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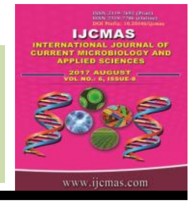

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Comparative Structure and Morphometry of Ultimate Rostral Segment of Different Aphid Species on Various Fruit Crops in Punjab, India

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A B S T R A C T

The study was based on field samples of six aphid species collected from three fruit crops namely citrus, peach and pear. The structure and morphometric data of ultimate rostral segment (URS) were compared based on 4th nymphal instar and alate adults. In fourth instar nymph, the maximum length of URS was observed in *T. aurantii* on citrus followed by *Toxoptera* spp. on pear but in alate adult, it was observed maximum in *A. gossypii* whereas *M. persicae* on citrus had minimum URS length in case of 4th instar nymph and alate adult.

Keywords

Alate, Aphid species, Nymph, URS, SEM imaging.

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Introduction

Aphids (Hemiptera: Aphididae) are truly interesting group of herbivorous insects and can affect plants directly or indirectly by feeding on the plant's sap (Blackman and Eastop 2007) as these are important pests of agriculture, horticulture and forestry having worldwide distribution. They have experienced some adaptations in relation to host plants so that many aphid taxa have biologically complex life cycles (Martin and Brown 2008). This has led to the occurrence of several distinct morphs in most aphid taxa which make their identification difficult. Taxonomists have frequently used morphological variations as primary parameters in the differentiation and separation of many natural populations of organisms and many species have been described based on the results of these studies (Mehrparvar *et al.*, 2012). Phytophagous insects comprise many cryptic species that have been recognized during last several decades (Feder *et al.*, 1998; Dres and Mallet 2002). Cryptic pest aphid species are highly dynamic and rapidly evolving groups with diverse ecological characters (Blackman and Eastop 2007; Lozier *et al.*, 2008). Most aphid species comprise a set of closely related populations which may have diverged genetically so that they could be considered as host races, incipient or sibling species (or subspecies) (Blackman and Eastop 2007). The recognition of these divergent populations can help to understand their ecology and evolution so that we could devise effective control programmes. Aphids are one of the most difficult genera to identify because of its morphological conservation and this problem is complicated by the fact that several species may live on a single host plant. In other word they show a very high degree of host-specific behavioural adaptations. The degree of phenotypic plasticity varies among phytophagous insects, because they have different capacities for morphological, physiological and behavioural adaptations in response to the nutritional, chemical and physical structure of their host plants (Scheiner 1993; Via *et al.*, 1995).

Morphological characters of aphids are very important in adaptation to various host plants. Due to this variability, some structures are considered of diagnostic value in description of new species. Among these, ultimate rostral segment which comprises of fourth and fifth rostral segments together play an important role in species identification but in many cases, fifth segment is hard to differentiate. No study is available on these morphometric parameters of the aphid species on various fruit crops in India as well as other countries. Thus, keeping in view the importance of aphids' species, the present studies were conducted at Punjab Agricultural University, Ludhiana during 2014-15 to elucidate the different morphs of any species.

Materials and Methods

For the morphometric studies of six aphid species, viz. *Toxoptera aurantii* (Boyer de Fonscolombe), *Aphis gossypii* Glover and *Myzus persicae* (Sulzer) on citrus; *Brachycaudus helichrysi* (Kaltenbach) and *M. persicae* on peach and *Schizaphis piricola* (Matsumura) and *Toxoptera* spp. on pear were collected from the College Orchard/Fruit Research Farm, Punjab Agricultural University, Ludhiana during 2014-15. The specimens so collected were then preserved in 75 per cent alcohol with glycerol in small glass vials which were subsequently labeled containing information like name, date of collection, host plant, location etc. Twenty-five specimens each of fourth nymphal instar and alate adult of each collection (i.e. out of 100 individuals preserved in vials) were observed individually for the preliminary segregation on the bases morphological similar characters. Any specimen found at variance was sorted out and kept in separate vial for further studies. These were dissected under compound microscope (Magnus MS 24) using different grades of absolute alcohol (30, 50, 70, 90 and 100 per cent). After preparing the slides, these were observed under Stereo Zoom microscope (4X) for their description.

