

Toxicological and Biochemical Effects of Four Bioactive Secondary Plant Metabolites Isolated from *Simmondsia Chinensis* (Link) and *Artemisia judaica* L. on White Albino Rats

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ABSTRACT

Four pesticidal botanicals were isolated (two from, *Simmondsia chinensis* and two from *Artemisia judaica*) and identified as simmondsin, simmondsin 2'-ferulate, piperitone and *trans*-ethyl cinnamate, respectively. Oral administration of single doses each of 250 or 500 mg/kg from each compound did not show any signs of toxicity or mortality to male albino rats after two days of treatment. The *in vivo* effects of these compounds on activities of alkaline phosphatase (ALP), gamma-glutamyl transferase (GGT), lactate dehydrogenase (LDH) and creatinine phosphokinase (CPK) in the serum of treated rats and concentrations of testosterone, follicle stimulation hormone (FSH) and triiodothyronine hormone (T3) were evaluated. Simmondsin 2'-ferulate displayed significant decrease in the activity of ALP enzymes at the two tested doses. Simmondsin caused decrease in the activity of the enzymes at 250 mg/kg and increase in the enzymes activity at 500 mg/kg. Piperitone and *trans*-ethyl cinnamate increased the activity of the enzymes at two tested doses. The tested compounds showed no significant effect on the activity of GGT except for *trans*-ethyl cinnamate at 500 mg/kg. Simmondsin and simmondsin 2'-ferulate displayed significant decrease in the activity of both LDH and CPK, while piperitone and *trans*-ethyl cinnamate caused increase in the activity of these enzymes. The tested compounds lowered the concentration of FSH. Simmondsin and simmondsin 2'-ferulate decreased the concentration of T3 and testosterone. Piperitone and *trans*-ethyl cinnamate increased the concentration of these enzymes.

Keywords: Biochemical effects; simmondsin; simmondsin 2'-ferulate; piperitone; *trans*-ethyl cinnamate; white albino rats; enzymes; hormones.

INTRODUCTION

Natural products have enormous potential to inspire and influence modern chemical research of agriculture and medicine. The increasing interest in the possible application of secondary metabolites to pest management has directed the investigation toward search for new sources of biologically active natural products with low mammalian toxicity, lack of neurotoxic mode of action, low persistence in the

environment and biodegradability, as well as delaying the development of pest resistance. Recent studies on this field showed the importance of natural chemicals as a possible source of non-phytotoxic, systemic and easily biodegradable alternative pesticides (Jacobson, 1989; Singh et al., 1997; Gonzalez-Coloma et al. 1997).

In our previous studies, simmondsin and simmondsin 2'-ferulate isolated from *Simmondsia chinensis*, a semi evergreen shrub, and piperitone and *trans*-ethyl cinnamate isolated from *Artemisia judaica*, a perennial fragrant shrub, showed promising insecticidal and antifeedant activities against the third instar larvae of the Egyptian cotton leafworm, *Spodoptera littoralis* (Abbassy et al., 2006 and 2007). In addition, these compounds revealed antifungal and molluscicidal activities against some of plant pathogenic fungi and a land snail *Eobania vermiculata* (Abbassy et al., 2007, Abdelgaleil et al., 2007). This strong pesticidal potential of the isolated compounds encouraged us to study the side effects of these compounds on mammals. Therefore, the present study describes the toxic effects of the isolated compounds; simmondsin, simmondsin 2'-ferulate, piperitone and *trans*-ethyl cinnamate on male rats and their *in vivo* effects on the activities of alkaline phosphatase, gamma-glutamyl transferase, lactate dehydrogenase and creatinine phosphokinase in the serum of treated rats and on the concentrations of testosterone, follicle stimulation hormone and triiodothyronine hormone.

MATERIALS AND METHODS

1. Plant materials

Seeds of *Simmondsia chinensis* (Link) Schneider (jojoba plant) and aerial parts of *Artemisia judaica* (L.) (Shih plant) were collected from Al-Bostan Region and Sinai Peninsula, respectively.

