

New Acarine Setal Receptors of *Varroa Destructor*

M. E. Zakaria* and Sally F. Allam**

*Apiculture Dept., Plant Protection Research Institute, Agriculture Research Center, Dokki, Giza, Egypt

**Dept. of Agric. Zoology and Nematology, Fac. Agric. Cairo Univ.

ABSTRACT

The female mite nymphs of *Varroa destructor* Anderson and Truman were collected from sealed worker and drone brood cells and prepared for the scanning electron microscopic examination to identify type and structure of the setae receptors present on the leg I. Eighteen setae types were recorded and described. Thought they can perceive bee associated volatiles by means of receptors in wall-pore sensory hairs of the tarsal pit organ and legs.

Key Words: *Varroa destructor*, Mite, Nymph, Honey bees, Sense setae receptors.

INTRODUCTION

The parasitic mite *Varroa destructor* Anderson and Truman is currently the most serious global threat to honey bee colonies. Within a few years it has become a serious problem causing economic losses to most beekeepers and apiaries. Finally, it leads to the death of colonies or migration of queen and worker bees. There is a possibility that *Varroa* is hitch on this information channel of the insect society. Indeed, chemical stimuli guide *Varroa* to the resource. It can be argued that the most sensitive detector for biologically active substances is the mite itself through adaptively "matched filters" of its sensory system (Nazzi *et al.*, 2001). Action potentials recorded from receptor cells in olfactory sensilla tell whether volatile products can be detected by the mite's sensory system as in other arachnid and insect ectoparasites (Steullet and Guerin, 1992; Guerin *et al.*, 2000). Substances perceived by the mites can then be tested as cues for odor-mediated behaviors in a bioassay. *Varroa* uses its front legs in the same way as insects use their antennae (Rickli *et al.*, 2000) Olfactory sensilla organs occurred on the foreparts of *Varroa* legs resemble as in other arachnids (Milani and Nanelli, 1989; Ramma and Böckeler, 1989). The most tarsal segment bears a pit organ with sensory hairs in it (Hess and Vlimant, 1982). The ultra-structure and histology of these sensilla in *Varroa* bear a striking similarity to olfactory sensilla of other arthropods. So it can be assumed that at least some of these sensilla house receptors involve in the perception of volatile host cues. In the laboratory behavioural assay, there were many considering of the *Varroa* mite behaviors in the honey bee colonies superintendent. Female *Varroa* mite can recognize between drone and worker brood cells and prefer enter into the first one to diffuse the strong kiromone odors.

MATERIALS AND METHODS

Natural infested honey bee colonies (*Apis mellifera*

carnica) with varroa mites were conducted for this study. Worker brood cells were randomly chosen and examined for the mite presence. All mite individuals either, attached to the bee immature or the specimens found on the cell wall were collected and classified according to its sex (Delfinado and Baker, 1984). The female deutonymphs were identified and prepared for the scanning electron microscopic examination (Joel GM4200) at the Applied center of Entomoneematodes (ACE), Faculty of Agriculture, Cairo University. Twenty nymphs were dehydrated in ethyl alcohol and dried using the critical point procedure, then individually affixed using double-sided sticky tape, and sputter coated with gold palladium according to Fashing *et al.*, (2000) method.

RESULTS AND DISCUSSION

Varroa deutonymphs were photographed from the ventral side to questionnaire setae receptors. Eighteen setae receptors in different parts of the leg I were recorded.

The following setae receptors were established and classified as follows;

- 1- Eupathidium seta.
- 2- Famulus seta.
- 3- Tactile setae; A- Tactile seta. B- Boat seta
- 4- Spatulate setae; A-Spatulate seta. B- Spatulate opening. C- Lateral spatulate seta.
- 5- Curved seta.
- 6- Pen setae; A- Short tip pen seta. B- Big hole pen.
- 7- Mushroom seta.
- 8- Flame seta.
- 9- Basiconica seta.
- 10- Broken seta.
- 11- Nodulus seta.
- 12- PG seta.
- 13- Cubital seta.
- 14- Trichodea type A.

Photographing sensitive setae of the female *Varroa* nymphs revealed presence of different setae receptors on different parts of all segments of leg I (2 -5 -8- 6- 4- 20) from the coxa to tarsus;

Coxa has 2 setae, their lengths (63.63- 103.02 μm) with mean 83.325 μm ; v1, ad1 tactile.

