Chapter 9

Natural Products

Although many of the toxic chemicals in the environment that worry the general public are man-made, there are also many hundreds of natural poisons of animal, plant, fungal and microbial origin. Indeed, the most toxic substances known to man are natural poisons such as botulinum toxin (Table 1.1) and it is certainly not reasonable to imply, as do some of the advertisements for health foods and herbal medicines, that natural substances are intrinsically harmless and safe. For example, allergies to natural constituents of food are known to occur just as they do to synthetic additives. Some of these natural, toxic substances have been known about for centuries and have been used for murder or suicide, or even sometimes misguided medical treatment (see Chapter 1).

Natural substances also still occasionally feature in accidental poisoning cases although this is relatively rare compared with poisoning by drug overdose. Natural toxins are of diverse structure and mode of action, and there are far too many categories to consider each individually in this book. Consequently we will simply examine a few interesting and important examples of toxic substances derived from plants, animals, fungi and micro-organisms.

Plant Toxins

There are many well known plant toxins ranging from the irritant formic acid found in nettles (and ants) to more poisonous compounds such as atropine in deadly nightshade berries *(Atropa belladonna),* cytisine in laburnam and coniine in hemlock. Let us consider a few less well-known plant toxins which have been studied recently.

Pyrrolizidine Alkaloids

There is a group of these alkaloids which are produced by plants of the *Senecio, Heliotropium* and *Crotolaria* species, many of which occur as weeds throughout the world. The plants have on occasion contaminated cereal crops and human consumption of flour made from them led to poisoning. This has occurred in

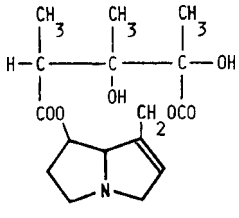
various parts of the world, especially where agricultural conditions are poor and the indigenous population may be forced to use the contaminated crops. For example, in South Africa during the 1930s poor whites suffered the toxic effects of these alkaloids because their staple diet was wheat which became contaminated, whereas their Bantu neighbours, who ate maize which was not contaminated, were not affected. More recently, poisonings have occurred in Tashkent, Central India and Northern Afghanistan. In one incident where 1600 poisoning cases were reported, the threshed wheat was found to be contaminated with *Heliotropium popovii* seeds giving an alkaloid concentration of at least 0.5 per cent. In the West Indies especially, these plants may also be used in traditional medicine to make herbal teas.

These alkaloids are interesting because after acute exposure, such as after the ingestion of herbal teas, they cause a particular form of liver disease known as veno-occlusive disease. The effect of chronic exposure to low doses is liver cirrhosis which can be seen in some members of the West Indian population, estimated to account for one third of the cirrhosis seen at autopsy in Jamaica. The constituent alkaloids, such as monocrotaline which has been extensively studied, (Figure 9.1), undergo metabolic activation to a reactive metabolite which damages the cells lining the liver sinusoids as well as the hepatocytes, leading to haemorrhagic necrosis and finally to the veno-occlusive disease. This blockage of the blood vessels in the liver eventually gives rise to alteration of the vasculature such that the liver blood supply is diverted and new blood vessels grow.

Animals may also be exposed and suffer the toxic effects. Where there is abundant vegetation for grazing, animals will ignore plants such as ragwort *(Senecio jacoboea)* which contain the alkaloids but in some countries, such as Australia, widespread losses of horses, cattle and sheep have occurred from heliotropium poisoning. This may also be another route of human exposure as the alkaloids can be detected in the milk of cows grazing on such plants.

Pennyroyal Oil

The Pennyroyal plant and the oil prepared from it have been used to induce abortions in the USA where it is possible to buy the oil 'over the counter'. The plant may be used to make a tea or the oil may be taken directly. Both may cause toxic effects, especially liver damage as well as inducing abortion. The



Monocrotaline

Figure 9.1. The structure of the pyrrolizidine alkaloid monocrotaline.

oil contains a number of terpenoid compounds and metabolic activation is believed to be required for the toxicity.

Ricin

Ricin is a highly toxic plant product found in the seeds of the castor oil plant. It achieved some notoriety when it was claimed that it had been used by the Bulgarian secret police to kill the Bulgarian journalist Georgi Markov in London in 1978. Although no trace of any poison was found in the victim's body clearly an extremely potent poison had been used and the symptoms were consistent with those of ricin poisoning. A tiny metal pellet was recovered from a wound on the victim's leg, seemingly inflicted accidentally by an umbrella. The pellet almost certainly was a reservoir for a toxic substance, but it could only contain a few nanograms of the substance.