2. Instruments

¹H and ¹³C NMR spectra were recorded at 500 MHz and 125 MHz, respectively, on a JEOL JNM ECD 500 Spectrometer in CDCl₃. IR spectra were performed with

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Perkin Elmer 1430 Ratio Recording Infrared Spectrometer in KBr disks. UV spectra were measured with HELIOS α UV-VL Spectrophotometer V4.60 in methanol.

3. Extraction, isolation and identification of the active principles from *S. chinensis* and *A. judaica*

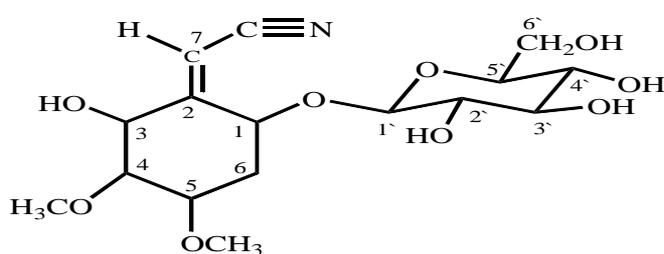
The chloroform extract of *S. chinensis* seeds (45.4 g) was subjected to silica gel column chromatography eluted by chloroform, 10% methanol/chloroform, 20% methanol/chloroform and methanol solvent systems. Thirty fractions of 200 ml were collected and pooled to two main fractions based on their TLC profiles. Fraction 1 (3.4 g) was further purified by silica gel column chromatography using 10% chloroform/methanol solvent system to give 2.1 g of a compound simmondsin 2'-ferulate (2), (Figure 1). Similar purification of the second fraction (7.3 g) followed by recrystallization from acetone/chloroform (1:4) gave 5.4 g pure prisms of a compound which identified as simmondsin (1).

Simmondsin (1): δ ^1H NMR 1.68 (1H, dt, J = 14.5 and 3.9 Hz, H-6a), 2.48 (1H, dt, J = 15.3 and 3.9 Hz, H-6b) 3.13 (1H, dd, 9.2 and 3.1 Hz H-2''), 3.22 (1H, m H-4), 3.29 (1H, t, J = 8.4 Hz, H-5''), 3.32 (1H, t, J = 9.2, H-4''), 3.36 (1H, t, J = 8.6 Hz, H-3''), 3.43 (3H, s, 5-OCH₃), 3.45 (3H, s, 4-OCH₃), 3.64 (1H, dd, J = 12.3 and 5.4 Hz, H-6'a), 3.81 (1H, brd, J = 12.2 Hz, H-6'b),

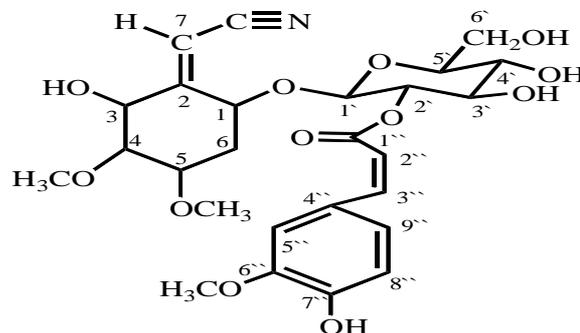
3.90 (1H, q, J = 3.7 Hz, H-5), 4.37 (1H, d, J = 8.4 Hz, H1''), 4.71 (1H, brd, J = 8.4, H-3), 4.86 (1H, t, J = 3.9, H-1) and 5.69 (1H, s, H-7).

