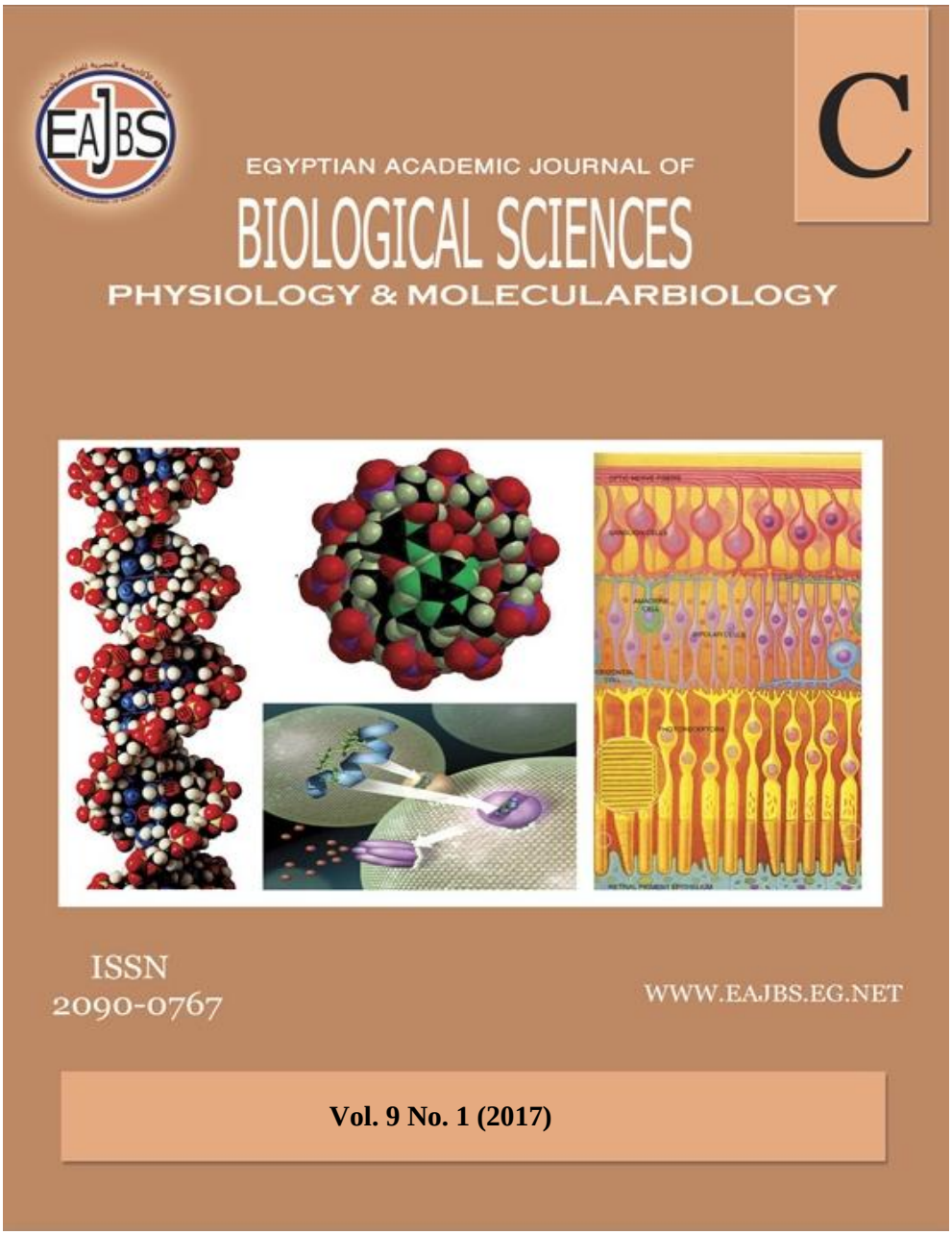

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Ameliorative Effects of Propolis Extract on Some Biochemical and Hematological Parameters of Burnt Skin of Male Guinea pigs.

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ABSTRACT

Burn injury, one of the most common diseases in primary care, is also a major cause of death and disability. The aim of this study was to evaluate the effect of Propolis in thermal burn wounds in guinea pigs and to compare its effects with those of silver sulfadiazine (SSD), the most widely used burn treatment. Burn injury was produced in guinea pigs by immersion of the shaved dorsal area to hot water. Male Guinea pigs of approximate (550 g body weight each) were divided into five groups. In the normal group; Guinea pigs were orally administered with 0.9% isotonic saline solution at a dose (10 ml/kg body weight). The second, served as positive control group which were orally administered with 0.9% isotonic saline solution at a dose (10 ml/kg body weight), then standard burns were obtained on the dorsal skin. The 3rd group was treated by Propolis topically (100 mg/kg b.wt), the 4th group treated with silver sulfadiazine topically (layer thickness of about 3–5 mm) and the 5th group treated with Propolis orally and topically (100 mg/kg b.wt). Every group contains 20 animals and sacrificed at 15 and 30 days post-treatment by Propolis or SSD (10 animals per each). The results are recorded after monitoring the CBC including (RBCs, WBCs, platelets, Hb content and HCT percentage), lipid profile including **(**total cholesterol (TC), triglyceride (TGs), high density lipoprotein (HDL), low density lipoprotein (LDL) and very low density lipoprotein (vLDL)**)** and the skin antioxidant status (catalase activity (CAT), superoxide dismutase (SOD) and serum nitric oxide (NO) beside malondialdehyde (MDA) concentration) were also monitored during the study compared to normal animals. The recorded results declared that, the treatment with Propolis has shown an ameliorative effect on burn healing. These observations and investigations were the pacemaker for the hypothesized ameliorating activity of Propolis in the present study.

