

BRIEF NOTE

ANTIGEN OF RETEST REACTION IN CONTACT
SENSITIVITY TO DNCB

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So-called retest reaction in tuberculin skin test was already reported in early years of this century, and the observation has been clinically and experimentally confirmed several times^{1,2,3}). However, few study of the reaction in contact sensitivity has been reported. The term retest reaction is used here to describe accelerated reaction which appears at previous positive skin test site by renewed local administration of antigen. An attempt was made to induce retest reaction in contact sensitivity and determine its antigen in this study.

Male Hartley strain guinea pigs were sensitized by an application of 0.1 ml of 5 per cent solution of 2,4-dinitrochlorobenzene (DNCB) in ethanol to both sides of inguinal skin surface, and tested with 0.01 ml of 0.2 per cent DNCB in ethanol 7 days later to result in positive patch test. Fourteen days after sensitization, applications of 0.01 ml of 0.2, 0.09, 0.05 or 0.01 per cent solution of DNCB in ethanol were given to previously tested sites. The results are illustrated in Figure 1. The intensity increased rapidly to a maximum 9 hours after patch testing at retest site and declined sharply after it. This time course is similar to those of retest reactions in delayed sensitivity to a variety of antigens shown by Arnason and Waksman³). At new sites (not previously tested) in the same animals, contact reaction increased slowly to reach maximal intensity at 9 hours and kept same degree of intensity for the next 15 hours. Until 24 hours, the reactions were significantly more intense at retest sites than at new sites. This experiment shows that retest reaction can be elicited in contact sensitivity.

The so-called flare-up reaction due to renewed systemic administration of antigen at the previous site of a reaction has been considered to be induced in the similar pathogenic mechanism⁴). In the previous study

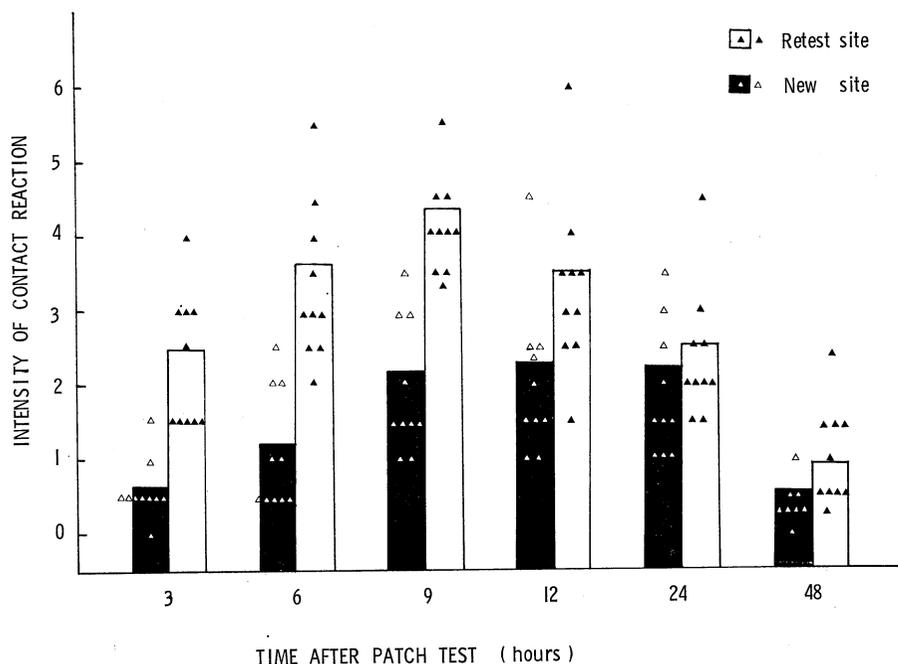


Fig. 1. Retest reaction at the site of contact reaction to DNCB. The reactions are assessed according to an arbitrary scale from 0 to 3, as described in the previous paper⁽⁷⁾. Intensities of the reactions and their means at each site are shown as the sum of the numerical readings obtained with four different test concentrations (▲, retest site, △, new site).

on the flare-up reaction, we have demonstrated evidence which shows that sensitized cells still remaining at the old test site react with the conjugate of hapten with epidermal proteins and provoke an antigen-antibody reaction which results in the flare-up.⁵⁾ Attempts were made to extend this concept to retest reaction.

