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# *Natural Plant Essential Oils for Controlling the Grasshopper (Heteracris littoralis) and their Pathological Effects on the Alimentary Canal*

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**Abstract.** In the present study, the toxic effect of three different natural essential oils of medicinal plants, namely Garlic (*Allium sativum)*, Mint (*Mintha pipereta)* and Eucalyptus (*Eucalyptus globulus)* were tested on 1st nymphal instar of the grasshopper (*Heteracris littoralis)*. The $LC_{50}$ values of the tested oils were estimated after 14 days from feeding on treated diet mixed with different concentrations of the oil. The $LC_{50}$ of the tested oils were arranged as follows: 0.067, 0.075 and 0.084ml. /100ml. diet for Garlic, Eucalyptus and Mint respectively. The effect of $LC_{50}$ concentration of the oils on the biological aspects and histological changes that observed on the alimentary canal and fat bodies were recorded. The normal development of the grasshopper was exhibited. Results cleared that there was statistical variable numbers of increased the nymphal periods, life cycle, adults longevity and life span comparing with the control test. Garlic oil inhibited egg lying by the resulting females offspring of the treated1st instar nymphs. High reduction in the deposited eggs and egg fertility caused by Eucalyptus or Mint oil and marked malformation were observed. Histological changes on the alimentary canal and fat bodies of the remaining nymphs after treatment with Garlic oil (the most effective oil) were detected by the light microscope have been recorded. The results suggest that the natural plant essential oils of Garlic, Eucalyptus and Mint may be used in IPM control program against *H. littoralis* grasshopper.

**Key words:** grasshopper, *Heteracris littoralis,* medicinal plants, *Allium sativum*, *Eucalyptus globulus, Mintha pipereta,* toxicity, histopathological changes, alimentary canal.

**Used abbreviations:** Crop (co); Gastric caecae (gc); Midgut (md); Malpighian tubule (mp); Ovary (ov); Testes (ts); Intestine (int); Rectum (rc); Anus (an); Epithelial cells (ep); Columnar cells (cc); Goblet cells (gc); Nucleus (n); Goblet cavity (gv); Regenerative cells (rgc); Microvilli (mv); Peritrophic membrane (pr); Circular muscles (cm); Longitudinal muscles (Lm);; Trachea (Tc); Vacuoles (V); Gastric caeca (gc); Intiman (In); Muscular layer (Ml); Fat cells (fc); Fat droplets (Fa); Degenerated epithelial cells (dep);; Basement membrane (bm), hindgut (hg); Lumen (lu) . Food particles (fo).

## Introduction

The grasshopper (*Heteracris littoralis*) considered one of the most harmful pests to different cultivated crops in Egypt. Its economic importance comes from attacking many vegetable cultivated areas even trees, feeding on it and causing great losses in quantity and quality of the attacked crops. In some cases thousands of cultivated hectares may be attacked by the swarms of grasshopper leaving it as a divested desert. The economic injury of *H. littoralis* in Egypt had been documented by MISTIKAWI (1929). Many grasshopper species could be found all over the year round and represent a pest status for many plants. One of these

grasshoppers is *H. littoralis* (IBRAHIM, 1983 & EL–SHAZLY, 1991). Essential oils are volatile, natural, complex compounds characterized by a strong odor and are formed by plants as secondary metabolites. In nature, essential oils play an important role in protection of the plants as antibacterial, antiviral, antifungal, insecticides and also against herbivorous by reducing their appetite for such plants. They also may, attract some insects to favor the dispersion of pollens and seeds or repel undesirable others (BAKKLI *et al*, 2008). The discovery and use of synthetic insecticides have reduced the interest in plant origin products. However, widespread use of these insecticides in public health and agriculture for the control of vector and pest species has created different problems, such as the development of physiological resistant in major vector species, environmental pollution and toxic hazards to human and other non- target organisms due to their broad spectrum of activity (WHO, 1992; 2005; HEMINGWAY & CRIAG, 2004; KOUL *et al*., 2008). As a result , there has been an increased interest in developing potential alternative or additional control methods or materials that are effective against the target vector species, environmentally safe, biodegradable, with low cost and can be used by individuals and communities in specific situation (REEDWANI *et al*., 2002). One of these potential alternatives or additional control methods or tools in the use of selective botanical derivatives against the target insect species (PERICH *et al*., 1995). Therefore, the use of essential oils extracted from aromatic plants to control insect pests has been investigated and is well documented (ISMAN, 2006; KOUL *et al*., 2008; RAJENDRAN & SRIEANJINI, 2008). For example, recently, the essential oil of Catnip (*Nepeta cataria* L.) was reported to have repellency against adult male cockroaches *Beriblanate germanica* L. (PETERSON *et al*., 2002). THAVARA *et al*. (2007) studied seven commercial essential oils for repellency against cockroaches and found *Citrus hystix* exhibited complete repellency against *P. americana* and *B. germanica*. TARE & SHARMA (2004) reported the larvicidal activity of essential oils of 11 plants against *Aedis aegypti* larvae. JANTAN *et al.* (2005) evaluated the leaf essential oils of eight *Cinnamomum* species for larvicidal activity against *A. aegypti* and *A. albopictus* they found five species of them to have significant larvicidal effects. MORIAS *et al.* (2006) evaluated the larvicidal activity of essential oil of four *Croton* species, they found three of them to be highly toxic against larvae of *A. aegypti.* Onion, Parsley and Cumin oils showed acceptable toxicity level against desert locust *Schestocerca gregaria* (ABDEL – HAMID, 2006). EZIAH *et al.* (2011) found that application of neem oil at 5 ml/l were effective dosage in preventing the development of *Ephestia cautell* larvae, mortality ranged from 32.5 – 55% after 96 hours of exposure period, the observed mortality can be attributed to the inherent properties of neem, mortality was dosage and time dependent.

