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Effectiveness of *Nigella sativa* (Ranunculaceae) seed extracts on the transaminase activity in certain tissues of *Schistocerca gregaria* (Orthoptera: Acrididae)

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Abstract
The present study aimed to investigate the disturbing effects of sublethal concentrations (15.0% and 7.5%) of methanol, petroleum ether and n-butanol extracts of *Nigella sativa* seeds on the Glutamic oxaloacetic transaminase (GOT) and Glutamic pyruvic transaminase (GPT) activities in haemolymph and fat bodies of last instar nymphs and newly emerged adult females of the desert locust *Schistocerca gregaria* which represents a dangerous pest for crops and natural pastures over almost 65 countries. In no certain trend, a predominant enhancing effect of extracts was unexceptionally exhibited on GOT activity in haemolymph along the last nymphal instar and in haemolymph of adults. GOT activity was remarkably disturbed in fat bodies of nymphs since it was induced or reduced, depending on the extract. With regard to fat bodies of adults, methanol and n-butanol extracts exerted contradictory actions, depending on concentration. Prevalent inducing effect of *N. sativa* seed extracts had been exhibited on GPT activity in haemolymph of nymphs, regardless to the extract. On the contrary, a general reducing effect on GPT activity was detected, with few exceptions, in haemolymph of adults. Concerning the fat bodies of nymphs, GPT activity was enhanced in early-aged nymphs but inhibited in mid-aged congeners, irrespective of the extract. In late-aged ones, petroleum ether and n-butanol extracts exerted reducing actions on the enzyme activity. In fat bodies of adults, methanol extract exhibited an extended inducing effect on GPT activity but other extracts exhibited reducing effects of it.

Keywords: n-butanol, methanol, petroleum ether, nymph, adult, GOT, GPT

1. Introduction
The desert locust (*Schistocerca gregaria*) represents a major threat for crops and natural pastures over almost 65 countries from northern Africa to the Arabian peninsula and India. In other words, it ravages crops and natural pastures in 20% of the surface area of the earth [1]. The capacity of migration enables it to adapt to different ecological situations [2-4]. The importance of this insect stems from the fact that under plague condition each locust can eat its weight/day, approximately 2g [5]. Under plague conditions, this pest generates a great socio-political pressure that draw out emergency responses at national and international levels [6]. The concern over the environmental risk of chemical control of locust has increased recently. It arose primarily from heavy reliance on chemical pesticides which pose many risks on the ecosystems and human health [6]. The use of chemical pesticides is the main insect controlling approach against various insect pests, including locusts, during the late decades. The overdoing and indiscriminate have resulted in significant drawbacks, such as the development of strain resistance to insecticides [7, 8], handling hazards, increased costs, insecticide residues, great threats to non-target fauna, and both human and environmental health [9-11]. These problems have necessitated the need to explore and develop alternative strategies using eco-friendly and biodegradable plant products. In other words, alternative methods to chemical pesticides for controlling *S. gregaria* could have considerable value.
Because the desert locust refuses completely or partially to eat some plants species [12, 13]. Plant extracts represent an alternative agent for pest control since different studies had shown their actions against many insect pests as toxicants, repellents, antifeedants, deterrents of oviposition, growth regulators with low pollution and quick degradation in the environment [14-24]. The possibility of using plant extracts and plant secondary metabolites against insect pests in general and against the desert locust in particular has generated a lot of work [25-41].
Nigella plants are widely distributed in countries which border the Mediterranean Sea, central Europe and western Asia [42]. There are many species classified in the genus *Nigella* (Ranunculaceae) [43, 44]. Among the most important medicinal crops in Egypt is *Nigella sativa* which is commonly called as known as black seed or black cumin [45] and "Habbat al-barakah" (the seed of blessing) in Arabic. Several constituents had been identified and isolated from *N. sativa* seeds, such as conjugated linoleic acid, thymoquinone, nigellone (dithymoquinone), melanthin, nigilline, damascenine, tannins, flavonoids, saponins, alkaloids, proteins, lipids, dithymoquinone carvacol and anethole 4-terpinole, carbohydrates, crude fiber, saponins, ash, fixed oils and essential oil [46-52]. Recently, Shokri [53] identified the major components of the essential oil as thymoquinone, p-cymene, trans-anethole, 2-methyl-5(1-methyl ethyl)-Bicyclo [3.1.0] hex-2-en and γ-terpinene.
Seeds of *N. sativa* and their oil have a long history of folklore usage in various systems of medicines. Different medicinal, pharmacological, traditional values and folk remedies of this herb had been reported [49, 51-53]. For pest control, Deshpande *et al*. [54] reported that oleic and linoleic acid as insecticidal components from *N. sativa* which were found to be toxic to *Callosobruchus chinensis* and similar results were obtained [55, 56]. *N. sativa* extracts exhibited toxic effects on *Spodoptera littoralis* [57] and *S. gregaria* [24]. They disrupted the growth, development and metamorphosis of the latter insect [24]. Also, Ahmad *et al*. [58] studied the insecticidal activity of extracts from

