

Pinnal Anaphylaxis: an Additional Anaphylactic Site

In some respects the mouse is a desirable experimental animal for immunological studies. Because of its small size and high reproductivity it can be used in statistically meaningful numbers. Availability of highly inbred strains minimizes variation in individual response and makes it possible to select strains of maximal sensitivity¹.

In other respects the mouse has certain drawbacks. One is its relative refractoriness to anaphylactic challenge. Another is the limited body-area available for local anaphylaxis.

Ovary² demonstrated the possibility of producing passive cutaneous anaphylaxis in the skin of the back of the mouse. He found that about 100 times as much rabbit antibody is necessary to induce local sensitization of the mouse skin as compared with the guinea pig. With high concentrations of reagents he was effectively limited to two injection sites, one on each side. His intravenous challenging dose of antigen was 1.2-mgm. hen ovalbumin.

The present investigation was undertaken with the objective of increasing the number of available sites for local anaphylactic challenge in the mouse and, perhaps, introducing more sensitive sites than the skin of the back. The ear was selected for several reasons. It is a translucent structure, in which collection of dye released by local anaphylactic reaction would be readily apparent. Though preferential inducement of capillary permeability in the ear, as compared with the skin of the back, is less marked in the mouse than in the rat and guinea pig, Feldberg and Miles³ did find a distinctly greater response to 48/80 over the head, shoulders and limbs than elsewhere on the mouse skin. Finally, as a paired structure, one ear can, if desired, serve as a control to the reaction in the other. This initial communication reports primarily on the production of passive anaphylaxis in the ear of the mouse, using a partially purified duck ovalbumin and the rabbit antiserum to it.

The skin over the ear is very fine, and it is impracticable to carry out intradermal injection. Therefore, it was decided to instil the sensitizing serum subcutaneously. The mouse was lightly anaesthetized with ether. Using 26 gauge $\times \frac{1}{2}$ -in. needles, a tenfold dilution of the antiserum was injected between the skin and cartilage on the outer aspect of one ear, while saline was injected similarly in the other. Volume was controlled by sight, a small bleb being raised. This approximates to 0.025 ml. of fluid per site.

At intervals varying from 30 min. to 24 hr. after sensitization the mice were injected intravenously in a tail vein with 0.25 ml. of a solution containing 1.25 mgm. of 'Pontamine' sky blue and 0.1 mgm. of the duck ovalbumin. Control mice were challenged with dye alone. A positive reaction, as exhibited by the local extravasation of blue dye in the ear, generally appeared within 10 min. and was complete after 30 min. The saline-injected control ears in antigen-challenged mice were uncoloured (apart from an occasional pin-prick of blue at the point of passage of the needle through the skin), as were both ears of the mice challenged with dye alone. Fig. 1 shows the complete results of a single experiment: no selection or elimination has been made. Positive reactions have been obtained when antigen challenge has been carried out as early as 30 min. and as long

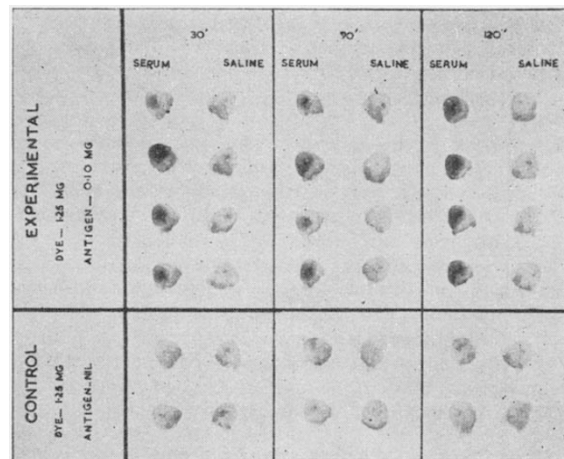


Fig. 1. Passive pinnal anaphylaxis in the mouse. Ears seen from inner aspect

as 24 hr. after passive sensitization of the ear, 2-3 hr. being the optimal time for production of uniformly strong reactions.

The term pinnal anaphylaxis seems most appropriate to the phenomenon as the entire thickness of the pinna appears to be permeated with the dye: the skin on both the inner and outer aspects of the ear, the subcutaneous spaces and the cartilage.

Passive pinnal anaphylaxis has also been produced in intravenously blued mice on subcutaneous challenge with as little as 0.5 mgm. of duck ovalbumin. It has also been demonstrated in the guinea pig both on intravenous and subcutaneous challenge with duck ovalbumin and grass pollen extracts, using appropriate antisera. Quantitative studies, active pinnal anaphylaxis and other extensions and applications of the phenomenon are under investigation.

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¹ Rothberg, R. M., and Talmage, D. W., *Fed. Proc.*, **19**, 215 (1960).

² Ovary, Z., *J. Immunol.*, **81**, 355 (1958).

³ Feldberg, W., and Miles, A. A., *J. Physiol.*, **120**, 205 (1953).

Cytochrome c in the Heart of the Turtle

In a thesis on the anaerobic metabolism and mechanical efficiency of the heart of the turtle, by R. Blake Reeves¹, there is evidence that this heart is able to work efficiently for any length of time without access to oxygen (nitrogen anoxia). In view of this remarkable quality we found it interesting to investigate the content of cytochrome c in the heart of the turtle (*Testudo graeca*). To our knowledge, no such data are available in the literature²⁻⁴, although reports on the cytochrome c content of the heart of the frog have been published by Keilin⁵.

Six turtles were killed by ether and the hearts were weighed (3.5 gm.). The cytochrome c was extracted according to a method described by one of us (G. B.⁴); instead of 'Amberlite XE-64', however, 'Duolite CS-101' was used in the present investigation.