Trochanter has 5 setae, their lengths (33.33-84.84 μm) with mean 52.72 μm ; V1, pl1 Trichodea, pl2 broken, ad1, ad2 tactile.

Femur has 8 setae, their lengths (36.36- 84,84 μm) with mean 68.175 μm . pl1, 2, 3, 4, 6, al1 tactile, pl5 spatulate, d1 pen seta, d2 pollen grain shape, v1 basoconical.

Genu has 6 setae their lengths (36.36- 66.66 μm) with mean 52.52 μm . d1, al1, al2,, pl3 tactile, d2 mushroom shape, d3 cubital shape.

Tibia has 4 setae, their lengths (48.48 - 60.60 μm) with mean 54.54 μm . al1,al2,pl1 and pl2 tactile.

Tarsus has 20 setae their lengths(27.27- 121.2 μm) with mean length 57.95 μm . P11 open spatulate, pl 2,4, d1, 3, 4, 5, 6, al1, 2, 3, 4, 5, tactile, pl1 open spatulate, pl3 spatulate, d2 eupathidium, d7 pen seta, d8 broken seta.

The following setae receptors were recorded on leg I and classified as follows;

1- Eupathidium seta (Epth):

A medium seta type with thin shape, pin edge (Fig. 3). No. 1 (Figs. 22, 23 and 24).

2- Famulus seta (Fam), as duck beak in the terminal edge.

A group of Actinopiline setae, situated on terminal segment of legs and palp (Fig. 4) No.2 (Figs. 22, 23 and 24).

3- Tactile setae

A- Simple tactile seta (Tct), medium to long setae with simple shape (Figs. 5 & 6) No. 3 (Figs. 22, 23 and 24).

B- Boat seta, boat shaped setae with deep groove, tapered edge and profound core in its base (Fig. 7) No. 4 (Figs. 22, 23 and 24).

Differences observed between decline and protrusion setae base are shown in Figs. 7&8.

4- Spatulate setae

A- Spatulate seta (S) (Fig.9) No. 5 (Figs 22, 23 and 24).

B- Spatulate opening (so) (Fig. 9) No. 6 (Figs. 22, 23 and 24) Spatulate seta with large opening hole.

C- Lateral spatulate (Ls) (Fig.10) No. 7 (Figs. 22, 23 and 24) Spatulate seta with small lateral orifice.

5- Curved seta (cv)

Seta with bent vertex (Fig.1) No. 8 (Figs 22, 23 and 24).

6- Pen setae

A- Short tip pen seta (Spn) (Fig. 12A) No. 9 (Figs. 22, 23 and 24).

B- Big hole pen (Bhp) (Fig.12B) No.10 (Figs. 22, 23 and 24).

7- Mushroom seta (Msh)

A seta has circle pinch shape just before ending its edge (Fig. 13) No. 13 (Figs. 22, 23 and 24).

8- Flame seta (Flm)

A short seta with flam shape at the top with deep circle groove in its base (Fig. 14) No. 12 (Figs. 22, 23 and 24).

9- Basiconica seta (Bas)

Dwarf seta with simple dipping (Fig. 15) No. 13 (Figs. 22, 23 and 24).

10- Broken seta (Brk)

Fracture seta without complete broken fracture (Fig.16) No. 14 (Figs. 22, 23 and 24).

11- Nodule setae (Nd)

Seta with small circle knots and simple hole in its base (Fig.17) No. 15 (Figs. 22, 23 and 24).

12- PG seta

Medium size seta with end like pollen grain shape (Fig.18) No. 16 (Figs. 22, 23 and 24).

13- Cubital seta (CT)

Short fragments seta with little hole (Fig. 19) No.17 (Figs. 22, 23 and 24).

14- Trichodea type A

Seta with smooth and curved top (Fig. 20).

Structure of the tactile seta as chemoreceptor organ was recorded (Fig. 21). It is worthy to note that female *Varroa* mites communicate with setae receptors on their legs. Trichobthria long, delicate and slender set in broad, shallow innervated sockets in the cuticle. Trichobthria are sensitive to vibration or near-field air movement and are a major sense organs of arachnids (Figs. 22, 23 & 24).

