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Protective and therapeutic effect of myrrh herb *(Commiphora molmol)* on inhibited immune system in experimental rats

Soha M. Ibrahim[1], Ekbal M. Mohamed[2], El-Sayeda G. E. El-Sahar[2] and Hala R.Ataya[2]
[1]Food Industries Department, Faculty of Agriculture, Ain Shams University
[2]Home Economics Department, Faculty of Specific Education, Ain, Shams University

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ABSTRACT

The present work aimed to study the therapeutic and protective effect of feeding myrrh against immunity inhibition causative to male rats**.** Forty eight rats were used in this study. They were divided into 8 groups, each one of 6 rats, the first group was the (-ve) control, the second group was left as a (+ve) control group. Groups (3, 4 & 5) were induced immunity inhibition for them at the end of experiment and received myrrh with (50, 100 & 150) mg/kg, respectively for four weeks. Groups (6, 7 & 8) were induced immunity inhibition for them at the beginning of the experiment and received myrrh with (50, 100 & 150) mg/kg, respectively for four weeks.

Results revealed that rats in the protective groups (3, 4 and 5), or therapeutic groups (6, 7 and 8) showed significant increased values in levels of their immunity protein (IgG) when compared with untreated rats (+ve group 2). Lymphocytes, total protein and albumin also increased significantly compared to untreated rats (+ve group 2), while their globulin, creatinine, total bilirubin, GPT, GOT and erythrocytes were significantly decreasing compared to control negative group. Group 5 (protective) that had myrrh (150mg/kg) showed highly increase in its immune efficiency.

The histopathological examinations of thymus gland and spleen showed relatively no alteration in their structure in rats that were received myrrh at the beginning of the experiment before induced immunity inhibition to them.

In conclusion myrrh can be used to increase the protective and therapeutic effect for immunity disorders, and to protect the body and raise its immune efficiency n rats.

Key words: Immune system - Myrrh-IgG- histopathological structure, thymus gland, spleen, rats.

INTRODUCTION

Immunity is the capability of multicellular organisms to resist harmful microorganisms. It involves both specific and nonspecific components. The nonspecific components act as barriers or eliminators of a wide range of pathogens irrespective of their antigenic make-up. Other components of the immune system adapt themselves to each new disease encountered and can generate pathogen-specific immunity (Sandra (2011). Nutrient availability has the potential to affect all aspects of the immune system. In general, deficiency of several nutrients will lead to impaired immune responses, and replenishment of those specific components will typically restore the affected responses (Fernandes *et al*., 2006). *Commiphora molmol*, known in folklore medicine as "myrrh" is one of the most common which has been used in the treatment of hypertension, hyperlipidemia, respiratory infections, ulcer and cancer. Myrrh is the dried resin of several species of Commiphora, Burseraceae, of small trees

of the arid and semiarid regions of East Africa, Arabia, and the Indian subcontinent. For many years, myrrh has been used for its healing qualities benefits during injuries clearly, the benefits of using the oleo-gum resin, *C. molmol*, in the middle east system of medicine have been proven in scientific studies (Musselman, 2007). However, the stimulatory role of myrrh, *C.molmol*, on the dynamic of the cellular component of the immune system has not been examined (Faraj, (2005) nor the role of myrrh on the behavior of leukocytes during healing is known (Tariq *et al*., 1985). Myrrh enhanced leukocytes proliferation before injury, it can be concluded that myrrh posse's antigenic-driven responses and that indicated some foreignness or toxicity of some constituents of myrrh. Because myrrh helped to maintain the relative rise of leukocytes counts throughout healing period and that implied it activated late steps of both proliferation and differentiation pathways for all types of leukocytes during effective phase of the specific immune responses (Al-Said, 2010).

MATERIALS AND METHODS

Materials:

1-Animals: Forty eight adult Albino male rats weighting (150-200) gm were obtained from the national research center Dokki Giza Egypt. Animals were clinically healthy and they randomized and housed in stainless steel wire bottom cages (3 rats/ cage) and maintained in air-conditioned room on a 12 light/dark cycle at 22 +2 ^{0}C. All rats were fed a standard diet and water ad-libitum.

2-Diet: The basal diet AIN-93 according to Reeves *et al*. (1993) the basal diet consists of the following: casein 20%, soy oil 4%, choline chloride 2%, vitamin mixture 1%, salt mixture 3.55%, fibers 5% (cellulose), sucrose 10 % and the remainder is corn starch. The basal Diet was obtained from Elgomhoria Pharmaceutical Company, Cairo, Egypt.

3-Myrrh *(Commiphora molmol)* : It was purchased from agriculture research center Giza.

4- Small glass bottles (42) were obtained from Elgomhoria Pharmaceutical Company, Cairo, Egypt.

5-Syrings were purchased from the local market for feeding and blood samples**.**

Methods:

Experimental Design:

After the adaptation period (7 days) rats were classified randomly into 8 groups (6 rats of each) as follow:

Group (1): A negative control group (-ve), they were fed on a basal diet only all the experimental period (4 weeks).

Group (2): Positive control group, were fed on abasal diet after inhibiting their immunity (+ve control).

Group (3): Fed on a basal diet and received 50 mg of myrrh/kg body weight and induced immunity inhibition for them at the end of experiment.

Group (4): As group 3 and was received 100 mg of myrrh/kg body weight, and induced immunity inhibition for them at the end of experiment.