this herb against the larvae of *Trogoderma granarium* under laboratory conditions. Khan *et al.* [59] reported the disturbing effects of the acetone seed extract of *N. sativa* on biology and invasion of the stored product pest *Tribolium castaneum*. *N. sativa* extracts showed the highest fumigant mortality against *Tribolium castaneum* [60]. Considerably toxic effects of *N. sativa* had been recorded against *Spodoptera littoralis* [61]. Recently, essential oil of *N. sativa* exhibited a moderate toxic effect on the scale insects *Ceroplastes rusci* and *Asterolcanium pustolans* [62]. *N. sativa* extracts exhibited disruptive effects on the adult performance [39], reproductive potential [40] and haemogram [41] of *S. gregaria*

Transamination has been demonstrated in a number of insect tissues, particularly that concerning glutamate, aspartate and alanine [63]. Glutamic oxaloacetic transaminase (=Glutmate Oxaloacetate Transaminase, GOT) has the official name: aspartate aminotransferase (AST or AsAT) and Glutamic pyruvic transaminase (=Glutamic Pyruvate Transaminase, GPT) has the official name: alanine aminotransferase, ALT or ALAT). These are key enzymes in the formation of non-essential amino acids, in metabolism of nitrogen waste, gluconeogenesis and correlated with protein anabolism and catabolism [64]. Moreover, transaminases, especially GPT, acts as a catalytic agent in carbohydrates metabolism [65].

In insects, the use of haemolymph as a medium for controlling insect pests has been made because the changes occurring in the haemolymph are quickly transferred to other portions of insect's body [66]. The exposure of an organism to xenobiotic product can modify the synthesis of certain metabolite and disturb the functionality of the organisms [68]. On the other hand, the fat body in insect is the main site for protein synthesis as well as the intermediating metabolism of amino acids, which are utilized for the production of hormones and enzymes and the composition of protein in the body as a whole may be greatly modified [68]. The current work was carried out to evaluate the effects of different extracts of *N. sativa* seeds on the transaminase activities in haemolymph and fat bodies of *S. gregaria* nymphs and newly emerged adult females.

2. Materials and Methods

2.1 Experimental Insect

The desert locust *Schistocerca gregaria* (Forskal) (Orthoptera: Acrididae) was used as an experimental insect in the present study. The present culture was originated by a sample of gregarious nymphs from Plant Protection Research Institute, Ministry of Agriculture, Giza. As designed by Hunter-Jones [69] and improved by Ghoneim *et al*. [70], insects were reared in wood formed cages (60 x 60 x 70 cm). The bottom was furnished with a sandy layer (20 cm depth) and provided with10-15% humidity to be suitable for egg laying. An electric bulb (100 watt) was adjusted to maintain a continuous photoperiod (12 L: 12 D) in each cage as well as in order to maintain an ambient temperature (32±2ºC). The insects were reared and handled under the crowded conditions. Fresh clean leaves of clover *Trifolium alexandrinum* were provided, as a food for insects, every day.