It could be summarized that testing leg I of the deutonymphs of *Varroa* mite (*V. destructor*) electroscanning, revealed presence of eighteen setae receptors. They are established and classified as follows:

- 1- Eupathidium seta.
- 2- Famulus seta.
- 3- Tactile setae; A- Simple tactile seta. B- Boat seta.
- 4- Spatulate setae; A-Spatulate seta. B- Spatulate

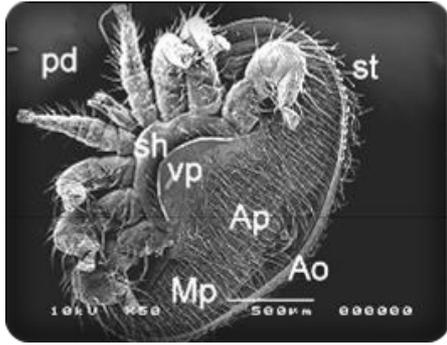


Fig.(1): Deutonymph of *Varroa destructor*
 Pd: Pedipalp. Vp: Ventral plate. Sh: Sternal shield. Ao: Anal opening. Ap: Anal plate. St: Setae. Mp: Metapodal plate.

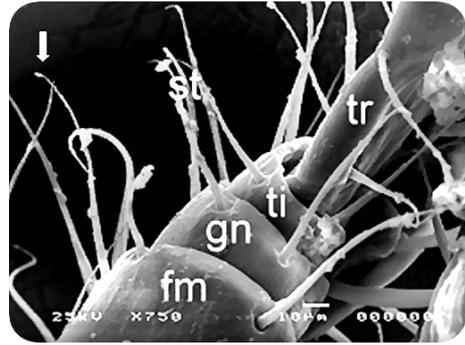


Fig. (2): Details of leg I of Deutonymph female *Varroa* mite.
 Tr: Tarsus Ti: Tibia Gn: Genu
 Fm: Femur St: setae receptors

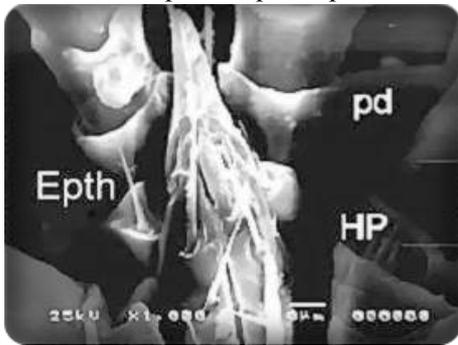


Fig. (3): Eupathidium seta (Epth) present on the gnathosoma of female *Varroa* nymph.
 Pd: Pedipalp. Hp: Hypostoma.

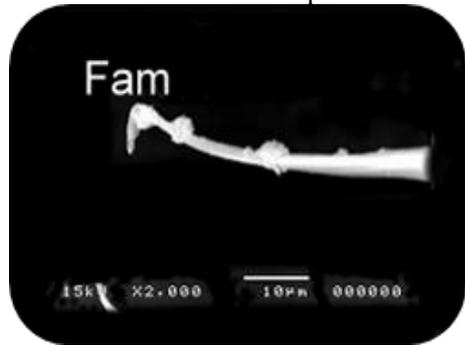
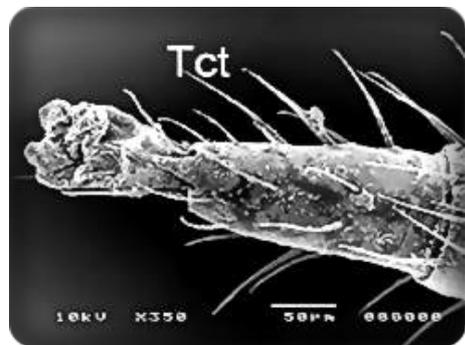
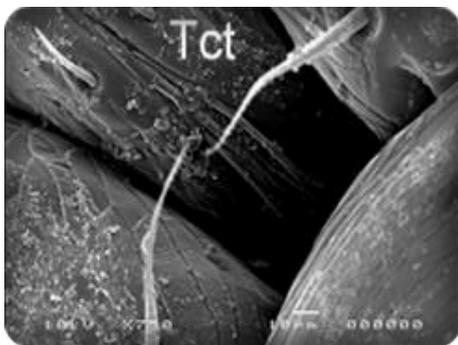


Fig. (4): Famulus seta (Fam).