Simmondsin 2'-ferulate (2): δ ^1H NMR 1.47 (1H, d, J = 12.3 Hz, H-6a), 2.39 (1H, d, J = 14.5 Hz, H-6b) 3.00 (1H, dd, 10.0 and 3.3 Hz, H-4), 3.23 (3H, s, 5-OCH₃), 3.35 (1H, m H-5''), 3.37 (3H, s, 4-OCH₃), 3.45 (1H, dd, J = 19.1 and 9.9, H-4''), 3.60 (1H, m, H-3''), 3.72 (2H, m, H-6''), 3.84 (3H, s, O-CH₃), 3.84 (1H, H-1''), 4.60 (1H, d, J = 7.7 Hz, H-3), 4.73 (1H, dd, J = 21.4 and 11.5 Hz, H-2''), 4.86 (1H, t, J = 9.2 Hz, H-1), 5.71 (1H, s, H-7), 6.34 (1H, d, J = 15.3, H-2''), 6.80 (1H, d, J = 5.1, H-8''), 7.04 (1H, d, J = 8.4 Hz, H-9''), 7.63 (1H, d, J = 15.3 Hz, H-3''), 7.76 (1H, s, H-5'').

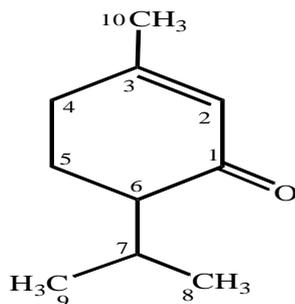
Extraction, isolation and identification of the active principles from *A. judaica* were done according the methods which adopted by Abbassy et al., (2007) and Abdelgaleil et al., (2007). Dried and powdered aerial parts of *A. judaica* were hydrodistilled in a Clevenger-type apparatus. The resulting essential oil (18 g) was chromatographed on silica gel column using hexane, 2.5% acetone/hexane, 10 % acetone/hexane and acetone solvent systems. The resulting fractions were concentrated under reduced pressure and examined by



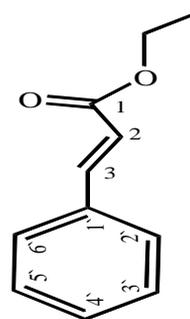
Simmondsin (1)



Simmondsin 2'-ferulate (2)



Piperitone (3)



trans-Ethyl cinnamate (4)

Figure1. Chemical structure of isolated compounds from *Simmondsia chinensis* (1 and 2) and *Artemisia judaica*(3 and 4)

TLC to offer two main fractions. The first fraction was further purified on silica gel column eluted with 1% acetone/hexane to give 2.05 g of a compound which identified as *trans*- ethyl cinnamate (4). The second fraction was subjected to silica gel column eluted with chloroform to offer 2.52 g of a compound which identified as piperitone (3).

Piperitone (3): δ ¹H NMR 0.50 and 0.60 (each 3H, t, J = 6.9 Hz, Me-8 and Me-9), 0.74 (1H, m, H-4a), 0.94 (1H, m H-5), 1.35 (1H, m, H-4b), 1.46 (m, H-6b), 1.59 (3H, d, J = 11.5 Hz, H-10), 1.66 (1H, m, H-7), 1.99 (1H, m, H-6a) and 5.46 (1H, d, J = 11.5 Hz, H-3).

***trans*-Ethyl cinnamate (4):** δ ¹H NMR 1.06 (3H, t, J = 6.9 Hz, CH₂CH₃), 3.98 (2H, q, J = 6.9 Hz, CH₂CH₃), 6.18 (1H, d, J = 17.6 Hz, H-2), 7.08 (3H, m, H-3', 4' and 5'), 7.23 (2H, m H-2' and H-6') and 7.44 (d, J = 19.4 Hz, H-3).

4. Animals

White male albino rats (*Rattus albinos*) were obtained from a breeding culture in the Animal Health Research Centre (Cairo, Egypt). The rats were kept under observation for two weeks before treatments.

5. Toxicological and biochemical effects of the isolated compounds on white male rats.

Two groups of adult's male albino rats (100 – 120 gm) were used in this study. The rats of the first group were given a single oral dose (250 or 500 mg / kg body weight) of each isolated compound in corn oil by stomach tube (single oral dose treatment). The rats of the second group (control group) were administered corn oil without any compounds. All treated rats were observed and mortality percentage was calculated in each treatment. The surviving rats treated with the single oral dose and control treatments were sacrificed two days after treatment and then blood samples collected in clean sterile vials and left till clotting occurred. Serum was collected and centrifuged at 4000 rpm for 10 min. The activities of alkaline phosphates, gamma-glut amyl transferase, lactate dehydrogenase, creatinine phosphokinase enzymes, and the concentrations of testosterone, follicle stimulation hormone and triiodothyronine hormone were determined in the serum.

