

Figure 1.5 Percentage of time spent actively swimming by *Gammarus pulex* exposed to different concentrations of zinc. Animals were observed via a remotely-controlled video camera to minimise disturbance

animals in an observation chamber, were considerably increased by exposure to the ambient zinc levels of the West Allen. These findings (for the Isopod crustacean *G. pulex*) were confirmed by Austin (1992) using a more sophisticated observation procedure based on analysis of remotely-controlled videotaped observations of the animals under similar conditions (Figure 1.5); and Crane (1995) also found that the feeding rate of *G. pulex* was reduced by 50% at a zinc concentration of 0.5 mg l⁻¹.

Whatever the underlying mechanism, a combination of increased activity combined with decreased feeding is potentially of profound ecological significance, leading for example to decreased growth rate and reproductive capacity, and may be associated with the ecological effects of the pollutants on the aquatic fauna. The increased activity levels may represent avoidance reactions on the part of the animals—it has been well documented that fish and invertebrates show measurable avoidance reactions to pollutants in laboratory choice chambers. In the field, hyperactive invertebrates are likely to suffer from increased rates of downstream drift, leading to depopulation of polluted stretches of river. Gilhooley (1988) showed this effect in artificial stream channels at the concentrations of zinc typically found in metalliferous rivers. Similar effects have been recorded in artificially-acidified streams in experiments to assess the ecological impact of acidification of waters (Hall *et al.*, 1980; Ormerod *et al.*, 1987).

Another aspect of sublethal toxicity is the possible accumulation of metals in the animal tissues. Animals may survive exposure to low levels of pollutants without apparent effect, but may continue to accumulate metal from solution, by ingestion

of or contact with particulates, or from their food, until harmful levels of the pollutant in the tissues are reached. These considerations relate not only to heavy metals but also to refractory organic pollutants including certain pesticides. One reason for concern about this phenomenon is that animal tissues containing heavy metals (and other pollutants) may cause harm to consumer organisms—fish consumed by humans are an obvious example—but there are obvious concerns relating to the passage of pollutants up the food chain which may affect all predators. If, however, we consider simply one species without regard to these aspects, we still need to know whether the level of metal in the tissues is harmful to the organism itself. For reasons explained in Chapter 4, this is not always an easy question to answer; however, the presence of the class of inducible metal-binding proteins known as metallothioneins (see Chapter 4) may be used as an indication that the level of metal in the organism under study is abnormally high. Metallothioneins have been tentatively identified as being present in the tissues of some invertebrates living in metalliferous rivers in the North Pennine Orefield (see Figure 4.18).

The effects of other poisons, and of environmental factors which modify toxicity, should also be considered. In many of the rivers referred to above, more than one metal was present in significant quantities. In the zinc-polluted river Nent in the North Pennine Orefield, substantial inputs of organic farm waste were recorded (Armitage, 1980), which may modify the toxicity of the heavy metals present (see Chapter 4). Clearly, some of the differences observed between different rivers may be attributable to such confounding factors. In addition, the toxicity of heavy metals is greatly influenced by levels of calcium, and indeed calcium itself may be a limiting factor; while it may be tempting to ascribe the absence of certain species (particularly molluscs and crustaceans) to the presence of zinc or other heavy metals, they may in fact be limited by levels of calcium lower than 10 mg l^{-1} (Edwards *et al.*, 1978). Even levels of sodium may be limiting to some species (Sutcliffe, 1967, 1983). It is particularly important to take account of such factors in upland, mineral-poor waters where faunistic differences may be too easily and simplistically ascribed to pollution effects whereas in fact they may reflect the effects of natural influences.

The partitioning of the zinc in the river environment is likely to have a great bearing on the correct interpretation of toxicological data. The precise form in which the zinc is present can have a large influence on its toxicity (see Section 4.2). Metals may exist, in the aquatic environment, in dissolved, colloidal or particulate form; in two or more different oxidation states; as simple ions, inorganic complexes or organometal complexes. The biological and toxic properties of these different forms may vary greatly. In laboratory tests, the metal is normally presented to the animals in dissolved form, but it is almost certainly never entirely in the form of simple ions. In the river, the measurements made of 'dissolved' metals are, more accurately, of that fraction of the metals which can pass through a $0.45 \text{ }\mu\text{m}$ filter, which is not necessarily the same thing. Another aspect of partitioning is

that many animals burrow into the substratum, and may be exposed to levels of zinc in the substratum which are much higher than those in the overlying water. Green (1984) devised an apparatus for sampling the interstitial water without contamination with overlying water, and in the Allens found zinc levels in the interstitial water up to 25 times higher than those in the water flowing over the substratum. All of these factors complicate the interpretation of toxicological data and their application to the situation in the field.

