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Efficacy of the toothpick weed *Ammi visnaga* L. (Apiaceae) fruit extracts on transaminase activity in certain tissues of *Schistocerca gregaria* (Forskal)(Orthoptera: Acrididae)

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Abstract
The present study aimed to investigate the disturbing effects of LC_{50} concentrations of ethanol, petroleum ether and n-butanol extracts (21.0, 12.0 and 22.5%, respectively) of *Ammi visnaga* fruits 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 dangerous desert locust *Schistocerca gregaria*. A predominant enhancing effect was exhibited on the GOT activity in haemolymph of nymphs, regardless to the extracts. In haemolymph of adults, GOT activity was induced or reduced depending on the extract. In fat bodies, n-butanol extract considerably prohibited the enzyme activity in both nymphs and adults. In contrast, petroleum ether extract enhanced the enzyme activity. The ethanol extract exhibited inducing or reducing effects on the enzyme activity, depending on the nymphal age but induced the enzyme activity in adults. GPT activity was reduced in haemolymph of both nymphs and adults by n-butanol extract. The ethanol extract, considerably enhanced it in nymphs but significantly prohibited it in adults. The petroleum ether extract exhibited an enhancing effect on the enzyme activity in haemolymph of nymphs and adults with few exceptions. GPT activity was elaborately induced or pronouncedly inhibited in fat bodies of nymphs, depending on the age and extract. In fat bodies of adults, GPT level was unexceptionally declined, irrespective of the extract.

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

1. Introduction
The desert locust, *Schistocerca gregaria* (Forskål), is characterized by a phase polymorphism (Uvarov, 1977) [1] enabling the transition from a solitary phase to an extremely dangerous gregarious one for the agricultural productions and pastures. *S. gregaria* is perhaps the most dramatic and potentially devastating species, and can devastate the cultures of a whole African continent (Lecoq and Mestre, 1988; Sanchez-Zapata *et al.*, 2007; Ammar *et al.*, 2009) [2, 4]. Most recent large-scale outbreaks of it occurred in 1986-1989 and in 2003-2005, mostly on the African continent (Latchininsky, 2013) [5]. In 2003-2005, to curtail the outbreak of this locust, 13 million ha were treated with broad-spectrum insecticides in 26 countries (Belaynech, 2005) [6].
Current locust control operations are mainly based on organophosphorus pesticides as a result of the banning of organochlorines (Lecoq, 2001) [7]. The widespread use of such synthetic pesticides has considerable drawbacks, such as the development of insect resistance to insecticides, increased costs, handling hazards, concerns about insecticide residues, and great threats to both human and environmental health (Garriga and Caballero, 2011) [8]. Therefore, many institutions have intensified their efforts in the search for integrated locust control measures. Much attention has been given to use plant extracts or plant constituents that have insecticidal effects (Schmutterer, 1990; Krall and Wilps, 1994) [9,10]. Although, hundreds of plant natural products have demonstrated deleterious effects on insects only a handful of botanical insecticides are currently approved for use in industrialized countries for several reasons (Isman, 1994, 2006) [11, 12].
Plant extracts, thus, 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 (Naqvi *et al.*, 1992; Bourguet *et al.*, 2000; Nakatani *et al.*, 2001; Farag, 2002; Schmidt and Assembe-Tsoungui, 2002; Sadek, 2003; Strand, 2008; Tavares *et al.*, 2009; Chermenskaya *et al.*, 2010; Vogelweith *et al.*, 2011; Lampert, 2012; Hamadah *et al.*, 2013) [13, 24]. Neem extracts, as for example, disturb the enzyme pattern in the insect body as reported by some researchers (Naqvi *et al.*, 1991; Hosseini-Naveh *et al.*, 2007) [25, 26]. The possibility of using plant extracts and plant secondary metabolites against the desert locust has generated a lot of work (Abbassi *et al.*, 2003a,b, 2004, 2005; Ould El Hadj *et al.*, 2006; Zouiten *et al.*, 2006; Idrissi and Hermes, 2008; Kemassi *et al.*, 2010; Abdellah *et al.*, 2013; Ghoneim, 2015; Ghoneim *et al.*, 2015 a,b,c, 2016) [27-40].
Ammi is a genus of 3-6 species of flowering plants in the family Apiaceae. *Ammi visnaga* Lamarck (Apiaceae = Umbelliferaceae) is native to Europe, Asia and North Africa but can be found throughout the world as an introduced species. The plant is endogenous to Egypt and other regions in the Middle East (Beltagy and Beltagy, 2015) [41]. Among Egyptian people, it is called "Khella" while in Europe the plant has often been referred to "Toothpick herb" or "Bishop's weed" (El-Fiky *et al.*, 1989) [42]. In the genus *Ammi*, coumarin and their derivatives, flavonoids, volatile oil and fixed oil are the major bioactive compounds of important biological activities (Abdul-Jalil *et al.*, 2010) [43]. The main chemical constituents of *A. visnaga* as couramins and furocumarins, the most important of which are khellin, visnagin, khellol, khellinol, and angular pyranocouramins including visnadin, samidin and dihydrosamidin (Ortel *et al.*, 1988) [44]. In fruits, khellin