The use of natural plant products is considered more sustainable, cheaper and safer. Natural pesticides are preferred for many reasons**:** 1 - They are cheaper compared with chemical pesticides. 2 - They are made from local resources and are readily available. 3 – They are safer to humans than chemical pesticides. The objective of the present work was mainly to evaluate in laboratory the toxicity and biological activity of three of plant essential oils Garlic (*Allium sativum*) , Eucalyptus (*Eucalyptus globulus*) and Mint (*Mintha pipreta*) for disrupting growth and development of *H. littoralis* and to evaluate the histological damages caused to the alimentary canal. The essential oils investigated in this study are used as pharmaceuticals and in flavoring and they are can use as safe bioinsecticides for controlling *H. littoralis*. However, the possibility of employing this natural insecticides in the management of the grasshopper is plausible, but is worthy of further investigation.

**Materials and methods**

Adults and nymphs of *H. littoralis* were collected from Giza governorate, Egypt. The colony was raised in laboratory stock and

reared in electrical heated wooden cages at constant temperature at 30 ± 1°C with fluctuating relative humidity (50 – 70%). Insects were fed on synthetic diet mentioned by SHARABY *et al.* (2010). For oviposition, cages were supplied with suitable oviposition pots. These pots were examined every day and, when laid in, were removed to glass jars (ca. 100 c.c.), hatched hoppers were transferred to large jars (ca. 7000 c.c.). After the fourth or fifth molt, hoppers were released in the larger cages. Biological notes were recorded including the developmental duration of each nymphal instar, number of instars, pre-Oviposition period, number of eggs per egg–pod, number of egg–pods per female, oviposition period and the duration of the post–oviposition period as well as effect of the treatments on development and reproduction. To study reproduction and longevity, ten pairs of newly emerged nymphs were used in pairs; each pair was placed in large glass jars. Each jar was provided with an oviposition pot and supplied with piece of synthetic diet for feeding replacing it every four days or when consumed. The experiment was conducted at 30°c and L.D. 12:12 and the relative humidity fluctuated between 50 - 60%.

*Plant essential oils*

Three essential oils: Garlic (*Allium sativum,* Liliaceae), Eucaluptus (*Eucalyptus globulus*, Myrataceae), Mint (*Mentha piperita*, Labiatae) were obtained from EL-Captain company (CAPPHARM), Alobour city (Cairo), Egypt. For determine the $LC_{50}$ concentration of the different plant essential oils on the 1St nymphal instar of *H. littoralis*, five descending concentrations that permit the computation of $LC_{50}$ was diluted on the basis of volume/volume (1, 0.5, 0.25, 0.13, 0.06%) from each plant oil were prepared by mixing known volume from the oil with 100 ml. diet during the diet preparation, one drop of Triton x100 was added as emulsifier for obtained the desired concentration. The treated diet poured into plastic box and kept in refrigerator till use. A piece of the treated diet was introduced into jars with containing 1St nymphal instar for feeding on it for seven days then remained treated diet replaced by untreated one, number of dead individuals were counted each day after treatment till 14 day (the end point) for calculating $LC_{50}$ values. For each concentration, 25 individuals were tested in five replicates, 5 nymphs each. Control was fed on untreated diet. $LC_{50}$ were determined according to (FINNEY, 1971) and mortality percent was corrected according to Abbott's formula (ABBOTT, 1925). After determined the $LC_{50}$ value of each oil, different biological aspects of the resulted insects have been recorded. The newly emerged 1St nymphal instar was fed on diet mixed with the prepared concentration of the oil for seven days then the diet replaced by untreated one till reached to the adult stage. The remaining adults were noticed for egg oviposition and egg hatching. The different biological aspects were recorded for each plant oil, for each test, 200 insects were used. Statistically, all data were subjected to Analysis of Variance (ANOVA) through SPSS Computer Program, To differentiate between means, Duncan's multiple range test (DUNCAN, 1965) was used ($P$ = 0.05).

*Histological examination*

The remaining treated nymphs after feeding for 7 days on the treated diet then on untreated diet for another seven days were used for morphological and histological examinations of the alimentary canal, control groups were also examined. Alimentary canal were dissected in 0.9 % NaCl solution and fixed in Bouin's solution for 24 hrs (HUMANSON, 1962) then dehydrated in ascending alcoholic series and cleared in xylen for few seconds then specimens were infiltrated in three changes of paraffin wax each lasted 20 minutes. Paraffin blocks were prepared and 6 u longitudinal or cross section were cut and stained with Ehrlich's acid haematoxylin and alcoholic eosin. The stained sections were dehydrated, cleared and mounted using D.P.X. for examination.