Figs. (5 & 6): Tactile setae (Tct).

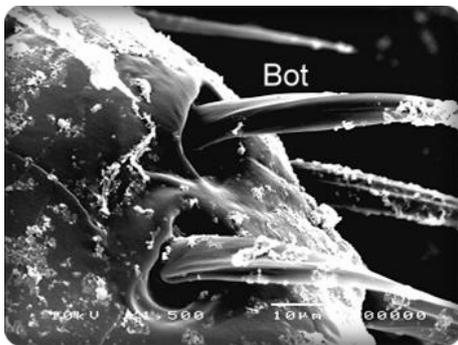


Fig. (7): Boat shaped seta has deep groove with tapered edges (Bot).

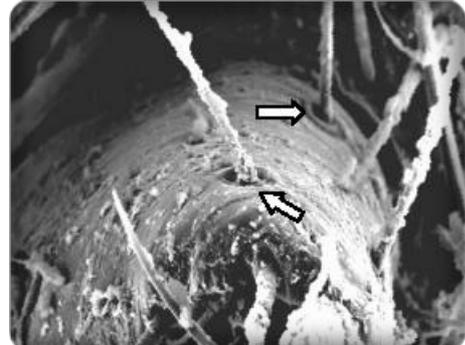


Fig. (8): Protrusion the seta base in comparable to another similar shown in Fig. (7).

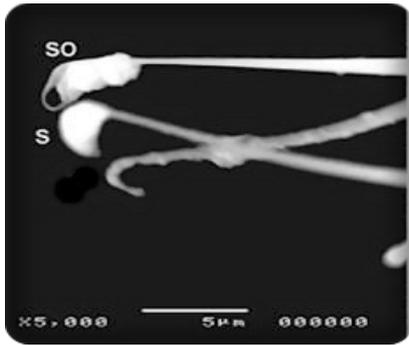


Fig. (9): Spatulate seta (s) and spatulate opening seta with large opening hole (So).



Fig. (10): Lateral spatulate seta (Ls).



Fig. (11): Curved seta (Cv).



Fig. (12 A): Pen seta: Short tip pen seta (pn).

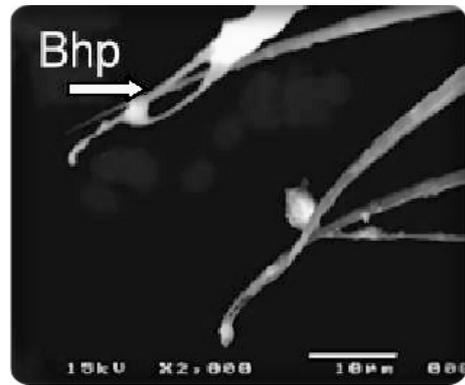


Fig. (12 B): Pen seta: Big hole pen seta (Bhp).

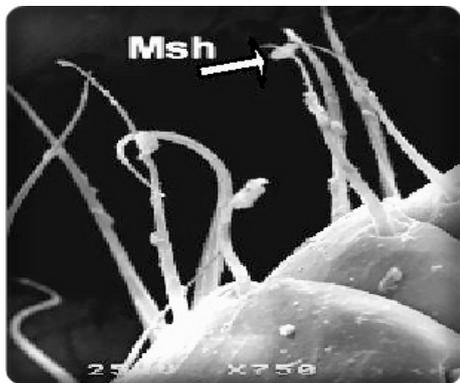


Fig. (13): Mushroom seta (Msh).

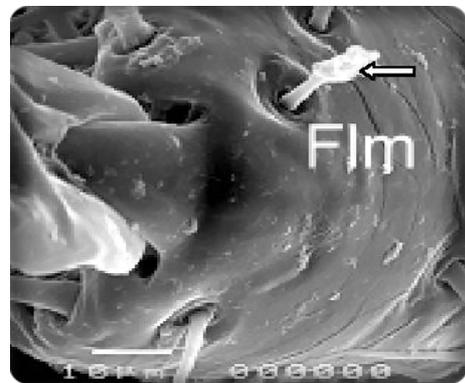


Fig. (14): Flame seta (Flm).