Group (5): As group 3 and was received 150 mg of myrrh/kg body weight,and induced immunity inhibition for them at the end of experiment.

Groups 3,4 and 5 are the protective

Group (6): As group 2, and were fed in basal diet and received 50 mg of myrrh/kg body weight, after inhibiting their immunity at the beginning of the experiment.

Group (7): As group 2 and were fed in basal diet and received 100 mg of myrrh/kg from body weight, after inhibiting their immunity at the beginning of the experiment.

Group (8): As group 2 and were fed in basal diet and received150 mg of myrrh/kg from body weight, after inhibiting their immunityat the beginning of the experiment.

Groups 6, 7 and 8 are therapeutic groups.

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Inducing immunity suppression:

This was done by treatment of each individual rat by placing it in a glass tube of the same size. The rat is not able to move and close the two sides with making breathing holes and swings on the back in a dark place at a temperature of 4-5 °C for 3-4 hours (Riley, 1981).

Biochemical analysis:

Total protein

The biuret reaction was the most extensively used method for quantifying serum protein. Serum protein reacts with copper sulphate in sodium hydroxide to generate a violet biuret complex, according to the reaction's principle. A DRE 3000 HACH spectrophotometer was used to quantify the intensity of the violet hue, which is proportional to the protein concentration (Bjorston *et al*., 2007).

Albumin and globulin:

Albumin was often quantified using a dye-binding approach that takes advantage of albumin's capacity to form a stable complex with the green dye bromocresol. At 546nm and 37°C, the absorbance of the samples and the standard were measured against a reagent blank. These containers, together with their contents, were combined and incubated at 37°C for 90 minutes. A DRE 3000 HACH spectrophotometer was used to estimate albumin level (g/dl).

For Globulin, the approach may overstate albumin by attaching to other proteins because the bromocresol green (BCG) albumin complex absorbs light at a different wavelength than the free dye. As a result, removing the albumin fraction from the total protein fraction yields the total globulin fraction. The data was statistically examined using analysis of variance (ANOVA) and the students T-Test (George, 2009).

.**Creatinine, GOT and GPT**:

The serum creatinine, GOT and GPT level were determined using the Reitman and Frankel method (1957). After utilizing Bio Meraux reagent kits, the color began to emerge and then fresh alkaline picrate was added to the mixture and it was calorimetrically measured at 520 nm (Reitman and Frankel, 1957)

Blood analysis:

Hemoglobin concentration:

Hemoglobin concentration (g/dI) was determined by colorimetric method using hemoglobin kits. The kits were obtained from the Egyptian American Company for laboratory services Determination was conducted spectro-photometerically according to the manufacturer (Waugh and Sarelius, 1996).

Hematocrit *(HCT):*

Blood was taken by hematocrit capillary tubes and was centrifuged for 10 minutes at 3000 r.p.m. for packed cell volume. Hematocrit is a measurement of the amount of red blood cells (HCT) and it was directly read and recorded as given by Waugh and Sarelius (1996).

Erythrocytes and Lymphocytes:

Calculation of the Erythrocytes is a simple method, part of the standard complete blood count. It was called Mean Corpuscular Volume (MCV) in hematology, expressed in femtoliters (fL=10-15 L=μm^3 (and was calculated using the following formula: MCV=10X hematocrit/RBC count. The hematocrit is the volume proportion (percentage) of RBCs in blood, and the RBC count is measured in millions per liter. On the other side, calculating the Surface Area (SA) is more difficult. It can be measured with micropipettes (Waugh and Sarelius, 1996). Absolute (Abs) lymphocytes = WBC count x 1000 x percent lymphocytes (expressed as a decimal). We can determine the

absolute lymphocyte count per mcL (2.5 x1000 x 0.30 = 750 lymphcyte/mcL.

Immunological parameters:

Analyzed immunoglobulin (IgG), Purified IgG fusion samples were frozen at 80°C after purification. Samples were either left untreated (referred to as "initial") or thawed and then frozen/thawed five times, then incubated for 2 weeks at 4°C. Visual inspection of the initial and treated samples was performed. Any symptoms of precipitation, and then reduced and examined observe cleavage of ZE3 or other proteins using non-reducing SDS-PAGE. Decomposition products to compare the images, Image J was utilized. Comparing the band intensity of a completely formed product to that of any other decomposition products, each sample was also subjected to a series of tests (Santi *et al*., 2008).

Histopathological examinations:

Autopsy samples were taken from thymus gland and spleen of the scarified rats and were fixed in 10% formalin saline for 24 hrs. Washing was done in with tap water then serial dilutions of ethyl alcohol were used for dehydration. Specimens were cleared in xylene and embedded in paraffin at 56^{o}C in hot air oven for 24 hrs. Paraffin bees wax tissue blocks were prepared for sectioning at 4 microns thickness by slide microtome. The obtained tissue sections were collected on glass slides, deparaffinized, and stained by hematoxylin and eosin stain for routine examination through the light electric microscope (Banchroft *et al*., 1996)

Statistical analysis:

This was carried out using analysis of variance (ANOVA) test with the statistical analysis system16 (SAS (1996) using a microcomputer program. Results were expressed as ±SD at $P<0.05$ of significance.