represents 1% and visnagin represents 0.3% (Martelli *et al.*, 1984) [45]. Alqasoumi *et al.* (2014) [46] qualitatively and quantitatively analyzed khellin, as the major component in fruits, for the commercial formulations. Recently, Beltagy and Beltagy (2015) [41] isolated and described the structure of khellin and visnagin in fruits. Flavonoids have been, also, reported (Bencheraiet *et al.*, 2011) [47]. The essential oil of *A. visnaga*, growing in Morocco, has been reported to contain linalool and aliphatic esters as the main components (Lamiri *et al.*, 2001a) [48]. Khalfallah *et al.* **(**2011) [49] identified different percentages of Isobutyrate, 2,2-dimethylbutanoic acid, croweacin and linalool as the main constituents of *A. visnaga*. Furthermore, Gas-liquid chromatographic analysis of the essential oil revealed the major components: isobutyl isobutyrate, linalool, thymol, fenchyl acetate, bornyl acetate, α-isophorone, 2,2-dimethyl butanoic acid and croweacin (Talaat *et al.*, 2014) [50]. Moreover, the unused parts of this plant were found to contain several compounds, such as β-sitosterol, visnadine, khellin, norkhellol, khellol, rhamnazin and cimifugin (Ashour *et al.*, 2013) [51].

A. visnaga is one of the most important medicinal plant species in the world. Its fruits have been used in folk medicine in Egypt many years ago to relief kidney stones by drinking teas of powdered fruits (Franchi *et al.*, 1985; Gunaydin and Beyazit, 2004) [52, 53]. As pointed out by several authors (Beltagy and Beltagy, 2015; Khan *et al.*, 2001; Jouad *et al.*, 2002; Cordero *et al.*, 2004; Whitton *et al.*, 2008; Lee *et al.*, 2010; Kwon *et al.*, 2010; Vanachayangkul *et al.*, 2010; Hilmi, 2013; Sabry *et al.*, 2014; Jan, 2014; Bhagavathula *et al.*, 2015) [41, 54, 64], *A. visnaga* extracts or some of their components have been used in modern therapeutics as anti-inflammatory, antibacterial, antifungal, antinociceptive, antihyperglycemic, vasodilator, antioxidant, enzymatic inhibiting, cytotoxic, anti-diabetic, anti-cancer and antihyperacidic agents, treatment of urolithiasis and hypertriglyceridemia, as well as for inhibition of oxalate nephrlithiasis.

