LETTER TO THE EDITOR

The Mechanism of Histamine Release from Mast Cells

SIR,—In the Symposium on Histamine[1] in honour of Sir Henry Dale (April, 1955), I drew attention (p. 401) to the importance of the lipid components of the mast cell, stressing in particular a similar behaviour of mast cells and the myelin of nerve sheaths, both of which undergo rapid hydration in the presence of alkali. Since then I have tested several lecithinases on mast cells—the α-toxin of *Cl. welchii*[2], purified bee venom and its heat-stable, non-enzymatic derivative, Melittin[3], and a powerful lecithinase which is present in the venom of the Indian cobra (*Naja naja*)[4]. All bring about disruption of mast cells parallel to their ability to attack lecithin prepared from nerves or egg yolk; Melittin appears to act as a histamine-liberator by virtue of its basicity and surface active properties. But this does not explain why a histamine-liberator, such as compound 48/80, specifically destroys the mast cells in the rat, leaving the myelin in the nerve sheaths intact; indeed, the few mast cells which are present in the nerve sheaths entirely escape the action of the liberator[5], which itself has no specific action on lecithin in model systems.

Professor Uvnäs[6] now offers a hypothesis which may resolve this difficulty. He suggests that there is normally an enzyme, a lecithinase, at the mast cell membrane and that this enzyme is prevented by an inhibitor from attacking the cell envelope. Compound 48/80, or other histamine-liberator specific for the species, removes the inhibitor and thus permits the lecithin of the membrane to be destroyed.

Perhaps we can theorise over this interesting new viewpoint a little further. Through the kindness of Dr. R. M. C. Dawson of Cambridge, chromatographic analysis for phospholipids has been carried out on a variety of tissues rich in mast cells, and upon mast cell granules isolated from the connective tissue of rats. In addition to lecithin, which is known to be present in some quantity in the mast granules of cattle[7], Dr. Dawson has found comparatively large amounts of phosphatidyl serine in all and, in the rat, some phosphatidyl ethanolamine also.

It will be recalled that in the original demonstration of the release of histamine from mast cells in the rat[8], the following morphological observations were made:

(i) The fluorescent diamidines can be seen to concentrate in the granules of the mast cells which then undergo a series of changes accompanying the release of histamine.

(ii) Pre-treatment with an antihistamine drug does not prevent the diamidine from reaching the granules but it does suppress the subsequent swelling, degranulation and disruption of the cell.

(iii) Histamine-release from the granules in the rat is accompanied by a curious vacuolation of the intergranular cytoplasm. Hill[9] has recently described this as a 'honeycomb' appearance.

The question thus arises, can the new hypothesis of Uvnäs help us to explain these morphological findings in terms of pharmacology?

If we modify the hypothesis slightly and postulate that the mast cell enzyme is a Type-C phospholipase, attacking the ester linkage between the phosphoric

acid and the alcohol (choline, serine, ethanolamine), then we may infer from known evidence

that the newly formed primary phosphatide (i.e., of the type R′—O—P(=O)—OH, with OH) will render the film leaky to histamine[10];

that an antihistamine drug will protect the newly formed film against the tendency of histamine to lower the surface tension[10];

and that if the substrate should include phosphatidyl ethanolamine, then the free ethanolamine so formed will give rise to an extraordinary cytoplasmic vacuolation. By a curious coincidence, this was also described as a 'honeycomb" appearance by its discoverers[11].

If a peritoneal 'window' of a freshly exteriorised portion of rat mesentery is coated with normal saline and a fragment of cover slip is applied, saline dilutions of the various split products of the phospholipids can be drawn across the microscope field and their effects on the mast cells observed. It is then evident that neither choline (10^{-3} as the chloride, prepared from lecithin) nor serine (10^{-3} from phosphatidyl serine) cause more than slight swelling of the mast cells, whereas ethanolamine, or the synthetic dimethylethanolamine (10^{-5} in saline), bring about a rapid swelling, vacuolation and even disruption of the cells. The microscopic appearances, in fact, closely resemble the 'explosions' which are seen when mast cells are treated with sodium hydroxide[12]. The point to be stressed, however, is that here the active base, ethanolamine, is formed intracellularly from a normal body constituent. Indeed, we are faced with the possibility that a single histamine-liberator applied extracellularly can produce *two* histamine-liberators intracellularly—a primary phosphatide (from lecithin, phosphatidyl serine or phosphatidyl ethanolamine) and free ethanolamine also when phosphatidyl ethanolamine is present.

Professor Uvnäs is to be congratulated on his ingenious explanation of the release of histamine from mast cells and more especially for giving us a hypothesis which can be tested in many fields of biological research.

My thanks are due to Dr. M. G. Macfarlane, London, for *Cl. welchii* α-toxin: to Dr. E. Habermann, Würtzburg, for purified bee venom and Melittin, and to Dr. R. Hirt, Bern, for samples of synthetic primary and secondary phosphatides.

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REFERENCES

1. Riley, *Histamine*, Ciba Symp., London, 1956, 401.
2. Macfarlane, *Biochem. J.*, 1948, **42**, 590.
3. Habermann, *Arch. Path. Pharm.*, 1955, **225**, 158.
4. Turner, *J. exp. Med.*, 1956, **104**, 517.
5. Riley and West, *J. Path. Bact.*, 1955, **69**, 269.
6. Uvnäs, *J. Pharm. Pharmacol.*, 1958, **10**, 1.
7. Hedbom and Snellman, *Exper. Cell Res.*, 1955, **9**, 148.
8. Riley, *J. Path. Bact.*, 1953, **65**, 471.
9. Hill, *Experientia*, 1957, **13**, 395.
10. Hirt and Berchtold, *Arzneimit.-Forsch.*, 1952, **2**, 453.
11. Brues and Jackson, *Cancer Res.*, 1941, **1**, 557.
12. Riley, *Pharmacol. Revs.*, 1955, **7**, 267.