Finally, the effects of the pollutant on the decomposer organisms of the river may be considered. The productivity of many aquatic ecosystems, particularly rivers, is sustained by the input of allochthonous organic matter (that is, organic material from outside the river) such as dead leaves. Decomposer organisms—bacteria and fungi especially—are thus of great importance in making available to the river fauna the major energy source represented by detritus. Indeed it is likely that most aquatic animals, like animals generally, cannot readily digest plant material unaided by microorganisms. An invertebrate may appear to be eating a dead leaf, but in reality is probably obtaining its nutrition from the bacteria, fungi, protozoa and microinvertebrates which have colonised the leaf and processed it into a form which is more accessible to detritivorous macroinvertebrates. Zinc is known as a fairly potent bactericide and fungicide. Possibly, therefore, one effect of the zinc is to interfere with the processing of detritus by decomposer organisms, thus depriving the invertebrates of a major food source and being indirectly responsible for the depletion of the fauna. Similar arguments have been adduced in relation to the acidification of surface waters (Haines, 1981; Howells, 1990; see Chapter 2). However, Chappell and Goulder (1994) studied the activity of extracellular microbial enzymes in the rivers East and West Allen, and found no evidence of reduced activity in the zinc-polluted river.

These and similar questions will be recurring themes in the following chapters. Even an apparently simple and straightforward case of a river polluted with a single toxic agent can clearly give rise to some questions which are difficult to answer. It is important, however, to understand the mechanisms by which the observed effects of the pollution are brought about, because this understanding may lead us to the means by which the effects can be remedied or at least ameliorated. The next case study shows that with a suitable combination of field and laboratory studies, detailed investigation of the effects of pollutants can provide an empirical basis for tackling practical problems relating to the control of pollution and the management of polluted waters.

1.3 Toxicity and the Status of Fisheries

Our second case study illustrates the way in which toxicological data obtained from laboratory investigations can be combined with chemical and biological data from field surveys into a predictive model which suggests specific measures to

protect or improve the status of fisheries in polluted waters. The example is particularly instructive because historically, toxicological research has been dominated by the measurement of lethal toxicity. There is, therefore, a widely-held view that a great deal of toxicological research is misdirected, irrelevant or of limited value in the actual management of polluted waters, since the most intractable problems in practice arise from sublethal, rather than lethal, toxic effects. The following example shows that this view is based on an inadequate understanding of the applications of toxicological data.

The account is based upon the application by Alabaster *et al.* (1972) of the results of a long series of investigations carried out by what was then the Water Pollution Research Laboratory, a Government establishment in Stevenage, UK. These investigations were based upon the work of a substantial number of people carried out over a period of nearly 20 years. The results can be synthesised into an empirical relationship between the presence of certain common toxic pollutants and the ecological status of aquatic communities which can be used for specific management purposes. For example, when several pollutants are present, it allows the identification of those which are responsible for the greatest adverse effects, so that pollution control measures can be selectively directed towards those pollutants whose removal would lead to the greatest improvement. It also allows predictions to be made of the likely effects of additional pollution, or of physical changes in the receiving water environment.

In the more heavily polluted rivers of Britain and of similar industrialised countries, the most abundant toxic pollutants are copper, zinc, phenol, cyanides and ammonia. The toxicity of these poisons to the rainbow trout, *Salmo gairdneri*, was first studied in great detail. The rainbow trout was chosen because it is a widely-available species, is amenable to life in the laboratory, and is a fish of considerable commercial importance. Consequently a great deal is known about many aspects of its biology. Additionally, it is sensitive to most toxic pollutants and reacts more quickly than most species to adverse environmental conditions. Within a few years, the lethal toxicity of the common pollutants to this species was reliably determined. The effects of common environmental variables on pollutant toxicity were also studied. In particular temperature, water hardness, pH and dissolved oxygen concentration were found to have significant effects on the toxicity of many pollutants. Finally, methods were devised for the study of the effects of fluctuating concentrations of poisons, and for determining the toxicity of poisons in different combinations of varying composition. Much of this work is described in greater detail in Chapter 4.