5.1. Measurement of Alkaline Phosphatase activity (ALP)

ALP activity was measured in the serum according to the method reported by Rec (1972) using Diamond Diagnostics Kits. Ten μ l of each sample was transferred into a test tube containing 1 ml of the working solution. The contents were mixed and the absorbance was measured after 0.5, 1, 2 and 3 minutes at 405 nm using

Jenway 6305 UV / VIS spectrophotometer. The following formula was used to calculate the alkaline phosphatase activity:

$$\text{Activity (U/L)} = 5454 \times \Delta A_{405 \text{ nm} / \text{min}}$$

5.2. Determination of gamma- Glutamyl Transferase activity (GGT)

GGT activity was carried out according to the method reported by Whitfield et al. (1973) using Liner Chemicals, S. L. Kits. 100 μ l of each serum sample was transferred into a test tube containing 1ml of the working solution. The resultant mixture was incubated for 1 min at 30°C and the change of absorbance per minute (ΔA /min) was measured for a period of 3 minutes at 405 nm using Jenway 6305 UV/VIS spectrophotometer. The following formula was used to calculate the gamma-glutamyl transferase activity:

$$\text{Activity (U/L)} = \Delta A / \text{min} \times 1111.$$

5.3. Determination of Lactate Dehydrogenase activity (LDH)

LDH activity was carried out according to the method reported by Rec (1970) using Diamono Diagnostics Kits. 50 μ l of each serum sample was transferred into a test tube containing 1.5 ml of the working reagent at 25°C. The absorbance was read after 30 sec. and repeated after 1, 2, 3 minutes at 340 nm using Jenway 6305 UV/VIS spectrophotometer. The following formula was used to calculate the lactate dehydrogenase activity:

$$\text{Activity (U/L)} = 4921 \times \Delta A_{340 \text{ nm} / \text{min}}$$

5.4. Determination of Creatinine Phosphokinase activity (CPK)

CPK activity was carried out according to the methods reported by Rec (1977) using Diamono Diagnostics Kits. 50 μ l of each serum sample was transferred into a test tube containing 1.5 ml of the working reagent at 25°C. The absorbance was read after 30 sec. and repeated after 1, 2, 3 minutes at 340 nm using Jenway 6305 UV/VIS spectrophotometer. The following formula was used to calculate the creatinine phosphokinase activity:

$$\text{Activity (U/L)} = 4921 \times \Delta A_{340 \text{ nm} / \text{min}}$$

5.5. Determination of Follicle-Stimulation hormone (FSH) Concentration

The concentration of FSH was carried out according to the method reported by Benidrane (1989) using International Immuno Diagnostics Kits. Assay procedure was as following; 1) secure the desired number of coated wells in the holder, 2) dispense 50 μ l of standard test samples and controls into appropriate wells, 3) dispense 100 μ l of FSH conjugate reagent into

each well, 4) thoroughly mix for 30 seconds. It is very important to have completed mixed in this setup, 5) incubate at 37°C in a water bath for 1 hour, 6) remove the incubation mixture by flicking plate contents into a waste container, 7) rinse and flick the micro titer wells 5 times with distilled water, 8) strike the wells sharply onto absorbent paper to remove all residual water droplets, 9) dispense 100µl TMB reagent into each well. Gently mix for 5 seconds, 10) incubate at room temperature (22 - 25°C) in dark for 30 minutes, 11) stop the reaction by adding 100µl of stop solution to each well, 12) gently mix for 30 seconds. It is important to make sure that all the blue color changes to yellow color completely, 13) read the optical density at 450 nm with a micro titer plate reader within 15 minutes using ELISA, GmbH model Jupiter.