INTRODUCTION

Burns led to insufficient blood volume, ischemia, and body damage caused by ischemic reperfusion. Burns can cause many changes in the systemic response-; the metabolic changes represent an important one among systemic response (Hettiaratchy *et al*., 2004). The antioxidant system of tissues is damaged by injury and cannot cope against reactive oxygen species (ROS) in the following period (Dubick *et al*., 2002). However, burn wound healing still remains a challenge to modern medicine. Methods such as sap therapy, wound cure, skin graft, and nutrition need simultaneous treatment because burn can lead to serious complications (Zhang *et al*., 2011).

Propolis, a complex resinous material collected by honeybees from buds and exudates of certain plant. Propolis includes; fatty and phenolic acids and esters, substituted phenolic esters, flavonoids (flavones, flavanones, flavonols, dihydroflavonols, chalcones), terpenes, β- steroids, aromatic aldehydes and alcohols, and derivatives of sesquiterpenes, naphthalene and stilbenes (Marcucci *et al*., 1996). The use of propolis goes back to ancient times, at least to 300 BC, and it has been used as a medicine in local and popular medicine in many parts of the world, both internally and externally. In recent years, propolis has attracted researcher's interest because of its many beneficial biological effects, such as hepatoprotective, antitumour, antiox-idant, antimicrobial, anti-inflammatory, antiviral, antifungal and antiparasite activities (Sforcin, 2007). The chemical composition of propolis depends on the specificity of the local flora at the site of collection.

Silver sulfadiazine (SSD) is the topical agent of choice in severe burns and is used almost universally today in preference to compounds such as silver nitrate and mafenide acetate. SSD cream, while being effective, causes some systemic complications including neutropenia, erythema multiforme, crystalluria and methaemoglobinemia (Hosnuter *et al*., 2004).

In this study, we examined the effects of propolis treatment on skin after thermal injury in an animal model compared to Silver sulfadiazine treated group. CBC counts, MDA levels, superoxide dismutase, catalase, Nitric oxide and lipid profile (cholesterol, HDL, LDL, vLDL and triglycerides) were measured 15 and 30 days post-treatment.

MATERIALS AND METHODS

Chemicals: Kits of cholesterol, triglycerides, HDL, LDL, MDA, SOD, NO and catalase were obtained from Biodiagnostic Company, Cairo, Egypt.

Propolis extraction:

Crude Propolis was obtained from an Egyptian honey bee keeper, Qena province, Egypt. Propolis was kept dry and freezed (-40°C) until used. Propolis samples were mixed with distilled water, heated gently and filtered through Whatman No:1 filter paper. Propolis was freshly prepared and the filtrate administered to animals by gavage at dose of 100 mg/kg b.wt. (El-Khayat *et al.* 2009).

Dermazine®cream (silver sulfadiazine) was used as a standard wound healing drug, manufactured by MUP company, EGYPT.

Animals: Adult Guinea pigs of approximate (550 g body weight each) were selected. The animals were housed in the Animal House of the Faculty of science, South Valley University, Qena, Egypt, for two weeks under natural day and night periods and supplied with a balanced diet and water *ad libitum*.

Induction of burns in Guinea Pigs skin:

Standard burns were induced on Guinea Pigs according to (salman, 1995). as the following:

At the time of experiment, the animal was shaved in a circle at the back of thoracic dorsum and a standard burn was induced under light diethyl ether anesthesia. At the center of the shaved area an opened glass tube with a diameter of 1.5 mm was fixed up right by hand in a vertical position. Five ml of continuously boiling water were poured in to the tube using a glass syringe within a very short time. The elapsed period of the hot water on the skin was justly approached to 30 seconds (the time was controlled by a stop watch) after which the tube was removed by tilting it away from the body.

Experimental Protocol:

Animals were divided in to 5 groups of 20 animals each. Animals were sacrificed every 15 or 30 days as the following:

Group1 (normal group): Each of 20 animals were orally administered with 0.9% isotonic saline solution at a dose (10 ml/kg body weight) once a day along the experimental period and served as a normal group.

Group 2 (Positive control group): Standard burns of Guinea pig skin are obtained wounds were treated orally with 0.9% isotonic saline solution at a dose (10 ml/kg body weight) once a day to observe the healing process occurring without treatment. Then10 guinea pigs were sacrificed after 15 days and the other 10 after 30 days.

Group 3 (Propolis topically treated group): Standard burns of skin are obtained wounds were treated by topical application of Propolis (100 mg/kg b.wt). 10 animals were sacrificed after 15days (i.e., after 15 doses of topical treatment). the other 10 animals were sacrificed after 30 days (i.e., after 30 doses of topical treatment).