In the pest control, research work on *A. visnaga* extracts, or some of its chemical constituents, against insect pests is unfortunately little. The available literature reported an ovicidal activity of its extracts against hessian fly *Mayetiola destructor* (Lamiri *et al.*, 2001b) [65], larvicidal activity against *Aedes aegypti*, *Anopheles stephensi* and *Culex quinquefasciatus* (Amer and Mehlhorn, 2006 a, b; Pavela, 2008) [66 68], as well as adulticidal and ovicidal activities against *Callosobruchus maculatus* (Tripathi *et al.*, 2001)[**69**]. These extracts had been reported, also, as a grain protectant against *Sitophilus granarius* (Abdel-Latif, 2004) [70] and *Rhizopertha dominica* (Halawa *et al.*, 2009) [71]. Hexane extract of *A. visnaga* exhibited toxic effect on *Sesamia cretica* (El-Hefny and Yacoub, 2010) [72]. The acaricidal activity of aqueous extracts of *A. visnaga* was reported against *Tetranychus urticae* (Pavela, 2016) [73]. With regard to *S. gregaria*, these extracts disruptively affected several physiological processes (Ghoneim *et al.*, 2014 a,b,c) [74-76].

Transamination has been demonstrated in a number of insect tissues, particularly that concerning glutamate, aspartate and alanine (Gilmour, 1965) [77]. 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 (Mordue and Golworthy, 1973) [78]. Moreover, transaminases, especially GPT, acts as a catalytic agent in carbohydrates metabolism (Katumuma *et al.*, 1968) [79].

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 (Pugazhvendan and Soundararajan, 2009) [80]. The exposure of an organism to xenobiotic product can modify the synthesis of certain metabolite and disturb the functionality of the organisms (Rodriguez-Ortega *et al.*, 2003) [81]. 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 (Arrese and Soulages, 2010) [82]. The present study aimed to evaluate the effects of different extracts of *A. visnaga* fruits on the transaminase activities in haemolymph and fat bodies of last instar nymphs and newly emerged adult females of *S. gregaria*.

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 **(**1961) [83] and improved by Ghoneim *et al.* (2009) [84], 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

A weight of 1.5 Kg *Ammi visnaga* fruits, which purchased from an Egyptian market, was thoroughly cleaned with tap water for disposing of impurities. The fruits were shade dried and then finely ground by a micromill. Solvents of different polarities were used for the extraction, as follows. The pulverized powder was macerated with ethanol in a closed container for a defined period with frequent agitation until soluble matter is dissolved as adopted from Ncube *et al.* (2008) [85]. The ethanol extract was divided into two parts: a part of the ethanol extract was evaporated for obtaining 25 gm dried extract. Another part was concentrated into 300 ml by rotary evaporator, and then diluted with 300 ml distilled water. Using a separating funnel, the dilute was fractionalized by petroleum ether (300 ml x 5) and n-butanol (300 ml x 5) giving 27 and 23 gm, respectively. From each of the crude ethanol extract and the fractionalized petroleum ether and n-butanol extracts, six concentrations were prepared: 80.0, 40.0, 20.0, 10.0, 5.0 and 2.5%. According to tests of toxicity, LC_{50} values were calculated as 21.0, 12.0 and 22.5%, for ethanol, petroleum ether and n-butanol extracts, respectively.

2.3 Nymphal treatments

LC_{50} concentrations of ethanol, petroleum ether, and n-butanol extracts of *A. visnaga* fruits had been applied on the newly moulted penultimate (4^{th}) instar nymphs of *S. gregaria* through the fresh food leaves of *Trifolium alexandrinum* dipped once in the extract for 3 minutes. A day after treatment, all nymphs (treated and control) were provided, individually, with untreated fresh food leaves. After treatment of penultimate instar nymphs with these concentrations, the successfully moulted last instar nymphs and newly 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. The nymphal ages were assorted as: early- (1-day old), mid- (4-day old) and late-aged (7-day old) nymphs.

2.4 Tissue sampling

For the determination of transaminase 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 transaminase 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 (1975) [86] 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 (Moroney, 1956) [87] for the test significance of difference between means.

3. Results

After treatment of penultimate instar nymphs of *S. gregaria* with LC_{50} concentrations of ethanol, petroleum ether and n-butanol extracts (21.0, 12.0 and 22.5%, respectively) of *Ammi visnaga* fruits, Glutamic oxaloacetic transaminase (GOT) and Glutamic pyruvic transaminase (GPT) activities had been determined in haemolymph and fat bodies of the successfully moulted last instar nymphs (of early-, mid- and late-ages) and newly emerged adult females.