**Results and Discussion**

The toxicity of the tested three different essential oils on the 1st nymphal instar of *H.*

*littoralis* is shown in Table 1. Latent toxicity was observed, for this $LC_{50}$ values were recorded after 14 days from the treatments. According to the $LC_{50}$ values it could be arranged as follows: Garlic 0.067 > Eucalyptus 0.075 > Mint 0.084 ml./100ml of diet. A parallel course was obviously seen, a mostly between the mortality and the oil concentration levels. The variable toxicity may be due to the constituents of each oil and disturbance or the hormonal regulations (AL-SHAROOK *et al*., 1991), 200 species of plants, which produce chemicals substances able to act against insects, are known. The substances can have poisonous and repellent effects and can work as phagorestrainer ovicide and can affect the insect's hormonal system. Moreover, a great number of essential oils can reduce the reproduction system of several insects and they can also hinder the growth the development and the reproduction of some herbivore insects (HILL, 1990; PARTES *et al*., 2000). KOUL *et al.* (2008) Found that Cineol and Limonene (terpes) and essential oils of *Eucalyptus globulus* and *E. canaldulensis* have shown poisonous effects through the cuticle and in ingestion and fumigation, causing 100% of mortality of *S. zeamais* in dilution 2:8 (essential oil: acetone). DUKE (2005) proved that *E. globulus* species has in its composition, 71% Cineol, pinen, terpinen, anethol, benzaldehyde, estragol, eugenol, limonene, linalool, menthol, methilchavicol, methilicinamate, ocimen, rutin and thimol. The same author recorded that the essential oil of *E. globulus* seem as natural or botanical potential insecticide, once the secondary plant metabolic have been used as pesticides or as models for synthetic pesticides, thus thy can cause poisonous interference in the biochemical and physiologic functions of herbivore insects. KWON & SANG (2005) studded the effect of plant essential oils from 29 plant species for their insecticidal activities against the Japanes termite, *Reticukitermes speratus* kolabe, using fumigation bioassay, they found good insecticidal effects with essential oils of *E. globulus* and other *E.species* and Garlic oil between the other tested oils, Garlic gave 100% mortality within 2 days of treatment, three major compounds from garlic oil were identified as tri and disulfide. NAGANAWA *et al.* (1996) mentioned that Garlic (*Allium sativum*) essential oil containing sulfoxide sulfated terpenoids (Allicin, Ajoene), they recorded inhibition of microbial growth by Ajoenen, a sulfur–containing compounds that derived from garlic.

**Table 1.** Susceptibility of the grasshopper *H. littoralis* 1st instars nymph to the natural plant essential oils mixed with the artificial diet.

| **Plant Essential oils** | **LC values mg,/100ml. diet** | | | **Fiducial limit** | | **Slope** |
|---|---|---|---|---|---|---|
| | **LC25** | **LC 50** | **LC 90** | **Upper** | **Lower** | |
| Garlic *Allium sativum* | 0.009 | 0.067 | 2.89 | 0.1038 | 0.031 | 0.7843 |
| Eucalyptus *Eucalyptus globulus* | 0.008 | 0.075 | 5.23 | 0.1187 | 0.0315 | 0.6951 |
| Mint *Mintha pipreta* | 0.001 | 0.084 | 215.62 | 0.1723 | 0.0053 | 0.376 |

The data presented in Table 2 cleared the biological aspects of the $LC_{50}$ values of the tested essential on *H. littoralis* nymphs, there were statistical significance differences between all nymphs instars periods. The total nymphal period was 68.6, 63.4 and 57 days for Garlic, Eucalyptus and Mint oil respectively which reduced to 46 day of the control taste. Life cycle increased in case of Eucalyptus 84.0 and Mint 77.8 and Garlic oil 68.6 days comparing with the control 65.6 days. The adults that resulting from 1st nymphal instars that fed on diet mixed with Garlic oil could not deposited egg pods at all, Garlic oil inhibited egg laying or reproduction, this may be as a result of its inhibition effect on oogenesis. Preoviposition period lasted 34.8 and 35.4 days for

Mint and Eucalyptus oil comparing with 20.6 in control. Oviposition period greatly decreased from 39.8 days on control to 21.2-23.4 days for oils treatments. The mean numbers of the deposited eggs decreased from 152 eggs in control to 56.6 - 43.6 eggs for Mint and Eucalyptus oils respectively. Also there were great reduction in the eggs hatchability from 24 and 45.2 comparing with 95%. Reduced fertility was observed due to a failure of many oocytes to mature. Longevity of the resulted adults increased in case of Garlic oil. There were percentage of mortality reached 50–52% and malformation ranged from 45.6- 51.6% on the oils treatment. Life span was affected by the oil treatments they take 171-186 days comparing to 164 days in control. Inhibition of egg reproduction by Garlic oil may be related to the effect of oil on the digestive and enzymatic system that reflect on hormonal release. These results agree with the finding of ZUDAIRE *et al.* (1998), who cleared that, the gut of locust disturbed and showed disrupted in the endocrine cells as affected by food nutrition content, insect age and stage. HUSSEIN *et al.* (1994) mentioned that the effect of plant extracts on maggot of *Erias insulana* may due to digestion and absorption of plant oil. FREISEWINKEL & SCHMUTTERER (1991) showed that the treatment of fifth instar nymphs of *L. migratiria* by neem oil led to increase mortality mainly during molts, prolonged periods of development, and reduced fitness. GHZAWI (2005) recorded that lower doses of Azadirachtin treatment the 4th and 6th nymphal instars of *H. littoralis* caused prolongation of the insect duration, leading finally to death. MOHAMMED & EL-GAMMAL (2002) reported that Azadirachtin caused prolongation in duration of the last instar nymphs of *S.gregaria*.

