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LETTER TO THE EDITOR

The redirection of glyceride and phospholipid synthesis by drugs including chlorpromazine, fenfluramine, imipramine, mepyramine and local anaesthetics

At present there is considerable interest in the possibility of controlling lipid metabolism through the use of drugs. This is immediately relevant to the treatment of obesity and hyperlipidaemia. In addition, the synthesis and turnover of phospholipids seems to be important for receptor function (Hawthorne, 1973; Michell, 1975), and many drugs which alter the functioning of the central nervous system seem to interfere with phospholipid metabolism or to interact with phospholipids themselves. In this letter we consider a group of amphiphilic cationic drugs and offer an explanation for many of their effects on lipid metabolism.

Compounds such as chlorpromazine, desipramine, cinchocaine, fenfluramine and mepyramine have been shown to inhibit phosphatidate phosphohydrolase (Brindley & Bowley, 1975a, b). The enzyme was from rat liver and the substrate was phosphatidate which was bound to endoplasmic reticulum membranes. This inhibition may be particularly significant since phosphatidate phosphohydrolase shows the properties of a regulatory enzyme in the synthesis of hepatic and possibly neuronal glycerolipids (see Brindley & Bowley, 1975a for refs). This enzyme lies at a branch-point in the biosynthetic pathways to glycerolipids (Fig. 1). The entire phosphatidate molecule can be incorporated via cytidine 5′-pyrophosphate (CDP)-diglyceride into phosphatidylinositol, phosphatidylglycerol and diphosphatidylglycerol (Route *a*). These lipids are all acidic, and bear a net negative charge. In *a*, the synthesis of triglyceride, phosphatidylcholine and phosphatidylethanolamine (Route *b*), however, the phosphate group is removed by phosphatidate phosphohydrolase and only the resulting diglyceride is incorporated into the final lipid molecule (see Fig. 1).

The effects of the amphiphilic cationic drugs on lipid metabolism in a number of tissues are compatible with an inhibition of phosphatidate phosphohydrolase. When liver slices are treated with a number of fenfluramine derivatives, the synthesis of triglycerides and of phosphatidylcholine (Route *b*, Fig. 1) is depressed and phosphatidate accumulates (Brindley & Bowley, 1975a, b). In other tissues, it is not phosphatidate, but the lipids which are derived from it via CDP-diglyceride (Route *a*, Fig. 1), which show marked accumulation. For example, using lymphocytes incubated with labelled phosphate or glycerol, several amphiphilic cationic drugs produce massive accumulations of radioactivity in phosphatidylinositol, and at the same time decrease the labelling of triglyceride, phosphatidylcholine and phosphatidylethanolamine. In these drug-treated lymphocytes the increased rate of phosphatidylinositol synthesis was sufficient to cause a doubling in the concentration of this lipid in the cells within 3·5 h (Allan & Michell, 1975). The other lipids which might also be expected to accumulate as a result of inhibiting phosphatidate phosphohydrolase were not detected in appreciable quantities in the lymphocyte experiments. However, Eichberg, Shein & Hauser (1973a); Eichberg, Shein & others (1973b) and Eichberg & Hauser (1974) have carried out similar experiments with pineal gland, using propranolol and local anaesthetics, and have detected a considerable increase in the labelling of CDP-diglyceride, phosphatidylglycerol and diphosphatidylglycerol. The drugs which have been reported to lead to the accumulation of acidic phospholipids in a

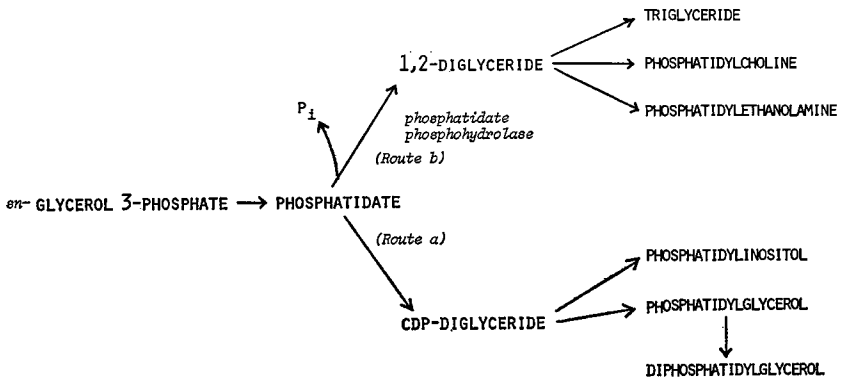

FIG 1.

variety of tissues include phenothiazine neuroleptics, imipramine antidepressants, local anaesthetics, fenfluramine and its derivatives, propranolol and some other β-adrenoceptor blocking agents, morphine and levorphanol (see Allan & Michell, 1975).