A crucial idea in the development of this approach is that of the unit of toxicity, or toxic unit. A toxic unit was defined as the concentration of a pollutant which would kill half of a sample of rainbow trout in 48 hours (i.e. the 48-hour median lethal concentration or 48 h LC50 as described in Chapter 4). Because trout react

quickly to most poisons at lethal concentrations, the 48 h LC50 is close to the lethal threshold concentration, that is, the concentration which would just kill half the sample of fish during an exposure of indefinite duration. A few simple examples will illustrate how toxic units are used.

Assume that, under a certain set of conditions, it is found that the concentration of zinc which will kill half the trout in 48 hours is 2 mg l^{-1} . Under these conditions, one toxic unit of zinc is 2 mg l^{-1} . Now, assume that the experiment is repeated under similar conditions, but in water which has a greater degree of hardness. It may be found that the 48 h LC50 for zinc is now 10 mg l^{-1} . Under these new conditions, one toxic unit of zinc is 10 mg l^{-1} , and 2 mg l^{-1} zinc is now equivalent to only 0.2 toxic units. Now assume that we wish to measure the toxicity of copper to trout. In soft water, one toxic unit of copper may be equal to 0.5 mg l^{-1} and in hard water 2.5 mg l^{-1} . Now assume that we wish to know whether, in the soft water, fish would survive in the presence of 1.5 mg l^{-1} zinc together with 0.25 mg l^{-1} copper. In this case, 1.5 mg l^{-1} zinc equals 0.75 toxic units, and 0.25 mg l^{-1} copper equals 0.5 toxic units. The total number of toxic units is therefore 1.25, whereas we know that by definition, 1 toxic unit will kill half the fish in 48 hours. Therefore we would expect that significantly more than half of the fish would die within 48 hours. In the hard water, however, zinc would contribute 0.15 toxic units, and copper 0.1 toxic units, a total of only 0.25 toxic units. We might therefore expect that the majority of fish would survive these concentrations of zinc and copper in the hard water. These expectations assume, of course, that the effect of the poisons in combination is neither more nor less than the sum of their individual effects. This point has been extensively investigated (see Section 4.2.3) and fortunately, with some important exceptions, appears to be generally true for this group of pollutants. It is not true, however, for all combinations of pollutants. The toxicity of mixtures of poisons is discussed more fully in Chapter 4.

The next stage is to use this technique to estimate the expected toxicity of polluted river water to fish, and to compare the results with the observed status of fish populations. It is not difficult to measure the concentrations of zinc, copper, phenol, cyanides and ammonia in samples of river water. The temperature, pH, hardness and dissolved oxygen concentration of the water at the time of sampling must also be determined. Using these data, the measured concentrations of each pollutant, in mg l^{-1} , can be converted into toxic units. Summing these 'fractional toxicities' will give a measure, in toxic units, of the total toxicity of the river water. Obviously, the toxicity of the river water will vary with time, depending upon the amount of effluent being discharged, the quantity of water available for dilution, and the prevailing environmental factors. It would not be sensible to try to associate the observed status of the fish population of a stretch of river with the toxicity of the water on any single occasion. It is necessary to take a relatively large number of samples over a reasonably long period of time, ideally a whole

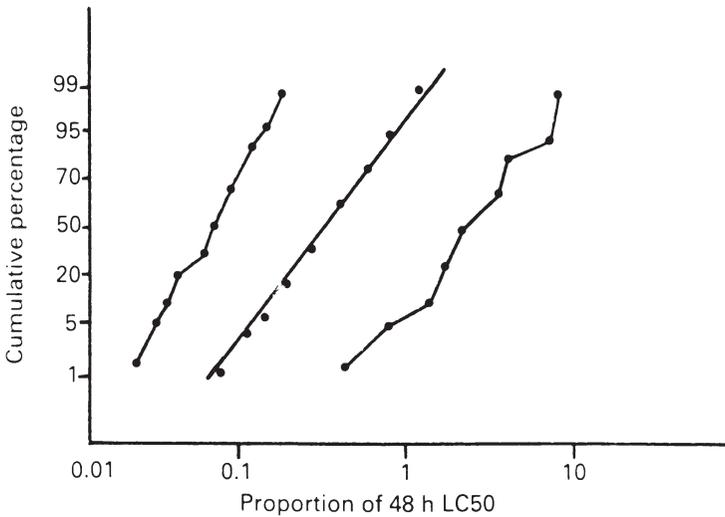


Figure 1.6 Distributions of toxicity calculated by the method of Alabaster *et al.* (1972). The line on the left is typical of a station which supports fish; the line on the right represents a fishless station; and the central line is reconstructed from the boundary distribution of toxicity between fishless and fish-supporting stations in the River Trent catchment. See text for explanation

year, and to calculate the toxicity of the water on each occasion. A graph of the kind shown in Figure 1.6 then is constructed. Each sampling station investigated generates one line on the graph.

