

Evaluation of the Activity of Essential Oil of *Origanum majorana* as an Antibacterial Agent in Vivo And Detect its Synergism Effect on Liver, Kidney, and Spleen Tissues in a Male of Rats.

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ABSTRACT

The present study was done in the Kufa University / Science College from aimed to investigate the potential effect of marjoram oil as an anti-bacterial agent for *Staphylococcus aureus* bacteria and study the synergism action on the vital organs which include (liver, kidney, and spleen) in male rats. twelve adult male rats were divided into four groups include three rats in every group:

Group 1: processed with normal saline for a two week.

Group 2: processed with (0.1) bacterial suspension.

Group 3: processed with (0.1) bacterial suspension and after one day processed with essential oil of *O. majorana* (0.16ml /kg) for a two week.

Group 4: processed with essential oil of *O. majorana* (0.16ml / kg) for a two week.

The histological inspection of our study show histopathological changes on G2 that treated with bacterial suspension compared with control group (G1) represented by granuloma like lesion, aggregation of lymphocyte in Liver, degeneration in the kidney tubule, fibrosis and Spleen white pulp hyperplasia, moreover the biochemical assay showed large change in the value of ALP levels (alkaline phosphatase), urea, creatinine, and ASAT level (aspartate aminotransferase).

In the G3 that treated with bacteria and oil was shows focal degeneration of portal triad, inflammatory cell and apoptotic bodies in liver, whereas the Kidney demonstrated the hyaline cast (pyelonephritis) while there was no any changes in the spleen and the biochemical study reported that little changes in the value of ALP levels, urea, creatinine and ASAT levels. As for G4 that treated with oil only, our results demonstrated normal effects in the Kidney, liver hepatocyte and tissue of spleen, and showed no any changes in value of (ALP), urea, Creatinine, (ASAT) and (ALAT).

Keywords

O. majorana: oil extraction, oil as antibacterial agent

Introduction

Medical plant considers the good source for antibiotic because it has antibacterial activity and safety and cheap and available. *Origanum majorana* one of the medical plants it is a member of the Lamiaceae family, an aromatic plant of great economic and industrial importance. It is known since antiquity for its therapeutic properties¹.

Volatile extract of marjoram is used in pharmacology, medicine, clinical The essential oil obtained from the flowering heads of marjoram has an aromatic smell and contain a high percentage of polyphenols and monoterpenes which are established antioxidants in microbiology, pathology, and food preservation, marjoram essential oil could protect liver and kidney². Several researcher was reported marjoram's antibacterial and antiviral properties³.

In recent years, sweet marjoram (*Origanum majorana*) leaves have antimicrobial and emmenagogue properties and be useful for the treatment of respiratory and gastrointestinal problems⁴.

The essential oil of the plant has been used for pains, gastrointestinal problems, and respiratory

tract disorders^{5,6}. Rosmarinic acid, sinapic acid, vanillic acid, ferulic acid, caffeic acid, syringic acid, p- and m-hydroxybenzoic acid, and coumarinic acid have been identified in the essential oil of sweet marjoram⁷. The essential oil and water extract of sweet marjoram have antimicrobial activity against bacteria and yeasts and the essential oil was higher effect than water extract⁸. cis-Sabinene hydrate in the essential oil of sweet marjoram has been claimed to be responsible for antibacterial effect⁹.

Materials and method

This study used essential oil of *Origanum majorana* which obtained from a local market in Najaf city.

Animals used in the experiment

Our study used 12 rats (male) rats' weight between (150-200 g). The rats were kept under normal healthy laboratory conditions; the temperature was adjusted at 25 ± 2 °C and 12-hour light-dark. animals were adapted on free access of water, and fed for one-week basal diet before the initiation of the experiment.

Experimental design

Sample from the liver, spleen, kidneys were collected from rats in all groups at the end of experiments (30 days), fixed in 10% formalin, then dehydrated in alcohol with deferent concentration, cleared in xylene and embedded in paraffin. Then sectioning with microtome and stained with Hematoxyline and Eosin¹⁰.

Histopathological examination

At the end of experimental period all processed groups were anesthetized, using a mixture of ketamine and xylazine i.m., and then they were sacrificed. Liver, spleen and kidneys were eradicated from rats in all groups after (30 days.) Fixed in formalin 10%, then dried in alcohol in the deferent concentration, cleared in xylene and embedded in paraffin, then sectioning with microtome and stained with Hematoxylin and Eosin¹⁰.

Study the influence of essential oil of *O. majorana* on the rats.

twelve adult male rats were divided into four groups include three rats in every group:

Group 1: processed with normal saline for a two week.

Group 2: processed with 0.1ml from suspension of bacteria .

Group 3: processed with 0.1ml from suspension of bacteria and after one day processed with essential oil of *O. majorana* (0.16ml /kg)¹¹ for a two week.

Group 4: processed with essential oil of *O. majorana* (0.16ml/ kg)¹¹ for a two week.