RESULTS

Table (1) illustrated the effect of Myrrh on serum total protein, globulin and albumin of male rats with induced immunity inhibition. The highest value (7.68±0.23 g/dl) of the total protein can be observed in rats of group (1), while the lowest value (4.58±0.32 g/dl) can be seen in group (2). Rats in group 2 (+ve control) showed significantly a decreased level compared to group 1 (-ve control). Rats in groups (3, 4, 5, 6, 7 and 8) showed the values of 6.49±0.23, 6.45±0.17, 5.75±0.15, 5.22±0.08, 6.31±0.17 and 7.62±0.20 g/dl, respectively. These values were highely significant compared to that of +ve control group (2). The best result in treated groups (7.62±0.20 g/dl) was found in rats of group (8) that were received 150 mg/kg body weight of myrrh, therapeutic.

The highest value (16.67±1.37 mmol/l) of globulin, was recorded in -ve control group (1), while the value (14.62±0.18 mmol/l) was seen in the group (2) of rats (+ve control), which showed decreased significantly value compared to group (1). Rats in groups (3, 4, 5, 6, 7 and 8) have the values of 13.85±0.49, 13.77±0.37, 12.27±0.31, 11.13±0.18, 13.46±0.36 and 13.69±0.42 mmol/l, respectively and showing a low significantly results compared to group (2) (+ve control). The best treatment result (13.85±0.49 mmol/l) was seen in rats of group (3) (150 mg/kg body weight of myrrh, protective).

The highest level of albumin (5.35±0.28g/dl) was recorded in rats of group (1) (-ve control) which was significantly high compared to group (2) (+ve control). Albumin values of groups (3, 4, 5, 6, 7 and 8) were 4.41±0.16, 4.39±0.12, 3.91±0.10, 3.55±0.06, 4.29±0.11 and 4.68±0.14 g/dl, respectively, which showed an increased significantly values compared to (+ve control) group (2). This means that the highest result in treated groups (4.68±0.14 g/dl) was seen in rats of group 8 (150 mg/kg therapeutic).

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Table (1): Effect of Myrrh on Total protein, globulin and Albumin of male rats with immunity inhibition

Tested Groups	Total protein (g/dl)	Globulin (mmol/l)	Albumin (g/dl)
	Mean ± SD	Mean ± SD	Mean ± SD
G1: –ve control	7.68±0.23[a]	16.67±1.37[a]	5.35±0.28[a]
G2: +ve control	4.58±0.32[e]	14.62±0.18[b]	3.68±0.13[e]
G3: 50 mg/kg protective	6.49±0.23[b]	13.85±0.49[c]	4.41±0.16[c]
G4: 100 mg/kg protective	6.45±0.17[b]	13.77±0.37[c]	4.39±0.12[c]
G5: 150 mg/kg protective	5.75±0.15[c]	12.27±0.31[d]	3.91±0.10[d]
G6: 50 mg/kg therapeutic	5.22±0.08[d]	11.13±0.18[e]	3.55±0.06[e]
G7: 100 mg/kg therapeutic	6.31±0.19[b]	13.46±0.36[c]	4.29±0.11[c]
G8: 150 mg/kg therapeutic	7.56±0.20[a]	14.69±0.42[b]	4.68±0.14[b]

Values were represented as mean + SD. Means with different superscript letters are significantly different at $p < 0.05$.

Effect of Myrrh on GPT and GOT enzymes of male rats with induced immunity inhibition

It was obvious from Table (2) group 1 (-ve control) rats has the normal value of GPT (43.17±1.30 U/l), while that in group 2 (+ve control) was 85.00±2.77 U/l which showed a significantly increasing values compared to -ve control group (1) and this due to suppressing their immunity. For rats in treated groups (3, 4, 5, 6, 7 and 8) GPT values were 40.89±1.45, 43.51±1.16, 36.88±0.94, 46.05±0.75, 40.61±1.08 and 35.76±1.03 U/l, respectively which showed significantly decreasing results compared to +ve control group (2). The best result in treated groups (35.76±1.03U/l) was seen in rats of group 8 (150 mg/kg therapeutic).

The result of GOT, showed the value of 44.67±1.37 U/l for group 1. Group 2 (+ve control) was the highly significant value (50.00±1.41) compared to group (1), (-ve control), while GOT values of rats in groups (3, 4, 5, 6, 7 and 8) were 41.84±1.49, 40.61±1.08, 31.06±0.79, 40.17±0.65, 42.54±1.13 and 34.79±1.00 (U/l), respectively, showing a significantly decreasing values compared to group (2), (+ve control). The best result in treated groups (42.54±1.13 U/l) was seen in rats of group 7 (100 mg/kg therapeutic).

Table (2): Effect of Myrrh on GPT enzyme and GOT enzyme of male rats with immunity inhibition.

Tested Groups	GPT (U/l)	GOT (U/l)
	Mean ± SD	Mean ± SD
G1: –ve control	43.17±1.30[c]	44.67±1.37[b]
G2: +ve control	85.00±2.77[a]	50.00±1.41[a]
G3: 50 mg/kg protective	40.89±1.45[d]	41.84±1.49[c]
G4: 100 mg/kg protective	43.51±1.16[c]	40.61±1.08[cd]
G5: 150 mg/kg protective	36.88±0.94[e]	31.06±0.79[f]
G6: 50 mg/kg therapeutic	46.05±0.75[b]	40.17±0.65[d]
G7: 100 mg/kg therapeutic	40.61±1.08[d]	42.54±1.13[c]
G8: 150 mg/kg therapeutic	35.76±1.03[e]	34.79±1.00[e]
F	48.5	42.3
Sig.	0.00	0.00

Values were represented as mean + SD. Means with different superscript letters are significantly different at $p < 0.05$.