3.1 Effects of *A. visnaga* fruit extracts on GOT activity

According to data assorted in Table (1), a predominant enhancing effect was exhibited on the GOT activity in haemolymph of nymphs, regardless to the *A. visnaga* fruit extracts. For some detail, the most potent enhancing effect was exhibited by petroleum ether extract on the enzyme activity (Increment%: 170.8) in haemolymph of late -aged nymphs, while the least enhancing effect (Increment%: 2.4) was exhibited by petroleum ether extract on it in early-aged nymphs. With regard to adults, both ethanol and n-butanol extracts exerted promoting actions on the enzyme activity in haemolymph (Increment% s: 133.8 and 92.6, respectively) while petroleum ether extract exerted a reducing action (Reduction%: 127.5).

In addition to the disturbance of GOT activity in haemolymph of nymphs and adults, it was disturbed in fat bodies of these developmental stages as exiguously shown in Table (2). Concerning the nymphs, n-butanol extract of *A. visnaga* fruits considerably prohibited the enzyme activity in both nymphs and adults (Reduction% s: 4.1, 6.1 and 5.5 in fat bodies of early-, mid- and late-aged nymphs, respectively, as well as 9.4 in adults). In contrast, petroleum ether extract enhanced the enzyme activity in fat bodies of both nymphs and adults (Increment% s: 162.4, 96.2 and 113.9 in fat bodies of early-, mid- and late-aged nymphs, respectively, as well as 111.1 in adults). In respect of the effect of *A. visnaga* ethanol extract, the enzyme activity was reduced in fat bodies of both early- and mid-aged nymphs but induced in late-aged congeners and adults (for detail, see Table 2).

3.2 Effects of *A. visnaga* fruit extracts on GPT activity

Depending on the data arranged in Table (3), GPT activity was insignificantly reduced in haemolymph along the nymphal instar and remarkably reduced in haemolymph of adults (21.4±1.9 vs. 43.2±2.1 U/L of control adults) by n-butanol extract. Regarding the disruptive effect of ethanol extract, GPT activity was slightly or considerably enhanced in haemolymph of nymphs but significantly prohibited in adults ($P<0.01$). As clearly shown in the same table, petroleum ether extract of *A. visnaga* exhibited an enhancing effect on the enzyme activity in haemolymph of nymphs and newly emerged adults with few exceptions.

In the light of data distributed in Table (4), GPT activity was elaborately induced in fat bodies of early-aged nymphs, regardless to the extract (Increment% s: 6.9, 50.6 and 30.8 by ethanol, petroleum ether and n-butanol extracts, respectively). On the contrary, the enzyme activity was pronouncedly inhibited in fat bodies of mid-aged congeners, regardless to the extract (Reduction% s: 40.6 and 30.9 by ethanol and petroleum ether, respectively) but induced by n-butanol extract (Increment%: 6.9). Concerning the late-aged nymphs, the enzyme activity was dramatically inhibited by petroleum ether extract (122.3±2.3 U/L, compared to 166.4±2.1 U/L of control nymphs) but considerably enhanced by other extracts (Increment% s: 21.1 and 5.3 by ethanol and n-butanol extracts, respectively). With regard to fat bodies of adults, GPT level was unexceptionally declined as response to the reducing action of *A. visnaga*, irrespective of the extract. The strongest reducing action was exerted by ethanol extract (Reduction%: 38.6) but the least reducing action was exerted by petroleum ether extract (Reduction%; 0.6, see Table 4).

Table 1. GOT activity (U/L) in haemolymph of *S. gregaria* nymphs and adults as affected by fruit extracts (LC_{50}) of *A. visnaga*.