The line on the left of Figure 1.6 represents the distribution of toxicity over time at a sampling station which sustains a fish population. It is constructed by plotting the cumulative frequency with which a given level of toxicity occurs in a number of samples taken from the site under investigations over a period of time. It is convenient to plot the graph using a logarithmic scale for the toxicity values and a probability scale for the y-axis. Thus, in the example shown, on 5% of sampling occasions the toxicity was equal to or less than 0.04 toxic units. On 20% of occasions, it was equal to or less than 0.055 toxic units, on 50% of occasions it was equal to or less than 0.08 toxic units, and so on. The line on the right of Figure 1.6 represents toxicity distribution typical of a fishless station. The increased toxicity of the water is represented by a shift of the line towards the right. Plotting a large number of these lines (one for each of the sampling stations studied) showed that the lines fell in various places on the diagram, depending upon the distributions of toxicity recorded at each station. However, lines corresponding to stations where fish were normally present were grouped to the left of the diagram, and lines corresponding to stations which were normally fishless were grouped to the right; a line can be constructed through the

Table 1.5 Coordinates of approximate boundary distribution of 48 h LC50 between fishless and fish-supporting waters in the River Trent catchment area, England. See Figure 1.5 and text for explanation. Data from Alabaster *et al.* (1972)

Per cent probability	1	5	10	25	50	75	90	95	99
Sum of proportions of LC50	0.07	0.10	0.13	0.18	0.28	0.41	0.60	0.73	1.1

narrow zone of demarcation between the two groups. From this line it is possible to read off the ‘coordinates of boundary distribution between fishless and fish-supporting waters’ (Alabaster *et al.*, 1972). The central line in Figure 1.6 has been reconstructed from the coordinates determined by Alabaster *et al.* (1972) for sampling stations in the catchment area of the River Trent. The values of the coordinates are shown in Table 1.5.

These data show that a water will only sustain a fish population if for at least 50% of the time the toxicity of the water is less than 0.28 toxic units; and if for 90% of the time its toxicity is less than 0.6 toxic units, for 95% of the time less than 0.73 toxic units, and so on. Where the toxicity of the water exceeds 1.0 toxic unit for as little as 2 or 3% of the time, fish will generally be absent. The simplest way to determine whether or not a sampling station is likely to support fish is to draw the line of toxicity distribution. If the line, or any substantial portion of the line, falls to the right of the boundary line, the water will be unlikely to support fish. The further to the left the line falls, the smaller is the likely effect of toxic pollution on the fish population. Where the line falls to the left of the boundary but close to it, the fishery may be of only marginal quality.

Although this relationship between toxicity and fishery status is purely empirical, it has considerable potential value in water management, as the following examples show. Assume that a stretch of river is fishless, and that we wish to know the most effective means of re-establishing a fish population. Chemical analysis of the water on a number of occasions reveals that several pollutants are present, and that the dissolved oxygen concentrations are generally low. Many poisons are more toxic at low dissolved oxygen levels. Could the fishery be restored by increasing dissolved oxygen levels, or is the removal of specific toxic substances required? In the toxicity distribution represented by the line on the right of Figure 1.6, the toxicity has been calculated using the dissolved oxygen calculations measured on each sampling occasion. These values can be recalculated, on the assumption that the dissolved oxygen concentration will be increased to any desired level. A new line can therefore be constructed using the hypothetical oxygen values, and this line will be displaced to the left.

If the displacement carries the line beyond the boundary distribution, it is expected that a fishery could be re-established by increasing the levels of dissolved oxygen in the water. This could be achieved relatively easily and cheaply, for example by imposing stricter controls on the discharge of sewage or organic wastes which cause reduced dissolved oxygen levels; by upgrading sewage treatment plants; or by utilising one of the various methods available for aerating rivers such as constructing a weir. (Any of these strategies are also likely to reduce problems associated with ammonia levels, since ammonia is produced in significant quantities by the decay of organic matter.)