Morphometry of ten slides of URS of 4th nymphal instar of six species and alate adult of three species like *B. helichrysi*, *M. persicae* (on citrus and peach) and *A. Gossypii* were recorded in micrometer under optical microscope (Zeiss-Axioskop 40 with Axiocam Camera and Axiovision software) at 50X. Photographs were taken with a Nikon 1300 camera attached on a Nikon SMZ 25 stereo zoom microscope (157.5X).

The structure of rostrum and ultimate rostral segment (URS) analyzed and imaged under Hitachi S-3400N Scanning Electron Microscope (SEM) operated at an accelerating voltage of 15.0 kV using secondary electron detector at the Electron Microscopy and Nanoscience (EMN) Laboratory, College of Agriculture, Punjab Agricultural University, Ludhiana as per standard protocol given by Bozzola and Russell (1999).

For comparison of means of different parameters of various species, critical difference was calculated separately for nymphs and alate adults by applying completely randomized block design using software CPCS-1.

Results and Discussion

Fourth instar nymph

The length of ultimate rostral segment (URS) of fourth instar nymph of different aphid species on leaves and flowers of various fruit plants (Table 1) revealed that *T. aurantii* had maximum length of ultimate rostral segment (12.01 ± 0.09 µm) followed by *Toxoptera* spp. and *A. gossypii* (11.70 ± 0.08 and 11.18 ± 0.03 µm, respectively). It is clear from the data that the length of URS of *A. gossypii* was statistically on par with *S. piricola* (11.09 ± 0.08 µm). Apart from these two, there was a significant difference among the aphid species. The minimum length (7.77 ± 0.09 µm) was observed in *M. persicae* on citrus which was significantly less to that of *B. helichrysi* and *M. persicae* on peach (10.24 ± 0.14 and 8.37 ± 0.07 µm, respectively). The comparative structure of segment IV of URS of 4th nymphal instar of different aphid species is given in Plate 1 (157.5X). The scan of the literature indicated that there is no information on this aspect.

The comparative structure of ultimate rostral segment (URS) (Plate 2) of different aphid species imaged under Scanning Electron Microscope revealed that in *A. gossypii*, URS had blunt apex and more numbers of setae than that of *T. aurantii* but setae were longer and thicker in nature. The URS was sharp and pointed in case of *S. piricola* with less and short setae, whereas it was short and heart shaped with more number of secondary setae in case of *M. persicae*. Regarding the length of rostrum, it is clear that *A. gossypii*, *T. aurantii* and *S. piricola* had the maximum rostral length than that of *M. persicae*.

Plate.1 Comparative structure of ultimate rostral segment (URS) of fourth instar Nymph of different aphid species (157.5X)

