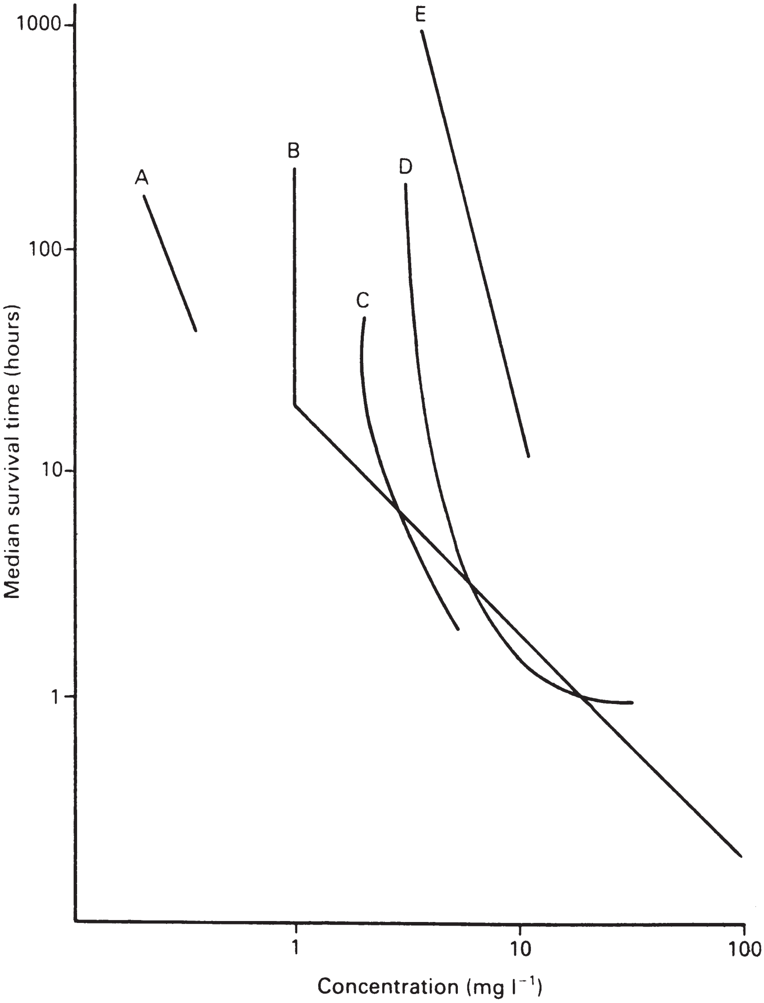

Figure 4.6 Some toxicity curves for various species of fish exposed to different detergents, redrawn from the originals to the same scale. A, *Salmo gairdneri*, linear alkylate sulphonate (Brown *et al.*, 1968); B, *Salmo salar*, polyoxyethylene lauryl ether ((Wildish, 1972); C, *Lepomis macrochirus*, linear alkylate sulphonate (Hokanson and Smith, 1971); D, *Gadus morrhua*, alkylbenzene sulphonate (Swedmark *et al.*, 1971); E, *Salmo gairdneri*, alkylbenzene sulphonate (Herbert *et al.*, 1957)

Thus the curve will become asymptotic to the time axis. The concentrations at which this occurs may be termed the *threshold median lethal concentration* or threshold LC50. (A synonymous term is *incipient lethal level* or ILL.) Further, even at very high concentrations death will not be instantaneous,