Group 4 (SSD topically treated group): Standard burns of skin are obtained wounds were treated by topical application of silver sulfadiazine (layer thickness of about 3–5 mm). 10 animals were sacrificed after 15 days (i.e., after 15 doses of topical treatment). The other 10 animals were sacrificed after 30 days (i.e., after 30 doses of topical treatment).

Group 5 (Propolis top. and oral treated group): Standard burns of skin are obtained wounds were treated by orally and topically applications of Propolis (100 mg/kg b.wt). 10 animals were sacrificed after 15 days (i.e., after 15 doses of topical and oral treatment). The other 10 animals were sacrificed after 30 days (i.e., after 30 doses of topical and oral treatment).

Collection of samples:

After 12 hrs Fasting the animals were sacrificed. Peripheral blood was divided into two portions (the first portion was used as whole blood for complete blood count (CBC), the second was used for separation of serum) also a part of the skin tissue was taken to prepare tissue homogenate as the following:

Skin Tissue Homogenate preparation:

Skin tissue was perfused with PBS (phosphate buffered saline) solution, pH7.4 containing 0.16 mg/ml heparin to remove any red blood cells and clots. Tissue then was homogenized in 5-10 ml cold buffer (i.e. 50 Mm potassium phosphate, pH 7.4, 1 mM EDTA and 1 ml/L Triton X-100) per gram tissue. After this the sample was centrifuged at 4,000 rpm for 15 minutes at 4 °C. Finally the supernatant was removed for assay and freeze at -80 °C (Nishikimi *et al.*, 1972).

Biochemical parameter of whole blood, serum and skin tissue:

Biochemical parameters; Lipid profile was estimated according to method of (Trivedi *et al.*, 2004) by enzymatic method. Malondialdehyde (MDA) according to (Ohkawa, *et al.*, 1979), Catalase CAT (Fossati *et al.*, 1980) and super oxide dismutase SOD (Nishikimi *et al.*, 1972), were analyzed also using available kits according the reported methods. Blood cell counts, hematocrit or packed cell volume (PCV) and hemoglobin (Hb) content were determined using Celltac α apparatus from NIHON KOHDEN (MEK-6410K).

Statistical analysis:

The variability degree of results was expressed as means ± standard deviation of means (Mean ± S.D). The significance of the difference between samples was determined using Graph Pad Prism 03n software, where appropriate. The difference was regarded significant at $P<0.05$.

RESULTS

Effect of treatment with propolis or silver sulfadiazine (SSD) on complete blood count (CBC) of the burned skin of male Guinea pigs:

As recorded in Table (1) and illustrated in Figure (1), the burned skin group (Positive control group) showed a significant decrease in RBCs, Hb, HCT and platelets after 15 days and a highly significant decrease after 30 days of the same parameters. In contrast, WBCs count was significantly increased after 15 days and highly significant increased after 30 days. After 15 days of topical treatment with propolis or silver sulphadiazine there was neither a significant increase in RBCs, Hb, HCT and Platelets nor significant decrease in WBCs when compared with the burned skin group.

Table 1: Effect of treatment with Top. Propolis, SSD alone and (Oral administration of Propolis +Top. Propolis) on (RBCs, Hb, HCT, WBCs, and Platelets) of male guinea pigs.

Groups	RBCs $\times10^{12}$/L	Hb g/dl	HCT %	WBCs $\times10^{9}$/L	Platelets $\times10^{9}$ /L
	Mean±S.D.	Mean±S.D.	Mean±S.D.	Mean±S.D.	Mean±S.D.
After 15 days					
Normal	6.55 ± 0.10	13.3± 0.40	41.00 ± 1.17	7.7 ± 0.24	400 ± 10.0
Positive control (Burned skin)	4.51^{-a} ± 0.14	9.1^{-a} ± 0.31	29.00^{-a} ±1.11	15.7^{+a} ± 0.37	290^{-a} ±7.0
Top.Propolis (100 mg/kg) treatment	5.24 ± 0.18	11.2 ± 0.34	34.9 ± 0.76	13.5 ± 0.31	332^{+b} ± 9.0
Top. SSD (layer thickness of about 3–5 mm) treatment	4.80 ± 0.21	10.5 ± 0. 35	33.4 ± 0.65	14.5 ± 0.31	312 ± 8.0
Top.+ Oral Propolis (100 mg/kg)	6.21^{+b} ±0.12	12.5^{+b} ±0.32	38.5^{+b} ±1.13	10.5^{-b} ±0.36	372^{+b} ±10.0
After 30 days					
Normal	6.60 ± 0.15	14.3 ± 0.37	42.5±1.15	7.9 ± 0.26	392 ± 8.0
Positive control (Burned skin)	4.05^{--a} ± 0.24	8.0^{--a} ± 0.23	24.05^{--a} ± 1.04	17.6^{++a} ± 0.34	275^{--a} ± 9.0
Top. Propolis (100 mg/kg) treatment	5.58^{+b} ±0.12	12.30^{+b} ±0.34	37.8^{+b} ± 1.6	12.5^{-b} ±0.33	345^{+b} ±10.0
Top. SSD (layer thickness of about 3–5 mm) treatment	5.18^{+b} ± 0.12	11.6^{+b} ± 0.28	36.3^{+b} ± 1.10	13.6^{-b} ± 0.32	330^{+b} ± 9.0
Top.+ Oral Propolis (100 mg/kg)	6.54^{++b} ± 0.11	13.9^{++b} ± 0.34	40.9^{++b} ± 0.97	8.5^{--b} ± 0.33	389^{++b} ± 11.0

Results are expressed as mean ± S.D. of 10 animals.

a = significantly different from the normal group. b = significantly different from the positive control group.