2.2 Plant Extraction

Samples of *Nigella sativa* seeds were purchased from an Egyptian market. The samples were air-dried, powdered and kept in tightly closed amber coloured glass containers for protecting from light, at low temperature. Dried and pulverized powder of *N. sativa* (2 kg) was exhaustively separately extracted with methanol (1.7 Lx3). The combined alcohol extracts were concentrated to 400 ml, diluted with 400 ml of water and the next successively extracted with petroleum ether (5x400 ml) was concentrated to dryness under reduced pressure giving (11 and 90 g). The n-butanol (5x400 ml) extracts were concentrated to dryness under reduced pressure giving (75 and 55 g).

2.3 Nymphal Treatments

According to Hamadah *et al.* [24], 15.0% and 7.5% were the sublethal concentrations of methanol, petroleum ether, and n-butanol extracts derived from *N. sativa* seeds. After treatment of the newly moulted penultimate (4th) instar nymphs of *S. gregaria* through the fresh food leaves of *T. alexandrinum* dipped once in each extract for 3 minutes, the successfully moulted last instar nymphs and emerged adult females were undergone to determine the influenced Glutamic oxaloacetic transaminase (GOT) and Glutamic pyruvic transaminase (GPT) activities in two tissues: haemolymph and fat body. Three ages of last instar nymphs were only used: early- (1-day old), mid- (4-day old) and late-aged (7-day old) nymphs.

2.4 Tissue Sampling

For the determination of phosphatase activity in the haemolymph, it was collected from last instar nymphs and newly emerged adult females. Haemolymph was drawn into Eppendorff Pipetman containing few milligrams of phenoloxidase inhibitor (phenylthiourea) to prevent tanning or darkening and then diluted 5× with saline solution 0.7%. For whole blood assays, the diluted haemolymph was frozen for 20s to rupture the haemocytes. The haemolymph samples were then centrifuged at 2000 r.p.m. for 5 min, and only the supernatant fractions were used for assay directly or frozen until use. Three replicates were used and the haemolymph of two individuals were never mixed. For the determination of phosphatase activity in the fat body, samples were collected from last instar nymphs (of the same ages) and newly emerged adults. The fat body samples were weighed and then homogenized in a saline solution (the fat body of one insect /1 ml saline solution 0.7 %) using a fine electric homogenizer, tissue grinder for 2 min. Homogenates were centrifuged at 4000 r.p.m. for 15 min. The supernatant was used directly or frozen until the use for the enzymatic determination. Three replicates were used and the fat bodies from two individuals were avoided to be mixed.

2.5 Determination of Transaminase Activities

GOT and GPT activities were determined in the nymphal and adult tissues according to the method of Harold [71] using a kit of Bioadwic. The enzyme was measured at wave length 546 nm by spectrophotometer.

2.6 Statistical Analysis

Data obtained were analyzed by the Student's *t*-distribution, and refined by Bessel correction [72] for the test significance of difference between means.

3. Results

After treatment of the penultimate instar nymphs of *S. gregaria* with 15.0 or 7.5% concentration of methanol, petroleum ether or n-butanol extract of *N. sativa* seeds, glutamate oxaloacetate transaminase (GOT) and glutamate pyruvic transaminase (GPT) activities had been determined in haemolymph and fat bodies of the early- (1-day old), mid- (4-day old) and late-aged (7-day old)

last instar nymphs as well as in the same tissues of newly emerged adult females.

3.1 Effects of *N. sativa* Seed Extracts on GOT Activity

A predominant enhancing effect of *N. sativa* seed extracts was unexceptionally exhibited on GOT activity in haemolymph along the last nymphal instar and newly emerged adults. As obviously shown in Table (1), no certain trend was attained for the enhancing effects of extracts. For some detail, at the high concentration of methanol extract, a curved-shape was observed for GOT level in haemolymph along the nymphal instar while a steady sloop curve-trend of activity was detected at low concentration. However, the most potent enhancing action on the enzyme activity was exerted by n-butanol extract, at high concentration, in the haemolymph of late-aged nymphs (233.3±36.2, compared to 40.0±8.7 U/ml of controls). The least inducing action on the enzyme activity (Increment %: 25.9) was exerted in mid-aged nymphs by the same extract at low concentration. With regard to the newly emerged adults, both the strongest and least promoting effects were exhibited after treatment of nymphs with n-butanol extract (245.0±45.8 and 108.3±11.5 U/ml at high and low concentrations, respectively, *vs*. 85.0 ±17.3 U/ml of controls congeners).

