of standing biomass (both weight and energy) is of a higher level than that of the R. Tean. However, whereas the biomass of the Tean is mainly controlled by the predation of fishes and natural phenomena, it is the intraspecific competition that must be uppermost in the regulation of the biomass of the Streams. Also, and perhaps more significant ecologically, is the inference that although the standing biomass in the Tean is lower than in the Streams the total production is probably much higher.

Interspecific competition has particularly attributed to the type of trophic relationships seen because in the first instance the streams were void of life at the start of the research. This is shown clearly in the next section pertaining to the seasonal innovation of species. As the trophic entities are compiled on the basis of the annual means (dry weight) from March 1973 to March 1974 when a great period of recruitment of species was occurring, they are descriptive of a changing community - a situation not true of the R. Tean whose ecological structure was obviously, due to time, fairly stable. Some of the major changes in species inhabiting the Streams occurred during the year in question and in particular numbers and weight of Gammarus, Asellus, Lymnaea, Ancylus, and chironomid larvae (Figs. 5.15 - 17). This is clearly because of the life histories and other biological phenomena, examples of which have been outlined in the discussion on the recovery of the Upper Cole from pollution. But a steady deposition of solids and detritus in the Streams has also affected the biotope with respect to inhabitable space; and competition, especially in Stream 3 where deposition is so severe, has shown itself in the reduction of numbers of many species, particularly chironomid larvae (Fig. 5.14). The biomass then, or more accurately the stability, which is obviously still in a state of change must naturally affect the trophic structures, and the trophic pyramids described only refer to a year of substantial ecological changes.

Nevertheless the pyramids of the trophic levels are not so very far removed in structure from those encountered on the R. Tean below the outfall and the major difference of carnivore composition is being rectified in the Streams with the appearances of the Hirudinea, which have been slow in establishing themselves.

The overall picture in the Experimental Streams then is one of eutrophication increasing the proportion of detrital feeders and reducing the proportion of grazers and carnivores, the Control Stream 2 containing 100% river water supplying clear evidence of this. The intensity of eutrophication, as determined by the proportions of river water to effluent in Streams 1 and 3, has profoundly differing effects upon the standing biomass. The degree of eutrophication encountered in Stream 1 issues a massive 'productivity' restrained principally by intraspecific competition while a greater degree of eutrophication, as shown by Stream 3, does not necessarily mean a greater 'productivity'; on the contrary, it is reduced in this case to 53% of that of Stream 1. Stream 3 does however have a greater productivity than the Control Stream, with not only intraspecific competition checking higher productivity but water quality in terms of oxygen, ammonia, and solids exerting a density independent effect also.

An analagous picture appears in the R. Tean at those stations below the outfall, the optimum standing biomass occurring some 300 yds. below the discharge as a result of both idealised conditions for <u>Simulium</u> production, and reduced fish predation owing to the water chemistry. The effects of eutrophication are more realistic at Stations

3 and 4 where the bacterial and hence Simulium proportions are reduced, and an active fish population is present.

The R. Ray at Moredon must support an equally productive fauna although only the dominant Asellus has been quantitatively assessed.

5.5 BENTHIC MACRO-INVERTEBRATES AND EUTROPHICATION.

It is intended to discuss the species and density of benthic macro-invertebrates in relation to various aspects of eutrophication as they occurred in the Experimental Streams and in the Rivers Tean and Ray. The seasonal data collected in the R. Tean and the Experimental Streams are shown in Tables 5.12 - 17. Mean numbers/$0.05m^2$/yr of the larger populations in the R. Tean have been drawn as histograms against the distance downstream in km. to its confluence with the R. Dove (Figs. 5.11 - 14). The major components of the benthic fauna inhabiting the Experimental Streams have been graphed against time to illustrate any seasonal fluctuations and colonisations made over the 18-month period (Figs. 5.15 - 20). Each point on these graphs represents the mean of the summed sample means for three consecutive samples thereby eliminating very small fluctuations in density, although where very small numbers were encountered in the Chironomidae actual numbers have been plotted.

The annual means of dry weight and numbers/$0.05m^2$ for species in the Experimental Streams (Tables 5.12 - 14) refer to the period 22.3.73 - 13.3.74, which excludes the very early data when the benthic communities were only just establishing themselves.

(a) Species distribution and the chemical effects of eutrophication.

The considerable amounts of allochthonous material encountered as a result of a 'point source' of nutrients like the sewage effluent in the R. Tean has been alluded to. The micro-organisms involved in the breakdown of organic material from the effluent and of animal and plant remains - especially that of *Cladophora* in the R. Tean, Ray, and Streams 1 and 2 - involve the utilisation of oxygen. Also some of the direct effects of the effluent has been mentioned with reference to ammonia and the limitation of species.

A combination of the 'oxygen sag' and ammonia concentrations observed in the Tean just downstream of the effluent (Fig. 5.5) is obviously responsible for the distribution of some sensitive species. The histograms (Fig. 5.12) give evidence of this when considered with the ammonia curve (Fig. 5.5). *Baetis rhodani*, *Gammarus pulex*, *Ancylus fluviatilis*, and *Rhyacophila dorsalis* show a reduction in numbers after Station 1 while the more tolerant species like *E. testacea* and *E. octoculata* appear below the effluent. However, the continuation of lower densities after the NH_3 peak which has subsided by Station 3 is attributable to the change in the dynamics of the food source and community structure, i.e. the increase in detritus inhibits the diatomaceous growth and herbivores like *Baetis* and *Ancylus* cannot increase their numbers to former proportions especially in the face of competition for space by the detrital feeders.

Lymnaea pereger is only found in abundance below the dairy effluent (Fig. 5.12), *E. testacea* and *E. octoculata* being dominant invertebrate carnivores at Stations 3 and 4 respectively. (This succession is reminiscent of gross

FIG.5.11. Histograms of mean numbers/0.05m^2/yr at Stations 1-5 of the R.Tean. Distance of Stations from confluence also shown.

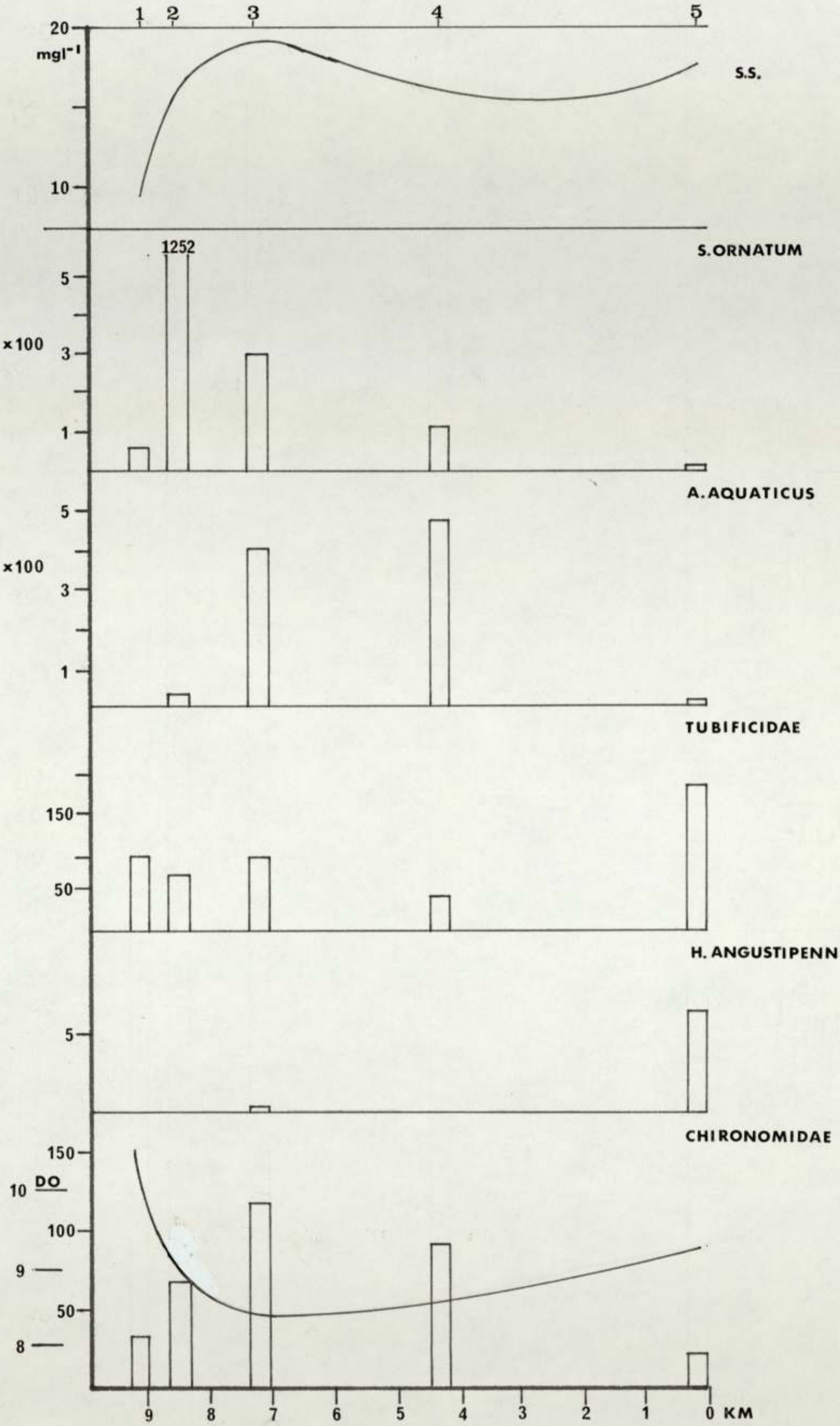


FIG.5.12.

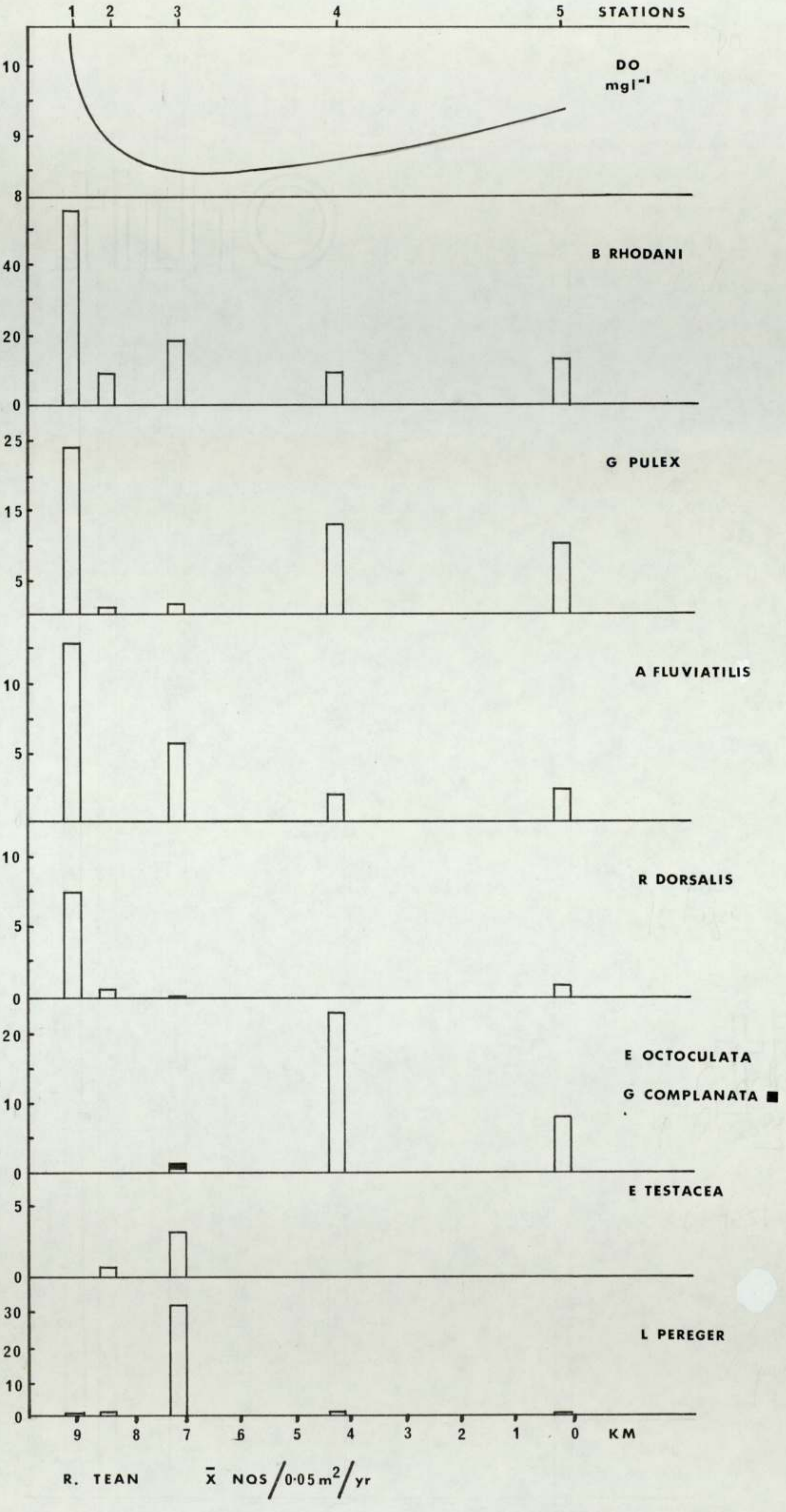

1
2
3
4
5
STATIONS
10
9
8
DO mgl^{-1}
40
20
0
B RHODANI
25
15
5
G PULEX
10
5
A FLUVIATILIS
10
5
0
R DORSALIS
20
10
0
E OCTOCULATA
G COMPLANATA ■
5
0
E TESTACEA
30
20
10
0
L PEREGER
9
8
7
6
5
4
3
2
1
0
KM
R. TEAN
$\bar{x}$ NOS / 0·05 m^2 / yr

FIG.5.13. Histograms of mean numbers of chironomid larvae per $0.05m^2$/yr. at Stations 1-5 of the R.Tean. Distance of Stations from confluence also shown.

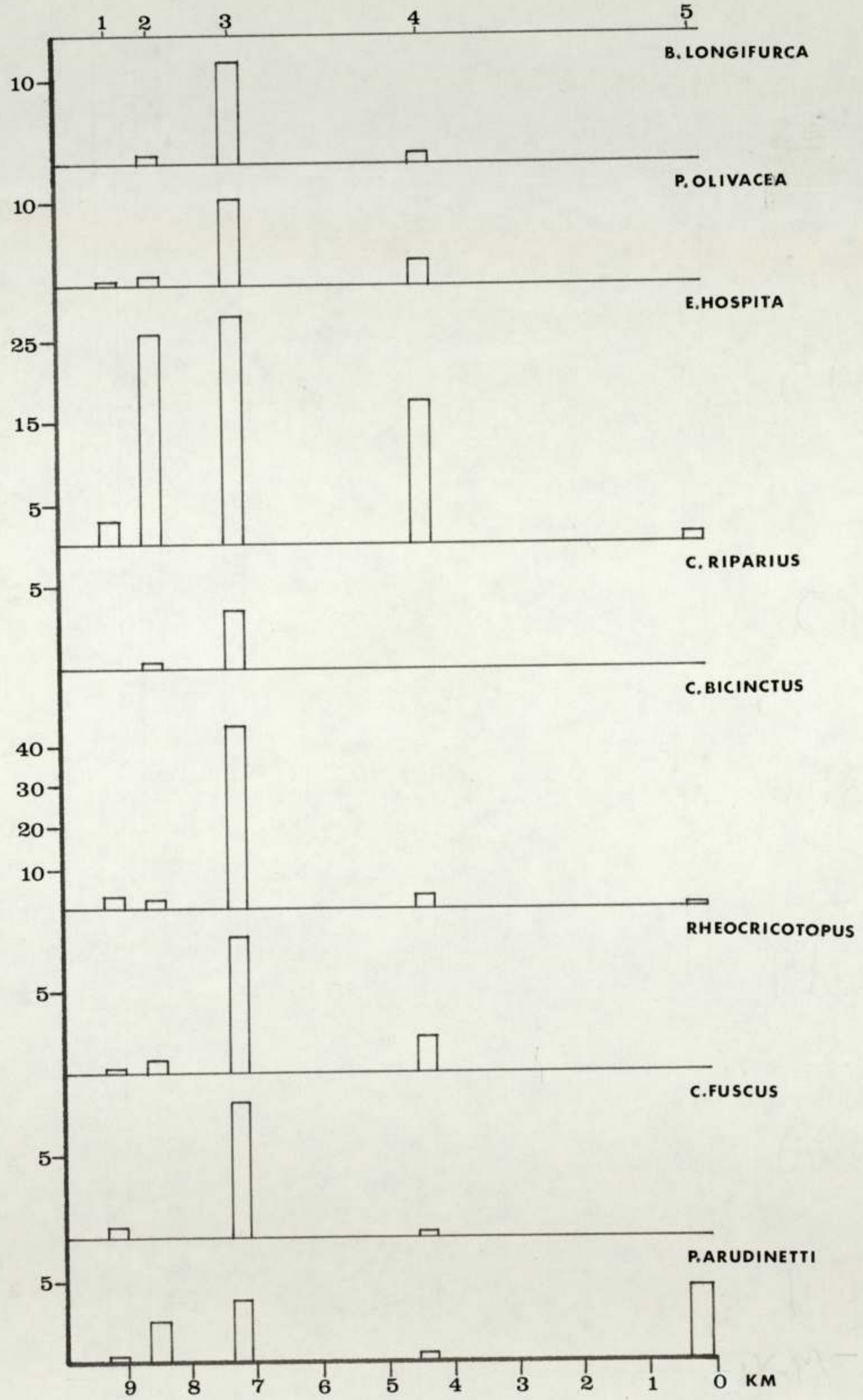
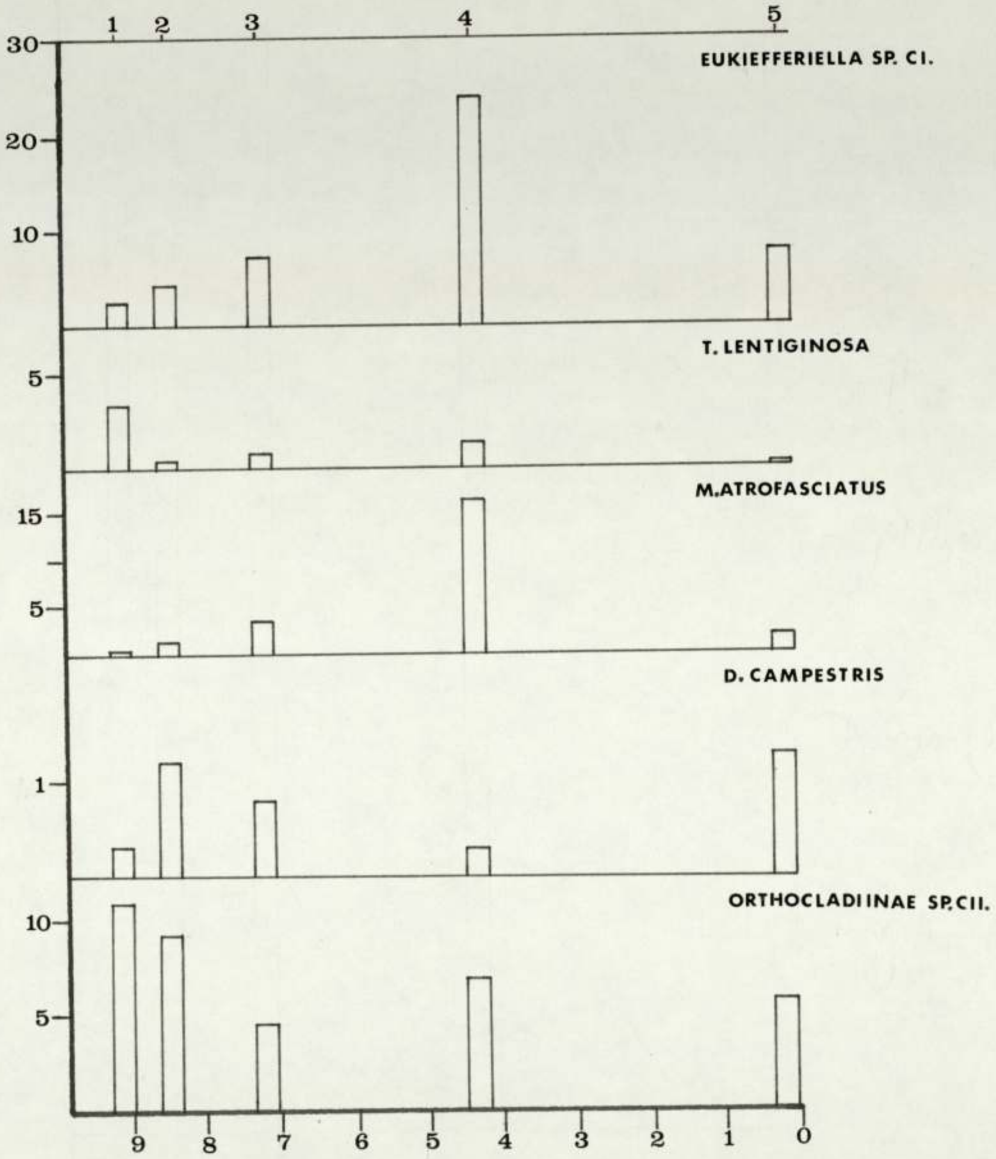


FIG.5.14. Histograms of mean numbers of chironomid larvae per $0.05m^2$/yr.at Stations 1-5 of the R.Tean. Distance of Stations from confluence also shown.



organic pollution).

The densities of G. pulex, Ephemerella ignita, Ecdyonurus venosus and occasional appearances of sensitive Trichoptera (Table 5.13) appear to be primarily distributed in the Experimental Streams by the water chemistry, as they are effectively absent in Streams 1 and 3 but not in Stream 2 (Fig. 5.15, Table 5.13). While the food supply is ample for Gammarus, the competition by Asellus is great and, while it is true that increasing siltation in Stream 3 denies Gammarus the habitat it requires, this is not the case in Stream 1 where abundant Leptodyctium, Fontinalis, and Cladophora offer suitable habitats. E. venosus, which has virtually the same diet as B. rhodani, is only recorded in Stream 2 as is E. ignita, whilst B. rhodani being more tolerant of lower oxygen levels does survive in Stream 1 and also in Stream 3 when algal growth in April and May are at a maximum (Fig. 5.15). Further evidence of chemical interference is supplied by the absence of E. ignita in Streams 1 and 3 as it is known that this species not only feeds on diatoms and desmids, but also mosses, which are abundant in Stream 1.

It is uncertain whether Rhyacophila dorsalis is being affected by the depressed oxygen or elevated ammonia levels in Stream 3, but they both seem unlikely when one considers the mean of $>$ 80% saturation in Stream 3 (87% sewage effluent) and the work of Davies (1971) who found R. dorsalis to be more tolerant of ammonia than C. riparius. In respect of the large numbers of chironomid larvae and tubificids as food sources, it may be correct to assume that either Rhyacophila is more dependent upon an algal supplemented diet than is commonly believed or that siltation is responsible, the latter seeming very likely when the numbers in Stream 3 are considered (Fig. 5.17).

(b) Species distribution and the allochthonous and microbial effects of eutrophication.

Allochthonous material or, more definitively, detritus, can be considered together with microbial fauna and flora for the obvious reason that they recycle the nutrients and amino acids of the former. However, the microbial population present in the effluent is a special case owing to its concentration and effects in allowing huge densities of Simulium larvae to exist. This is most striking when the histograms of Simulium ornatum in the Tean and graphs of the Streams are examined in relation to the amount of suspended solids in the water (Figs. 5.11 and 5.16).

The expansion of the populations of macro-phagous detritus feeders like Asellus aquaticus, Gammarus pulex, Tubificidae, and the Chironomidae, together with the filtering Chironomidae and trichopteran Hydropsyche angustipennis are evident in the Streams and the Tean alike. Nevertheless it is equally likely that their distribution is determined by the occurrence of filamentous algae such as Vaucheria and Cladophora, and the Bryophyta. Exceptions in the cases of tubificids, some chironomid larvae, and Asellus in the Experimental Streams are noticeable when the numbers continue to increase despite the Cladophora and moss growths having receded in winter (Figs. 5.16., 5.17).

Clearly the numbers and species of chironomid larvae are increased with this aspect of eutrophication and

FIG.5.15. Seasonal numbers per $0.05m^2$ of three species of invertebrates in Experimental Streams 1, 2 and 3. Points are the mean values of three consecutive months means.

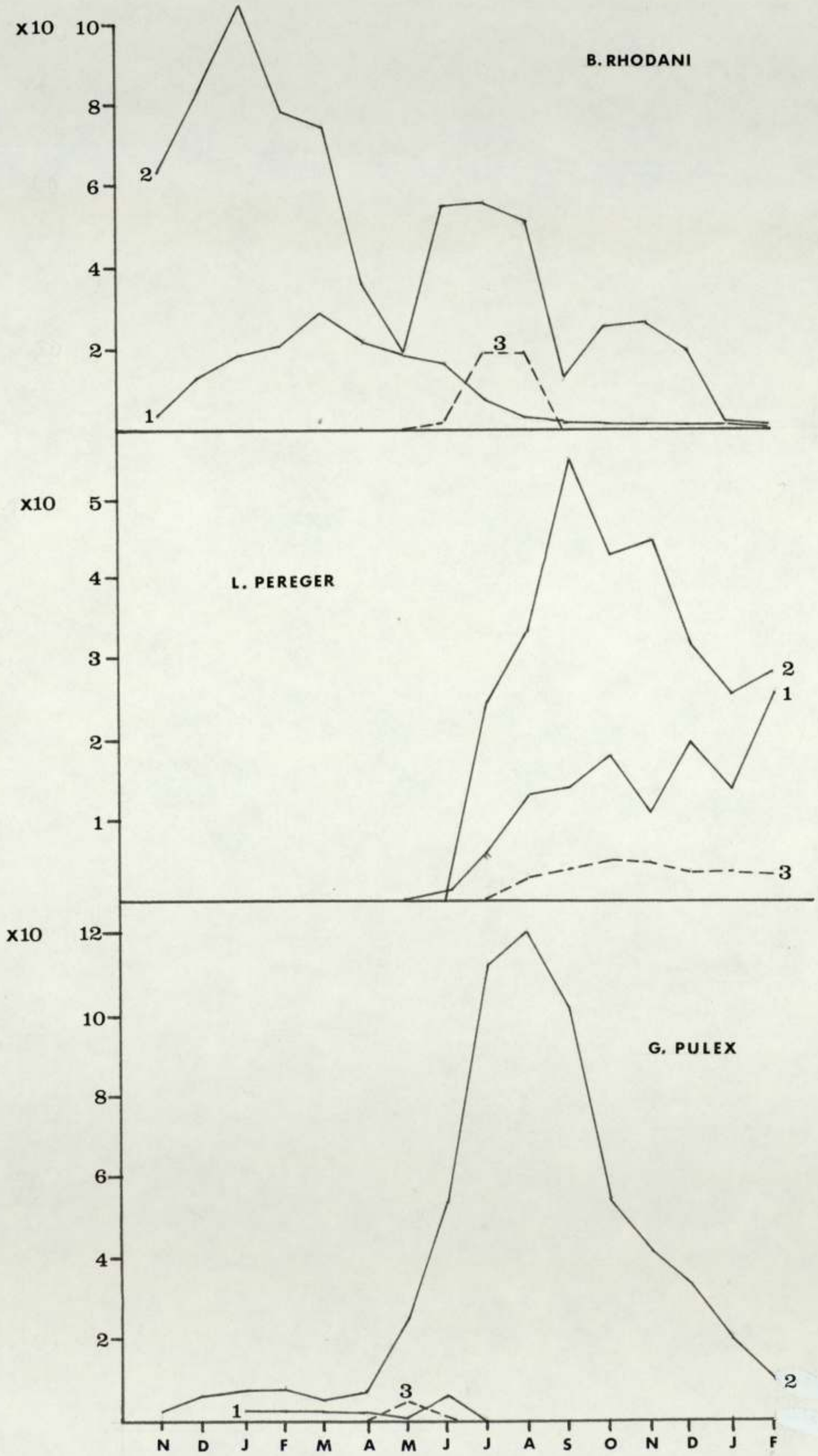


FIG.5.16. Seasonal numbers per $0.05m^2$ of two species of invertebrates in the Experimental Streams. Points are the mean values of three consecutive months means

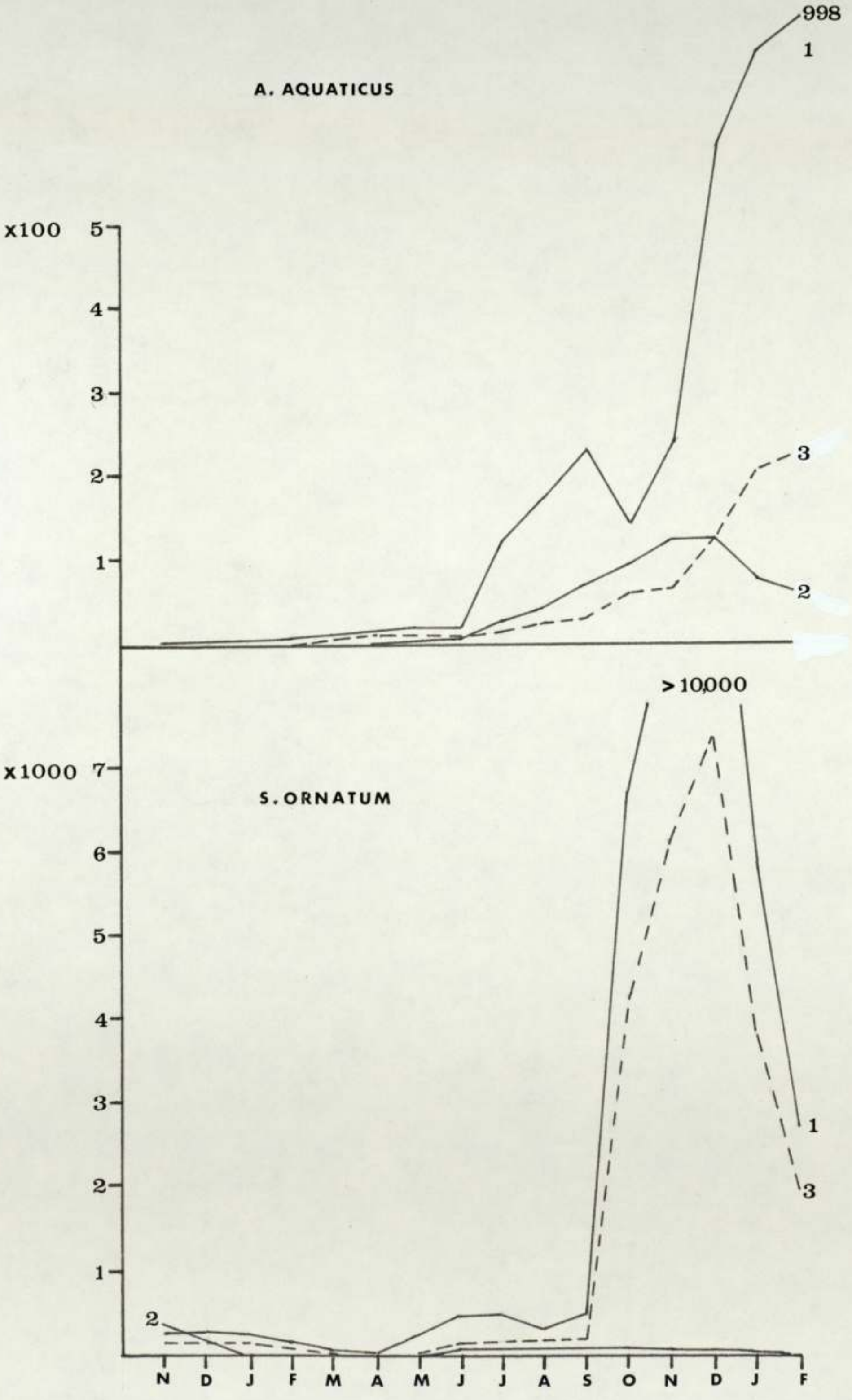
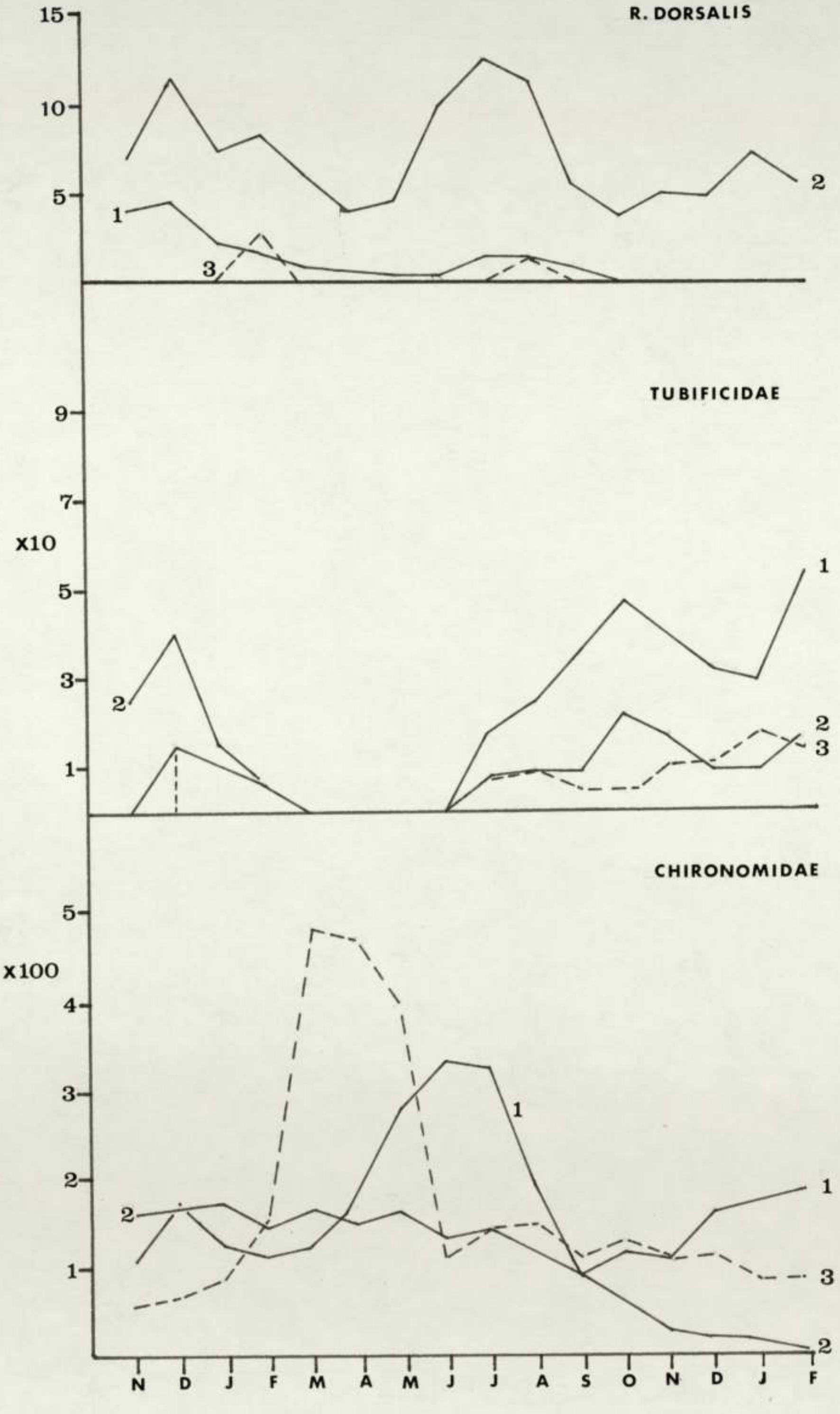


FIG.5.17. Seasonal numbers per $0.05m^2$ of one species and two families of invertebrates in Experimental Streams 1,2 and 3. Points are the mean values of three consecutive months means.



reach a peak at Station 3 of the R. Tean, no doubt because of the supplemented solids from the dairy effluent. The histograms of summed means of chironomid species density plotted against distance correlates well not only with the curve of suspended solids against distance but the inverse of the 'oxygen sag' curve (Fig. 5.11) thereby demonstrating their ability to withstand depressed oxygen levels while taking advantage of reduced competition and increased food. Similarly mean annual numbers in the Experimental Streams show the relative abundance of the larvae in Streams 1 and 3 as opposed to those in Stream 2 (Fig. 5.9).

The peak in solids which occurs as a result of both the effluent and dairy discharges into the Tean causes maximum density in the following chironomid species (also see Figs. 5.13 and 5.14). Brillia longifurca, Prodiamesa olivacea, Eukiefferiella hospita, Chironomus riparius, Rheocricotopus sp, Cricotopus (Cricotopus) fuscus, and Cricotopus (Cricotopus) bicinctus. Stream 3, which has a similar suspended solids loading, also has five of these species seasonally abundant.

Table showing seasonally dominant chironomid larvae at Station 3 (Tean) and in Stream 3

	Station 3	Stream 3
B. longifurca	+	+
P. olivacea	+	
E. hospita	+	+
C. riparius	+	+
Rheocricotopus sp.	+	
C. (Cricotopus) ex. gr. fuscus	+	+
M. atratulus		+
Lymnophyes sp.		+
D. ex gr. vitellinus		+
C. bicinctus	+	+

FIG.5.18. Seasonal numbers per $0.05m^2$ of 5 species of chironomid larvae in Experimental Streams 1, 2 and 3. Points are the mean values of three consecutive months means with the exception of the Brilla spp. which are actual monthly means.

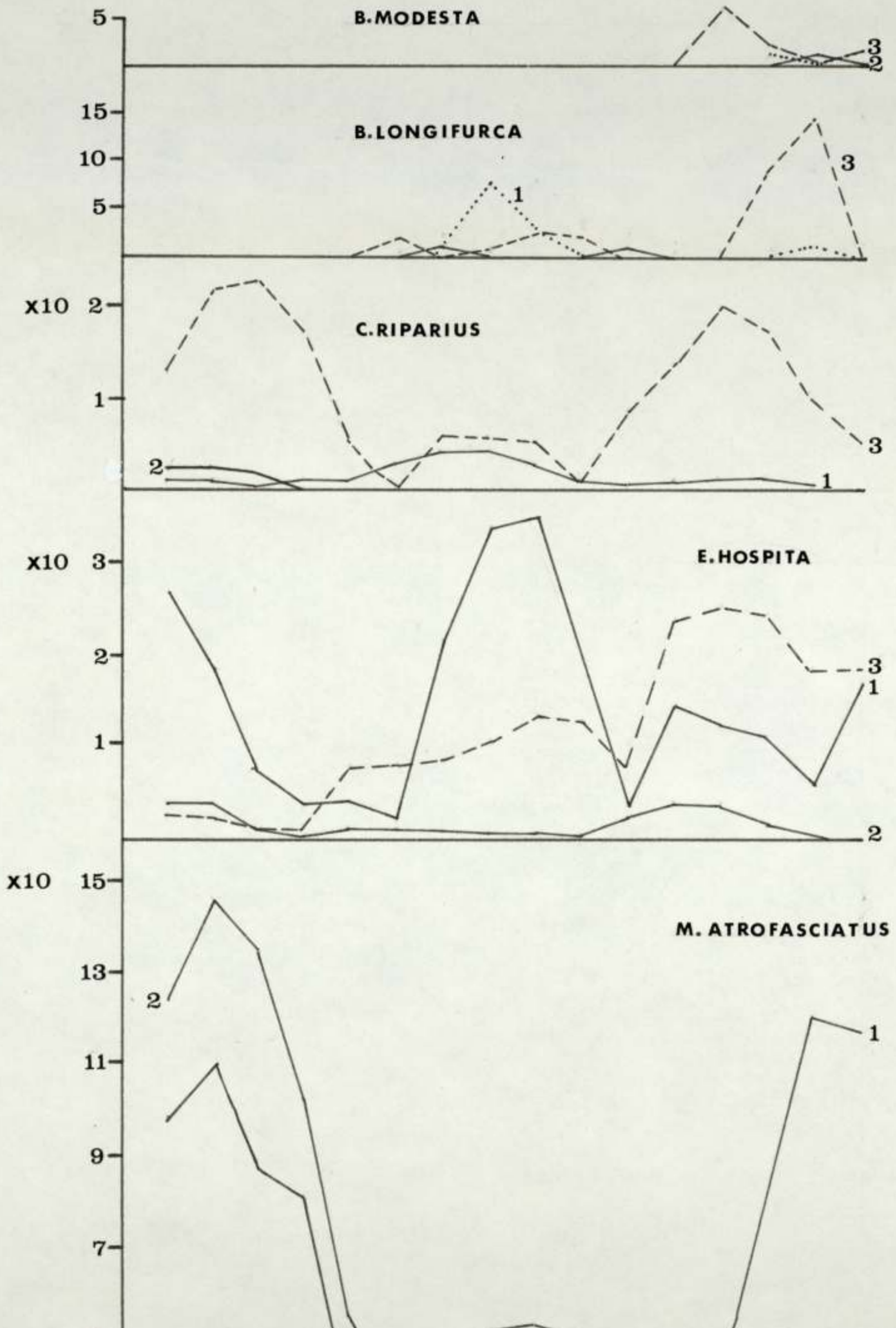


FIG.5.19. Seasonal numbers per 0.05m^2 of 4 species of chironomid larvae in Experimental Streams 1,2 and 3. Points are the mean values of three consecutive months means.

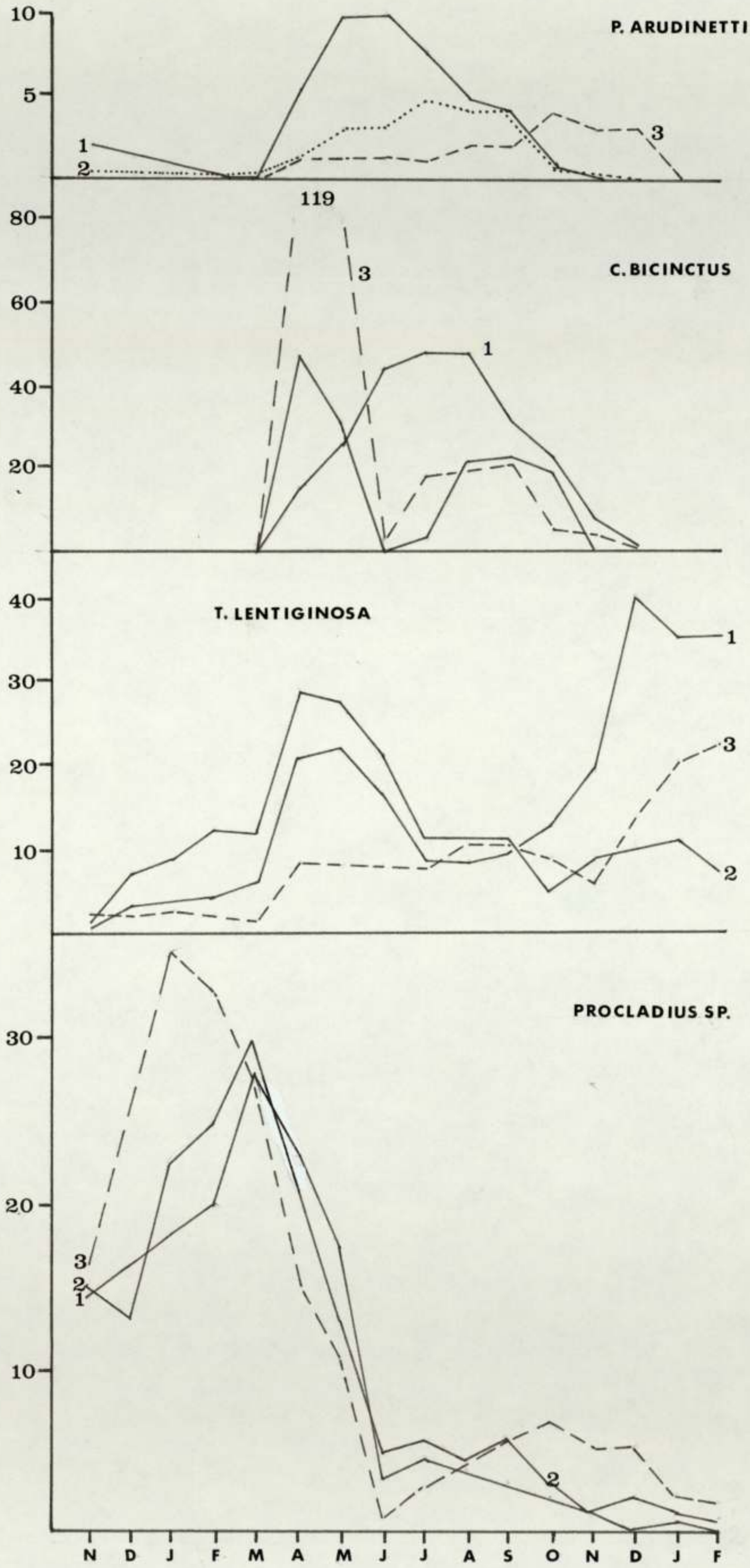
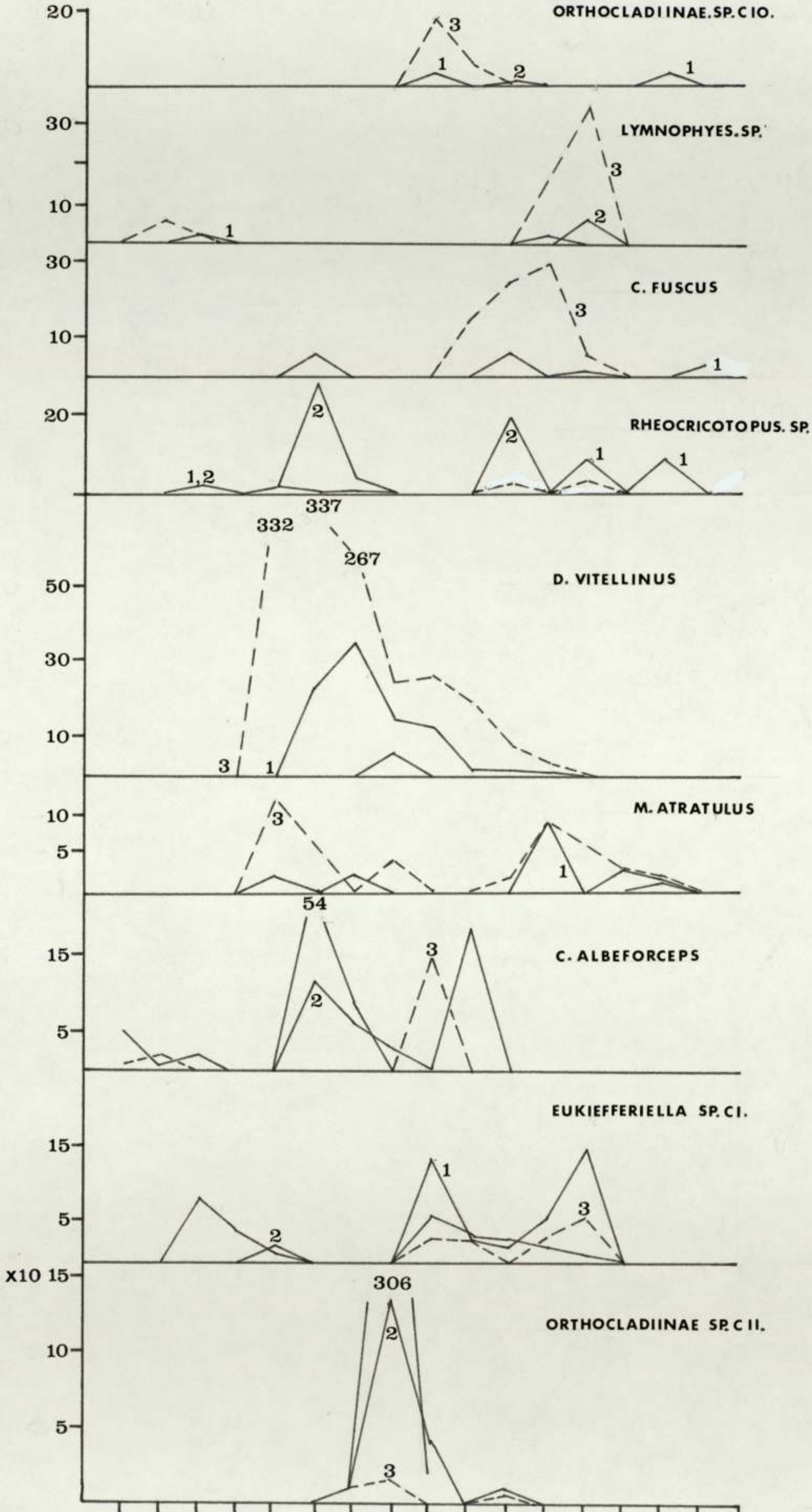


FIG.5.20. Seasonal numbers per $0.05m^2$ of 9 species of chironomid larvae in Experimental Streams 1,2 and 3. Points are the actual monthly means with the exception of D.vitellinus which are the mean values of three consecutive months means.