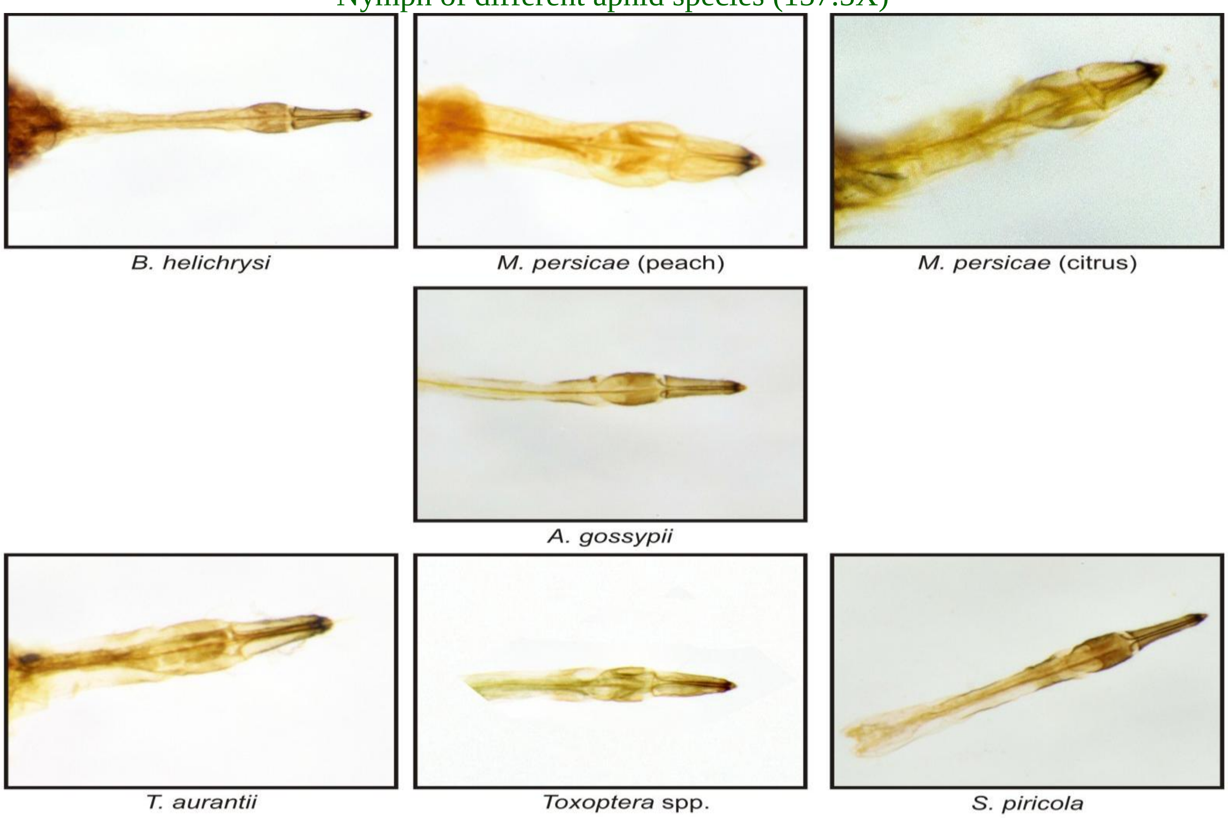

Plate.2 Comparative SEM imaging of ultimate rostral segment of different aphid species

