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# SCANNING ELECTRON MICROSCOPIC (SEM) OF HYPOPHARYNGEAL AND MANDIBULAR GLANDS OF HONEY BEE WORKERS INFESTED BY VARROA MITES (*Varroa destructor*)

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## ABSTRACT

Hypopharyngeal and mandibular glands of healthy, heavily infested and deformed newly emerged honey bee workers with varroa mites were dissected and examined by Scanning Electron Microscope (SEM). The hypopharyngeal glands of heavily infested newly emerged workers were atrophied with little number of their lobules, accompanied with presence of abnormal shaped of globular growths of their lobules. These changes were clearly observed in the deformed infested bees. Infestation with varroa mites particularly in the deformed infested bees showed obvious shrinkage of mandibular glands and smaller structures of the reservoir glands.

***Key words:*** *Apis mellifera, hypopharyngeal gland, mandibular gland, Varroa destructor.*

## 1. INTRODUCTION

The honey bee parasitic mite *Varroa destructor* (Anderson and Trueman, 2000, formerly known as *Varroa jacobsoni*) is the most serious pest of managed honey bees (*Apis mellifera* L.) world wide. Varroatosis causes several losses in apiaries in Egypt and other

countries in the world (De Jong *et. al.*, 1982; Abd- El Wahab, 1996). The varroa mite is an obligate ectoparasite that feeds on the haemolymph of larval and pupal bee in capped brood cells and adult bees (Achou & Soltani 1997; Anderson & Trueman, 2000). The mite sucks blood from immature and adult bees, introducing viral pathogens that cause deformities and poor bee health (Kanga and James, 2002). The hypopharyngeal and mandibular glands are the most important protein-producing glands in honey bee workers producing royal jelly and digestive enzymes for honey processing (Nour, 1988). Guards and forager bees produce 2-heptanone in their mandibular glands which have two pheromone functions, alarm releasing behaviour and releasing sting behaviour (Lensky and Cassier, 1995 and Zakaria, 2002). Several different methods have been developed to estimate the hypopharyngeal gland activity (Hassanein, 1952, Brouwers, 1982, Huang *et al.*, 1989, Hrassning and Cralsheim, 1998 and Zakaria, 2002). The morphology and secretory cycle of the hypopharyngeal glands of diseased honey bee workers (*Apis mellifera* L.) were affected by varroa mite infestation (Youssif-Khalil, 1992 and Zakaria, 2002). The secretory globules of infested workers were extremely reduced, leading to malformation which renders them unable to secrete royal jelly to feed larvae and queens (Wang and Moeller, 1971). It is necessary to study how far the damage caused to the important glands from varroa infestation of honey bee workers (De Jong, *et al.*, 1982 and Weinberg and Madel, 1985).

This work aimed to study the external changes occurring to hypopharyngeal and mandibular glands of honey bee workers infested with varroa mites using Scanning Electron Microscope (SEM).

## 2. MATERIALS AND METHODS

Heavily infested honey bee colonies of Carniolian hybrid (Not less than 3 colonies) were used for this study during autumn season, 2003. Another three healthy colonies were used as control. The mean percentage of infested worker brood by varroa mites in the heavily infested colonies was 20% according to the method of De Jong *et al.*, 1982. These colonies did not receive any chemical control against varroa infestation during the experimental period.

For SEM studies, samples of worker bees from each infested colony (each sample including newly emerged worker bees infested

with more than 7 female mites / bee, as well as infested deformed bees), as well as from healthy colonies were dissected for obtaining the hypopharyngeal (HG) and mandibular glands (MDG) according to the method of Snodgrass (1956). These glands were fixed with 4% Glutaraldehide in O-2-M phosphate buffer at pH of 7.4 and temperature of 4° for 12 hrs., then they were fixed in 2% O2 O4 at room temperature for 3 hrs. according to the technique of Liu, *et al.*, (1989).

The different samples of hypopharyngeal and mandibular glands were examined in Jeol JXA 840 A SEM in the National Research Center Dokki, Giza, Egypt.

## 3. RESULTS AND DISCUSSION

Scanning Electron Microscope (SEM) of the hypopharyngeal glands (HG) of healthy newly emerged honey bee workers showed the presence of many large regular lobules (Figs. 1A &2A). The (HG) from heavily infested workers were clearly atrophied and wrinkled, while little density of their lobules were found as shown in (Figs 1B & 2B). The hypopharyngeal glands of deformed infested newly emerged workers showed abnormal pear shaped lobules with adenoma of the glandular germinates as shown in Figs. (1C &2C), accompanied with the presence of a lower number of lobules (Fig. 1C).