Results and Discussion

From this study we showed increasing in value of alkaline phosphatase (ALP) in the group processed with bacteria suspension and reach to 616 (U/L) comparisons with the control group which was 390 (U/L) further, our results showed little height in the value of ALP in the group processed with (0.1ml) of bacteria suspension and Marjoram Oil (0.16ml /kg) that was reached to 406 (U/L) in comparisons with group that processed with bacteria suspension only, this finding was agreement with^{12,13} who showed to the harmful roles of bacteria suspension in liver organ .

For the aspartate aminotransferase (AST), all groups were near to monitoring group excepting the group deal with bacteria suspension showed increasing in value of AST and reached to 250 (U/L) comparisons with the control group which was 216 (U/L) this attributed to the effect of bacteria on the liver which caused in released the AST into the bloodstream and led to raise in the

level of enzyme ¹⁴. These results agreed with ¹⁵ who showed that the beginning of bacterial infections was correlated with the appearance of severe complexity such as hepatic impairment, organ failures and acute kidney injury, Infections of bacteria elevated of mortality rates in any phase of the liver illness also in chronic liver failure.

whereas the Alanine transaminase (ALT) in all treatments were near to control group that were reached to (209, 215 and 211 U/L) respectively comparison with the control group which was (209 U/L).

For urea , all groups were near to the monitoring group while group that processed with bacteria suspension showed decreasing in value of urea and reach to 63(mg/dl) comparisons with the control group which was 78(mg/dl) this decreasing may attribute to it is largely confined to advanced liver disease ¹⁶ and the liver of this group has many problems because of infection with bacteria (table 1).

For creatinine, all treatments were near to monitoring group while the group processed with bacteria showed decreasing in value of creatinine and reach to 60 (mg/dl) comparisons with the control group which was 70 (mg/dl) this decreasing may attribute the synthesis of creatinine occurs in the kidney with amino acid arginine and glycine then, this product is add to the methyl group from methionine after transferred to the liver to forming creatinine. ^{17,18} or attributed to the creatinine capable to destroyed variety species of bacteria either Gram-positive and negative wall, moreover to the variety of antibiotic-resistant bacterial strains ¹⁹.

Table1: The effect of different treatment groups in biochemistry assay

Parameter treatment	ALP (U/L)	Urea (mg/dl)	Creatinine (mg/dl)	AST (U/L)	ALT (U/L)
Control group	390	78	0.70	216	209
processed with bacteria	616	63	0.60	250	215
processed with bacteria +oil	406	73	0.68	225	211
processed with oil	390	75	0.72	218	209

The result of histological study shows that there was no histological changes in the group1 which represent the control group processed with normal saline only. (Fig1) Which shows normal histological feature liver like normal hepatocyte, central vein, portal vein, and hepatic cord moreover the kidney shows normal proximal and distal convoluted tubule and normal glomerular, the spleen shows normal white and red pulp.

The current study shows the sever changes in group which with processed bacterial suspension(Fig2). include liver granuloma like lesion, aggregation of lymphocyte, kidney degeneration in the kidney tubule and spleen white pulp hyperplasia this results agreement with ²⁰ who indicated to the toxic role action of α -toxin produced by S.aureus, As the rapid depletion of adenosine triphosphate (ATP) , these toxin lead to release of TNF- α from monocyte , IL-I β and induce cardiac depression in sepsis, Degeneration in some organs were seen in liver, kidney , stomach, probably due to cytotoxic effects of S.aureus products , furthermore ²¹ showed to the harmful role of Staphylococcus aureus (S. aureus) that manufacture many species of toxins which

causes Varity diseases, which range from moderate infections in the skin to systemic, life-threatening diseases.

O.vulgare species has many effectiveness associated to medical ,within the properties related to medical importance are the following , kidney and liver diseases, metabolic, hormonal and neuronal disorders ²².

These plants have antibacterial activities which give them a very important role not only for treating infectious bacterial diseases ²³.

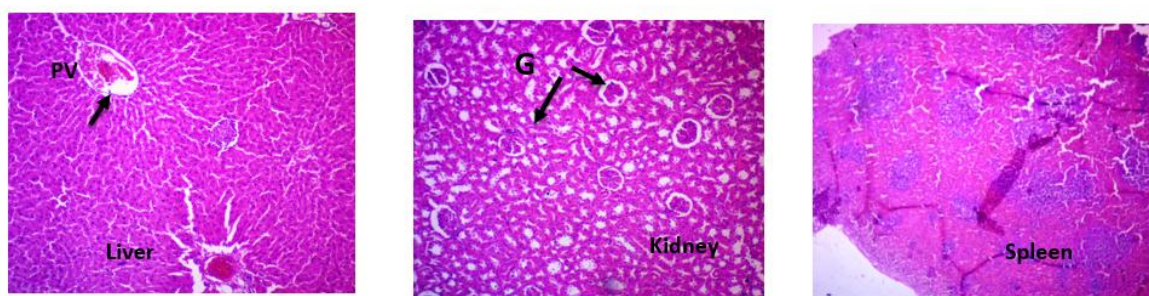


Figure 1: show control groups, normal histological feature of liver, (PV) portal vein, normal proximal tubule, distal tubule, glomeruli of the kidney, normal white and red pulp of Spleen.