According to data assorted in Table (2), GOT activity was remarkably disturbed in fat bodies of nymphs, because it was induced or reduced, depending on the extract. For some detail, the enzyme level considerably elevated in nymphs, regardless to their age, by petroleum ether extract (Increment %s: 296.6 and 193.7 in early-aged nymphs, 18.0 and 41.2 in mid-aged nymphs, 37.5 and 17.0 in late-aged nymphs, at high and low concentrations, respectively). Furthermore, the enzyme activity was promoted in adults by the same extract.

In contrast, inhibitory effects on the GOT activity in fat bodies had been exhibited by methanol and n-butanol extracts. The highest reduction % in the enzyme activity was calculated as 54.6 in late-aged nymphs, at low concentration of n-butanol extract but lowest reduction % as 0.1, at the low concentration of methanol extract. Contradictory effects of methanol and n-butanol extracts had been exhibited on the enzyme activity in fat bodies of adults, depending on the concentration. As for example, the enzyme activity was induced (Increment%: 46.1) at high concentration of methanol extract but declined (Reduction%: 1.0) at its low concentration.

3.2 Effects of *N. sativa* Seed Extracts on GPT Activity

In the light of data arranged in Table (3), prevalent inducing effects of *N. sativa* seed extracts had been exhibited on GPT activity in haemolymph of nymphs, regardless to the extract. However, the strongest promoting effect was exhibited on the enzyme activity in haemolymph of late-aged nymphs (Increment %: 421.7) at high concentration of n-butanol extract. In respect of the adults, a general reducing effect on GPT activity was detected in haemolymph, regardless to the extract with an exception of failure to affect it at low concentration of n-butanol extract.

Data distributed in Table (4) clearly show a predominant inducing effect on GPT activity in fat bodies of the early-aged nymphs but reducing effect in mid-aged nymphs, regardless to the extract. Concerning the fat bodies of late-aged nymphs, methanol extract exerted a promoting action on GPT activity (Increment %s: 35.6 and 40.8, at high and low concentrations, respectively) but petroleum ether extract (Reduction %s: 2.7 and 3.2, at high and low concentrations, respectively) and n-butanol extract (Reduction %s: 11.4 and 7.0, at high and low concentrations, respectively) exhibited reducing effects on it. With regard to the fat bodies of adults, methanol extract exhibited an extended inducing effect on the enzyme activity (Increment %s: 30.6 and 29.3, at high and low concentrations, respectively). On the other hand, other extracts exhibited reducing effects on the enzyme activity with a maximum (Reduction %:-15.4) at low concentration of n-butanol extract, and a minimum (Reduction %: 1.5) at low concentration of petroleum ether extract (Table 4).

Table 1: Effects of *N. sativa* seed extracts on GOT activity (U/ml) in haemolymph of *S gregaria*.