The two species that are not common in Stream 3 while being abundant at Station 3, i.e. P. olivacea and Rheocricotopus, are probably prevented from establishing large numbers because of the relative chemical severity of Stream 3. Rheocricotopus is seasonally abundant in Stream 2, and respirometric work on P. olivacea reported in Chapter 7 shows it to have a similar 'incipient limiting point' to E. testacea, the latter being unrecorded in Stream 3 yet abundant at Station 3.

(c) Species distribution and filamentous algal and bryophytic growths.

The effect of increased fertilisation often results in the excessive growths of algae, bryophytes, and phanerogams when other conditions thought to be important, like flow rate, carbon and trace elements, are met also. Flowering plants like Ranunculus, Potamogeton, and Sparganium were common at Station 3 together with patches of Fontinalis antipyretica - this moss also being common at Station 4 and in Experimental Streams 1 and 2. The greatest plant cover was afforded by growth of the blanket weed Cladophora, and in particular at Station 4 where 80% cover was recorded during July and August. During summer this stretch of river is controlled by weir gates and frequently the water level is only 5 - 8 cm. deep, which likens it to the depth in Streams 1 and 2 where Cladophora also grows. The cover in Streams 1 and 2 during summer was about 70% in most parts of the riffle, which is in contrast to the small tufts that grow at Station 1 despite the possession of the same water chemistry as Stream 2.

Percival and Whitehead (1929) have shown the preference of many fauna for filamentous algae. Cladophora and mosses, in particular, collect large amounts of detritus also; and it is common to find large populations of Naididae, Tubificidae, and Chironomidae feeding on the trapped detritus. A number of the Chironomidae must feed upon the alga as well and, whilst it is difficult to tell which are doing so without a gut analysis, some species of the Orthocladiinae, which possess no pigmentation, are noticeably green under the microscope. Eukiefferiella (sp. C.1) is one of these and appears along with Micropsectra atrofasciatus at Station 4 where Cladophora cover is extensive (Fig. 5.14). Eukiefferiella (sp. C.1.) and Micropsectra (Fig. 5.18) are also prevalent in Streams 1 and 2 during the Cladophora season from May to October along with other species of Orthocladiinae; Cricotopus (Cricotopus) albeforceps? and Orthocladiinae (sp. C. 11) (Fig. 5.20). However whether they are strict herbivores is doubtful, for during taxonomic examination it was frequently observed that most guts contained particles of mud and sand and/or pieces of animal and plant debris despite the sometimes numerous diatoms in the Orthocladiinids.

The density of the herbivores B. rhodani and E. ignita in the R. Tean at Station 1 was greater than in the Experimental Stream 2 while Lymnaea and Ancylus were more in abundance in the latter after they managed to establish themselves (Fig. 5.15 & Table 5.13). It has already been suggested that the distribution of Lymnaea and also Ancylus are favoured by the artificial situation and, while the types of epiphytic and epilithic algae are probably the same in both the Tean (Station 1) and Stream 2, the density of some species in the Experimental Streams were greater owing to the asbestos channels used for algal investigations. Whereas B. rhodani does not appear to increase in density with the growing season of Cladophora at Station 4 (Table 5.17) the occurrence of Gammarus pulex does (Fig. 5.15). Percival and Whitehead (1929) suggest that G. pulex thrives best in loose moss; and just such a biotope occurs in Streams 1 and 2, but although numbers increase during the growing season so do the numbers of G. pulex through breeding.

The association of Asellus aquaticus and large growths of Cladophora with eutrophication has been mentioned in connection with the R. Tean at Station 4 (Fig. 5.11) and the R. Ray at Moredon. Given an ideal medium in which to live and reproduce, it was noticed that the majority of individuals had discarded their classical role of scavenging detritus for the new one of grazing filamentous algae. Populations using the alga as a support medium must be tolerant to the low oxygen levels encountered overnight during Cladophora respiration particularly when the blankets are thick, thus preventing efficient water exchange and subsequent oxygen replenishment. Also when decomposition and spates break up the mats in autumn, the mounds of algae strip the water of oxygen. Asellus is very tolerant to low oxygen levels and it was shown, using the apparatus described in the following chapter, that 11% of a population of 19 survived fluctuating diurnal oxygen concentrations of

12 hours 0.5 mgl^{-1} and 12 hours 6 mgl^{-1} over a period from 30.11.73 to 9.12.73. The conditions were particularly severe, especially with respect to the temporal nature and while they probably do not occur naturally in the R.Ray, it demonstrates the tolerance that the species exhibits.

An idea of the weight of Cladophora in the R. Ray during the peak of the growing season is given below with percentage covers for the remaining seasons. The weights and density of the Asellus population at the same Station are given although they are the mean of three samples, not all of which were taken in the Cladophora blankets.

Table showing mean dry weights and percentage cover of Cladophora, with mean dry weights and density of Asellus taken in the River Ray during the year May 1972 - April 1973.

Date	Cladophora g/m^2	Cladophora % cover	Asellus g/m^2	Asellus nos./m^2
17.5.72	* 38		11.56	10,058
17.6.72	* 218		25.64	10,866
19.7.72	* 210		6.16	7,085
14.8.72	Cladophora disintegrating		4.60	9,025
20.9.72		25	10.76	13,621
26.10.72		35	23.1	21,032
23.11.72		35	7.87	14,844
31.1.73		55	9.09	11,093
7.3.73		75	13.00	19,707
9.4.73		80	5.10	3,867
			$\bar{x}$ 11.69	$\bar{x}$ 12,119

* results of Urquhart (1972)
personal communication.

The graphs of mean dry weight, percentage occurrence of male, female, ovigerous and juvenile entities of the same Asellus population (Figs 8.1 - 8.3 Chapter 8) show that the weight of the population is a result of reproduction rather than the incidence of Cladophora, although the latter, whose growing season spans both broods, obviously aids in protecting the young from predators and supplies food and shelter. The greatest weights occur in June and October as a result of the heavier adult proportion, and the lowest when the numerous juveniles predominate.

By executing length/frequency analyses of the Asellus populations in the R. Ray and the Experimental Streams over the period of one year, it was intended to discover whether the differing amounts of Cladophora and hence fluctuating oxygen levels within the matrix of filaments would be sufficient to incur any changes in the percentage composition and production of the populations. However, delays occurred in the building of the Experimental Streams of nearly one year and, following completion, the colonisation of the streams by Asellus was protracted until the first spring brood established itself in July 1973. The remaining nine months for investigation was not sufficient time for indicative observations to be made especially while the population was steadily increasing in size as a result of its recent colonisation. Observations on the R. Ray population has therefore been included into a separate Chapter (8). Somewhat unrealistic annual mean figures of weight and density have been recorded in the Streams for the same reasons, but they do demonstrate the relative proportions between the Streams and show the mean weight and density in

Stream 1 approaching those for the R. Ray.

Table showing annual means of Asellus numbers and dry weight in the Experimental Streams and the River Ray.

	Dry Weight g/m^2	Number/m^2
Stream 1	10.27	7,116
" 2	2.40	1,190
" 3	4.25	1,696
River Ray	11.69	12,119

(d) Later colonisation, interspecific competition and the effects of siltation.

Those species which had the advantages of mobility, multivoltinism, or were brooding or ovipositing at the time the Experimental Streams became operational were obvious candidates for swift colonisation of the new Streams. Some of these have been mentioned at the beginning of the chapter where it was noted that B. rhodani, Simulium, and chironomid larvae were particularly adept.

Others, like Asellus, were present in very small numbers within a couple of months but could not increase in density until the time for reproduction. The seasonal graphs (Figs. 5.15, 5.16) show how from March onwards the numbers of Asellus escalate in all three Streams and Gammarus in Stream 2. Whereas the numbers of Asellus have continued to rise in Streams 1 and 3 over the whole period of investigation, the density in Stream 2 where it is in competition with Gammarus has fallen in accordance with the curtailment of the autumn brooding. It appears that in the other

Streams recruitment is still occurring in such large numbers that only extended reproduction or grossly unequal distribution within the riffle could account for it. The fluctuation in numbers of Gammarus due to reproduction is more normal, although the virtual return in January 1974 to the density of January 1973 is suggestive of the instability of the population and thought to be the result of increasing sedimentation with inert river solids, thus decreasing the area available for inhabitation. During the growing season Cladophora and mosses the inhabitable biotope is increased again. The Tean at Station 1 only supports a very low density population of Gammarus all the year round, a feature indicative of the predation by fish (Fig. 5.12).

Lymnaea pereger did not appear in the Streams until April 1973 (Fig. 5.15) and similarly the mollusc Ancylus fluviatilis was characteristically slow in colonising the new areas (Table 5.13). The success of the sensitive latter species in Stream 2 is evident while the more tolerant Lymnaea is very successful in both Streams 1 and 2, their being virtually indifferent to the increasing sedimentation.

The Tubificidae established themselves in an unexpected manner. The densities were expected to be highest in the two eutrophicated Streams but initially the numbers were highest in the 100% river water Stream (Fig. 5.17) and subsequently, after their disappearance from the Streams altogether from February to May 1973, their numbers remained highest in Stream 1 as opposed to the greater eutrophication of Stream 3 where densities were expected to be highest. The increasing siltation, with time, of Stream 3 was greater than in the other Streams and of what appeared to be of a different nature. While Streams 1 and 2 contained inert river solids and some detrital material, the bottom of Stream 3 by the winter of 1973 was smothered by an amalgam of clay particles and organic solids which gave a consistency similar to a paste. It was this material that was thought to be unsuitable and responsible for the reduction in numbers of many invertebrate species and also for the productivity of the Stream.

The Tubificidae exhibit an interesting relationship with their predators *Thienemannimyia lentiginosa* and *Procladius spp*. All in instars of the former and all but the very young first instars of the latter are carnivorous preying primarily on the oligochaetes. In the latter quarter of 1972 when the tubificid population was greatest in Stream 2, the density of *T. lentiginosa* was greatest in this Stream also, reaching a peak in February 1973 (Fig. 5.19). After March 1973 when the greatest density of tubificids was in Stream 1, the density of *T. lentiginosa* was also greatest in this Stream. Whether the virtual absence of tubificids between February and May 1973 accounts for the crash in *Procladius* numbers alone is uncertain. Emergence with the warmer weather must have reduced the numbers of larvae, but the continuing low density populations in all the Streams is seen as a result of interspecific competition and changing substratum. The graph (Fig. 5.19) also shows the preference of *Procladius* to eutrophicated conditions, an an example not shared by *Thienemannimyia.*

It was demonstrated in the R. Cole that the aerial advantage possessed by the Chironomidae led to swift recolonisation of the badly polluted areas subsequent to the abatement of the pollutant. This advantage is evident in the colonisation of the Streams, their occurrence being seasonal in most cases which is corroborated by reference to their seasons in the R. Tean. *Polypedilum arundineti*, *Cricotopus* (*Cricotopus*) *bicinctus*, *Eukiefferiella hospita*, *Micropsectra atrofasciatus*, *Brillia longifurca*, *Rheocricotopus*, *Eukiefferiella* (*sp. Cl*), and *Diamesa campestris* appear in the Streams (Figs. 5.18 - 20) according to their seasonal abundance which may be determined from their occurrence in the R. Tean (Figs. 5.13 and 14). *Dyscampocladius ex*. *gr*. *vitellinus* (Fig. 5.20) is only recorded once in the R. Tean, i.e. one individual at Station 1 in March 1973. In the Streams, however, they have excelled in density, first appearing in January in Stream 3 and reach a mean density of 337 per $0.05m^2$ by March 1973. It has also remained throughout most of the year and some aspect of the Streams artificiality or instability is considered responsible. The same factors are thought to be pertinent to the density and occurrence of *Metriocnemus atratulus*. *Cricotopus* (*Cricotopus*) *albeforceps?*, and *Cricotopus* (*Cricotopus*) *fuscus*.

The physiological diversity of the chironomid larvae has, then, allowed large populations of different species to thrive in the eutrophicated streams. This has been demonstrated in Streams 1 and 3 (Fig. 5.9) where they are present in equal abundance, both being greater by a factor of 2 than the density in Stream 2. Using the criterion of physiological diversity, the only reasons for the relative paucity of numbers in Stream 2 and Station 1 of the Tean can be interspecific competition and relative oligotrophy. The artificial conditions prevailing in the Experimental Streams, particularly with regard to laminar flow and the lack of fish predation all year round, are seen as reasons for the differences in chironomid abundance between the Streams and the Tean Stations. The extra input by the dairy of solids and a subsequently slightly silted substratum must be responsible for the maximum abundance occurring at Station 3; while solids are high at Station 2 also, the shifting nature of the substratum and lack of flora precludes such a heavy colonisation.

The colonisation of the Experimental Streams by all the Chironomidae has been graphed (Fig. 5.17) and shows the relative speed with which species and densities have accumulated in Stream 3, soon to be followed in Stream 1. In fact all the Streams were colonised quickly but the eutrophicated conditions of Streams 1 and 3 allowed seasonal colonisation in terms of numbers to occur rapidly - the greater the eutrophication the greater the density of individuals. The fact that the seasonal fluctuations have not reached the prior abundances of February and March 1973 (Stations 3 and 1 respectively) is a result of increased stability, inter- and intraspecific competition, and increased siltation, especially in Stream 3. The gradual decline in density over the whole period of investigation in Stream 2 is thought to be indicative of increasing diversity and stability of the whole benthic community, although the mean number of chironomid species in Stream 2 is seen as lowest of the three streams.

Mean number of species of Chironomid Larvae over the period 22.3.73. - 13.3.74 in the Experimental Streams.

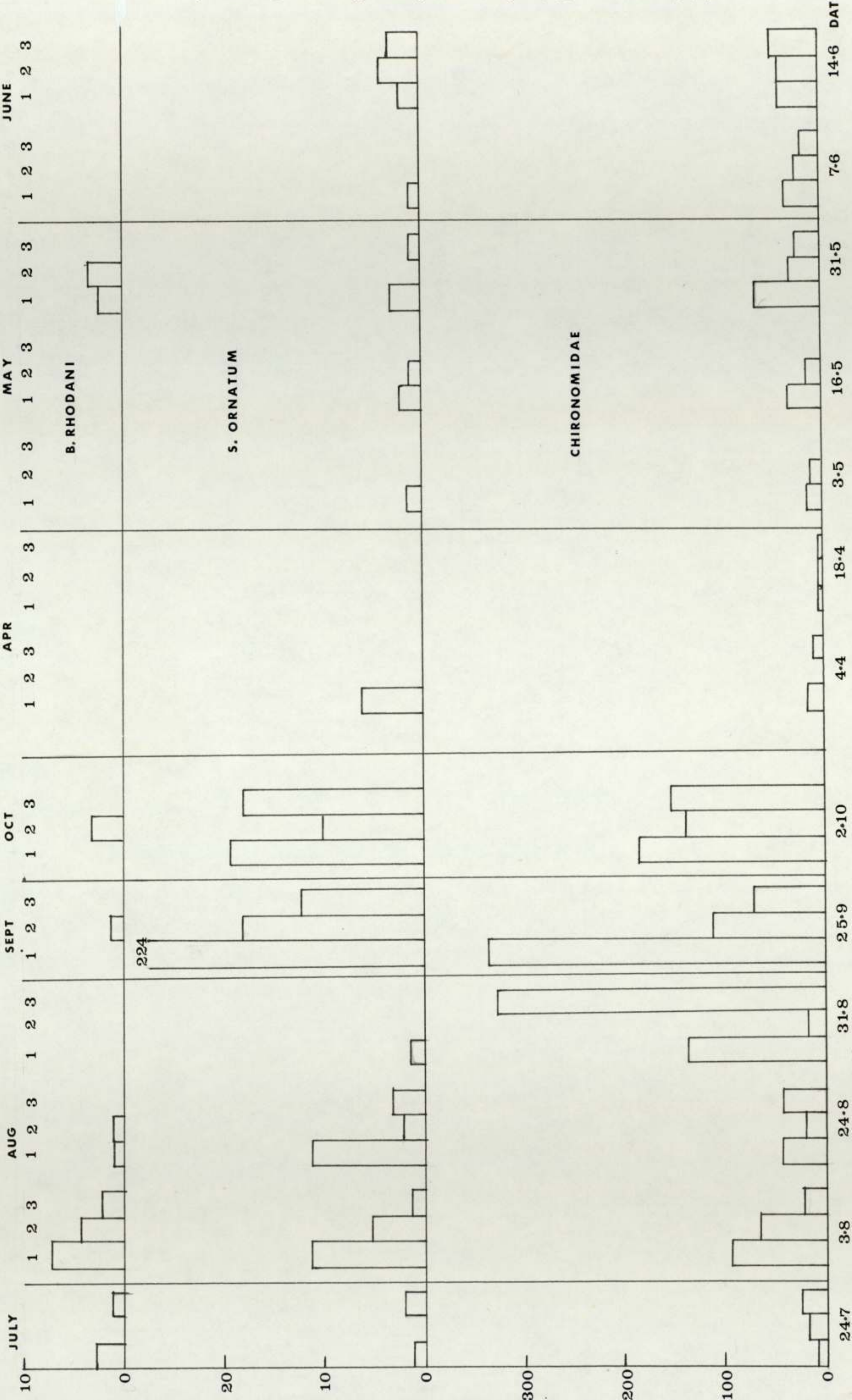
Stream (1)	Stream (2)	Stream (3)
9.75	7.0	9.0

(e) Emergence.

Fig. 5.21 shows the density of *Simulium ornatum*, *Baetis rhodani*, and Chironomidae imagines which were taken in the Experimental Streams during the emergence periods from July 1973 - June 1974. The dry weights of each catch together with the biomass of 'other' unidentified imagines appear in Table 5.23.

The Chironomidae appear to emerge all the year round except for the period November - March when they are overwintering as larvae. Very few adults were taken in November 1973 when trapping for this year was subsequently abandoned. It was resumed in March 1974 when small numbers appeared to be emerging. Most larvae emerged to oviposit between late August and early October and a similar pattern of emergence existed for the *Simulium* adults. The extended period of emergence from 3.8.73 - 2.10.73 of *S. ornatum* suggests that a number of generations were present and this is in accordance with its multivoltinism. *B. rhodani*

Fig.5.21. Number of adults trapped per $1.8x10^3cm^2$ in the Experimental Streams 1,2 and 3 during the year July 1973 - June 1974.



emerged in greatest numbers in August and this was the only occasion that they were taken from Stream 3 - following the high density population taken in the Stream (Fig. 5.15).

A number of chironomid adults have been seen floating downstream just before take off and therefore the numbers taken in the traps do not strictly reflect the numbers present as larvae beneath the traps, despite efforts to use a trap which is nearly touching the water surface. The number trapped is further complicated for the same reasons by the size of the populations inhabiting the asbestos channels used in algal growth experiments (Plate 2). Similarly, the adults of S. ornatum float downstream before take off aided by an air bubble in the pupal skin. If the distance they float is greater than the distance from Y to the Stream number (Plate 2) where the traps are situated, then erroneous estimates would be expected. It may be for reasons like these that erratic results bearing little relation to the standing crops shown by the substratum sampler arise and that there does not appear to be any evidence of differential emergence caused by the $1^{o}C$ increments between the Streams 2, 1, and 3, see tables below: -

Table showing the number of S. ornatum adults taken in the emergence trap ($0.18m^{2}$) and the number of larvae taken with the cylinder sampler from the substratum ($0.05m^{2}$) corrected to $0.18m^{2}$.

Stream	July		August		Sept.	(2.10.73) Sept/Oct.	Oct.
	trap	sampler	trap	sampler	trap	sampler	trap
2	0	94	3	187	18	198	10
1	1	76	8	360	224	2656	29
3	2	54	1	126	12	975	18

Table showing the number of chironomid adults taken in the emergence trap (0.18m^2) and the number taken in the cylinder sampler from the substratum (0.05m^2) corrected to 0.18m^2.

	July		August		Sept	Sept/Oct (2.10.73)	Oct.
Stream	trap	sampler	trap	sampler	trap	sampler	trap
2	14	453	33	457	110	144	136
1	5	378	87	306	335	313	185
3	22	622	126	273	67	324	152

Table showing the ratio of numbers of adult S.ornatum to larval numbers (0.18m^2)

Stream	July	August	Sept.	Oct.
2	-	1:62	1:11	1:20
1	1:76	1:45	1:11	1:91
3	1:27	1:126	1:81	1:54

Table showing the ratio of numbers of adult Chironomidae to larval numbers (0.18m^2)

Stream	July	August	Sept.	Oct.
2	1:32	1:14	1:1	1:1
1	1:75	1:4	1:1	1:2
3	1:28	1:2	1:5	1:2

The total dry weights (mg) per $0.18m^2$ for the various taxa trapped over the year July 1973 - June 1974 in the three Streams are tabulated below: -

	(1)	(2)	(3)
Chironomidae	77.9	47.1	51.7
S. ornatum	197.8	24.8	26.9
B. rhodani	12.1	20.8	3.0
Others	14.3	5.5	10.2
Totals	302.1	98.2	91.8

The individual and total weights of adults emerging from the Experimental Streams, as expected, reflects the patterns of mean standing crop of invertebrates for the respective Streams.

5.6 CONCLUSIONS AND SUGGESTIONS FOR FURTHER WORK.

1. The differing trophic structures in the three Experimental Streams and at the five Stations in the River Tean are the result of the relative degree of eutrophication under which the communities exist.

2. The importance of allochthonous material and relative subordinate role of autochthonous supply is realised in the standing biomass of invertebrates determined as a consequence of the magnitude of eutrophication.

3. The manifestation of eutrophication in the Experimental Streams and the River Tean is one of increased scavenging and filtering populations utilising allochthonous material, with concomitant reduction in the proportion of grazers and carnivorous populations.

4. Benthic macro-invertebrate communities living in eutrophicated conditions are subject to the density independent factors of toxicity, low oxygen, spates and fish predation. The communities in the Experimental Streams are only subject to toxicity and low oxygen as independent factors, intraspecific competition regulating the production to a greater extent.

5. The larger standing biomass in the Experimental Streams reflects the artificiality of the situation compared with the natural situation of the River Tean.

6. The productivity in terms of standing biomass is dependent upon the magnitude of eutrophication and does not necessarily increase with increasing fertilisation.

Experimental Stream 1 was twice as productive as Stream 2 and Stream 3 although the latter was richer in nutrients and allochthonous material. Examples from the River Tean corroborate the conclusion with the potential energy source being greatest just downstream of the outfall and diminishing further downstream.

7. A great proportion of the total standing biomass may be attributable to one or two components of the invertebrate community. *Simulium ornatum* forms greater than half the standing biomass in Streams 1 and 3, and in the River Tean at Station 2. *Asellus aquaticus* has an extensive role in production in Streams 1 and 3, and Stations 3 and 4 of the River Tean also. Such domination by a few species necessarily presents an ecologically unstable situation.

8. The stability of the benthic communities is determined by the degree of eutrophication. The ecological stability of Streams 1 and 2 is greater than that of Stream 3. Similar gradients of stability are apparent with levels of fertilisation in the River Tean.

9. The standing biomass in the Streams, and in particular Stream 3, was slowly reduced as a result of siltation - another facet of the artificiality of the Stream systems.

10. The distribution of a number of species is the result of a multiplicity of factors arising from eutrophic conditions. Physical and chemical factors restrict the range of *B. rhodani*, *E. ignita*, *E. venosus*, *G. pulex*, *A. fluviatilis*, and *R. dorsalis*.

The increased microbial and allochthonous input, both directly from sewage effluent and indirectly from breakdown of primary producers, favours the populations of *S. ornatum*, *A. aquaticus*, *Tubificidae* and Chironomidae. A number of species from the latter family are especially able to take advantage of these conditions: *Brillia longifurca*, *Prodiamesa olivacea*, *Eukiefferiella hospita*, *Chironomus riparius*, *Cricotopus* (*Cricotopus*) *fuscus*, *Cricotopus* (*Cricotopus*) *bicinctus*, *Rheocricotopus sp*. and *Dyscampocladius vitellinus*.

The associated growths of bryophytes and filamentous, epiphytic and epilithic algae with increased fertilisation provide protection and food favouring a number of species: *E. ignita*, *G. pulex*, *L. pereger*, *A. fluviatilis*, *Micropsectra atrofasciatus* and *Eukiefferiella sp.C.1*. *A. aquaticus* has been seen to change its trophic status within the confines of the *Cladophora* blankets.

11. Observations on the short term colonisation of the Experimental Streams have shown similar results to those of other workers. Longer term studies have shown colonisation to be a function of the life history of the species and include such factors as those displayed in Chapter 4, section 4.3.(1).

12. The number of chironomid larvae increases with eutrophication. This is thought to be a result of the ecological imbalance caused by this form of pollution.

13. The amount of drift from the Streams alone, as determined by the 24 hr. sample, is negligible and may be related to the constant flow of water over the riffles and possibly the assumed low level of production - there being

no predation by fish and plenty of food amounting to less reason for dispersal.

14. It is suggested that the rates of production are greater in the River Tean despite the larger standing biomass in the Experimental Streams. Useful further work would include the introduction of suitable fish to the Streams while monitoring the monthly standing biomass. It would not only prove useful as a pilot scheme for fish culture, production and farming in eutrophicated rivers, but clarify the position of relative secondary production with regard to the Allen paradox.

As the reproductive capacities of ecosystems are not constant from season to season and year to year, further research into the seasonal biomass of the Experimental Streams' benthic invertebrate communities would be useful as the relatively new populations begin to oscillate around inter and intraspecific equilibria.

15. The significance of stream research on this scale has managed to liberate ecological investigation from some of those problems associated with laboratory designs as outlined in Section 5.1. paragraphs 2 - 3, and, while there are no pretensions about the problems still involved, a transcendence of many conceptual dangers inherent with laboratory ecosystems and their interpretation to the natural situation is thought to have been achieved.

TABLE 5.1.

Monthly means of physical and chemical analyses of Stream 1 (mgl^{-1}).
Means and standard errors are for the year January - December, 1973.

	pH	^oC	S.S.	D.O.	% Sat.	Cl^-	Alk.	NH_3	O.N.	T.I.P.	P.V.	HDNS	DET.	B.O.D.
Aug. 72	7.9	13.9	20.1	10.0	99.9	48.0	143	0.4	11.0	3.1				
Sept.	7.7	11.5	6.4	10.3	93.6	56.2	146	1.0	11.4	3.3				
Oct.	7.8	10.5	9.4	10.7	95.6	54.7	154	0.5	12.5	3.6				
Nov.	7.5	8.5	19.2	10.9	92.6	51.4	126	1.1	11.6	2.8	3.2			
Dec.	7.5	7.3	14.6	10.3	85.9	45	129	2.2	9.9	2.1	2.3			
Jan. 73	7.3	6.6	11.4	10.8	87.5	51	156	4.8	11.2	3.1	2.2			
Feb.	7.5	6.3	11.5	10.9	89.3	52	145	5.7	7.0	2.3	1.7		0.02	
Mar.	7.4	8.3	18.2	10.9	93.6	52	169	7.0	6.2	2.2	1.9		0.02	
Apr.	7.6	10.4	20.2	11.0	97.8	49	172	5.7	8.3	1.9	2.2			
May	7.5	13.0	22.1	9.6	90.8	49	131	1.8	11.9	3.8	2.6			
June	7.5	15.1	12.5	9.9	98.8	48	140	2.2	12.9	3.6	2.3			5.4
July	7.6	15.8	24.5	9.2	93.4	41	125	1.2	-	2.2	2.6	254		
Aug.	7.7	15.5	10.1	9.7	97.4	49	140	1.1	11.2	3.8	1.4	276		
Sept.	7.6	13.3	10.8	8.8	83.2	50	126	0.9	-	3.8	1.9	259		
Oct.	7.4	10.8	-	9.3	80.7	50	122	0.7	11.4	3.2	2.2	228		
Nov.	7.5	7.1	14.2	9.2	75.8	51	116	1.2	9.5	3.5	2.1	213		
Dec.	7.5	6.6	16.9	8.8	76.1	56	123	2.2	6.6	2.4	2.1	213	0.2	
Jan. 74	7.4	6.5	21.1	9.1	75.0	49	129	2.9	8.8	2.6	1.5	214	0.01	
Feb.	7.4	6.7	21.5	9.4	79.4	54	131	1.6	8.7	3.3	1.6	209	0.04	
Mean	7.5	10.7	14.9	9.8	88.7	49.8	139	2.9	9.5	3.0	2.1	237		
S.E.	0.03	1.0	1.8	0.2	2.4	1.0	5.4	0.7	0.7	0.2	0.09	9.8		

TABLE 5.2.

Physical and chemical analyses of Stream 2. (mgl^{-1}).

Means and standard errors are for the year January - December 1973.

	pH	°C	S.S.	D.O.	% Sat	Cl^-	Alk	NH_3	O.N.	T.I.P.	P.V.	HDNS	DET	BOD.
Aug. 72	8.2	13.0	11.4	11.0	107.3	35	160	0.1	3.9	0.2				
Sept	8.0	10.2	5.0	11.0	97.0	38.4	158	0.3	3.3	0.4				
Oct.	8.0	9.6	5.9	11.5	99.9	35.9	164	0.2	3.6	0.4				
Nov.	7.8	7.6	19.2	11.3	94.7	37.2	126	0.5	4.7	0.5	2.2			
Dec.	7.8	6.6	14.0	11.3	91.3	34.5	124	0.5	4.3	0.3	1.4			
Jan. 73	7.8	5.7	7.2	11.9	94.5	37	131	0.6	5.3	0.3	1.1			
Feb.	7.8	5.5	8.5	12.8	101.1	36	123	0.7	4.4	0.4	1.4		ngl	
Mar.	8.1	7.6	13.8	13.4	114.8	36	133	0.8	3.5	0.4	1.0		ngl	
Apr.	8.2	10.1	7.4	13.0	116.4	33	144	0.7	4.0	0.4	0.7			
May	5.0	12.5	12.9	11.6	109.2	36	142	0.6	2.9	0.5	1.3			
June	8.1	14.7	8.1	11.8	116.3	33	154	0.6	3.4	0.2	1.0			2.4
July	8.0	14.9	26.2	10.6	105.5	33	134	0.6	-	0.7	2.1	240		
Aug	8.1	14.5	7.3	10.7	105.2	34	145	0.4	3.8	0.5	0.9	276		
Sept.	8.0	12.2	8.8	10.6	95.6	38	137	0.4	-	1.0	1.2	255		
Oct.	7.9	9.2	-	10.6	92.5	35	126	0.3	4.9	0.1	1.3	222		
Nov.	7.8	6.1	19.4	11.3	90.3	36	119	0.6	3.8	0.5	1.7	212		
Dec.	7.8	5.8	21.3	10.7	87.7	44	115	0.9	4.3	ngl	1.7	206	0.001	
Jan. 74	7.7	5.8	17.3	10.8	86.7	39	118	1.3	4.0	0.7	1.1	216	0.001	
Feb.	7.8	6.5	20.6	11.0	90.6	42	123	0.8	4.2	0.8	1.1	199	0.001	
Mean	7.9	9.9	13.2	11.6	102.4	35.9	134	0.6	4.0	0.4	1.3	232		
S.E.	0.04	1.08	1.87	0.3	3.0	0.89	3.4	0.00	0.2	0.00	0.1	9.7		

TABLE 5.3

Physical and chemical analyses of Stream 3. (mgl^{-1})
(monthly means)
Means and standard errors are for the year Jan - Dec. 1973.

	pH	°C	S.S.	D.O.	% Sat.	Cl^-	Alk	NH_3	O.N.	T.I.P.	P.V.	HDNS	DET	BOD
Aug. 72	7.8	15.3	9.9	8.6	88.6	70.0	134	1.0	21.9	6.9				
Sept.	7.4	12.9	8.5	9.2	86.7	71.7	137	0.6	20.3	7.1				
Oct.	7.6	11.8	13.3	9.3	85.9	77.7	130.7	0.7	22.6	7.1				
Nov.	7.3	9.7	15.2	9.4	82.9	68	121	1.6	20.4	5.7	4.5			
Dec.	7.4	8.4	11.3	5.9	75.8	58	132	3.5	18.7	4.8	3.5			
Jan 73	7.4	7.8	16.5	9.2	76.8	73	191	9.5	16.3	5.3	3.9			
Feb.	7.5	7.4	21.7	9.2	76.5	72	186	9.9	10.0	4.2	3.5		0.04	
Mar.	7.6	9.3	25.2	9.2	85.9	70	209	13.5	9.0	3.8	3.2		0.04	
Apr.	7.6	10.8	30.3	9.3	84.0	69	196	11.6	13.7	3.6	4.0			
May	7.4	13.7	22.1	9.1	88.0	64	126	2.1	19.6	6.6	3.9			
June	7.4	16.2	16.7	9.2	92.6	66	124	3.3	21.2	6.8	4.1			6.0
July	7.5	16.7	16.6	8.8	91.1	59	118	1.4	-	2.6	3.7	264		
Aug.	7.6	16.8	11.9	8.9	92.5	66	137	2.4	18.1	6.7	3.4	295		
Sept.	7.4	13.6	12.1	8.3	82.4	64	113	1.3	-	6.4	3.0	285		
Oct.	7.3	11.9	-	7.7	71.0	66	119	0.9	17.4	6.4	2.9	244		
Nov.	7.4	8.2	11.7	8.3	70.3	63	116	2.1	14.9	6.0	2.8	238		
Dec.	7.3	7.6	13.2	8.0	67.0	72	134	4.1	11.2	5.4	2.8	233	0.55	
Jan. 74	7.3	7.5	15.7	8.6	72.6	64	149	4.9	9.1	6.4	2.7	253	0.025	
Feb.	7.2	8.0	15.1	8.3	70.1	76	151	3.4	15.8	6.2	2.4	233	0.05	
Mean $\bar{X}$	7.45	11.7	17.8	8.76	81.5	67	147	5.2	14.6	5.3	3.4	259		
S.E.	0.00	1.06	1.7	0.17	2.6	1.2	10.6	1.3	1.3	0.4	0.1	8.9		

TABLE 5.4

Monthly means of physical and chemical analyses of the R. Tean water abstracted for the purposes of mixing with effluent in Streams 1 and 3. (mgl^{-1})

Means and standard errors are for the year Jan. 73 - Dec. 73.

	pH	°C.	S.S.	D.O.	% Sat.	Cl^-	Alk.	NH_3	O.N.	T.I.P.	P.V.	HDNS	DET	BOD
Aug.72	8.0	12.3	11.5	8.7	94.1	36	164	0.07	3.8	0.2				
Sept.	7.7	9.9	6.2	10.0	88.2	37	162	0.3	3.5	0.5				
Oct.	7.9	9.4	10.6	9.8	84.8	36	162	0.2	3.8	0.5				
Nov.	7.7	7.6	22.0	10.6	87.9	37	130	0.5	4.8	0.5	2.4			
Dec.	7.7	6.6	17.2	10.9	88.1	35	123	0.8	4.2	0.3	0.8			
Jan. 73	7.8	5.7	10.3	11.0	87.5	38	133	0.7	5.3	0.3	1.3			
Feb. 73	7.9	5.4	16.7	11.2	88.4	39	114	0.5	4.3	0.4	1.5		ngl	
Mar.	8.1	6.7	16.5	11.6	94.9	36	132	0.8	3.1	0.5	0.9		ngl	
Apr.	8.2	9.5	5.3	12.8	112.6	33	145	0.7	4.2	0.4	0.9			
May	7.9	11.8	11.4	11.7	109.2	37	142	1.8	3.0	0.5	1.4			
June	7.9	13.7	6.9	9.6	92.6	33	154	0.3	3.5	0.2	0.9			2.2
July	7.8	14.1	38.9	9.2	88.9	34	136	0.6	-	0.4	2.0	239		
Aug.	7.9	13.5	5.9	9.5	90.7	34	149	0.4	3.9	0.4	0.8	263		
Sept.	7.8	11.7	8.0	8.3	84.2	38	135	0.4	-	1.2	1.2	251		
Oct.	7.7	9.4	-	9.6	83.2	35	126	0.3	4.9	0.02	0.9	229		
Nov.	7.7	6.1	15.9	10.4	82.9	37	119	0.7	3.5	0.5	1.3	215		
Dec.	7.7	5.9	14.8	10.5	82.7	42	115	1.0	3.7	ngl.	1.6	199	.001	
Jan. 74	7.6	6.2	18.0	10.6	85.1	38	119	1.3	3.9	0.7	1.2	218	.001	
Feb.	7.8	5.8	27.4	10.9	87.5	42	129	0.7	4.4	0.7	1.1	198	0.001	
Mean	7.9	9.5	14.1	10.5	91.4	36	133	0.68	3.9	0.4	1.2	231		
S.E.	0.1	0.98	2.6	0.3	2.87	0.29	3.8	0.1	0.2	0.00	0.1	8.4		

TABLE 5.5.

Physical and chemical analysis of the effluent used for the production of mixtures in Streams 1 and 3 (mgl^{-1}). (Monthly means)

Means and standard errors are for the year Jan 1973 - Dec. 1973.

Date of Sample	pH	^{o}C.	S.S.	D.O.	% Sat.	Cl^-	Alk.	NH_3	O.N.	T.I.P.	P.V.	HDNS	DET	BOD.
Aug. 72	7.6	15.5	16.4	7.5	77.6	78	131	0.03	28.1	8.8				
Sept.	7.1	13.5	7.9	7.6	73.3	86	131	0.9	27.8	9.2				
Oct.	7.4	12.5	15.8	8.3	77.4	87	125	1.0	29.5	8.7				
Nov.	7.2	10.6	16.3	8.3	74.7	78	118	1.8	25.3	6.9	5.8			
Dec.	7.3	9.3	10.8	7.5	64.9	68	136	4.4	23.4	6.6	4.2			
Jan. 73	7.3	8.6	21.4	7.4	52.8	83	205	12.1	23.4	5.9	5.3			
Feb.	7.6	7.8	21.7	7.2	59.6	82	207	12.6	12.0	5.3	4.0		0.06	
Mar.	7.6	9.5	19.8	7.0	61.3	78	222	13.9	10.7	4.8	3.9		0.07	
April	7.5	10.5	26.3	7.0	62.9	78	211	12.3	14.7	4.5	4.8			
May	7.3	13.2	21.6	6.7	64.1	70	122	3.2	24.4	8.0	5.1			
June	7.0	15.6	19.6	6.7	66.9	76	114	3.9	30.3	8.1	5.3			9.0
July	7.2	16.4	14.3	7.2	73.2	68	113	2.9	-	6.0	3.8	278		
Aug.	7.4	16.6	12.9	7.0	72.2	69	139	2.5	21.9	8.2	3.9	295		
Sept.	7.2	14.8	15.4	6.3	63.4	73	116	1.4	-	7.8	3.7	291		
Oct.	7.1	12.7	-	6.6	62.3	73	116	1.1	22.2	8.1	3.2	251		
Nov.	7.3	9.4	12.3	7.3	64.0	71	117	2.2	14.8	7.0	3.5	237		
Dec.	7.1	8.8	14.9	7.1	63.3	81	139	5.3	12.8	7.1	3.4	257		
Jan. 74	7.2	8.1	12.8	6.9	58.0	70	157	5.9	19.7	7.9	3.1	266	0.089	
Feb.	7.1	8.4	14.5	6.9	58.0	78	154	3.6	18.9	6.8	2.8	246	0.06	
$\bar{X}$	7.3	11.99	17.8	6.95	63.8	75	152	6.1	18.8	6.8	4.15	268		
S.E.	0.00	0.94	1.31	0.1	1.56	1.53	12.9	1.44	1.88	0.41	0.22	8.1		

TABLE 5.6

Physical and chemical data for Station 1 of the R.Tean.

Date	pH.	°C	S.S	D.O	% Sat	Cl^-	Alk	NH_3	ON	TIP	PV	BOD.
1972												
21/11	7.7	6.5	18.0	10.8	86.7	37	100	0.4	4.0			
20/12	7.1	6.0	1.0	10.8	86.7	34	150	1.2	4.7	0.2		
1973												
19/1	7.2	5.5	6.0	11.5	89.8	35	135	0.4	4.9	0.2		2.6
8/2	8.2	8.0	11.5	-	-	35	140	6.0		2.3		
15/2	7.6	2.5	15.0	11.9	88.4		125				0.7	
22/2	7.9	6.0	4.5	11.0	88.4	37	115			0.4		
26/2	7.0	5.0	5.0	11.6	90.8	30	120	0.3	5.6	0.3		1.5
8/3	7.8	5.5	44.0	12.0	96.3	32	115				0.6	
15/3	7.9	6.3	24.0	12.0	96.3	33	140	5.9	5.3	0.5	0.4	
22/3	7.9	6.0	6.0	12.4	99.6	32	155	1.0	6.6	0.4	0.5	
29/3	7.8	8.0	7.0	12.9	108.9	28	135	1.1	5.3	0.5		1.2
19/4	8.0	7.8	9.0	12.0	101.4	34	145	0.5	2.3	0.5	0.2	
26/9	8.0	9.0	2.5	13.6	117.7	32	145	1.0	7.8	0.5		
3/5	8.1	7.5	6.5	11.5	97.1	32	145	2.2	2.3	-		
17/5	8.5	11.0	8.0	15.2	137.9	32	155	negl	4.4	0.4	-	
25/5	7.6	11.5	11.5	11.2	103.9	46	135	negl	2.9	0.6	1.6	
31/5	8.0	11.3	11.5	9.1	82.6	30	135	0.3	2.8	0.6	3.2	
7/6	7.9	13.5	5.0	9.7	94.3	32	160	0.6	3.5	0.6	1.1	
14/6	7.8	12.0	4.0	10.2	94.7	33	135	0.4	3.5	2.0		
21/6	7.7	14.0	6.5	9.3	90.3	36	150				1.9	
28/6	7.8	14.0	6.0	9.1	88.4	34	150				1.0	
5/7	8.0	15.3	8.5	9.2	91.3	31	165					
12/7	7.7	14.0	3.5	-	-	33	160					
19/7	7.7	13.0	27.5	8.6	81.7	33	110	2.0	2.3	0.42	2.6	
26/7	7.9	13.3	6.5	10.3	97.8	33	140	0.4	3.8	0.3	0.4	
1/8	8.0	14.5	7.5	9.1	90.4	35	155	1.0	3.1	0.2	-	
9/8	7.7	13.0	7.5	9.3	88.3	30	125	0.6	3.4	0.3	1.0	
23/8	7.8	13.0	4.5	10.3	97.8	36	155	1.7	8.7	2.1	0.4	
30/8	7.8	12.5	5.5	9.3	88.3	33	155	1.2	2.3	0.3		
7/9	8.1	14.5	5.0	9.2	91.4	35	160	0.7	3.5	0.2		
20/9	8.0	11.5	-	8.6	79.8	31	125	0.7	3.6			
27/9	7.6	-	-	10.0		39	150	0.8	2.8	0.9		
4/10	7.9	-	-	9.5	-	38	160		4.2	negl	0.3	
11/10	7.7	9.0		6.8	58.9	27	85	4.1	4.5	negl	0.2	
18/10	7.8	6.0		9.6	77.1	34	130	.7				
25/10	7.7	7.8		10.4	87.8	34	120	1.4	4.0	negl	negl	
1/11	7.8	9.0		9.8	84.8	35	140	1.1	3.4	negl	0.1	
8/11	7.5	9.0		9.3	80.5	61	115	1.3			0.6	
15/11	7.5	7.3	84.0	9.4		27	85	3.7			0.6	

TABLE 5.7

Physical and chemical data for Station 2 of the R. Tean.