Effect of Myrrh on serum Creatinine and Total bilirubin of male rats with induced immunity inhibition:

It was clear from Table (3) that the highest value (2.48±0.24 Umol/l) of serum creatinine, can be seen in group (2) rats, while it was (1.85±0.10 Umol/l) in group (1). Rats in group 2 (+ve control) showed significantly increased results compared to group 1 (-ve control). Rats in groups (3, 4, 5, 6, 7 and 8) showed the values of 1.81±0.06, 1.55±0.04, 1.65±0.04, 1.57±0.03, 1.16±0.03, and 1.16±0.03 g/dl, respectively. These values decreased significantly, compared to +ve control group (2). This means that the best result in treated groups (1.16±0.02) was seen in rats of group 7 (100 mg/kg therapeutic).

The highest value (6.83±0.46 mmol/l) of the total bilirubin (mmol/l), was in -ve control group (1), while the value (5.70±0.06 mmol/l) was seen in group (2) of rats (+ve control), which is low significantly value compared to group (1). Rats in groups (3, 4, 5, 6, 7 and 8) have the values of 4.93±0.18, 4.90±0.13, 4.37±0.11, 3.97±0.06, 4.79±0.13, and 5.23±0.15 (mmol/l), respectively and showed a decreased significantly results compared to group 2 (+ve control). This means that the lowest result in treated groups (3.97±0.06 mmol/l) was seen in rats of group 6 (50 mg/kg therapeutic).

Table (3): Effect of Myrrh on serum Creatinine and Total bilirubin of male rats with immunity inhibition.

Tested Groups	Creatinine (Umol/l)	T. bilirubin (mmol/l)
	Mean ± SD	Mean ± SD
G1: –ve control	1.85±0.10^{b}	6.83±0.46^{a}
G2: +ve control	2.48±0.24^{a}	5.70±0.06^{b}
G3: 50 mg/kg protective	1.81±0.06^{b}	4.93±0.18^{d}
G4: 100 mg/kg protective	1.55±0.04^{cd}	4.90±0.13^{d}
G5: 150 mg/kg protective	1.65±0.04^{c}	4.37±0.11^{d}
G6: 50 mg/kg therapeutic	1.57±0.03^{c}	3.97±0.06^{e}
G7: 100 mg/kg therapeutic	1.16±0.02^{e}	4.79±0.13^{d}
G8: 150 mg/kg therapeutic	1.16±0.03^{e}	5.23±0.15^{c}
F	1.61	4.34
Sig.	0.00	00.0

Values were represented as mean + SD. Means with different superscript letters are significantly different at $p < 0.05$.

Effect of Myrrh on Erythrocytes and Lymphocytes of male rats with induced immunity inhibition:

Data in Table (4) indicated that the count of erythrocytest (x^6/mcl) in rats of group (1) (-ve control) was (8.17±0.69), while the highest value (15.67±1.49) was seen in group (2) of +ve control rats, which increased significantly compared to -ve control group. Rats in groups (3, 4, 5, 6, 7 and 8) have the values of 5.48±0.19, 6.19±0.16, 6.83±0.17, 7.05±0.11, 6.34±0.17 and 7.58±0.22 (x^6/mcl), respectively showed significantly decreasing values compared to group (2) (+ve control). The best value was detected in group 8 (150 mg/kg therapeutic) which showed the value of 7.58±0.22.

The lowest value of Lymphocytes (%) was seen in group (2) 11.83±1.46%. This value showed a significant decreasing compared to group (1). Rats in groups (3, 4, 5, 6, 7 and 8) showed the values of 17.12±0.61, 19.34±0.52, 21.35±0.55, 22.05±0.36, 19.82±0.53 and 23.68±0.68 (%), respectively. These values were significantly increased when compared to group 2 (+ve control). The highest value (23.68±0.68 %) was seen in rats of group 8 (150 mg/kg therapeutic).

Table (4): Effect of Myrrh on counting of Erythrocytes and Lymphocytes

Tested Groups	Erythrocytes (x^6/mcl)	Lymphocytes (%)
	Mean ± SD	Mean ± SD
G1: –ve control	8.17±0.69^{b}	24.50±0.96^{a}
G2: +ve control	15.67±1.49^{a}	11.83±1.46^{f}
G3: 50 mg/kg protective	5.48±0.19^{f}	17.12±0.61^{e}
G4: 100 mg/kg protective	6.19±0.16^{e}	19.34±0.52^{d}
G5: 150 mg/kg protective	6.83±0.16^{d}	21.35±0.55^{c}
G6: 50 mg/kg therapeutic	7.05±0.11^{c}	22.05±0.36^{b}
G7: 100 mg/kg therapeutic	6.34±0.17^{e}	19.82±0.53^{d}
G8: 150 mg/kg therapeutic	7.58±0.22^{b}	23.68±0.68^{a}
F	9.87	19.85
Sig.	0.00	00.0

Values were represented as mean + SD. Means with different superscript letters are significantly different at $p < 0.05$.

Effect of Myrrh on immunity protein (IgG) of male rats with immunity inhibition.

