increased activity in glycolytic pattern after 20 s ischaemia, as was observed in normal hearts.

Relative to earlier findings in fluoracetate treated hypothyroid rats (Paterson, 1971) these results indicate that phosphofructokinase responds to growth hormone rather than to thyroid hormones with the treatments used, but hexokinase and pyruvate kinase respond to either. It may therefore be concluded that both these hormones are necessary for maintaining a normal cardiac response to ischaemia of short duration as well as to long-term hypertrophy induced in the rat heart (Beznak, 1967).

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Mitogenic and inflammatory activities produced by antigen activation of guinea-pig lymphocytes

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Lymphocytes can be stimulated *in vitro* by several agents to release materials into the culture medium with a wide variety of biological actions (Bloom & Glade, 1971). After specific antigen activation of cells from animals (or man) with delayed hypersensitivity, a class of materials is generated which has been proposed to mediate cellular immunity and termed lymphokines (Dumonde, Wolstencroft, Panayi, Matthews, Morley & Howson, 1969). Two of the biological activities observed in these preparations are (a) the appearance of inflammation after intradermal injection into normal animals and (b) the ability to increase DNA synthesis of cultured lymphocytes. In the present experiments, an attempt has been made to correlate the inflammatory activity of these preparations with their mitogenic activity. Inflammatory activity was assessed by the measurement of accumulation of ^{125}I-labelled serum albumin after injection into guinea-pig skin over a period of 4 hours. Mitogenic activity was assessed by measurement of the increased incorporation of tritiated thymidine, induced in cultured guinea-pig lymph node lymphocytes.

Guinea-pigs were sensitized with antigen (100 μg of bovine gamma-globulin in Freund's complete adjuvant). Lymphocytes collected 2 weeks after sensitization were cultured in serum-free Eagle's medium for 18–24 h in the presence of antigen (1 mg/ml). Cells were separated by centrifugation and the culture fluid stored at −20° C.

In such preparations, biological activities were greatest in preparations obtained by stimulating cells with specific antigen when compared with preparations obtained from cells cultured either in the absence of antigen or with a non-cross reacting antigen (for example ovalbumin). Both inflammatory and mitogenic activities of such preparations were greater than those due to extracts from equivalent numbers of cells

(extracted by sonication or freeze thawing) (Fig. 1). Inflammatory activity of lymphokine preparations usually showed only slight differences from control preparations, whereas the mitogenic activity was clearly demonstrable. However, inflammatory activity is regularly observed when these preparations are concentrated by dialysis and freeze drying and tested at higher concentrations.

Removal of the antigen and any immunoglobulin by sequential ammonium sulphate precipitation leaves the bulk of the mitogenic and inflammatory activity in the fraction remaining in solution at 50% ammonium sulphate saturation, with increased specific activity of at least ten for both activities.

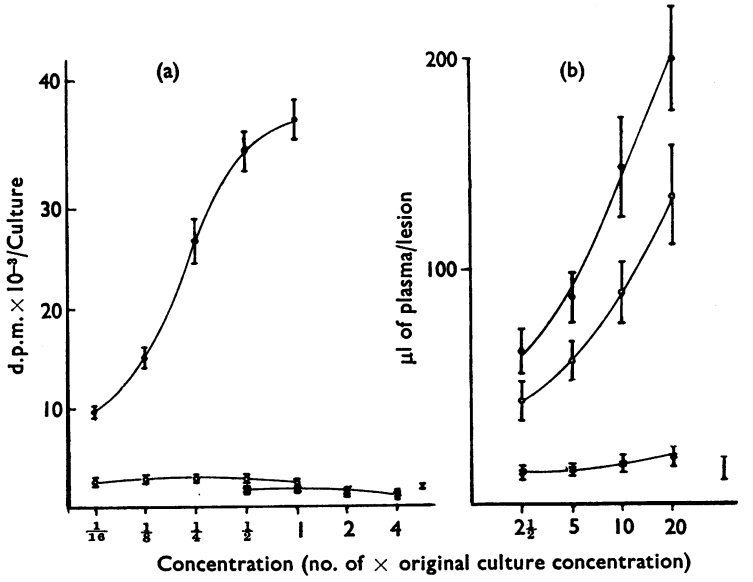

FIG 1. Increased DNA synthesis (a) and increased vascular permeability (b) produced by increasing concentrations of materials derived from sensitized lymphoid cells; (●), by culture with antigen; (○), by culture without antigen, the antigen being added only at the termination of the culture; (×) by extraction of equivalent numbers of lymphoid cells. Figures along the abscissa refer to the degree of concentration of the test material; (I) indicates the response range to diluent alone.

Lymphocyte activation results in the release of material that will reproduce certain features of responses of delayed hypersensitivity, and the inflammatory and mitogenic activities cannot be attributed to release of performed material from lymphoid cells. Partial purification of these preparations has not yet resulted in clear separation of inflammatory and mitogenic activities. However, the partial suppression of the inflammatory response by antihistamine and the considerable inflammatory activity of control preparations in which there is little or no demonstrable mitogenic activity, suggests that a substantial proportion of the inflammatory activity of these preparations is not related to their mitogenic activity.

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