**Table 2.** Biological parameters of *H. littoralis* treated as 1st instar nymph with the $LC_{50}$ values of the tested natural plant essential oils that mixed with the artificial diet.

| Biological parameters | Plant essential oils Mean ± S.E | | | | F-value |
|---|---|---|---|---|---|
| | Garlic | Mint | Eucalyptus | Control (Artificial diet) | |
| 1st instar nymph | 12.00±0.32 a | 9.00±0.63 b | 10.80±0.58 a | 8.20±0.37 b | 12.082 ** |
| 2 nd instar nymph | 12.60±1.08 a | 9.20±0.73 b | 11.80±0.86 a | 8.80±0.37 b | 5.499 ** |
| 3rd instar nymph | 13.60±1.36 a | 11.40±0.51 ab | 12.60±1.03 a | 9.20±0.37 b | 4.321 * |
| 4th instar nymph | 14.00±0.84 a | 12.60±1.08 a | 13.00±1.14 a | 9.60±0.40 b | 4.321 * |
| 5th instar nymph | 16.40±1.03 a | 14.80±1.24 a | 15.20±0.86 a | 10.20±0.37 b | 8.502 ** |
| Total nymphal period (in days) | 68.60±1.69 a | 57.00±2.70 b | 63.40±2.29 a | 46.00±0.45 c | 24.265 ** |
| Life Cycle(in days) | 68.60±1.69 b | 77.80±3.01 a | 84.00±2.21 a | 65.60±0.87 b | 16.261 ** |
| Pre-Oviposition period (in days) | 0.00±0.00 c | 34.80±1.62 a | 35.40±2.23 a | 20.60±1.08 b | 125.936 ** |
| Oviposition period (in days) | 0.00±0.00 c | 21.20±1.07 b | 23.40±1.08 b | 39.80±1.20 a | 285.276 ** |
| Post-Oviposition period (in days) | 0.00±0.00 b | 37.20±1.02 a | 40.40±2.40 a | 38.00±0.95 a | 193.794 ** |
| No. of deposited eggs/female | 0.00±0.00 c | 56.60±5.86 b | 43.60±4.26 b | 152.00±6.44 a | 174.535 ** |
| Percentage of Egg hatchability | 0.00±0.0 d | 45.20±6.13 b | 24.00±3.67 c | 95.00±1.41 a | 123.269 ** |
| Longevity (in days) | 118.00±1.41 a | 93.20±2.60 b | 99.20±3.40 b | 98.40±1.47 b | 21.039 ** |
| Total mortality percentage | 52.40±1.29 a | 50.00±3.16 a | 51.60±1.17 a | 0.00±0.00 b | 202.695 ** |
| Malformation percentage | 51.60±0.93 a | 45.60±2.23 b | 50.40±0.98 a | 0.00±0.00 c | 360.991 ** |
| Life span (in days) | 186.60±2.73 a | 171.00±0.84 b | 183.20±4.72 a | 164.00±1.34 b | 13.790 ** |
| Longevity (in days) | 118.00±1.41 a | 93.20±2.60 b | 99.20±3.40 b | 98.40±1.47 b | 21.039 ** |
| Total mortality percentage | 52.40±1.29 a | 50.00±3.16 a | 51.60±1.17 a | 0.00±0.00 b | 202.695 ** |
| Malformation percentage | 51.60±0.93 a | 45.60±2.23 b | 50.40±0.98 a | 0.00±0.00 c | 360.991 ** |
| Life span (in days) | 186.60±2.73 a | 171.00±0.84 b | 183.20±4.72 a | 164.00±1.34 b | 13.790 ** |

**Legend:** **= Highly significant; *= Significant; Means followed with the same letter(s) within the horizontal column were not significantly different $p<0.05$.