Drugs which elicit these effects possess both an ionizable amine group and a hydrophobic region; the more massive the apolar substituent the more effective the drugs are in inhibiting phosphatidate phosphohydrolase in liver (Brindley & Bowley, 1975a) and in enhancing phosphatidylinositol synthesis in lymphocytes (Allan & Michell, 1975). Such drugs are known to interact with membranes and with isolated lipids. In particular, their positively charged amine groups interact with the anionic headgroups of acidic phospholipids such as phosphatidate, phosphatidylinositol and phosphatidylserine. This interaction would lead to the neutralization of charge on the phosphate group and the expulsion of the tightly bound divalent cations such as calcium (Dawson & Hauser, 1970); changes in the concentration and distribution of calcium ions within cells are known to have profound physiological consequences.

The hypothesis that the amphiphilic drugs inhibit phosphatidate phosphohydrolase through interactions with phosphatidate and other acidic phospholipids is supported by the observation that the concentration of chlorpromazine required to produce half maximum synthesis of phosphatidylinositol by lymphocytes is approximately sufficient to cause neutralization of half of the acidic phospholipids in the assay system used (Allan & Michell, 1975). The effect of this inhibition of phosphatidate phosphohydrolase is to increase the synthesis of acidic phospholipids, thus tending to make up the charge deficit produced by neutralization of existing acidic phospholipids. Although lipids such as phosphatidate and phosphatidylinositol are minor components of membranes, they may have considerable importance in relation to the receptor mechanisms whereby cells respond to extracellular stimuli (see Hawthorne, 1973; Michell, 1975).

Inhibition of phosphatidate phosphohydrolase seems likely to be only an example of a general situation in which a cationic drug interacts with an acidic phospholipid leading to modification of the activity of an enzyme or receptor. Such an enzyme or receptor may require the acidic phospholipid for its activity, although it need not itself be directly involved in lipid metabolism. A wide range of amphiphilic amines was shown to inhibit phosphatidate phosphohydrolase (Brindley & Bowley, 1975a; Allan & Michell, 1975) and the inhibition is therefore relatively non-specific. However, the possibility remains that in other interactions the structures of the hydrophobic regions of the drugs may confer greater selectivity in their effects. Additional selectivity *in vivo* might also arise from the different tissue distributions and metabolic modifications of the drugs.

One possible value of a drug which is a potential inhibitor of phosphatidate phosphohydrolase is to inhibit glyceride synthesis. Of the active amphiphilic amines tested, fenfluramine and its derivatives have this effect and in addition appear to be hypotriglyceridaemic agents (see Brindley & Bowley, 1975a for references). In the metabolic treatment of obesity it is not only necessary to inhibit triglyceride synthesis, but ideally a drug should also enhance lipid catabolism. Fenfluramine is known to increase the levels of free fatty acids and acyl-CoA esters in cells (Wilson & Galton, 1971) which could well lead to increased fatty acid oxidation. It is also interesting that chlorcyclizine diverts fatty acid metabolism away from triglyceride synthesis and increases β-oxidation in the livers of ethanol-treated rats (Wooles & Weymouth, 1968). This prevents the appearance of the ethanol-induced fatty liver. Chlorcyclizine is an antihistamine related in structure to mepyramine, and the latter compound was shown to inhibit phosphatidate phosphohydrolase (Brindley & Bowley, 1975a).

In this letter we present a number of seemingly related events observed with a variety of drugs which are in widespread clinical use and which can be classified as amphiphilic cations. The relevance of these observations to the known therapeutic effects, or side effects, of these drugs cannot yet be assessed. However, it seems reasonable to suggest that, at least at high therapeutic doses, their effects on lipid metabolism should be considered in future discussions of the mechanisms of action of amphiphilic cationic drugs. It has been known for some time that these drugs affect membrane phenomena such as movement, fusion, permeability, transport and receptor function; it is thought that these effects are partly due to interactions between the drugs and membrane lipids. It now seems likely that not only do they interact with lipids, particularly acidic phospholipids, but that they also have marked effects on lipid metabolism.

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