

was devoid of any similar cell and displayed only slight metachromasia.

The appearance of mast cells in both of the cellular layers of the cyst of *H. taeniaeformis* indicates strongly that the entire cyst wall is of host origin. Since the mast cell is a connective tissue cell probably derived from undifferentiated mesenchymal cells, it is less likely that the cyst wall is elaborated from modified host-liver cells, although the latter may be involved in this reaction. Bogitsh(5), on the basis of histochemical tests, found a similar host-mesenchymal origin for the cyst wall of *Neoeccchinorhynchus cylindricus* (a nematode parasite of fish) in liver.

The present demonstration of mast cells in the cyst wall contributes to the further identification of its cellular components. Moreover, it may provide an additional method of determining whether the cyst of a particular larval helminth is of dual host-parasite origin, or entirely produced by the host (Types I and II respectively, as described by Hunter and Dalton(6)).

Therefore, the function of the mast cell would appear, not unexpectedly, to include participation in some aspects of the host reaction to parasitic invasion.

Summary. Mast cells have been found in the cyst wall of a larval cestode parasite (*Hydatigena taeniaeformis*). The mast cells were present in the outer 2 layers of the cyst wall. This indicates that the entire cyst wall is of host-inflammatory origin and not derived from parasite tissue nor solely from modified host-liver cells. It is suggested that detection of mast cells as host-mesenchymal cellular units may aid in determining origins of cysts produced by other parasites.

The authors are grateful to Dr. L. A. Stauber, Rutgers, the State Univ. of New Jersey, for identification of the parasite; to Dr. W. Montagna, Brown University, for critical reading; and to Mr. J. Sanders for photomicrographs.

1. Kelsall, M. A., Crabb, E. D., *Lymphocytes and Mast Cells*, Williams & Wilkins, Baltimore, 1959.
2. Montagna, W., Chase, H. B., Melaragno, H. P., *J. Nat. Cancer Inst.*, 1951, v12, 591.
3. McManus, J. F. A., *Stain Technol.*, 1948, v23, 99.
4. Ahlqvist, J., *Acta, Pathol. et Microbiol. Scand.*, 1960, v50, Suppl. 142.
5. Bogitsh, B. J., *Proc. Helm. Soc. Wash.*, 1961, v28, 75.
6. Hunter, G. W., III, Dalton, H. C., *ibid.*, 1939, v6, 73.

Received September 12, 1962. P.S.E.B.M., 1963, v112.

Quantitative Relation Between Certain Fibrinolysis Factors and Inflammation. (28068)

E. ASCHHEIM,* V. TSULUCA AND A. L. COPLEY

Department of Pathology, New York University School of Medicine and Hemorheology Laboratory,
Department of Physiology and Pharmacology, New York Medical College

This communication deals with the quantitation of the antiinflammatory activity of bovine and human fibrinolysin (plasmin) preparations. This effect described by Martin(1) and Copley and Tsuluca(2-7) represents a physiologically significant instance of the antiphlogistic action of proteolytic enzymes, since the activation of the profibrino-

lysin (plasminogen)-fibrinolysin system accompanies tissue injury(8).

Materials and methods. Male albino rats were anesthetized with sodium pentobarbital (35 mg/kg body weight), and their clipped abdominal skin injected at various sites with either the phlogogen alone or a mixture of phlogogen and fibrinolysin. The substances were injected in a volume of 0.02 ml, injection sites were rotated and no animal received twice the same dose of the antiinflammatory agent. Histamine dihydrochloride in

* Present address: Depts. of Dermatology and Physiology, Stanford Univ. Med. Center, Palo Alto, Calif.

a dose of 2 μ g, or 0.2 mg of a purified preparation of profibrinolysin (Merck, Sharp and Dohme Research Laboratories, West Point, Pa.) were employed as inflammatory agents. The profibrinolysin preparation was found previously(2,3,5) to increase locally the vascular permeability to proteins in a manner analogous to the action of histamine, serotonin or bradykinin. As an antiinflammatory agent, human fibrinolysin (Ortho Research Foundation, Raritan, N. J.) in doses of 100, 200, 400 and 800 units was used.