- = significant decrease at P< 0.05 -- = highly significant decrease at P< 0.01

+ = significant increase at p< 0.05 ++ = highly significant increase at P< 0.01

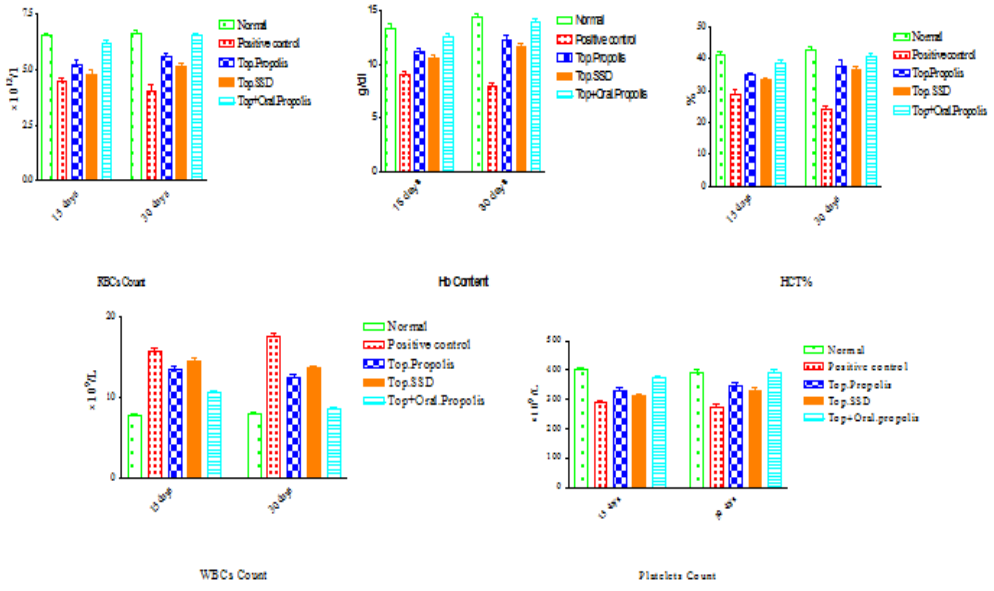

Fig. 1: Effect of treatment with Top. Propolis, SSD alone and (oral administration of Propolis +Top. Propolis) on (RBCs, Hb, HCT, WBCs and Platelets) of male guinea pigs.

While in the case of topical and oral treatment with propolis (after 15 days) there was a significant increase in RBCs, Hb, HCT and Platelets while WBCs recorded a significant decrease. Also in comparing animals with the burned skin group the RBCs, Hb, HCT and Platelets showed a significant increase after 30 days of topical treatment with propolis or silver sulphadiazine and a corresponding significant decrease in WBCs. Similarly a highly significant increase was observed in RBCs, Hb, HCT and Platelets and a corresponding highly significant decrease in WBCs.

Effect of treatment with propolis or Silver sulfadiazine (SSD) on serum lipid profile in burned *Guinea Pigs*:

The recorded results in Table (2) showed that TGs, T. Cholesterol, LDL and vLDL levels in burned male *Guinea pigs* (positive control group) after 15 days increased significantly ($P<0.05$) when compared with normal group.

Table 2: Effect of treatment with Top. Propolis, SSD alone and (Oral administration of Propolis +Top. Propolis) on (Chol., TGs, HDL-C, LDL-C and vLDL) of male guinea pigs.

Groups	Cholesterol mg/dl	Triglycerides mg/dl	HDL-C mg/dl	LDL-C mg/dl	vLDL-C mg/dl
	Mean ± S.D.	Mean ±S.D.	Mean ±S.D.	Mean ± S.D.	Mean ±S.D.
After 15 days					
Normal	105.0 ± 3.96	100.6 ± 4.20	45.2 ± 0.85	39.8 ± 1.55	20.2 ± 1.10
Positive control (Burned skin)	160.6^{+a} ± 3.9	166.5^{+a} ± 4.07	26.7^{-a} ± 1.06	100.8^{+a} ± 2.00	33.2^{+a} ± 1.03
Top.Propolis (100 mg/kg) treatment	129.5 ± 3.76	139.2 ± 3.9	32.8 ± 0.66	69.2 ± 2.90	27.8 ± 0.72
Top. SSD (layer thickness of about 3–5 mm) treatment	134.0 ± 4.5	147.5 ± 3.1	29.8 ±0.66	74.4 ±2.10	29.6 ± 0.69
Top.+ Oral Propolis (100 mg/kg)	122.0^{-b} ±3.01	127.7^{-b} ± 4.3	37.6^{+b} ± 0.82	58.4^{-b} ± 2.59	25.6^{-b} ± 0.71
After 30 days					
Normal	95.3 ± 2.88	102.5 ± 3.11	46.5 ± 1.14	28.6 ± 2.01	20.4 ± 0.99
Positive control (Burned skin)	181.0^{++a} ± 4.24	186.0^{++a} ± 3.54	21.9^{--a} ± 1.04	121.8^{++a} ± 3.18	37.2^{++a} ± 0.67
Top.Propolis (100 mg/kg) treatment	119.0^{-b} ± 3.31	128.0^{-b} ± 3.08	34.7^{+b} ± 0.78	58.4^{-b} ± 2.17	25.6^{-b} ± 0.77
Top. SSD (layer thickness of about 3–5 mm) treatment	125.0^{-b}± 2.21	135.0^{-b}± 3.050	31.5^{+b}± 0.78	66^{-b}± 1.51	27.0^{-b} ± 0.69
Top.+ Oral Propolis (100 mg/kg)	101.7^{--b} ± 3.73	107.0^{--b} ± 3.09	42.0^{++b} ± 0.91	38.6^{--b} ± 2.95	21.4^{--b} ± 1.28