The mandibular glands (MDG) from healthy workers showed elongated and strong sac in each side of the head with an epithelium of secretory cells lined with a thin cuticular intima seems to be divided to two large parts, one for secretion and another acting as a reservoir (Snodgrass, 1956 and Stort *et al.*, 1986) as shown in Fig. (3A).

Heavily infested honey bee workers with varroa mites had disunited structures and stunts in the reservoir of the mandibular glands as shown in Fig. (3B), in spite of no differences recorded in the secretion cells between healthy and infested workers (Figs. 4A&B). Severe shrinkage of the glandular cells was subjected with deformed bees (Figs. 3C &4C), while unrestricted shaped of the reservoir glands was remarked as shown in Fig. (3C).

From the obtained observations it could be concluded that the hypopharyngeal and mandibular glands were deeply affected by infestation with varroa mites in both newly infested and deformed worker bees. The parasitic mite sucked the haemolymph of the

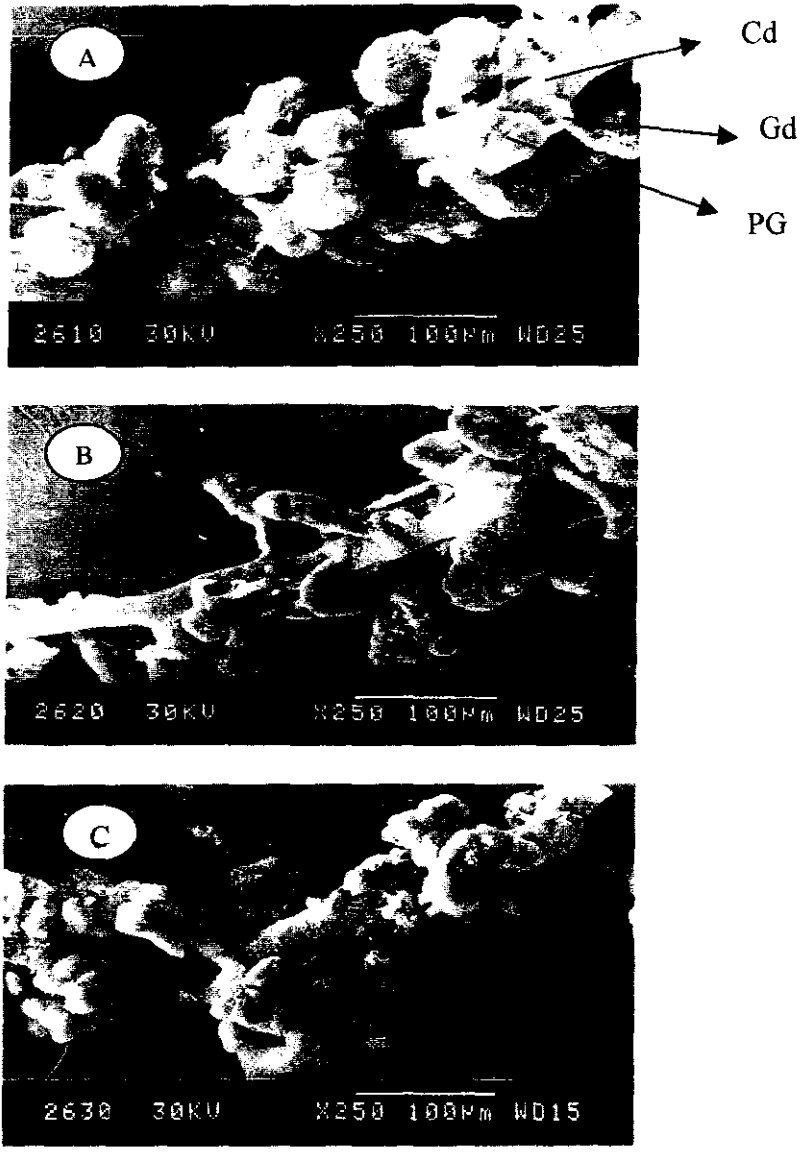


**Fig.(1) :Scanning electron microscope view of hypopharyngeal gland of healthy newly emerged honey bee workers (A); shrinkage and wrinkles on the surface of hypopharyngeal gland of heavily infested bee workers (B) and adenoma pear shape of the hypopharyngeal gland lobules of deformed workers (C).**
**Cd: Common duct Gd: Glandular duct PG: Pharyngeal glandular lobule**