5.6. Determination of Testosterone hormone concentration

Testosterone hormone concentration was carried out according to the method reported by Turkes (1979) using International Immune Diagnostics Kits. Similar procedure of FSH was followed except for adding testosterone conjugate reagent and incubation times of some steps were varied.

5.7. Determination of Triiodothyronine (T3) hormone concentration

Triiodothyronine (T3) hormone concentration was carried out according to the method reported by Cavalieri and Rapoport (1977) using International Immuno Diagnostics Kits. Similar procedure of FSH was followed except for adding T3 conjugate reagent and incubation times of some steps were varied.

5.8. Statistical analysis

Enzyme activities and hormone concentrations were subjected to one-way analysis of variance (ANOVA) followed by Student-Newman-Keuls test (Cohort software Inc., 1985) to determine the significant

differences among mean values at the probability level of 0.05.

RESULTS AND DISCUSSIONS

1. Isolation and structure determination of active principles

The chloroform extract of *S. chinensis* seeds was fractionated on silica gel column chromatography and the most active fractions were further purified on a silica gel column followed by recrystallization to give the two glucosides; simmondsin (1) and simmondsin 2'-ferulate (2). In addition, a monoterpenoid piperitone (3) and a phenylpropanoid *trans*-ethyl cinnamate (4) were isolated from *A. judaica* essential oil by repeated silica gel column chromatography. The chemical structures of the isolated compounds (1-4) were elucidated by using spectroscopic methods, including UV, IR, ¹H NMR, ¹³C NMR and MS. Extensive studies of spectroscopic data of the isolated compounds allowed us to confirm the four isolated compounds as simmondsin (1), simmondsin 2'-ferulate (2), piperitone (3) and *trans*-ethyl cinnamate (4). The two glucosides 1 and 2 isolated from *S. chinensis* have been reported before as main components of jojoba seeds (Van Boven et al., 1994 and Van Boven et al., 1995). On the other hand, the two isolated compounds 3 and 4 represented the major constituents of the oil of *A. judaica* as stated by El-Massry et al., 2002.

2. Toxic and biochemical effects of the isolated compounds

Oral administration of single oral doses each of 250 or 500 mg/kg of each compound did not show any signs of toxicity or mortality in treated rats after two days of treatment.

Results in Table 1 show that oral administration of single doses of simmondsin, simmondsin 2'-ferulate, piperitone and *trans*-ethyl cinnamate to rats revealed

Table 1. *In vivo* effect of isolated secondary plant metabolites on some enzyme activities in the blood serum of treated rats

Compound / Treatment (mg/kg)	ALP (U/L)	GGT (U/L)	LDH (U/L)	CPK (U/L)
Control	146.9±0.25 ^e	11.6±0.25 ^{bc}	225.2 ± 0.36 ^e	82.6 ± 0.25 ^e
Simmondsin 250	144.0±0.21 ^g	11.5±0.15 ^c	222.9 ± 0.31 ^f	74.0 ± 0.35 ^f
Simmondsin 250	149.0±0.20 ^d	11.8±0.36 ^{bc}	219.4 ± 0.25 ^h	72.7 ± 0.35 ^g
Simmondsin 2'-ferulate 250	143.7±0.36 ^g	11.4±0.25 ^c	223.0 ± 0.35 ^f	74.1 ± 0.20 ^f
Simmondsin 2'-ferulate 500	145.5±0.35 ^t	11.6±0.15 ^{bc}	220.1 ± 0.40 ^g	73.9 ± 0.20 ^t
Piperitone 250	147.7±0.20 ^e	11.8±0.15 ^{bc}	225.9 ± 0.20 ^d	86.8 ± 0.35 ^d
Piperitone 500	151.9±0.31 ^b	12.3±0.37 ^{ab}	230.1 ± 0.10 ^b	89.5 ± 0.15 ^b
<i>trans</i> -Ethyl cinnamate 250	150.3±0.25 ^c	12.3±0.25 ^{ab}	227.3 ± 0.20 ^c	88.2 ± 0.20 ^c
<i>trans</i> -Ethyl cinnamate 500	158.2±0.64 ^a	12.8±0.36 ^a	234.1 ± 0.35 ^a	92.1 ± 0.10 ^a

ALP = alkaline phosphatase; GGT = gamma-glutamyl transferase; LDH = lactate dehydrogenase; CPK = creatinine phosphokinase.

