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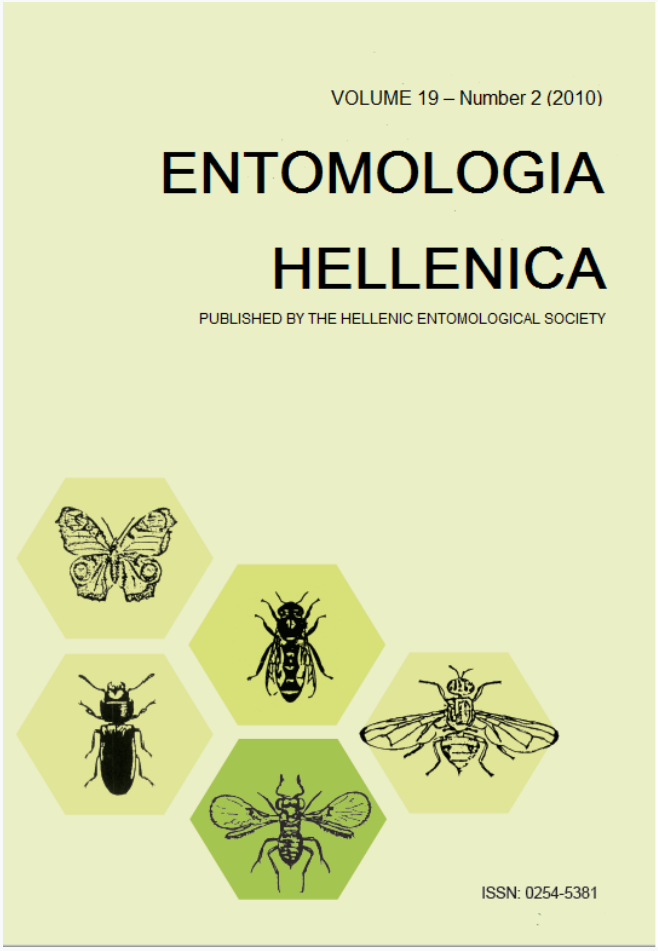

Scanning electron microscope observations on the multilocular disc-pores and dermal projections of adult female Porphyrophora tritici and P. cynodonti

H.A. Vahedi, F. Gholami Mahfar

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Scanning electron microscope observations on the multilocular disc-pores and dermal projections of adult female *Porphyrophora tritici* and *P. cynodontis*

H.A. VAHEDI* AND F. GHOLAMI MAHFAR

Plant protection Department, Agricultural Collage, Razi University, Kermanshah, Iran

ABSTRACT

The morphology of the multilocular disc-pores from the anterior part of the body (abdominal segments I-III and all thoracic segments, dorso-venterally) of two species of *Porphyrophora* (*P. tritici* and *P. cynodontis*: Hemiptera: Coccoidea: Margarodidae) were examined using a scanning electron microscope. The multilocular disc-pores of both species have 1 or 2 (rarely 3) rings of evenly or unevenly distributed loculi but the more central rings are almost always incomplete. Each outer ring of the more anterior disc-pores of *P. tritici* was complete, with 5-13 loculi, and the inner ring had 0-4 unevenly distributed loculi. Each disc-pore of *P. cynodontis* had a complete outer ring of 6-9 evenly distributed loculi and an inner ring with only 0-1 loculi. A bright central zone is absent in most cases. These two species are similar in that both have a dense network of small, globular projections throughout the derm surface. These observations are discussed in relation to those of light microscope studies.

KEYWORDS: SEM study, multilocular disc-pores, Hemiptera, Margarodidae, adult female, *Porphyrophora* spp.

Introduction

The genus *Porphyrophora* (Coccoidea: Margarodidae) contains about 105 species (Ben-Dov et al. 2010). It appears to be particularly abundant in the Middle East and in Central Asia, with 5 species in Iran where several are of some economic importance (Vahedi 2002, Vahedi and Hodgson 2007). Three of these species, namely *P. cynodontis* (Archangelskaia), [as a source of dyes], *P. tritici* (Bodenheimer) and *P. jashenkoi* Vahedi occur on cereals. They are not easy to separate either in the field or on microscope slides and the purpose of this research project was to see if there were obvious differences between the species in pore structure under the electron microscope.