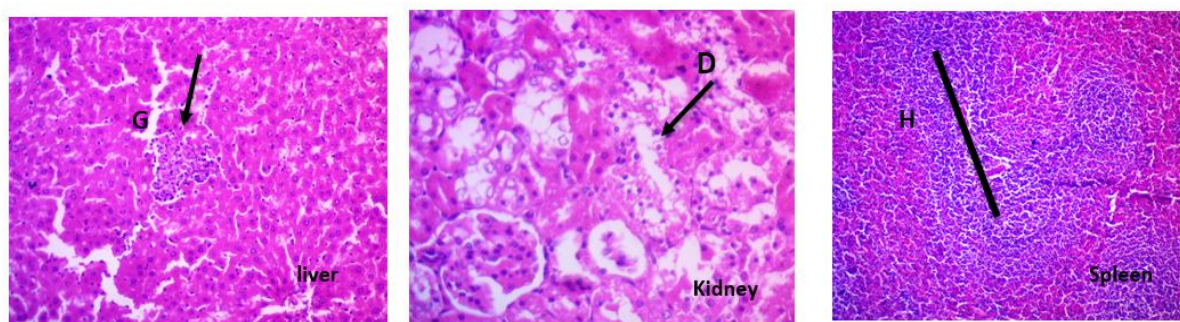


Figure 2: show the treated groups with bacterial suspension only. the section of liver shows that granuloma like lesion, aggregation of lymphocyte. degeneration in Kidney tubule, white pulp hyperplasia of Spleen

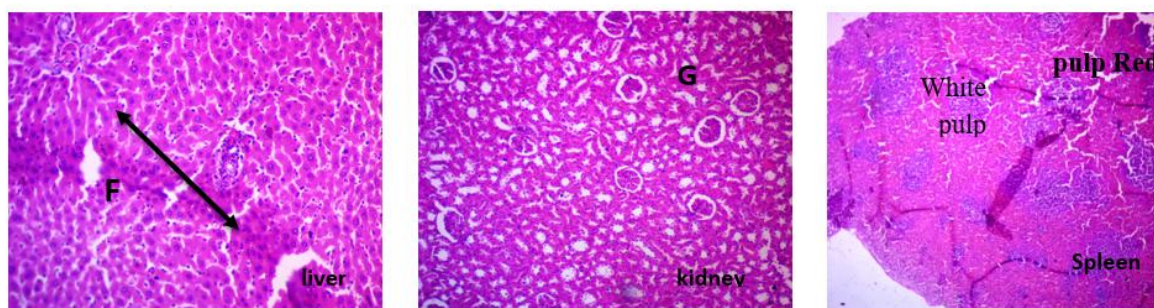


Figure 3: the treated groups with oil only show, normal kidney, focal aggregation in the liver while no changes appear in the Spleen

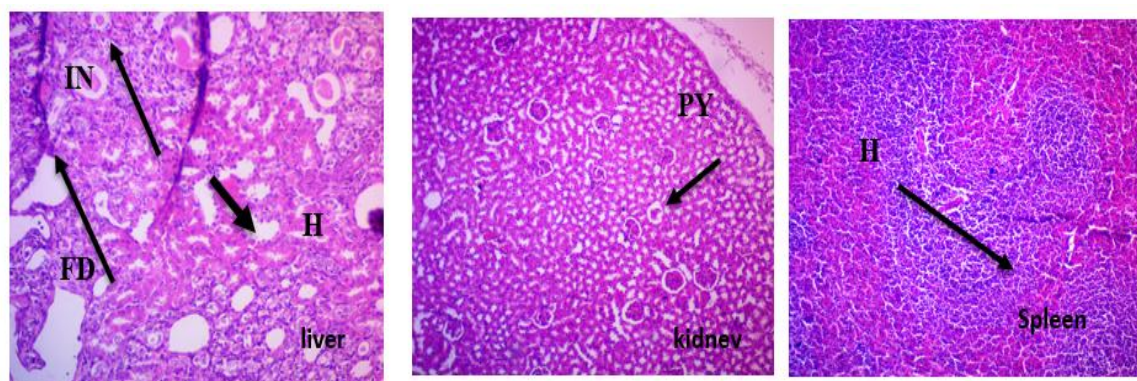


Figure 4: the treated groups with bacterial suspension and oil. shows focal degeneration of portyriad , inflammatory cell m apoptotic bodies of the liver. Haylincastpyelonephritis , fibrosis in the kidney . Spleen show white pulp hyperplasia.

Conclusion

The present study revealed the synergism effect of *O.majorana*oil for vital organ against bacterial infection furthermore this study shows the sever changes in liver histology represented by liver granuloma like lesion, aggregation of lymphocyte, kidney degeneration and spleen white pulp hyperplasia.

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