Results are expressed as mean ± S.D. of 10 animals

a = significantly different from the normal group. b = significantly different from the positive control group.

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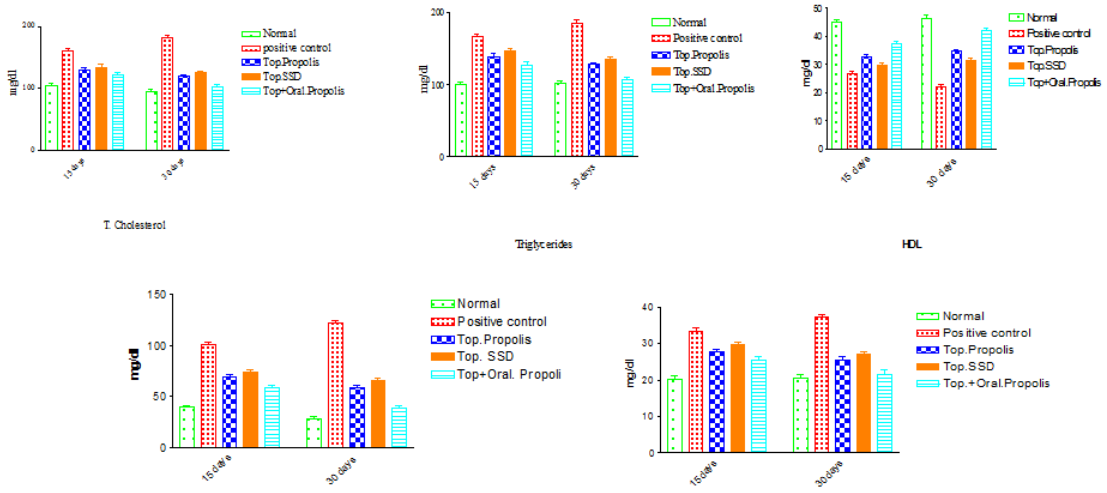

Fig. 2: Effect of treatment with Top. Propolis, SSD alone and (Oral administration of Propolis +Top. Propolis) on (Chol., TGs, HDL-C, LDL-C and vLDL) of male guinea pigs.

While after 30 days a highly significant (P<0.01) increase in their levels were recorded. In contrast, HDL level was significantly decreased after 15 days and was highly significant (P<0.01) decreased after 30 days.

Daily topical treatment with propolis or Silver sulfadiazine (SSD) for 15 days had a non-significant change in serum TGs, T. Cholesterol, LDL and vLDL levels in comparing with the positive control group. While after 30 days, a significant decrease was recorded (P< 0.05) in comparing with the corresponding positive control group.

Also the increase of HDL level was non-significant while the increase became significant (P<0.05) after 30 days. Treatment topically and orally with propolis for 15 days had a significant decrease (P<0.05) in serum TGs, T.Cholesterol, LDL and vLDL levels in comparing with the positive control group. While after 30 days, the decrease became highly significant (P<0.01) in comparing also with the corresponding positive control group. Also the increase of HDL level after 15 days was significant (P<0.05) while, the increase became highly significant (P<0.01) after 30 days, as shown in Table (2) and fig(2).

Effect of treatment with propolis or silver sulfadiazine (SSD) on antioxidant status and MDA concentration in burned skin tissue of Guinea pigs:

Effect of treatment with propolis or silver sulfadiazine (SSD) on antioxidant parameters catalase (CAT), superoxide dismutase (SOD) and nitric oxide (NO):

The activity of CAT and SOD in burned skin tissue of the positive control group after 15 days were significantly decreased (P<0.05) and highly significant decreased (P<0.01) after 30 days as recorded in Table (3) and illustrated in fig (3). In addition the concentration of serum NO in the positive control group was significantly decreased (P<0.05) after 15 days and a highly significant decreased (P<0.01) after 30 days as shown in Table (3) and Fig (3).

There was a significant increase (P<0.05) in CAT, SOD and serum NO of the positive control group after 30 days of treatment with propolis or silver sulfadiazine (SSD) topically. The treatment with propolis topically and orally showed a significant increase (P<0.05) in NO, SOD activity and CAT activity after 15 days as recorded in table 3 and highly significant increase (P<0.01) after 30 days, as shown in Table (3) and fig (3).

Effect of treatment with propolis or silver sulfadiazine (SSD) on malondialdehyde level (MDA):

The level of MDA in burned skin tissue of the positive control group was significantly increased (P<0.05) after 15 days and a highly significant increased (P<0.01) after 30 days as recorded in Table (3) and illustrated in Fig (3).