If, however, the displacement does not carry the line beyond the boundary distribution, the desired effect is likely to be achieved only by the removal of specific toxic substances, which may be more difficult and expensive. A similar line of reasoning can be applied in situations where, for example, it is proposed to site a new discharge on a river and we wish to predict the likely effect of the new discharge. The new discharge may contain additional toxic pollutants, and/or may alter the temperature, dissolved oxygen or other environmental characteristics of the receiving water. If the effect of the proposed new conditions is significantly to shift the existing distribution of toxicity towards the boundary conditions, or to carry the existing distribution beyond the boundary conditions, the decision may well be made that the proposed new discharge is unacceptable, or should be subjected to more rigorous control.

A further application of this technique is the determination of the relative contributions of each of several pollutants to the overall toxicity of the water. It is frequently the case that several pollutants are present in significant quantities, but chemical analysis alone cannot reveal which of them is exerting the most serious adverse effect on the biota. If the most significant pollutants can be identified, specific control measures may be directed against those, rather than against pollutants whose biological impact may be small, thus affording greater efficiency in the allocation of resources. Alternatively, we may wish to know whether the addition of a new pollutant, or an increase in the expected concentration of an existing pollutant, is likely to have any serious effect. Since the fractional toxicities of each pollutant are initially determined separately, plotting them in the manner shown in Figure 1.7 allows these questions to be answered.

In one example given by Alabaster *et al.* (1972), the toxicity distribution of ammonia alone was plotted, giving a line similar to that shown on the left of Figure 1.7. A second line, showing the toxicity distribution of metals and ammonia combined, displaced the line significantly to the right. This indicates that metals were contributing substantially to the overall toxicity of the water. A third line, showing the effect of including cyanide in the calculation, similarly indicated a smaller, but substantial contribution from cyanide. Phenol, however, contributed little to the overall toxicity, although it was present in the water; its inclusion in the calculation gave only a very slight displacement to the right. Using the same method, the authors were able

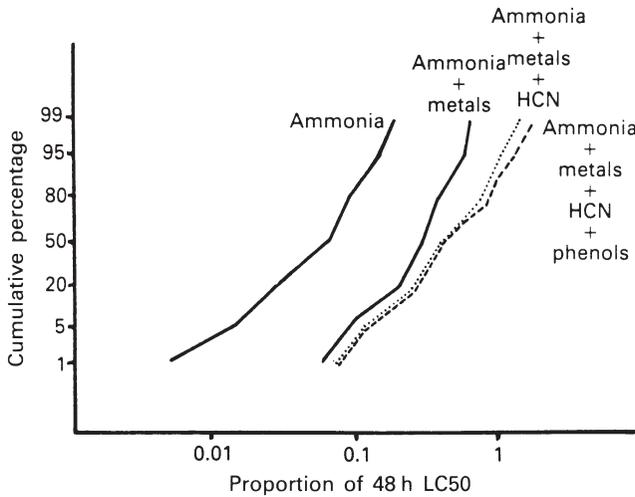


Figure 1.7 Contribution to toxicity from ammonia, metals, hydrogen cyanide and phenols at a heavy-polluted site in the River Trent catchment (Alabaster *et al.*, 1972)

to show that among the metals present, copper and zinc were identifiable as major pollutants in the sites they studied, whereas nickel, chromium and cadmium contributed little to the overall toxicity of the water. This was in spite of the fact that nickel alone formed 20% of the total metal present in chemical analyses. However, nickel and, in particular, cadmium are known to be very slow-acting poisons (see Chapter 4). The model in use relies upon the 48-h LC50 as the definition of the toxic unit, and this is only satisfactory if the 48-h LC50 is a good approximation of the lethal threshold concentration. Since this is not the case for nickel and cadmium, the calculations were revised using the lethal threshold concentration as the definition of the toxic unit. Under these circumstances, the results indicated that nickel and cadmium (but not, in the cases studied, chromium) were significant pollutants in some instances.

The synthesis of data from field and laboratory studies into the potentially useful model for the management of polluted waters has many advantages. Toxicological data, particularly those derived from the study of lethal toxicity, can rapidly be accumulated and replicated under controlled conditions, but are difficult to apply to real situations in the field because our knowledge of ecotoxicological mechanisms is inadequate. We might expect, for example, that if the level of pollution in the water was equal to the 48-h LC50, that half or more of the fish would die and that sublethal toxicity and mechanisms involving the relationships of fish with other members of the aquatic community might be sufficient to account for the total absence of fish from a particular site. The empirical finding that fish are absent if the toxicity exceeds 1.0 toxic unit for 1% of the time, or exceeds 0.07 toxic units for 99% of the time (Table

1.5) is, however, more directly useful to us. It does not actually matter, in the day-to-day management of the water, why exactly this happens—whether, for example, it arises through sublethal toxicity, avoidance by the fish of specific adverse conditions which they have detected, or because the pollutants are adversely affecting the food organisms of the fish. Knowledge of ecotoxicological mechanisms is ultimately valuable because it allows the interpretation and application of data in the frequent circumstances that decisions have to be made on the basis of incomplete information. In these same circumstances, purely empirical relationships are also useful for precisely the same reasons.