Solvent	Conc. %		Last instar nymphs			Newly emerged adults
			Early-aged	Mid-aged	Late-aged	
Methanol	15.0	Mean ± SD	163.3 ± 14.4 d	206.7 ± 27.5 c	135.0 ± 20.0 c	206.7 ± 27.5 c
		Change %	+117.3	+113.8	+237.5	+143.2
	7.5	Mean ± SD	223.3 ± 31.8 c	171.7 ± 14.4 c	53.3 ± 34.0 a	188.3 ± 14.4 c
		Change %	+197.7	+77.6	+33.3	+121.5
Petroleum ether	15.0	Mean ± SD	156.7 ± 22.5 c	163.3 ± 14.4 c	141.7 ± 11.5 d	198.3 ± 31.8 c
		Change %	+108.9	+68.9	+254.3	+133.3
	7.5	Mean ± SD	216.7 ± 31.8 c	135.0 ± 20.0 a	150.0 ± 22.8 d	225.0 ± 40.9 c
		Change %	+188.9	+39.6	+275.0	+164.7
n-butanol	15.0	Mean ± SD	206.7 ± 27.5 c	163.3 ± 14.4 c	233.3 ± 36.2 d	245.0 ± 45.8 c
		Change %	+175.6	+68.9	+483.3	+188.2
	7.5	Mean ± SD	101.7 ± 11.5 b	121.7 ± 11.5 a	101.7 ± 11.5 c	108.3 ± 11.5 a
		Change %	+35.6	+25.9	+154.3	+27.4
Controls		Mean ± SD	75.0 ± 8.7	96.7 ± 17.6	40.0 ± 8.7	85.0 ± 17.3

Conc.: Concentration level; mean ± SD followed with the letter (a): not significantly different ($P>0.05$), (b): significantly different ($P<0.05$), (c): highly significantly different ($P<0.01$), (d): very highly significantly different ($P<0.001$).

Table 2: Effects of *N. sativa* seed extracts on GOT activity (U/ml) in fat bodies of *S gregaria*.

Solvent	Conc. %		Last instar nymphs			Newly emerged adults
			Early-aged	Mid-aged	Late-aged	
Methanol	15.0	Mean ± SD	310.2 ± 5.9 d	244.7 ± 5.1 d	173.0 ± 5.8 a	334.7 ± 6.6 d
		Change %	-24.2	-15.7	-7.8	+46.1
	7.5	Mean ± SD	313.8 ± 12.3 c	211.9 ± 6.7 d	187.4 ± 4.4 a	226.8 ± 7.4 a
		Change %	-23.4	-27.0	-0.1	-1.0
Petroleum ether	15.0	Mean ± SD	1624.1 ± 36.1 d	342.6 ± 5.4 d	258.0 ± 9.4 d	306.2 ± 8.2 d
		Change %	+296.6	+18.0	+37.5	+33.7
	7.5	Mean ± SD	1202.5 ± 24.0 d	409.9 ± 7.8 d	219.5 ± 6.5 c	392.7 ± 7.2 d
		Change %	+193.7	+41.2	+17.0	+71.4
n-butanol	15.0	Mean ± SD	263.9 ± 13.3 d	270.1 ± 11.6 b	169.2 ± 5.6 b	228.1 ± 6.9 a
		Change %	-35.6	-7.0	-9.8	-0.4
	7.5	Mean ± SD	289.9 ± 5.8 d	262.4 ± 7.4 b	85.2 ± 6.3 d	229.8 ± 11.8 a
		Change %	-29.2	-9.6	-54.6	+0.3
Controls		Mean ± SD	409.5 ± 17.4	290.3 ± 7.5	187.6 ± 7.7	229.1 ± 5.5

Conc., a, b, c, d: See footnote of Table 1

Table 3: Effects of *N. sativa* seed extracts on GPT activity (U/ml) in haemolymph of *S. gregaria*.

Solvent	Conc. %		Last instar nymphs			Newly emerged adults
			Early-aged	Mid-aged	Late-aged	
Methanol	15.0	Mean ± SD	61.7 ± 7.6 c	46.7 ± 7.6 c	18.3 ± 2.9 a	58.3 ± 2.9 d
		Change %	+184.3	+179.6	+120.5	-43.6
	7.5	Mean ± SD	75.0 ± 8.7 c	18.3 ± 2.9 a	8.3 ± 5.8 a	53.3 ± 7.6 d
		Change %	+245.6	+9.6	0.0	-48.4
Petroleum ether	15.0	Mean ± SD	58.3 ± 2.9 c	51.7 ± 5.8 d	26.7 ± 5.8 b	63.3 ± 5.8 c
		Change %	+168.7	+209.6	+221.7	-38.7
	7.5	Mean ± SD	51.7 ± 5.8 c	38.3 ± 7.6 b	21.7 ± 7.6 a	56.7 ± 2.9 d
		Change %	+138.2	+129.3	+161.4	-45.1
n-butanol	15.0	Mean ± SD	43.3 ± 2.9 b	71.7 ± 10.6 d	43.3 ± 2.9 d	46.7 ± 7.6 d
		Change %	+99.5	+329.3	+421.7	-54.8
	7.5	Mean ± SD	26.7 ± 11.5 a	20.0 ± 8.7 a	10.0 ± 8.7 a	103.3 ± 5.8 a
		Change %	+23.0	+19.8	+20.5	0.0
Controls		Mean ± SD	21.7 ± 7.6	16.7 ± 2.9	8.3 ± 5.8	103.3 ± 5.8