The result of (IgG) in Table (5) indicated that group 1 (-ve control) showed the highest value (1835.17±24.93mg/dl), while the lowest one (1201.67± 29.30mg/dl) was seen in group (2) of +ve control rats, which decreased significantly compared to group (1). Rats in groups (3, 4, 5, 6, 7 and 8) have the values of 1312.38±46.68, 1571.10±41.86, 1768.25±45.17, 1420.76±23.12, 1377.74± 36.71 and 1662.38±47.98 mg/dl, respectively which showed significantly increasing values compared to group 2 (+ve control). The best value was detected in group 5 (150 mg/kg protective) which showed the value of 1768.25±45.17.

Table (5): Effect of Myrrh on immunity protein (IgG) of male rats with immunity inhibition.

Tested Groups	IgG (mg/dl)
	Mean ± SD
G1: –ve control	1835.17±24.93^{a}
G2: +ve control	1201.67±29.30^{h}
G3: 50 mg/kg protective	1312.38±46.68^{g}
G4: 100 mg/kg protective	1571.10±41.86^{d}
G5: 150 mg/kg protective	1768.25±45.17^{b}
G6: 50 mg/kg therapeutic	1420.76±23.12^{e}
G7: 100 mg/kg therapeutic	1377.74±36.71^{f}
G8: 150 mg/kg therapeutic	1662.38±47.98^{c}
F	1519.78
Sig.	0.00.

Values were represented as mean + SD. Means with different superscript letters are significantly different at $p < 0.05$.

Histopathological examination of the thymus:

Microscopically, thymus of rats from group 1 revealed normal structure (Fig. 1). Thymus of rats from group (2) with induced immunity inhibition showed lymphocytic necrosis with appearance of tangible body macrophages (Fig. 2).

Thymus of rats from groups (3) received 50 mg of myrrh/kg body weight and induced immunity inhibition for them at the end of experiment showed slight lymphocytic necrosis with appearance of

tangible body macrophages (Fig. 3). Whereas thymus of rats from rats of groups (4 &5) that were received 100 and 150 mg of myrrh/kg body weight, respectively and induced immunity inhibition for them at the end of experiment showed no histopathological changes (Figs. 4&5).

Thymus of rats in group (6) that were received 50 mg of myrrh/kg body weight at the beginning of experiment showed slight lymphocytic necrosis with appearance of tangible body macrophages (Fig. 6). However, thymus of rats from group (7) that were received 100 mg of myrrh/kg body weight at the beginning of experiment revealed no histopathological changes (Figs 7). However, Thymus of rats in group (8) that were received 150 mg of myrrh/kg body weight at the beginning of experiment showed slight lymphocytic necrosis with appearance of tangible body macrophages (Fig. 8).

These results indicated that thymus gland of treated rats with Myrrh in groups (4,5 & 7) compared to the control positive group showed no alteration in thymus structure which indicated that myrrh protect it from the damage which caused by immunity inhibition.

Histopathological examination of the spleen:

Microscopically, spleen of rats in group 1 (-ve) has normal structure as shown in Figure (9). However, spleen of rats in group 2 (+ve control) showed lymphocytic necrosis and depletion with appearance of tangible body macrophages (Fig. 10). Similar observation was found for rat's spleen tissue in group (3) that was received 50 mg of myrrh/kg body weight and induced immunity inhibition for them at the end of experiment (Fig. 11). While, there were slight lymphocytic necrosis, depletion and tangible body macrophages in section of rats from group (4) that were received 100 mg of myrrh/kg body weight and induced immunity inhibition for them at the end of experiment (Fig. 12). However sections of spleen in rats of group (5) that were received 150 mg of myrrh/kg body weight and induced immunity inhibition for them at the end of experiment revealed no histopathological alterations (Fig. 13).

Spleen of rats with induced immunity inhibition in group (6)) that were received 50 mg of myrrh/kg body weight at the beginning of experiment showed slight lymphocytic necrosis and depletion with appearance of tangible body macrophages (Fig. 14). On the other hand spleen of rats with induced immunity inhibition in groups (7 & 8) treated rats with Myrrh at the beginning of experiment revealed no histopathological alterations (Figs. 15 and 16).

These results mean that spleen of treated rats with Myrrh in groups (5, 7 & 8), compared to the control positive group showed no alteration in spleen tissue structure which indicated that myrrh protect spleen from the damage which caused by immunity inhibition.