Margarodid scale insects have a variety of different types of wax glands (microducts, simple pores, cicatrices and multilocular disc-pores). These secrete a diverse range of waxes, which vary in quantity, shape, size and chemical composition, and whose main function is probably protective (Hamon et al. 1975, Foldi 1981, Foldi 1997, Foldi 2005). These loculate disc-pores are distributed throughout the body and vary in their structure, abundance and distribution, both between genera and species although, within any given taxon, there tends to be a high degree of uniformity. This variability can provide important information for diagnosing taxa at all levels (Williams and Kosztarab 1972, Gimpel et al. 1974, Foldi 1978, Gullan 1979,

**Corresponding author, e-mail: Vnassah@yahoo.com*

Miller and Kosztarab 1979, Foldi 1981, Foldi and Cassier 1985, Foldi and Pearce 1985, Takagi 1990, Qin and Gullan 1992, Hodgson 1994, Foldi 1997, Vahedi and Hodgson 2007, Unruh 2008, Unruh and Gullan 2008a,b). Several of these workers have suggested that the differences in pore ultrastructure might be useful in separating closely related species and such differences have been used recently to revise the generic placement of some species (Takagi 1990, Ülgentürk and Willems 2001, Unruh 2008, Unruh and Gullan 2008a,b). The structure of these pores appears to be complicated and we considered that it might be better elucidated using the electron microscope. The external ultrastructure of the multilocular disc-pores of the adult females of two *Porphyrophora* species from Iran is described below.

Materials and Methods

The preparation techniques for scanning electron microscope (SEM) used here were mainly those used by Eisenbeis and Wichard (1987). Adult female specimens of *P. tritici* and *P. cynodontis* were fixed in 2% gluteraldehyde, post-fixed in osmium tetroxide and then rinsed and taken through a series of alcohols of increasing concentration and finally critical-point dried before mounting on SEM stubs, using double-sided sticky tape. The specimens were then coated with gold for 6-8 min in a Sputter Coater, and finally studied using scanning electron microscope at 15ev.

Results and Discussion

As my previous studies on adult female (Vahedi 2002, Vahedi and Hodgson 2007), *Porphyrophora tritici* and *P. cynodontis* are similar in that both are red and have fossorial prothoracic legs, long setae, many multilocular disc-pores (Fig. 1A-F) and a dense network of small, globular projections throughout the derm surface (Fig. 1C-D). These dermal projections usually disappear on slide mounted specimens. Vahedi (2002) and Vahedi and Hodgson (2007) noted that the derm of mounted material showed a reticulate pattern of small humps, particularly on young specimens. A similar pattern of globular projections has been noted on adult female *Matsucoccus feytaudi* Ducasse (Matsucoccidae) (Foldi 2004). It is likely that these "projections" or dermal papillae allow the derm of the adult female to expand as she matures and her ovaries swell. Similar papillae are a common feature of immature and young adult coccoids, allowing the derm to expand as each stage grows between moults.

The loculate disc-pores are circular and generally have 5-17 loculi in 1 or 2 rings of O-shaped loculi. The rings may form a single, evenly spaced loculi (Fig. 1B, C, E) or may be unevenly distributed (Fig. 1A, F). The loculate pores of some Margarodidae have a central ring of pores; these are never present on *Porphyrophora* species. Some disc-pores on *P. tritici* and *P. cynodontis* have a bright central zone surrounded by an area without loculi when seen under the light microscope but this zone was not visible on these pores under the SEM (Fig. 1A-F). The multilocular disc-pores of *P. tritici* are large, 12 µm, each with a 1 or 2 rings of loculi, the outer ring with usually 5-13 loculi and the inner ring with 0-4 unevenly distributed loculi (Fig. 1A-C). The disc-pores of *P. cynodontis* are smaller, 8 µm, and almost always have a complete outer ring of 6-9 evenly distributed loculi and an inner ring of 0-1 loculi (Fig. 1D-F). Whilst, these differences may seem small, this study does suggest that specimens which have 10 or more loculi in the outer ring and more than a single loculus in the inner ring are likely to be *P. tritici*; whilst those with 9 or fewer loculi in the outer ring and a single loculus in the inner ring are likely to be *P. cynodontis*.