Extract		Last instar nymphs			Newly emerged adults
		Early-aged	Mid-aged	Late-aged	
Ethanol	Mean±SD	202.9 ±2.5 d	217.3 ±1.8 d	111.0±2.3 d	156.4±2.3 d
	Change (%)	+149.3	+121.1	+101.5	+133.8
Petroleum ether	Mean±SD	161.1±2.4 d	175.5 ±1.8 d	149.2 ±2.1 d	152.2±2.2 d
	Change (%)	+002.4	+078.5	+170.8	-127.5
n-butanol	Mean±SD	171.4±2.7 d	154.9±1.6 d	101.1±2.2 d	128.9±2.2 d
	Change (%)	+110.6	+057.5	+083.5	+092.6
Control		081.4±2.0	098.3±1.9	055.1±2.3	066.9±2.6

d: very highly significantly different (P<0.001).

Table 2: GOT activity (U/L) in fat bodies of *S. gregaria* nymphs and adults as affected by fruit extracts (LC_{50}) of *A. visnaga*.

Extract		Last instar nymphs			Newly emerged adults
		Early-aged	Mid-aged	Late-aged	
Ethanol	Mean±SD	342.3±2.2 c	281.4±2.1 d	221.5±1.9 c	251.3±2.2 b
	Change (%)	-009.1	-09.7	+011.6	+002.9
Petroleum ether	Mean±SD	988.3±1.8 d	611.4±2.1 d	424.6±2.0 d	515.4±2.3 d
	Change (%)	+162.4	+96.2	+113.9	+111.1
n-butanol	Mean±SD	361.3±2.1 b	303.7±2.3 b	187.5±1.8 b	221.3±2.3 c
	Change (%)	-04.1	-06.1	-005.5	-09.4
Control		376.6±1.7	311.7± 2.2	198.5±2.5	244.2±2.5

b: significantly different (P<0.05), c: highly significantly different (P<0.01), d. See footnote of Table (1).

Table 3: GPT activity (U/L) in haemolymph of *S. gregaria* nymphs and adults as affected by fruit extracts (LC_{50}) of *A. visnaga*.

Extract		Last instar nymphs			Newly emerged adults
		Early-aged	Mid-aged	Late-aged	
Ethanol	Mean±SD	033.2±1.9 a	027.5±1.8 b	020.3±1.8 b	026.4±1.9 c
	Change (%)	+01.6	+07.7	+16.7	-39.0
Petroleum ether	Mean±SD	041.2±2.1 b	022.3±2.1 a	020.1±2.1 a	048.5±1.8 b
	Change (%)	+26.0	-12.7	+15.7	+12.3
n-butanol	Mean±SD	028.5±1.8 a	023.3±2.2 a	013.4±1.7 a	021.4±1.9 c
	Change (%)	-12.8	-08.6	-05.7	-50.5
Control		032.7±2.8	025.5±2.1	017.4±2.1	043.2±2.1

Mean ± SD followed with a: not significantly different (P>0.05), b, c: See footnote of Table (2).

Table 4: GPT activity (U/L) in fat bodies of *S. gregaria* nymphs and adults as affected by fruit extracts (LC_{50}) of *A. visnaga*.

Extract		Last instar nymphs			Newly emerged adults
		Early-aged	Mid-aged	Late-aged	
Ethanol	Mean±SD	136.4±1.9 b	117.3±1.9 c	201.4±1.9 c	185.4±2.2 b
	Change (%)	+06.9	-40.6	+21.1	-11.4
Petroleum ether	Mean±SD	191.6±2.3 c	136.6±2.3 c	122.3±2.3 c	128.5±1.8 d
	Change (%)	+50.6	-30.9	-26.6	-38.6
n-butanol	Mean±SD	166.4±2.3 c	211.3±2.1 b	175.2±1.9 c	208.6±2.3 b
	Change (%)	+30.8	+06.9	+05.3	-00.6
Control		127.2±2.1	197.6±2.2	166.4±2.1	209.3±2.2

b, c: See footnote of Table (2), 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 (Etebari *et al.*, 2005**)** [88]. 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 (Mordue and Golworthy, 1973) [78]. 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 (Azmi *et al.*, 1998; Giboney, 2005; Etebari *et al.*, 2007) [89, 91]. In other words, they may play an important role in the insecticidal poisoning (Abd El-Mageed, 2002; Hassan, 2002) [92, 93].