Ricin is a small protein consisting of two polypeptides, a short A chain and a longer B chain. The A and B chains are linked via a disulphide bridge. The B chain attaches the ricin molecule to the outside of the mammalian cell by binding to the galactose part of a glycoprotein. The cell membrane invaginates and the ricin is taken into the cell inside a vacuole. The ricin molecule is released from the glycoprotein and the A and B chains then break at the disulphide bridge. The B chain makes a channel through the vacuole cell wall, allowing the A chain to enter the cytoplasm and reach the ribosomes where it blocks protein synthesis and kills the cell. One molecule of ricin is sufficient to kill one cell.

Bracken

The bracken fern contains a substance, ptaquiloside, which degrades into a compound which is carcinogenic. In Japan, the shoots of the bracken fern are eaten and this may explain the high incidence of throat cancer among the Japanese. Animals which eat the fern as fodder suffer from bladder and intestinal cancer. The breakdown product of ptaquiloside reacts with DNA, specifically the base adenine, and this is lost with the result that the DNA chain breaks.

Fluoroacetate

For a description of the toxicity of this naturally occuring plant toxin see Chapter 7.

There are many more well-known substances derived from plants such as the drugs heroin, morphine, cannabis, nicotine and digitalis.

Animal Toxins

As with plant toxins, animal toxins comprise a diverse range of structures and modes of action (Figure 9.2). A simple and well-known example is formic acid

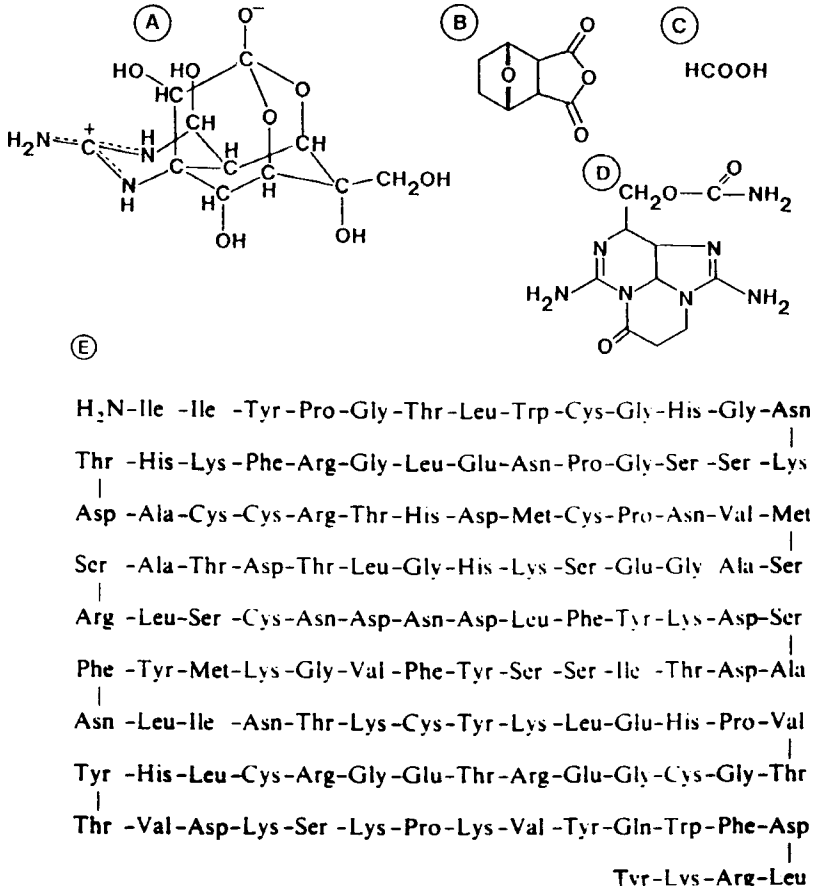


Figure 9.2. The structures of various animal toxins. A: tetrodotoxin; B: cantharidin; C: formic acid; D: saxitoxin; E: Amino acid sequence from honey bee venom phospholipase A.

which is found in ants (the name is derived from the Latin word, *formica,* for an ant). Other examples are tetrodotoxin found in the Puffer Fish and saxitoxin found in shellfish and fish which have consumed certan dinoflagellates. Animal toxins are often mixtures of complex proteins. Most of us suffer from animal toxins at some time in our lives even if it is only a wasp or bee sting. However, in some countries death and illness due to animal poisons represents an important proportion of poisoning cases and a significant cause of illness and death.