Conc., a, b, c, d: See footnote of Table 1

Table 4: Effects of *N. sativa* seed extracts on GPT activity (U/ml) in fat bodies of *S. gregaria*

Solvent	Conc. %		Last instar nymphs			Newly emerged adults
			Early-aged	Mid-aged	Late-aged	
Methanol	15.0	Mean ± SD	124.8 ± 7.8 a	118.3 ± 5.6 d	222.2 ± 5.0 d	279.5 ± 2.9 d
		Change %	+1.8	-41.3	+35.6	+30.6
	7.5	Mean ± SD	126.0 ± 5.1 a	87.1 ± 6.4 d	230.8 ± 3.8 d	276.8 ± 6.2 d
		Change %	+2.8	-56.8	+40.8	+29.3
Petroleum ether	15.0	Mean ± SD	188.9 ± 30.9 b	167.4 ± 8.1 c	159.5 ± 5.3 a	197.1 ± 6.0 d
		Change %	+54.1	-17.0	-2.7	-7.9
	7.5	Mean ± SD	330.9 ± 36.0 d	85.3 ± 10.2 d	158.7 ± 3.7 a	210.8 ± 6.1 a
		Change %	+169.9	-57.7	-3.2	-1.5
n-butanol	15.0	Mean ± SD	123.4 ± 9.3 a	159.0 ± 6.2 c	145.2 ± 3.2 c	185.0 ± 5.8 c
		Change %	+0.7	-21.2	-11.4	-13.6
	7.5	Mean ± SD	125.8 ± 4.9 a	62.9 ± 7.0 d	152.4 ± 8.0 a	181.0 ± 10.0 c
		Change %	+2.6	-68.8	-7.0	-15.4
Controls		Mean ± SD	122.6 ± 6.3	201.7 ± 6.4	163.9 ± 6.0	214.0 ± 7.2

Conc., a, b, c, d: See footnote of Table 1

4. Discussion

Transaminases (GOT= AST and GPT= ALT) help in the production of energy and serve as a strategic link between the carbohydrate and protein metabolism [73]. These are key enzymes in the formation of non-essential amino acids, in metabolism of nitrogen waste, gluconeogenesis and correlated with protein anabolism and catabolism [64]. It is of interest to mention that GOT and GPT activities are known to be altered during various physiological and pathological conditions in insect body such as microorganism infections, damage to some tissues or being a toxic material [74-76]. In other words, they may play an important role in the insecticidal poisoning [77, 78].

4.1 Disturbed GOT Activity in *S. gregaria* by *N. sativa* Seed Extracts

Contradictory results of disturbed GOT activity in several insects by various botanicals had been reported in the available literature. Enhancement or prohibition of the enzyme activity

usually depends not only on the insect species but also on its developmental stage, age, tissue, nature of the botanical and method of treatment [79-88].