Date	pH	$^{\circ}C$	S.S	D.O	% Sat.	Cl^-	Alk	NH_3	ON	TIP	PV	BOD
1972												
Nov.												
21	7.6	6.5	16.5	10.2	81.9	41	120	0.1	7.0			
Dec.												
20	7.1	7.0	17.0	10.0	82.4	45	160	0.4	9.8	2.1		
1973												
Jan.												
19	7.05	5.5	29.0	9.7	75.7	54	160	1.9	12.5	3.2		13.5
Feb.												
8	7.6	7.5	17.0	-	-	56	175	5.3		2.3		
15	7.3	3.5	27.0	10.2	77.8		140	1.9	5.2	1.6	1.7	
22	7.6	6.5	14.0	10.3	84.9	55	150	5.2	5.5	2.0		
26	6.8	6.0	7.0	10.6	85.1	46	165	6.5	6.3	1.8		5.4
Mar.												
8	7.5	6.5	45.0	10.4	85.7	47	165		6.1		2.0	
15	7.6	7.3	24.0	10.5	86.6	48	175	7.4	7.4	2.1	1.3	
22	7.6	8.3	8.0	9.9	83.6	50	200	11.3	6.3	3.4	1.7	
29	7.2	10.0	12.0	-	-	48	190	8.2	4.6	1.9		5.4
Apr.												
19	7.5	9.0	17.5	8.3	71.9	56	185	7.4	8.9	2.4	2.2	
26	7.6	10.0	8.5	10.6	93.9	52	185	6.3	6.4	2.4		
May												
3	7.7	7.5	20.0	8.9	75.2	51	170	10.8	10.1	3.5		
17	7.9	11.8	14.5	11.0	102.1	53	155	2.0	11.7	3.1	1.3	
25	7.1	13.0	19.0	8.3	78.8	54	145	0.8	10.2	3.1		
31	7.8	12.0	15.0	7.4	68.7	43	135	1.1	8.7	2.6		
June												
7	7.5	15.5	10.0	7.6	77.2	55	200	1.2	10.3	3.5		
14	7.3	13.3	7.0	9.1	86.4	61	170	1.1	10.8	3.9		
21	7.3	14.8	16.5	7.0	69.5	55	160				3.6	
28	7.4	15.0	10.0	8.0	79.4	43	130				1.6	
July												
5	7.7	16.3	8.0	9.1	92.4	46	165					
12	7.5	15.0	11.0	-	-	62	150					
19	7.4	14.3	37.0	7.5	72.9	36	105	1.4	6.4	1.5	4.2	
26	7.6	14.5	10.5	8.5	84.9	52	130	0.7	10.5	3.6	3.7	
Aug.												
1	7.5	16.5	10.5	8.0	82.9	65	145	0.5	10.5	5.1	-	
9	7.7	14.5	10.0	8.2	81.4	44	125	0.7	7.7	2.1	2.0	
23	7.7	15.0	14.5	9.0	89.4	68	200	10.0	7.9	5.0	1.6	
30	7.6	14.0	12.0	8.6	83.6	47	140	1.5	9.7	-	1.6	
Sept.												
7	7.5	16.0	7.5	8.3	84.2	59	135	0.8	11.2	4.9		
20	7.4	13.5	-	8.7	84.5	53	120	1.0	10.1	3.1		
27	7.7	-	-	9.9	-	39	165	1.0	12.8	5.9	0.7	
Oct.												
4	7.6	-	-	8.3	-	62	140		11.0	5.2	0.3	
11	7.7	9.5		7.6	67.3	30	90	3.8	4.6	0.6	0.3	
18	7.6	8.0		8.6	72.6	55	125	2.1				
25	7.3	9.0		9.9	85.7	46	115	2.0	8.7	negl	negl	
Nov.												
1	7.5	10.5		7.6	68.9	58	125	2.1	10	3.7	0.3	
8	7.6	10.0		7.4	65.6	55	125	1.7			0.6	
15	7.4	8.0	70.0	9.1		32	85	3.2				

TABLE 5.8

Physical and chemical data for Station 3 of the R. Tean.

Date	pH	°C	S.S	D.O	% Sat.	Cl⁻	Alk	NH_3	ON	TIP	PV	BOD
1972												
Nov.												
11	7.6	6.5	14.0	9.9	79.5	45	130	0.1	7.3			
Dec.												
20	7.2	7.5	7.0	9.6	79.1	45	165	0.6	6.9	1.8		
1973												
Jan.												
19	7.05	5.0	17.0	9.4	73.4	53	170	2.1	11.3	2.9		16.5
Feb.												
8	7.7	8.0	20.0	-	-	53	185	2.6		1.5		
15	7.6	4.0	27.0	10.7	81.6		145	2.1	5.3	1.8	1.9	
22	7.8	7.3	19.5	9.8	80.8	54	155	4.9	5.4	2.0		
26	6.9	6.0	9.0	9.8	78.7	42	185	5.7	6.1	1.9	2.1	3.6
Mar.												
8	7.7	7.3	58.0	9.8	80.8	46	160		6.4		1.8	
15	7.9	8.5	58.0	8.6	74.5	47	175	8.3	3.7	1.3	2.3	
22	7.9	8.5	11.0	9.7	83.9	47	200	-	-	1.3	1.4	
29	7.3	10.5	11.0	9.9	87.8	45	205	7.1	-	1.7		6.9
Apr.												
19	8.3	9.5	19.0	9.4	83.3	44	185	3.6	1.8	1.5	1.8	
26	7.8	11.0	15.0	10.0	90.7	45	185	4.5	7.8	1.9		
May												
3	7.7	9.0	18.0	8.8	76.2	47	175	4.4	9.1	2.6		
17	8.0	12.5	21.0	9.7	92.1	54	160	2.0	11.7	3.5	1.0	
25	7.2	13.5	20.5	8.1	78.7	51	145	1.9	9.3	2.8		
31	7.9	12.5	26.0	7.6	72.2	42	145	0.9	8.4	2.6		
June												
7	7.5	16.0	17.5	7.8	79.2	50	170	0.9	10.0	3.1		
14	7.4	14.5	12.0	8.5	84.4	55	140	1.1	11.4	4.2		
21	7.4	16.0	12.5	6.7	68.0	51	145				2.8	
28	7.5	15.5	18.5	6.8	69.0	38	160				2.9	
July												
5	8.0	16.8	13.5	7.7	79.8	41	185					
12	7.6	15.5	18.0	-	-	49	165					
19	7.4	14.5	26.5	7.3	72.5	35	120	1.7	6.9	1.78	3.3	
26	7.6	14.5	19.0	8.6	85.4	45	150	0.5	7.5	2.3	3.2	
Aug.												
1	7.9	16.5	12.5	8.0	82.9	51	175	0.5	7.3	3.5	-	
9	7.7	14.3	14.5	8.4	81.6	43	145	0.6	6.1	1.5	1.3	
23	7.7	15.0	13.5	8.7	86.4	46	200	3.9	4.8	2.0	1.2	
30	7.6	13.8	9.5	8.1	78.7	40	180	1.1	6.3	2.6	2.4	
Sept												
7	7.8	15.8	13.5	8.0	81.2	-	160	0.7	8.0	2.4		
20	7.5	14.0	-	7.2	69.9	50	135	1.0	8.9	2.5		
27	7.0	-	-	6.1	-	67	125	0.8	6.7	1.8	1.8	
Oct.												
4	7.4	-	-	6.7	-	56	165		7.2	3.3	negl	
11	7.7	9.8		7.8	69.1	30	90	-	5.9	0.6	0.1	
18	7.7	8.3		8.4	70.9	46	150	1.5				
25	7.4	9.0		7.7	66.6	45	135	2.2	7.9	1.2		
Nov.												
1	7.7	10.0		8.0	70.9	48	165	1.9	7.8	1.3	negl	
8	7.6	10.3		8.4	74.4	73	135	2.1			0.5	
15	7.5	8.0	78.0	9.1		30	100	3.6				

TABLE 5.9

Physical and chemical data for Station 4 of the R. Tean.

Date	pH	°C	S.S	D.O	% Sat.	Cl^-	Alk.	NH_3	ON	TIP	PV	BOD
1972												
Nov.												
21	7.7	6.5	22.5	10.2	81.9	43	120	nl	4.2			
Dec.												
20	7.4	7.0	8.0	10.0	82.4	40	185	0.4	3.1	0.8		
1973												
Jan.												
19	7.2	4.5	12.0	9.8	74.6	52	185	1.3	7.3	-		8.1
Feb.												
8	8.1	8.0	18.5	-	-	45	190	2.6		-		
15	7.8	3.5	28.0	11.1	84.6		150	1.3	4.9	0.8	1.6	
22	8.0	6.3	19.0	10.0	80.3	64	160	2.5	5.4	1.1		
26	7.1	5.5	10.0	10.5	84.3	122	165	2.1	4.1	0.7		3.9
Mar.												
8	8.1	6.5	53.0	10.2	84.0	49	160		6.6		1.7	
15	8.3	7.3	46.0	9.7	79.9	40	195	2.2	6.8	1.2	0.9	
22	8.2	6.8	8.0	10.3	84.9	42	210	4.2	6.6	1.6	0.9	
29	7.6	10.0	10.0	9.7	86.0	87	185	3.2	-	1.4		3.9
Apr.												
19	7.9	8.5	8.5	9.0	77.9	47	190	4.1	7.6	1.8	1.3	
26	7.9	10.0	6.0	9.8	86.8	45	190	4.1	7.7	1.4		
May												
3	7.7	8.3	9.5	6.7	56.6	48	180	2.8	9.1	2.3		
17	8.1	12.0	12.0	10.5	97.5	48	175	0.8	10.0	2.0	1.2	
25	7.5	13.0	15.5	8.6	81.7	44	160	0.9	7.1	2.1		
31	7.9	12.0	14.0	7.0	64.9	36	180	0.7	4.7	1.5		
June												
7	7.8	15.0	12.5	7.9	78.5	49	210	0.8	9.6	2.9		
14	7.7	13.3	12.5	8.7	82.6	48	195	0.7	10.2	2.9		
21	7.7	15.0	8.0	8.1	80.4	46	150				2.6	
28	7.6	15.5	11.0	7.1	72.1	40	170				2.1	
July												
5	8.0	16.5	9.5	7.9	81.9	43	190					
12	7.7	15.3	7.0	-	-	54	175					
19	7.6	14.0	35.0	7.1	68.9	60	145	0.5	7.1	1.5	3.6	
26	7.6	14.0	15.0	8.1	78.7	45	165	0.6	7.7	2.1	2.2	
Aug.												
1	8.0	16.3	17.0	7.8	79.2	53	180	0.5	5.9	2.5	-	
9	7.9	14.0	11.0	9.0	87.5	135	150	0.5	5.3	1.1	1.7	
23	7.6	14.5	18.0	7.1	70.5	53	185	1.5	9.3	2.9	0.8	
30	7.6	14.0	10.0	5.4	52.5	51	195	2.8	9.4	4.8	3.0	
Sept.												
7	7.8	16.0	10.5	7.6	77.2	48	175	0.9	9.1	3.1		
20	7.8	14.0	-	7.3	70.9	47	145	1.0	8.7	1.9		
27	7.4	-	-	7.6	-	48	175	0.9	8.5	2.9	1.4	
Oct.												
4	7.7	-	-	7.6	-	54	170	[illegible]	10	3.2	0.1	
11	7.7	9.5		7.1	62.9	29	90	5.0	5.6	0.5	0.3	
18	7.7	7.0		8.4	69.2	49	160	1.1				
25	7.7	8.3		9.4	79.4	115	150	2.2	7.2	0.7	0.1	
Nov.												
1	7.7	9.5		8.5	75.4	47	170	1.6	8.2	1.5	0.2	
8	7.7	9.5		9.0	79.8	42	150	2.1			0.6	
15	7.6	7.5	65.0	9.7		30	100	3.2				

TABLE 5.10

Physical and chemical data for Station 5 of the R. Tean.

Date	pH	^{o}C	S.S	D.O	% Sat.	Cl^-	Alk	NH_3	ON	TIP	PV	BOD
1972												
Nov.												
21	7.8	6.5	22.5	10.4	83.5	57	135	nl	4.2			
Dec.												
12	7.5	6.0	13.0	10.6	93.9	40	185	0.3	8.5	1.3		
Jan.												
19	7.3	3.5	12.0	10.4	77.1	44	190	2.8	8.6	1.9		1.9
Feb.												
8	8.1	8.0	22.5	-	-	66	190	-		-		
15	7.9	2.5	34.0	11.0	81.7		145	2.8	5.3	0.9	1.6	
22	8.1	6.3	22.0	10.9	83.5	59	160	2.5	5.5	0.9		
26	7.2	5.0	18.0	10.7	83.8	122	165	2.1	4.5	0.9		3.9
Mar.												
8	8.2	6.3	56.0	10.7	85.9	48	160		7.2		1.8	
15	8.3	7.0	38.0	10.5	86.6	44	190	3.0	6.1	1.3	1.1	
22	8.2	7.3	8.0	11.4	93.9	45	200	3.1	5.3	-	1.2	
29	7.8	10.0	8.0	11.1	98.4	45	180	2.3	4.4	1.3		3.0
Apr.												
19	7.1	8.5	11.5	9.5	82.3	44	190	3.0	8.4	1.6	1.2	
26	8.2	10.5	11.0	11.5	104.4	45	185	2.2	8.0	1.8		
May												
3	7.8	8.5	11.0	8.4	72.7	50	185	1.8	9.4	2.0		
17	8.7	12.5	10.5	12.0	113.9	47	170	0.9	10.3	2.3	0.8	
25	7.5	13.8	16.5	9.7	94.3	46	170	1.3	7.2	2.1		
31	8.2	12.0	20.0	8.4	77.9	54	180	0.9	6.6	1.6		
June												
7	8.1	16.3	14.0	8.7	88.3	51	190	0.6	9.6	2.7		
14	7.9	14.5	11.0	9.1	90.4	46	180	-	10.0	3.1		
21	7.8	16.0	12.5	8.5	86.3	45	150				2.7	
28	7.9	16.5	12.5	8.7	90.2	61	165				1.9	
July												
5	8.5	18.0	12.0	11.1	117.3	51	195					
12	8.1	16.5	6.5	-	-	58	180					
19	7.6	14.3	25.0	7.6	73.9	98	140	1.4	6.0	1.45	2.1	
26	7.7	15.0	20.5	8.4	83.4	70	160	0.6	8.6	2.1	2.5	
Aug.												
1	8.2	17.5	19.5	88.3	87.7	57	175	negl	7.5	2.1	-	
9	8.1	14.3	21.5	8.8	85.5	57	150	negl	6.1	1.1	2.1	
23	7.9	15.0	14.5	10.1	100.3	52	185	0.6	8.8	2.7	1.0	
30	8.1	14.5	12.5	8.5	84.4	45	205	0.8	7.9	3.5	2.5	
Sept.												
7	8.1	16.5	15.0	8.6	89.1	50	175	0.8	8.8	3.5		
20	7.9	14.5	-	7.7	76.5	48	130	0.8	9.4	1.6		
27	7.4	-	-	6.3	-	49	180	0.8	9.1	3.7	1.5	
Oct.												
4	7.6	-		7.5	-	53	180		8.9	3.1	0.1	
11	7.7	9.5		6.3	55.8	40	85	7.0	5.6	0.5	0.2	
18	7.8	7.0		7.6	62.6	51	155	1.3				
25	7.8	8.5		9.9	85.7	56	145	2.0	7.7	0.6	0.1	
Nov.												
1	7.7	9.5		7.3	64.7	56	175	2.3	8.0	1.8	0.1	
8	7.6	10.0		8.4	74.4	46	150	2.0			0.4	
15	7.7	7.5	113.0	9.2		40	100	4.4				

TABLE 5.11

Physical and Chemical data from Moredon Bridge - R. Ray. Mean = year 9.2.72. - 31.1.73.

Date	ON	TIP	NH_3	pH	S.S.	^{o}C	D.O.	% Sat.	Cl^-	Alk.	P.V.	B.O.D.	HDNS
9.2.72.	9.9	1.8	3.0	7.2	39	8	8.0	67.4	104	300	7.4	11.1	
15.3.72.	7.5	2.7	1.5	7.2	27	11	7.9	71.8	69	290	7.6	12.6	440
19.4.72	8.8	2.9	3.2	7.2	19	11	8.9	80.9	82	255	6.0	6.0	360
17.5.72	10.0	3.7	1.4	7.3	35	13	-	-	90	295	5.6	16.5	270
17.6.72	10.8	5.6	2.7	7.2	9	18	9.9	103.7	91	280	6.6	5.1	306
19.7.72	10.8	-	0.1	7.3	7	17.5	4.9	51.4	104	280	5.8	5.4	330
14.8.72	15.6	6.4	3.2	7.2	31	19	8	85.6	77	295			
20.9.72	16.2	5.7	1.8	7.2	11	15.5	9.5	98.3	121	275			
26.10.72	3.3	6.1	2.6	7.2	31	15	9.5	94.3	106	225			
23.11.72	9.1	4.16	1.9	7.2	69	10	7.2	63.6	91	225			
31.1.73	11.6	3.5	2.1	7.3	25	7	7.6	62.7	72	260			
7.3.73	12.5	2.4	0.5	7.2	6.3	8.5	9.3	80.5	89	250			
9.4.73	12.5	3.0	3.1	7.3	13	7	10.7	88.2	79	235			
MEAN	10.3	4.2	2.1	7.2	25.3	12.0	7.4	70.9	84	248	6.5	9.5	341

TABLE 5.12.

Mean monthly dry wt. per 0.05 m^2. in Experimental Streams 1, 2 & 3 (mg.)
Annual mean values = 22.3.73. - 17.3.74.

	G. complanata			*A. aquaticus*			*G. pulex*			*B. rhodani*		
	1	2	3	1	2	3	1	2	3	1	2	3
1973												
Jan. 16	2.1			3.8				15.5		1.8	17.8	2.2
Feb. 19				20.1		88.4	11.6	15.1	64.2	27.8	44.0	4.6
Mar. 22				46.2	5.0	39.2		16.6		13.6	30.4	
Apr. 30				51.6		32.6		66.0	36.3	7.2	32.6	
May 30				116.6	18.1	12.5		205.4		13.3	17.8	2.5
June 25				26.1	8.7	7.4		205.4		13.1	185.4	0
July 22				424.2	94.9	52.2	6.5	684.0		6.3	6.6	36.2
Aug. 31				243.5	0	47.6		123.9	10.0			
Oct. 2				273.9	117.2	30.3		50.7			38.4	
Nov. 12				1094.4	293.2	155.4		275.8			2.1	
Dec. 11				686.0	242.5	99.0		142.2				
1974												
Jan. 9				1993.2	97.4	413.1		56.6			1.8	
Feb. 11				390.0	760.7	306.5		36.3				
Mar. 13				818.6	452.0	1356.1	23.6	32.5				
Mean				513.7	119.9	212.6	2.5	158.0	3.8	4.6	26.3	3.2

continued ...

TABLE 5.12

(continued)

	Diptera (ephydridae)			Chironomidae			*E. ignita*			*E. venosus.*		
	1	2	3	1	2	3	1	2	3	1	2	3
1973												
Jan. 16	2.1		1.8	17.3	32.9	47.6						
Feb. 19	4.8		4.8	61.7	67.8	137.6						
Mar. 22	6.0	4.2		120.8	72.0	331.8						
Apr. 30				73.6	51.2	32.9						
May 30				128.4	64.8	20.9		2.8			19.0	
June 25				36.1	25.4	36.7		93.3				
July 22		7.2		31.7	46.9	43.4	7.1	21.5			15.9	
Aug. 31		1.2		26.7	23.7	21.0						
Oct. 2	3.3	2.9		17.1	9.0	42.6						
Nov. 12	3.3			32.7	1.6	17.3						
Dec. 11					3.0	21.8						
1974												
Jan. 9				45.0	4.3	48.9						
Feb. 11		0.9		79.5	9.1	51.8						
Mar. 13	3.8			69.0	20.9	16.8						
Mean	1.4	1.4	0	55.8	27.6	57.1	0.6	9.8	0	0	2.9	0

continued ...

TABLE 5.12

(continued)

	R. dorsalis			*H.angustipennis*			*S. ornatum*			Tipulidae		
	1	2	3	1	2	3	1	2	3	1	2	3
1973												
Jan. 16	1.8	18.8			9.2		7.8				91.1	
Feb. 19	7.6	13.6	5.6				5.5		3.6		42.5	
Mar. 22		22.8										
Apr. 30	27.1	37.5										
May 30		85.0					354.7	2.5	4.6			
June 25	24.4	113.3					573.0	11.3	319.1		41.7	
July 22	25.0	115.1	48.0				12.6	8.1	4.9	75.6	213.7	46.1
Aug. 31		40.7					51.2	12.1	12.4		23.9	
Oct. 2		5.3					1177.2	25.5	298.9		51.3	
Nov. 12		20.8					4103.6	5.2	2681.5	335.	72.6	72.0
Dec. 11		51.4					2413.1	5.9	1636.2	113.7	194.4	19.0
1974												
Jan. 9		12.1			3.8		317.7	3.5	454.2	416.2	91.5	71.7
Feb. 11	3.8	44.4					314.4	10.9	461.7	46.1	126.8	42.1
Mar. 13		23.9			21.6		296.0	–	76.3	23.9	81.0	93.1
Mean	6.7	47.7	4.0	0	2.1	0	801.0	7.0	495.8	76.6	75.2	28.7

continued ...

TABLE 5.12
(continued)

	Sialis			Dicranota			Tubificidae			L. pereger		
	1	2	3	1	2	3	1	2	3	1	2	3
1973												
May 30		10.5			6.							
June 25					6.8							
July 22							9.8			214.8	535.5	77.7
Aug. 31		14.1					20.1	3.8	2.5	608.8	221.7	125.8
Oct. 2			14.1				16.2	9.5	0	89.4	941.8	78.2
Nov. 12			52.2				27.9	6.6	0	762.9	759.2	245.1
Dec. 11		17.0					8.9	0	10.4	97.7	802.7	408.0
1974												
Jan. 9							15.0	1.7	5.4	1198.8	1098.7	0
Feb. 11					5.7		0	4.6	0	137.1	234.2	73.1
Mar. 13				10.9	6.8		106.5	46.9	36.3	1291.5	1799.2	24.3
Mean		3.5	5.5	0.9	2.4	0	17.0	6.1	4.6	366.7	532.7	86.0

TABLE 5.12

(continued)

	E.testacea			A.fluviatile			E.octoculata			Coleoptera larvae			Lumbriculus		
	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3
1974															
Aug. 31	13.6			80.2	6.2										
Oct. 2				29.4	47.6			9.9							
Nov. 12					98.7						9.7				
Dec. 11				11.6	157.5		24.5	24.6						29.3	
1974															
Jan. 9					50.2			33.0					36.9		75.9
Feb. 11					102.0									14.8	
Mar. 13					231.0			88.0	18.7						
Mean	1.1			10.1	57.8		2.0	12.9	1.6		0.8		3.1	3.7	6.3

TABLE 5.13

Mean monthly numbers of invertebrates per $0.05m^2$ in the Experimental Streams 1, 2 and 3.

Annual mean values = 22.3.73 - 13.3.74.

	B. rhodani			*G. pulex*			*S. ornatum*			*A. aquaticus*		
Date	1	2	3	1	2	3	1	2	3	1	2	3
1972												
Oct. 10	3	64	0		2	1	235	673	71	0	0	0
Nov. 10	27	141	0		3	0	339	32	255	1	5	1
Dec. 9	8	44	0		12		328	12	88	8	2	0
1973												
Jan. 16	20	128	0	2	6		98	0	0	2	0	0
Feb. 19	35	65	2	2	5	4	11	0	8	12	1	2
Mar. 22	30	32	0	2	2	0	0	0	0	12	2	24
Apr. 30	2	12	0	2	12	4	0	0	0	15	2	6
May 30	25	15	3		58		639	6	8	30	5	3
June 25	17	138	1		91		795	32	430	14	9	11
July 22	5	15	54	5	189	0	21	26	15	328	75	45
Aug. 31	1	0	0		83	2	100	52	35	185	43	21
Oct. 2	3	21	0		34		1,377	59	507	189	101	33
Nov. 12	0	54	0		48		18,690	41	12,213	66	141	129
Dec. 11	0	2	0		42		11,700	36	5,721	467	136	40
1974												
Jan. 9	3	1	0		11		1,713	12	4,200	1,245	104	195
Feb. 11	0	0	0		9		5,181	18	1,737	441	0	390
Mar. 13	0	0	0	3	7	0	1,284	1	48	1,278	96	120
Mean	7.2	24.1	4.8	1.0	48.8	0.5	3,458	23.5	2,076	355.8	59.5	84.8

continued ...

TABLE 5.13

(continued)

Date	Tubificidae			*L. pereger*			Tipulidae			*A. fluviatile*		
	1	2	3	1	2	3	1	2	3	1	2	3
1972												
Oct. 10	0	25	0									
Nov. 10	15	40	75				4	3	1			
Dec. 9								2	2			
1973												
Jan. 16	6	6					2					
Feb. 19		few						1				
Mar. 22											2	
Apr. 30												
May 30				3							1	
June 25	15	15	18	0	1		1	3				
July 22	18	0	3	15	72	3	3	15	3	3		
Aug. 31	40	14	3	24	27	6		3		4	3	
Oct. 2	48	27	9	3	65	3		2		9	8	
Nov. 12	54	23	0	27	36	6	9	5	3	3	22	
Dec. 11	18	0	21	2	32	5	6	13	1	3	35	
1974												
Jan. 9	24	6	9	30	27	0	9	9	3	3	10	
Feb. 11	45	23	21	9	17	6	3	6	3	0	19	
Mar. 13	100	19	15	39	39	3	0	7	1	8	31	0
Mean	30.2	10.6	8.3	12.7	26.3	2.7	2.6	5.3	1.2	2.8	10.9	-

continued ...

TABLE 5.13

(continued)

	R.dorsalis			Chironomidae			Ephydra			Coleoptera Larvae			Dicranota		
Date	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3
1972															
Oct. 10	9	17	0	218	111	38									
Nov. 10	3	4	0	116	205	78									
Dec. 9	2	10	0	186	184	88									
1973															
Jan. 16	2	8	6	78	120	96									
Feb. 19	1	7	2	82	130	290									
Mar. 22	0	3		216	232	1056	4	0	4						
Apr. 30	1	2		132	88	78	0	2	2	0	1	1			
May 30	0	9		489	171	52									
June 25	1	19		379	134	200					1		0	12	0
July 22	3	11	5	105	126	173	3	12	3		3			3	
Aug. 31	0	4		85	127	76	1	0	0		2				
Oct. 2	0	2		87	40	90	6	2	3						
Nov. 12	0	5		188	12	216	6				5				
Dec. 11	0	8		48	23	25									
1974															
Jan. 9	0	2		249	13	91	3								
Feb. 11	3	12	0	226	15	140	6								
Mar. 13	0	5p.	0	92	26	29	6	1	0						
Mean	0.7	6.8	0.4	191	84	186	2.9	1.4	1.0	0	1.2	0	0	1.3	0

continued ...

TABLE 5.13

(continued)

Date	Sialis			E. testacea			E. octoculata			H. angustipennis			E. venosus		
1973	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3
Mar. 22														2	
Apr. 30															
May 30	0	2	0					1						1	
June 25		1													
July 22														3	
Aug. 31	0	1	0	2	0	0									
Oct. 2	0	1	2					1							
Nov. 12	0	0	1												
Dec. 11		2					1	3							
1974															
Jan. 9								5							
Feb. 11															
Mar. 13								3			1				
Mean	0	0.6	0.3	0.2				1.1						0.5	

1973	E. ignita			Anabolia sp.			P. felina			Plectrocnemia sp.			Psychomyiidae		
	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3
May 30		4													
June 25		36													
July 22	12	9	0												
Aug. 31															
Oct. 2															
Nov. 12					1			1							
Dec. 11		1													
1974															
Jan. 9		1									1			1	
Feb. 11															
Mar. 13															
Mean	1.0	4.3	0												

TABLE 5.14

Mean monthly numbers of chironomid larvae per 0.05 m^2 in Experimental Streams 1, 2 and 3.

Annual mean values = 22.3.73 - 13.3.74.

	M.atrofasciatus			E.hospita			Procladius sp.			C. riparius			T. lentiginosa		
Date	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3
1972															
Oct. 10	130	89	16	40	5	5	10	4	7	0	0	7	0	0	0
Nov. 10	65	159	30	13	2	0	19	28	26	3	4	10	1	2	4
Dec. 9	134	192	10	2	0	2	20	8	46	0	2	22	8	20	2
1973															
Jan. 16	64	52	22	6	0	0	16	32	34	0	2	34	2	4	2
Feb. 19	46	62	10	2	1	0	23	34	18	1	0	18	2	14	2
Mar. 22	4	50	0	4	2	24	44	24	30	2	0	0	14	18	0
Apr. 30	1	0	0	0	0	0	2	6	0	0	0	0	47	56	24
May 30	9	0	3	60	0	1	6	9	2	6	0	1	6	9	1
June 25	144	41	54	42	1	30	2	0	1	8	0	19	0	0	0
July 22	5	26	0	3	0	9	6	8	6	0	0	0	20	24	22
Aug. 31	1	10	0	8	0	0	4	5	5	1	0	0	6	10	10
Oct. 2	39	14	3	6	7	14	0	4	6	0	0	2	3	0	0
Nov. 12	72	0	24	30	3	60	3	0	9	0	0	24	30	5	18
Dec. 11	17	0	6	0	0	2	1	0	1	2	0	15	26	22	0
1974															
Jan. 9	156	4	15	3	1	12	3	1	6	0	0	21	66	4	24
Feb. 11	189	4	36	15	0	42	0	1	0	0	0	9	16	7	39
Mar. 13	8	3	0	33	1	0	0	0	0	0	0	1	27	11	6
Mean	54	13	12	17	1.3	16	6	5	5	1.6	0	7.7	22	14	12

continued ...

TABLE 5.14
(continued)

Date	P. arundineti			Eukiefferiella sp. c.l.			C.albeforceps			C.bicinctus			D.campestris		
	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3
1972															
Oct. 10	0	2	0	0	0	0	5	0	1						
Nov. 10	1	2	0			0	1	0	2				1	0	0
Dec. 9	0	0	0	8	10	0	2	2	0						
1973															
Jan. 16	0	0	0	4	0	0							2	4	0
Feb. 19	1	0	0	1	2	0							2	5	0
Mar. 22	0	0	0				54	12	0	2	94	234	2	0	0
Apr. 30	15	4	4				9	6	0	28	0	4			
May 30	12	5	0				0	3	0	48	0	0			
June 25	2	0	0	13	6	3	0	0	15	66	0	3			
July 22	9	11	3	3	3	0	0	19	0	29	9	52			
Aug. 31	3	1	3	2	3	0				43	54	3	1	0	0
Oct. 2	0	1	0	6	2	3				21	5	9	6		
Nov. 12	0	0	9	15	1	6	1	0	0	3	0	3		1	
Dec. 11														1	
										0	0	3		2	1
1974															
Jan. 9															
Feb. 11	1	2	0												
Mar. 13										7	0	0	1	1	0
Mean	3.5	2.0	1.6	3.3	1.3	1.0	5.3	3.3	1.3	.5	12.8	25.9	0.8	0.4	0.08

continued ...

TABLE 5.14
(continued)

Date	*P.olivacea* 1	2	3	*M.atratulus* 1	2	3	*Rheocricotopus* sp. 1	2	3	*D. vitellinus* 1	2	3	*B. longifurca* 1	2	3
1972															
Oct. 10				0	1	0									
Nov. 10	1	0	0												
Dec. 9	0	4	2				2	2	0						
1973															
Jan. 16	2	2	2												
Feb. 19							2	2	0	0	0	240			
Mar. 22				2	0	12	0	28	0	68	0	756			
Apr. 30				0	0	6	1	4	0	0	0	15	0	0	2
May 30				0	2	0				36	6	28	0	1	0
June 25				0	0	4				0	0	31	8	0	1
July 22										5	0	20	3	0	3
Aug. 31							5	20	3			6	0	0	2
Oct. 2				0	0	2							0	1	0
Nov. 12				9	0	9	9	1	3			3			
Dec. 11															
1974.															
Jan. 9	3	0	0	3	0	3	9	0	0						9
Feb. 11				2	1	3							1	0	15
Mar. 13	4	5	0	0	3	4	3	2	2			1	6	0	0
Mean	0.6	0.4	0	1.3	0.5	3.6	2.3	4.6	0.7	9.1	0.5	71.6	1.5	0.2	2.7

continued ....

TABLE 5.14

(continued)

Date	Orthocladiinae sp. c.10			Orthocladiinae sp. c. 11			B. modesta			C. fuscus			C. (isocladius) sp.		
	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3
1973															
Mar. 22										6	0	0			
Apr. 30				12	0	12							0	4	0
May 30				306	135	16									
June 25	4	0	18	22	38	0							0	1	0
July 22			6				0	0		0	0	15			
Aug. 31		1		0	11	8				6	0	25			
Oct. 2										0	0	30			
Nov. 12							0	0	6	1	0	6			
Dec. 11							2	0	1						
1974															
Jan. 9	3	0	0					1					3	0	6
Feb. 11							1	0	0				1		
Mar. 13												15			
Mean	0.6	0.08	2.0	28.3	15.3	3.0	0.3	0.08	0.6	1.0	0	7.5	0.3	0.4	0.5

continued ...

TABLE 5.14

(continued)

Da

Date	*Lymnophyes*			*C. trifascia*			*Eukiefferiella* s. C.2.			*Chironominae* sp. C.2.		
	1	2	3	1	2	3	1	2	3	1	2	3
1972												
Oct. 10			1									
Nov. 10			6	0	2	0						
Dec. 9	2	0	2							2	0	0
1973												
Jan. 16												
Feb. 19												
Mar. 22												
Apr. 30												
May 30												
June 25				34	21	3						
July 22												
Aug. 31												
Oct. 2	0	2	18									
Nov. 12	6	0	36									
Dec. 11												
1974												
Jan. 9												
Feb. 11												
Mar. 13												
Mean	0.6	0.2	5.3	2.8	1.9	0.3						

TABLE 5.15

Mean monthly dry wts. per 0.05m^2 (mg) of invertebrates at Station 1 - 5 of the River Tean.

Date	Tubificidae					*E. octoculata*					*E. testacea*				
1972	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
Dec. 20	37.0	157.5	45.9		44.5				63.1	54.6		93.5	41.8		
1973															
Jan. 19	20.4	15.9	52.9		54.3				220.6			276.8	82.0		19.2
Feb. 26	114.2	9.2	22.5		167.0	11.4			93.0	93.3					
Mar. 29					70.5				88.3	40.5					
Apr. 30		22.4	19.3		69.0				62.5	33.7					
May 28	22.7		8.9		45.3				463.6	57.1		27.3	42.8		
July 25			78.3		30.9				177.9	116.1			209.4		
Sept. 4	23.7	15.3	22.2	22.8	54.1				90.0	67.8			181.5		
Oct. 4	20.3	39.6	24.6	5.3	11.9			192.6	142.8	65.1					
Nov. 7	21.8	3.1	8.0		35.1				85.8	82.4			77.4		
Mean	26.0	26.3	28.3	2.8	58.3	1.1	0	19.3	148.7	61.1	0	39.8	63.5	0	1.9
S.E.	10.5	15.1	7.6		13.3				38.6	10.3			24.2		

continued ...

TABLE 5.15

(continued)

Date	*G. complanata*					*A. aquaticus*					*G. pulex*				
1972	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
Dec. 20	88.1		26.0				2.5	412.5	151.4	3.0	12.8		9.4	3.7	15.3
1973 Jan. 19		9.4					4.6	304.9	468.4	12.9	79.5		8.6	2.2	15.9
Feb. 26							111.8	769.2	316.7		13.4				14.1
Mar. 29								334.5	259.8	5.0	19.7				13.1
Apr. 30							65.0	387.3	198.5	175.5	83.4		26.7	152.2	33.1
May 28	6.2		12.2		4.2		9.1	303.7	302.7	20.3	33.6			145.3	16.5
July 25							23.9	510.0	476.4	13.5	9.1			39.0	61.8
Sept. 4			16.5				9.9	626.7	468.0	20.3	44.1			84.0	13.7
Oct. 4						1.9	75.2	648.6	570.3		89.9			8.6	29.3
Nov. 7							176.4	978.3	167.9	6.0	42.2				25.3
Mean	9.4	0.9	5.5	0	0.4	0.2	47.6	527.5	338.0	25.7	42.8	0	4.5	43.5	23.8
S.E.							18.6	71.2	46.9	16.8	9.8			19.4	4.7

continued ...

TABLE 5.15

(continued)

	B. rhodani					R. dorsalis					H. angustipennis				
Date 1972	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
Dec. 20	22.1	1.7	12.1	2.0	2.7	31.9									
1973															
Jan. 19	27.3	8.5	26.4	10.4	20.5	31.3									33.6
Feb. 26	6.6		127.5		5.4	108.6	4.8								14.3
Mar. 29	36.4		16.8	13.8	6.3	22.7	38.8								26.6
Apr. 30	6.2	32.0	37.9	18.1	19.3	48.7				28.3					
May 28	12.0	2.9	3.8			41.2	108.5								
July 25	0.8	14.2		5.4	13.3	36.5	3.3			7.0					
Sept. 4	9.0		22.2		2.6	81.3									5.2
Oct. 4	22.1		7.2	6.2	47.9	122.0				40.2			5.7		44.3
Nov. 7	64.5	4.0		3.1		76.2	6.2								15.5
Mean	20.7	6.3	25.4	5.9	11.8	60.0	16.2	0	0	7.6	0	0	0.6	0	13.9
S.E.	5.9	3.2	11.9	1.9	4.7	11.0									

continued ...

TABLE 5.15

(continued)

Date	*S. ornatum*					Chironomidae					*A. fluviatilis*				
1972	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
Dec. 20	0	17.8	127.6	16.5	2.0						73.9				
1973															
Jan. 19	1	2102.0	112.5	136.6	21.5		6.0	36.1	29.6	9.5	44.2			44.2	
Feb. 26		2333.8	196.8	269.4	14.6		5.9	64.5	17.5	2.2	29.8				
Mar. 29		3330.8	211.9	234.5	78.6	1.6	23.4	64.2	21.2	6.4	59.2				24.3
Apr. 30	2.0		4.2			5.7	3.9	32.0	10.2	5.3					
May 28	1.6	86.8	0.4	6.6		22.0	50.4	40.8	21.3	8.1	25.6			30.0	
July 25	13.7	139.9	148.2	45.3	62.2			2.3	49.1	0.4					
Sept. 4	78.6	270.3	317.1		8.0	9.1	1.2	23.1	2.4		63.0				
Oct. 4	18.8	2214.9	408.9	2.0	5.6	1.1	6.0	19.8	0.8	0.3	48.2			9.7	
Nov. 7		143.5	79.5	1.6		1.2	5.8	12.1	2.3	0.5	70.0		22.4		20.0
Mean	11.6	1063.8	160.7	71.3	19.3	4.6	11.4	32.8	17.1	3.6	41.4	0	2.2	8.4	4.4
S.E.	7.7	403.6	40.9	32.9	8.9	2.4	5.3	7.2	5.2	1.2					

continued ...

TABLE 5.15

(continued)

Date	*L. pereger*					Other Diptera					Limnephilidae				
1972	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
Dec. 20			162.9			4.2									
1973															
Jan. 19						4.7					16.3				
Feb. 26										0.7					
Mar. 29										1.3					
Apr. 30															
May 28															
July 25			438.0												
Sept. 4			1208.1	48.5											
Oct. 4		31.5	766.2			4.1									
Nov. 7			32.0	206.4											
Mean	0	6.4	278.2	4.9	0	1.3	0	0	0	0.2	1.6	0	0	0	0
S.E.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

continued ...

TABLE 5.15

(continued)

Date			H. jenkinsi					Lumbriculus					E. ignita		
1972	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
Dec. 20								38.8			4.1				
1973															
Jan. 19							52.5								
Feb. 26															
Mar. 29															
Apr. 30															
May 28											8.4				
July 25											19.9				
Sept. 4															5.1
Oct. 4			175.5			11.4	4.5								
Nov. 7										48.6					
Mean	0	0	17.6	0	0	1.1	5.3	3.9	0	4.9	3.7	0	0	0	0.5
S.E.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

			Atherix					Tipula			
1973											
Sept. 9					40.8						
Oct. 4						14.5					
Mean					4.1	1.5					
S.E.					0	0					

TABLE 5.16

Mean monthly numbers of chironomid larvae per $0.05m^2$ at Stations 1 - 5 of the River Tean.

Date	*C. riparius*					*E. hospita*					*M. atrofasciatus*				
1972	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
Nov. 21	1	3	29			2	8	3		1					
Dec. 20			2					3	1						
1973															
Jan. 19			4				2	9	22	1		1	4	68	6
Feb. 26							26	20	28			8	3	24	6
Mar. 29							174	120	57	6			3	9	
Apr. 30						1		1	8				1		
May 28						23	12		55	4			6	59	3
July 25									1				4	1	3
Sept. 4							3	9					3	12	
Oct. 4			3			2	11	63	2		4	1	12	5	
Nov. 7			3			2	44	76	19	2		3			
Mean	0.09	0.3	3.6	0	0	2.7	25.4	27.7	17.5	1.3	0.4	1.4	3.3	16.2	1.6

continued ...

TABLE 5.16

(continued)

	T. lentiginosa					Eukiefferiella sp. C.l.					P. olivacea				
Date	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
1972 Nov. 21											4	4	20		
Dec. 20			2			1	1	13	29		2		13	1	
1973 Jan. 19						2	25	38	164	67		2	15		
Feb. 26							6	6	52	5		2	40	2	
Mar. 29	1		3				3	7	4	1			3		
Apr. 30	4	3	2	9	1							1	4		
May 28	4			1				12	9	4			12		
July 25				3				2	3	2				26	
Sept. 4	24					3							6		
Oct. 4	1					13	3		1	3					
Nov. 7	1					8	9		7	2		1	1		
Mean	3.2	0.3	0.6	1.2	0.09	2.5	4.2	7.0	24.4	7.6	0.5	0.9	10.3	2.9	0

continued...

TABLE 5.16

(continued)

	P. arundiaeti					*B. longifurca*					*C. sylvestris*				
Date	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
1972															
Nov. 21								6					2		
Dec. 20	2							22	2						
1973															
Jan. 19								15					3		
Feb. 26		2	2				6	12					3	4	
Mar. 29		6	2		8		3	31	4					2	
Apr. 30		5	4		11			3			8				
May 28	1	12	12		3										
July 25	1		17	6	3				1						
Sept. 4		3	3		8			18	3				84		
Oct. 4					6			18							1
Nov. 7					9			8							
Mean	0.4	2.5	3.8	0.5	4.4	0	0.8	12.0	0.9	0	0.7	0	8.4	0.5	0.09

continued ...

TABLE 5.16

(continued)

	Rheocricotopus sp.					C. bicinctus					D. campestris				
Date	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
1972 Nov. 21															1
Dec. 20			2												
1973 Jan. 19			3									3			3
Feb. 26			3									10	2		3
Mar. 29	2	6	73	10		1	24	130			1		2		3
Apr. 30			5	4				49	17		1				
May 28	1			9				12	16	2					2
July 25									3				2		2
Sept. 4		3	6			27	3	27						3	
Oct. 4						6		258					3		
Nov. 7	1	1		1		1		13			1				
Mean	0.4	0.9	8.4	2.2	0	3.2	2.5	44.5	3.3	0.2	0.3	1.2	0.8	0.3	1.3

continued ...