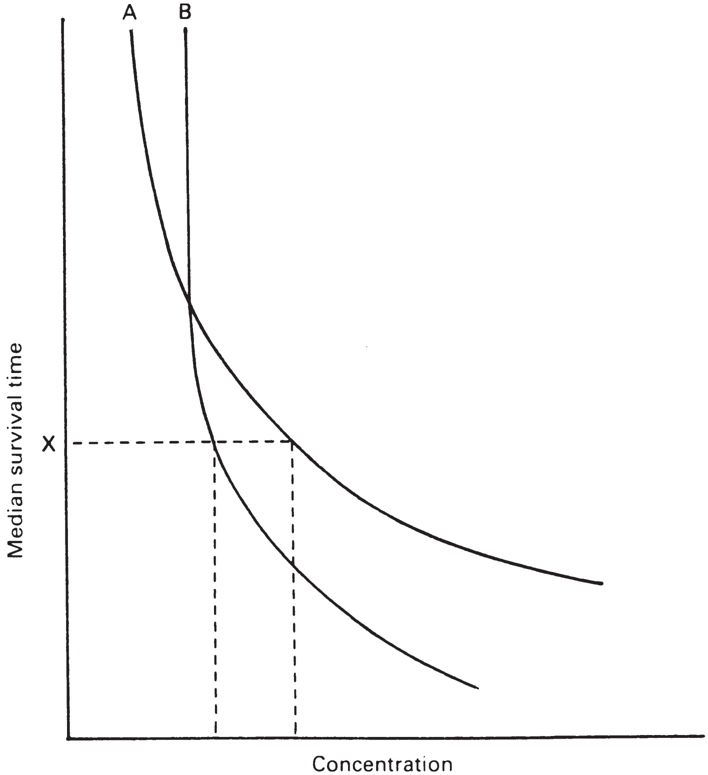

Figure 4.7 Two hypothetical toxicity curves. Substance A is clearly more toxic than substance B, since the lethal threshold concentration of A is smaller. However, had the test been discontinued at time X, B would have appeared more toxic than A

but will take a finite period of time to occur. Thus the curve will become asymptotic to the concentration axis. Between these two asymptotes, the curve generally will show a decrease in survival time as concentration increases. Whether the toxicity curve is a straight line or a curve is therefore of no significance. Where a straight line is obtained, it may be considered as a segment of a larger curve. This point is illustrated by Brown (1973), and also in Figure 4.6. Here, toxicity curves for several species of fish exposed to some synthetic detergents have been redrawn from the originals to the same scale. Clearly, no significance can be attached to the fact that, for example, curves A and E are rectilinear while C and D are curvilinear. Most of these curves, and A and E in particular, cover only part of the range over which survival time changes with poison concentration. More extensive testing over a wider range of concentration is required to establish the true shape of the curves.

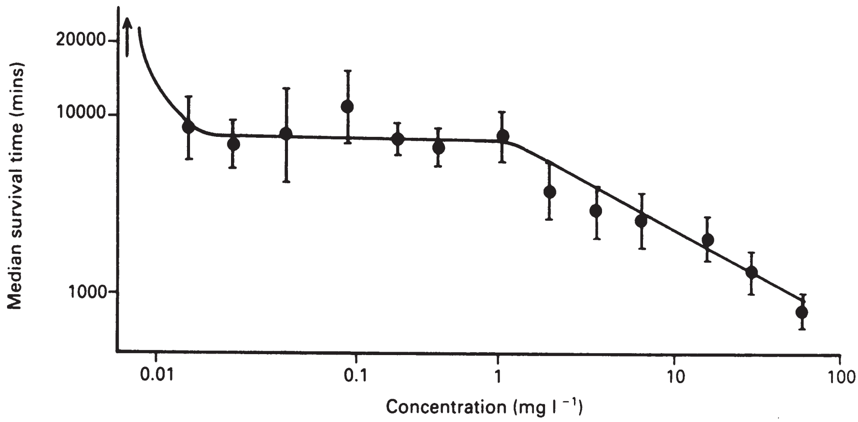

Figure 4.8 Toxicity curve for rainbow trout (*Salmo gairdneri*) exposed to cadmium (Ball, 1967b)

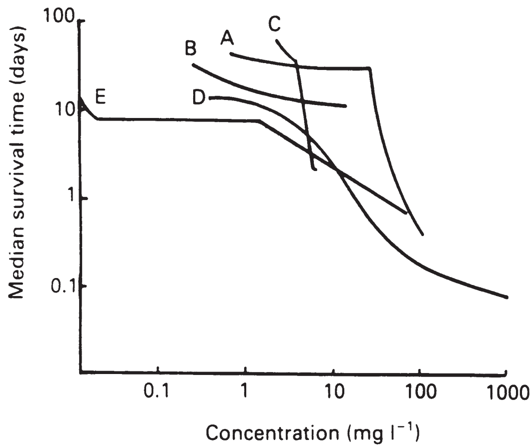

Figure 4.9 Toxicity curves for several fish species exposed to cadmium, redrawn to the same scale. Note that none of the curves shows a clear threshold concentration, despite the long duration of most of the tests. Inflections in the curves, and segments of curve over which mortality is apparently unrelated to poison concentration, commonly occur. Finally, the curves for different species frequently intersect: which species appears most, or least, sensitive depends upon the duration of the toxicity test. A, *Tilapia aurea* (Abel and Papoutsoglou, 1986); B, *Cyprinus carpio* (Abel and Papoutsoglou, 1986); C, *Noemacheilus barbatulatus* (Solbé and Flook, 1975); D, *Gasterosteus aculeatus* (Pascoe and Cram, 1977); E, *Salmo gairdneri* (Ball, 1967b)

