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DETERMINANTS OF TOXICITY

ANDRÉ E. M. MCLEAN
Department of Experimental Pathology, University College Hospital Medical School, University Street, London WCIE 6JJ

Abstract—The factors that render individuals or species susceptible to toxins are considered. Toxic chemicals combine with cell sites, either to exert their toxic effects, or else to undergo an activation step. This initial interaction will usually depend on the protein structure of the site and so be determined genetically, by the process of differentiation, and by enzyme induction. These concepts are examined with reference to the effects of nutritional state and enzyme induction on the toxicity of carbon tetrachloride.

IT HAS been thought for a long time that there must be some fit between the highly specific poisons and their target cells. In some instances the toxin interacts directly with a specific protein and inhibits a necessary metabolic pathway (e.g. fluorocitrate–aconitase[1]). In other instances the poison that is administered is itself a fairly inert chemical which is converted to the active form by an enzyme system, the metabolite then reacting with other vital parts of the cell. The destructive end of the process is largely unknown, perhaps membranes, nucleic acids, or proteins are altered and some aspect of cell behaviour is lost, but at the beginning is a specific interaction.[2,3]

When a group or population are exposed to some hazard, such as infection, a poison inadvertently put into the food (e.g. "Epping Jaundice"[4]) or, a carcinogen in the environment, (dimethylnitrosamine in the drinks,[5] smoking, or aflatoxin) some members of the exposed population die, or are severely injured, while others suffer little or nothing. This variability can be attributed in part to different degrees of exposure, but we know from clinical, and experimental experience that even with equal exposure to hazard, the response remains very variable. This variance in resistance or sensitivity is clearly contributed by the target organism; the very term "median lethal dose" recognises that there is a wide range of doses for any toxin, within which a proportion of poisoned animals will die while others recover.

For many years this phenomenon has been designated "biological variation", but three concepts developing in separate fields of biology can now be seen to provide some explanation for the variation of response to poisons.

There is abundant evidence that the amino acid sequence of homologous proteins varies between species and often between individuals of the same species, and this variation is genetically determined.[6–9] Such variation will allow a poison to attack only the species or individual that carries the right binding site. When a poison is far more toxic for one species or strain than another we can look for a difference between the proteins that form the site of initial interaction of the toxin. Moreover if we know which protein a poison must interact with, in order to produce its toxic effect in one species, we can look at other species, such as man, and see if they possess the relevant proteins.

Secondly, the organ specificity of poisons can now be linked to the organ specificity of proteins, which develops during differentiation.[10] The other structures of cells, such as lipid membranes and nucleic acids, seem to undergo less specific differentiation, and poisons that attack these structures would be expected to have less organ specificity. The third concept which helps us to understand the variability of intoxication is that of enzyme induction.

Organisms react to their environment, and that environment determines the enzymatic structure of the organism. It is no longer possible to speak of a "normal" rat or a "normal" man. We can speak of a rat in a laboratory environment, fed a particular diet with a particular amount of DDT and protein in that diet. Or we can speak of European man with his high protein, high fat diet and contrast him with his American or African counterpart with their very different diets.[11]

Our chemical environment is predominantly made up of food or the lack of it, this together with social patterns of rest, sleep and exercise determine the enzyme patterns with which toxic agents will interact.

The determinants of toxic action are first, the genetically determined protein composition of the species and individual, secondly the way this is varied during organ differentiation, and thirdly the influence of enzyme induction. All these go to providing the appropriate binding site for toxin–host interaction.

If we look at a specific instance we can test out this scheme. Carbon tetrachloride (CCl_4) reacts with the cytochrome P450 of liver microsomes.[12] The cytochrome is part of a hydroxylating enzyme system found mainly in liver but also in kidney, lung and to a small extent in other tissues.

The interaction between CCl_4 and the cytochrome can be detected by a marked shift in the absorption spectrum of the heme, and also by the rapid conversion of CCl_4 to CO_2 by the microsomal hydroxylating enzyme system.[13]

The newborn rat has practically no microsomal hydroxylating enzymes in the liver, and is immune to CCl_4 hepato toxicity.[14–16] In this instance the stage of differentiation of the animal determines the pattern of cell proteins, and hence the susceptibility to cell injury by CCl_4.

The adult rat, and sheep when fed usual stock diets are well supplied with liver cytochrome P450, and suffer severe liver damage when exposed to CCl_4. However, when fed a low protein diet liver cytochrome P450 falls, and CCl_4 toxicity is greatly reduced. Exposure to phenobarbitone or DDT induces synthesis of P450, and toxicity of CCl_4 is enhanced.[17, 18]

In qualitative terms the scheme fits. More P450 leads to more liver damage after

TABLE 1. THE EFFECT OF DIET AND ENZYME INDUCTION ON THE TOXICITY OF CARBON TETRACHLORIDE

Diet	Treatment	Microsomal cytochrome (P450 nmoles/g liver)	LD_{50} CCl_4 (ml/kg)
Stock	+ Phenobarbitone	135	0·5
Stock	+ DDT	57	4·2
Stock	—	20	6·4
No protein	+ DDT	28	4·3
No protein	—	6	14·7

CCl_4 as judged histologically, by measurement of liver glycogen and water content, or measurement of serum enzymes or bilirubin.

In the single species for which we have much information, the rat, the amount of CCl_4 that is metabolised seems to be the only determining factor. There is no evidence for instance, that the protein depleted animal is more susceptible to death from a given amount of liver damage, than is the well fed rat. This is surprising; one would expect some "general factor" of resistance to be influenced by protein depletion. One instance where some such general factor is found, is in the adrenalectomised animal. Animals are notoriously unable to resist injury after adrenalectomy. The detailed mechanism of this effect is not clear. We have a long way to go before we can fit a description of toxicity and response to injury within normal biochemistry and physiology.

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