The inflammatory reaction was quantitated by measuring the amount of extravasated radioiodinated human serum albumin (RISA, Abbott Laboratories, Chicago, Ill.). The method was described previously(9,10,11). Briefly, the anesthetized animals were injected intravenously with approximately 2 microcuries of radioiodinated human serum albumin and a 1% solution of Evans blue in a total volume of 1 ml. The dye served better to delineate the inflamed areas. Ten to 15 minutes later 6 abdominal sites were injected intracutaneously with the testing materials. Exactly 10 minutes following the intracutaneous challenge, the animals were killed, the injected skin sites excised and a blood sample obtained. Radioactivity of the skin is given in counts per minute per one gram of dry skin (drying overnight at 100°C) and related to the blood radioactivity. The value thus obtained expresses the amount of blood in milliliters cleared completely of radioactive albumin by one gram of dry skin during the experimental period, and is given by:

$$C = \frac{\text{counts/min/g dry skin}}{\text{counts/min/ml blood}}$$

This value is taken as a measure of the intensity of the inflammatory reaction.

Results. Fig. 1 shows the effect of injecting the fibrinolysin preparation together with histamine. Each point represents the average of 5 determinations and its standard error. The inflammatory reaction due to histamine was significantly reduced whenever this agent was injected together with the fibrinolysin preparation.

Fig. 2 presents similar data with regard to

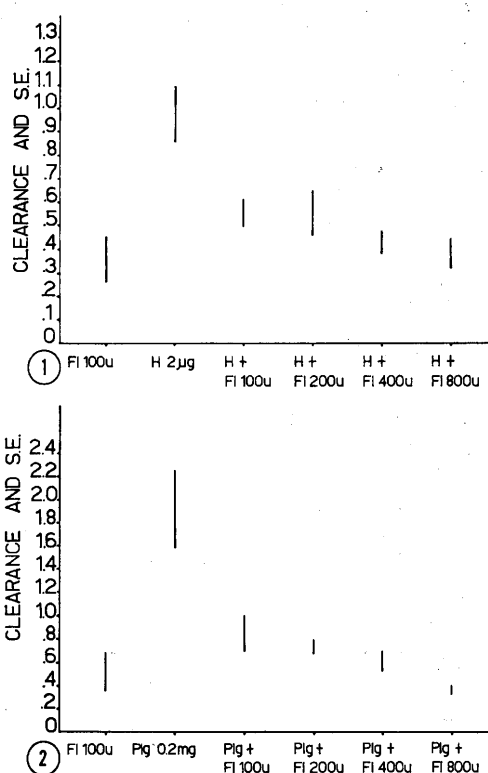


FIG. 1. Histamine induced capillary permeability inhibited by fibrinolysin.

FIG. 2. Plasminogen induced capillary permeability inhibited by fibrinolysin.

the inhibition of the inflammatory action of the profibrinolysin product by the fibrinolysin preparation. Here also the inclusion of the fibrinolysin preparation in the injected material decreased significantly the amount of extravasated protein. In both cases, an amount of the preparation containing 400 units of fibrinolysin brings the inflammatory reaction down to control levels.

Discussion. The antiinflammatory activity of proteolytic enzymes appears to be established(1-7,12-15,18,19), although the nature of this effect is not understood. Proteolysis may diminish the extent of intra- and extravascular clotting which is responsible for the fixation of substances at inflamed sites (16,17). Lysis of fibrin clots may also result in greater circulatory flushing of the inflamed site and thus promote the dilution of the irritant with a consequent diminution of its activity. On the other hand, the beneficial antiphlogistic action of systemically ad-

ministered proteolytic enzymes is observed in the absence of any detectable increase in proteolytic activity of the blood(18). This finding suggests an indirect influence of proteolysis on the inflammatory process, although it does not exclude a possible concentration of the enzyme at the inflamed site with a consequent rise in local activity as compared to low systemic levels. The possibility is not excluded, however, that systemically administered proteolytic enzymes act indirectly by activating other systems which in turn counteract inflammation. The problem appears further complicated by the observation that both trypsin and soya bean trypsin inhibitor are effective in suppressing the inflammatory reaction(19).