The most important feature of a toxicity curve is the indication it gives of the threshold median lethal concentration, and wherever possible tests should be continued until a lethal threshold is apparent. Figure 4.7 shows clearly the importance of determining lethal threshold concentrations. In this diagram, two toxicity curves are shown. These may represent two poisons tested against the same species, or one poison tested against one species under different environmental conditions. Clearly A is more toxic than B, since the lethal threshold concentration of A is smaller than that of B. If, however, the experiment had been terminated arbitrarily at time X, as shown on the diagram, it would be erroneously concluded that B is more toxic than A. Therefore unless complete toxicity curves are obtained, and lethal thresholds established, the results of any test must be interpreted with caution. This is particularly important where comparisons between species, poisons or environmental conditions are involved. For example, Ball (1967a) found that in tests lasting one day, trout were more sensitive to ammonia than coarse fish. However, when the tests were continued for up to five days, all fish were found to be equally sensitive, the difference between trout and coarse fish being simply that the latter were more slow to react.

Apart from being the only proper basis for comparative studies of lethal toxicity, lethal thresholds are also useful, in conjunction with other information, in setting water quality standards (see Chapter 6). A major criticism of many investigations of lethal toxicity is that experiments are not continued long enough. Sprague (1969) examined 375 published measurements of lethal toxicity and found that only 211 of these showed a lethal threshold within four days. In 122 cases the time required to show a lethal threshold was between four and seven days, and 42 cases required longer than this. Although the results of shorter experiments are by no means invalid, it is nevertheless clear that unless lethal thresholds are clearly established the interpretation which may validly be put on the results is strictly limited. Further, useful toxicological information may be lost. Ball (1967b) reported an example of the advantages of continuing tests as long as is economically or practically feasible (Figure 4.8). The toxicity curve for rainbow trout exposed to cadmium was linear over a concentration range between 1 and 64 mg $l\text{-}^{1}$ and a time period of about six days. Continuing the test for 14 days revealed that cadmium continued to act lethally down to 0.01 mg l^{-1} and that the threshold concentration may lie as low as 0.008 mg l^{-1}. Fish exposed to concentrations between 0.01 and 1.0 mg l^{-1} continued to die throughout the latter part of the experiment, and between these two concentrations survival time did not increase as the cadmium concentration decreased. These results demonstrated that cadmium was a very slow-acting poison which was considerably more toxic than shorter tests had previously indicated. Toxicity curves for five fish species exposed to cadmium have been compared (Abel and Papoutsoglou, 1986) and show some interesting features (Figure 4.9).

4.1.5 Alternative Methods for Measuring Lethal Toxicity

As indicated earlier, there are many different reasons for measuring the toxicity of pollutants to aquatic organisms, and it is important to use a method which is appropriate to the purpose for which the results are required. The procedures described above are unnecessarily elaborate for some purposes, and similarly there are circumstances in which these conventional methods provide inadequate information. It is therefore sometimes necessary to consider alternative methods, on the one hand simpler, and on the other more complicated, than those discussed already. The modifications may be to the experimental apparatus, to the collection of data, to the methods of data analysis, or to the duration of the test. The most common reasons for modifying a test procedure are:

1 *The purpose of the test:* for example, routine screening and monitoring of effluents do not necessarily require the same level of complexity as toxicity measurements designed to contribute to the formulation of a water quality standard. Tests required for legal or quasi-legal purposes such as certification of toxic chemicals for use in water, or for purposes of meeting consent conditions for discharge of wastes, usually must conform to the relevant procedures laid down by the regulatory authority. This type of test is usually kept as simple as possible, consistent with producing useful data, as the results may become subject to legal challenge on the grounds that the procedures were incorrectly followed. However they are usually not suitable for any purpose other than that for which they were designed.

2 *The characteristics of the pollutant:* for example, certain substances have unusual physical or chemical properties, such as low solubility, immiscibility with water, volatility, susceptibility to adsorption or degradation, or other reasons which affect their behaviour in the test and which will require special steps to be taken to ensure that the final result is relevant to the way in which the chemical will behave in the environment. Examples include oil dispersants and certain pesticides. It is also important in some circumstances to consider the pattern of use or discharge of the chemical. For example, a test which assumes continuous exposure of organisms to the chemical will generally produce false results if the organisms are exposed only intermittently. This is particularly relevant to certain forms of pesticide applications, and to estimating the impact of effluent plumes or mixing zones on the receiving water biota (Abel, 1980a, b; Abel and Garner, 1986).

3 *The characteristics of the test organisms:* for example, some organisms have special requirements by virtue of their size, behaviour or environmental requirements which may dictate modifications to the test procedure.