Data are expressed as means ± S.D. from experiments with three replicates.

Means within a column sharing the same letter are not significantly different at the 0.05 probability level.

Table 2. *In vivo* effect of isolated secondary plants metabolites on some hormone concentrations in the blood serum of treated rats

Compound/ Treatment (mg/ kg)		T3 (ng/dL)	FSH (mU/mL)	Testosterone (ng/mL)
Control		225.1 ± 0.95 ^c	4.24 ± 0.02 ^a	3.72 ± 0.03 ^a
Simmondsin	250	220.1 ± 2.05 ^d	3.99 ± 0.20 ^b	3.63 ± 0.02 ^b
	500	118.4 ± 0.45 ^e	3.85 ± 0.15 ^b	3.58 ± 0.04 ^b
Simmondsin 2'-ferulate	250	220.6 ± 0.70 ^d	4.00 ± 0.06 ^b	3.65 ± 0.05 ^b
	500	119.8 ± 0.60 ^c	3.87 ± 0.10 ^b	3.61 ± 0.03 ^b
Piperitone	250	225.5 ± 0.60 ^c	3.45 ± 0.01 ^c	3.74 ± 0.03 ^a
	500	229.0 ± 0.35 ^b	3.27 ± 0.04 ^d	3.79 ± 0.02 ^a
<i>trans</i> -Ethyl cinnamate	250	225.8 ± 0.40 ^c	4.02 ± 0.03 ^b	3.73 ± 0.04 ^a
	500	232.6 ± 0.40 ^a	3.85 ± 0.11 ^b	3.77 ± 0.02 ^a

FSH = follicle stimulation hormone; T3 = triiodothyronine hormone.

Data are expressed as means ± S.D. from experiments with three replicates.

Means within a column sharing the same letter are not significantly different at the 0.05 probability level

varied effects on the activity of serum alkaline phosphatase enzymes (ALP). Simmondsin 2'-ferulate displayed significant decrease in the activity of enzymes at the two tested doses compared with control. Simmondsin caused decrease in the activity of the enzymes at 250 mg/kg while it caused increase in the enzymes activity at 500 mg/kg. The activity of enzymes was increased when rats treated with piperitone and *trans*-ethyl cinnamate at 250 and 500 mg/kg. All of the tested compounds had no significant effect on the activity of gamma-glutamyl transferase (GGT) except for *trans*-ethyl cinnamate which increased the enzyme activity at 500 mg/kg.

On the other hand, simmondsin and simmondsin 2'-ferulate displayed significant decrease in the activity of both lactate dehydrogenase (LDH) and creatinine phosphokinase (CPK) enzymes at the two tested doses, while piperitone and *trans*-ethyl cinnamate caused increase in the activity of the tested enzymes. The increase or decrease of the activity of each enzyme was more pronounced in the rats given the high dose of each compound (500 mg/kg) than that of the rats given 250 mg/kg body weight. It has been reported that the extracts of and latex of *Calotropis procera* as well as the isolated compounds uscharin and voruscharin caused elevation or reduction of the activities of liver enzymes (El-Sheikh et al., 1991; Basu et al., 1992; El-Sebae, 1996).

Results in Table 2 revealed that oral administration of simmondsin and simmondsin 2'-ferulate caused significant decrease in the concentration of triiodothyronine hormone (T3) at the two tested doses while piperitone and *trans*-ethyl cinnamate caused slight increase of hormone concentration at 250 mg/kg and significant increase at 500 mg/kg. All of the tested compounds lowered follicle stimulation hormone (FSH) concentration without significant differences between treatments. The tested compounds simmondsin and simmondsin decreased the concentration of testosterone hormone while piperitone and *trans*-ethyl cinnamate

elevated the concentration of the hormone. Again the higher dose of each botanical compound (500 mg/kg) caused higher decrease or increase in the concentration of each hormone than the lower dose (250 mg/kg body weight).