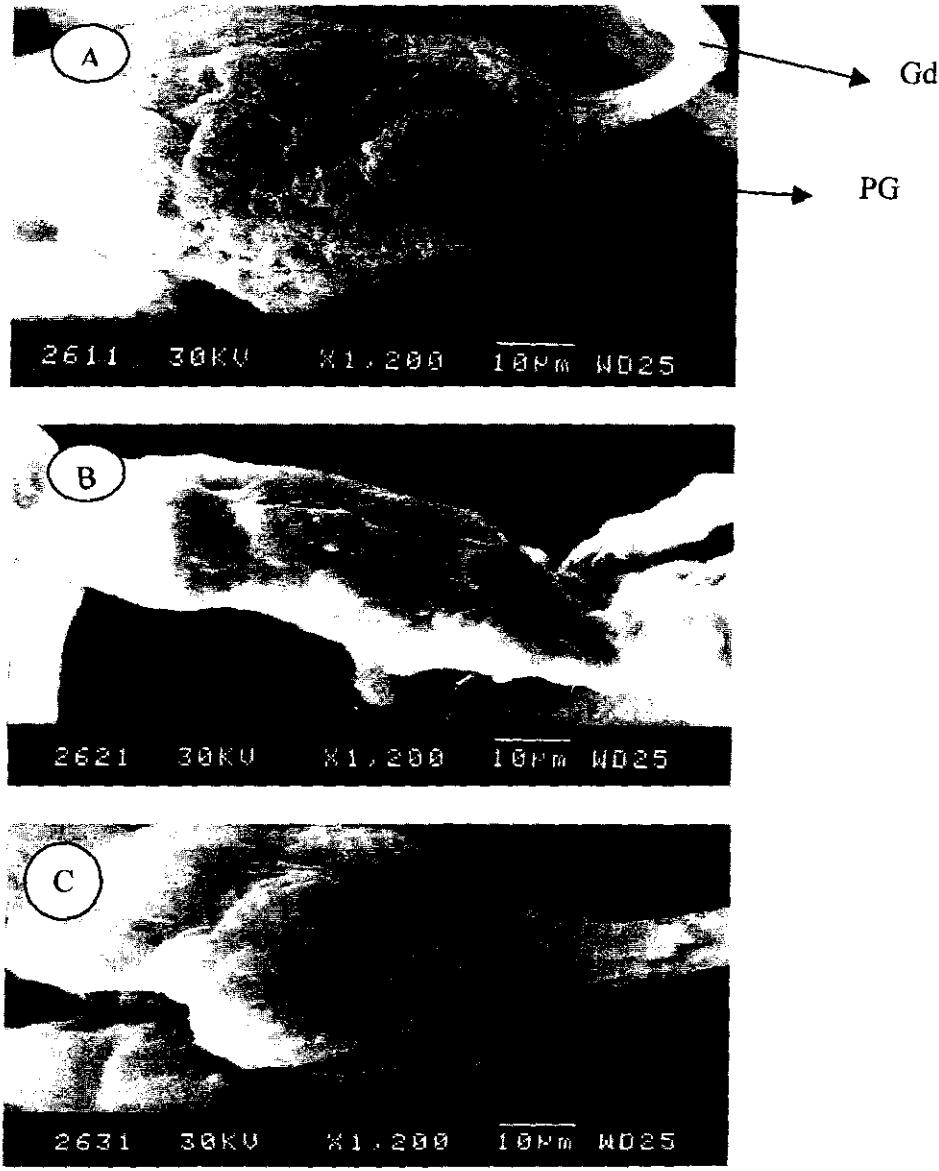


**Fig. (2): Scanning electron microscope view of hypopharyngeal gland lobule (A) of healthy emerged honey bee worker; atrophied shaped of glandular lobule (B) of heavy infested honey bee worker and adenoma pear shape of the gland lobule of deformed newly emerged honey bee worker (C).**
**Gd: Glandular duct PG: Pharyngeal glandular lobule**