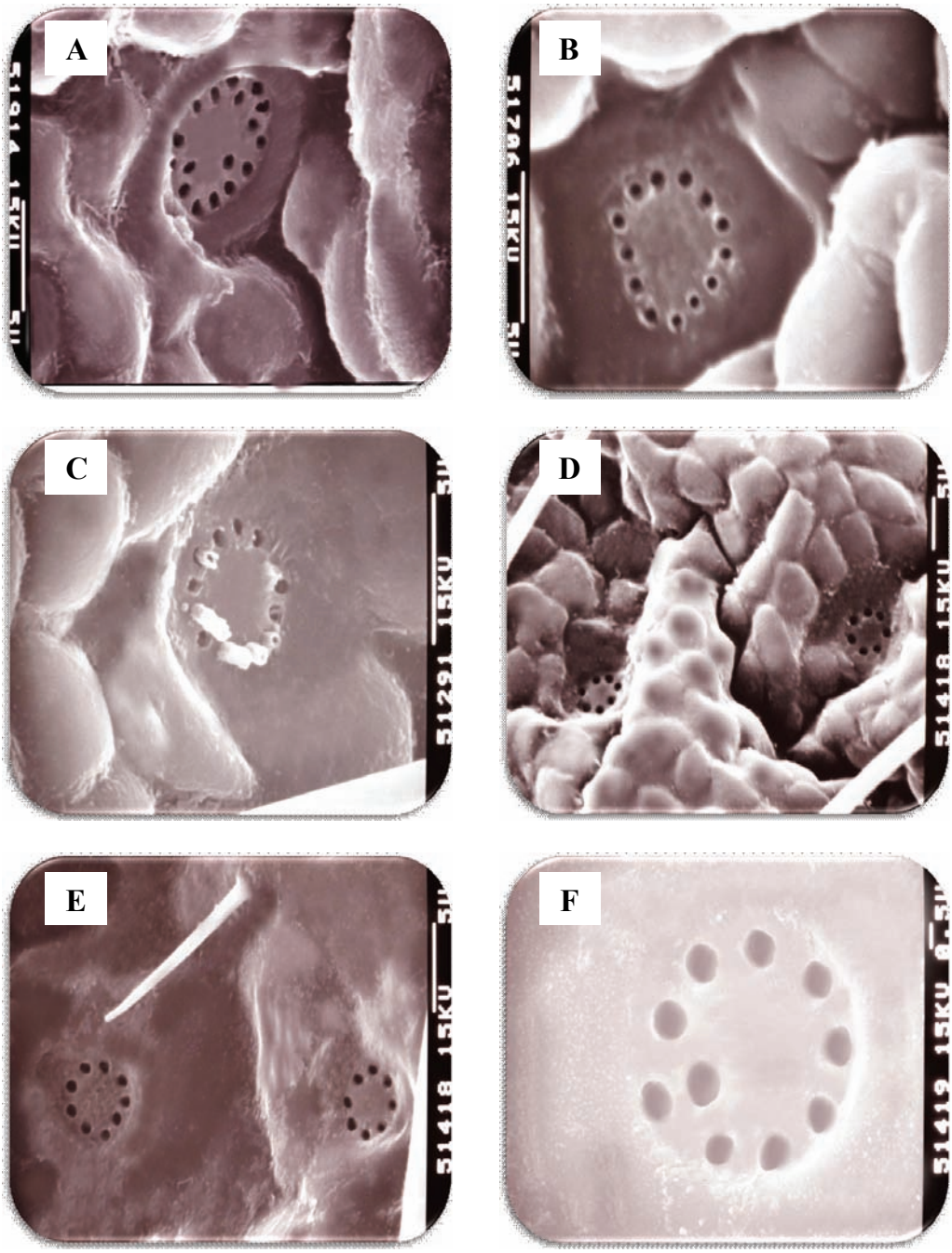

FIG. 1. Scanning electron micrograph showing external views of the multilocular disc-pores and the globular dermal papillae A-C: Those of *P. tritici*; C also shows some loculi producing cottony filamentous wax and the globular dermal papillae (in the left side). D-F: Those of *P. cynodontis*; D also shows a dense network of small projections or papillae [bar = 5μm].