*Histological changes in the alimentary canal*

The alimentary canal is a long straight tube running from the mouth to the anus, it fills most of the internal cavity of the grasshopper nymph, it is divided into the foregut, the midgut and the hindgut Fig. 1A. The foregut is considered to consist of four sections, the pharynx, the oesophagus, the crop and the proventriculus. The pharynx is the first part of the foregut and apart from being a tube that connect s the interior of the mouth area (Buccal cavity). The oesophagus is basically a tube leading to the midgut via the crop and the preventriculus. The Crop is simply a storage area, and the proventriculus is a muscular extension of the crop. This organ contains tooth-like denticles (known as the intima) that grind and pulverize food particles in addition to protect the foregut tissues from abrasion by food particles. The hard denticles inside the proventriculus are made from the chitin materials same of the external integument of the insects. Next is the stomach to which are attached six double finger–shaped digestive glands, the gastric carecae pouch–like structure arranged in a ring–like manner around the anterior end of the midgut Fig. 1A. The anterior lobe of each pair of the caecae extended over the crop while the posterior lobe extended over the midgut, gastric caecae produce enzymes that are secreted into the stomach to aid digestion. The gastric caeca serve to increase the surface area of the midgut, thus increase both its ability to secrete digestive enzymes and its ability to extract useful products from the partially digestive food. The useful proteins, vitamins and fats that are released by the digestive process pass across the wall of the midgut into the body cavity. The midgut runs from the gastric caeca to the Malpigahian tubules, a series of long thin tubes, in between these two the stomach or ventriculus or midgut which is the area of most active digestion (CHAPMAN, 1985). The digested food absorbed through the midgut tissue into the surrounding haemocoel then diffused to the different body parts. The hindgut comprises the intestine which compresses the undigested food and waste products, extracts more water before it is passed through the anus as faeces. The hindgut is lined by intima like the foregut. The digestive tract continues as the intestine, a thin tube without accessory structures. It leads to the short rectum which opens to the exterior via the anus. The hair–like tubules lying over the intestine are malpighian tubules, the excretory organs Fig. 1B.

Histological examination for the alimentary canal of the treated nymphs that remained after feeding on diet treated with the $LC_{50}$ of the most effective essential oil (Garlic) were recorded. After desiccation the digestive tract became flabby, softness were observed, swollen in the gastric caeca and preventriculus, elongation of the midgut Fig. 2A&B.

Histopathological changes that observed through the alimentary canal were as follows: in the crop tissues were separation of the muscular layer, disorganization of the cells were observed and partial rupture in the intima that lined the lumen Fig. 10, 13 and 14 comparing with the control section Fig 8 and 9.

The midgut of the control last instar nymphs of *H. littoralis* consists of a unicellular layer (epithelium) resting upon a basement membrane. This membrane is surrounded externally by circular and then by longitudinal muscle fibers. The epithelium consists of columnar and goblet cells with clusters of small regenerative cells each of which contains a relatively large nucleus and strongly basophilic cytoplasm Fig. 3 and 4. The epithelium is, also, protected from the food particles by a detached sheath, peritrophic membrane, surrounding lumen Fig. 4. Also as seen in Fig. 4 is the control (normal) midgut appears with luminal surface of the epithelium which is provided with a striated border constituting long microvilli Fig. 4. Such microvilli protect inwards into the lumen to increase the absorption surface of the cells, as well as, the spaces between them act as a kind of sieve. The histopathological effects of Garlic oil on the epithelium of the treated nymphs of *H. littoralis* are demonstrated in Fig. 5 and 6). The microscopically studies show a destruction of the cell vacuolization

and a rupture of the cell wall, some signs of morbidity in both nuclei and cytoplasm of the epithelial cells, microvilli of the columnar cells were curled and ruptured, vacuolated cytoplasm with large area of necrosis Fig 6. Elongation of the columnar and goblet cells was observed in Fig 7, rupture of pritrophic membrane Fig 5, 6 and 7). CLARKE *et al*. (1977) mentioned that diflubenzuron blocked the production of peritrophic membrane in *Locusta migratoria*. TAPPOZADA *et al*. (1968) investigated the histological and cytological changes in the mid gut of *Spodoptera littoralis* by some insecticides such as elongation of the epithelial cells, fading of cell boundaries and degeneration of some cells, decamethrin, diflubenzuron and methomyl caused vacuolation, elongation and breakdown of the epithelium, separation and detachment of peritrophic membrane of the pink bollworm *Pectinophora gossypiella* (SAAD *et al*., 1985). Large vacuoles were observed in the epithelial layer of treated *chironomid* midges larvae (*Chironomous decorus* and *Tanypus gradhaus*) fed on diflubenzuron and triflumuron (PELSU, 1985). Similarly, in adult *Tribolium castaneum* fed on triflumuron, epithelium vacuoles were observed in the midgut (PARWEEN, 1997). The vacuolation, elongation and disintegration of the epithelial cells, as well as the disappearance of muscular and regenerative cells, detachment of the basement membranes, had been observed in the mid gut of *S. exigua* by the action of diflubenzuron, malathion and cypermethrin (YOUNES *et al*., 2000). Many investigations have been conducted on the antifeedant effects, growth inhibition and abnormal development in various insects caused by using natural insecticides including *Boxus chinensis* oil and precoceneII (NASSAR, 1995; BREAM *et al*., 2001). In the present studies we could postulate that Garlic oil may affect the neuroendocrine system of insects which affect the production of ecdysone and consequently disturb the molting process and finally cause insect death as mentioned by GUJAR & MEHROTRA (1983); MOSTAFA *et al.* (1995). NASIRUDDIN & MORDUE (1993) studied the histological and ultrastructure changes caused by azadirachtin on the midgut of locust. His findings revealed necrosis of epithelial cells, enlargement of cytoplasmic inclusions and small sized striated borders. Various aspects of histopathology in the midgut of locust *Heteracris arnulosa* caused by neem seed extract were discussed by NAQVI *et al. (*1994). ZUDAIRE *et al.* (1998) cleared that, the gut of locusts disturbed and showed disrupter in the endocrine cells as affected by food nutrient content, insect age and stage.