Snake venoms

Snake bites are one of the most common forms of poisoning by natural toxins worldwide. Many snake venoms are similar in their mode of action and constituents, being mixtures of proteins or polypeptides. The proteins may be enzymes, especially hydrolytic enzymes. Some of the more important are

proteinases, phospholipases, ribonucleases, deoxyribonucleases, phosphomonoesterases and phosphodiesterases, and ATPases. The toxicity of some snake venoms is shown in Table. 9.1. The venoms may be mixtures and consequently cause a variety of effects. For example, the presence of foreign proteins may cause an anaphylactic reaction, although this is rare, and such allergic reactions may cause death in minutes. The enzyme components can digest various tissue constituents either at the site of action, causing local necrosis, or elsewhere causing systemic effects. For example, the bite of the diamondback rattlesnake, the most poisonous snake in the USA produces a very painful swelling within minutes. Nausea, vomiting and diarrhoea may occur and cardiac effects, such as a fall in systemic arterial blood pressure and a weak, rapid pulse, may be seen. The central nervous system can be affected, leading to respiratory paralysis. Haemolytic anaemia and haemoglobinuria sometimes occur, and there may be thrombosis and haemorrhage. Vascular permeability and nerve conduction can change, and cerebral anoxia, pulmonary oedema and heart failure also develop. The phospholipases found in snake venom sometimes cause intravascular haemolysis by direct action on the red cell membrane.

Most snake venoms contain a phosphodiesterase which attacks polynucleotides.

Tetrodotoxin

This toxin is found in the Puffer Fish and also in the Californian newt and has been studied extensively. The fish is eaten as a delicacy in Japan and provided it is properly prepared is edible and safe. However, fatalities have occurred which resulted from incorrect preparation of the fish and about 60 per cent of poisoning cases are fatal. The tetrodotoxin and another toxin ichthyocrinotoxin are found in the roe, liver and skin of the fish. Tetrodotoxin is a very potent nerve poison, lethal at doses of around 10 μg kg^{-1} body weight. Initial effects are a tingling in the mouth followed within 10–45 minutes by muscular inco-ordination, salivation, skin numbness, vomiting, diarrhoea and convulsions. Death results from skeletal muscle paralysis. Sensory as well as motor nerves are affected and it is believed that

Table 9.1. Comparative toxicity of snake venoms.

Snake venom	Yield (g)	LD_{50} i.v. mg/kg
Copperhead	40–72	10.92
African puff adder	130–200	3.68
Mojave rattler	50–90	0.21
Russell's viper	130–250	0.08
Sea snake	7–20	0.01

Source: F.W.Oehme,J.F.Brown and M.E.Fowler (1980), Toxins of animal origin,, in *Cassarett and Doull's Toxicology,* J. Doull, C.D.Klaassenand M.O.Amdur (Eds), 2nd edition, New York: Macmillan.

tetrodotoxin selectively blocks the sodium channels along the axon, preventing the inward action potential current.

Fungal Toxins

Many fungi produce toxins of a variety of chemical types and these can cause acute or chronic poisoning. Poisonous mushrooms may be confused with the edible varieties and hence accidental acute poisoning may occur. Poisoning may also occur through the intentional eating of fungi believed to contain psychoactive substances. Several fungal toxins have been fully identified and characterized. The toxic effects vary from relatively mild gastrointestinal disturbances to severe organ damage. Some, such as the psychoactive constituents mescaline and psilocin, affect the central nervous system. Certain fungal products, such as the aflatoxins which are discussed in Chapter 6, are potent carcinogens.

Death Cap Mushroom

The Death Cap mushroom, *Amanita phalloides* is probably the most poisonous mushroom in Britain. It is occasionally eaten by mistake, but poisoning with this mushroom is rare in the UK. The mushroom contains a number of toxins: the phallotoxins, including phalloidin, phalloin and phallolysin, and the amatoxins (α, β and γ amanitin). The phallotoxins cause violent gastroenteritis which occurs rapidly (4–8 hours) after the mushroom is eaten.