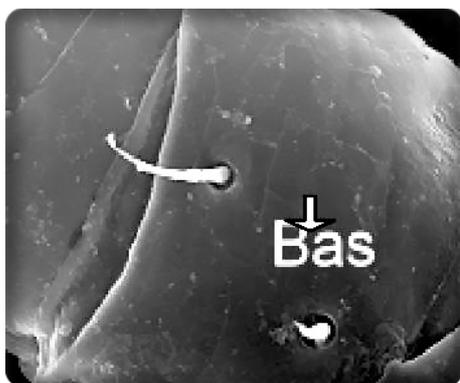


Fig. (15): Basophile seta (Bas).

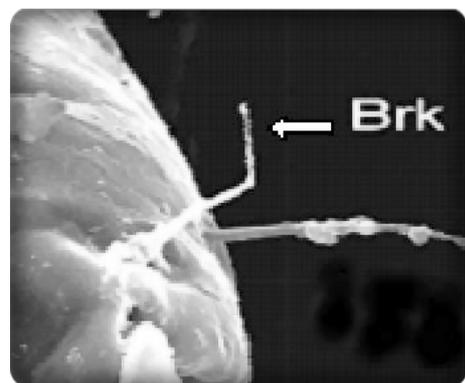


Fig. (16): Broken seta (Brk).

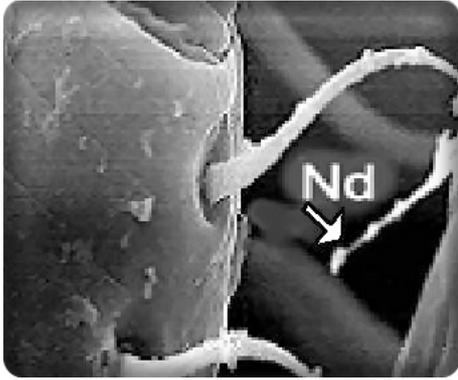


Fig. (17): Nodule seta (Nd).

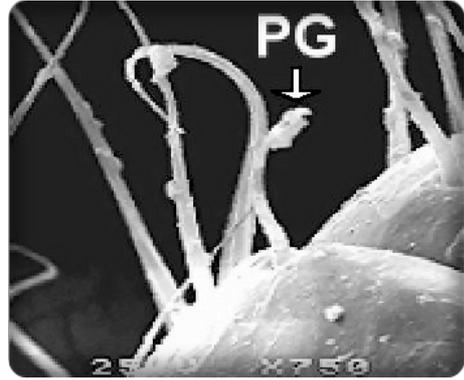


Fig. (18): PG seta (as pollen grain).

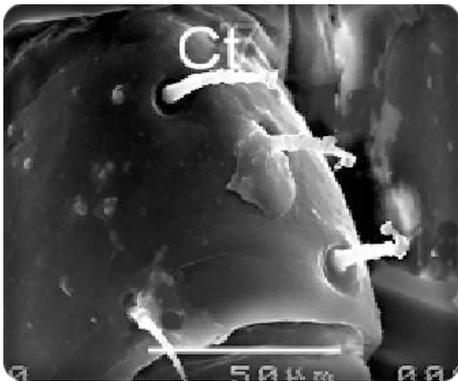


Fig. (19): Cubital seta (Ct).



Fig. (20): Trichodea type A(TA).

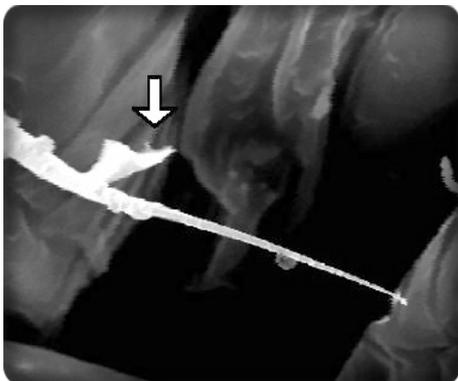


Fig. (21): Structure on the tactile seta as chemoreceptor organ.