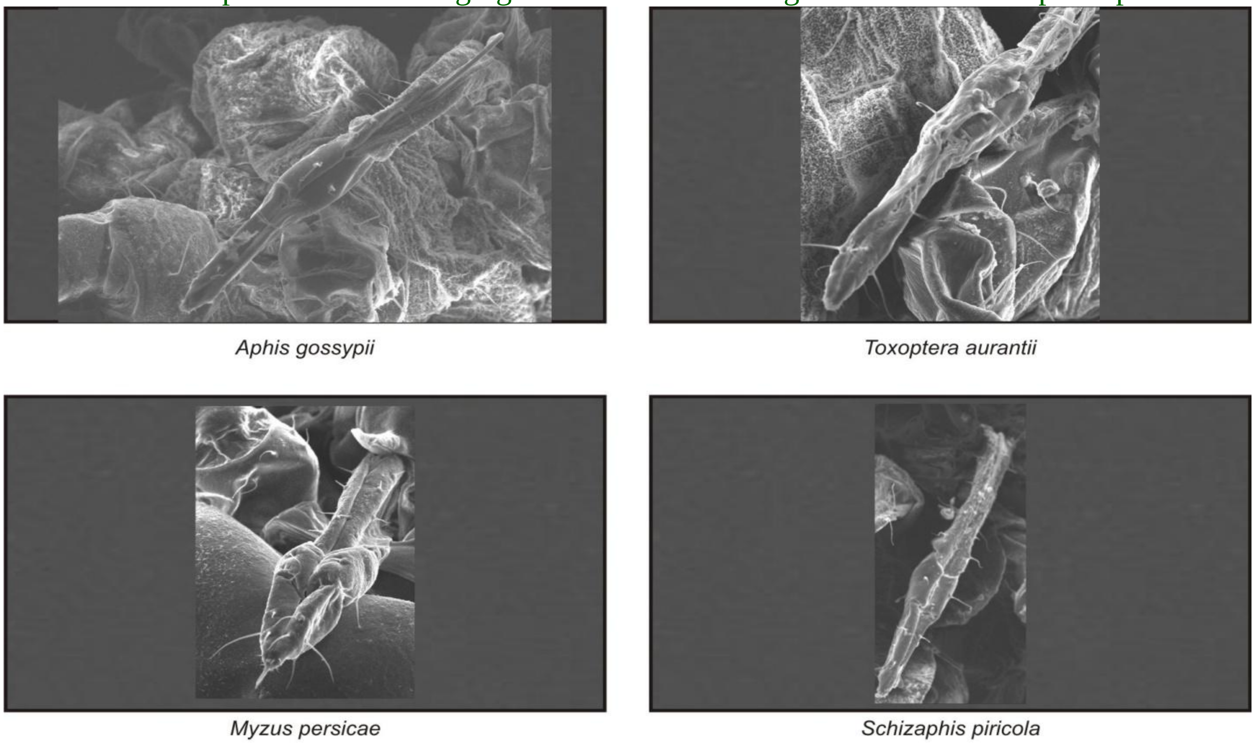

Plate.3 Comparative structure of ultimate rostral segment (URS) of alate adult of Different aphid species (157.5X)