TABLE 5.16

(continued)

	Orthocladiinae sp. C.11					C. (isocladius) sp.					B. modesta				
Date	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
1972															
Nov. 21															
Dec. 20															
1973															
Jan. 19									6				1	2	
Feb. 26									4	2			2		
Mar. 29				58	32					1			6	2	
Apr. 30			2	10	8	1	4							1	
May 28	121	102	48	10	24										
July 25															
Sept. 4															
Oct. 4															
Nov. 7										1	1	1	4		
Mean	11.0	9.3	4.6	7.1	5.8	0.09	0.4	0	0.9	0.4	0.09	0.09	1.4	0.5	0

TABLE 5.17

Mean monthly numbers of invertebrates per 0.05m^2 at Stations 1 - 5 of the River Tean.

	Tubificidae					A. aquaticus					G. pulex				
Date	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
1972															
Nov. 21	189	202	71	16	48	4	17	383	1739	3	73	1		4	4
Dec. 20	330	323	195	40	150		6	353	292	4	8		4	5	1
1973															
Jan. 19	55	61	123	40	246		7	211	692	10	19	3	4	4	4
Feb. 26	70	21	45	150	775		87	271	255	6	4	4	2		6
Mar. 29	90		20		187		8	162	205		5		2		4
Apr. 30		56	40	150	200	2	36	295	192	3	19	2	4	27	5
May 28	33	30	66	20	78		12	417	174	1	15			28	9
July 25	45	6	294	24	82		21	360	432	18	13			48	26
Sept. 4	93	21	33	64			6	249	481	17	30			20	17
Oct. 4	90	93	24	5	20	1	66	693	443	3	46			3	17
Nov. 7	62	7	35	25	100	1	104	1059	258	4	19	1		1	9
Mean	96.0	74.5	86.0	48.5	171.4	0.7	33	404	469	6.2	23.0	1.0	1.4	12.7	9.2
S.E.	27.4	30.3	26.0	16.0	64.7	0.3	10.7	77.8	136.5	1.8	6.2	0.4	0.5	4.8	2.3

continued ...

TABLE 5.17

(continued)

	R. dorsalis					A. fluviatilis					E. octoculata				
Date	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
1972															
Nov. 21	2					2		6	7	3			1	35	3
Dec. 20	7	1			1	15		15	2	2				28	12
1973															
Jan. 19	8					14		1	3					50	5
Feb. 26	18	2				12		12			1			16	15
Mar. 29	6	2				7		2	2	4				45	23
Apr. 30	6		1		2	5				1				8	4
May 28	5	1				9			2					35	2
July 25	4	1			2	20								12	4
Sept. 4	9					21		9						8	7
Oct. 4	13				5	14		9	6	14			3	9	5
Nov. 7	6	1				20		9	1	2				9	4
Mean	7.6	0.7	0.09	0	0.9	12.6	0	5.7	2.1	2.3	0.09	0	0.4	23.1	7.6
S.E.	1.3	0.2	0	0	0.5	1.9	0	1.6	0.7	1.2	0	0	0	4.8	1.9

continued ...

TABLE 5.17

(continued)

	E. testacea					*E. ignita*					*H. angustipennis*				
Date	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
1972															
Nov. 21			6												19
Dec. 20		2	2			10									
1973															
Jan. 19		4	3			2									17
Feb. 26						1									15
Mar. 29		2	1												8
Apr. 30															
May 28						46									
July 25			6			23	1								
Sept. 4			12			3				3					2
Oct. 4						6							3		8
Nov. 7			5			4									2
Mean	0	0.7	3.2	0	0	8.6	0	0	0	0.3	0	0	0.3	0	6.4
S.E.	0	0	1.1	0	0	4.3	0	0	0	0	0	0	0	0	

TABLE 5.17

(continued)

	S. ornatum					L. pereger					B. rhodani				
Date	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
1972															
Nov. 21		108	28	10	1		1	22	2		7	5			
Dec. 20		105	587	76	11			44			107	8	30	8	7
1973															
Jan. 19		5259	410	312	60	2	1	27			89	27	48	20	30
Feb. 26		1088	67	430	29			6			18	1	21	6	10
Mar. 29		2460	200	270				8			59	2	36	16	12
Apr. 30	2		4					33	2		8	16	27	26	10
May 28	5	609	9	46	7			12	4	1	18	9	12		
July 25		198	291	138	70		1	38			5	23	9	6	43
Sept. 4	567	882	510	6	3			78	3		33	3	15		3
Oct. 4	117	1350	762	3	8		3	69			64		6	6	30
Nov. 7	4	1720	429	17	2	1	2	12			215	8	1	19	3
Mean	63.1	1252	299.7	118.9	17.4	0.2	0.7	31.8	1.0	0.09	56.6	9.2	18.6	9.7	13.4
S.E.	51.4	463	78.6	45.4	7.5	0	0	7.3	0	0	19	2.7	4.6	2.7	4.3

continued ...

TABLE 5.17
(continued)

	G. complanata					Naididae					Limnephilidae				
Date	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
1972															
Nov. 21							1	12					1	1	2
Dec. 20	1		1			300	4	0			4		3	1	3
1973															
Jan. 19		1	1			20	6				4				
Feb. 26			2			210					8	1	2		2
Mar. 29						80					2		3		1
Apr. 30						2000								1	
May 28															
July 25	1														
Sept. 4			3												
Oct. 4			9												
Nov. 7															
Mean	0.02	0.09	1.4	0	0	237	1	1	0	0	1.6	0.09	0.8	0.3	0.7
S.E.	0	0	0	0	0	178.9	0	0	0	0	0	0	0	0	0

continued ...

TABLE 5.17

(continued)

	Hydracarina					Lumbriculidae					Chironomidae				
Date	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
1972															
Nov. 21	1										11	21	60	4	6
Dec. 20											5	1	59	33	
1973															
Jan. 19								1		2	2	33	92	262	77
Feb. 26							1	2				64	93	114	16
Mar. 29						1		1		1	6	222	373	144	53
Apr. 30	12										15	15	69	59	20
May 28	3					1					151	144	102	160	42
July 25					3						1		25	47	10
Sept. 4			3		2						57	12	156	18	8
Oct. 4		3				1					32	18	360	10	12
Nov. 7						1				1	15	16	108	34	14
Mean	1.6	0.3	0.3	0	0.5	0.4	0.1	0.4	0	0.4	26.8	53.6	136.1	80.5	23.5
S.E.											13.4	20.9	35.8	24.3	7.2

	H. jenkinsi
July 25	6
Sept 4	3
Oct. 4	42
Mean	5.1

TABLE 5.18

Annual mean dry weights (mg) of invertebrates per 0.05m^2 at the five Stations of the R. Tean. Taxa separated into trophic entities.

	(1)	(2)	(3)	(4)	(5) Stations
Decomposers/ Detrital feeders					
A. aquaticus	0.2	47.6	527.5	338.0	25.7
G. pulex	42.8	0.1	4.5	43.5	23.8
H. angustipennis	0	0	0.6	0	13.9
S. ornatum	11.6	1063.8	160.7	71.3	19.3
Tubificidae	26.0	26.3	28.3	2.8	53.8
Lumbriculus	1.1	5.3	3.9	0	4.9
Chironomidae	4.0	11.3	32.7	16.8	3.6
	85.7	1154.4	758.2	472.4	145.0
Grazers/ Herbivores					
L. pereger	0	6.4	278.6	4.9	0
B. rhodani	20.7	6.3	25.4	5.9	11.8
H. jenkinsi	0	0	17.6	0	0
E. ignita	3.7	0	0	0	0.5
A. fluviatilis	41.1	0	2.2	8.4	4.4
Limnephilidae	1.6	0	0	0	0
	67.1	12.7	323.8	19.2	16.7

continued ...

TABLE 5.18

(continued)

	(1)	(2)	(3)	(4)	(5) Stations
Carnivores					
R. dorsalis	60.0	16.2	0	0	7.6
E.octoculata	1.1	0	19.3	148.7	61.1
E. testacea	0	39.8	63.5	0	1.9
G.complanata	9.4	0.9	5.5	0	0.4
Tipulidae	1.5	0	0	0	0
Chironomidae	0.6	0.06	0.14	0.25	0.01
	72.6	56.9	88.4	148.9	71.0

TABLE 5.19

Annual mean dry weights (mg) of invertebrates per $0.05m^2$ in the Experimental Streams. Taxa separated into trophic entities.

	(1)	(2)	(3) Stream
Decomposers/ Detrital feeders			
A. aquaticus	513.7	119.9	212.6
G. pulex	2.5	158.0	3.8
H. angustipennis	0	2.1	0
S. ornatum	801.0	7.0	495.8
Tubificidae	17.0	6.1	4.6
Lumbriculus	3.1	3.7	6.3
Chironomidae*	47.3	20.9	51.7
	1384.6	317.7	774.8
Grazer/Herbivores			
L. pereger	366.7	532.7	86.0
B. rhodani	4.6	26.3	3.2
E. ignita	0.6	9.8	0
A. fluviatilis	10.1	57.8	0
E. venosus	0.6	9.8	0
	382.6	636.4	89.2
Carnivores			
R. dorsalis	6.7	47.7	4.0
E. octoculata	2.0	12.9	1.6
E. testacea	1.1	0	0
Tipulidae	76.6	75.2	28.7
Sialis	0	3.5	5.5
Dicranota	0.9	2.4	0
Coleoptera larvae	0	0.8	0
Chironomidae*	8.37	6.7	5.3
	95.67	149.2	45.1

*Mean wt. of Chironomidae in the trophic entities has been estimated from the % of larvae in these entities and the total mean weight of Chironomidae for each Stream.

TABLE 5.20.

Mean Calories/ash-free gram for invertebrates encountered in the Experimental Streams and the R. Tean.

Species	Cal/ash free g.	% ash.
E. octoculata	6.07	7.86
H. angustipennis	6.27	10.9
R. dorsalis	6.39	11.1
S. ornatum	5.8	14.5
A. aquaticus	5.29	31.75
G. pulex	5.65	34.58
E. testacea	6.4	4.8
Tipula	5.96	13.9
Sialis	5.5/g	-
A. fluviatilis	5.57	49.8 (body)
L. pereger	5.39	18.8 (body)
Lumbriculus	6.05	16.6
Tubificidae	6.51	20.3
B. rhodani	6.49	14.3
E. ignita	5.82	6.4
E. venosus	5.8/g	-
Chironomidae	5.10	6.6

TABLE 5.21

Mean standing biomass of macro-invertebrates (mg. dry wt/ $0.05m^2$/yr) at Stations 1 - 5 of the River Tean.

(1)	(2)	(3)	(4)	(5)
225.4	1224.1	1170.0	640.6	233.0

Mean standing biomass of macro-invertebrates (mg. dry wt/ $0.05m^2$/yr) in Experimental Streams 1, 2 and 3.

(1)	(2)	(3)
1863.8	1097.8	909.2

Mean number of chironomid larvae per $0.05m^2$/yr at Stations 1 - 5 of the River Tean.

(1)	(2)	(3)	(4)	(5)
25.6	50.3	137.2	79.0	22.8

Mean number of chironomid larvae per $0.05m^2$/yr in the Experimental Streams.

(1)	(2)	(3)
184.7	77.3	180.3

Percentage of carnivorous chironomid larvae (as a percentage of total number of larvae per $0.05m^2$/yr).

Tean	(1)	(2)	(3)	(4)	(5)
	12.5	0.6	0.44	1.5	0.3
Streams	(1)	(2)	(3)		
	15.0	24.5	9.4		

TABLE 5.22

9.5.73. INVERTEBRATE DRIFT PER $0.04m^2$/15 min. IN THE EXPERIMENTAL STREAMS.

Incoming drift is counted in the first columns of each stream and outgoing drift in the second column.

	4.30pm		3.45pm		5.15pm		8.05pm		7.15pm		8.50pm		12.5am		11.30pm		12.20am.	
Experimental Stream	1	1	2	2	3	3	1	1	2	2	3	3	1	1	2	2	3	3
G. pulex			2	1										1				
B. rhodani			2						1	2			1		2	1	1	
A.aquaticus					1		3						1					
R. dorsalis	1																	
Hydracarina	1		1															
Naididae			3															
S. ornatum								1										
Ephydia															1			
Tipula																		
M. atratulus	7	1	3	1	1	11	1	4	1	1	3	2		1		1	5	13
M. atrofasciatus	1	1		2	1			1		2		1		1	2			
E. hospita	1	1					2	1			2		3	3		1	1	
Rheocricotopus		1				2					1					1	1	
Procladius. sp.			2	1										1		4		1
Thienemannimyia			1		1				1		1				2	1	3	
P.arundineti		1					1											
P. longimanus				1					2				2					
P. olivacea																		
B. longifurca													1				1	
C. albeforceps?	3						7	1					1				4	
C.(isocladius)sp.			5								1					1		
D. ex.gr.vitellinus																		
C. bicinctus	3	4					1	2				2	7		2	1		
Ortho sp. C10		1		13	1	1		3										
" " C11		2	1			1			3	1	1		10	1				
Adults					8	77	7	51	9	10	34	65	11	22	3		33	49
Pupae	4			1	3	7	3	9	3	2	1	5	3	6	2		2	3

TABLE 5.22

(continued)

	3.50am		2.30am		4.30am		6.55am		6.00am		7.30am		10.4am		9.15am		10.50am	
Experimental Stream	1	1	2	2	3	3	1	1	2	2	3	3	1	1	2	2	3	3
G. pulex			1											3				
B. rhodani	1		1		1		2								1	2		
A.aquaticus								1						1				
R. dorsalis																		
Hydracarina																		
Naididae																		
S. ornatum																		
Ephydia																		
Tipula			1															
M. atratulus		1	1			3	2	4		1	2		1	1			7	3
M. atrofasciatus				2										1			1	
E. hospita							1	1		1			1					
Rheocricotopus										1						1		
Procladius. sp.	1		1			1												
Thienemannimyia					1			1		2		7				1	1	
P. arundineti								1						1				
P. longimanus								1							1			
P. olivacea																		
B. longifurca	1																	
C. albeforceps?	3	1			1		1	4		1			1				4	
C.(isocladius)sp.		2					1									1		
D. ex.gr.vitellinus						1								1				
C. bicinctus											2	1		2				
Ortho sp. C10							1					2						
" " C11	5	1	2	2	1						2		3	1				
Adults	14	13	1	1	12	21	6	36		4	19	35	12	41	6	3	18	21
Pupae	2	3		1	3	3	1	8			2	7	1	8				

TABLE 5.23

INSECT EMERGENCE:

Numbers and dry weights (mg) per 1.8×10^3 cm^2 taken over 24 hrs. in the Experimental Streams

		Simulium ornatum		Chironomidae		Baetis rhodani		Others	
Stream	Date	Number	Wt.	Number	Wt.	Number	Wt.	Number	Wt.
1	24.7.73	1	-	5	-	3	-	3	-
2		0	-	14	-	0	-	0	-
3		2	-	22	-	1	-	1	-
1	3.8.73	11	8.7	89	9.1	7	7.8	0	3.0
2		5	4.1	64	6.1	4	6.2	2	-
3		1	-	14	1.6	2	3.0	1	3.0
1	24.8.73	11	8.4	38	6.3	1	1.2	0	0
2		2	1.5	17	0.3	1	2.0	2	0.9
3		3	2.2	39	9.1	0	0	2	2.8
1	31.8.73	1	1.2	135	12.4	0	0	11	2.6
2		0	0	19	2.4	0	0	3	0.6
3		0	0	327	17.1	0	0	5	1.0
1	25.9.73	224	151.5	335	17.0	0	0		
2		18	12.6	110	4.9	1	1.1		
3		12	10.4	67	3.7	0	0	1	-
1	2.10.73	29	18.9	185	7.6	0	0		
2		10	6.6	136	7.8	3	4.1		
3		18	11.4	152	6.8	0	0		
1	4.4.74	6	5.1	15	1.9	0	0	2	1.5
2		0	0	8	1.2	0	0	0	0
3		0	0	7	1.2	0	0	0	0

TABLE 5.23 (continued)

		Simulium ornatum		Chironomidae		B. rhodani		Others	
Stream	Date	Number	Wt.	Number	Wt.	Number	Wt.	Number	Wt.
1	18.4.74	0	0	1	–	0	0	0	0
2		0	0	2	–	0	0	0	0
3		0	0	22	–	0	0	0	0
1	3.5.74.	1	1.0	16	1.9	0	0	0	0
2		0	0	10	1.0	0	0	1	0
3		0	0	0	0	0	0	0	0
1	16.5.74.	2	–	32	4.9	0	0	4	5.6
2		1	–	13	1.5	0	0	2	1.7
3		0	0	4	0.2	0	0	0	0
1	31.5.74.	3	2.3	65	8.4	2	3.0	0	0
2		0	0	33	4.2	3	7.4	2	0.5
3		1	1.4	25	4.2	0	0	1	1.9
1	7.6.74	1	–	36	3.3	0	0	1	0.8
2		0	0	26	2.5	0	0	3	1.0
3		0	0	21	2.5	0	0	0	0
1	14.6.74.	2	0.7	43	5.1	0	0	1	0.8
2		4	1.7	46	6.2	0	0	1	0.8
3		3	1.5	4.9	5.3	0	0	1	1.5

CHAPTER 6.

THE EFFECTS OF DIURNAL FLUCTUATIONS IN OXYGEN TENSIONS ON BENTHIC MACRO-INVERTEBRATES

6.1. INTRODUCTION.

Reference has been made in the Introduction (Chapter 1) to the effect of increased photosynthesis during the day, and respiration during the night as a result of enlarged growths of algae and bryophytes caused by the increased nutrient supply with eutrophication. It was pointed out that eutrophic rivers could revert from 5% saturation with oxygen at night to super-saturation during the day, although figures would be dependent upon the oxygen demands of pollutants, the amount of re-aeration and solar radiation. In Chapter 5 (5.5.c.) the large quantities of the blanket weed Cladophora in the Rivers Tean and Ray and in the Experimental Streams were commented upon, with particular reference to the oxygen tensions within the matrix of the filaments where circulation of water would be poor.

In a grossly organically polluted river like the Upper Cole prior to 1972 (Chapter 4), very low night determinations of oxygen are expected due to the increased organic degradation by micro-organisms, while during the day, respectable levels of oxygen are recorded as a result of photosynthesis. Examples of fluctuations from both eutrophic and organically polluted conditions are given in the table below: -

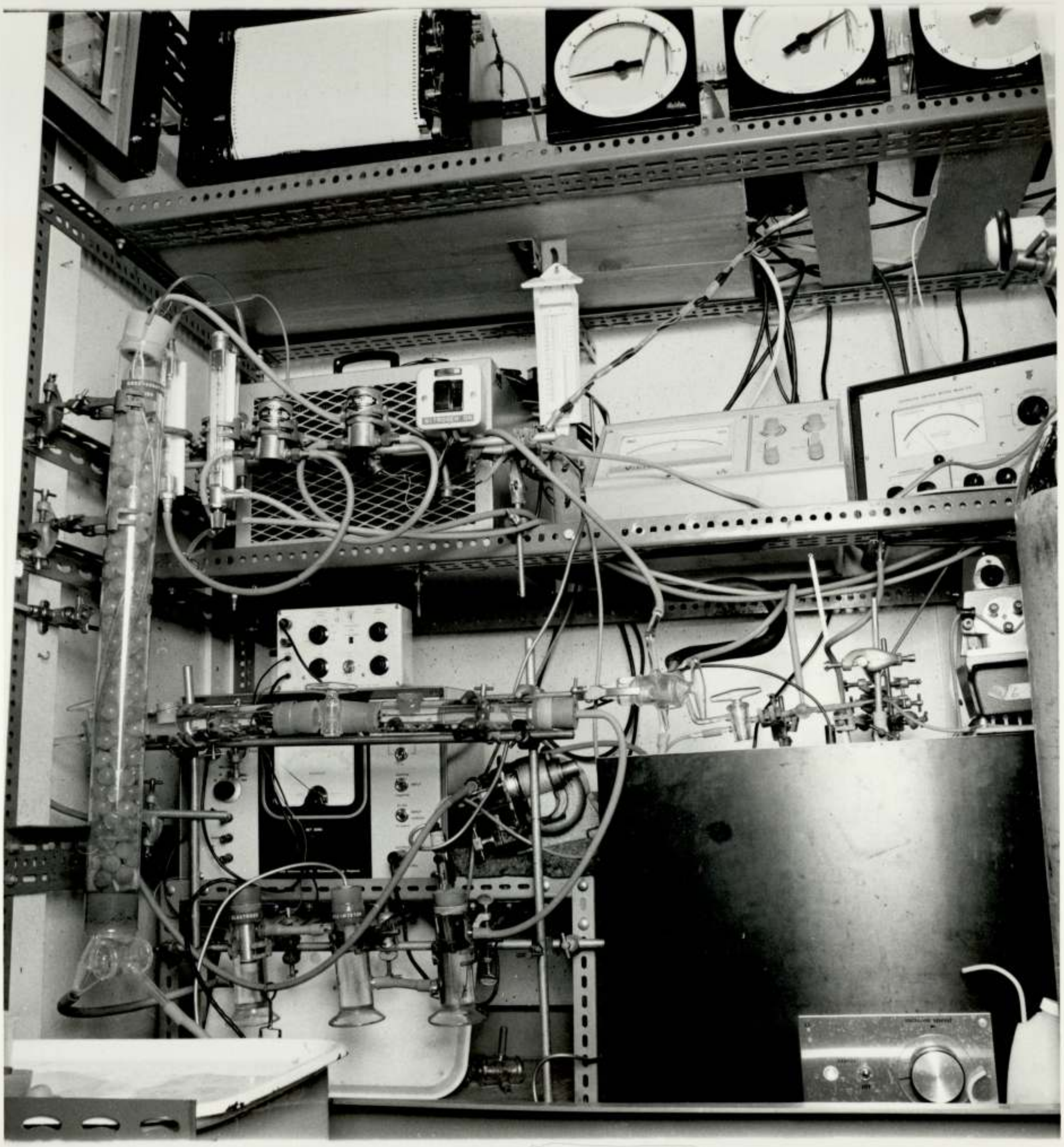
TABLE 6.1. showing examples of the maximum and minimum oxygen concentrations in mgl^{-1} occurring at night and during the day in the R. Cole (from Davies, 1971) and the R. Ray.

River	Maximum(day)	Minimum(night)	Difference
Cole (Midlands)	7.0	1.0	6.0
Cole	11.0	6.0	5.0
Cole	5.0	0.5	4.5
Ray (Wiltshire)	18.0	4.0	14.0
Ray	8.0	3.5	4.5
Ray	16.5	4.5	12.0
Ray	11.0	3.0	8.0

The time that a certain oxygen concentration is maintained in a river is also a function of the intensity of solar radiation, time of year, and in the case of some polluted rivers the extent and severity of the discharge. For example, even in summer when 18 hours of light prevail there may be many hours of a low oxygen tension due to the existence of pollution, Hawkes and Davies (1970) having furnished an example where the R. Cole (29.6.67) maintained approximately 1 mgl^{-1} of oxygen for over 12 hours. As a rule a stable concentration of oxygen is not maintained for long but long periods above an arbitary concentration may be.

For these reasons it was decided to experiment using three different temporal periods at a number of different oxygen concentrations (Fig. 6.1.) The effects of two temperatures were investigated while keeping the pH of

plate 9



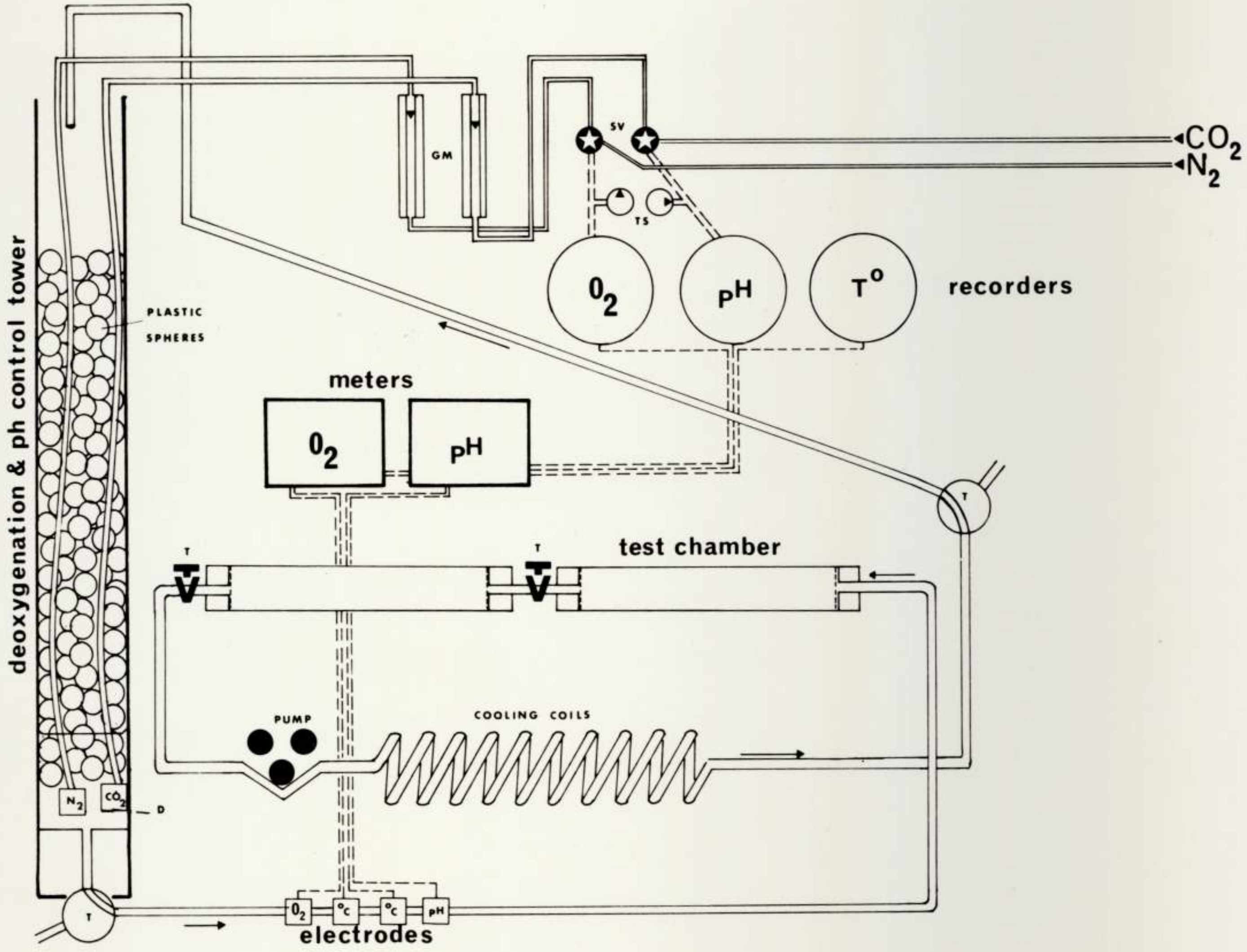


PLATE 10

Apparatus controlling parameters during diurnal oxygen fluctuations the water constant throughout each experiment. Photoperiods were induced to coincide with the natural situation of high oxygen levels during daylight and lower oxygen levels during the dark.

The synergistic effects of temperature, pH, and fluctuating oxygen concentrations with respect to intensity and time upon benthic macro-invertebrates were considered worthy of investigation as oxygen fluctuations such as these were thought to affect their distribution and range in nature. An apparatus was developed in which macro-invertebrates could be subjected to accurately controlled conditions of temperature, pH and oxygen concentrations, the latter with respect to time periods also. Initially, the survival of A.aquaticus and G. pulex under such regimes were tested, but A.aquaticus was found to be so tolerant that the experimental programme could not have been completed in the time available and tests with this animal were abandoned. G. pulex, an oxygen sensitive species does inhabit eutrophicated rivers living in loose moss and between stones, and seemed an ideal animal for preliminary investigations.

6.2. DESIGN OF APPARATUS AND EXPERIMENTAL METHODS.

The diagram of the apparatus (Plate 10) and a photograph (Plate 9) show the essential components of the system designed to induce diurnal fluctuations in oxygen concentrations within the animal chambers while keeping the temperature and pH of the water constant throughout the experiments.

Fluctuations in oxygen concentration were presented with respect to time and intensity which can best be described by referral to Fig. 6.1. High and low oxygen concentrations have been presented within any experiment on a circadian basis. The lowest concentration in any one test was varied in time between experiments. Four, eight and twelve hour periods of low oxygen levels were tried to show the effect of increasing temporal exposure on the experimental animals. The difference between these periods and 24 hr. constituted the other (high) part of the oxygen fluctuation, which was either to an oxygen concentration of 6.0 mgl^{-1} or to 10 mgl^{-1} at 10°C. The experiments were duplicated at 20°C to observe the thermal effect, and in this case the high part of the oxygen fluctuation was either 6.0 mgl^{-1} or 8.0 mgl^{-1}. The differences in the highest oxygen concentrations were the result of the re-aeration of the experimental water only reaching about 88% of oxygen saturation at both temperatures. The re-aeration procedure is described later on.

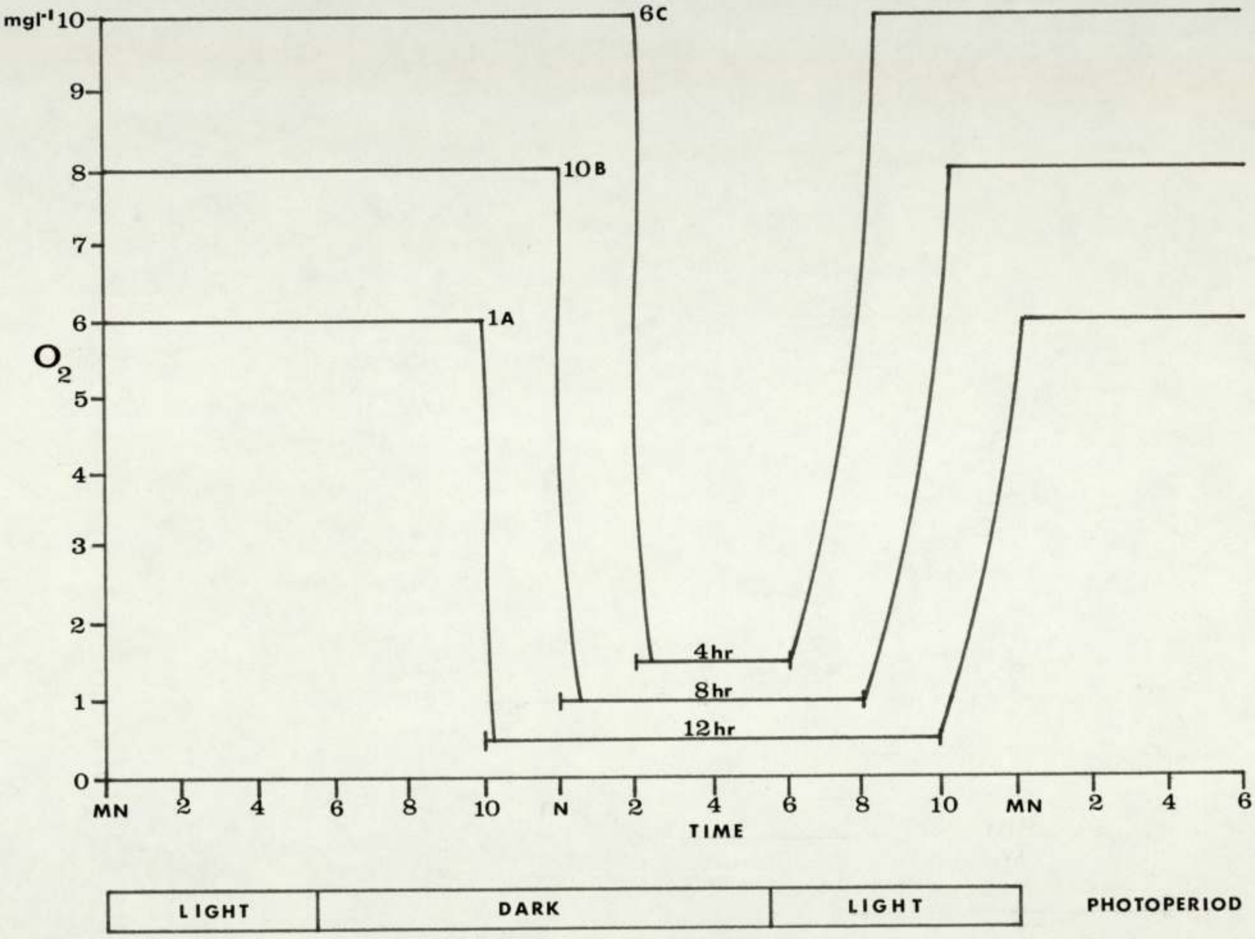
6.2.1. Control of parameters.

a) Control of oxygen tensions.

The oxygen tension required was achieved by the use of an oxygen electrode (Mackereth type), an E.I.L. 15A Dissolved Oxygen Meter, and a Fielden 24 hr. recorder equipped with limit switches. The concentrations required were set on the recorders limit switches; for example, 0.5 mgl^{-1} O_2 on the lower limit switch and 6.0 $mgl^{-1}O_2$ on the upper limit switch in the case of Experiment 1 (Table 6.2). The oxygen electrode and its compensating thermistor were placed as near to the de-oxygenation and pH control tower as possible (Plate 10) so that accurate control of the concen-

FIG.6.1. illustrating diagrammatically examples of diurnal oxygen fluctuations in both concentration (mgl^{-1}) and time (hr). Experiments 6.c.,10.b., and 1.a. are shown centralised for ease of comprehension but in fact all oxygen stripping commenced at 10am regardless of the length of the fluctuation. The curves demonstrate the relative speed of oxygen stripping compared with the slower recovery by re-aeration.

Fig.6.1.



trations could be maintained. The oxygen electrode detected the oxygen tension in the experimental water and, when the pre-set oxygen limits were reached, the limit switches of the recorder operated, closing the circuit and operating a solenoid valve via a relay. The operation of the solenoid valve (s.v.) permitted the passage of a metered amount of nitrogen (80 cc/min) through a diffuser situated in the de-oxygenation tower, thus stripping off oxygen until the electrode detected the concentration to be within the limit. By careful control of the water volume, re-aeration (achieved by trickling over the plastic spheres in the tower), carbon dioxide level and nitrogen flow, the oxygen levels were controlled to within 1% of saturation at the lower oxygen levels and 2% of saturation at the higher oxygen levels.

b) Diurnal fluctuations.

The duration of upper and lower oxygen levels was controlled by placing a time switch (t.s.)between the limit switches of the recorder and the solenoid valve (Plate 10).

c) Control of pH.

The effect of diffusing nitrogen through the experimental water is to alter the partial pressures of gases above the water and cause oxygen and carbon dioxide to come out of solution. The carbonic acid - carbonate - bicarbonate equilibrium is altered with the free carbon dioxide loss with a concomitant change in pH. The overall effect of oxygen stripping by nitrogen, therefore, is to increase the pH.

The pH was maintained at 7.2 during all experiments in a similar fashion to the oxygen regulation. An E.I.L.

pH electrode and temperature compensator were inserted into the system just downstream of the oxygen control electrodes and were thus in a position for exacting control in the de-oxygenation and pH control tower (Plate 10) to be achieved. Metering of the pH was accomplished with an E.I.L. Vibret pH Meter and 24 hr. recordings were made on a Fielden recorder equipped with limit switches. Only one limit (7.2) was set, above which a solenoid valve was activated via a relay, allowing a metered amount of carbon dioxide gas (100 cc/min) to be supplied to the diffuser in the tower.

As the carbon dioxide was always being removed during oxygen stripping, pH control could be achieved to within ∓ 0.1 of a unit. During the times of high oxygen periods when no de-oxygenation was occurring, little pH control was necessary. The amount of carbon dioxide remained about an average after the first burst of gas via the solenoid valve. It decreased after the commencement of oxygen stripping and changed again after stripping was over and atmospheric partial pressures of carbon dioxideand oxygen were resumed. These were the same for each experiment.

d) Control of temperature.

The 10 and 20°C experimental temperatures were maintained by passing the water through glass cooling coils immersed in a refrigerated water bath. Control was accurate to 0.01°C. The whole apparatus was situated inside a constant temperature room maintained at either 10° or 20°C.

e) Re-oxygenation.

Re-oxygenation was a matter of diffusion of oxygen from the air in the de-oxygenation and pH control tower aided by percolation of the water over plastic spheres. Continual re-oxygenation occurred therefore, more so during stripping, which aided the process of control and replenishing the loss caused by the electrode when stripping was not occurring.

The oxygen and pH meters were re-calibrated every weekday using air equilibriated water, barometric pressure and saturated water vapour pressure to achieve 100% saturation on the oxygen meter, and 7.0, 7.4, and 8.0 pH buffers for calibration of the pH meter. Twenty-four hour continuous recordings were made of pH oxygen and temperature thus allowing the efficiency of the system to be checked every day.

f) Photoperiods.

It was convenient to subject the constant temperature room to 12 hr. dark: 12 hr. light, with the latter from 17.30 hrs to 05.30 hrs, thereby coinciding with times of the high oxygen levels both in the test chambers and in accordance with the coincidence of light and high oxygen levels in nature. The dark period was applied during the working day when oxygen levels were at their lowest. Observations on the mortality rate were made using a red bulb.

The experimental animals (Gammarus pulex) were kept for at least two weeks in another constant temperature room under the same light regime before use.

6.2.2. The Experimental Water.

Water was taken in large quantities from Langley Pool - a small lake feeding Langley Brook from where the experimental animals were taken. The water was filtered through Whatman GF/C glass fibre filter papers and stored at 10° and 20°C. The alkalinity was measured (260 mgl^{-1} $CaCO_3$) from time to time but little variation was apparent outside titremetric error. This batch of water was used throughout each experimental programme.

6.2.3. Experimental Animals.

Gammarus pulex (L) subsp. pulex (Sch) were collected from Langley Brook by heel sampling and kept in buckets at 10 and 20°C with food and aeration. At least two weeks before use they were transferred into small glass aquaria and induced into the photoperiod described above. Some of the animals used in November and March were in pre-copula. As far as possible, a random selection of size and sex was made for animals used in the experiments.

6.3. PROCEDURE.

As the constant temperature room never maintained exactly the same temperatures within all parts of it, heat was often lost or gained by the apparatus making it necessary to run the experimental water through the apparatus for about 10 min. before the animals were introduced. About 20 animals were introduced into each test chamber through bunged apertures in their sides. The test chambers initially held 20 A. aquaticus in one and 20 G. pulex in the other but when Asellus experiments were terminated after one trial run because of their great tolerance, 20 Gammarus were placed in both chambers.

The pH of the water into which they were placed was already under carbon dioxide control and the oxygen saturation was always approaching 90%. The animals were then acclimated to their new surroundings for over 24 hrs. before the diurnal fluctuations were induced. Food was provided in the form of circles of decaying alder leaf cut from the lamina with a cork borer, and shelter was accomplished by previously placing a layer of gravel followed by small stones on the floors of the chambers. The water was circulated throughout the whole system by a Multifix peristaltic pump acting on 6.5 mm I.D. silicone tubing. The pump tube was oiled externally for longer life and changed every 7 days. The flow rates were adjusted and maintained in the test chambers at 2 cm/sec. for all experiments.

6.3.1. The Oxygen concentration/time fluctuations.

The oxygen intensities were chosen using rationales arising from observations of diurnal patterns in some Midland Rivers. The lowest levels tested were 0.5, 1.0, and 1.5 mgl^{-1} oxygen. These were varied temporally for 4, 8 and 12 hr, the remaining period of each 24 hr. being spent at 6.0 or 10.0 mgl^{-1} oxygen at 10^oC; 6.0 or 8.0 mgl^{-1} oxygen at 20^oC.

TABLE 6.2. showing the 36 experiment plan denoting the ranges of oxygen fluctuation and the times spent at each oxygen level (a, b, and c).

Temps	Oxygen			Oxygen		Oxygen		Oxygen (mgl^{-1})
10°C	1.	0.5	-	6.0mgl^{-1}	2.	0.5	-	10.0mgl^{-1}
"	a)	12hr	-	12hr	a)	12hr	-	12hr
"	b)	8hr	-	16hr	b)	8hr	-	16hr
"	c)	4hr	-	20hr	c)	4hr	-	20hr
"	3.	1.0	-	6.0mgl^{-1}	4.	1.0	-	10.0mgl^{-1}
"	a)	12hr	-	12hr	a)	12hr	-	12hr
"	b)	8hr	-	16hr	b)	8hr	-	16hr
"	c)	4hr	-	20hr	c)	4hr	-	20hr
"	5.	1.5	-	6.0mgl^{-1}	6.	1.5	-	10.0mgl^{-1}
"	a)	12hr	-	12hr	a)	12hr	-	12hr
"	b)	8hr	-	16hr	b)	8hr	-	16hr
"	c)	4hr	-	20hr	c)	4hr	-	20hr
20°C	7.	0.5	-	6.0mgl^{-1}	8.	0.5	-	8.0mgl^{-1}
"	a)	12hr	-	12hr	a)	12hr	-	12hr
"	b)	8hr	-	16hr	b)	8hr	-	16hr
"	c)	4hr	-	20hr	c)	4hr	-	20hr
"	9.	1.0	-	6.0mgl^{-1}	10.	1.0	-	8.0mgl^{-1}
"	a)	12hr	-	12hr	a)	12hr	-	12hr
"	b)	8hr	-	16hr	b)	8hr	-	16hr
"	c)	4hr	-	20hr	c)	4hr	-	20hr
"	11.	1.5	-	6.0mgl^{-1}	12.	1.5	-	8.0mgl^{-1}
"	a)	12hr	-	12hr	a)	12hr	-	12hr
"	b)	8hr	-	16hr	b)	8hr	-	16hr
"	c)	4hr	-	20hr	c)	4hr	-	20hr

6.3.2. Observation of mortality.

Hourly inspections of the test chambers for mortalities were made during 20°C runs at the lower oxygen levels; i.e. 0.5, 1.0, and 1.5mgl^{-1} O_2. Hourly inspections during the progress of 10°C runs when 12 hr periods of the same oxygen levels were being administered were also made, otherwise inspections were made at approximately two hourly intervals during the day. An electric torch with a red filter was used to count the number of dead animals in the otherwise dark constant temperature room. Dead animals and remains were removed after the low oxygen fluctuation period was over and percentage saturation was above 50% - the water circulation had to be switched off for up to three minutes to facilitate their removal.

The numbers removed were checked against the numbers recorded dead with the torchlight and then recorded against time; zero time being the beginning of the first descent to low oxygen level. Experiments were continued until 100% mortality had occurred with the exception of Experiment 3b which was terminated at 70% cumulative mortality, when, after 280 hr, fuses blew and killed the remainder.

Death was indicated by a typical orange/yellow colouration if it had occurred overnight, but this was extremely rare, most deaths occurring during the low oxygen fluctuation period or within a couple of hours afterwards. Deaths within 1 or 2 hr periods were recorded if there were no respiratory movements for over 5 min, or if there was no response to jolting - achieved by pinching and releasing the rubber tubing supplying the water to the test chambers.

6.3.3. Controls.

Controls for the experimental or mechanical effects were executed at 10° and 20°C adopting exactly the same experimental methods used above with the exception that no fluctuations in oxygen levels were given. The pH was maintained throughout the experiments at 7.2 ∓ 0.1 and aeration was achieved by diffusion in the de-oxygenation and pH control tower. This meant that oxygen saturation levels were approximately 90% at both temperatures, or 8.0 mgl^{-1} at 20°C and 10.0 mgl^{-1} at 10°C.

While all the experiments with oxygen fluctuation were completed within the period November 1973 to April 1974, i.e. with the effects of death from natural causes after mating (in males) and brooding in some females, minimised, the controls were started in May and finished in July. Thus some mortalities may have been due to natural causes following the spring brooding; but the extensive survival times of the control experiments prevented their being executed in November or indeed for every experimental run, i.e. paired controls. One control experiment was therefore accomplished for each temperature with each test chamber containing a population of 20 G. pulex.

6.4. RESULTS.

With the exception of experiment la and lc when only one test chamber contained G. pulex, the mortalities observed at a particular time in each test chamber were averaged. This gave a mean mortality rate appropriate to two populations each of 20 individuals.

A number of experiments were not performed because

100% mortality had occurred within the time covered by the previous experiment; for example, Experiment 1b (8 hr at 0.5 $mgl^{-1}0_2$ - 16 hr 6.0 $mgl^{-1}0_2$) was not performed because 100% mortality of the G. pulex population had occurred within 6 hr of experiment 1a (12 hr at 0.5 $mgl^{-1}0_2$ - 12 hr at 6.0 $mgl^{-1}0_2$) avoiding the need to repeat the experiment for an 8 hr time period in Experiment 1b. Other experiments not executed directly for the same reasons were 2a, 2b, 7b, 7c, 8a, 8b, 8c, and 10a.

At 10°C some of the experiments involving shorter time periods of exposure to greater oxygen concentrations appeared to have similar mortality rates to the 10°C control, i.e. the effects of the fluctuating oxygen intensities on mortality were not apparently greater than those caused by experimental or mechanical effects, and because of the large time scales involved (up to 700 hr) they were either terminated (Experiments 5a and 6a) or omitted (Experiments 4b, c, 5b, c, and 6.)