In contrast, the treatment with propolis or silver sulfadiazine (SSD) topically recorded a significant decrease (P< 0.05) after 30 days as shown in table (3) and fig (3). While treatment with propolis topically and orally showed a significant decrease (P<0.05) after 15 days and highly significant decrease (P< 0.01) after 30 days, as recorded in Table (3) and illustrated in Fig. (3).

Table 3: Effect of treatment with Top. Propolis, SSD alone and (Oral administration of Propolis +Top. Propolis) on (MDA, SOD CAT and NO) of male guinea pigs.

Groups	MDA nmol /g tissue	SOD U/gm tissue	CAT U/gm tissue	NO µmol/L
	Mean±S.D.	Mean ± S.D.	Mean ± S.D.	Mean ± S.D.
After 15 days				
Normal	177.4 ± 3.89	80.10± 2.01	3.21 ± 0.12	45.50 ±1.10
Positive control (Burned skin)	320.5^{+a} ± 7.24	35.13^{-a} ±1.80	1.40^{-a} ± 0.06	20.57^{-a}±1.04
Top.Propolis (100 mg/kg) treatment	232.4 ± 5.61	57.59 ±1.50	2.41 ± 0.09	28.79 ± 1.10
Top. SSD (layer thickness of about 3–5 mm) treatment	257.2 ± 5.21	53.78 ± 1.40	2.11 ± 0.10	24.99 ± 1.50
Top.+ Oral Propolis (100 mg/kg)	$^{-b}$ ± 8.79200.6	64.75^{+b} ±2.12	2.59^{+b} ± 0.12	35.26^{+b} ±1.01
After 30 days				
Normal	181.0 ± 5.01	85.01 ± 2.11	3.45 ± 0.10	41.98 ± 1.02
Positive control (Burned skin)	405.6^{++a} ± 12.00	21.36^{--a} ±1.90	1.00^{--a} ± 0.089	13.12^{--a} ±1.01
Top.Propolis (100 mg/kg) treatment	210.7^{-b} ± 7.94	70.31^{+b} ±1.70	2.72^{+b} ± 0.09	34.10^{+b} ±1.06
Top. SSD (layer thickness of about 3–5 mm) treatment	235.6^{-b}± 6.88	56.3^{+b}1 ± 1.80	2.49^{+b} ± 0.10	29.20^{+b}± 1.05
Top.+ Oral Propolis (100 mg/kg)	186.0^{--b} ± 8.88	81.41^{++b} ±1.92	3.21^{++b} ± 0.10	39.92^{++b} ±1.04

Results are expressed as mean ± S.D. of 10 animals.

a = significantly different from the normal group b = significantly different from the positive control group.

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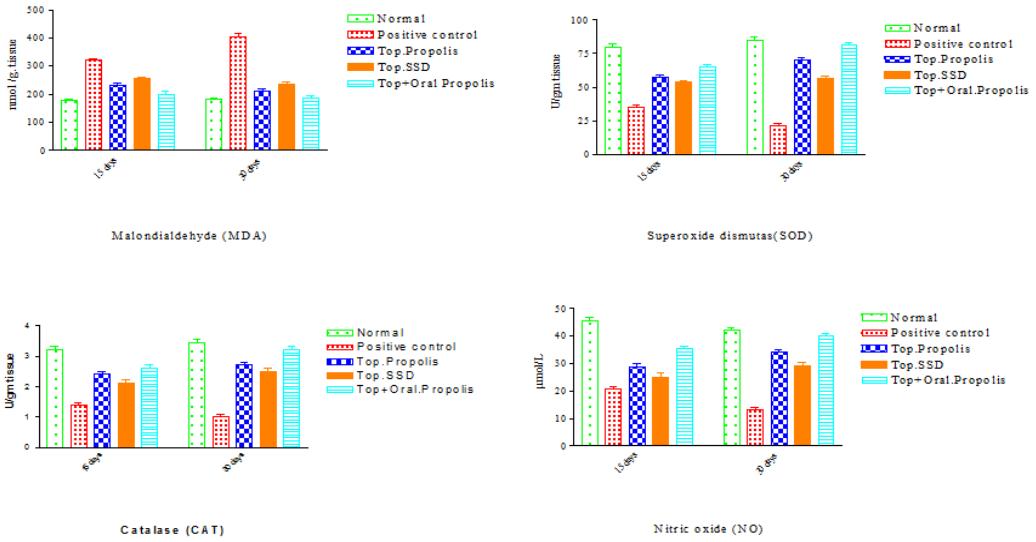

Fig, 3: Effect of treatment with Top. Propolis, SSD alone and (Oral administration of Propolis +Top. Propolis) on (MDA, SOD CAT and NO) of male guinea pigs.