The amatoxins have a delayed toxic effect, with the liver and kidney as target organs; liver necrosis and destruction of renal tubular cells may result. Both the phallotoxins and the amanitins are strongly bound to plasma proteins yet are toxic in this form. Consequently, treatment involves displacement from the proteins by a drug such as a sulphonamide or benzylpenicillin which will reduce toxicity. This is probably due to increased excretion of the unbound form as protein binding of foreign compounds slows excretion (see Chapter 2).

After the mushroom is eaten sometimes there may not be any symptoms for up to a day then vomiting and diarrhoea occur possibly followed by jaundice, hypoglycaemia, acidosis and other effects on blood chemistry. In severe cases hepatic failure can result.

Aflatoxins

These fungal toxins have already been discussed under food contaminants (Chapter 6). Aflatoxin B1 causes liver damage after high doses but in humans chronic exposure to lower doses via the diet is a more likely occurrence which can cause liver tumours. The toxin is most likely to occur in food infected with the mould *Aspergillus flavus* or prepared from infected food or ingredients.

Microbial Toxins

There are many toxins produced by bacteria. As with other natural toxins, they are of a variety of chemical types and consequently they cause a variety of different toxic effects, ranging from gastrointestinal effects to severe and fatal effects on the nervous system. We will consider just one well known toxic syndrome, botulism.

Botulism and Botulinum Toxin

The toxic bacterial product botulinum toxin is produced by the bacterium *Clostridium botulinum*. The syndrome this causes is known as botulism and results from oral ingestion of the toxin. The bacteria thrive under anaerobic conditions and produce a mixture of six heat-labile toxins. Consequently, botulism is often produced by a bacterial infection of home- canned or bottled non-acid foodstuffs which have become infected during preparation, have been inadequately preserved and not refrigerated, and have not been adequately heated prior to eating. The toxin itself is destroyed by heating although the spores of the bacterium are quite heat-resistant.

This toxin is one of the most potent known to man, with an LD_{50} of around 0.01 μg kg^{-1} and so less than a microgram would be lethal for a human. It acts on the nerve terminal, binding irreversibly to it and preventing the release of acetylcholine. The result of this is that the muscle behaves as if it was denervated and the victim suffers paralysis and fatal cessation of breathing if severe. Although botulism may prove fatal fortunately such poisoning is relatively uncommon.

Question

1. Write short notes related to the toxicology of three of the following:

   (a) pyrrolizidine alkaloids;
   (b) ricin;
   (c) snake venoms;
   (d) tetrodotoxin;
   (e) *Amanita phalloides.*

Bibliography

Emsley, J. (1994) How bracken's deadly chemical breaks the back of DNA, *New Scientist,* **142** (1921), 16.

Habermeyl, G.G. (1981) *Venomous Animals and Their Toxins,* Berlin: Springer Verlag.

Harris, J.B. (Ed.) (1986) *Natural Toxins, Animal, Plant and Microbial,* Oxford: Oxford University Press.

HMSO (1987) Mycotoxins, Food Surveillance Paper no. 18, London: HMSO.

Lampe, K.F. (1991) Toxic effects of plant toxins, in *Cassarett and Doull's Toxicology,* Amdur, M.O., Doull, J. and Klaassen, C. (Eds), 4th edition, New York: Pergamon Press.

Moffat, A.C. (1980), Forensic pharmacognosy—poisoning with plants, *Journal of Forensic Science Society,* **20,** 103.

Rechcigl, M. (Ed.) (1983) *Handbook of Naturally Occurring Food Toxicants,* Boca Raton: CRC Press.

Russell, F.E. and Dart, R.C. (1991) Toxic effects of animal toxins, in *Cassarett and Doull's Toxicology,* Amdur, M.O., Doull, J. and Klaassen, C. (Eds), 4th edition, New York: Pergamon Press.

Twigg, L.E. and King, D.R. (1991) The impact of fluoracetate-bearing vegetation on native Australian fauna: a review, *Oikos,* **61,** 412–430.

Wedin, G.P. (1993) Poisons of animal origin, in *General and Applied Toxicology,* Ballantyne, B., Marrs, T. and Turner, P. (Eds), Basingstoke, UK: Macmillan.