6.4.1. Presentation.

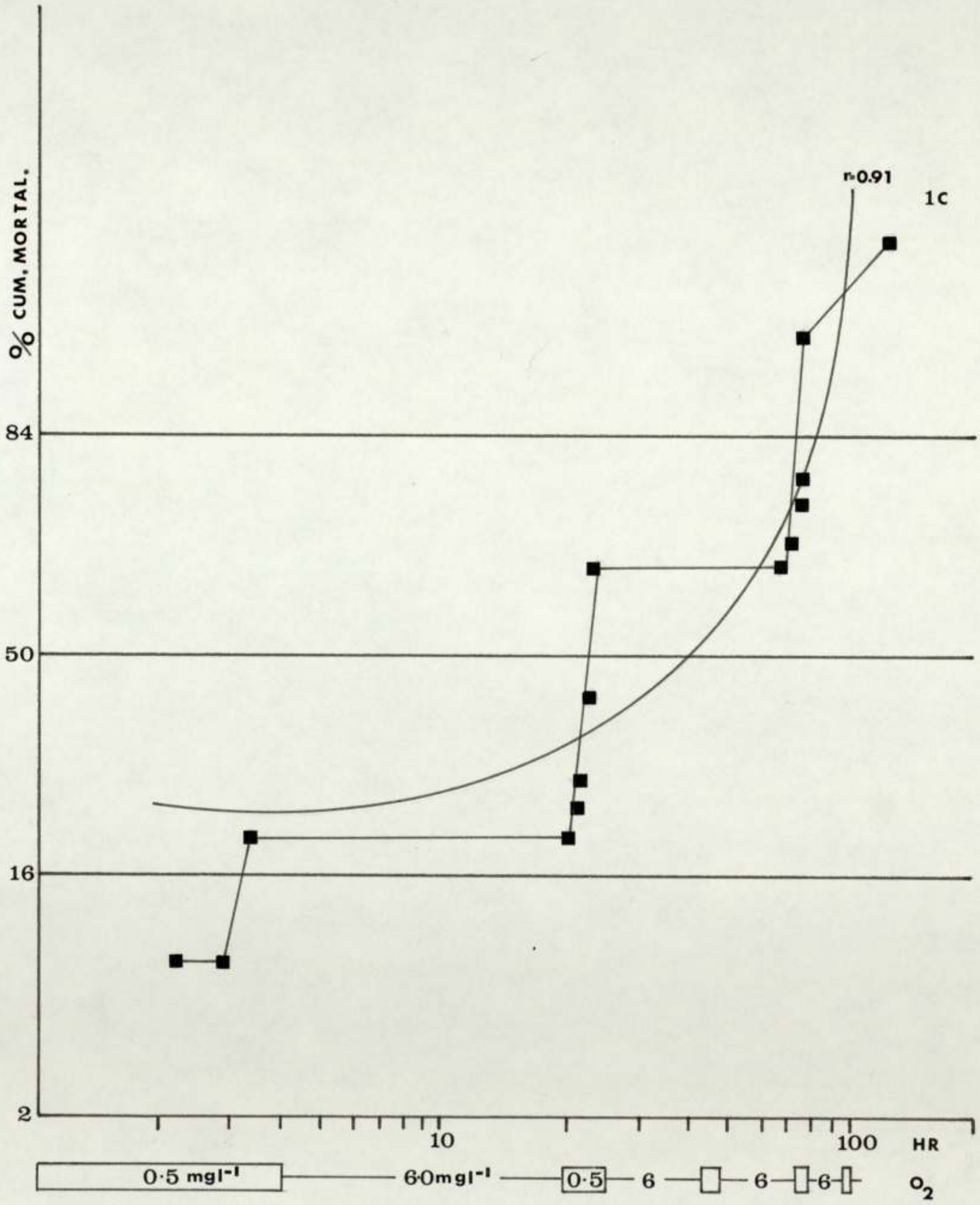
There is, apparently, no work of this type in the existing literature on which to base methods for expressing the results to achieve conformity. An original method has therefore been proposed.

The raw data was first transformed into figures appropriate to an infinite population rather than an average of two populations of 20 individuals, using the method suggested by Finney (1952). The individuals were expressed as a percentage of the total population; i.e. when n=20 one mortality = 5% of the total. The cumulative % mortality was thus calculated by multiplying the total number of deaths by 5 and subtracting 2.5 to transform results into a form representative of an infinite population. If this data was than plotted against time on probability/logarithm paper or linear/logarithm paper (log time) the type of graph formed has been illustrated in Fig. 6.2 - black squares. The horizontal lines between the periods of low oxygen concentrations (denoted by blocks along the time axis) show that nearly all the mortalities occur within these periods of low oxygen levels. The correlation coefficients (r) for the regression of y upon x were calculated for a number of experiments on data in the arithmetical form and all were significant, most at the 0.001 level. However, it seemed logical to omit those times when no mortalities were expected; i.e. outside the 12, 8 or 4 hr periods of low oxygen levels, and present the data as a line representing periods of low oxygen concentrations running concurrently. This would then render the line suitable for the determination of slope and comparison for parallelism with other slopes (Litchfield, 1949; Litchfield and Wilcoxon 1949) or indeed comparison of L.T.50's - the median lethal time in which 50% of the population dies under the conditions of the experiment.

Data was therefore reworked into a form that omitted the recording of the same mortality at different times, i.e. between those periods of higher oxygen fluctuations represented by the horizontal line (Fig. 6.2). If, infrequently, there were mortalities during this period (rarely, and never more than one) they were shifted to the last point in time of the preceeding low oxygen fluctuation; which was considered immediately responsible for the death(s). The regression

Experiment 1.c. ■= plot of % cumulative mortality (probit scale) of <u>G.pulex</u> against logarithmic time including mortalities outside the 4hr period of low oxygen tension (clear blocks below tine axis). The curve illustrates the regression of y upon x for the same data with the mortalities outside the 4hr periods of low oxygen tension shifted to the last point in time of the low oxygen fluctuation, and also omitting the recording of the same mortalities at different times. See Section 6.4.1.

Fig.6.2.



line now represented the results of low oxygen fluctuations running concurrently (Fig. 6.2 - curve). The r values are approximately the same but now the degrees of freedom are reduced and consequently the significance of the correlation is higher.

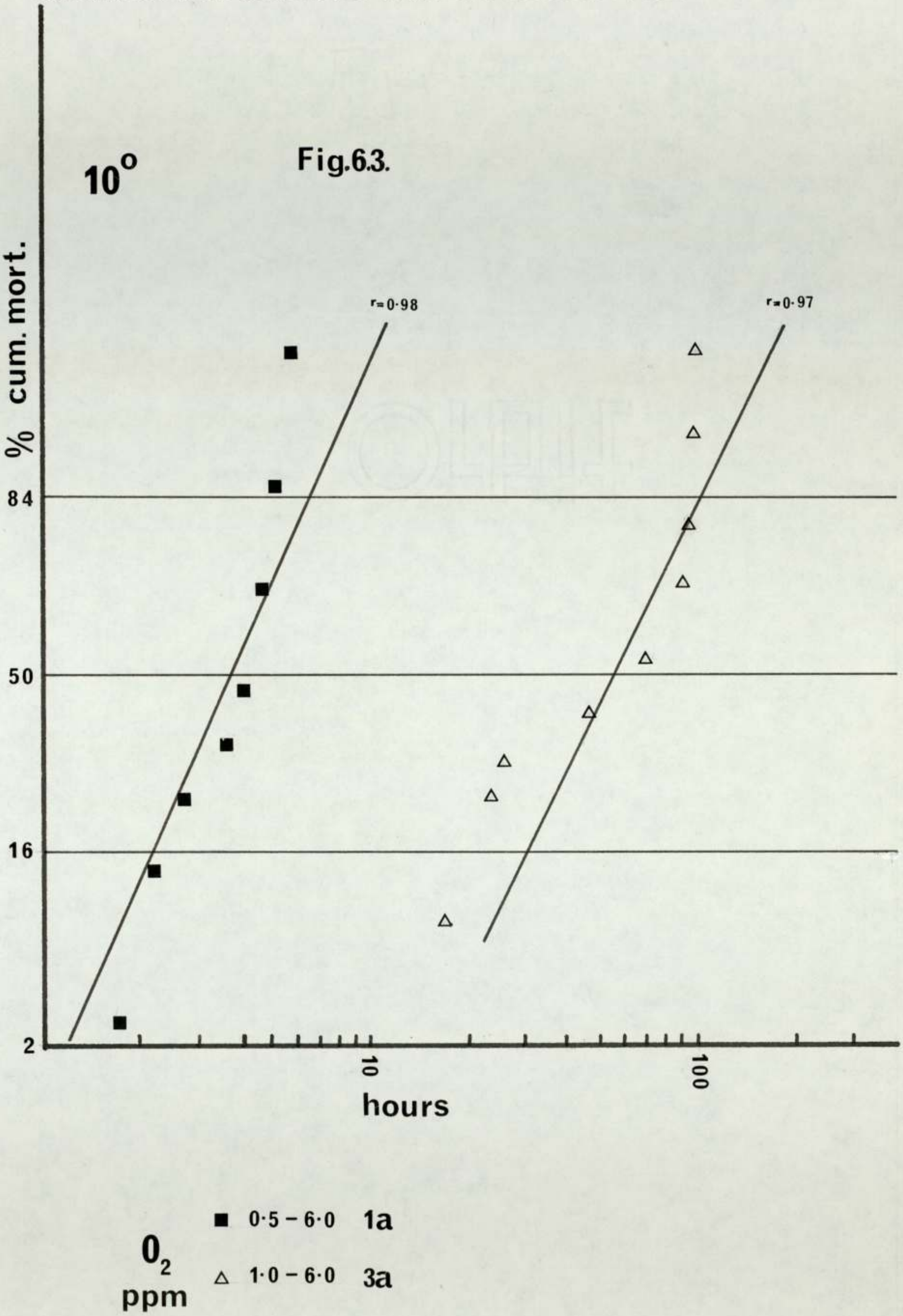
The cumulative percentage mortality resulting from continuous, i.e. unfluctuating doses of low oxygen, when plotted against time usually results in a skewed sigmoid curve. The skew is usually eliminated by using a logarithmic time scale and the sigmoid faction straightened by plotting percentage mortality on a probability scale (Sprague, 1969). Those experiments where 100% mortality was achieved within the first period of lower oxygen levels, i.e. 1a, 7a and 9a are examples where the log/probit analyses may be used (Fig. 6.3., 6.4). When the mortalities were spread over a whole range of diel fluctuations of oxygen concentrations however, e.g. 3a and 11a, the usual skewed sigmoid form does not appear, as reflected by the correlation coefficient which is calculated on the arithmetic data. If this data is plotted on log/probit paper (Figs. 6.3 and 6.4 - Experiments 3a and 11a) it is patently unsuitable, and thus all data has been plotted on an arithmetic/log scale which reveals the natur nature of the correlation. (Linear scales were not used because of the large time scales involved in some experiments).

All the time-per cent effects have therefore been treated in a manner that represents the results of the low oxygen tensions running concurrently and have then been plotted on logarithm (time)/linear (mortality) paper.

For the purposes of comparison of the parameters temperature, oxygen tension to oxygen tension and temporal

Logarithm/probability plot of Expts.1a and 3a. Probability scale = % cumulative mortality of <u>G.pulex</u>. Lines and r values calculated from arithmetic data. See Section 6.4.1.

Fig.6.3.



Logarithm/probability plot of Expts.7.a.,9.a., and 11.a. Probability scale = % cumulative mortality of <u>G.pulex</u>. Lines and r values calculated from arithmetic data. See Section 6.4.1.

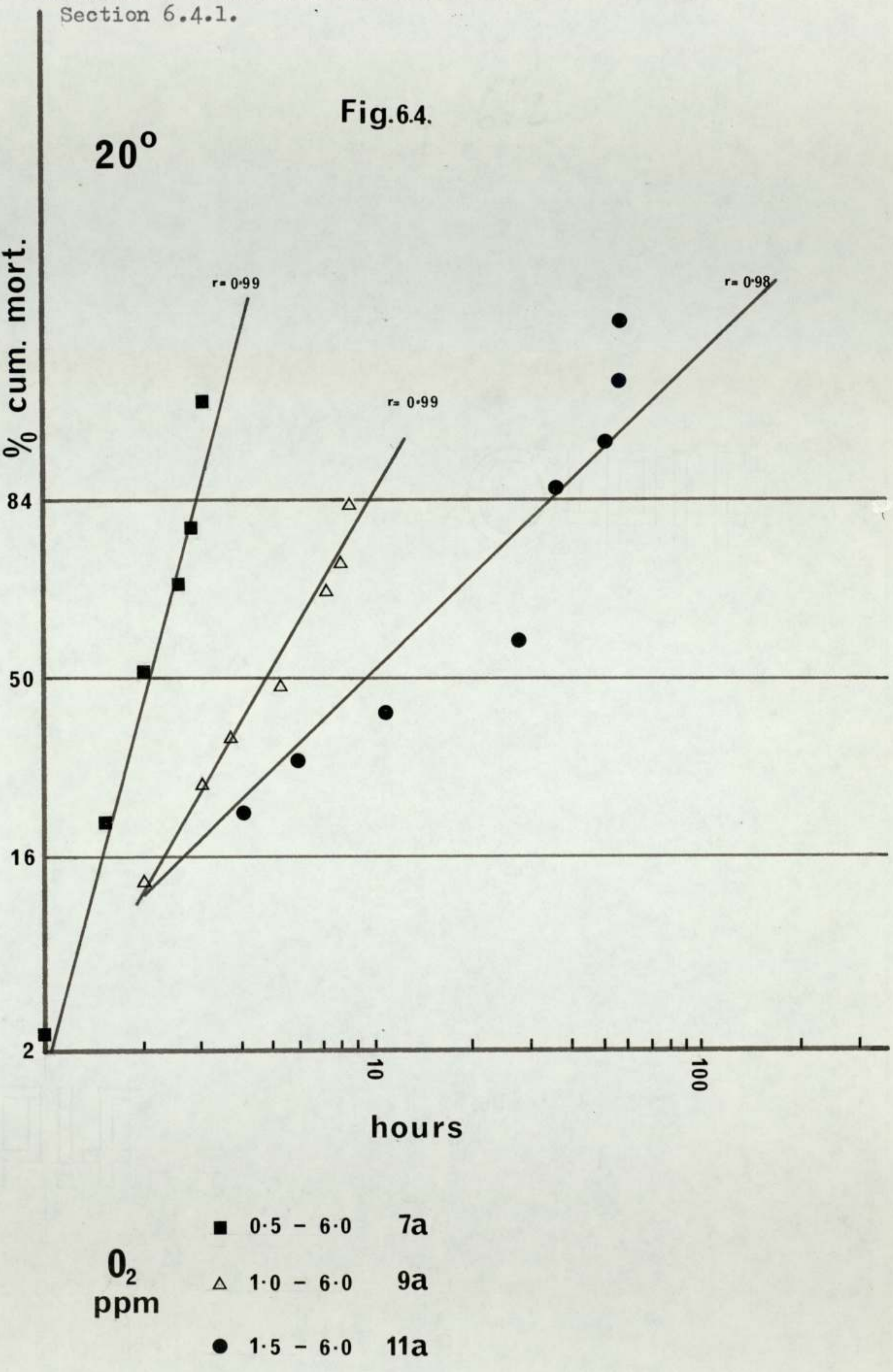


Fig.6.4.

fluctuations, the L.T. 50 for the whole experiments have been tabulated.

TABLE 6.3. Showing the L.T. 50's (hr) for the experiments listed (see Table 6.2). Those with asterisks denote their derivation from a preceeding experiment.

(a = 12hr/12hr, b = 8hr/16 hr, c = 4hr/20hr.)

	10°C			20°C	
$O_2(mgl^{-1})O_2$	Expt.	L.T.50	$O_2(mgl^{-1})O_2$	Expt.	L.T.50
0.5 - 6.0	1a	3.6	0.5 - 6.0	7a	2.05
	1b	3.6*		7b	2.05*
	1c	41.0		7c	2.05*
0.5 - 10.0	2a	3.6*	0.5 - 8.0	8a	2.05*
	2b	3.6*		8b	2.05*
	2c	15.0		8c	2.05*
1.0 - 6.0	3a	58.0	1.0 - 6.0	9a	5.4
	3b	230.0		9b	8.4
	3c	a.c.		9c	35.0
1.0 - 10.0	4a	52.3	1.0 - 8.0	10a	5.4*
	4b	a.c.		10b	14.5
	4c	o.		10c	10.5
1.5 - 6.0	5a	a.c.	1.5 - 6.0	11a	18.5
	5b	o.		11b	41.0
	5c	o.		11c	68.0
1.5 - 10.0	6a	a.c.	1.5 - 8.0	12a	24.5
	6b	o.		12b	33.0
	6c	o.		12c	83.0
	Control	380.0		Control	260.0

a.c. = approximated control situation see Section 6.4.

o = omitted.

6.5. DISCUSSION.

The essential considerations for assessment of the results from the experimental programme may be classified into four main areas. Those results or L.T.50's concerned with the effects of temperature; those pertaining to the effects of oxygen fluctuation; the level of oxygen fluctuation, and finally the result of returning from a low oxygen fluctuation to either a medium level of oxygen concentration (6.0 mgl^{-1}) or a high level of oxygen concentration (10.0 mgl^{-1} at 10°C, 8.0 mgl^{-1} at 20°C).

It is well known that increasing water temperature elevates the respiratory rates of a number of freshwater invertebrates, (Berg 1952, Berg and Ockelmann 1959, Edwards 1958b), and that increasing metabolism, changes in the life history and other factors (see Section 2.3.c.), results in their distribution and range being restricted (Walshe 1948, Hawkes 1962, 1969, Macan 1963, Hynes 1966). Also, if Hynes (1966) is to be believed, the autecological importance of temperature with respect to percentage saturation of water with oxygen is greater than the actual dissolved oxygen concentration. But the effect of temperature on the resistance to low oxygen concentrations was very clear, and while it has been demonstrated for G. pulex only, it is certain that most aquatic invertebrates would respond in a similar manner.

The L.T.50's for the control populations of G. pulex at 10° and 20°C showed how the temperature effect alone resulted in a 28% higher median survival time at 10°C than at 20°C (Table 6.3). No doubt part of the discrepancy in the survival times was caused by the experiments having to be conducted in slightly differing seasons, i.e. the 10°C control in May/June and the 20°C control in June/July. The seasonal condition of the animal with reference to reproductive status and subsequent changes in metabolic requirements has been demonstrated for Asellus and Ancylus by Lang (1951) and Berg and Ockelmann (1959). Moreover, although many rivers have annual mean temperatures of approximately 10°C (e.g. Tables 5.6 and 5.11), means of 20°C would be extremely rare and with this latter temperature being used as an arbitary figure for the purposes of comparison between oxygen fluctuations, it was not surprising to find the 20°C control mortalities heavier with respect to time.

Inspection of the L.T.50's for the 10° and 20°C components (Table 6.3) with oxygen and temporal fluctuations, i.e. horizontally, clearly shows the decreased survival times with an increase of 10°C. Also, for an increment of 0.5 $mgl^{-1}O_2$ at 10°C the effect of temperature on the L.T.50 was greater than a similar increment at 20°C. For example, the difference in L.T.50's between Expts. 1a and 7a (which only differ with respect to operating temperature) was only 1.55 hr but with the low oxygen fluctuation incremented by 0.5 $mgl^{-1}O_2$ in Expts. 3a and 9a, the difference is one of 52.6 hr. These differences in median lethal times were further disproportionately increased when the temporal effect of the fluctuation in low oxygen levels were considered; for example, comparing Expts. 1b and 1c, with 7b and 7c, the ratios of their L.T.50's were 1:11 and 1:1 respectively. The ratio of L.T.50's of Expts. 3a to 3b, was 1:4 while that of Expts. 9a to 9b was only 1:1.5, thus showing the temperature effect at different temporal exposures. Fig. 6.5 demonstrates the temperature effect graphically.

These results stress the importance of temperature to the survival of G. pulex in the environment, especially one depleted of oxygen diurnally and for different periods of time. The temperature effect is very marked between 10° and 20°C and even if differences of this magnitude were uncommon between seasons in all rivers, the effect of only a few degrees - which could be caused by effluents alone - could substantially restrict the range of this and other species in eutrophicated or organically enriched watercourses.

It has been demonstrated that fluctuations in dissolved oxygen of the magnitude used in the experimental programme do occur in nature (Table 6.1) although obviously not under the rigid regime that the experiments exemplify. There were two types of oxygen fluctuations encountered in the investigations: those which varied from a low oxygen concentration to a high oxygen concentration (even Experiment numbers) and those which varied from low oxygen concentrations to a medium oxygen concentration (odd Experiment numbers). At 10°C the difference in mortality rates between a fluctuation from a low oxygen concentration to a medium (6.0 mgl^{-1}) or high (10.0 mgl^{-1}) concentration of oxygen was expected to be virtually negligible owing to observations made in the field of G. pulex populations surviving well under 6.0 $mgl^{-1}O_2$ and the tolerance of aquatic invertebrates to such medium levels of oxygen at this temperature. While differences in the L.T.50's for such fluctuations were apparently the same in Expt. 3a and 4a, this was not the case with Expts. 1c and 2c when the low oxygen fluctuation only lasted for 4 hr. It was at first thought that the differences between the two L.T.50's

Fig.6.5. The effect of temperature (10 and 20^{o}C) on the LT$_{50}$ of G.pulex populations subjected to low concentrations of oxygen(0.5,1.0,1.5 mgl^{-1}) for 12hr alternated with high concentrations(6.0mgl^{-1})for 12hr. The control LT$_{50}$(10oC) has been used for Expt. 5a(1.5-6.0 mgl^{-1}O$_2$,12hr) which the latter approximated.

Fig.6.6. The effect of duration of the low oxygen fluctuation (0.5,1.0,1.5 mgl^{-1}alternating with high fluctuations of 6.0mgl^{-1}) on the LT$_{50}$ of G.pulex populations at 20^{o}C. ■=4hr. □=8hr. △=12hr.

Fig.6.7. The effect of duration of the low oxygen fluctuations(0.5,1.0,1.5 mgl^{-1} alternating with high fluctuations of 8.0mgl^{-1}O$_2$) on the LT$_{50}$ of G.pulex populations at 20^{o}C. ■=4hr. □=8hr. △=12hr.

fig 6.5.

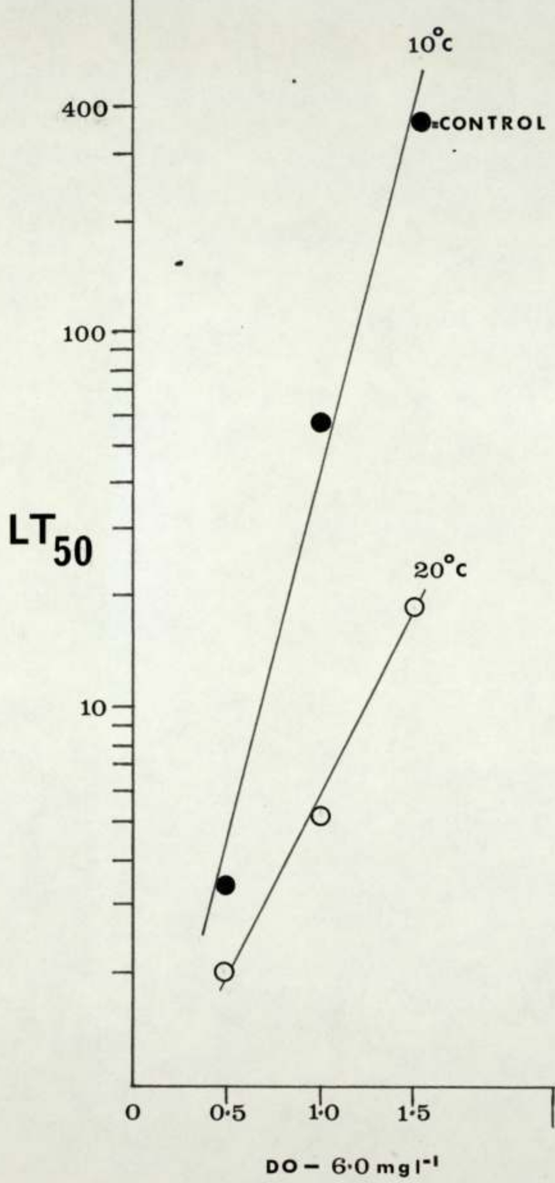


fig 6.6.

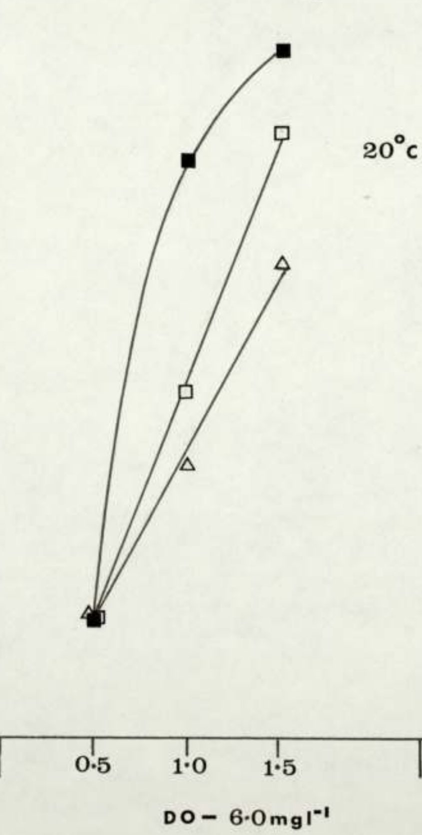
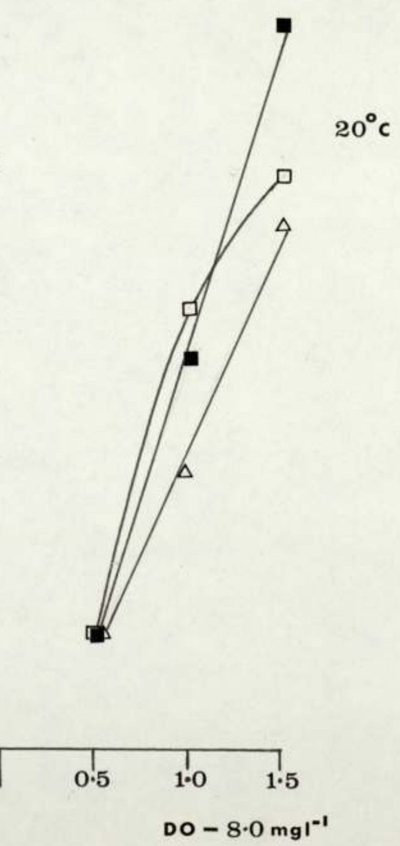


fig 6.7.



for the latter two experiments (41 hr for 1c, 15 hr for 2c) might have been the result of faulty selection or handling of animals but when a similar situation arose for Expts. 9c (35hr) and 10c (10.5 hr) it was thought that the greater shock consequent with the reduction of 9.5 $mgl^{-1}O_2$ (2c) as opposed to 5.5 $mgl^{-1}O_2$ for Expt. 1c might have been responsible. However, this patternwas not repeated in Expts. 11c and 12c where the L.T.50 of the population subjected to the larger drop in oxygen concentration (12c) was actually 20% higher than its 11c counterpart. Similarly unpredictable results occurred for Expts. 9b and 10b, 11b and 12b, i.e. sometimes mortality rates were decreased with greater oxygen drop and sometimes they were increased. No definite patterns were detectable but the deviations in L.T.50's were mostly small (about 20%).

The level to which the dissolved oxygen concentration dropped naturally determined, to a much greater extent, the mortality rate of G. pulex and differences of only 0.5 $mgl^{-1}O_2$ had profound effects upon this rate. Comparisons of the L.T.50's for typical examples at both 10° and 20°C illustrated the exponential effect that 0.5 $mgl^{-1}O_2$ increments made, e.g.: -

10°C	Expt.	O_2 level	L.T.50(hr)	20°C	Expt.	O_2level	L.T.50
	1a	0.5mgl^{-1}	3.6		7a	0.5mgl^{-1}	2.05
	3a	1.0	58.0		9a	1.0	5.4
	5a	1.5	360*		11a	1.5	18.5

* the control L.T.50 that the experiment approximated to.

The difference in magnitude of the L.T.50's at 10°

and 20°C are immediately seen as clearly illustrating the role of temperature as a synergist to oxygen tolerance. The mortality curves for the above and the other experiments are seen in Figs. 6.8 - 6.11 where the effect of low oxygen level variance can be appreciated visually.

Response of G. pulex populations to the effects of temporal fluctuations are denoted by the three lines drawn on Figs. 6.6 and 6.7 corresponding to 12, 8 and 4 hr periods of low oxygen fluctuations. The shorter the periods of exposure to low oxygen levels the greater the survival time of the populations became. There was only one exception to this rule which was shown by the L.T. 50 of Expt. 10c (Table 6.3., Fig. 6.7). This exceptionally low and unexpected figure, when considered with the L.T.50 of Expt. 10b, was obviously due to experimental or mechanical error and while no errors were made in the selection, photoperiodic induction or handling procedure prior to the experiment, no abnormal diurnal traces for pH, temperature or oxygen were recorded during the experiment either.

Table showing the L.T.50's (hr) for G. pulex populations subjected to different temporal periods of exposure and three concentrations of dissolved oxygen(mgl^{-1}) at 10° and 20°C.

	10°C				20°C			
	period of exposure				period of exposure			
	12hr	8hr	4hr	low O_2 conc.	12hr	8hr	4hr	low O_2 conc.
Expt.	1a	1b	1c	0.5mgl^{-1}	7a	7b	7c	0.5mgl^{-1}
L.T.50	3.6	3.6	41.0		2.05	2.05	2.05	
Expt.	3a	3b	3c	1.0	9a	9b	9c	1.0
L.T.50	58.0	230.0	360*		5.4	8.4	35.0	
Expt.	5a	5b	5c	1.5	11a	11b	11c	1.5
L.T.50	360*	360*	360*		18.5	41.0	68.0	

* = control L.T.50 for 10°C.

Fig.6.8.Cumulative % mortality rate of control G.pulex populations(above) and, (below) populations subjected to 12hr $1.0mgl^{-1}$ O_2 and 12hr $10.0mgl^{-1}$ at 10^oC - i.e.Expt.4a.

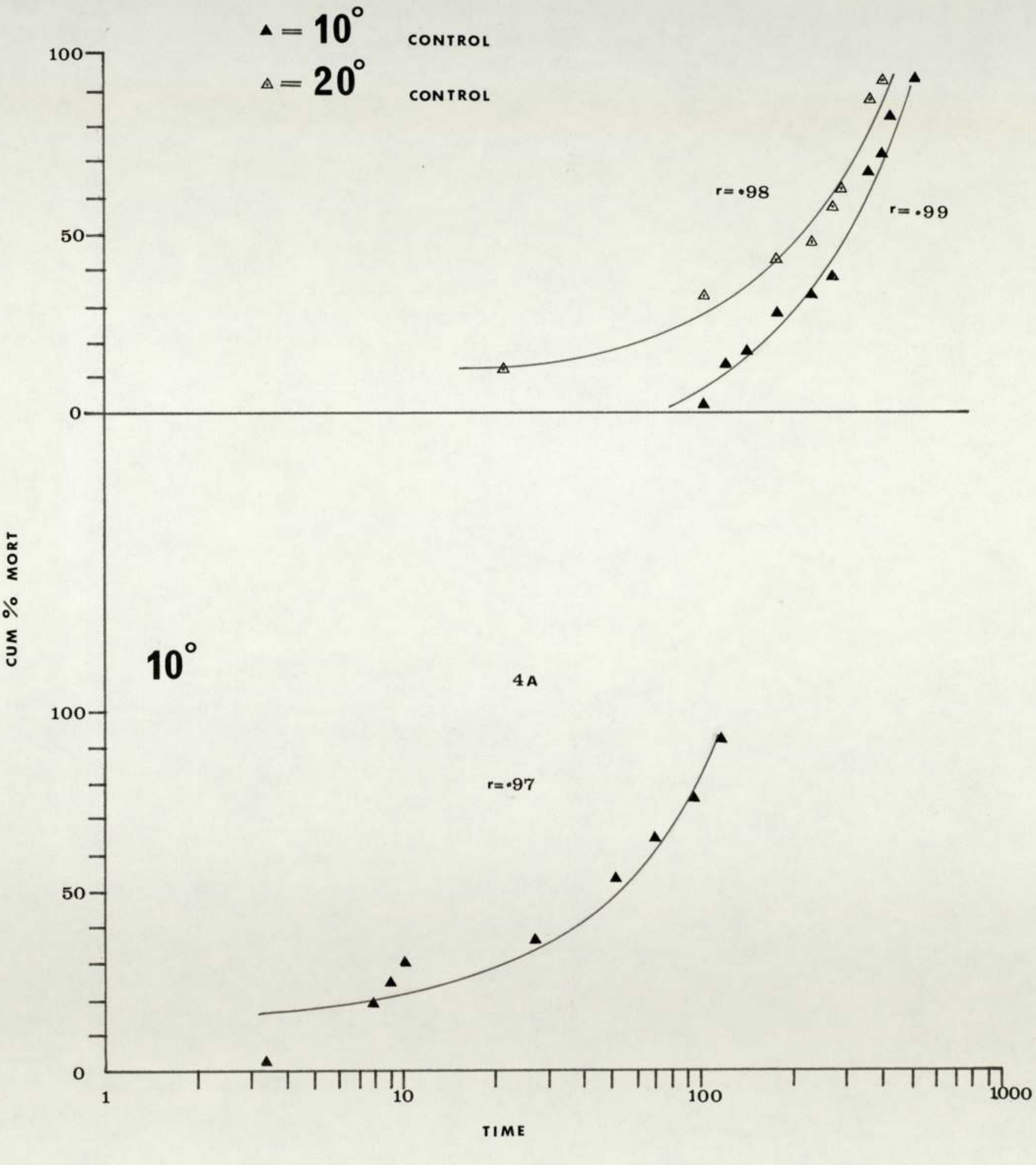


Fig.6.9. Cumulative % mortality rates of <u>G.pulex</u> populations.

1c= 4hr 0.5mgl^{-1} O_2 - 20hr 6.0mgl^{-1} O_2
2c= 4hr 0.5mgl^{-1} O_2 - 20hr 10.0mgl^{-1} O_2
3b= 8hr 1.0mgl^{-1} O_2 - 16hr 6.0mgl^{-1} O_2
1a= 12hr 0.5mgl^{-1} O_2 - 12hr 6.0mgl^{-1} O_2
3a= 12hr 1.0mgl^{-1} O_2 - 12hr 6.0mgl^{-1} O_2

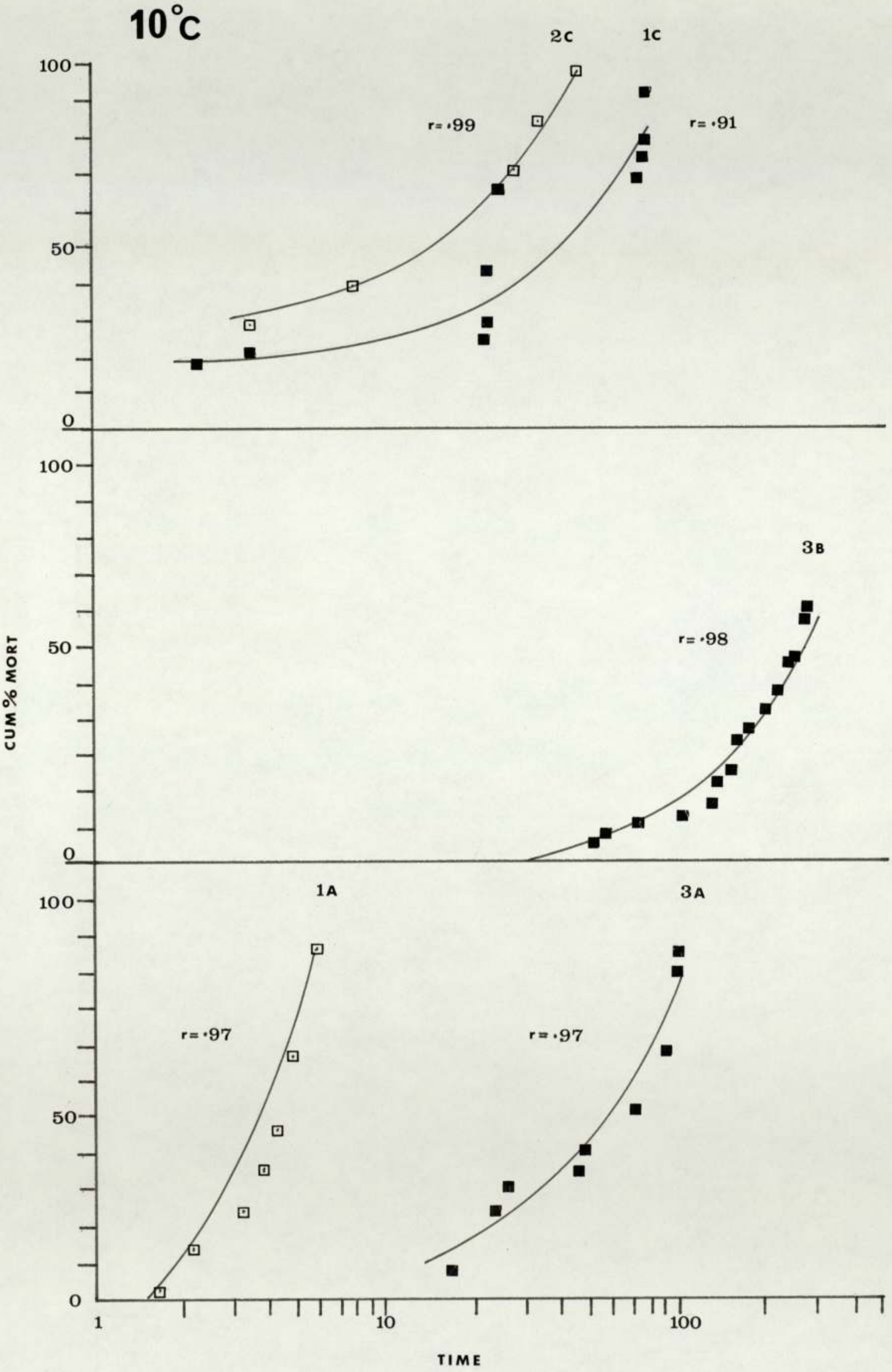


Fig.6.10.Cumulative % mortality rates of *G.pulex* populations at 20°C.

9c= 4hr 1.0mgl^{-1} O$_2$- 20hr 6.0mgl^{-1} O$_2$. 11c= 4hr 1.5mgl^{-1} O$_2$- 20hr 6.0mgl^{-1} O$_2$
9b= 8hr 1.0mgl^{-1} O$_2$- 16hr 6.0mgl^{-1} O$_2$. 11b= 8hr 1.5mgl^{-1} O$_2$- 16hr 6.0mgl^{-1} O$_2$
7a= 12hr 0.5mgl^{-1} O$_2$- 12hr 6.0mgl^{-1} O$_2$. 9a= 12hr 1.0mgl^{-1} O$_2$- 12hr 6.0mgl^{-1} O$_2$
11a= 12hr 1.5mgl^{-1} O$_2$ - 12hr 6.0mgl^{-1} O$_2$

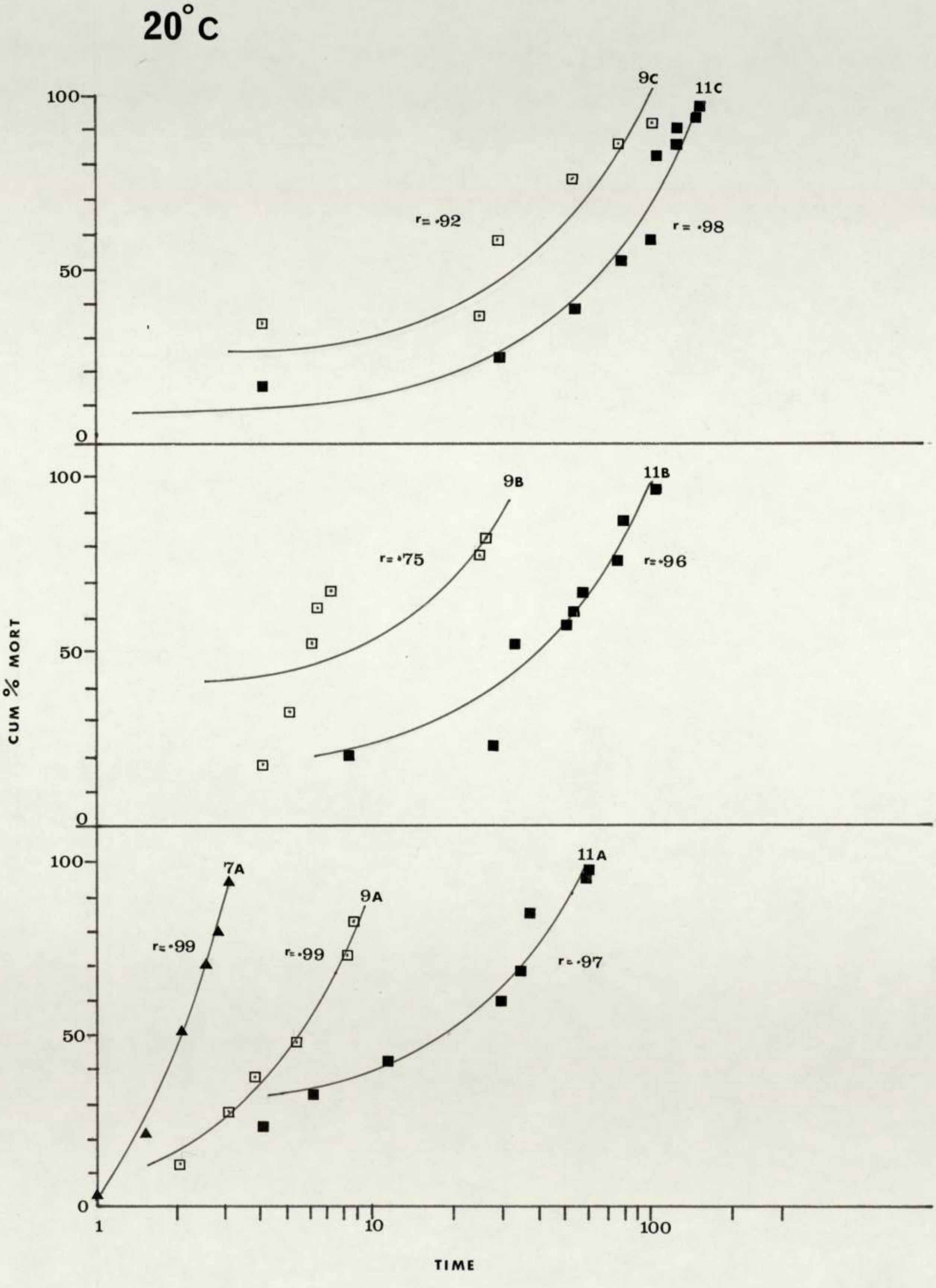
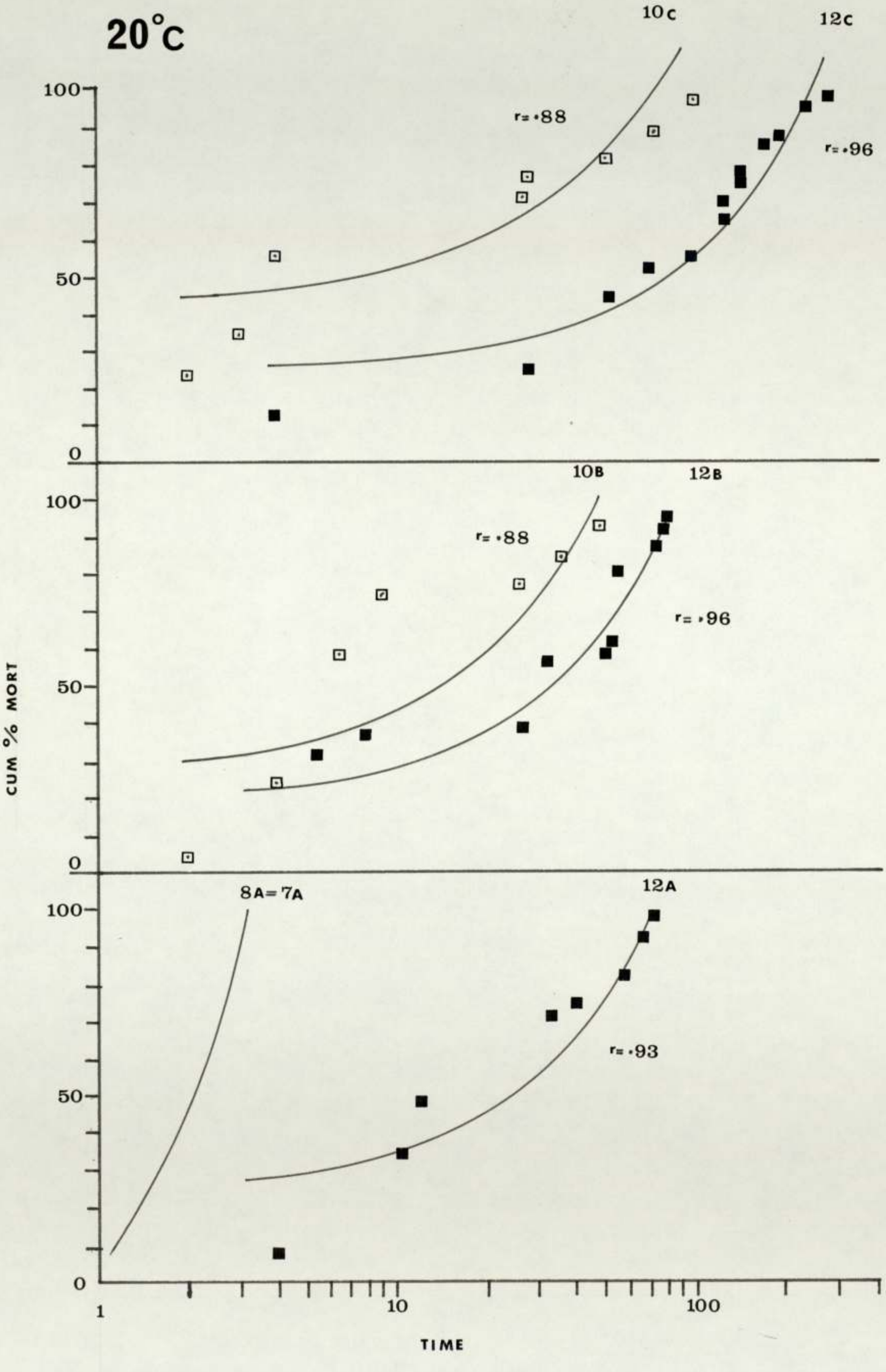


Fig.6.11.Cumulative % mortality rates of G.pulex populations at 20°C.
10c= 4hr 1.0mgl^{-1} O_2- 20hr 8.0mgl^{-1} O_2. 12c= 4hr 1.5mgl^{-1} O_2-20hr 8.0mgl^{-1} O_2
10b= 8hr 1.0mgl^{-1} O_2- 16hr 8.0mgl^{-1} O_2. 12b= 8hr 1.5mgl^{-1} O_2-16hr 8.0mgl^{-1} O_2
8a= 12hr 0.5mgl^{-1} O_2- 12hr 8.0mgl^{-1} O_2. 12a= 12hr 1.5mgl^{-1} O_2-12hr 8.0mgl^{-1} O_2



The L.T.50's from the above table demonstrate the rule relating to the temporal effects of exposure to low oxygen periods. Here also, as with low oxygen concentration, existed an exponential increase in survival times with the effect of increasing temperature clearly enhancing the toxic effects of reduced dissolved oxygen.