Animals were initially applied epicutaneously 0.1 ml of 50 per cent solution of DNCB in acetone to the nape skin and given intradermal injection of 0.1 ml of Freund's complete adjuvant (FCA) to the previously painted area. The animals were tested intradermally with 50 μ g of 2, 4-dinitrophenyl (DNP) conjugates prepared *in vitro* from the extract of guinea-pig epidermis (GPE) and 0.1 μ g of purified protein derivatives of tubercle bacillus (PPD), and tested epicutaneously with 0.2% DNCB. They were retested epicutaneously with DNCB in four different concentrations and intradermally with DNP-GPE, DNP-guinea pig serum and PPD at the previous test sites 14 days after sensitization. Skin testing with DNP-

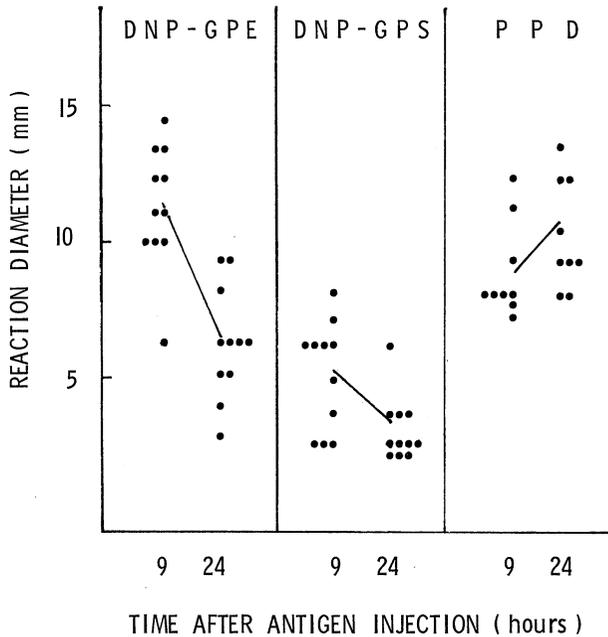


Fig. 3. Reactions on retest with DNP-GPE, DNP-GPS and PPD at sites of prior contact reactions to DNCB. Diameters of erythematous indurations in millimeters are shown.

TABLE

Patch test with DNCB at site of delayed reaction to DNP-GPE in animals sensitized with DNP-GPE

Sensitized with	Positive patch test with DNCB at	
	9 hours	24 hours
DNP-GPE plus FCA	(8 animals)	
Retest site		
DNP-GPE	7	7
PPD	2	4
New site	4	7
DNP-GPS plus FCA	(5 animals)	
Retest site		
DNP-GPS	0	0
PPD	0	0
New site	0	0

24 hours after testing. DNP-GPS was not effective to induce contact sensitivity as described previously⁷⁾. Consequently, retest reaction, maximal at 9 hours and waning some extent at 24 hours, was elicited only at the site of reaction to DNP-GPE.

We do not at present know the mechanism by which the retest reaction could be produced. Arnason and Waksman have indicated that, despite its unique gross and histological features, it is closely allied to delayed sensitivity⁸⁾. It has been shown by Polak *et al.* that antigen-sensitive cells previously involved in the test reaction could still remain in the cell infiltrate at the site of the old contact reaction⁴⁾. It is plausible to explain as follows: Epicutaneously applied hapten forms a complete antigen by conjugation with epidermal proteins as shown elsewhere⁹⁾. Thereafter the antigen-sensitive cells remaining at the skin test site following the previous contact reaction react with its complete antigen to produce contact reaction.

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