From a standpoint of pesticides chemistry and toxicology the most important aspect in the search for new pest control chemical is to find compounds which are selectivity toxic or biologically active against pest and have low toxicity to mammals. From such standpoint, the more promising pesticidal secondary plant metabolites in this work, simmondsin; simmondsin 2'-ferulate of *S. chinensis* and piperitone and *trans*-ethyl cinnamate of *A. judaica* volatile oil did not show any mortality or any signs of toxicity in rats given single oral doses from each compound up to 500 mg/kg body weight. However, the biochemical results show that dosing rats with a single oral dose either 250 or 500 mg/kg body weight of each botanical compound did cause significant effect on the activities of ALP, CPK, LDH enzymes and on concentrations of FSH, T3, Testosterone in the serum of treated rats.

REFERENCES

- Abbassy, M.A.; Belal, A.H.; Abdelgaleil, S.A.M; Abdel Rasoul, M.A.A. (2006). Insecticidal and antifeedant activities of two glucosides isolated from the seeds of *Simmondsia chinensis* (link) Schneider against cotton leafworm, *Spodoptera littoralis* (Boisd.). J. Pest Cont. Environm. Sci. 14, 271-282.
- Abbassy, M.A.; Abdelgaleil, S.A.M; Belal, A.H.; Abdel Rasoul, M.A.A. (2007). Insecticidal, antifeedant and antifungal activities of two glucosides isolated from the seeds of *Simmondsia chinensis* (link). Ind. Crops Prod. 26, 345-350.
- Abdelgaleil, S.A.M.; Abbassy, M.A.; Belal, A.H.; Abdel Rasoul, M.A.A. (2007). Evaluation of molluscicidal properties of extracts and active principles from two Egyptian plants against the terrestrial snail, *Eobania vermiculata*. J. Pest Cont. Environm. Sci. 15, 101-112.

- Basu, A.; Sen, T.; Ray, N. R.; Chaudhuri, N. K. A. (1992). Hepatoprotective effects of *Calotropis procera* root extract on experimental liver damage in animals. *Fitoterapia*, 63: 507-514.
- Benidrane, M.E. (1989). Production of monoclonal antibodies complementary to an antibody-antigen complex. *J. Immunol. Methods*. 111:189.
- Cavalieri, R.R.; Rapoport, B. (1977). Impaired peripheral conversion of thyroxine to triiodothyronine. *Ann. Rev. Med.*, 28:57-65.
- Cohort Software Inc., (1985). *Costat User's Manual*. Version 3. Cohort Tucson, Arizona, USA.
- El-Massry, K.F.; El-Ghorab, A.H.; Farouk, A. (2002). Antioxidant activity and volatile components of Egyptian *Artemisia judaica* L. *Food Chem.* 79:331-336.
- Gonzalez-Coloma A.; Gutierrez C.; Cabrera R.; Reina M. (1997). Silphinene Derivatives: Their effects and modes of action on Colorado potato beetle. *J. Agric. Food Chem.* 45: 946-950.
- El-Sebae, M. A. (1996). Evaluation of the efficacy of some new compounds as pesticidal activities of some cardenolides isolated from *Calotropis procera* plant. Alex. Univ. Faculty of Agriculture, Ph.D.
- El-Sheikh, H. A.; Ali, B. H.; Homeida, A. M.; Hassan, T.; Idris, O. F.; Hapke, H. J. (1991). The activities of drug-metabolizing enzymes in goats treated orally with the latex of *Calotropis procera* and the influence of dieldrin pre-treatment. *J. Com. Pathol.*, 104: 257-268.
- Jacobson M. (1989). Botanical Pesticides Past, Present and Future. In *Insecticides of Plant Origin*; Arnason JT, Philogene BJR, Morand P, Eds; *ACS Symposium Series 387*, Washington, DC; pp 95-109.
- Rec, G.S.C. (1970). *J.Clin.Chem.Clin. Biochem.* 8: 658.
- Rec. G.S.C. (1972). *J. Clin. Chem. Clin. Biochem.* 10:182.
- Rec, G.S.C. (1977). *J.Clin.Chem.Clin. Biochem.* 15: 255.
- Shrestha, M. K.; Peri, I.; Smirnoff, P.; Birk, Y.; Goldhirsh, A. G. (2002). Jojoba seed meal proteins associated with proteolytic and protease inhibitory activities. *J. Agric. Food Chem.* 50: 5670-5675.
- Singh M.; Khokhar S.; Malik S.; Singh R. (1997). Evaluation of neem (*Azadirachta indica* A. Juss) extracts against American bollworm, *Helicoverpa armigera* (Hubner). *J.Agric. Food Chem.* 45, 3262-3268.
- Turkes, A. (1979) *J. Endocrinol.* 81(2):165.
- Van Boven, M.; Toppet, S.; Cokelaere, M.; Daenens, P. (1994). Isolation and structural identification of a new simmondsin ferulate from jojoba meal. *J. Agric. Food Chem.* 42: 1118-1121.
- Van Boven, M.; Daenens, P. Cokelaere, M. (1995). New simmondsin 2'ferulates from jojoba meal. *J. Agric. Food Chem.* 43: 1193-1197.
- Whitfield, J.B.; Moss, D. W.; Neale, G.; Orme, M. Breckenridge, A. (1973). *J. Brit. Med.*, 1:136.