An alternative approach would be to rely entirely upon data from field surveys. In principle, any particular management decision could be formulated with reference to experience of what has happened previously in a similar situation elsewhere. In practice, the database of previous information which would be required is so vast that even if it existed, the task of collating it and analysing it in such a way as to extract from it the specific information required would be impossible. In any case, new situations are constantly arising; and water bodies differ so widely in their physical, chemical and biological characteristics that purely anecdotal evidence is of limited predictive value without some attempt to understand and apply fundamental principles. In practice, the control and amelioration of the problems raised by water pollution are best achieved by a combination of approaches—field based and laboratory based, empirical and fundamental.

Sources and Effects of Water Pollutants

There are hundreds, perhaps thousands of pollutants whose effects are of actual or potential concern. Their numbers increase annually, as new compounds and formulations are synthesised. A substantial minority of these find commercial applications and become significant pollutants of water during their manufacture and in subsequent use. It is clearly impossible within the scope of a short book to discuss all of these in detail; instead, the sources and effects of broad categories of pollutants will be discussed in general terms, and where appropriate reference will be made to further sources of information.

2.1 The Nature of Effluents

Water pollution is most commonly associated with the discharge of effluents from sewers or sewage treatment plants, drains and factories. Outfalls of this kind are known as ‘point-source discharges’. Most cases of accidental, negligent or illegal discharge are also from point sources. The concentration of pollutant in the receiving water is initially high, decreasing as the distance from the point of discharge increases. The effects of the pollution are therefore frequently easy to observe. Some of the more serious forms of pollution arise, however, from ‘diffuse’ sources, that is the pollutant does not enter the water from a single point. For example, in agricultural areas, surface water runoff and groundwater infiltration into lakes and rivers can introduce plant nutrients (from fertilisers) and pesticides in substantial quantities to water bodies. The effects of pollution from diffuse sources can be serious, but are often less immediately obvious than those from point sources as there is no adjacent unpolluted area with which comparisons may

Table 2.1 Analysis of sewage effluents after primary and secondary treatment. Figures given are in mg l^{-1} , and are the range of values found at four different treatment works, as summarised by Bond and Straub (1974). Treatment plants operating under adverse conditions yield higher values than those shown here, but values at the upper end of the ranges shown would in many countries exceed modern effluent standards

Analysis	Range
Total solids	640–1167
Suspended solids	15–51
Biological oxygen demand	2–70
Chemical oxygen demand	31–155
Organic carbon	13–20
Anionic detergents	0.75–1.4
Ammonia	1.9–22
Nitrate	0.25–38
Nitrite	0.2–1.8
Chloride	69–300
Sulphate	61–270
Phosphate	6.2–9.6
Sodium	144–243
Potassium	20–26

be made. Many pollutants also enter water through fallout from the atmosphere. Historically, control and prevention of water pollution have concentrated on point sources as these are more obvious, easily identifiable and in theory easier to regulate at the point of origin. As awareness has increased of the significance of diffuse sources of pollution, control strategies have been under development but are based more on the application of good practices designed to reduce pollutant impact rather than on regulation of specific sources of input (see, for example, MAFF, 1991).

Most effluents are complex mixtures of a large number of different harmful agents. These include toxic substances of many kinds, extreme levels of suspended solids, and dissolved and particulate putrescible organic matter. In addition, many effluents are hot, of extreme pH value, and normally contain high levels of dissolved salts. Detailed compilations of data on the composition of sewage and industrial effluents of many kinds are given by Bond and Straub (1974), and by Sittig (1975). Some representative values for treated sewage effluent are given in Tables 2.1 and 2.2. Most effluents also vary in their strength and composition, on a seasonal, diurnal or even hourly basis. Most sewage treatment plants report regular diurnal peaks and troughs in their output according to patterns of water use. Sometimes storm-water drains are connected to the sewerage system, so the strength of the sewage effluent will vary with rainfall. Alterations in the