However, the above descriptions of the disc-pores of *P. tritici* and *P. cynodontis* differ from the light microscope study of Vahedi (Vahedi 2002, Vahedi and Hodgson 2007). On *P. tritici*, the light microscope study suggested that the multilocular disc-pores had 1-3 rings of evenly or unevenly distributed loculi, each outer ring complete with 7-14 loculi; the middle and inner rings almost always incomplete, with 8-10 loculi in each middle ring and with the inner ring mostly absent but occasionally with 1 or 2 loculi. The posterior disc-pores, particularly around genital opening, tended to be larger, usually with 3 rings of loculi, the outer ring with 14-16 loculi, middle ring with 6-8 loculi and inner ring 1 or 2 loculi. The situation with *P. cynodontis* was very similar. The light microscope study found that the number of loculi varied depending on the position on the body, with the most anterior disc-pores having 1 ring of evenly distributed loculi, each ring mainly with about 12 loculi, while the more posterior disc-pores tended to be larger, almost always with 2 rings of loculi, outer ring with about 16 loculi and inner ring with 8 loculi.

Several workers have suggested that the differences in pore ultrastructure might be useful in separating closely related species and such differences have been used recently to revise the generic placement of some species of scale insect families, such as Coccidae, Diaspididae and Monophlebidae (Takagi 1990, Ülgentürk and Willems 2001, Unruh 2008, Unruh and Gullan 2008a,b). This scanning electron microscope (SEM) study on the ultrastructure of multilocular disc-pores of *Porphyrophora* species has provided a clearer understanding of their structure.

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Παρατηρήσεις σε ηλεκτρονικό μικροσκόπιο των πολυσημειακών δισκοειδών πόρων και προεξοχών του δερματίου σε ενήλικα θηλυκά άτομα των ειδών *Porphyrophora tritici* και *P. cynodontis*

H.A. VAHEDI ΚΑΙ F. GHOLAMI MAHFAR

Plant protection Department, Agricultural Collage, Razi University, Kermanshah, Iran

ΠΕΡΙΛΗΨΗ

Η μορφολογία των πολυσημειακών δισκοειδών πόρων από το πρόσθιο μέρος του σώματος (κοιλιακά άρθρα I-III και όλα τα θωρακικά) των ειδών *Porphyrophora* (*P. tritici* και *P. cynodontis*: Hemiptera: Coccoidea: Margarodidae) εξετάστηκε με τη χρήση ηλεκτρονικής μικροσκοπίας. Οι πολυσημειακοί δισκοειδείς πόροι και στα δύο είδη έχουν 1 ή 2 (σπάνια 3) δακτυλίους με ομοιόμορφα ή μη κατανεμημένα σημεία αλλά οι πιο κεντρικοί δακτύλιοι είναι σχεδόν πάντα μη πλήρως σχηματισμένοι. Οι εξωτερικοί δακτύλιοι των πρόσθιων δισκοειδών πόρων του *P. tritici* ήταν πλήρεις, με 5-13 σημεία, και ο εσωτερικός με 0-4 σημεία. Κάθε δισκοειδής πόρος του *P. cynodontis* είχε ένα πλήρη εξωτερικό δακτύλιο με 6-9 σημεία και ένα εσωτερικό με 0-1 σημεία. Αυτά τα δύο είδη είναι παρόμοια στο ότι έχουν ένα πυκνό δίκτυο από μικρές σφαιρικές εξοχές σε όλη την επιφάνεια του δερματίου. Τα παραπάνω ευρήματα συζητούνται σε σχέση με τις παρατηρήσεις μελετών σε οπτικό μικροσκόπιο.