4.1 Disturbed GOT activity in *S. gregaria* by *A. visnaga* fruit 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 (Ghoneim *et al.*, 2016; Saha *et al.*, 1986; Tabassum, 1994; Tabassum *et al.*, 1998; Bakr *et al.*, 2002; Zohry, 2006; Abdel-Ghaffar and Ghoneim, 2007; Al-Dali,

2008; Ezz and Fahmy, 2009; Hamadah, 2009; Tanani *et al*., 2009) [40, 94, 103].

In the present study, a predominant inducing effect was exhibited by all *A. visnaga* fruit extracts on the GOT activity in haemolymph of *S. gr*egaria nymphs. Also, the enzyme activity was induced in haemolymph of adults by ethanol and n-butanol extracts. In fat bodies of both nymphs and adults, petroleum ether extract enhanced the enzyme activity. These results are in agreement with some reported results of induced GOT activity in various insects by different plants extracts since Ghoneim *et al.* (2014d) [104] recorded enhanced activity in haemolymph of last instar nymphs and in fat bodies of adults of *S. gregaria* by *Punica granatum* peel extracts. In no certain trend, a predominant enhancing effect of *Nigella sativa* seed extracts was unexceptionally exhibited on GOT activity in haemolymph along the last nymphal instar and in haemolymph of adults of the same locust (Ghoneim *et al.*, 2016) [40]. Also, methylene chloride extract of *Azadirachta indica* enhanced the enzyme activity in the same locust (Asiri, 2015) [105]. Sublethal doses of Basil essential oil *(Ocimum basilicum*) promoted the enzyme activity in *Sitophilus granarius* adults (Abo El-Makarem *et al*., 2015) [106].

In the current work, enhanced GOT activity in haemolymph of last instar nymphs and adults of *S. gregaria* by certain extracts of *A. visnaga* fruits, as well as in fat bodies of both nymphs and adults by 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 (Zeba and Khan, 1995) [107]. This suggestion may be conceivable since several chemical constituents had been identified in *A. visnaga* fruits, such as khellin, visnagin, flavonoids, linalool, aliphatic esters, Isobutyrate, 2,2-dimethylbutanoic acid, croweacin, isobutyl isobutyrate, linalool, thymol, fenchyl acetate, bornyl acetate, α-isophorone, 2,2-dimethyl butanoic acid and croweacin (Beltagy and Beltagy, 2015; Martelli *et al*., 1984; Alqasoumi *et al*., 2014; Bencheraiet *et al*., 2011; Lamiri *et al*., 2001a; Talaat *et al*., 2014) [41, 45, 48, 50]. 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.* (2013) [108] for some bioinsectcides against *Spodoptera 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* (Uma devi and Sujatha, 2013) [109], *Rhyzopertha dominica* adults by hexane extract of *Capparis deciduas* (Upadhyay, 2013)[110], *Pieris rapae* by methanolic extract of *Silybium marianum* (Hasheminia *et al.*, 2013) [111], *S. gregaria* fat bodies of nymphs and adults by *P. granatum* peel extracts (Ghoneim *et al.*, 2014d) [104], *T. castaneum* larvae by essential oils of *Wedelia trilobata* and *Melissa officinalis* (Khater and El-Shafiey, 2015) [112] and by garlic oil (*Allium sativum*)(Beltagy and Omar, 2016) [113]. In consistent with these reported results, GOT activity was inhibited in haemolymph of *S. gregaria* adults by petroleum ether extract of *A. visnaga* fruits, in the present study. Also, ethanol extract exhibited a reducing effect on the enzyme activity in fat bodies of both early- and mid-aged nymphs. Moreover, n-butanol extract considerably prohibited the enzyme activity in fat bodies of both nymphs and adults. However, the reduced GOT activity in the present investigation can be attributed to difficulty in the formation of dissociable enzyme-inhibitor complexes which reduce the specific enzyme activity (Dragomirescu *et al*., 1979) [114] or to a disturbance of the link between the carbohydrate and protein metabolism.