Although the responses of G. pulex to such experimental conditions cannot be compared with those expected from the oxygen-sensitive Plecoptera or the haemoglobin containing Chironomidae, it is a sensitive species which inhabits not only well oxygenated troutbecks but 'recovery zones' (Chapter 4) and eutrophicated rivers (Chapter 5). In this respect it is capable of living in a range of water qualities and was therefore potentially useful as a guide of invertebrate response to the experimental programme. Severe conditions, as administered in the experimental scheme, probably never occur in natural rivers and streams even when large amounts of allochthonous material are deposited in autumn. However, as diurnal patterns of this degree do arise in eutrophic and organically polluted lotic systems, the response of G. pulex is valuable from the autecological point of view and may help to explain not only its distribution under such conditions but the distributions of species often associated with it, e.g. B. rhodani. Nevertheless only from experimental trials with other species will the actual nature of the responses be elicited. Of substantial interest was the unexpected tolerance of G. pulex to very low oxygen concentrations, even though exposure was only for 8 or 4 hr periods.

6.6. CONCLUSIONS.

The effects of diurnal oxygen fluctuations on the amphipod G. pulex were observed and these results considered useful as a guide to the autecology of this animal in lotic eutrophic waters or watercourses displaying such oxygen fluctuations.

1. The temperature effect during the diurnal oxygen fluctuations was very marked: - the median survival times were decreased with an increase of 10°C, and were disproportionately increased with low oxygen increments of $0.5 mgl^{-1}$. i.e. for every $0.5\ mgl^{-1}O_2$ rise at 10°C the survival times were much greater than times for equivalent increments at 20°C; also, and similarly, L.T.50's were disproportionately increased with shortening exposure to low oxygen levels at 10° and 20°C.

It is suggested that the effect of only a few degrees rise about a mean of 10°C could thus restrict the distribution of G. pulex and other species in eutrophic rivers.

2. The mortality rate due to differences in fluctuations between low oxygen fluctuation and either a medium or high oxygen concentration were neither very important nor predictable. Sometimes mortalities were increased with higher oxygen deficits, and sometimes increased by lower oxygen deficits but deviations in the L.T.50's under these conditions were relatively small.

3. The low level to which the diurnal oxygen fluctuation dropped determined to a large extent the mortality rate of G. pulex. Differences of only $0.5\ mgl^{-1}O_2$ at these levels

had profound and exponential effects on the L.T.50.

4. The duration of the low diurnal oxygen fluctuation was of equal importance to survival times as were the absolute levels of low oxygen. The increases in survival time of G. pulex with shortening of the diurnal oxygen fluctuation were exponential.

5. The most important parameters affecting the survival time of G. pulex populations were therefore temperature, intensity and duration of oxygen fluctuation.

6. The apparatus proved successful in achieving and maintaining accurately the required environments for testing the effect of diurnal oxygen fluctuations on macro-invertebrate populations.

7. Further work with macro-invertebrates would be useful for estimating thresholds of tolerance to diurnal oxygen levels and consequently for attributing possible ecological significance to the range and distribution of species in eutrophic and organically enriched rivers.

CHAPTER 7.

RESPIROMETRIC STUDIES ON THE LARVAE OF SOME CHIRONOMIDAE (DIPTERA).

7.1. INTRODUCTION.

Respiratory studies are necessary for the understanding of the energy flow within populations. The amount of energy expended during respiration is a necessary pre-requisite for the calculation of production determinations figuring the measurements of consumption, absorption and assimilation. Energy losses by respiration can be simply measured by calculating the oxygen uptake and transforming it into an energy equivalent of food, by means of an oxy-calorific coefficient (the approximate mean for different energy sources used usually being 4.8 Cals/ml O_2 at N.T.P.) The ecological value of respiratory studies for deciding when oxygen pressures in water become limiting to the ranges of species are highly regarded by some authors (see Section 2.7, Chapter 2). The 'incipient limiting point' is often described as being autecologically significant in the animal's distribution; this point being reached when the oxygen level is just sufficient for the maintenance of the activities of the animal, the latter not being limited by the amount available.

Respiratory studies are useful, therefore, when concerned with the de-oxygenating effects of pollution. The amount of oxygen needed by an animal to maintain its growth, reproduction, etc., may explain its absence in water with consistently, or, indeed, only temporally low oxygen concentrations. In measurements of respiratory rate for the greater understanding of both energy flow and autecological distribution, it is necessary to realise the shortcomings of the methods used. Ideally, in situ measurements such as those of Odum (1957) and Knowles _et al_ (1962) would be the most desirous but difficulties encountered with respiration and photosynthesis of other organisms present prevent easy determinations. Consequently most workers have concentrated upon laboratory methods and their application to the real situation.

The importance of chironomid larvae as fish food make the understanding of chironomid population dynamics and energetics of fundamental importance. This, together with the notion that knowledge of the respiration rate with varying oxygen pressures of water may enhance the explanations for the chironomid distributions in the artificial streams, R. Tean, R. Cole, and general geographical ranges, led to an attempt to develop an apparatus capable of such measurements with sufficient accuracy. Previous attempts have been restricted; in the first instance because of taxonomic difficulties, and secondly due to an inability to overcome elementary problems of measurement, substratum and realism. Most work has been done with the haemoglobin-containing _Chironominae_ (Walshe, 1948; Fox and Taylor, 1955; Edwards, 1958b; Augenfeld, 1967).

After the apparatus had been developed and tested, determinations of the relationship between oxygen consumption and oxygen concentration of chironomid larvae were undertaken.

7.2. RESPIROMETRIC APPARATUS.

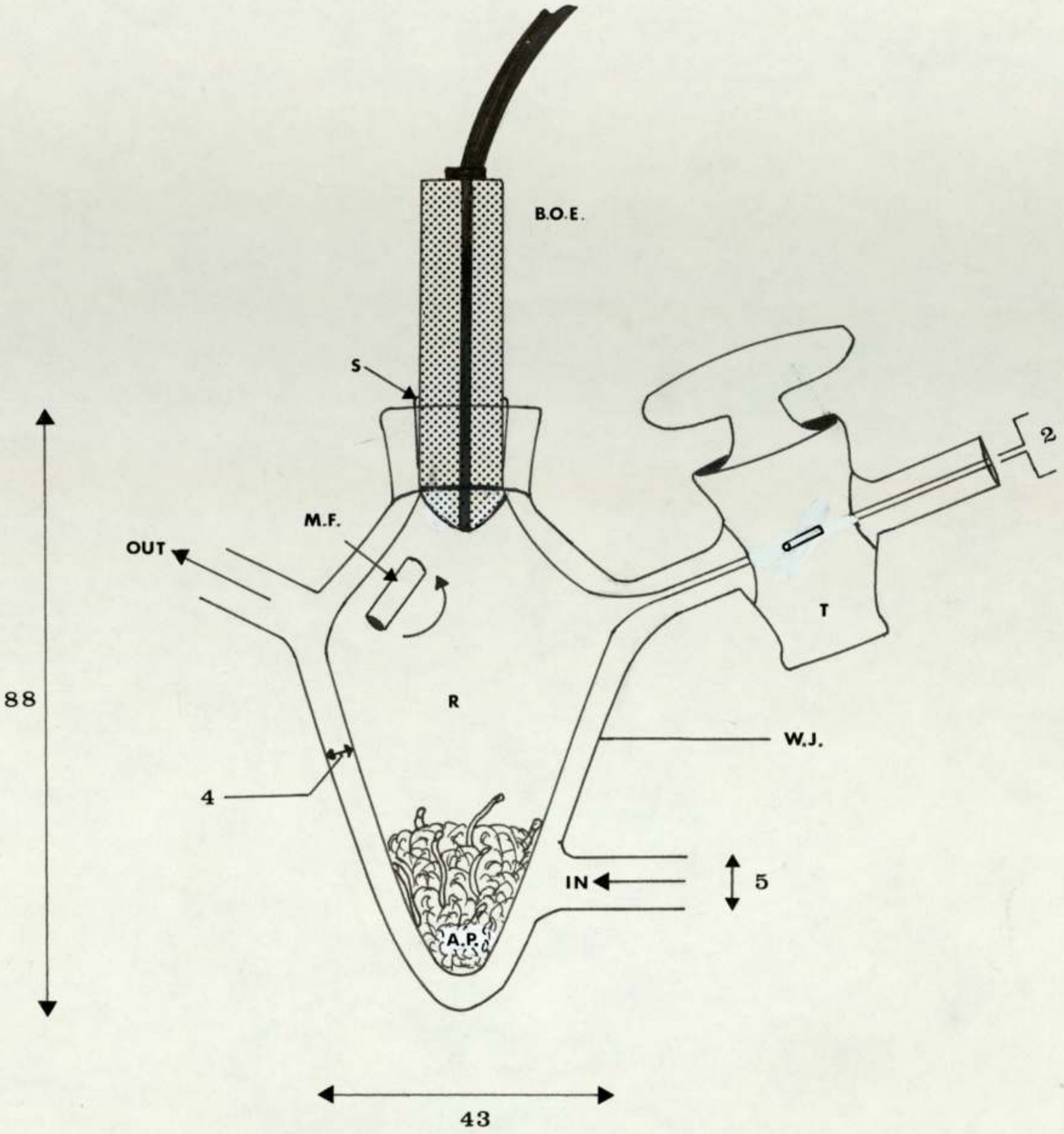
The methods for measuring oxygen in water continuously have been briefly mentioned in Chapter 2. Manometric methods were disregarded because a continuously running or stirred system was desired rather than the more artificial methods of shaking, which also require carbon dioxide absorption. The Scholander gasometric methods (1947, 1955) requiring the use of a multiple gas analyser were precluded. The choice was then between a membrane covered solid electrode, as described by Clark _et al_ (1953), and a dropping mercury electrode. The ready availability of a solid electrode together with an accurate electrometer with 'back off' facilities led to its utilisation.

The electrode was of the Bishop pattern(1958, 1960), marketed by E.I.L. (model number SOH33), and utilising a membrane 0.002" (0.005 cm.) thick. This gives an initial response time in the order of two to four minutes. Electrode systems of this kind are temperature, flow, and pressure sensitive which precluded the use of peristaltic pumps as a means of circulation of the respiratory water. A D48A oxygen accessory unit (E.I.L.) was used to relay a polarising voltage of 650 mv to the electrode, with the reduction of oxygen at the cathode producing a current proportional to the amount of oxygen dissolved. This current was passed via the unit to an E.I.L. 33B Vibron Electrometer. The current/partial pressure relationship was linear over the range 0.1 atmosphere that it was being used at. This allowed the use of two gases only to determine the relationship numerically. Air from a compressor and nitrogen from a cylinder were used for this purpose.

All laboratory methods suffer from the unnatural situation of the animals being constrained within a vessel, but considerable adjustments may be made to the chamber to alleviate some of the worst symptoms. The apparatus had to be transparent or translucent to allow photoperiods or certain light intensities of choice to be administered. Glass was the material used for the respirometer chamber. The final pattern chosen was one of pear shape (Plate II) which allowed stirring in the widest part, and inhabitation by chironomids in the tapering part. In this way the chironomids were not overstimulated by the effects of the circulation yet the oxygen tensions around them were kept constant. The larvae were supplied with an asbestos substratum; asbestos being chosen beacuse of its inert nature, its properties of support, no oxygen consumption, and its versatility. Augenfeld (1967) used this material successfully as a substratum for rearing chironomid larvae. The 'plug' of asbestos rested neatly in the tapered portion of the chamber, prevented from dislocation by the action of the current.

Stirring has the advantages of not producing

plate 11



Respirometer

All measurements in mm.

'in' and 'out' indicate the direction of cooling water.

A.P.= asbestos plug

B.O.E.= Bishop oxygen electrode

M.F.= magnetic stirrer

R.= respirometric chamber

S.= rubber seal

T.= tap

W.J.= water jacket

pressure effects as do peristaltic and reciprocating pumps and preventing the build up of dead space both around the electrode and around the animals. The animals receive a certain amount of stimulation from the stirring (which may be regulated) in an attempt to simulate the conditions under which the larvae naturally live, thus reducing the risk of recording the basal metabolism, which still bottle experiments like those of Mann (1961, 1956), Fox et al (1937) and Berg (1952, 1958) are likely to measure.

The chamber was blown to an approximate 25 ml volume and surrounded with a 4 mm wide water jacket, the former having a stopcocked 2 mm tube to allow displacement of water upon the entry of the electrode. The jacketing was necessary as the chamber was only partially submerged in a water bath in order to facilitate more control over the magnetic stirring device which was less sensitive when submerged in the bath. The thermostatically controlled and refrigerated bath was accurate to 0.001°C and all operations were performed in a constant temperature room at 10°C.

Control runs were made at all oxygen tensions in case of interference by use of oxygen at the electrode, bacterial respiration or epiphytic photosynthetic effects, and corrections were made when necessary. Barometric readings were taken daily for corrections to N.T.P. and calibration of the electrode system.

7.3 PROCEDURE.

Acclimation of animals to temperature (Sprague, 1963) oxygen levels (Teal, 1971) and photoperiods prior to experiments is necessary but not always possible; e.g. there is a likelihood of interference from instar change or emergence with time (Walshe, 1948). Also the ecological significance of acclimation should be understood. For example, industrial spills causing de-oxygenisation of temperature increases are without warning in nature. It seems better to measure respiration immediately after collection and later after acclimation to the new conditions to discover the nature of the short and long term acclimation effects. The rate recorded after collection or movement of the animals is inevitably elevated as their activity is increased; Newell and Northcroft (1967) demonstrated this phenomenon in marine Crustacea. Rates may also vary diurnally, seasonally, with water movement, illumination, water chemistry, instar size and age. Meticulous studies for production estimates should be designed to include these variables, but studies for autecological distribution trends need not be so elaborate.

Chironomid larvae were collected in a hand net, stored in polythene bottles and transferred to the laboratory as fast as possible. After sorting into the species required, groups of between 10 and 20 (depending upon size) of third and fourth instar stage were placed in 15 ml glass vessels containing a pre-weighted plug of asbestos, and subsequently covered with boulting cloth secured with rubber bands. Some of these were placed in a constant temperature room with adjusted photoperiod (to the environmental conditions at the time) at 10°C, and at the oxygen pressure of the experiment to follow. They acclimated to their new conditions for 24 hrs. before experimentation began.

The respiratory rates of these larvae were to be measured at different oxygen concentrations of filtered lake water (the same water as used in the fluctuating oxygen pressure experiments of Chapter 6). They were primarily subjected to a range of concentrations of oxygen: - 11.26, 5.64, 2.82, 1.5, 1.0, and 0.5 mgl^{-1}. Extra determinations were made to show more precisely where the inflexure in the curve of oxygen consumption vs. oxygen concentration occurred (if at all), i.e. 'the incipient limiting point'. The uptakes were measured at the highest oxygen concentrations first, the time for acclimation at the oxygen pressure of the next experiment being shown in Fig. 7.1 and Table 7.1. The experimental water was kept in a two litre beaker immersed in a water bath with the oxygen and carbon dioxide concentrations being controlled by the same apparatus used in the control of the oxygen fluctuation experiments (see 6.2.1.a, Chapter 6) - the desired oxygen tension being set upon the limit switch and a pH of 7.2 also being set upon a limit switch. Oxygen stripping was thus accomplished and maintained by diffusing carbon dioxide through the water, and pH maintained by diffusing carbon dioxide through the water. 100% saturation for calibration purposes was set upon the E.I.L. 15A oxygen meter twice a day after diffusing compressor air through the experimental water for at least 30 mins. and applying the equation: -

$$\% \text{ saturation} = \frac{P - v}{760 - v} \times 100$$

where P = barometric pressure; v = saturated water vapour pressure, which in this case was always 9.2 mmHg as all determinations of uptake were executed at 10°C. The Vibron 33B Electrometer was similarly calibrated in the experimental water using the 'back off' facilities to reach approximately 95% F.S.D. Winkler determinations were used as a check for both meters.

The animals were removed from their glass containers by lifting out the asbestos plug in which they were enmeshed, and placing them into the bottom of the respirometer. The latter was then filled slowly after fitting the electrode aperture with a narrow bore tube, submerging it in the experimental water, and controlling the output of water by the rate of air released from the side-arm. Hence the turbulence of filling was minimised; of particular importance at low oxygen pressures. The polythene covered magnetic stirrer was added followed by the insertion of the solid electrode. The electrode, and the respirometric chamber were kept at the experimental temperature for over one hour beforehand as the output of the electrode is temperature sensitive. The stopcock of the side-arm was closed and the chamber was clamped into position in the bath, aligning the stirrer and connecting up the water jacket.

Previously the meter scale had been adjusted to approximately 95% of the F.S.D., at the experimental oxygen pressure and only fine adjustments were needed to compensate for heat transmitted during handling of the electrode, chamber and stirrer. The uptake was recorded on a continuous chart and when the reading was seen to be steady (usually 15 - 45 mins) the consumption was measured over a one hour period. The 'back off' facilities of the electrometer allowed the expansion of the scale resulting in very accurate measurement. The calibration of the meter was re-checked (from zero to F.S.D. - 0-30 mv) at the end of each experiment.

Technical and seasonal considerations had restricted the amount of time available for determinations to be made with the consequence that after the testing of the apparatus, relatively few determinations of uptake could be made. Recollecting the variations inherent when dealing with whole organism respiration as being in the order of 5 - 10% (Teal, 1971) together with errors due to measurement, artificial environments, and differences between weight (Edwards, 1958b) within an instar stage, a substantial number of determinations are required. Errors were reduced by using the same sample groups during at least five oxygen pressures always in order of decreasing pressure, with a final check by duplicity of the 5.64 mgl^{-1} uptake. As the oxygen concentration in water changes according to its ionic content, the same sample of experimental water was used throughout.

7.4 RESULTS.

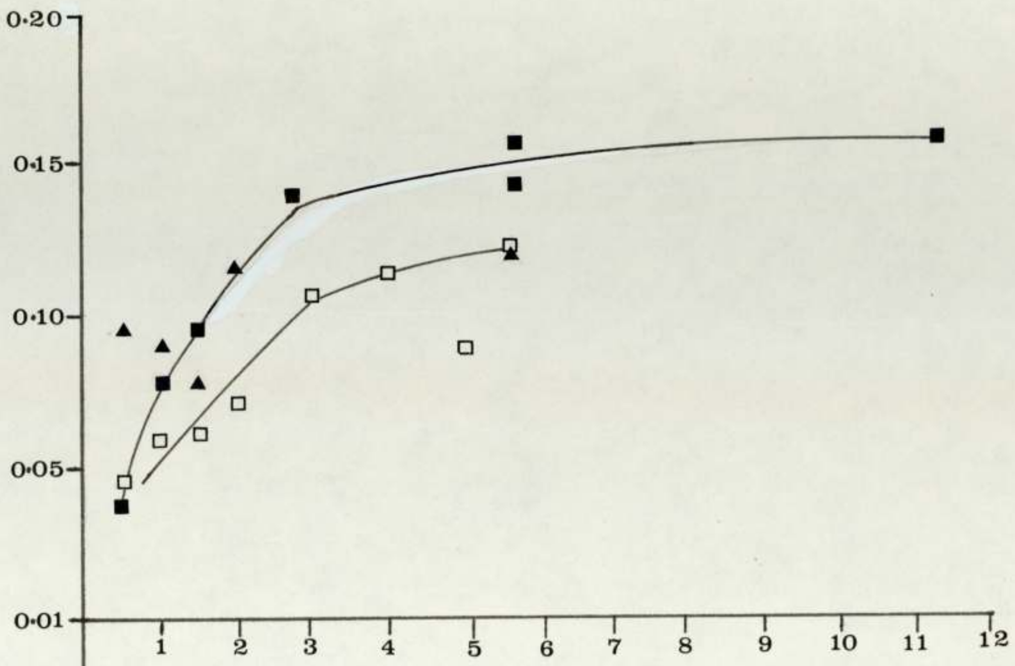
The uptake was expressed in concentration of oxygen (mgl^{-1}) per mg. dry wt. per hour.

$$q = \frac{(t_1 - t_2)}{w} = mgl^{-1} O_2 \; hr^{-1} \; mg \; dry \; wt^{-1}.$$

where; w = dry wt in mg; t_1 = initial units of oxygen (mgl^{-1}), t_2 = units after 1 hr.

Owing to the difficulty of inserting the electrode to the same depth into the chamber every experiment, measuring the relatively large volume of the instars accurately, and the inevitable loss of some fragments of the asbestos plug that adhere to the sides of the glass vessels after transference to the respirometric chamber, volometric determinations would have been slightly inaccurate. To overcome this, the expression of oxygen uptake that was utilised, as in Fig. 7.1., was $mgl^{-1} O_2$ mg dry wt^{-1} hr^{-1}. These units, although not usually applied to respiratory uptake, are independent to the measurement of the 'incipient limiting point' as it is the inflexure of the curve against oxygen concentration that is important. The method of calibrating

fig 7.1 The relationship between oxygen uptake and oxygen concentration in <u>Prodiamesa olivacea</u>.



■ 24hr.acclimation. 20min.acclimation between O_2 reductions. $\bar{X}$wt=0.42, 0.55mg

□ ditto 1hr.acclimation between O_2 reductions. $\bar{X}$wt=0.82, 1.19mg

▲ ditto no acclimation between O_2 reductions. $\bar{X}$wt=0.63mg.

fig 7.2 The relationship between oxygen uptake and oxygen concentration in <u>Micropsectra atrofasciatus</u>.

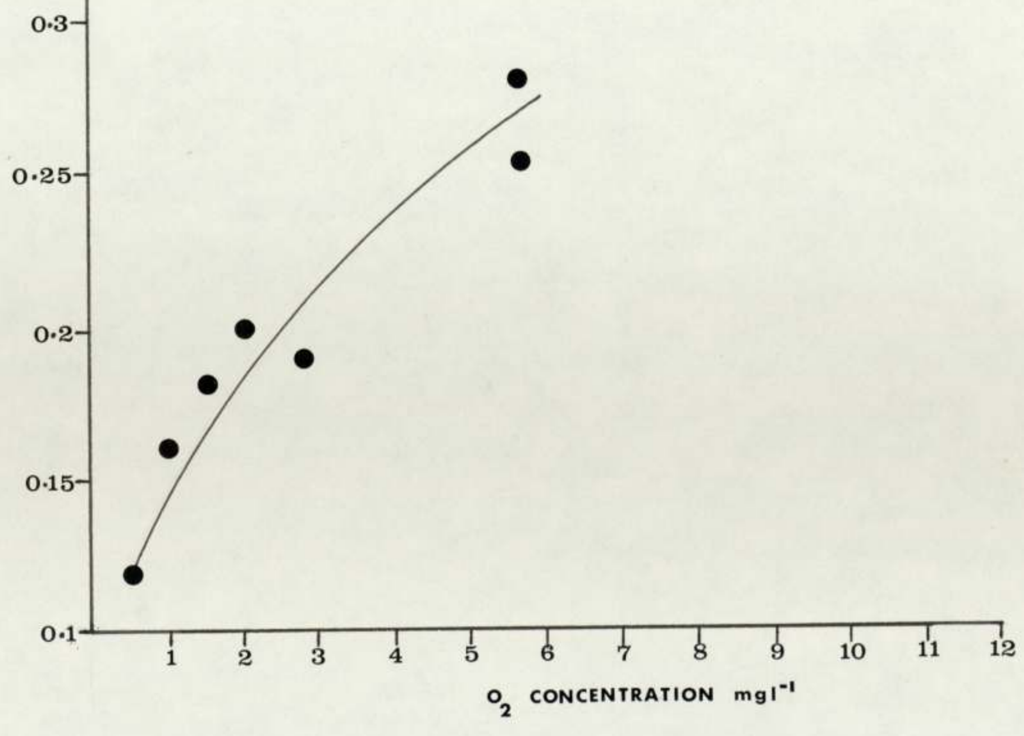


TABLE 7.1. SHOWING RESULTS OF THE RESPIRATION OF P. OLIVACEA AT DIFFERING OXYGEN TENSIONS.

Prodiamesa olivacea 3rd and 4th instars 24 hr acclimation. 20 min. between O_2 reductions.

Date	Number	$0^{o}C$.	mmHg	O_2 tension mgl^{-1}	uptake mgl^{-1} hr^{-1}	dry wt mg.	uptake $mgl^{-1}hr^{-1}mg$ dry wt^{-1}	$\bar{X}$ dry wt mg per animal.
12/6/74	15	10	759.5	11.26	1.02	6.4	0.159	0.43
12/6/74	15	10	759.5	5.64	0.93	6.4	0.145	0.43
12/6/74	15	10	759.5	2.82	0.91	6.4	0.142	0.43

Prodiamesa olivacea 3rd and 4th instars 24 hr acclimation. 20 min. between O_2 reductions.

13/6/74	14	10	757.8	1.5	0.737	7.7	0.096	0.55
13/6/74	14	10	757.8	1.0	0.661	7.7	0.079	0.55
13/6/74	14	10	757.8	0.5	0.293	7.7	0.038	0.55
13/6/74	14	10	757.8	5.64	1.28	7.7	0.116	0.55

Prodiamesa olivacea 3rd and 4th instars 24 hr acclimation. 20 mins (*=1hr) between O_2 reductions.

19/6/74	12	10	753.9	5.64	0.915	7.6	0.120	0.63
19/6/74	12	10	753.9	2.0	0.875	7.6	0.115	0.63
19/6/74	12	10	753.9	1.5	0.605	7.6	0.079	0.63
19/6/74	12	10	753.9	1.0	0.69	7.6	0.091	0.63
19/6/74	12	10	753.9	0.5	0.75	7.6	0.098	0.63
19/6/74	10	10	753.9	5.64	1.48	11.9	0.124*	1.19
19/6/74	10	10	753.9	2.0	0.82	11.9	0.069*	1.19
19/6/74	10	10	753.9	1.5	0.076	11.9	0.064*	1.19
20/6/74	10	10	755.7	1.0	0.725	11.9	0.061*	1.19
20/6/74	10	10	755.7	0.5	0.59	11.9	0.049*	1.19

Prodiamesa olivacea 3rd and 4th instars 24 hr acclimation. 1 hr between oxygen reductions.

20/6/74	11	10	755.7	5.0	0.79	9.1	0.087	0.83
20/6/74	11	10	755.7	4.0	1.03	9.1	0.113	0.83
20/6/74	11	10	755.7	3.0	0.97	9.1	0.107	0.83

continued ......

TABLE 7.1. (continued)

Micropsectra atrofasciatus 3rd and 4th instars 24 hr. acclimation. None between O_2 reductions.

Date	Number	0^{o}C.	mmHg	O_2 tension mgl^{-1}	uptake mgl^{-1} hr^{-1}	dry wt mg.	uptake $mgl^{-1}hr^{-1}mg$ dry wt^{-1}	$\bar{X}$ dry wt mg per animal
17/6/74	21	10	757.8	5.64	0.78	3.0	0.26	0.014
17/6/74	21	10	757.8	2.82	0.57	3.0	0.19	0.014
17/6/74	21	10	757.8	1.5	0.56	3.0	0.186	0.014
17/6/74	21	10	757.8	1.0	0.48	3.0	0.16	0.014
17/6/74	21	10	757.8	0.5	0.36	3.0	0.12	0.014
19/6/74	17	10	753.9	5.64	0.57	2.0	0.283	0.117
19/6/74	17	10	753.9	2.0	0.41	2.0	0.20	0.117

the electrode, accounting for changing barometric pressure and saturated water vapour pressure, keeps the units of concentration between experiments comparable.

7.5 DISCUSSION.

The plots from the graph of uptake vs. oxygen concentration (Fig. 7.1) indicate that two curves may possibly be drawn, the distinguishing feature between the two apparently being that of instar size. It has long been recognised that the rates of oxygen consumption are generally lower in larger animals of the same or closely related species and recent studies have confirmed an inverse relationship between metabolic rate and body size. The relationship is not linear; rather the rate of oxygen consumption is correlated with body weight as an exponential function, $\text{ml}O_2$ consumed = kW^x where k is the species constant, W is body weight and ^{x} is an exponent ranging from 0.66 to unity. Edwards (1958b) has demonstrated the relationship between body weight and uptake for the chironomid C. riparius.

The lower curve (Fig. 7.1) illustrates the synthesis of uptakes by the heavier instars ($\bar{X}$ wt 0.82 and 1.19 mg/individual) and the upper the result of greater respiration by the less heavy instars ($\bar{X}$ wt 0.42 and 0.55 mg/individual) which is in accordance with the surface law. The uptakes of those larvae whose mean weight (0.63) lies between these two extremes, and indicated by the symbol Δ, are seen to be scattered between the two lines. This scatter may have been amplified by the lack of acclimation to oxygen concentration during the reduction of the partial pressure between experiments. Similarly, the differences between acclimation times to oxygen concentration could have resulted in a higher respiratory rate in those less acclimated (top curve) as opposed to those acclimated for longer (bottom curve). Nevertheless, the importance of the points lies in the position where the inflexure straightens out with respect to the abscissa, i.e. the incipient limiting point. Regardless of weight or acclimation then, this point appears to occur at about $3mgl^{-1}$ of oxygen.

This figure in some cases can be used to show how the species is adapted to living in certain environmental conditions. P. olivacea therefore appears to have an independent type of relationship with oxygen concentration, i.e. it can maintain its normal respiratory rate until approximately $3mgl^{-1}$ when the rate is actively reduced as a survival mechanism. However, depressing the metabolic rate for long periods will almost certainly have an adverse effect upon the survival of the individual, and, with fecundity reduction inevitable, survival of the population. P. olivacea possesses no haemoglobin or other respiratory pigments hence it is most probable that the relationship is independent, with its feeding activities being depressed for metabolic reasons below the incipient limiting point. It would appear, therefore, that P. olivacea is capable of maintaining its metabolic rate down to a level of oxygen usually only encountered in polluted milieu.

The ecological significance of this independent relationship and the incipient limiting point is demonstrated when the results of Davies (1971) and the author (Chapter 4) are considered. Davies showed that P. olivacea was most abundant at stations 4 and 6 on the R. Cole where pollution was fairly severe, but less severe than at station 2 where the dissolved oxygen often dropped to 1 mgl^{-1} (and lower at night).

P. OLIVACEA OCCURRENCE IN THE R. COLE 1966-1967.

Adapted from Davies (1971).

Station	Nos./$0.01m^2$/yr	Dissolved oxygen
2	0	below $2mgl^{-1}$ for 4 mths of year
4	350	$2mgl^{-1}$ for only part of 1 mth.
6	400	never dropped below $4mgl^{-1}$.

P. olivacea were absent at Station 2 and his conclusions were, in general, the degree of tolerance to organic pollution shown by P. olivacea was similar to E. testacea, H. stagnalis and Brillia longifurca. Mann (1956, 1961) showed the inflexion in the uptake vs. oxygen pressure relationship for E. testacea and H. stagnalis to occur at at 2.5mgl^{-1} and 4.0 - 2.0mgl^{-1} respectively, which compares very favourably with the 3mgl^{-1} figure for P. olivacea. The authors work on the recovery of the R. Cole demonstrates habitation of all the stations 1 - 6 by P. olivacea in 1972-1973, it being recorded in similar numbers at Station 2 (now averaging 8.89 mgl^{-1} D.O.) as at stations 4 and 6.

The number of determinations of oxygen consumption of Micropsectra atrofasciatus at differing oxygen concentrations are few, partly because the required instar size could not be collected in great enough quantity, and partly because time for experimentation would have run out before the next generation of larvae could have developed. Therefore nothing conclusive about the curve in Fig. 7.2 can be elicited, but the preliminary results point to a smooth curve, which may indicate a dependent relationship from at least 6 mgl^{-1} O_2 to 0.5 mgl^{-1} O_2. If the relationship of oxygen uptake to oxygen concentration is independent, the 'incipient limiting point' may be a high one, or, even if completely dependent, suggestive that the species might be sensitive to reduction in oxygen pressures. Observations in the Experimental Streams show this species to be most abundant in Stream 2 (100% river water, mean D.O. 11.6 mgl^{-1}) followed by Stream 1 (37% effluent, mean D.O. 9.8 mgl^{-1}) and least abundant in Stream 3 (87% effluent, mean D.O. 8.76 mgl^{-1}). Also, M. atrofasciatus only became dominant in the benthic macro-fauna of the R. Cole in the recovery stages and after the cessation of the effluent, and therefore field observations also point to a pollution sensitive species.

7.6. CONCLUSIONS.

1. From preliminary results, the respirometric apparatus developed for the measurement of oxygen uptake by chironomid larvae has proved successful.

2. The advantages of this apparatus over some others lay in the elimination of basal metabolism alone being measured by virtue of the stirring action which subsequently omitted 'dead spaces'. The use of an asbestos plug as an inert support medium was useful in expediting the transference of the larvae to the respirometric chamber with the minimum disturbance.

3. From the number of determinations of oxygen uptake of Prodiamesa olivacea at different oxygen concentrations an 'incipient limiting point' appeared at about 3.0 mgl^{-1} O_2. This figure was shown to be apposite to its autecology.

4. Nothing conclusive about the relationship between oxygen uptake and oxygen concentration for Micropsectra atrofasciatus could be elicited from the restricted number of determinations although a 'dependent' type relationship may be indicated, at least between 6.0 mgl^{-1} and 0.5 mgl^{-1} O_2. Field observations also suggest that this species is pollution sensitive.

5. It is suggested, therefore, that the experimental 'incipient limiting point' figure can be a valuable measure,

substantiable with field observations of the situation.

6. Determinations of this brevity are only sufficient to show the type of relationship the animal displays with varying oxygen concentrations. Measurements of rates of uptake in other seasons would add weight to the conclusions and detect any variations in response.

CHAPTER 8.

SOME OBSERVATIONS ON THE POPULATIONS OF ASELLUS AQUATICUS IN THE R. RAY (WILTSHIRE) AND EXPERIMENTAL STREAM 1.

8.1. INTRODUCTION.

The highly eutrophicated state of the R. Ray resulting from the Rodbourne S.D.W. effluent encourages large growths of the blanket weed Cladophora and the mosses Leptodyction and Fontinalis. Urquhart (1972, personal communication) has recorded up to 218 g/m^2 of Cladophora and 51 g/m^2 of mosses at Moredon Bridge during the late spring and summer of 1972. Asellus aquaticus is typically associated with large growths of Cladophora and mosses as these form excellent media for support while trapping substantial amounts of detrital material as food. Also, Asellus is known to feed on the Cladophora filaments (see Chapter 5, Section 5.5.c.). It was considered feasible that the large diurnal fluctuations in oxygen/carbon dioxide which accompanies excessive growths of these weeds might modify the rate of development, production and perhaps the timing of the reproduction of animals living under such conditions. These assumptions were made knowing that metabolism, and therefore growth, fecundity and possibly production, is affected by the amount of oxygen available. Some night time determinations of oxygen concentration at Moredon Bridge are shown in Chapter 6, Table 6.1., and while these may be in the region of 3 mgl^{-1}, the actual concentration in the matrix of filaments might be expected to be much lower (see Section 6.1).

It was proposed, therefore, to conduct a survey of the reproductive biology and standing biomass of A. aquaticus populations in the R. Ray at Moredon Bridge and in Experimental Stream 1 for comparative purposes, the latter being less eutrophicated and possessing high diurnal levels of oxygen (Fig. 5.6). A survey of the Asellus populations in the Ray was conducted from October 1971 to April 1973 with determinations of density, biomass and length/frequency analyses being made, but a similar survey of Experimental Stream 1 was not possible due to the slow colonisation of the animal (Section 5.5.c.). Only standing biomass and density were recorded in the Experimental Stream and as the populations did not appear to have reached a self-regulating level, i.e. they were still rapidly expanding (Fig. 5.16), the results were not comparable with those from the R. Ray.

The results of the length/frequency analyses and biomass determinations on the Asellus populations of the Ray have therefore been included in a small chapter.
The methods used in the analyses have been described in Chapter 3.

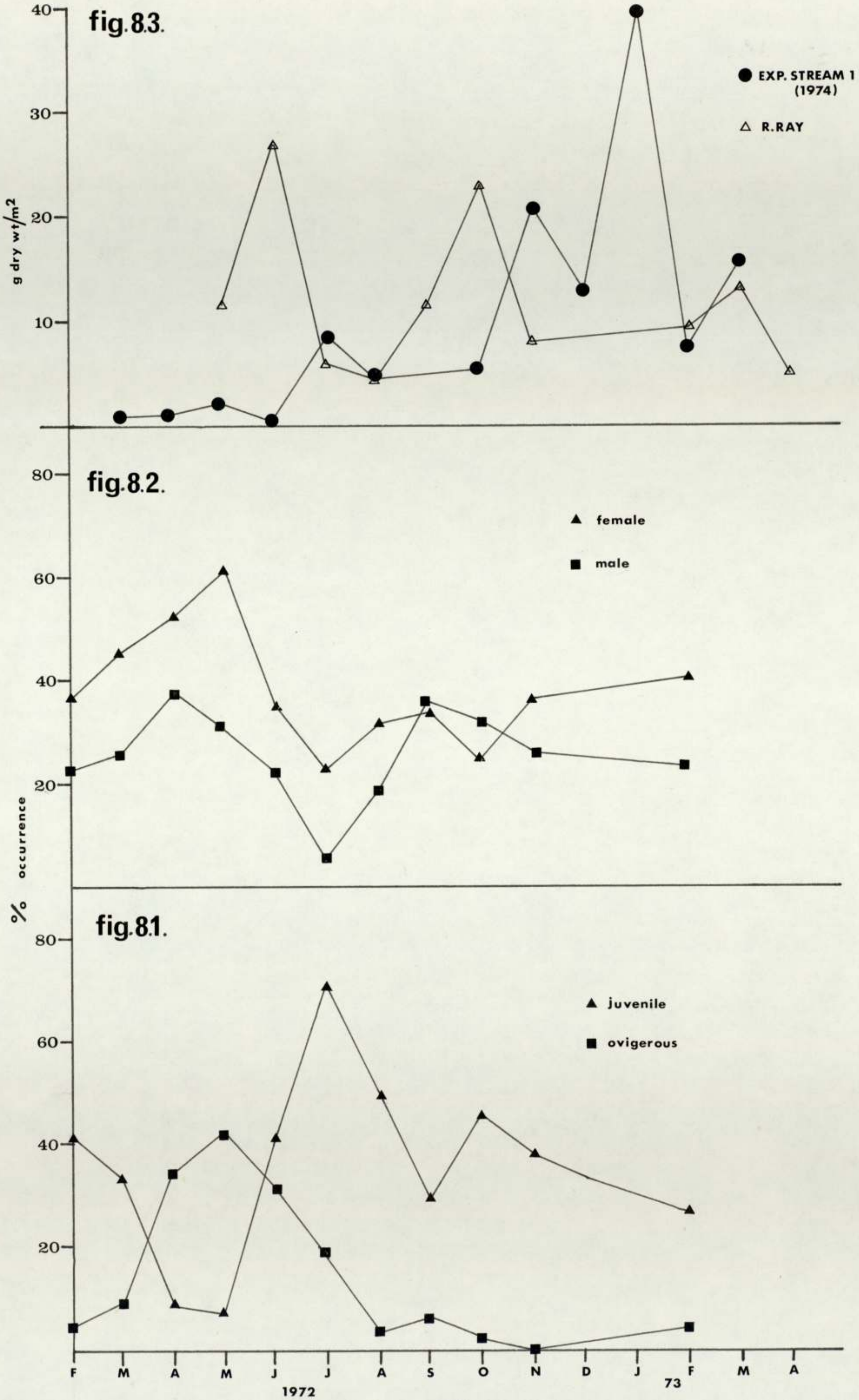
The timing of breeding, sizes of broods and other aspects of the life history of Asellus aquaticus have therefore been compared, as far as possible to that ascertained by Steel (1961) for a population of Asellus in the R. Thames at Reading. Little is known about the water quality of the Thames at Reading, but in 1958 it is fairly certain that it was not as nutrient rich as the Ray in 1971 - 1973 and, being some 50 to 80m wide and up to 4m deep there was probably little Cladophora.

8.2. LIFE HISTORY.

This was ascertained for the R. Ray population using length/frequency analyses with range classes of 1mm distinctions and samples ranging between 200 and 500 individuals. The majority of samples came from the Cladophora blankets which virtually covered the river bed in the growing season. The histograms of the results (Fig. 8.6) are, for the most part, similar to those of Steel. Fig. 8.1 shows that breeding begins around February and finishes in late autumn, with one large peak occurring in spring and a smaller one in September, which, for all intents and purposes may be referred to as the spring and autumn broods. With copulation starting in February, the juvenile population is seen to greatly increase some two months afterwards, reaching a peak in July. At 4 mm they are easily distinguishable into sexes. The second or autumn peak is seen resulting from the September increase in ovigerous females. Fig. 8.2 shows a predominance of the female population with the exception of the period from August to October when the proportions are approximately the same. It appears, however, that the males die after copulation in spring, and increase from July (when the sexes are distinguishable) at a greater rate than the females. The increase is not due to an unequal sex ratio recruitment but to the females dying after brooding. Indeed, a sex ratio of 1:1 is demonstrated in September when four months after the spring peak the juveniles have reached a sexually differential state.

Steel also found the breeding season began February, after a break from October, when no ovigerous females were found. The older and thus longer females brooded first, with younger ones breeding as the season progressed. In the Ray, breeding was still occurring in October 1971 with 16.2% ovigerous females and October 1972 with 11.7% ovigerous females (expressed as percentage total females) and 2.6% in January 1973.

Seasonal fluctuations in the structure (Figs 8.1. and 8.2.) and biomass (Fig 8.3.) of the A.aquaticus population in the R.Ray, and in the biomass of the A.aquaticus population in Experimental Stream 1.



The table below (1) shows how the peaks of breeding differ by approximately one month, the spring peak in the eutrophic Ray occurring in May whilst the peak in the Thames occurred in April. Both rivers show the marked secondary breeding in September (see also Fig. 8.1 showing September ovigerous females and subsequent juvenile recruitment in October).

Table 1. showing ovigerous females as a percentage of total females seasonally in the River Ray and Thames.

	Thames (Steel, 1961)		Ray
Oct. '58	0	1971	16
Nov.	0		–
Dec.	0		–
Feb. '59	11	1972	4.6
Mar.	49		9
Apr.	67		35
May	44		42
June	54		31
July	30		17
Aug.	38		11
Sept.	58		20
Oct.	5		11
Nov.	–		0
Jan.	–	1973	3

The table on the next page (2) demonstrates a difference in the lengths of the smallest breeding females from March onwards in the R. Thames and R. Ray: -

Table 2. showing the smallest breeding females (in mm.) in the R. Thames and R. Ray in their respective years.

	Thames (Steel, 1961)		Ray
Feb. '59	6.0	1972	6.0
Mar.	6.0		4.0
Apr.	5.5		4.0
May	5.0		4.0
June	4.0		4.0
July	4.0		4.0
Aug.	3.5		4.0
Sept.	3.5		4.0
Oct.	-		4.0
Nov.	-		-
Jan.	-	1973	5.0

In the Thames, sexual maturity of the autumn brood is reached when the animal is 6 mm long, while in the Ray the animal is only 4 mm long. It is not until June that 4 mm ovigerous females appear in the Thames population, this being the result of the faster attainment of sexual maturity of the spring brood. It is possible that the oxygen concentrations, at night in particular, from September to February are low enough to depress the rate of growth, and whilst temperature and availability of food could also have the same effect, the Rodbourne S.D.W. 2.1 km. upstream of the sampling area maintains elevated temperatures and food supplies all the year round. No figures relating to temperature are given for the R. Thames but it is thought unlikely that the temperature or food supply would have been greater, especially as the effluent from Rodbourne accounts for a major part of the Ray's flow - and almost all in summer.