In the present study, a predominant enhancing effects of *N. sativa* seed extracts (*viz*. methanol, petroleum ether and n-butanol extracts) were unexceptionally exhibited on GOT activity in haemolymph of last instar nymphs and newly emerged adults of *S. gregaria*, but in no certain trend. Also, GOT activity was remarkably disturbed in fat bodies of nymphs, depending on the extract. Petroleum ether extract exhibited a promoting effect on the enzyme activity in nymphs, regardless to the age, as well as in adults. These results are in agreement with some reported results of increased GOT activity in various insects by different plants extracts since Ghoneim *et al*. [89] recorded enhanced activity in haemolymph of last instar nymphs and in fat bodies of adults of *S. gregaria* by *Punica granatum* peel extracts. Also, methylene chloride extract of *Azadirachta indica* enhanced the enzyme activity in the same locust [90]. Sublethal doses of Basil essential oil *(Ocimum basilicum*) promoted the enzyme activity in *Sitophilus granarius* adults [91]. In addition to botanicals, increasing GOT activity was reported in *Spodoptera littoralis* after treatment with some insect growth regulators (IGRs) such as pyriproxyfen, flufenoxuron or chlorfluazuron [92]; flufenoxuron [83] and Novaluron, Cyromazine or Diofenolan [93]. In addition, increasing GOT activity was reported in *Musca domestica* by pyriproxyfen [94] and *Culex pipiens* by Cyromazine [95]. GOT activity was significantly induced after treatment of 4th instar larvae of *S. littoralis* with bioinsecticides (*viz*. emamectin benzoate, abamectin and spinosad) [96] and after injection of the 5th instar larvae of the same species with spores of *Beauveria bassiana* and *Metarhizium anisopliae* [97]. In the current work, enhanced GOT activity in haemolymph of last instar nymphs and adults of *S. gregaria* by *N. sativa* seed extracts, as well as in fat bodies as response to the action of petroleum ether extract suggests the mobilization of amino acids during the stress exerted by certain toxic components in these extracts to meet the energy demands [98] since several chemical constituents had been identified, such as thymoquinone, nigellone (dithymoquinone), melanthin, nigilline, damascenine, tannins, flavonoids, saponins, alkaloids, thymoquinone, p-cymene, trans-anethole, 2-methyl-5(1-methyl ethyl)-Bicyclo[3.1.0]hex-2-en and γ-terpinene and anethole 4-terpinole [46-50, 53, 99]. In addition, this increasing activity of GOT may be due to the occurrence of reversible binding between these extracts and enzymatic site of action on the enzyme surface as suggested by Megahed *et al*. [96] for some bioinsectcides against *S. littoralis*.

As reported in the available literature, GOT activity was inhibited in several insect pests by various botanicals, such as *Tribolium castaneum* by different extracts of *Curcuma longa* [100], *Rhyzopertha dominica* adults by hexane extract of *Capparis deciduas* [101], *Pieris rapae* by methanolic extract of *Silybium marianum* [102], *S. gregaria* fat bodies of nymphs and adults by *P. granatum* peel extracts [89], *T. castaneum* larvae by essential oils of *Wedelia trilobata* and *Melissa officinalis* [103] and by garlic oil (*Allium sativum*) [104]. In consistent with these reported results, GOT activity was remarkably disturbed in fat bodies of *S. gregaria* nymphs by the *N. sativa* seed extracts, in the present study, since it was induced or reduced, depending on the extract. Methanol and n-butanol extracts exhibited significant inhibitory effects on the enzyme activity in fat bodies of nymphs. Methanol and n-butanol extracts exerted contradictory actions on the enzyme activity in fat bodies of adults, depending on concentration. However, the reduced GOT activity in fat bodies of *S. gregaria* by some seed extracts of *N. sativa*, in the present investigation, can be attributed to difficulty in the formation of dissociable enzyme-inhibitor complexes which reduce the specific enzyme activity [105] or to a disturbance of the link between the carbohydrate and protein metabolism.