4.2 Disturbed GPT activity in *S. gregaria* by *A. visnaga* fruit extracts

In the present study, GPT activity was enhanced in haemolymph of *S. gregaria* nymphs by ethanol extract of *A. visnaga* fruits. In addition, petroleum ether extract exhibited a similar effect on the enzyme activity in haemolymph of nymphs and adults, with few exceptions. The enzyme activity was elaborately induced in fat bodies of early-aged nymphs, regardless to the extract. In fat bodies of the late-aged nymphs, the enzyme activity was considerably enhanced by ethanol and n-butanol extracts. These results are in agreement, to some extent, with 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 (Ghoneim *et al.*, 2014d) [104], in whole body homogenate of *S. gregaria* nymphs by methylene chloride extract of *A. indica* extract (Asiri, 2015)[105] and in adults of *S. granarius* by sublethal doses of essential oils of *Eugenia aromatic* or *O. basilicum* (Abo El-Makarem *et al.*, 2015)[106]. 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 (Assar *et al.*, 2010) [115], *Bactrocera zonata* by methoxyfenozide or lufenuron (Mosleh *et al.*, 2011) [116] and *C. pipiens* by Cyromazine (Assar *et al.*, 2012) [117] and *S. littoralis* by spores of *B. bassiana* and *M. anisopliae* (Mirhaghparast *et al.*, 2013) [118] or by emamectin benzoate (Abd-El-Aziz, 2014) [119].

In the present study, enhanced activity of GPT in *S. gregaria* by some extracts of *A. visnaga* fruits 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 (Etebari *et al.*, 2005) [88].

The available literature, on the other hand, clearly reported GPT reduction in different pests by some botanicals. For some detail, suppressed enzyme activity in *T. castaneum* was observed after treatment with different extracts of *C. longa* (Uma devi and Sujatha, 2013) [109], in *R. dominica* after treatment with hexane extract of *C. deciduas* (Upadhyay, 2013) [110], in *P. rapae* after treatment with methanolic extract of *S. marianum* (Hasheminia *et al.*, 2013) [111], in *Aedes aegypti* larvae after treatment with methanolic extract of *L. camara* (Rajan, 2013) [120], in haemolymph of mid-aged nymphs and in fat body of adults of *S. gregaria* after treatment with *P. granatum* peel extracts (Ghoneim *et al.*, 2014d) [104], in *T. castaneum* larvae after treatment with essential oils of *W. trilobata* and *M. officinalis* (Khater and El-Shafiey, 2015) [112] or garlic oil (*A. sativum*). (Beltagy and Omar, 2016) [113], in *Callosobruchus analis* after treatment with *Acorus calamus* (essential oil) or Biosal (Neem preparation) (Arif *et al.*, 2015) [121], etc. In accordance to those results, GPT activity was slightly reduced in haemolymph along the nymphal instar of *S. gregaria* but remarkably reduced in haemolymph of adults by

n-butanol and ethanol extracts, in the present study. The enzyme activity was considerably inhibited in fat bodies of mid-aged nymphs by ethanol and petroleum ether extracts. A similar inhibitory effect was exhibited on the enzyme activity in fat bodies of late-aged nymphs by petroleum ether extract. Furthermore, GPT level was unexceptionally declined in fat bodies of adults, irrespective of the extract.

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 (Abulyazid *et al.*, 2005) [122]. Several chemicals had been identified in *A. visnaga* fruits (Beltagy and Beltagy, 2015; Martelli *et al.*, 1984; Alqasoumi *et al.*, 2014; Bencheraiet *et al.*, 2011; Lamiri *et al.*, 2001a; Talaat *et al.*, 2014) [41, 45, 48, 50]. 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 *A. visnaga* fruit 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 (Nath, 2000) [123], or the effect of certain effective components in these extracts on the neurosecretory hormonal pattern.

5. Conclusions

The disturbance in GOT and GPT levels is 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 these transaminases in nymphs and adults of *S. gregaria* as responses to *A. visnaga* fruits 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 tested extracts can be used as a part of Integrated Pest Management Program against this dangerous locust *S. gregaria*.

6. References

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