Another feature of the Ray Asellus population is the smaller percentage of ovigerous females (expressed as a percentage of total females). The peak of the population's spring breeding in the Ray is only 62% of the Thames' equivalent. Similarly, the number of eggs per brood in the Asellus population from the Ray are reduced compared with the number in a similar range class for the Thames population. Only one range class was analysed for the Ray population (5 - 6 mm) and whether the difference is significant is unknown as Steel only gives the mean and range: -

Table 3. showing the number of eggs and range per brood in the Asellus populations of the Ray and Thames.

	Thames				Ray		
Size mm.	N	$\bar{X}$	Range	Range Class mm.	N	$\bar{X}$	Range
5.0	68	30	54	5 - 6	14	25	35
5.5	30	40	51				
6.0	18	43	79				

Observations on the size of the juvenile when released from the brood pouch were the same as those from the Thames population, i.e. about 1 mm. in length.

Table 4. showing the length of young released from the brood pouch - from the Asellus population of the Ray.

Female length (mm)	No. of young	Size of young
5.5	18	0.8, 0.9 1.0x2, 1.1x7, 1.2x2, 1.3x3, 1.4x2.
4.8	13	0.7, 0.8, 0.9, 1.0x6, 1.1x4.
5.4	16	0.8x2, 0.9x4, 1.0x8, 1.1x2.

Differences in timing of the spring brood (Table 1) may be related to temperature, although while the Ray is probably warmer and more likely approximating to environmental temperatures, the breeding peaks occur later than in the Thames populations. The growths of Cladophora are continually enlarging in the R. Ray from April onwards and peak around June and July, so while this alga is such a good medium for Asellus to inhabit, the timing of the broods may be delayed until the Cladophora blankets are large. Alternatively the Cladophora habitat may bias the percentage occurrence in favour of the ovigerous females, the filaments seemingly supplying protection for the young. Also, the length/frequency histograms (Fig. 8.6) all show a degree of skew to the left, particularly during the Cladophora season. This is to be expected as it is also the breeding season, but even over winter when few juveniles are being recruited in the R. Ray (none in the Thames) the frequency distribution is weighted on the left. The larger individuals on the right may therefore avoid Cladophora as a habitat, and, as the larger individuals are nearly all males, would explain the imbalance in the sex ratios demonstrated in Fig. 8.2.

8.3. STANDING BIOMASS.

The data from Table 5 is shown graphically - Figs. 8.3 and 8.4. Similar data for the period 22.3.73 - 13.3.74 for the Asellus population in Experimental Stream 1 is shown in Tables 5.12 and 5.13 and again graphically (Figs. 8.3 and 8.4.) after being transformed to mean density per m^2 and mean dry weight per m^2. Fig. 8.3 demonstrates the expected fluctuations in biomass (dry wt.) appropriate to the life history of the Ray population. The weight is greatest in March when the overwintering population is rapidly expanding in size and breeding. There is a decrease in weight and numbers (Fig. 8.4) soon after as the heavier males die after copulation and the females die after brooding (March - October). Numbers increase again during July as the young appear and as these grow rapidly to sexual maturity the weight also increases after a lag, i.e. during August, reaching another peak in October. More males and females from the previous cohort and those of the spring brood die after the autumn brood reducing the numbers and weight accordingly.

The Asellus population in Experimental Stream 1, however, is difficult to characterise as it is a young and continually expanding population with no great pressures from predation. Figs. 8.3 and 8.4 show the very fast

Fig.8.4 Seasonal fluctuations in density of the A.aquaticus populations in the R.Ray 1972-3 and Experimental Stream 1 1973-4.(Thin line)

Fig.8.5 Calorific values of A.aquaticus range classes.

fig.8.4.

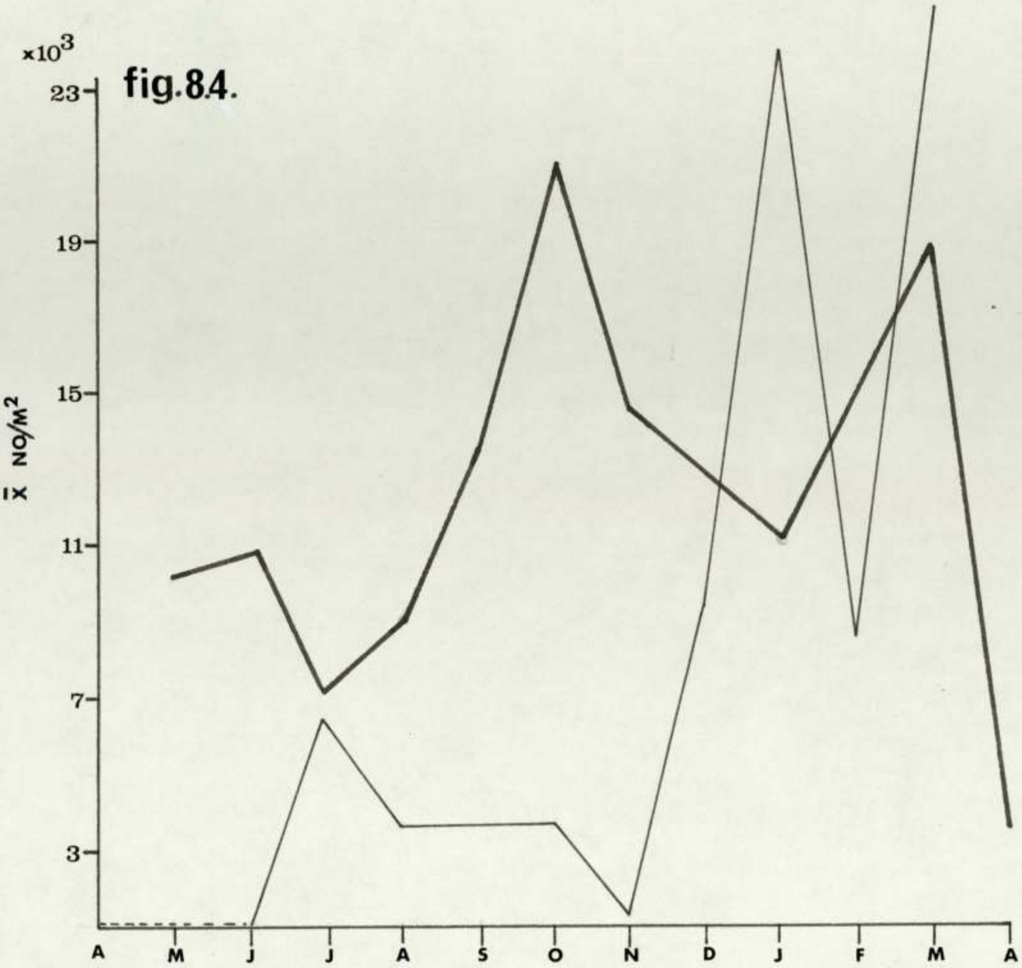


fig.8.5.

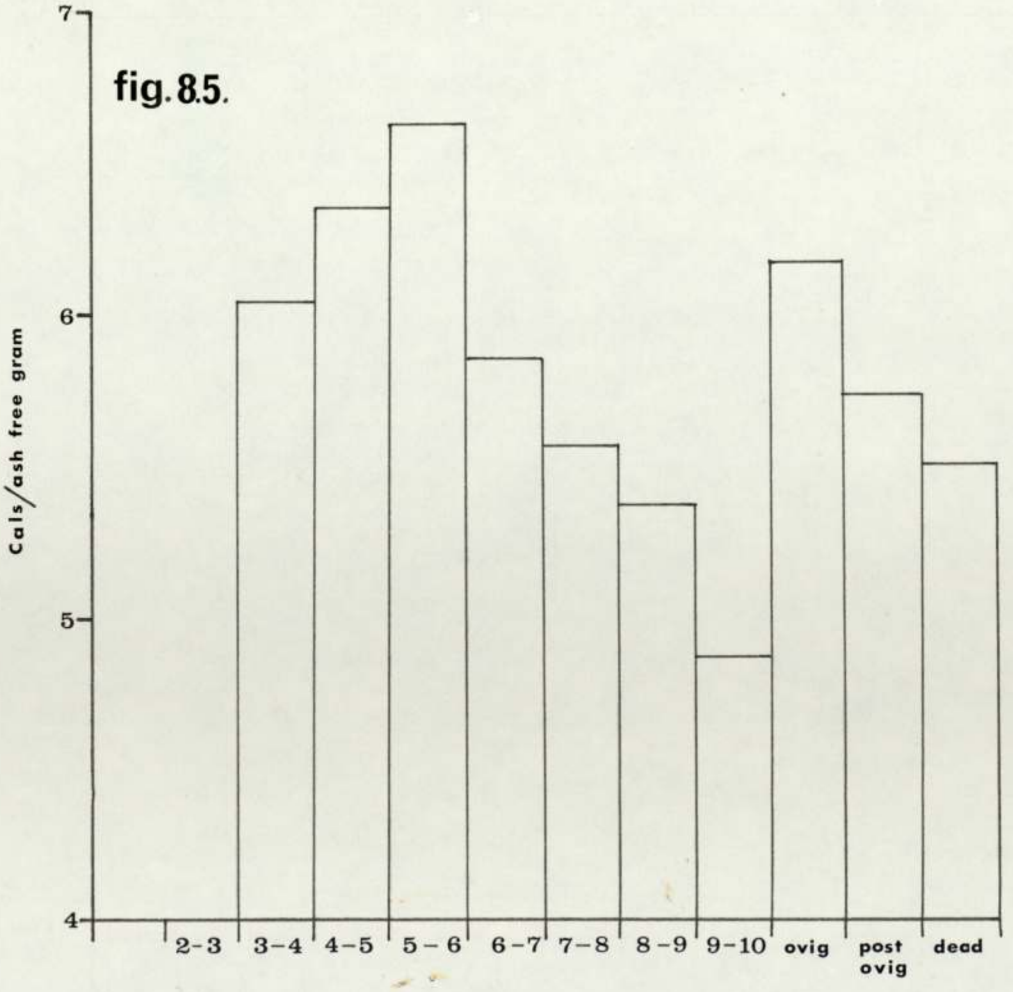
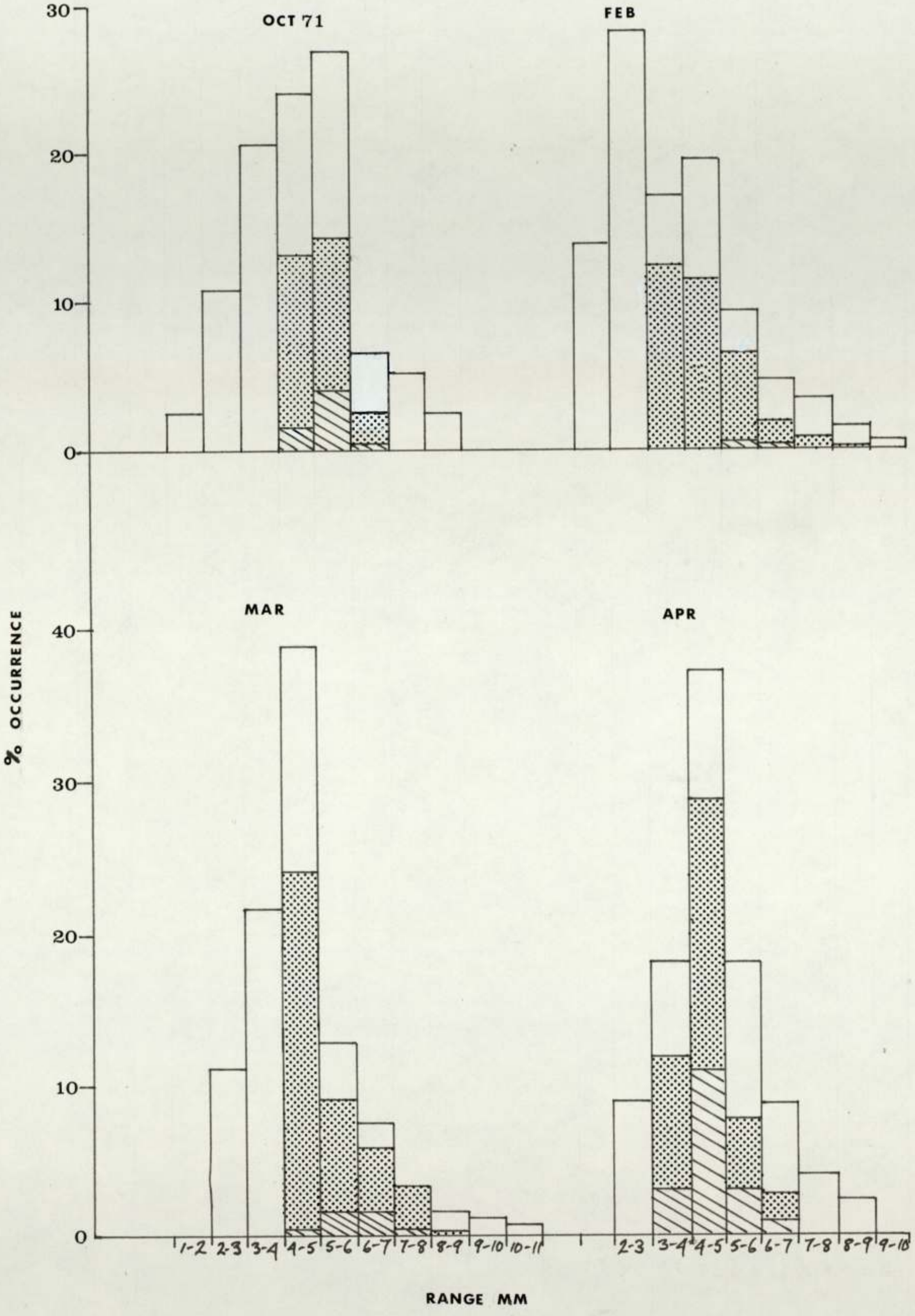
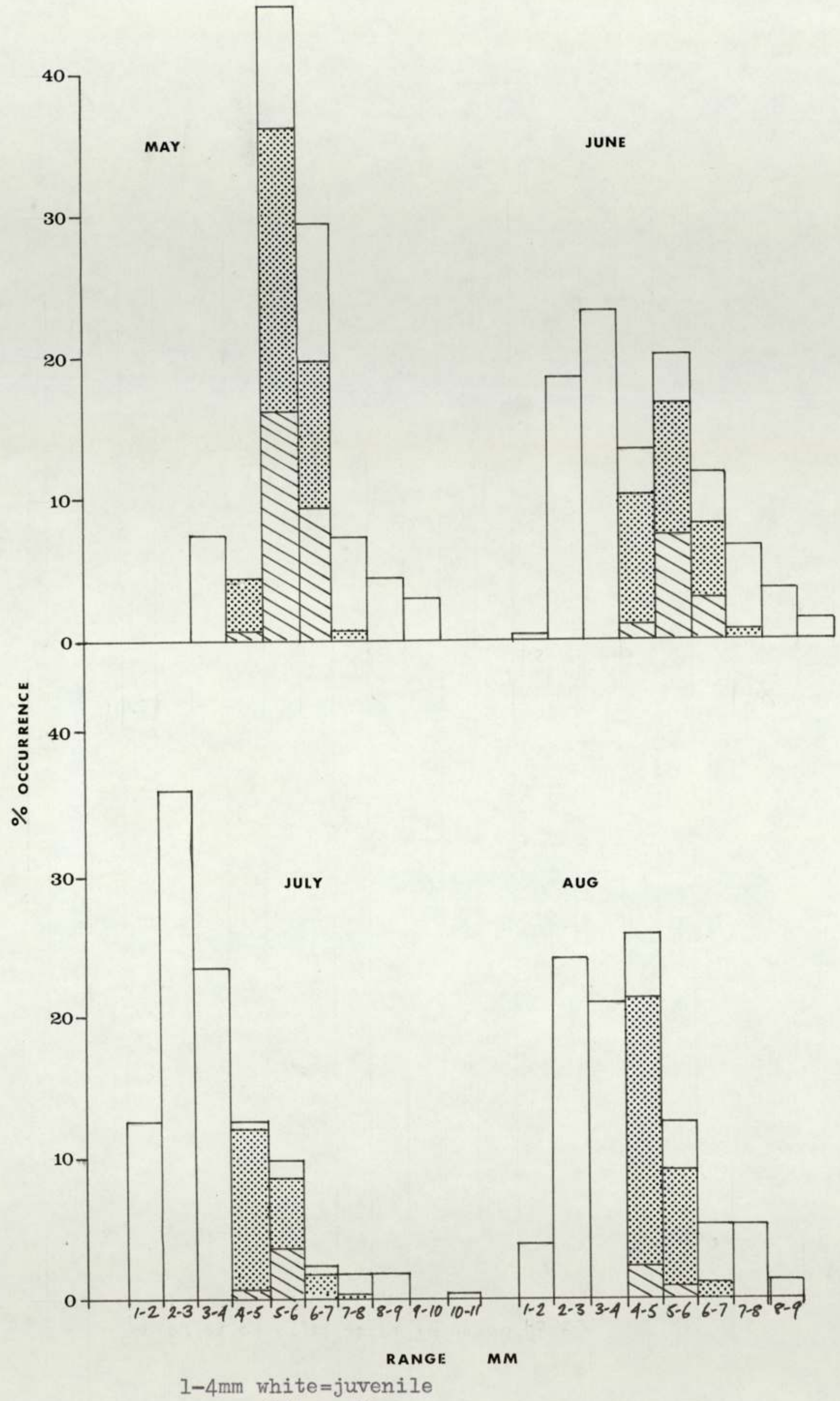


Fig.8.6 Length/frequency histograms of the A.aquaticus population in the R.Ray from October 1971 - January 1973 (continued overleaf). Each range class is expressed as a % of the total sample.



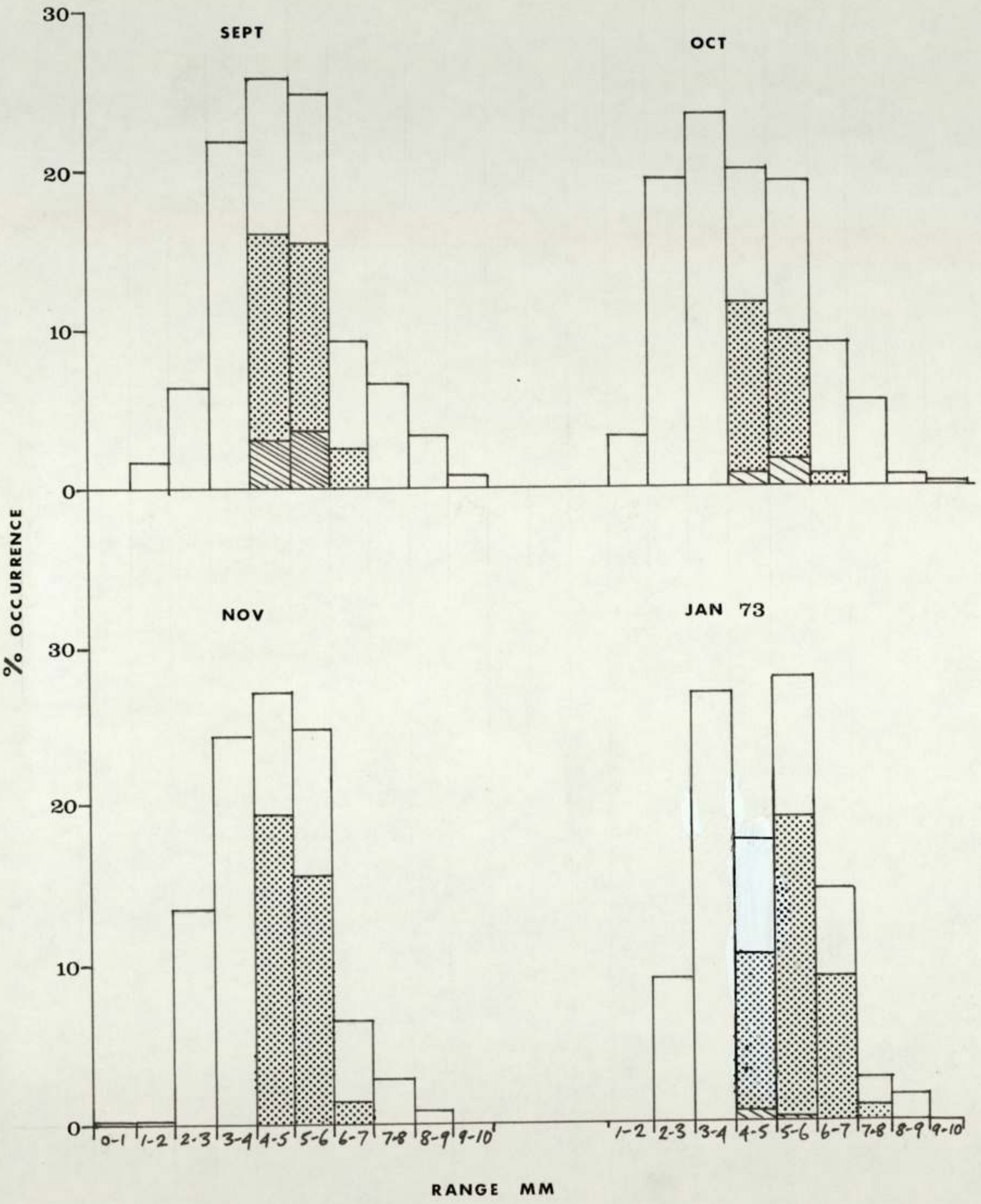
1-4mm white= juvenile
white= male
stipple = female
stripe = ovigerous

Fig.8.6 (cont)



1-4mm white=juvenile
white=male
stipple=female
stripe=ovigerous

Fig.8.6(cont)



1-4mm white=juvenile
white=male
stipple=female
stripe=ovigerous

Table 5. Density and weight of the Asellus population in the R. Ray at Moredon Bridge May 1972 - April 1973.

Date	Mean No. $/m^2$	Mean dry wt. g/m^2	Mean dry wt. individual mg.
17.5.72	10,085	11.56	1.15
17.6.72	10,866	25.64	2.63
19.7.72	7,085	6.16	0.87
14.8.72	9,025	4.60	0.51
20.9.72	13,621	10.79	0.74
26.10.72	21,032	23.1	1.10
23.11.72	14,844	7.87	0.53
31.1.73	11,093	9.09	0.82
7.3.73	19,707	13.0	0.66
9.4.73	3,867	5.10	1.32
Mean	12,122 m^2	11.69 g/m^2	
"	606/0.05 m^2	580mg/0.05m^2	

22.3.73 - 13.3.74.

Mean no. Asellus/0.05m^2 in Experimental Stream 1 = 355.8

Mean dry wt. mg/0.05m^2 " " " 1 = 513.7

growth of the population. The increase in numbers and biomass (dry wt.) after the spring brood is not very marked but by the end of autumn they have exploded to proportions equivalent to those of the Ray. The apparent stability of the population between June and October points to the possibility of sampling error, the gregariousness of these newly colonised animals accounting for the anomaly.

Table 5 shows the mean annual biomass (dry wt.) of Asellus populations in the Ray and Experimental Stream 1 to be 580 and 514 mg/0.05 m^2 respectively. While these are approximately the same, the density of individuals determined over the same periods differ substantially, with the Ray's population numbers exceeding those of the Experimental Stream by almost 50% (Table 5). This also points to the conclusion that the Ray's population, which was mostly sampled from Cladophora blankets, contains a higher proportion of juveniles, the older members living apart from the alga. The rapid increase of the Asellus population witnessed in the Experimental Stream has therefore approached the proportions of the more highly eutrophicated Ray's population. Bearing in mind that there is no fish predation in the Experimental Stream the standing biomass of its Asellus population will probably exceed that of the Ray's in time, but whether the production will ever equal that of the predated Ray is thought unlikely - n.b. the large densities of juveniles.

The calorific values of the 1mm range classes have been determined for populations of Asellus from the Ray (Fig. 8.5). The histograms are the results of only one determination per range class, each sample being a composite of more than 50 individuals of the range (both sexes and ovigerous) with the exception of the scarce 9 - 10 mm class which only comprised 2 individuals of approximately 5 mg each. The results show that more accuracy in the estimations of production should be made using length/frequency analysis (where necessary) and calorific values for the same range classes instead of the usual method of dry weight per range class multiplied by a coefficient for the species.

Other salient observations from Fig. 8.5. are the expected higher calorific values of the younger range classes as a result of inherited fats, and the ovigerous females as a result of egg production. The difference due to eggs and embryos is manifest in the calorific value of the post-ovigerous females. The importance of keeping animals alive until the last minute, if calorific determinations are to be conducted, is shown from the histogram of 'dead' samples. This is the mean of three determinations of animals from the 5 - 7 mm ranges dead for eighteen hours.

8.4. CONCLUSIONS.

1. Breeding of Asellus populations in the eutrophicated Ray continued for one month longer than populations in the Thames at Reading recorded by Steel (1961).

2. The peaks of breeding in the Ray and Thames differed by one month, occurring in May and April respectively. Sexual maturity of the Asellus autumn brood was reached earlier in the Ray populations but the fecundity was lower than in populations from the Thames.

3. The above differential effects may have been caused by differences in dissolved oxygen concentrations in which the animals existed, being synchronous with the degree of eutrophication of the rivers. It is also suggested that the timing of the broods may be influenced by <u>Cladophora</u> blankets which provide shelter.

4. The annual mean standing biomass of the <u>Asellus</u> populations in the Ray and in Experimental Stream 1 were approximately the same but a greater proportion of biomass in the Ray was accounted for by juveniles as a result of apparent habitat preferences with age.

5. Length/frequency analyses, together with calorific determinations for range classes, would give a more accurate figure of production for <u>Asellus</u> and probably other invertebrates whose age class cannot be readily distinguished.

6. Calorific determinations should be made on freshly killed and dried animals.

APPENDIX.

THE TAXONOMIC FEATURES OF THE FRESHWATER LARVAL CHIRONOMIDAE IDENTIFIED FROM THE MIDLAND RIVERS AND EXPERIMENTAL STREAMS.

The importance of the larval Chironomidae has been noted with reference to production studies, fisheries and biological indicators. These non-biting midges form a significant contribution to the benthic ecosystems at all trophic levels, as seen in Chapters 4 and 5, but because a number of species remain undescribed or are only known in the adult form, the keys were not considered entirely credible and confidence in their use by some workers was never gained. This, together with the difficult and specialist taxonomy, led to much valuable information inherent in hydrobiological research work being lost.

With more keys and information available now, the task of identification is eased a little but it is still advisable to make drawings of the important elements of the species taxonomy. This appendix includes thirteen plates containing the main taxonomic features of twenty-eight species in the event that subsequent work may be continued or that misidentification has been made. Those species like Chironomus riparius and Prodiamesa olivacea that are easily identifiable have not been illustrated.

Information thought to be helpful for methodological purposes has also been compiled.

i) Methods.

Preserved specimens often loose their colour - i.e. haemoglobin containing larvae and those with guts full o of algae - although, of course, the body itself is not pigmented in most species. For this reason it is a good idea to examine the colours of both preserved and unpreserved bodies. Preliminary sorting in white ceramic dishes according to the colour of the body, hooks of the prolegs, head capsule (especially clypeus, mandible, area around the eye and the occipital area) and eye shape is helpful. Other features discernable with a low power binocular microscope may be the length and slimness of the posterior proleg (n.b. Eukiefferiella spp.) and the hairyness' of the abdomen and thorax.

Larvae with imaginal plates showing through the thorax should be slected for high power identification as the full larval features will have been attained. Specimens may then be mounted for high power identification in Berlese's fluid after separating the head from the thorax, mounting these apart at each end of a glass slide. Very thin cover glasses are recommended. (The use of an alkali to dissolve away muscle is necessary until familiarity with the larvae is achieved). Unidentified specimens should also be drawn and numbered so that similar larvae may be likewise numbered until an identification is made. Sometimes the degree of sclerotisation of the head capsule and procercus is useful for taxonomic purposes.

While the Berlese fluid is still tacky, the x40 and x100 objectives may be used to compress the head to reveal details of the antenna, anterior setae etc. (as long as the lenses are recessed within the outer casing). A good x100 non oil immersion lens is ideal with x5 ocular magnification.

Drawings for the plates 12 - 24 were made on a Nikon microscope with x10 Vickers eyepiece, x40 and x100 Nikon objectives together with a camera lucida.

ii) Notes on the taxonomy.

Species Orthocladiinae C.10 and C.11 (15 and 30 respectively on the plates) remained unidentified. Additional information -

C.10 Width of median tooth is equivalent to the distance between teeth 1 - 3 (numbering from below median). Sometimes the first tooth of the mandible is seen to be sharper. The antennal shaft is tapered in this species. The prolegs are slightly extended but not to the extent of *Eukiefferiella* spp. The body is hairless and green/grey with a yellowish thorax (preserved).

C.11 The sides of the median tooth are straighter than those of *C. (Cricotopus) albeforceps?* and the antenna possess larger Lauterborn organs. A mandibular spine was observed on some specimens as were mandibular crenulations. There are no serrations on the inner edge of the mandible - cf. *C.* (*Cricotopus*) *ex*. *gr*. *bicinctus* Meig. The body is hairless and green/grey with a more yellowish thorax. The antennal blade is not always the same length as the antenna at their tips.

Key to Symbols.

a	antenna
ag	anal gill
c	paralabial comb
ep	epipharynx
gl	glossa (lingua)
h	head capsule
l	labrum
lp	labial plate
m	mandible
mb	mandibular brush
mp	maxillary palp
pm	premandible
ppl	posterior pro-leg
sa	anterior labral setae
sp	striated paralabial plate
w	procercus

Measurements are in microns (μ)

CHIRONOMID LARVAE: TAXONOMIC FEATURES OF THE HEAD CAPSULE AND ABDOMEN.

Key to the Plates.

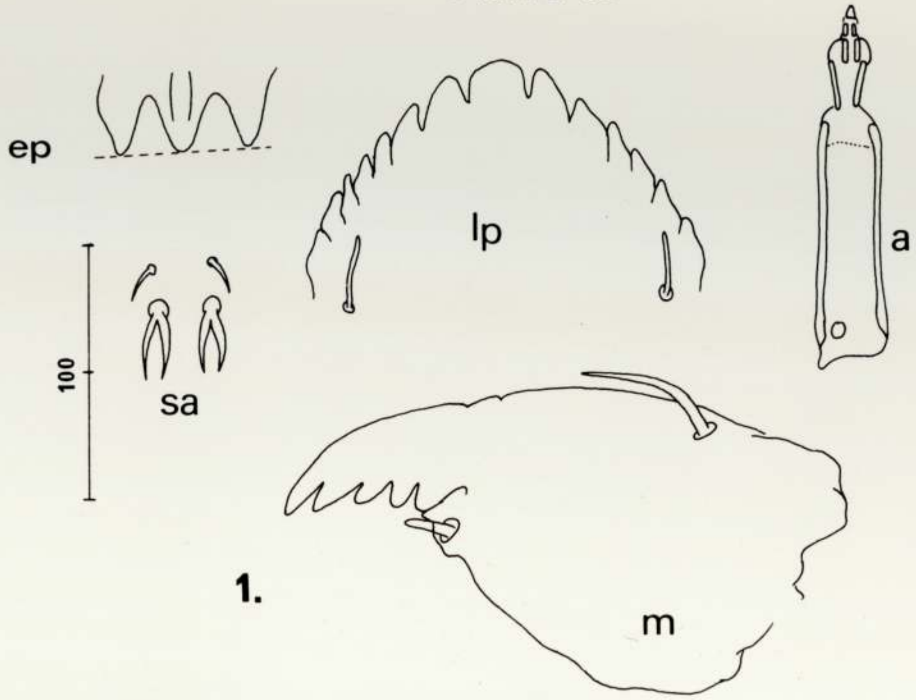
Plate 12	1. Cricotopus (Isocladius) sp?
	2. Rheocricotopus sp.
Plate 13	3. Cricotopus (Cricotopus) ex. gr. fuscus Meig.*
	4. Cricotopus (Cricotopus) trifascia Edw.*
Plate 14	5. Cricotopus (Cricotopus) ex. gr. bicinctus Meig.*
	6. Diplocladius cultriger Kieff.
Plate 15	7. Eukiefferiella hospita Edw.
	8. Lymnophyes sp. 2 tooth.
	9. Lymnophyes sp. 3 tooth.
Plate 16	10. Eukiefferiella sp. (C.1.)
	11. Cricotopus (Cricotopus) albeforceps?
Plate 17	12. Coryenura sp.
	13. Synorthocladius semivirens Kieff.
	14. Eukiefferiella sp. - subtype 1.B. lavrica Zavrel 1939.
Plate 18	15. Orthocladiinae sp. (C.10)
	16. Brillia modesta Meig.
Plate 19.	17. Procladius sp. (choreus?)
	18. Thienemannimyia ex. gr. lentiginosa Fries.+
Plate 20	19. Brillia longifurca Kieff.
	20. Diamesa campestris Edw. (Potthastia longimanus)
Plate 21.	21. Polypedilum ex. gr. pedestre Meig. (arundineti Goet)
	22. Metriocnemus atratulus Zett.

Plate 22	23. Micropsectra atrofasciatus Kieff.
	24. Dyscamptocladius ex. gr. vitellinus Kieff.
Plate 23	25. Stictochironomus ex. gr. histrio Fabr.
	26. Polypedilum nebeculosum Meig.
Plate 24.	27. Polypedilum scaelinum Shrank.
	28. Orthocladiinae sp. (C.11.)
	29. Prodiamesa olivacea Meig. (not illustrated)
	30. Chironomus riparius Meig. (not illustrated)

* A number of keys use the genus *Trichocladius* instead of *Cricotopus*.

\+ from Fittkau, Die Tanypodinae Stuttgart 1964. = *Pentaneura melanops* Meig.

PLATE 12



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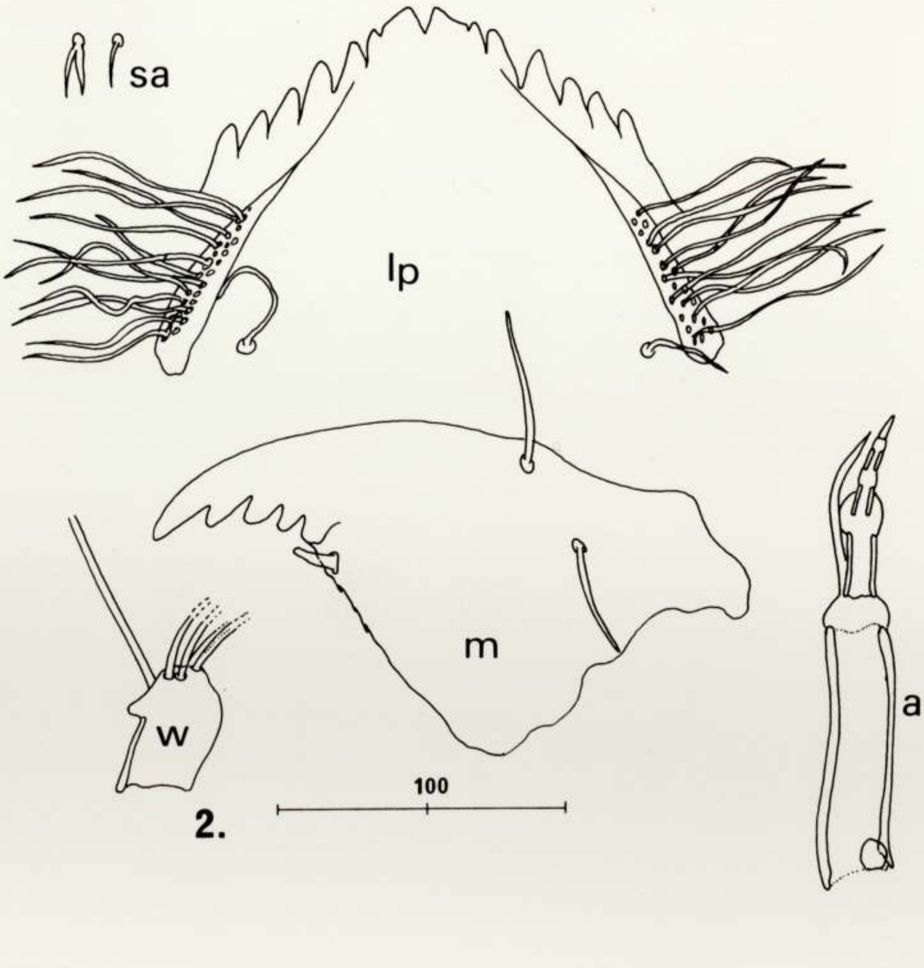
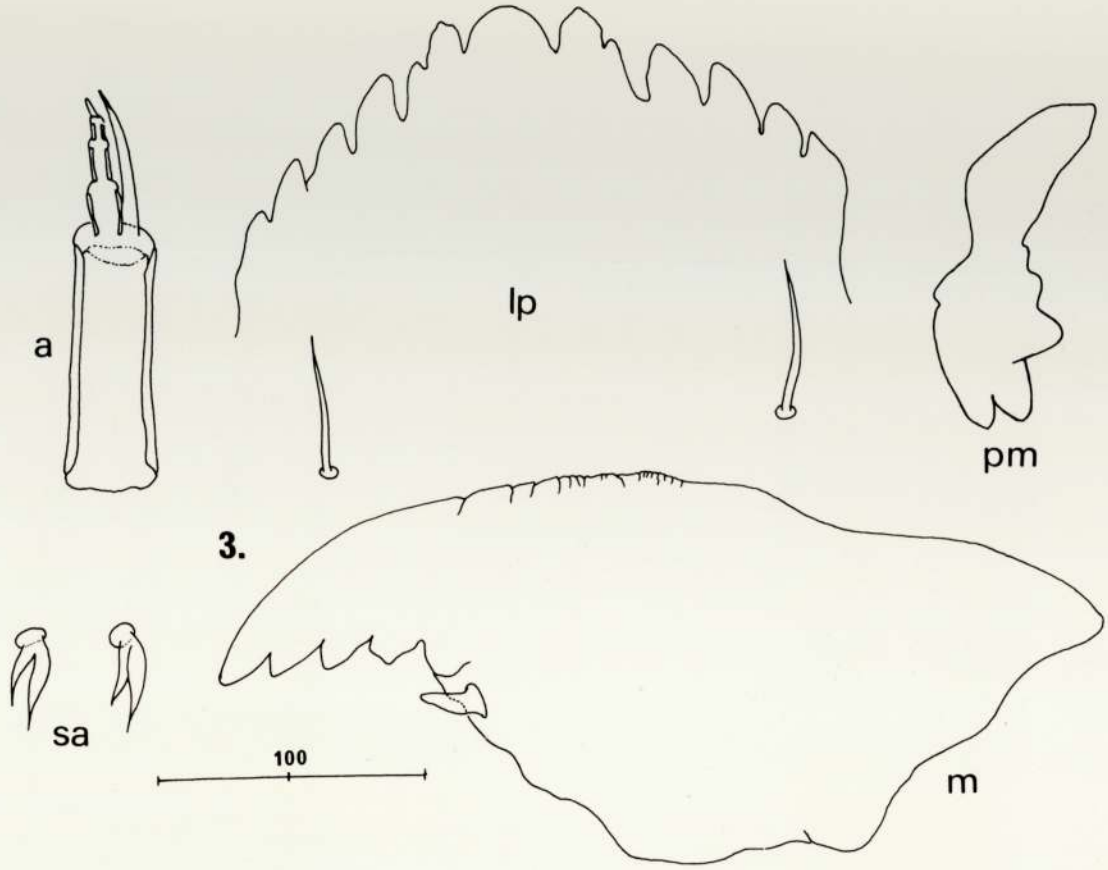


2.

PLATE 13

3.