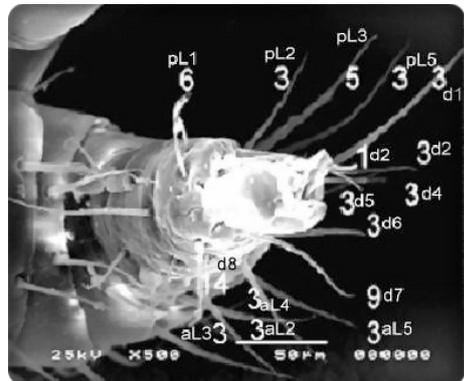


Fig. (22): Distribution of setae on genu and tarsus.

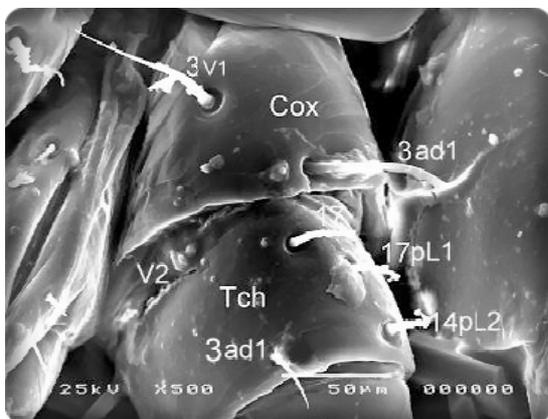


Fig. (23): Distribution of setae on coxa and trochanter.

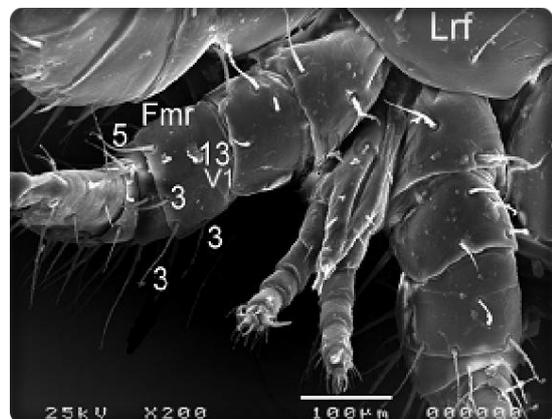


Fig. (24): Distribution of setae on femur.

- opening. C- Lateral Spatulate seta.
 5- Curved seta.
 6- Pen setae; A- Short tip pen seta. B- Big hole pen.
 7- Mushroom seta.
 8- Flame seta.
 9- Basiconica seta.
 10- Broken seta.
 11- Nodules seta.
 12- PG seta.
 13- Cubital seta.
 14- Trichodea type A.

Dasher and Smith, (2008) found relationship between olfactory interference in honey bee workers and a form of backward pairing-induced inhibitory learning memory. Electrophysiological action potentials from receptors on the olfactory sensilla on the first leg of *Varroa* mite were recorded (Dillier *et al.*, 2003). Hess and Vlimant, (1982) reported that the most distal tarsal segment bears a pit organ with sensory hairs in it. The ultra-structure and histology of these sensilla in *Varroa* bear a striking similarity to olfactory sensilla of other arthropods. So it can be assumed that at least some of these sensilla house receptors involve in the perception of volatile host cues. Volatiles from worker and drone brood are relevant for brood cell invasion by *Varroa* in a behavioral assay where mites move from nurse bees to honeybee brood cells in absence of contact by the nurse bees with the brood. Electrophysiological studies affirmed that *V. destructor* can perceive bee-associated volatiles and volatiles from air. The small dimension of the olfactory pit organ makes it very difficult to get good recording from the sensory cells in the olfactory sensilla (Endris and Baker, 1993). Among the synthetic compounds that provoke olfactory responses, benzaldehyde is known as a volatile in royal jelly and adult drones. Benzaldehyde and methylsalicylate are also constituents of pollen, flowers and honey (Maga, 1983). Receptors for these products are known from arachnids. A benzaldehyde receptor is described in olfactory sensilla on the tarsus of the tick *Amblyomma variegatum* Fabricius (Steullet and Guerin, 1994). Methylsalicylate receptors occur on the mite *Phytoseiulus persimilis* (de Bruyne *et al.*, 1991). Brood cell invasion behavior is probably influenced by a multiple of cues detected by different sensory pathways in *V. destructor*. This leads to an average ten fold higher infestation rate of drone brood than worker brood of *Apis mellifera* L (Martin, 1998).