The gastric caeca presents notable morphological swollen Fig.2A, and histological modification Fig. 10, 12 and 13, there were elongation, hypertrophy and hyperplasia clearly observed in Fig 12. comparing to the control section Fig.11, there dilation showing increasing in to lumen surface accompanied by epithelial bursting and fall height of the epithelial folds. This modification would be probably due to the destruction occurred in the muscular layer surrounding the caeca. For changes on the hindgut a partial loss of cellular integrates which reaches even the muscular bases leading to loss of integrity of the basement membrane of the cells.

In the hindgut there were partial changes was observed including the intima layer that lined the cells was affected to some extents, a partial abortion was occur-ing Fig. 16 also degeneration of small number of their cell was noticed comparing with the control Fig. 15. These observations are to be brought closer the observation on *Schisticerca* reported by ARMAH *et al.* (1999).

The untreated nymph show a visceral fat body made up of cellular clusters which often take the form of cellular cords Fig. 17. The cells appear rectangular with a core and cytoplasm filled with many lipid droplets (not colored vacuoles) that noticed in Fig. 17. The treated fat cells became smaller and more colored, became less provided with a significant fall of the number of the fat cells Fig. 18. This tissue loss is often accompanied by cellular exudates. The change of the size is probably due to a change of permeability causing a loss of the cellular content. CHAIEB *et al.* (2007) mentioned that the destruction

of the cellular structure is probably due to the membrane destabilization followed by the bursting of the membrane and the discharge of the cellular contents. The same author revealed a cytotoxic effect of *Certum perqui* saponins on the fat body of *S. littoralis* larvae, the cells of this tissue decrease in size and became more colored by loss of their cytoplasmic content. They also noted a cell destruction of the foregut and the gastric caeca of *Schisticerca*, they concluded that saponins interact with membrane cholesterol, this causes a membrane destabilization and that provokes cell death. Finally the histological changes that observed and recorded in this studies revealed disturbances in the foregut, midgut, hindgut and gastric caeca caused disturbances in the metabolic process of digested food and decrease the distribution of the nutritive metabolites materials into the haenocoel and effect on the stored lipid contents in the fat bodies which recognized as a powerful of energy for all the physiological properties of the insects, accordingly metamorphosis, development and reproduction of the insects was greatly affected. Thus, it could be concluded that essential oil of Garlic plant and may be the other tested oils Eucalyptus and Mint oils could be used as an effective natural products to be included in the integrated pest management program of *H.littoralis* grasshopper and other pests as well.


**Fig. 1.** Untreated alimentary canal in normal nymph of *H. littoralis* (1A - anterior part and 1B - posterior part). **Fig. 2.** Treated alimentary canal (2A and 2B). Crop (co) white arrow