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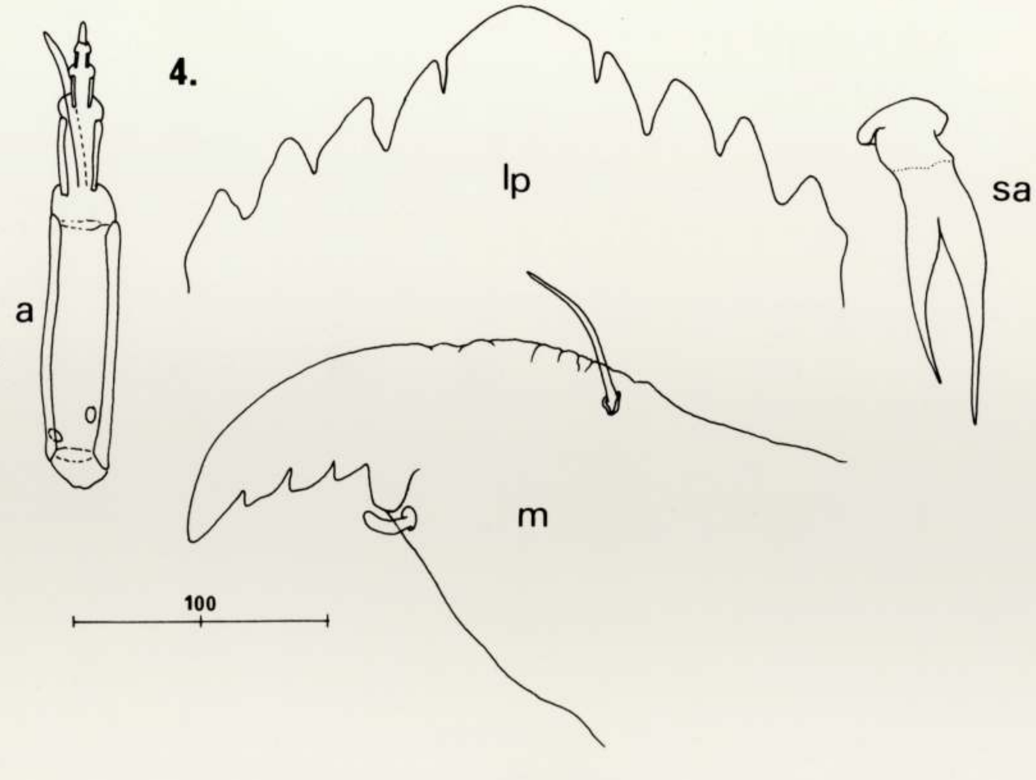
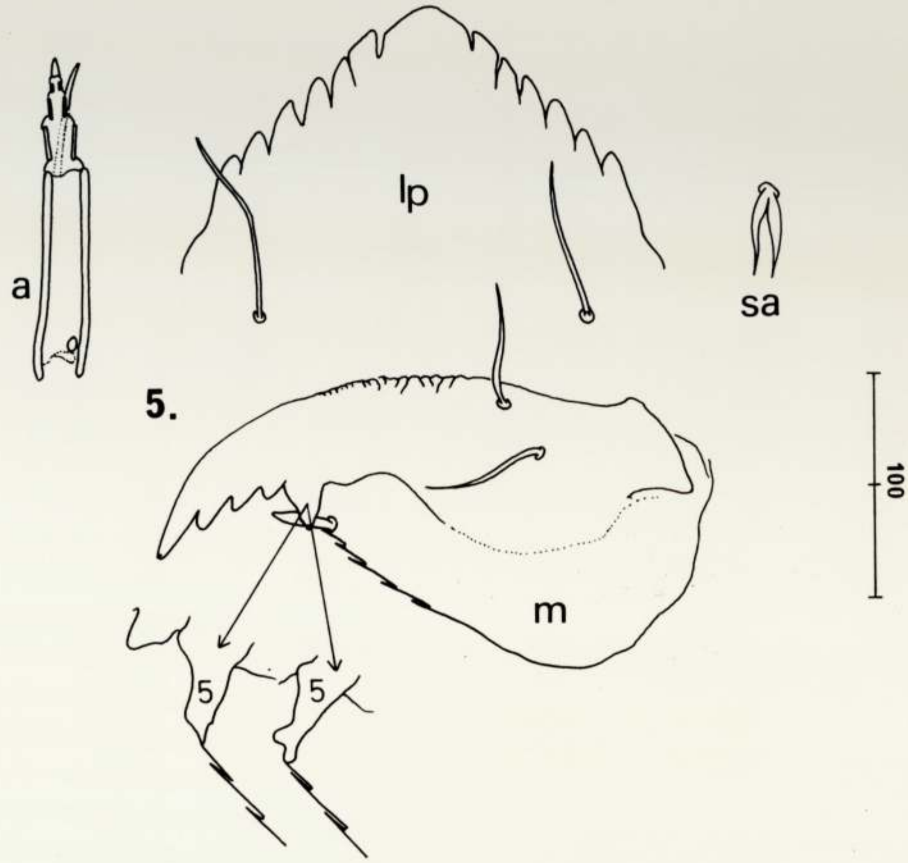


PLATE 14



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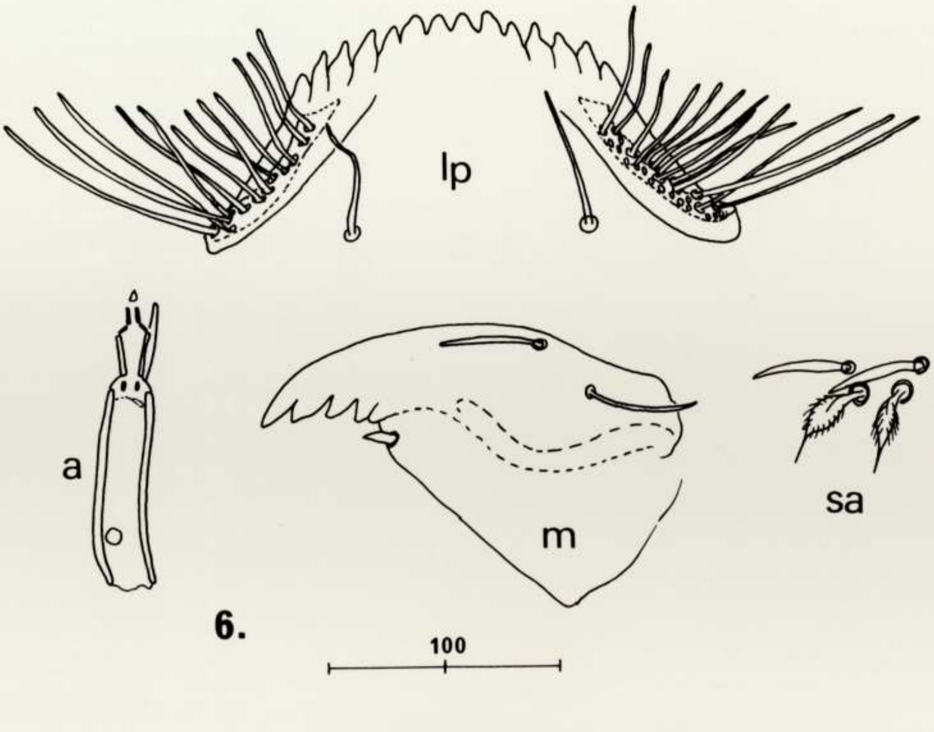

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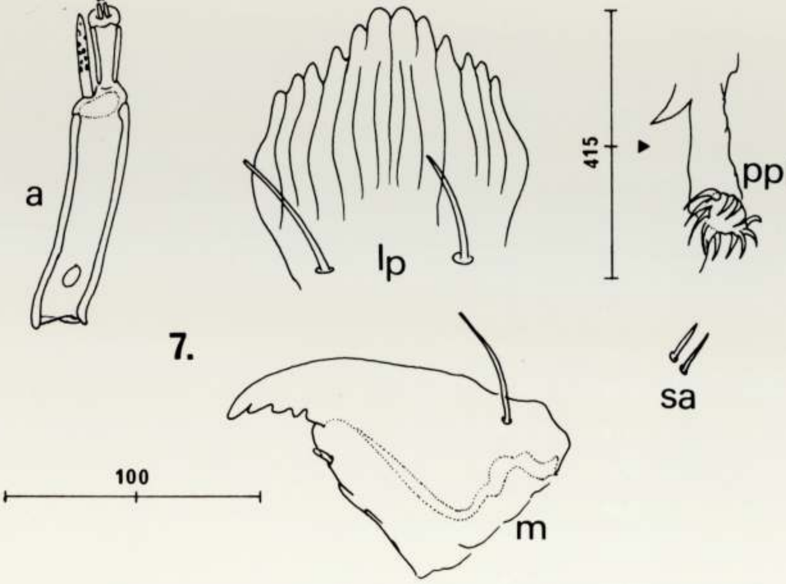
PLATE 15



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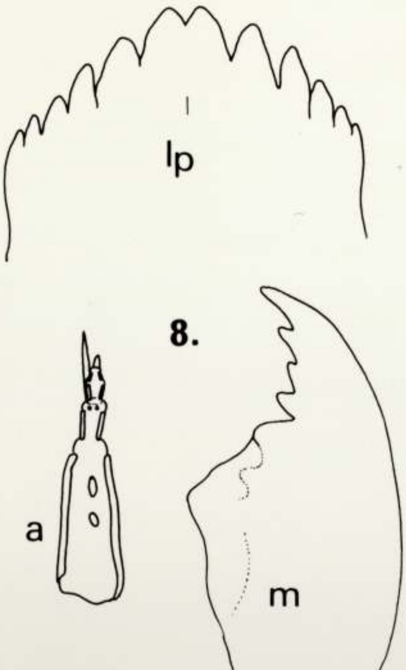

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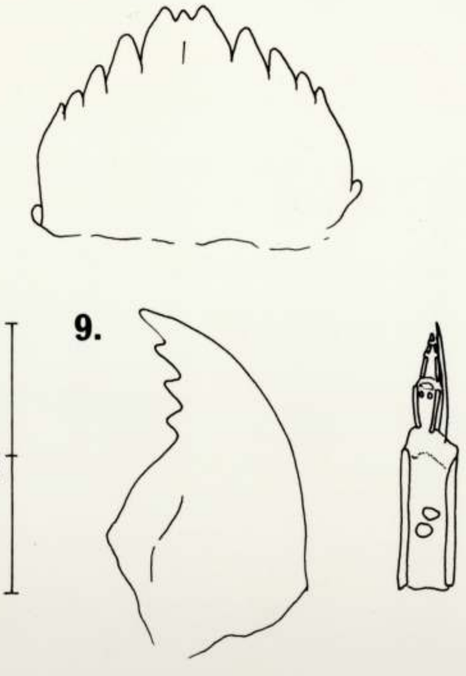

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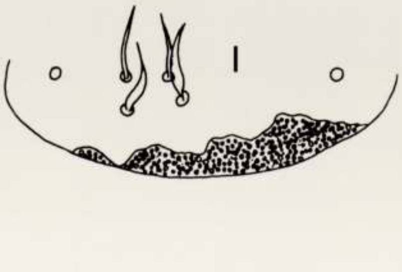
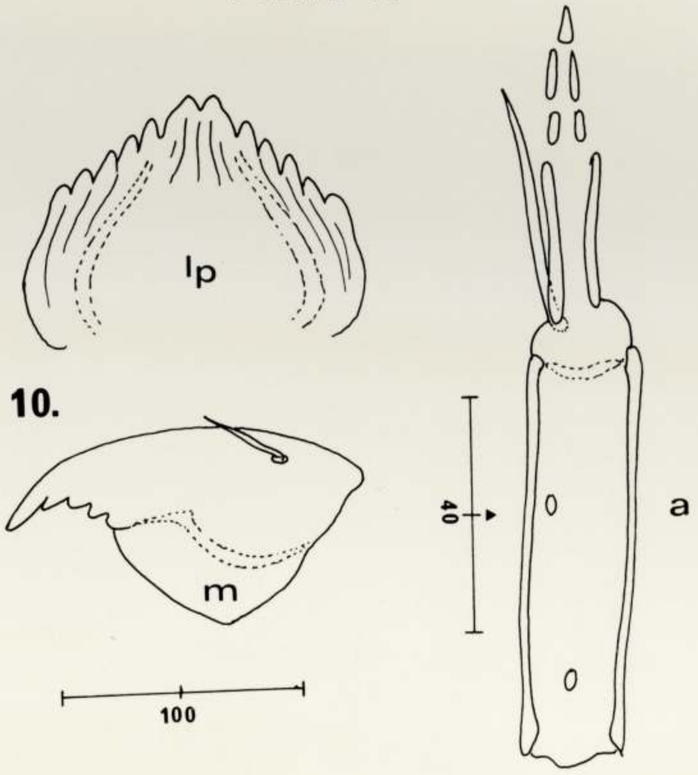


PLATE 16



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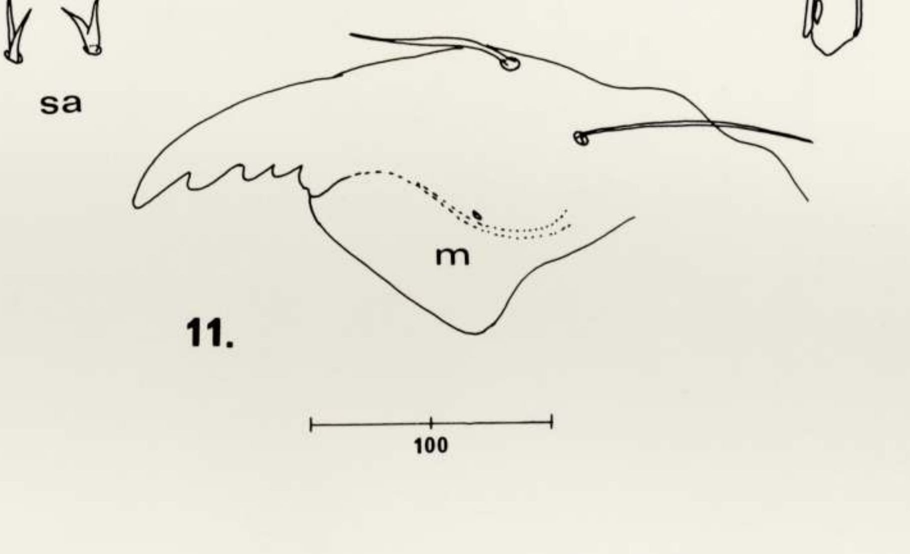
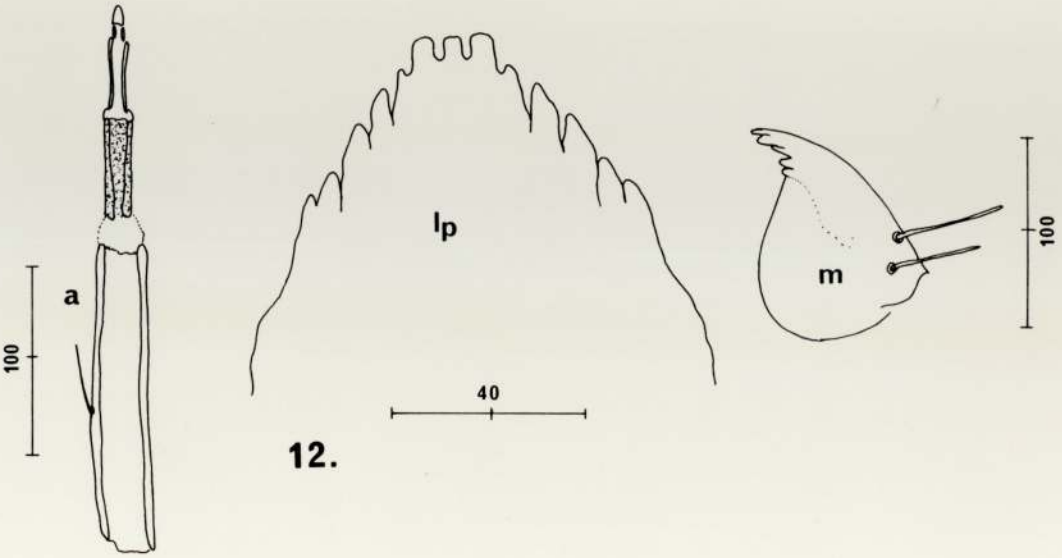


11.

PLATE 17



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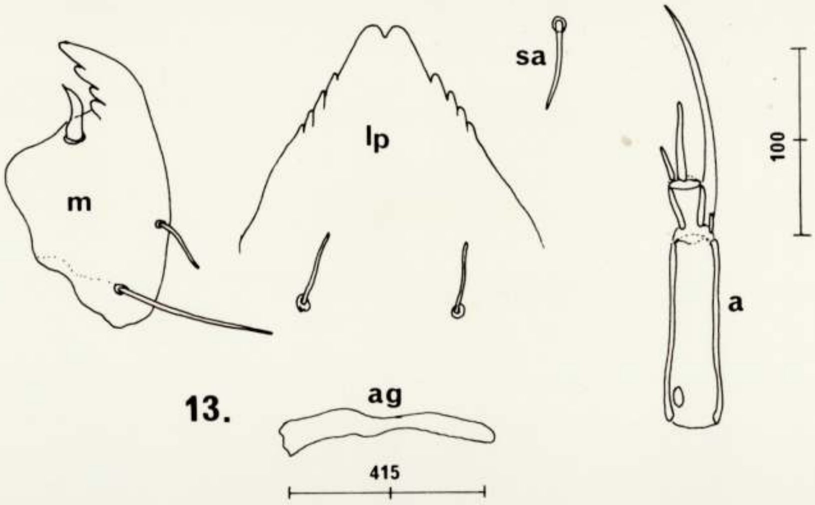

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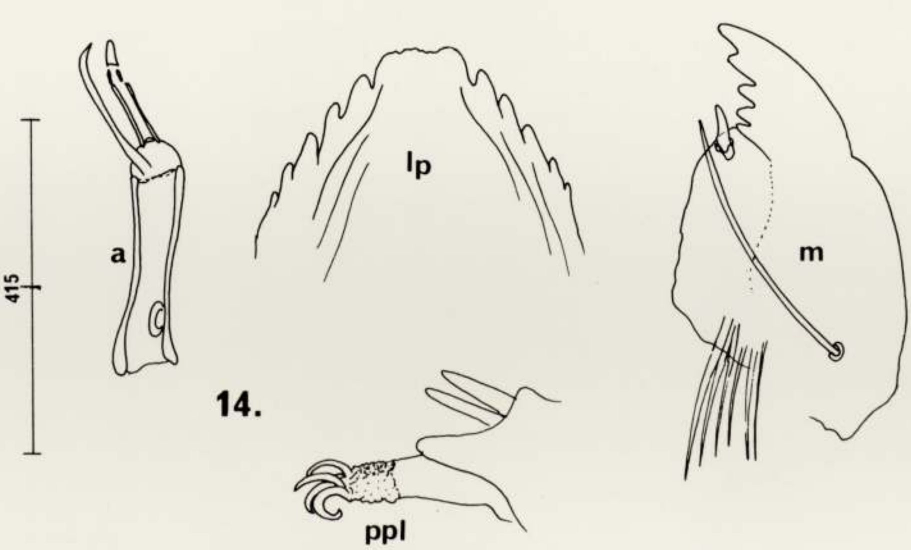

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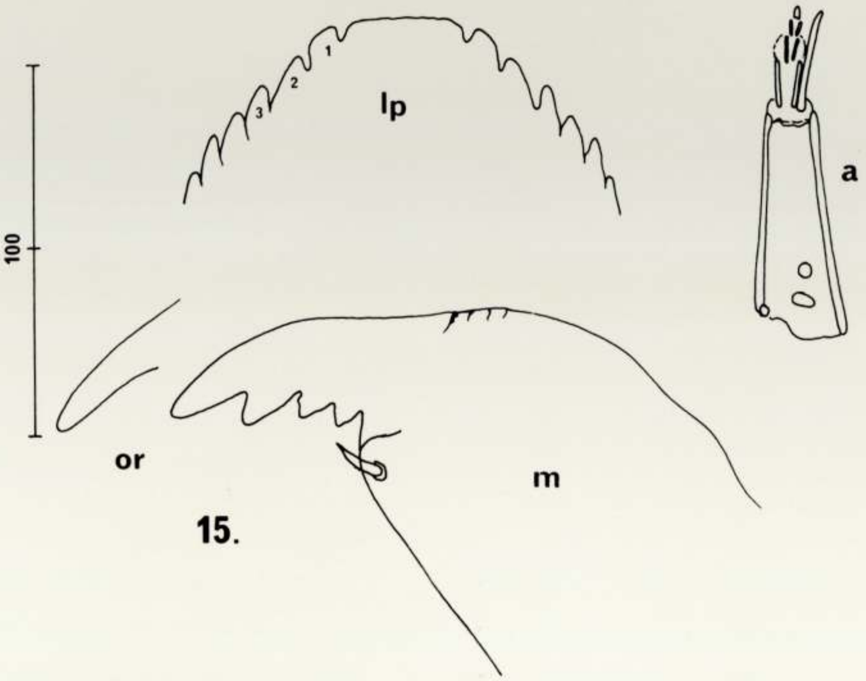

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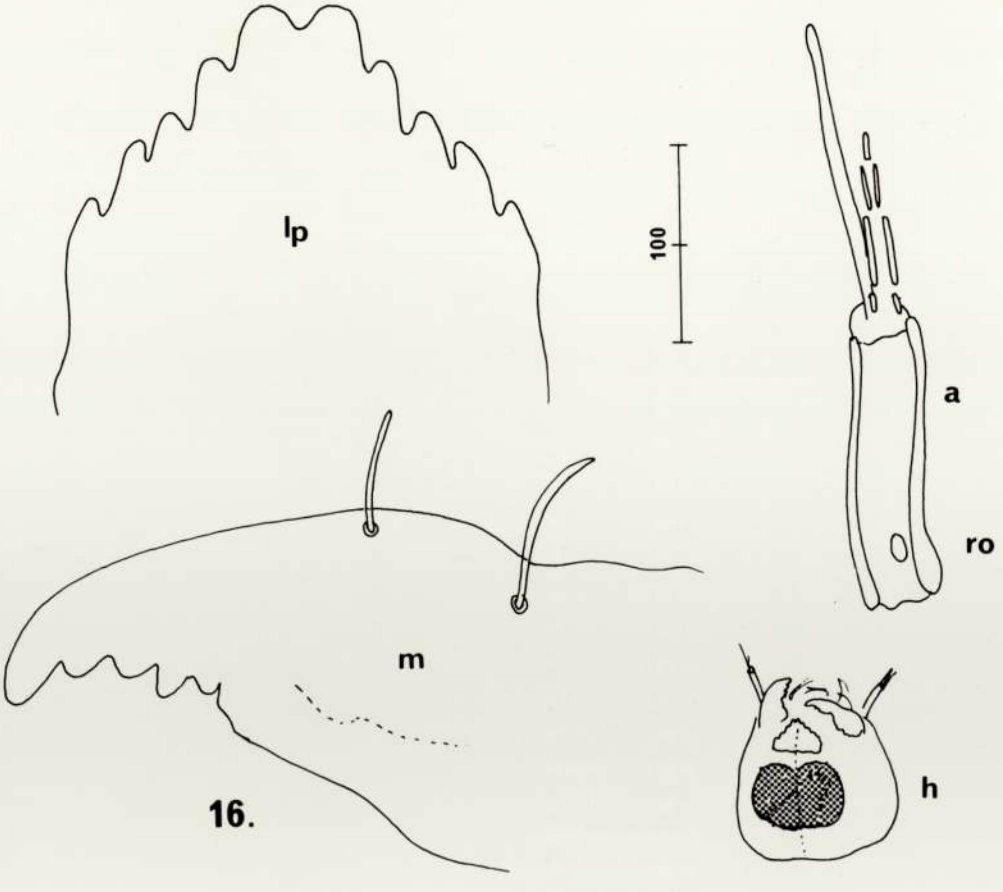

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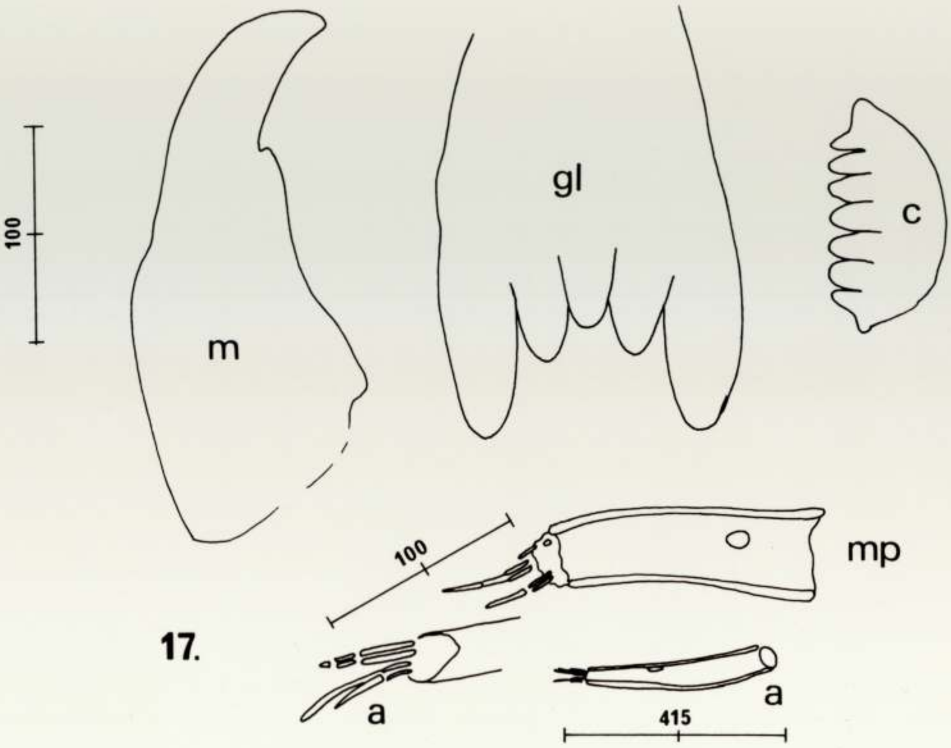

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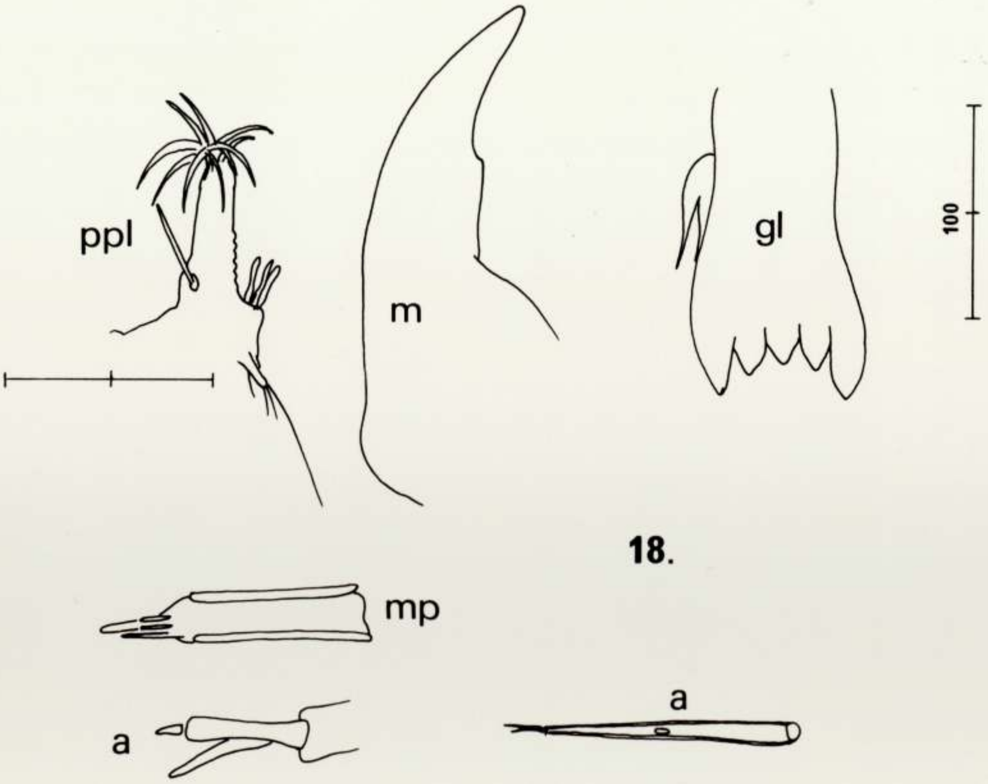

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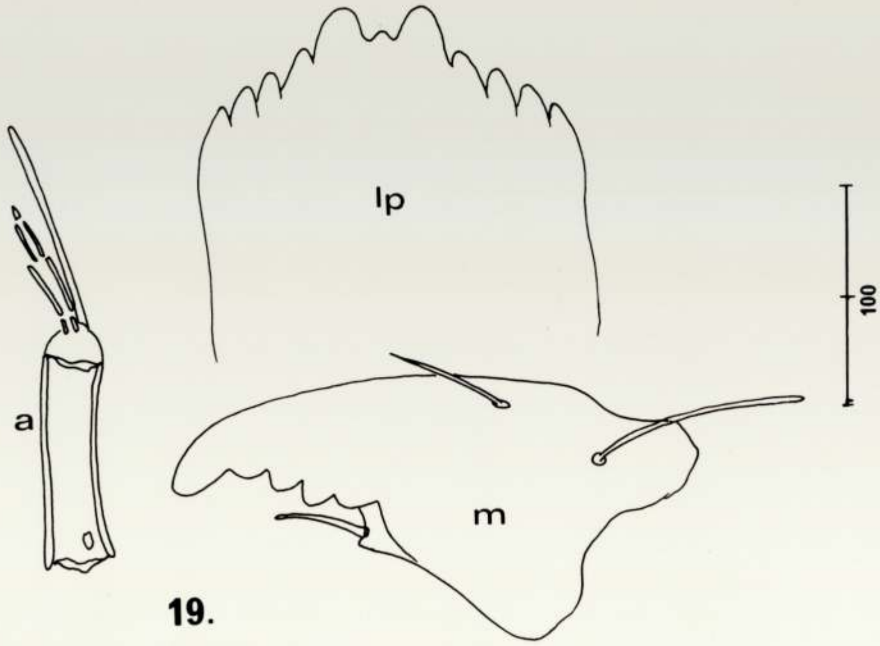

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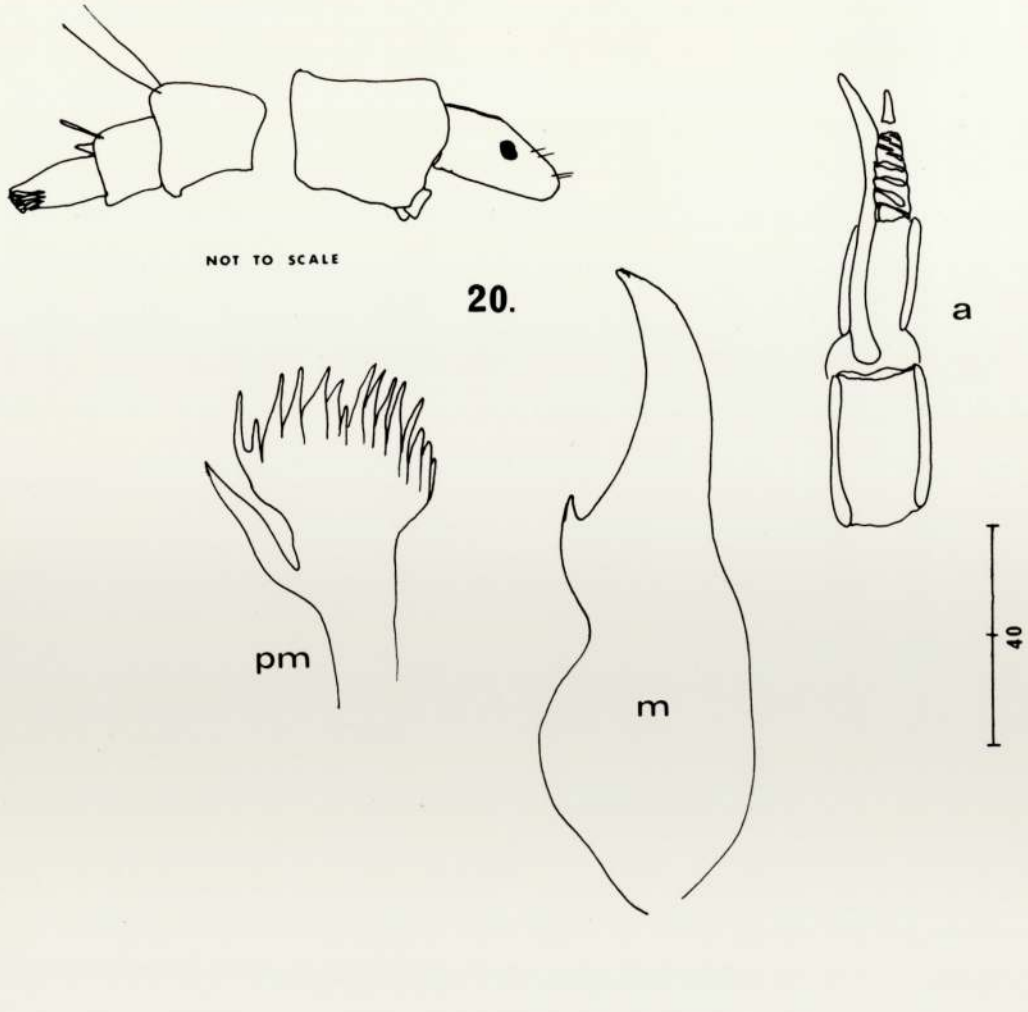

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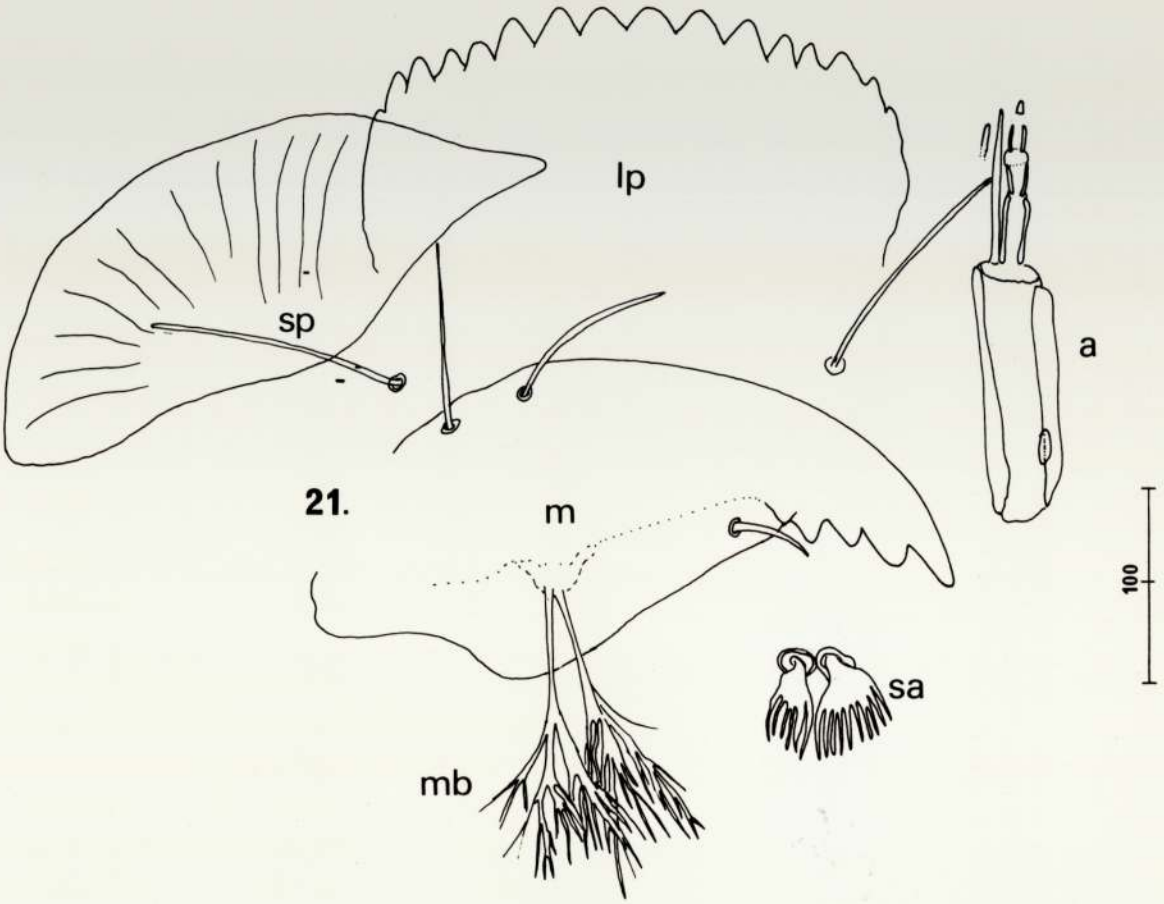

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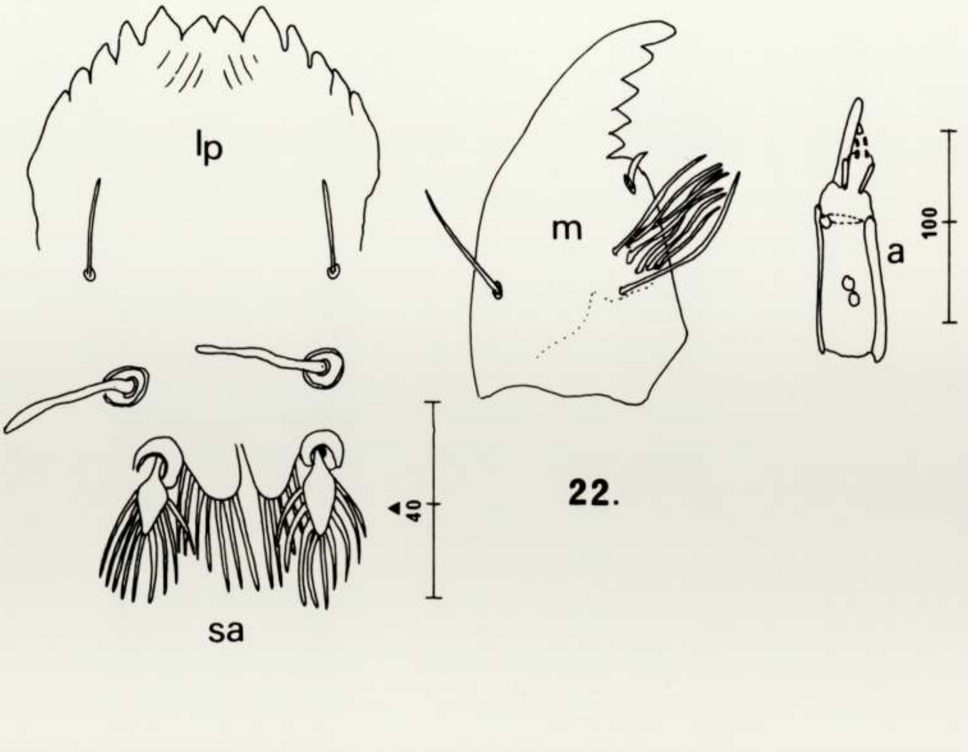

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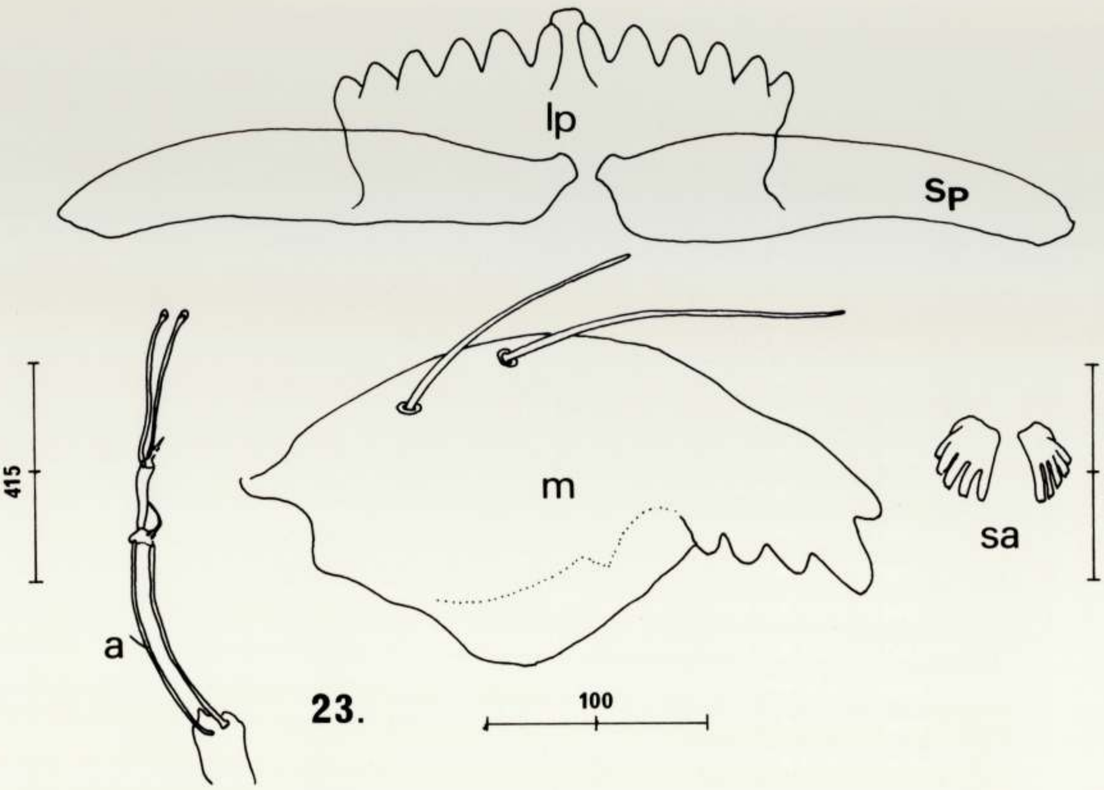

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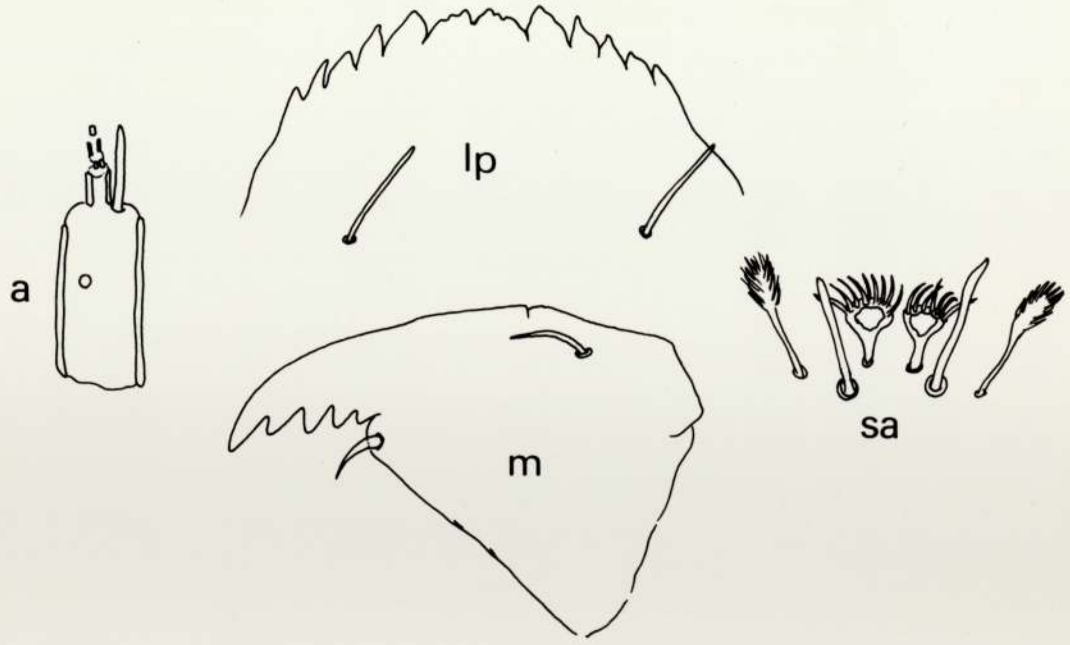

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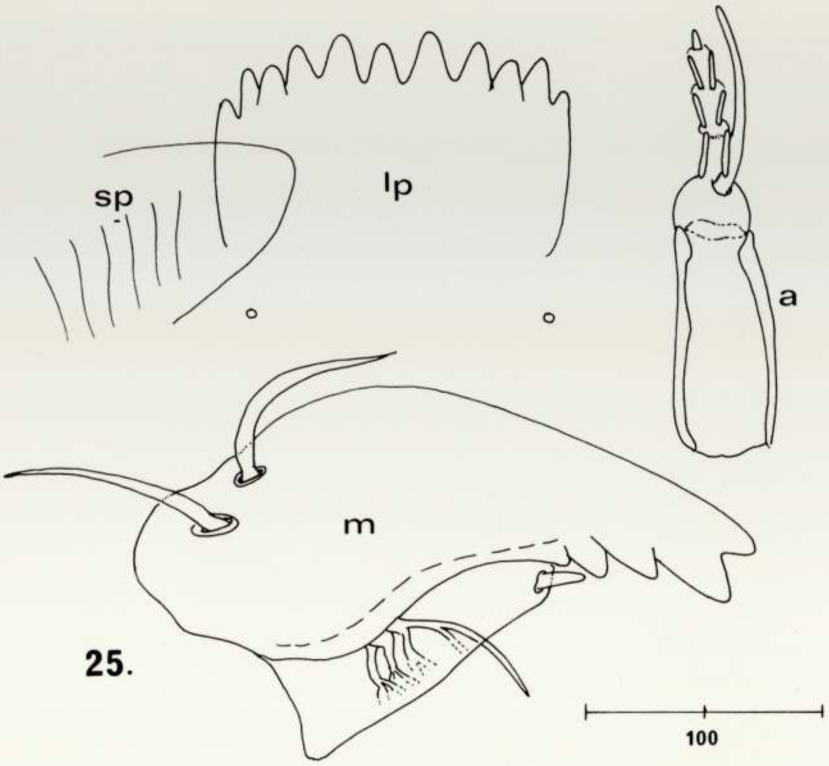

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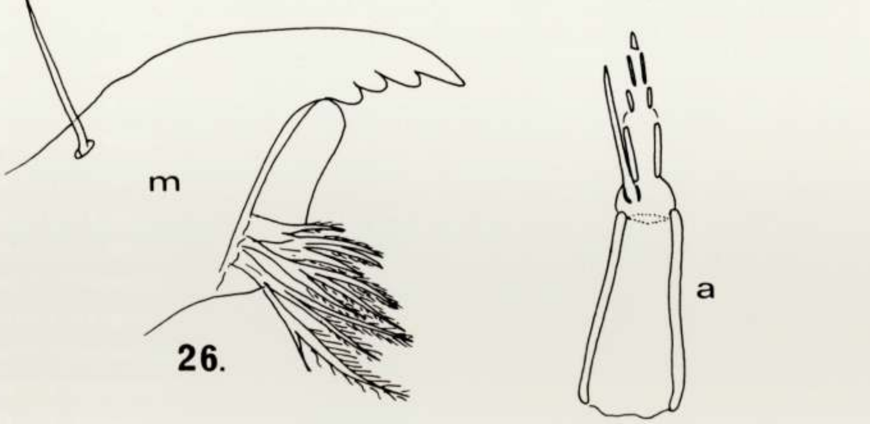

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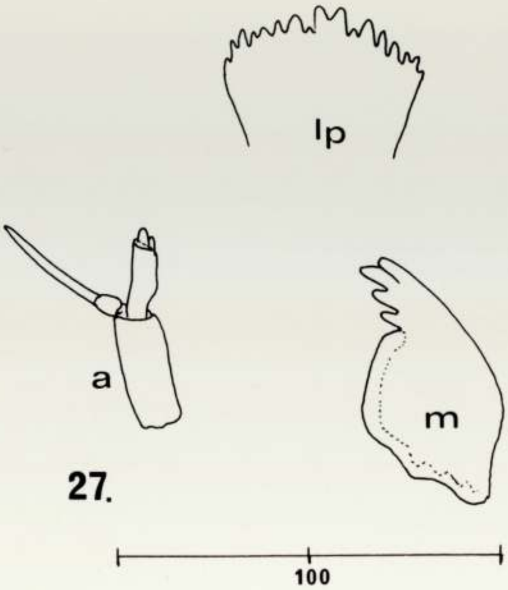

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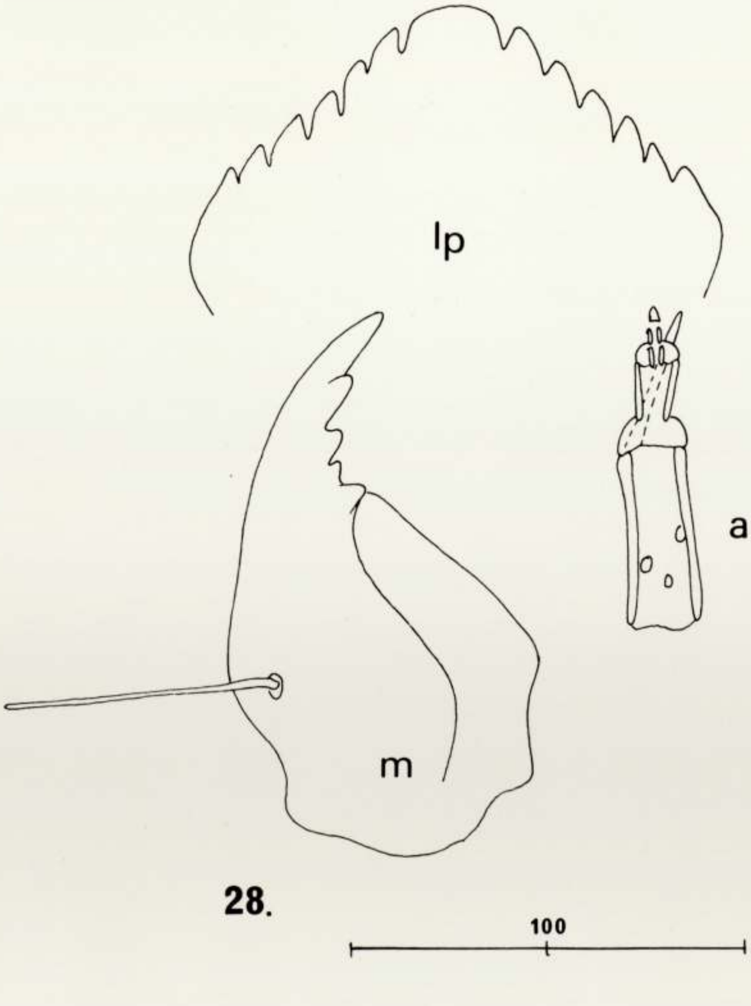

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