REFERENCES

- Dasher, M. and Smith, B.H. 2008. Olfactory Interference during Inhibitory backward pairing in honey bees. *Plos one*, 3(10): 1-14.
- De Bruyne, M.; Dicke, M. and Tjallingii, W. F. 1991. Receptor cell responses in the anterior tarsi of *Phytoseiulus persimilis* to volatile kairomone components. *Exp. App. Acarol.*, 13: 53-58.
- Delfinado-baker, M. 1984. The nymphal stages and male of *Varroa jacobsoni* Oud. a parasite of honey bees. *Inter. J. Acarol.*, 10: 75-80.
- Dillier F. X.; Guerin, P.; Fluri, P.; Imdorf, A.; Meyre, P. B.; Vilmant, M. and Mitteilung, P. A. D. 2003. Odour-mediated transfer to brood cells of its honey bee host, *Apis mellifera* and olfactory cell responses of *Varroa destructor* to volatiles. *Swiss Bee Research Center*, 217-219.
- Endris, J. J. and Baker, T. C. 1993. Aktionspotentiale, abgeleitet am ersten Laufbein von *Varroa jacobsoni*, nach olfaktorischer Stimulierung. *Apidologie*, 24: 488-490.
- Fashing, N. J.; Oconnor, B. M. and Kitching, R. L. 2000. *Lamingtona carus*, a new genus of Algophagidae (Acari: Astigmata) from water filled treeholes in Queensland. Australia. *Invertebrate Taxonomy*, 14: 591-606.
- Guerin, P. M.; Körber, T.; McMahon, C.; Guerenstein, P.; Grenacher, S.; Vlimant, M.; Diehl, P. A.; Steullet, P. and Syed, Z. 2000. Chemosensory and behavioral adaptations of ectoparasitic arthropods. *Nova Acta Leopoldina NF 83*, 213-229.
- Hess, E. and Vlimant, M. 1982. The tarsal sensory system of *Amblyomma variegatum* Fabricius (Ixodidae, Metatrata), Wall pore and terminal pore sensilla. *Rev. Suisse Zool.*, 89: 713-729.
- Maga, J. A. 1983. Honey flavor. *Lebensmittel-Wissenschaft und Technologie*, 16: 65-68.
- Martin, S. 1998. A population model for the ectoparasitic mite *Varroa jacobsoni* in honey bee (*Apis mellifera*) colonies. *Ecological Modelling*, 109: 267-281.
- Milani, N. and Nanelli, R. 1989. The tarsal sense organ in *Varroa jacobsoni* Oud.: SEM observations, In: Cavolloro R. (Ed.) Present status of Varroa infestation in Europe and Progress in the *Varroa* mite Control. Commission of the European communities, Luxembourg: 71-82.
- Nazzi, F.; Milani, N.; Della Vedova, G. and Nimis, M. 2001. Semiochemicals from larval food affect the locomotory behaviour of *Varroa destructor*. *Apidologie*, 32(2): 149-155.
- Ramma, D. and Böckeler, W. 1989. Ultrastrukturelle Darstellungen der Sensillen in der Vordertarsengrube von *Varroa jacobsoni* (Acari). *Zool. Jb. Anat.*, 119: 221-236.
- Rickli, M.; Guerin, P. M. and Diehl, P. A. 2000. Palmitic acid released from honeybee worker

- larvae attracts the parasitic mite *Varroa jacobsoni* on a servosphere. *Naturwissenschaften*, 79: 320-322.
- Steullet, P. and Guerin, P. M. 1992. Perception of vertebrate breath components by the tropical bont tick, *Amblyomma variegatum*. II. Sulfide receptors. *Journal of Comparative Physiology A*, 170: 677-685.
- Steullet, P. and Guerin, P. M. 1994. Identification of vertebrate volatiles stimulating olfactory receptors on tarsus I of the tick *Amblyomma variegatum* Fabricius (Ixodidae). I. Receptors within the Haller's organ capsule. *Journal of Comparative Physiology A*, 173: 27-38.