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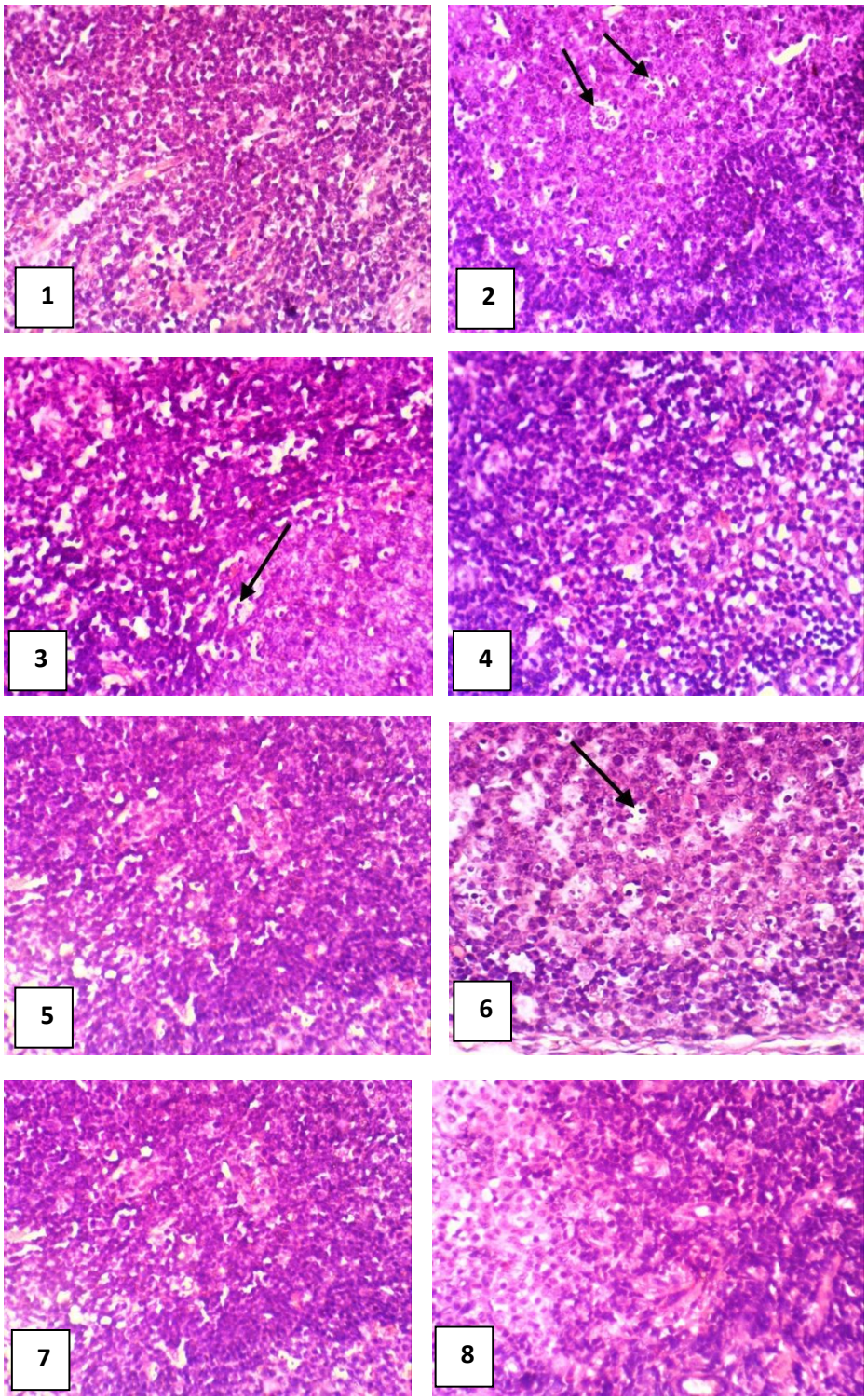
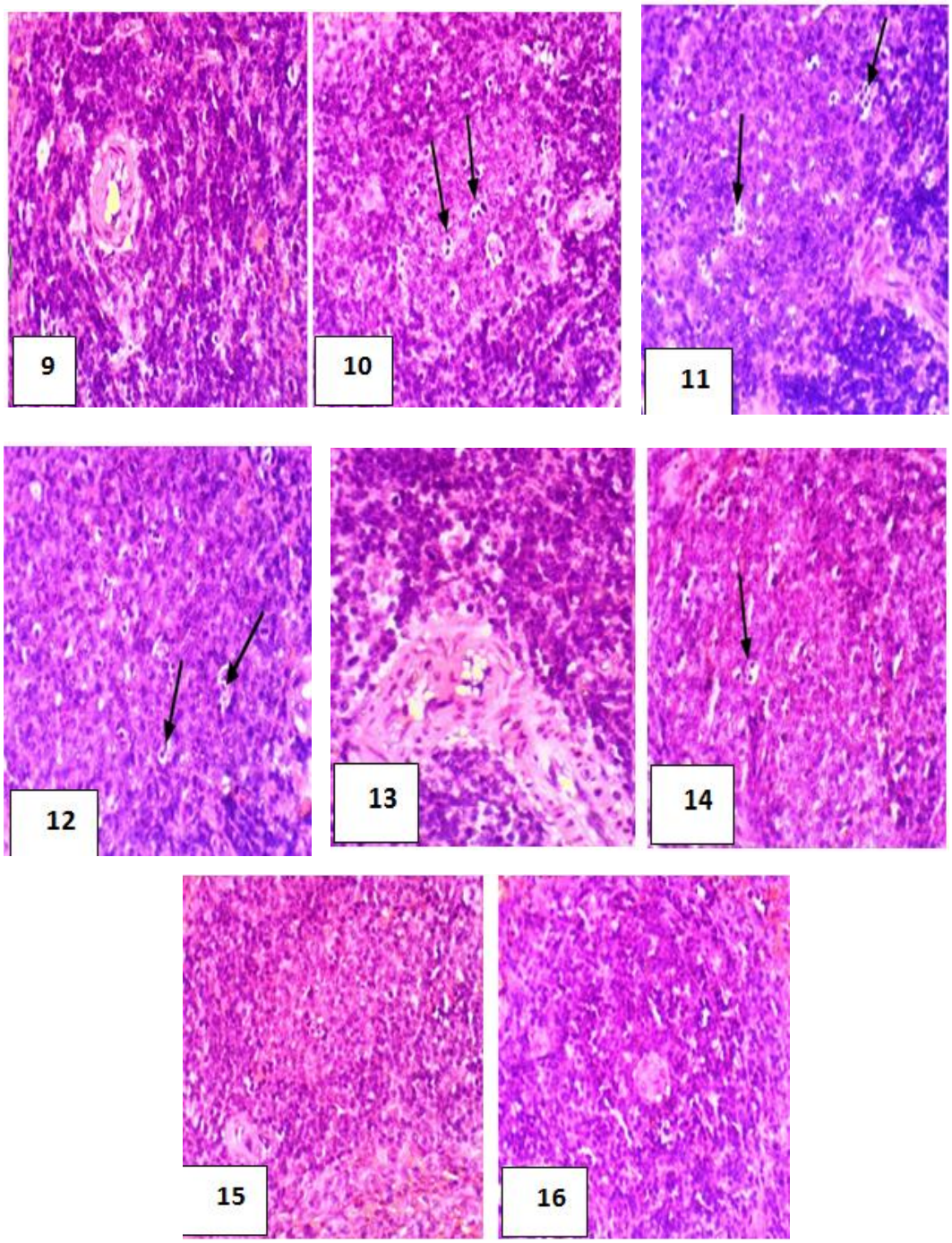