الملخص العربي

التأثيرات السامة و البيوكيميائية لأربع مركبات طبيعية معزولة من نباتى الجوجوبا

والشيخ على فتران التجارب البيضاء

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إنزيم ALP عند الجرعتين المستخدمتين. مركب simmondsin سبب خفض فى نشاط الإنزيم عند الجرعة 250 مجم/كجم وزيادة فى النشاط عند الجرعة 500 مجم/كجم. مركب piperitone و *trans-ethyl cinnamate* أظهرت زيادة فى نشاط الإنزيم عند الجرعتين المستخدمتين. المركبات المختبرة لم تظهر تأثير معنوى على نشاط إنزيم GGT عدل مركب *trans-ethyl cinnamate* عند الجرعة 500 مجم/كجم. مركب simmondsin و *simmondsin 2`ferulate* أحدثتا خفض معنوى فى نشاط إنزيمى LDH و CPK بينما مركب piperitone و *trans-ethyl cinnamate* أحدثتا إرتفاع معنوى فى نشاط هذين الإنزيمين. المركبات المختبرة خفضت تركيز هرمون FSH عند الجرعات المستخدمة. مركب simmondsin و *simmondsin 2`ferulate* قلل تركيز هرمونى testosterone و T3 فى حين مركب piperitone و *trans-ethyl cinnamate* سببا زيادة غير معنوية فى تركيز هذين الهرمونين.

تم عزل أربع مركبات طبيعية: أثنين من نبات الجوجوبا هما *simmondsin* و *simmondsin 2`ferulate* وأثنين من نبات الشيخ هما *piperitone* و *trans-ethyl cinnamate*. تم إعطاء الفتران جرعة واحدة عن طريق الفم وهى إما 250 مجم/كجم أو 500 مجم/كجم من وزن الجسم من كل مركب. المعاملة بهذه الجرعات لم تظهر أى أعراض تسمم أو موت للفتران بعد يومين من المعاملة. تأثير هذه المركبات على نشاط إنزيمات (ALP) *alkaline phosphatase* و *gamma-glutamyl transferase* (GGT) و *lactate* و *dehydrogenase* (LDH) و *creatine phosphokinase* (CPK) وكذلك تركيز هورمونات testosterone و *follicle stimulation hormone* (FSH) و *triiodothyronine hormone* (T3) فى سيرم دم الفتران المختبرة تم تقييمه. النتائج أظهرت أن مركب *simmondsin 2`ferulate* سبب خفض معنوى فى نشاط