4.2 Disturbed GPT Activity in *S. gregaria* by *N. sativa* Seed Extracts

In the present study, a prevalent inducing effect of *N. sativa* seed extracts had been exhibited on GPT activity in haemolymph of nymphs of *S. gregaria*, regardless to the extract. Concerning the fat bodies, the enzyme activity was enhanced in the early-aged nymphs, irrespective of the extract. In respect of adults, methanol extract exhibited an inducing effect on GPT activity in fat bodies. These results are in accordance to those reported results of GPT increasing in certain tissues of some insects by extracts of different plant species, such as in fat bodies of the early-aged nymphs of *S. gregaria* by *P. granatum* peel extracts [89], in whole body homogenate of *S. gregaria* nymphs by methylene chloride extract of *A. indica* extract) [90] and in adults of *S. granarius* by sublethal doses of essential oils of *Eugenia aromatic* or *O. basilicum* [91]. In addition to botanicals, induced GPT activity in the present study agree, to some extent, with those reported results of GPT induction in various insect pests by some IGRs, such as *M. domestica* by pyriproxyfen [94], *Bactrocera zonata* by methoxyfenozide or lufenuron [106] and *C. pipiens* by Cyromazine [95] and *S. littoralis* by spores of *B. bassiana* and *M. anisopliae* [97] or by emamectin benzoate [107]. The enhanced activity of GPT in *S. gregaria* by *N. sativa* seeds extracts, in the present study, may be attributed to the occurrence of reversible binding between the tested extracts and enzymatic site of action on the enzyme surface. This may be due to the fact that the relationships between protein synthesis and transaminase levels were affected by the hormonal control of protein synthesis and neurosecretory hormones which involved in the regulation of transaminase levels [73].

The available literature, on the other hand, clearly reported GPT reduction in different insects by some botanicals. For some detail, suppressed enzyme activity in *T. castaneum* was observed after treatment with different extracts of *C. longa* [100], in *R. dominica* after treatment with hexane extract of *C. deciduas* **[101]**, in *P. rapae* after treatment with methanolic extract of *S. marianum* [102], in *Aedes aegypti* larvae after treatment with methanolic extract of *L. camara* [108], in haemolymph of mid-aged nymphs and in fat body of adults of *S. gregaria* after treatment with *P. granatum* peel extracts [89], in *T. castaneum* larvae after treatment with essential oils of *W. trilobata* and *M. officinalis* [103] or garlic oil (*A. sativum*) [104], in *Callosobruchus analis* after treatment with *Acorus calamus* (essential oil) or Biosal (Neem preparation) [109], etc.. In agreement with those results, GPT activity was inhibited in fat bodies of mid-aged nymphs of *S. gregaria*, in the present study, irrespective of the extract of *N. sativa* seeds. In fat bodies of late-aged congeners, petroleum ether and n-butanol extracts exerted reducing actions on the enzyme activity. In respect of adults, a general reducing effect on the enzyme activity was detected in the haemolymph, with few exceptions. In their fat bodies, petroleum ether and n-butanol extracts exhibited reducing effects on the enzyme activity. These observed cases of GPT inhibition, in the present work, may be attributed to the intervening of certain chemical

components of these extracts in the hormonal control of protein synthesis and neurosecretory hormones involved in the regulation of transaminase levels [110]. Several chemicals had been identified in *N. sativa* seeds [46-50, 99]. Certain chemicals are responsible for the inhibition of GPT activity but the exact mode of action of tested extracts on the transaminase regulation is still controversial until now!!

In general, the diverse effects of *N. sativa* seed extracts on GPT activity in certain tissues or stages of *S. gregaria*, in the present study, can be understood in the view of effect on synthesis or functional levels of this enzyme directly or indirectly by altering the cytomorphology of the cells [111], or the effect of certain effective components in these extracts on the neurosecretory hormonal pattern.

5. Conclusions

The transaminases form a link between the metabolism of amino acids, lipids and carbohydrates. Accordingly, the disturbance in GOT and GPT levels will be closely related to metabolism of proteins and amino acids. Thus, it will disrupt many physiological functions and ultimately lead to death, in other way control the pest. The disturbed activities of GOT and GPT in nymphs and adults of *S. gregaria* as responses to *N. sativa* seed extracts, in the present study, need to be fully understood in the view of results of further investigation upon the chemical constituents of these extracts, separately. However, the present *N. sativa* seed extracts can be used as a part of Integrated Pest Management Program against this dangerous locust.

6. References

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