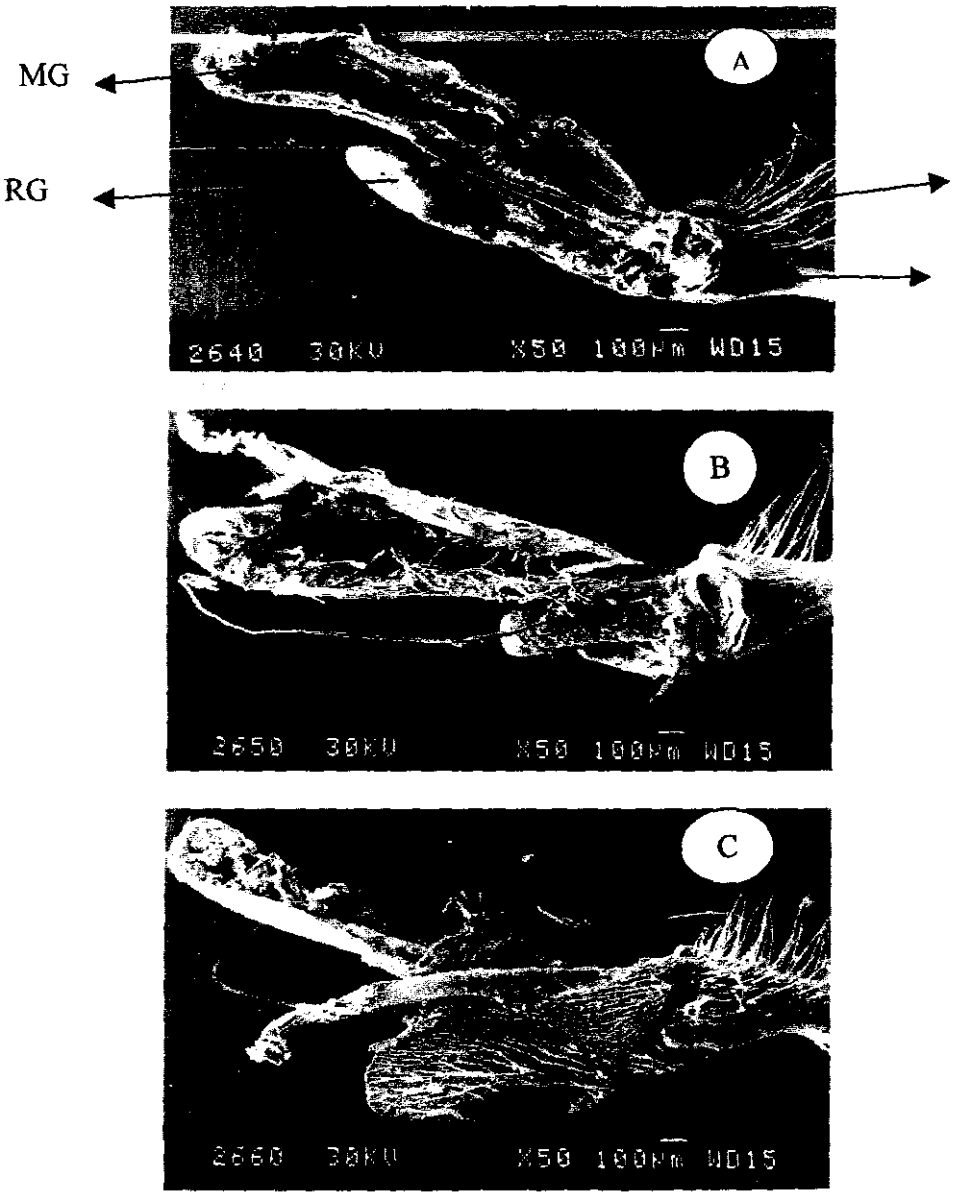


**Fig. (3): Scanning electron microscope view of mandibular gland of healthy newly emerged honey bee workers (A); disturbances in the mandibular gland structure of severely infested workers (B) and disunited structure of mandibular gland of deformed workers (C).**
**MG: Mandibular gland RG: Reservoir gland S: Setae**
**Md: Mandible**