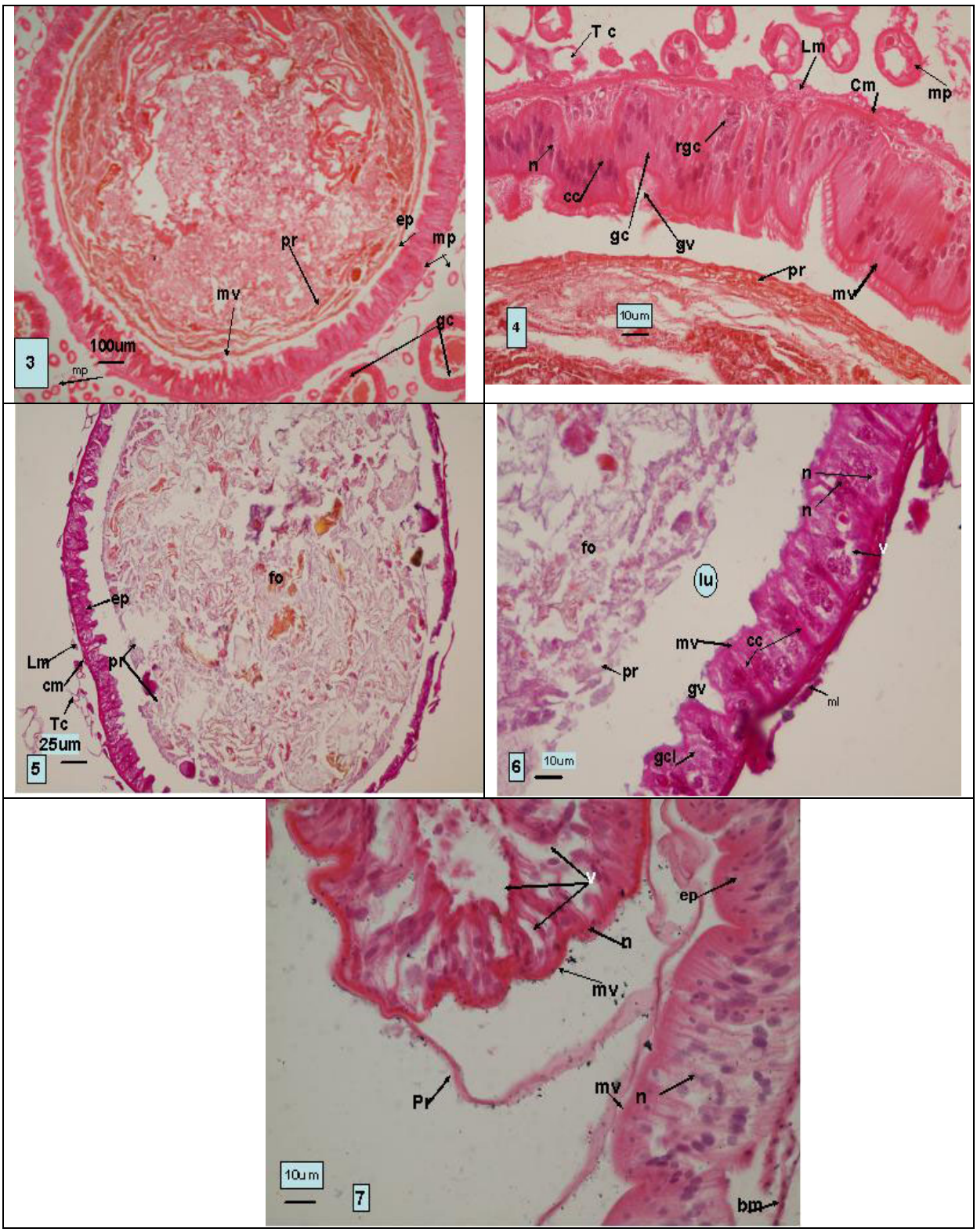


**Fig. 3-7:** Transverse section (T.S); Fig. 3. Normal midgut; Fig. 4. Magnified portion from Fig. 3; Fig. 5. Treated mid gut; Fig. 6 and 7. Magnified portion from Fig. 5. showing hypertrophy and hyperplasia of the epithelial cells of the treated midgut.