Figs. 1-8: section in thymus of rats from 8 groups, stained with H & E, X400.

Fig. 1: Group 1 showing no histopathological changes.
Fig. 2: Group 2 showing lymphocytic necrosis (arrow) with appearance of tangible body macrophages.
Fig. 3: Group 3 showing slight lymphocytic necrosis (arrow) with appearance of tangible body macrophages
Figs. 4& 5: Groups 4 & 5 showing no histopathological changes.
Fig. 6: Group 6 showing slight lymphocytic necrosis (arrow) with appearance of tangible body macrophages.
Fig. 7: Group 7 showing no histopathological changes.
Fig. 8: Group 8 showing lymphocytic necrosis with appearance of tangible body macrophages.



Figs. 9-16: Section in spleen of rats from 8 groups, stained with H & E, X400.
Fig. 9: Group 1 showing normal structure.
Figs. 10, 11: Groups 2 and 3 showing lymphocytic necrosis (arrow) and depletion with appearance of tangible body macrophages.
Fig. 12: Group 4 showing slight lymphocytic necrosis (arrow) and depletion with appearance of tangible body macrophages.
Fig. 13: Group 5 showing no histopathological alterations.
Fig. 14: Group 6 showing slight lymphocytic necrosis (arrow).
Figs. 15, 16: Groups 7 and 8 showing no histopathological changes. Note normal lymphoid follicle.

DISCUSSION

Results of the present study revealed that rats with induced immunity inhibition could be protected and treated with diet supplemented by myrrh with different levels (50, 100, 150 mg/kg), where it's had ability to enhancement immunity system as compared to unprotected and untreated rats in control positive group (+ve), this result agreed with Al-Said (2010) who revealed that myrrh can directly kill bacteria, as well as stimulate the immune system to make more white blood cells.

The observed significant increase in serum levels of total protein and albumin in the treated groups compared to (+ve) and decreased in globulin for all treated groups compared with group (-ve) in the present study can be related to an increase in the body's resistance to raising immunity. These results agreed with Maha *et al*. (2013) who showed increased levels of serum proteins and albumin in treated group by myrrh when compared with the control positive group that suffering from experimental *trichinella spiralis* infection, in addition to decrease in globulin levels.

The present investigation indicated a significant decrease in levels of serum GPT, GOT, creatinine, total bilirubin in treated rats compared to (+ve) control, this finding agreed with Khaled *et al*. (2010) who found that rabbits received supplemented diet with myrrh increased aminotransferases activity, it is a potent antioxidant, and can protect against PbAc-induced hepatic oxidative damage and immunotoxicity by reducing lipid peroxidation and enhancing the antioxidant and immune defense mechanisms.

The significant decrease in levels of serum creatinine and total bilirubin in the treated rats compared to (+ve) control can be related to the fact that administration of myrrh improved liver and kidney function, this finding agreed with that of Ashry and El-Ashmawy (2005) who found that myrrh partially ameliorates the toxicity induced by carbon tetrachloride in mice by improving ALT, AST and protein profiles. Therefore, further investigation is required to ascertain its antioxidant potential and thus determine if it can be used as an antioxidant drug. Also, Harbi *et al*. (1994) and Abeer *et al*. (2016) recorded reduced in AST, ALT, urea, and creatinine levels compared to mice in group received trichinosis in a study aimed to investigate the effects of ivermectin and myrrh obtained from the aloe-gum resin of *Commiphora molmol* on experimental trichinosis.

On the other hand, the present results indicated significant increase in lymphocytes count in all treatment groups of rats compared to (+ve) control as results of autoimmunity due to the stimulation of the drug represented in myrrh. This finding agreed with Moniuszko *et al*. (2007) and Mohamed *et al*. (2005) who recorded increasing in lymphocytes count in rats received myrrh during healing from gastric ulcer or skin injury compared to rats without treating with myrrh.