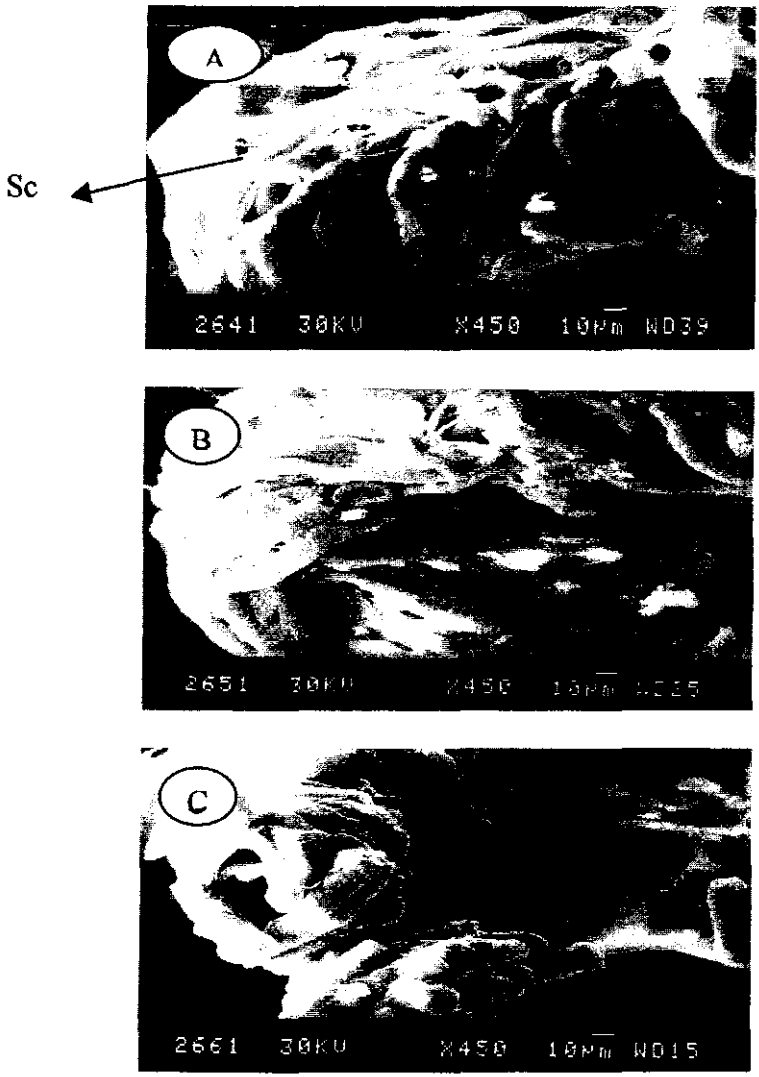


**Fig. (4): Scanning electron microscope view of secretory glandular cells of mandibular gland of healthy newly emerged honey bee workers (A); disturbances in the secretory glandular cells of severely infested bee workers (B) and stunts in the secretory cells of deformed honey bee workers (C). Sc: Secretion cells.**

infested worker bees which contains protein components and resulted in severe damage in the both tested glands (HG & MDG). These bees are unable to perform their functions inside and outside the colony and this is considered one of the main reasons for decline products of the honey bee colony. The activity of the diseased colonies may be reduced including the ripening process of honey and the production of royal jelly (Graham, 1992). Youssif-Khalil (1992) and Zakaria (2002), found that the infestation of honey bee workers by varroa mites decreased significantly the HG development. The secretory activity of these glands produces less amounts and lower quality of the royal jelly.

Severe damage in Hypopharyngeal and Mandibular glands was clearly observed in deformed bees due to the heavily infestation by varroa mites in comparison with newly emerged infested bees. Hrassning and Grailsheim (1998), reported that HG was often used to describe the physiological status of honey bee workers. Getchev and Belemezove (1997), found that newly emerged workers infested with varroa mites had many pathological effects in the HG. Wang and Moeller (1969) and Liu *et al.*, (1989) mentioned that nosema diseased bees are influenced concerning the size of the hypopharyngeal gland lobules.

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## مسح بالميكروسكوب الإلكتروني لغدة الغذاء الملكي والغدة الفكية في شغالات نحل العسل المصابة بطفيل الفاروا ( *Varroa destructor* )

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## ملخـــص

تم فصل غدد الغذاء الملكي والغدد الفكية لشغالات نحل العسل حديثة الفقس المصابة بشدة والمشوهة نتيجة الإصابة باكاروس الفاروا وفحصها لمقارنتها بالغدد السليمة بواسطة الميكرسكوب الإلكتروني.

أوضحت النتائج المتحصل عليها أن غدد الغذاء الملكـــي فـــي الشـــغالات المصابة بشدة بطفيل الفاروا بها ضمور مع قلة في عدد فصوص الغدة مصحوبـــة بظهور شكل غير طبيعي في النمو الكروي لفصوص الغدة.

وجد كذلك انكماش واضح في الغدد الفكية وصغر في مكونات مخزن الغدة في الشغالات المصابة خاصة الشغالات المشوهة نتيجة الإصابة.

المجلة العلميـــة لكلية الزراعة – جامعة القاهرة – المجلد (٥٥) العـــدد الثـــالـــث (يوليو ٢٠٠٤):٣٧٥-٣٨٤.