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REACTION OF HOST TO THE TICK-BITE I

BY

J. NOSEK, J. RAJČÁNI and O. KOŽUCH

INTRODUCTION

Man and animals in which he has a vital nutritional or economic interest represent to the tick a source of animal protein. During the tick-feeding an enormous variety of pathoergonts may be transmitted to the host by ixodid saliva. Ixodid saliva has a tripple role: A cement is secreted by granule secreting alveoli of II and III type to provide firm attachment for the feeding tick on the host, pharmacologically active substances did in feeding, and excess dietary water and electrolytes are eliminated by secretions into the host.

Tick may act as vectors of protozoan, rickettsial and viral diseases of man and animals (Horsfall et al. 1965, Philip 1967, Hoogstraal 1966, 1967, 1973, Balashov and Daiter 1973). Heavy infestation with certain species of ixodid ticks may result in severe disorders of haematopoetic system and sometimes in fatal anaemia (Jellison and Kohls 1938, Riek, 1956). Tick paralysis of the ascending type is the most enigmatic of the many diseases caused by ticks (Gregson, 1973).

Interaction of saliva from subsequent series of parasites proceeded on the principle antigen-antibody and may result in the skin immunity. After the primary exposure to the parasite acquired resistance mechanisms of the host become important. In the majority of cases acquired resistance is immunological in whole or at least in part (Tatchell, 1969, Musatov, 1970).

Reports about acquired immunity of hosts to tick bites are contradictory. Trager (1939) and Feldman-Muhsam (1964) observed that the first feeding may sensitize the host, i.e. that a higher percentage of ticks feed on the second feeding than on the first. Only the second and subsequent feedings immunize the host progressively, i.e. that the proportion of feeding ticks decreases from feed to feed. Enigk and Grittner (1953) claim that one feeding of larvae on rabbit's ear suffices to immunize the rabbit at least partly, because on the second feeding a smaller proportion of larvae attaches, and those which attach feed more slowly. In our feeding experiments we have observed that the first feeding of *Ixodes ricinus* larvae can immunize the white mouse at least partly but only in the case if the first dosis of larvae is very high (more

than 100 larvae per one individual). This finding is in agreement with very frequent observation in nature where in areas with high occurrence of larvae and nymphs of *I. ricinus* tick, some adult individuals of free living rodents, e.g., *Apodemus flavicollis* were non or very low infested but the marks after tick feeding were still visible.

The nature of tick saliva and salivary gland function have been studied by Gregson (1960, 1967, 1969) and Tachell (1967, 1969); Tachell et al. (1972), Moorhouse (1969). The histology of the salivary gland was described in detail by Chinery (1965) in *Haemaphysalis spinigera* ticks and by Balashov (1967) in ticks of the subfamily Ixodinae and Amblyommatinae. Recently, the fine structure of the salivary glands of unfed male *Dermacentor variabilis* was excellently demonstrated by Coons and Rosdy (1973). Microscopic and submicroscopic studies of the various salivary alveoli are essential to reveal the nature and composition of the cell components as a basis for studying relationship pathoergont-tick-host. More recent studies seem to indicate that the cement substance is wholly of salivary origin (Gregson 1960, Chinery 1973) as was also established in trombiculid mites (Voigt 1968). The aim of present study was established the local reaction of viraemic host during the feeding of noninfectious tick.

MATERIAL AND METHODS

Ixodid ticks *Dermacentor reticulatus* and *Haemaphysalis concinna* originated from laboratory bred were used in feeding experiments. The larvae were kept at temperature 22°C and r.h. 95%.

To induce viraemia white rats weighing 90-100 g were infected subcutaneously with 0.2 ml of a 10% brain suspensions containing virus in titre $10^{7.5}$ LD₅₀ per 0.03 ml. TBE virus hedgehog strain J 13, 9th mouse passage was used for infection of rats. The skin biopsies with attached mouthparts were carried out after 15, 39, 72, hours and fixed in Bouin's fixative. Semi-serial sections were cut at 100 µm and for general histological features stained in Azan or heamatoxylin erythrosine, respectively. Paralelly the larvae fed on viraemic and non-viraemic mice.