DISCUSSION

Early, adequate and repeated investigation of the circulating blood parameters is therefore an essential tool in the treatment of severe burns (Kramer, 2002). Muir, (1961) has shown that a general relationship exists between the extent of deep burn and the amount of red cell destruction. While Baxter, (1978) has observed a shorter life span of RBCs after burns. The progressive deficit in red cell mass may be accompanied by the appearance of multiple abnormal red cells contributing to early post burn anemia. In the present study, erythrocyte count and blood hemoglobin content significantly decreased in the burn group in comparison with the normal group. The improvement of RBCs, hemoglobin and hematocrit after treatment with propolis, are in accordance with other research. Propolis administration in female albino rats indicated an increase in RBCs, hematocrit and hemoglobin (Cetin *et al*., 2010). The administration of water soluble propolis derivatives in

rabbits clarified a significant increase in RBCs count, hemoglobin and packed cell volume (Hager, 2010).

burned guinea pigs In the present study show decrease in platelet count and rise in WBCs in the initial post-burn days This finding coincides with similar observation in the studies by other authors (Maduli *et al*., 1999; Sarda *et al.*, 2005; Pavic and Milevoj, 2007 and Belba *et al*., 2015). The results of present study clearly demonstrate that propolis treatment exerts a protective effect on platelets and WBCs by increasing platelet count and decreasing WBCs to the normal levels on subsequent post-burn days. The effects of propolis on the platelets and WBCs may occur through its anti-bacterial properties. It has been reported that propolis displays antibacterial action against different pathogenic bacteria (Kujumgiev *et al*., 1999).

Dyslipidemia after burn injury is one of the important alterations that resulted from many factors like hypermetabolic state in burn, release of hormones and inflammatory mediators and organ dysfunction (Birke *et al*., 1972 and Coombes *et al.*, 1980). Furthermore, data obtained by others, showed that the plasma TGs level increase in burned patients due to mainly increased availability of free fatty acids released by stimulated lipolysis in adipose stores due to catecholamines, cytokines, tumor necrosis factor alpha, interleukin- 1, interferon-alpha, beta and gamma; and recently interleukin-6, growth factors such as platelet derived growth factor, transforming and colony-stimulating factors; all modulate the lipid metabolism and may be the cause of these changes (Cree and Wolfe, 2008). In the current study, a marked increase in the concentrations of serum total cholesterol, triglycerides, LDL-C with a decrease in the level of HDL-C was found in the burned groups compared with normal group. Different studies indicated that propolis alleviated the high blood lipids, high total cholesterol and arteriosclerosis (Castaldo and Capasso, 2002). This result is in agreement to the present study with animals treated with propolis and this represents the powerful influence of propolis to reduce the risk of hyperlipidemia. Several studies are in agreement with the present study (Nirala *et al*., 2008) which proved the modulating effect of propolis on total cholesterol and triglycerides levels with a significant increase in total proteins content after beryllium toxicity and the improvement of serum level of HDL-C by propolis in a dose-dependent manner.

Oxygen radical formed and inflammation produced by heat burn causes lipid peroxidation (Hiramatsu *et al.*, 1942). Cetinkale *et al.*, (1997) reported that lipid peroxidation leads to serious damage of the endothelium, resulting to increased capillary permeability, damage of skin tissue, and necrosis. Furthermore, lipid peroxidation is essential for wound and shock caused by burns. Our results showed that lipid peroxidation was increased following burn injury. Propolis treatment, however, decreased MDA activity levels indicating inhibition of lipid peroxidation.

The antioxidant defense system is known to inhibit lipid peroxidation in mammalian tissues by destroying some of ROS that has an important role in initiation of the lipid peroxidation process. The antioxidant defense system operates through enzymatic and nonenzymatic components. The system is affected by burns. It has been reported that nonenzymatic antioxidants, such as glutathione, α-tocopherol and selenium, are decreased in the serum and tissues after thermal injury (Bekyarova and Yankova, 1998). Some authors have reported that SOD and CAT activities gradually decrease after burns (La Londe *et al.*, 1996 and Youn *et al*., 1998). The tissue antioxidant enzyme activities were only decreased in the positive control group when compared to the normal

group. This decrease may be related to the consumption of activated enzymes against oxidative stress. Propolis treatment resulted in improved enzyme activities. Shinohara *et al*., (2002) demonstrated that propolis was found to modulate antioxidant enzymes and decrease lipid peroxidation processes in plasma, liver, lungs, and brain of mice in a dose and tissue dependent manner.

Nitric oxide (NO) is involved in several processes in the skin including wound healing and pigmentation (Weller, 2003). In recent years, NO has emerged as a critical molecule in wound healing, where it increases collagen content in experimental wounds (Thornton *et al*., 1998 and Shi *et al.*, 2000). It is worth to mention that, in other study the wound healing in diabetic guinea pigs are improved by injection of bradykinin potentiating factor (BPF) which is the stimulator of NO (Elshater *et al.*, 2011). So in this study, the burned skin healing in guinea pigs are improved by propolis. Propolis stimulation of the NO may be connected with the ability of the apitherapeutic agent to enhance the expression of transforming growth factor-β (TGF-β) (Ansorge *et al*., 2003). Propolis, NO and TGF-β1 play central roles in collagen synthesis and can cross-regulate each other (Khorasgan *et al.*, 2010 and Olczyk *et al*., 2013).

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ARABIC SUMMARY

التأثيرات التحسينية لمستخلص الصمغ على بعض القياسات البيو كيميائية والدموية في ذكور خنازير غينيا المحترق الجلد.