Significant increase in levels of immunoglobulin G (IgG) in treated groups compared to +ve group is an indicator that administration of myrrh produced a protective and therapeutic effect for immunity system disorders, this result was confirmed by the result of Lymphocytes count. The increase in the level of IgG was due to the body's attempt to raise the autoimmunity as a result of taking myrrh plant after suppressing the immune system, and then the level of IgG decreases again. This result was in a line with Mohamed *et al.* (2005) who observed that myrrh can improve the cellular immunity of schistosomiasis infected mice. Also, with Kadry and Mohamed (2016) who showed that IgG levels was significantly increased after treatment with

myrrh. However, IL-12 levels were significantly decreased after treatment with artesunate. The results demonstrated that artesunate or myrrh treatment could give a level of protection against *S. mansoni* infection and modulate the levels of some Th1 and Th2 cytokines in mice infected with *S. mansoni.*

Histopathological examination of the Thymus of treated rats with Myrrh, compared to unprotected rats, showed decrease in appearance of lymphocytic necrosis with appearance of tangible body macrophages and congestion of blood vessel. Whereas, decrease was detected in groups (3 & 4). These mean that myrrh with 100 & 150 mg/kg giving more therapeutic and protective effect for immunity system. This result agreed with lab results.

Histopathological examination of the spleen of treated rats with myrrh groups (5,7 &8), compared to unprotected rats, showed significant decrease in appearance of lymphocytic necrosis and depletion with appearance of tangible body macrophages. This result agreed with El-Ashmawy *et al*. (2006) who showed that myrrh protect spleen from the damage which caused by immunity inhibition.

Conclusion:

Consumption of Myrrh is very useful as its protective and has a therapeutic effect for immunity disorders where it increases the values of immunoglobulin (IgG), Lymphocytes, total protein, albumin, globulin and hemoglobin. Also, its protection against immunity disorders was important before its occurrence and helping to protect the body and raise its immune efficiency.

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التاثيرالوقائي والعلاجي لعشبة المرةعلى الجهازالمناعي المثبط في فئران التجارب

سوها محمد ابراهيم حسن[1], إقبال محمود محمد صالح[2], السيدة غندور السيد السحار[2],هالة راشد عطايا[2]
1 - قسم الصناعات الغذائية، كليةالزراعة، جامعة عين شمس
2 - قسم الاقتصاد المنزلي, كليه التربيه النوعيه , جامعه عين شمس

المستخلص
هدف البحث الى دراسة التأثيرالعلاجي والوقائي لعشبة المره ضد تثبيط المناعة الذي تم أحداثه في ذكورالفئران البالغة. تم تقسيم 48 من الفئران الي 8 مجموعات كل مجموعة 6 فئران كالاتي: المجموعة الاولي الضابطة السالبة, المجموعة الثانية الضابطة الموجبة التي تم أحداث تثبيط مناعي لها دون أخذ تدعيم بعشبة المرة, المجموعات (3, 4 , 5) تلقت عشبة المرة بنسب (50, 100, 150 ملجم/كجم) علي التولي لمدة 4 أسابيع مع أحداث تثبيط المناعة في نهاية التجربة, المجموعات (6, 7, 8) تلقت عشبة المرة بنسب (50, 100, 150 ملجم/كجم) علي التوالي لمده أربعه أسابيع مع أحداث تثبيط المناعة في بداية التجربة. أظهرت النتائج أن تغذية الفئران علي عشبة المره في المجموعات التي تم أحداث الاصابة لها في نهاية التجربة (المجموعات الوقائية 3, 4, 5) بنسب (50, 100, 150 ملجم/كجم) علي التوالي وكذلك المجموعات التي تم أحداث أصابة لها مع بداية التجربة (المحموعات المعالجه 6, 7, 8) بنسب (50, 100, 150 ملجم/كجم) علي التوالي زياده معنوية في مستويات بروتين المناعة (IgG) عند مقارنتها بالفئران غيرالمعالجة (المجموعة الثانيه). كما زادت الخلايا الليمفاوية والبروتين الكلي والألبومين بشكل ملحوظ مقارنة بالمجموعه الثانيه الضابطه الموجبه، كما أظهرت التحاليل البيوكيميائية حدوث أنخفاض معنوي في مستويات الجلوبيولين ,الكرياتينين,البيليروبين, أنزيمات الكبد (GPT, GOT) وكريات الدم الحمراء مقارنه بالمجموعه الثانيه. وكانت أفضل النتائج في المجموعه الخامسه (الوقائيه) التي تناولت 150 ملجم/كجم من المره حيث أظهرت كفاءه الجهاز المناعي بمعدل أكبر . وأظهرالفحص الهستوباثولوجي للغدة الزعترية في الفئران التي تناولت عشبة المره انخفاضًا ملحوظًا في نخر الخلايا اللمفاوية وأحتقان الأوعية الدموية مقارنة بفئران (المجموعة الضابطة الموجبة). كماأظهرت نتائج الفحص الهستوباثولوجي للطحال أنخفاضًا في نخر الخلايا اللمفاوية مقارنةً بفئران (المجموعة الضابطة الموجبة). الخلاصه:أن تناول عشبة المره مفيد للغاية حيث أن له تأثير وقائي وعلاجي يعزز جهاز المناعه و الوقايه من المرض من البدايه أمر هام يحمي الجسم ويرفع كفاءه الجهاز المناعي.

الكلمات المفتاحية: المرة – الجهاز المناعي – بروتينات المناعة- فئران التجارب