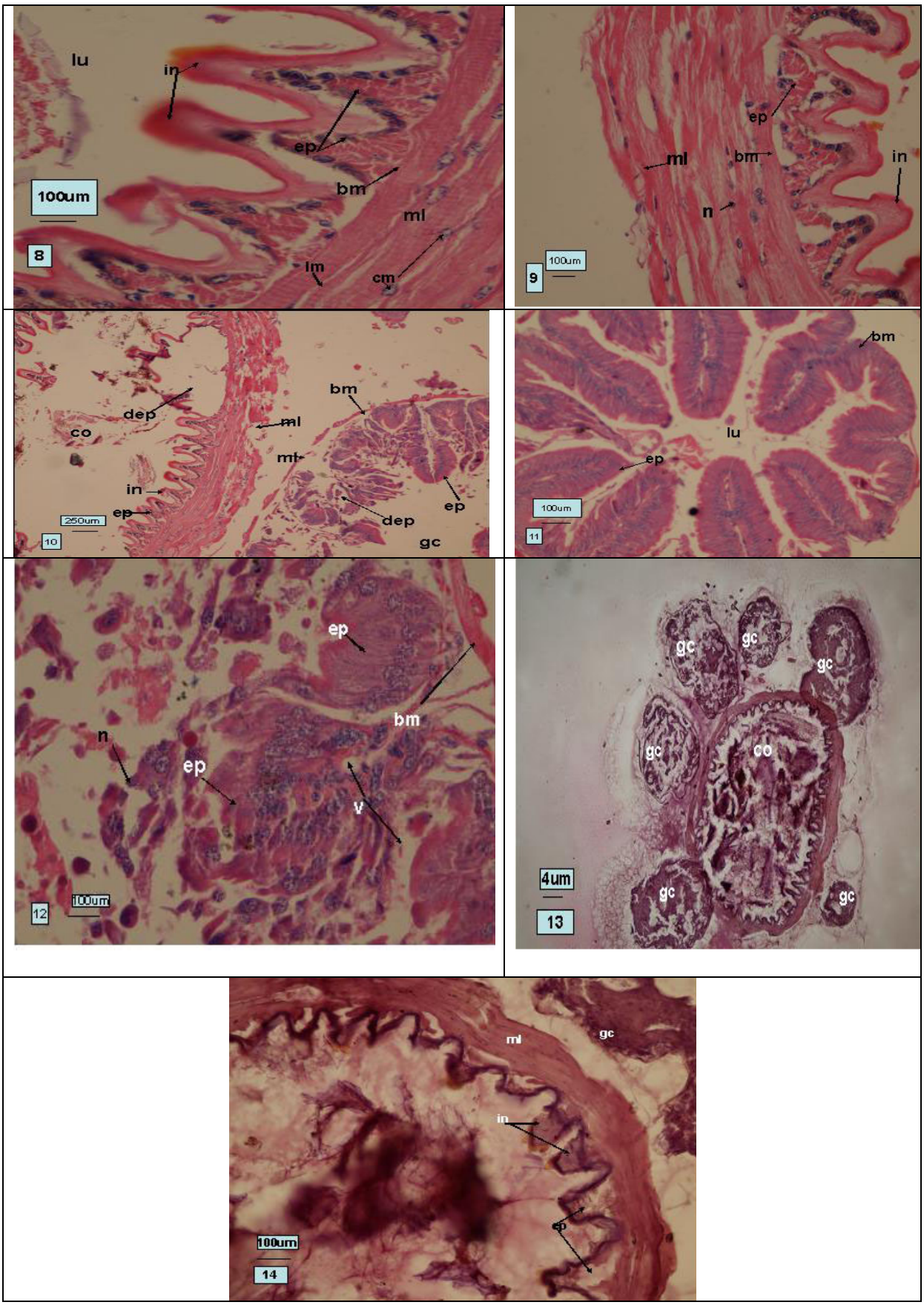


**Fig. 8-14:** Fig. 8 and 9. Normal crop; Fig. 11. Normal gastric caecae; Fig. 10, 13 and 14. Treated crop; Fig. 10 and 13. Treated crop and gastric caecae; Fig. 12 and 13. Treated gastric caecae.

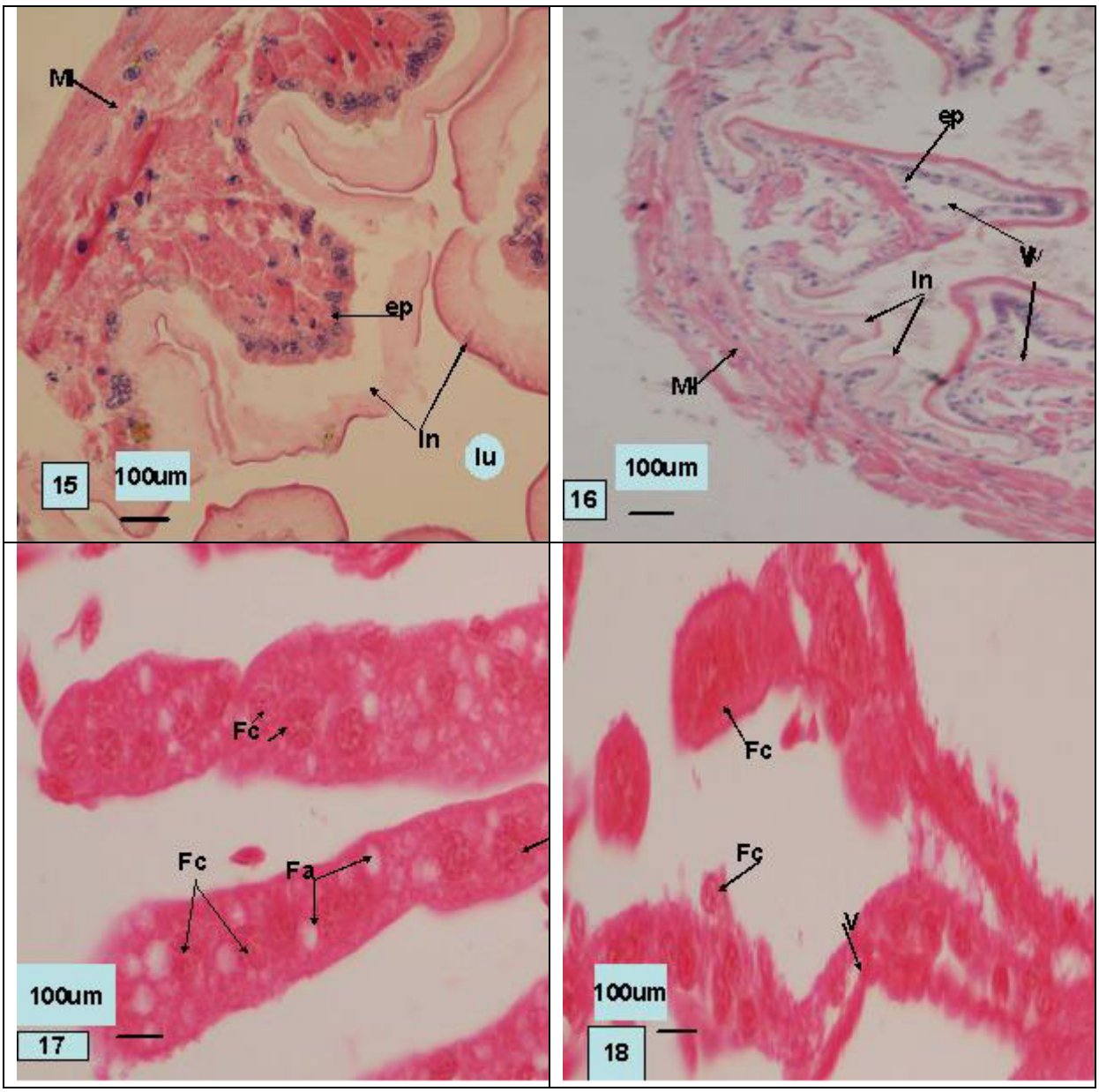


**Fig. 15-18:** Fig 15. Normal intestine; Fig. 16. Treated intestine; Fig. 17. Normal fat body; Fig. 18. Treated fat body.

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