RESULTS

Virus neutralizing antibodies in white rats were found 6 weeks after infection in titre 1:16-1:64. Attachment of species examined is accompanied by the secretion of cement, and leads to a marked hypertrophy of epidermis (Malpighian layer) beneath and adjacent to the cement. The details showed the penetration through epidermis to the superficial layers of corium (Figs 1 a,b). Deeply in dermis a slight infiltration from mononuclear cells was observed. The venulae and lymphatic vessels were dilated (Fig. 1b). The superficial cement cone completely invested the mouth-

parts, hypostome and chelicerae (Fig. 1b). Conversely, skin biopsies taken 3 days after attachment on the back of a rat showed the penetration through the stratum corneum and preservation of stratum germinativum (Malpighian layer) and mononuclear infiltration in corium (Figs. 2,3). The small lesions in Malpighian layer were observed (Fig. 2). These lesions are present also in the early feeding, e.g., 5 hours after attachment.

The reaction of viraemic host to the tick-bite observed in light microscope seems to be identical with that of non-viraemic host. Sections through the salivary glands of the cement-secreting ticks, contained cells with granular inclusions.

Species *Haemaphysalis*, *Dermacentor*, have cement which is mainly superficial and the mouthparts do not penetrate into the dermis. The infection rate of nymphs amounted 60 per cent.

DISCUSSION

Meredith and Kaufmann (1973) suggest that the group III acinus contributes most of the fluid portion of the saliva and that the main cell type involved is so called water cell. The granule-cells possibly secrete the cement by which the tick secures its mouthparts to the host, and the vacuolar cells possibly produces a protein rich secretion. In vacuolar cells also the multiplication of TBE virus was observed (Nosek et al., 1972).

It seems that the cement substance is characteristic of most genera of slow-feeding ticks. With reference to the results obtained in the study of trombiculid mites (Schumacher and Hoeppli 1963) it is concluded that the cement of tick similarly as stylostome of trombiculid mites are essentially a product of the parasite which serves rather a mechanical than a chemical function in the feeding process. Gregson (1960) and Chinery (1965) were of opinion that cement is wholly of salivary origin. It is the function of this rapidly hardening secretion to secure the attachment of the mite to the host. The skin reaction are probably caused by another secretion which remains fluid and spreads rapidly into the dermis, leaving the epidermis unaffected. The differences in the methods of attachment within the family Ixodidae suggested that a comparative study of this mechanism in this family would be of interest, especially in structurally very primitive species *Haemaphysalis inermis* which belongs to the short-feeding ticks.

SUMMARY

In the insertion-places of tick mouthparts reactive changes of dermis were observed characterized by slight mononuclear infiltration. Hypostome and chelicerae penetrated across epidermis and were surrounded with cement. No differences between the feeding pattern of ticks on the viraemic and non-viraemic host were seen.

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PLANCHE I

FIG. 1a. — Section through the rat's ear showing the pattern of attachment of *Dermacentor reticulatus* larvae during the final stages of feeding (39 hours after attachment) arrow = Infiltration (Magnification $\times 160$).

FIG. 1b. — The same section at the magnification $\times 400$. Penetration of mouthparts to the superficial layer of corium. The dermis is characterized with slight infiltration of mononuclear and polynuclear cells, and dilatation of venulae and lymphatic vessels ($\times 400$) C = cement cone, E = epidermis, D = dermis, sc = stratum corneum, sg = stratum germinativum (= Malpighian layer), h = hypostome and chelicerae.

PLANCHE II

FIG. 2a. — Section from the rat's skin on the back on the 3rd day after attachment. Mouthpart-penetration of *Dermacentor reticulatus* larva beneath stratum corneum; epidermis enough preserved. Mononuclear infiltration in corium. fl = feeding lesion ($\times 220$).

FIG. 2b. — The same section at the magnification $\times 880$. Detail of mononuclear infiltration.



