From the Department of Zoology, Sri Venkateswara University, Tirupati; A. P., India

A COMPARATIVE STUDY OF THE ARACHNID CUTICLE
II. CHEMICAL NATURE*

By
A. KRISHNAKUMARAN

(Received February 19, 1961)

Introduction

The demonstration of the role of sulphur in the hardening of the cuticle in a scorpion, *Palamnaeus* (*Heterometrus*) *swammerdami* (KRISHNAN 1953) has indicated a diversity in the mode of hardening of the arthropod cuticles. Earlier LAFON (1943) reported the occurrence of sulphur in the cuticles of *Xiphosura*. Based on the sulphur content he suggested that the cuticular proteins in this form resemble the vertebrate keratins. RICHARDS (1956) confirmed the presence of sulphur in this form but concluded that most of the sulphur present is in the form of inorganic salts. TIEGS and MANTON (1958) considered that the occurrence of sulphur in the cuticles may be characteristic of the chelicerate subphylum. HUGHES (1959) reported the occurrence of sulphur in the cuticle of the tick, *Acarus* (*Tyroglyphus*) *sirô* and considers that this sulphur might play a role in the binding of chitin and protein fibres. Further, the presence of sulphites inhibits the autoxidation of hydroquinone (JAMES and WEISSBERGER 1939). Agreeing with RICHARDS (1956) that some of the sulphur occurs as sulphites what effect it has on phenolic tanning in such cuticles is little known. With a view to elucidate a few of these features the cuticles of five different arachnids have been chosen for the present study. The results pertaining to structure and staining properties have been presented earlier (KRISHNAKUMARAN 1961a).

Materials and methods

*Thelyphonus* (Pedipalpi), *Rhipicephalus* (Acarina), *Argiope* (Araneae), *Galeodes* (Solifugae) and *Carcinoscorpius* (Merostomata) have been used in the present investigation.

For the histochemical investigations the routine tests have been used. (For procedures see LISON (1953) and PEARSE (1954)). Xanthoproteic, Millon’s and Biuret tests were employed for the identification of proteins. Chitosan reaction for chitin; ninhydrin for amino acids; Sudan dyes for lipids; Azide-iodine reaction

* Forms part of a thesis submitted to the University of Madras for the award of the degree of doctor of philosophy.
(Feigl 1954) for sulphur; Baker's test, Ferric chloride reaction and also argentaffin-reaction for aromatic substances particularly the phenolic substances; were amongst the few tests employed during the present study. In addition diazo-benzene-sulphonic acid and phosphomolybdic acid reactions have been applied for the histochemical studies.

Acid hydrolysis was done by heating at 110°C with 6N hydrochloric acid in a sealed tube. (However such a treatment does not digest the cuticle in Carcinoscorpius completely.) The cuticle has been cleaned of all the attached tissues before hydrolysis.

The histochemical reactions were performed on fresh or gelatin embedded frozen sections. For the latter the cuticle has been fixed in 5% formaldehyde and was impregnated and embedded in gelatin. The gelatin blocks were hardened in formalin.

Observations

a) Thelyphonus. The cuticle as described elsewhere (1961a) is constituted of an epicuticle, coloured exocuticle, a mesocuticle and an endocuticle the last of which is further subdivided into two regions. The histochemical reactions indicate that epicuticle is a nonchitinous, lipoprotein layer while the exocuticle, mesocuticle and endocuticle are made up of chitin-protein complex. The pigmentation of the cuticle is partly due to the occurrence of melanins as is evident from the partial bleaching obtained by treatment with ethylene chlorhydrin as well as hydrogen peroxide and acidified potassium permanganate solution. Phenolic tanning appears to be partly responsible for the pigmentation as indicated by the chromophobic nature, the effect of detanning agents such as alkaline stannite as well as the amber colouration. The mesocuticle which is acidophilic is rich in proteins containing tyrosine (strong positive xanthoproteic and Millon's reactions) and phenolic and lipidal substances (positive argentaffin, Baker's and Sudan reactions) thereby resembling a transitory stage described in Schistocerca (Schatz 1952) and Periplaneta (Dennell and Malek 1955). The endocuticle is evidently the unmodified procuticle it being composed of chitin-protein complex. The cuticle in Thelyphonus does not give any indication of the presence of sulphur.

The histochemical and other reactions are summarized in Table 1.

b) Rhipicephalus. The abdominal cuticle of female tick differs from that of Thelyphonus in the absence of a pigmented exocuticle. Thus there are outer and inner epicuticle, mesocuticle and endocuticle (cf. Krishnakumaran 1961a). The fundamental pattern of the arthropod cuticle is however maintained. The epicuticle is a non-chitinous, lipoprotein layer while the lamellated portions are derived from the chitin-protein complex procuticle. Although there is no evidence of tanning in the abdominal cuticle of the female tick, in the scutellum the cuticle is amber coloured and also gives evidence for the occurrence of phenolic