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تعد الحروق من أكثر الإصابات التي تتم معاينتها طبيا في المستشفيات والعيادات الطبية، ولما لها من مخاطر معقدة فقد كانت دافعا للعديد من المراكز السريرية والبحثية للحصول على مادة من شأنها أن تكون لها الوظائف العلاجية الأساسية.

قد اثبتت الدراسات الحديثة ان صموغ النحل لها العديد من التأثيرات الحيوية الفعالة, على سبيل المثال المقاومة ضد البكتيريا والفيروسات والفطريات والطفيليات والاورام وعوامل الاكسدة والالتهابات. لذلك كان الهدف من هذه الدراسة هو تقييم تأثير مستخلص الصمغ في معالجة الحروق لذكور خنازير غينيا ومقارنته بعقار سلفاديازين الفضية المستخدم على نحو واسع في معالجة الحروق.

ومن ثم فقد تم تقسيم الحيوانات (ذكور خنازير غينيا) الى خمسة مجموعات كل مجموعة تتكون من عشرين حيوان:

المجموعة الأولى (المجموعة الطبيعية):- أعطيت محلول فسيولوجي (كلوريد الصوديوم) وكانت تلك المجموعة بدون حروق.

المجموعة الثانية (المجموعة الغير معالجة): تم حرق الجلد بحروق متساوية (قياسية) على الظهر ثم تركت بدون علاج.

المجموعة الثالثة (المجموعة المعالجة سطحيا بمستخلص الصمغ): تم حرق الجلد بحروق متساوية (قياسية) على الظهر ثم عولجت سطحيا بمستخلص الصمغ (١٠٠ مليجرام/كيلوجرام).

المجموعة الرابعة (المجموعة المعالجة سطحيا بسلفاديازين الفضية): تم حرق الجلد بحروق متساوية (قياسية) على الظهر ثم عولجت سطحيا باستخدام سلفاديازين الفضية (طبقة سمكها ٣- ٥مليمتر).

المجموعة الخامسة (المجموعة المعالجة سطحيا وفميا بمستخلص الصمغ): تم حرق الجلد بحروق متساوية (قياسية) على الظهر ثم عولجت سطحيا وفميا باستخدام مستخلص الصمغ (١٠٠مليجرام/كيلوجرام).

ولقياس المعدلات الدموية والبيوكيميائية ومتابعة عملية الشفاء تم ذبح الحيوانات بعد ١٥ و ٣٠ يوما من العلاج وتم جمع جزء من عينات الدم في أنابيب ايدتا وذلك لعد خلايا الدم الحمراء وخلايا الدم البيضاء والصفائح الدموية وتقدير محتوى الهيموجلوبين ونسبة الهيماتوكريت. وكذلك تم فصل المصل لتقدير كلا من مستوى الكوليسترول الكلى (TC)والكوليسترول عالي الكثافة (HDL) والكوليسترول منخفض الكثافة (LDL)والدهون الثلاثية (TGs) وكذلك النيتريك اوكسيد (NO).تم اخذ عينات من الجلد متساوية في الوزن لتقدير نشاط إنزيمى الكتاليز(CAT) وسوبر أكسيد ديسميوتاز(SOD)وقياس مستوى مالون داى ألدهيد(MDA) في أنسجة الجلد لذكور خنازير غينيا.

ولقد اظهرت النتائج والتحاليل الاحصائية ان المجموعة الغير معالجة كان عدد كريات الدم الحمراء والصفائح الدموية ومحتوى الهيموجلوبين ونسبة الهيماتوكريت منخفضة جدا يقابلها زياده كبيرة في عدد كرات الدم البيضاء. بينما انخفض نشاط كلا من انزيم الكتاليز والسوبر أكسيد ديسميوتاز والنيتريك اوكسيد انخفاضا معنويا وارتفعت كثافة المالون داي ألدهيد مما يدل على وجود اجهاد تأكسدي في انسجة الحيوانات الغير معالجه. وكان مستوى الكوليسترول والدهون الثلاثية والكوليسترول منخفض الكثافة مرتفع جدا نتيجة للتغيرات الايضية التي تنتج عن الحروق وترتب على ذلك انخفاضا معنويا ايضا في مستوى الكوليسترول عالي الكثافة.

اما المجموعة التي تم علاجها بمستخلص الصمغ سطحيا وفميا فقد اظهرت تحسنا معنويا في عدد كريات الدم الحمراء والبيضاء والصفائح الدموية ومحتوى الهيموجلوبين ونسبة الهيماتوكريت ومستوى الدهون وكان لها تأثيرا واضحا في خفض الزيادة الملحوظة في تركيز المالون داى الدهيد ورفع نسبة نشاط إنزيمي الكتاليز وسوبر أكسيد ديسميوتاز في انسجة الجلد لخنازير غينيا بالإضافة الى رفع مستوى النيتريك اوكسيد بمصل الدم ذلك مقارنة بالمجوعة الغير معالجه ويلاحظ ايضا ان التحسن في المجموعتين المعالجتين سطحيا فقط باستخدام مستخلص الصمغ او سلفاديازين الفضية اقل مقارنة بالمجوعة المعالجة سطحيا وفميا بمستخلص الصمغ.

ومما سبق يتبين ان هذا المستخلص يمكن ان يطبق في علاج الحروق لما له من تأثيرات فعالة وذلك عن طريق المعالجة الفمية والسطحية.