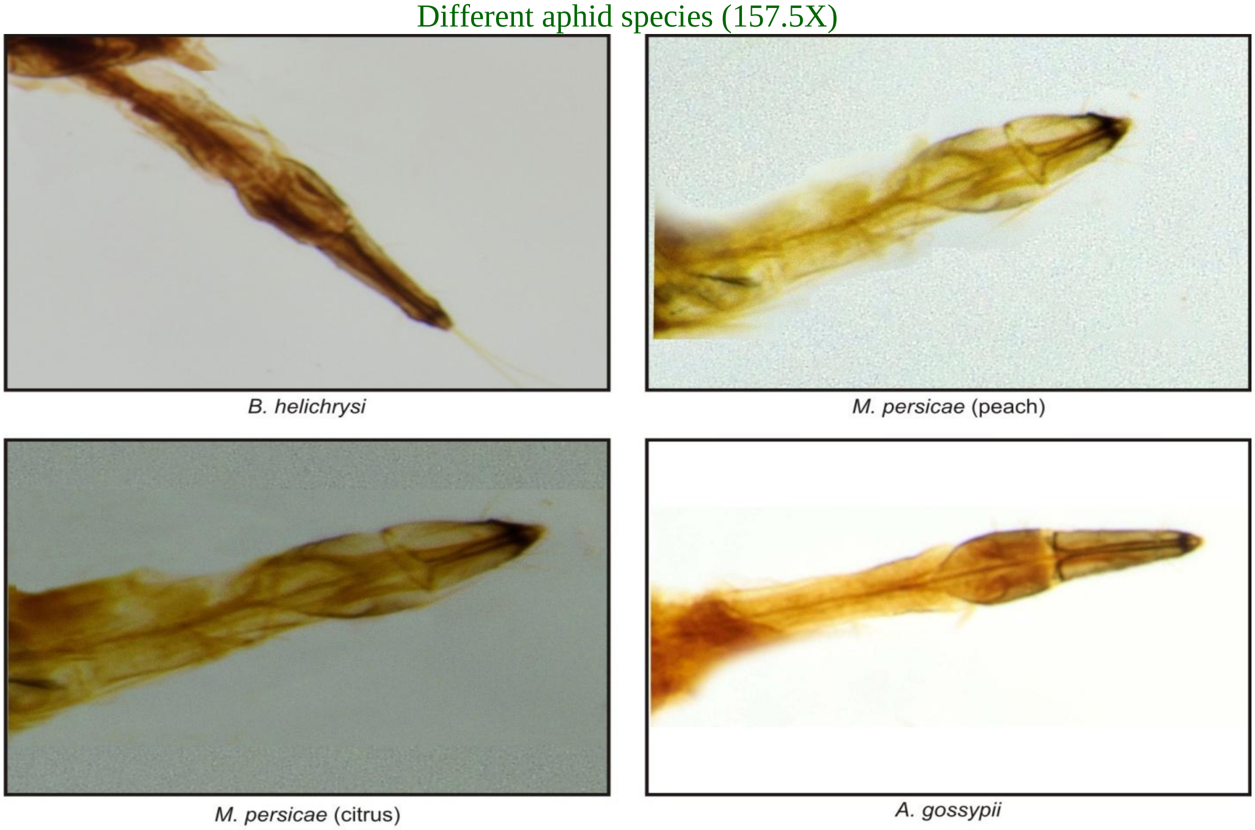

Table.1 Measurement of ultimate rostral segment (µm) of 4th nymphal instar of different aphid Species on leaves and flowers of various fruit crops during 2014-15

Aphid species	Host	Length (µm)*(157.5X)[1]
B. helichrysi	Peach	10.24 ± 0.14
M. persicae	Peach	8.37 ± 0.07
M. persicae	Citrus	7.77 ± 0.09
A. gossypii	Citrus	11.18 ± 0.03
T. aurantii	Citrus	12.01 ± 0.09
Toxoptera spp.**	Pear	11.70 ± 0.08
S. piricola??	Pear	11.09 ± 0.08
CD (p=0.05)		0.25

*Mean of 10 specimens ± SE; ** Specimens being identified; Species needs confirmation
[1]Total magnification = Zoom x M_{obj} x $M_{eyepiece}$

Table.2 Measurement of ultimate rostral segment (µm) of alate adult of aphid Species on leaves of various fruit crops during 2014-15

Aphid species	Host	Length (µm)*(157.5X)[1]
B. helichrysi	Peach	10.04 ± 0.08
M. persicae	Peach	8.32 ± 0.12
M. persicae	Citrus	7.93 ± 0.11
A. gossypii	Citrus	11.02 ± 0.14
CD (p=0.05)		0.32

*Mean of 10 specimens ± SE; [1]Total magnification = Zoom x M_{obj} x $M_{eyepiece}$

Alate adult

The length of URS (Plate 3) of alate adult of aphid species on leaves of various fruit plants (Table 2) indicated that *A. gossypii* had maximum length of URS (11.02 ± 0.14 µm) followed by *B. helichrysi* (10.04 ± 0.08 µm). It is evident from the data that there were significant differences among *B. helichrysi*, *M. persicae* and *A. gossypii*. The minimum length of URS was observed in *M. persicae* on citrus (7.93 ± 0.11 µm) which was non-significantly less to that of *M. persicae* on peach (8.32 ± 0.12 µm). Due to the hardy nature of citrus plant, the length of URS was slightly longer for piercing and sucking in case of *A. gossypii* than that of *M. persicae* which feeds on the younger leaves of peach plant.

In conclusion, the present studies showed that there was clear structural difference in the morphometry of URS obtained from SEM and stereo microscope imaging of different aphid species on the three different fruit crops which acts as an important diagnostic parameter for species description. Fourth instar nymph of *T. aurantii* had maximum length of ultimate rostral segment but in alate adult it was observed maximum in *A. gossypii* whereas *M. persicae* on citrus had minimum URS length in case of 4th instar nymph and alate adult. The present study clear indicated that within a species, URS of *B. helichrysi, A. gossypii, Toxoptera aurantii, Schizaphis piricola* and *Toxoptera* spp. did not show any variation and thus there are no nymphs or races of these species. However, URS of *M. persicae* on peach and citrus showed the variation and there could be different morphs on these host plants.

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