The nature of the phlogogenic action of the profibrinolysin preparation is obscure. In addition to profibrinolysin the product contains also gamma globulin(20) which may be responsible for this effect. It is likely that the activity of the permeability increasing factor of the preparation is not destroyed by fibrinolysin, since the latter affects in a like manner the reactions induced by histamine. The properties of this factor must await further characterization.

Summary. Application of radioisotope technics to the study of the inflammatory reaction confirmed the antiinflammatory properties of fibrinolysin (plasmin) preparations. The effect was studied in both histamine and profibrinolysin (plasminogen) induced inflammation. In both cases 400 units of fibrinolysin neutralized the response bringing it down to control levels.

1. Martin, G. J., *Ann. N. Y. Acad. Sci.*, 1957, v68, 70.
2. Copley, A. L., *Proc. 8. Congr. Europ. Soc. Haemat.*, Vienna, 1961, Basel-New York, S. Karger, 1961, 357; *Hémostase* (Paris), 1963, in press.
3. Copley, A. L., Tsuluca, V., *Fed. Proc.*, 1962, v21, 64.
4. ———, *Life Sci.*, 1962, v6, 243.
5. ———, *Proc. 1. Internat. Pharmacol. Meeting*, Stockholm, 1961, Vol. 9, M. Rocha e Silva and P. Lindgren, Ed., Oxford, Pergamon Press, in press; *Biochem. Pharmacol.*, 1962, v10, 67.
6. ———, *Handbook*, 22. *Internat. Congr. Physiol. Sci.*, Leiden, 1962, v2, 132.
7. Copley, A. L., *Handbook*, 2. *Europ. Conf. on Microcirculation*, Pavia, 1962, p7; *Proc.*, Basel-New York, S. Karger, in press.
8. Ungar, G., Hayashi, H., *Ann. Allergy*, 1958, v16, 542.
9. Aschheim, E., Zweifach, B. W., *Circ. Res.*, 1961, v9, 349.
10. ———, *Fed. Proc.*, 1961, v20, 100.
11. ———, *Am. J. Physiol.*, 1962, v202, 554.
12. Seligman, B., *Angiology*, 1955, v6, 208.
13. Innerfield, I., *Surgery*, 1956, v39, 426.
14. Moser, K. M., *New Eng. J. Med.*, 1957, v256, 258.
15. ———, *ibid.*, 1957, v256, 303.
16. Menkin, V., *PROC. SOC. EXP. BIOL. AND MED.*, 1933, v30, 1069.
17. ———, *Dynamics of inflammation; an inquiry into the mechanism of infectious processes*, New York, Macmillan Co., 1940.
18. Miller, J. M., *Ann. N. Y. Acad. Sci.*, 1957, v68, 185.
19. Hladovec, J., *Experimentia*, 1958, v14, 146.
20. Rife, U., Milgrom, F., Shulman, S., *Fed. Proc.*, 1962, v21, 65.

Received October 18, 1962. P.S.E.B.M., 1963, v112.

Studies on the Metabolism of Meprobamate. (28069)

J. F. DOUGLAS, B. J. LUDWIG AND NANCY SMITH (Introduced by F. M. Berger)

Wallace Laboratories, Division of Carter Products, Inc., Cranbury, New Jersey

The urinary excretion products obtained after administration of meprobamate (2-methyl-2-propyl-1,3-propanediol dicarbamate) to warm-blooded animals include unchanged drug(1), an oxidized derivative, recently identified as 2-methyl-2-(β -hydroxypropyl)-

1,3-propanediol dicarbamate(2), and a glucosyluronide conjugate(1-5). Although the first 2 substances have been well characterized, the literature reveals only 2 references to their quantitative relationship(5,6). We have investigated the excretion pattern of