

Antioxidant, Antibiofilm & Biological Activities of Summer Savory

***Samar H.M. Mohamed^{1*}, Ihab S. Ashoush¹,
Hesham M. Elhariry¹, Eshak M. El-Hadidy²***

¹Food Science Department, Faculty of Agricultural,
Ain Shams University, Cairo, Egypt.

²Food Technology Res. Inst., Agricultural Research Center, Giza.

Abstract

The Hepatic disease all over the world had a more attention towards the prevention methods; planned diet can effective and protective. Therefore, this study was aimed to examine the antioxidant and hepatoprotective role of the dried savory leaves (1 and 2%) and its dried hydroethanolic extract (25 and 50 ppm) against carbon tetrachloride (CCl₄) induced liver damage in rats. Also, the effect of savory leaves extract on biofilm formation of some microbial strains was studied. Liver injury group revealed significant increase in serum liver parameters such as AST, ALT and ALP activities and malondialdehyde (MDA) levels. Whereas, reduction in superoxide dismutase (SOD) activity was done. Furthermore, the liver injury rats recorded hepatic histological several and severe alteration due to hepatocellular damage by CCl₄. Treatments by the dried savory leaves and its extract at different levels resulted significantly improved the liver function and oxidative stress parameters; also, suppresses the histological alteration in liver. Both Gram-positive and Gram-negative bacteria have the ability to form biofilm with varying degrees. The savory ethanolic extract has an antibiofilm effect against the studied bacteria strains. Also, savory leaves extract exhibited scavenging activity due to their containing from phenolic

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and flavonoids, responsible for their antioxidant, antibiofilm and hepatoprotective properties. It could be concluded that the dried savory leaves and their extracts could be used as ingredients in functional foods for hepatoprotective.

Keywords: Summer Savory; Antioxidants; Antibiofilm;
Hepatoprotective

Introduction

Savory(*Satureja hortensis* L.), commonly known as summer savory or garden savory, belongs to family *Lamiaceae*. It is also well known traditional medicine as a remedy for various diseases and have been used for centuries as culinary herbs and spices (**Zargari, 1990**). Recently, summer savory had many properties like, antimicrobial, antioxidant, antihyperglycemia, antihyperlipidemic and protecting the body against oxidative stress effect. Plant leaves are widely used in food as a spice and flavor component **Exarchou et al., (2002); Hajhashemi et al., (2002) and Gulluce et al., (2003)**. The pharmacological examination, extracts of savory have been reported to have antibacterial, antifungal, antioxidant, hepatoprotective and anti-inflammatory effects according to many literature studies like that **Sahin et al., (2003); Mosaffa et al., (2006); Mahboubi and Kazempour (2011); Panjehkeh and Jahani (2011) and Hajhashemi et al., (2012)**.

The extract obtained from savory leaves represents the subject of significantly fewer studies, when compared with the essential oils; the composition of the summer savory extracts revealed a variation of the total phenolic and total flavonoids content (**Boroja et al., 2018**).

The aim of our study was to examine the potential ameliorating effect of dried summer savory leaves and its dried hydroethanolic extract against CCl₄-induced oxidative damage in hepatic tissues. Antioxidants, antimicrobial and antibiofilm properties of the savory leaves extracts have been also carried out.

Materials and Methods

Materials:

The dry ground leaves of the summer savory plant (*Satureja hortensis*) has been obtained from Horticultural Research Institute, Agricultural Research Center, Giza. Whereas, the carbon tetrachloride (CCl₄), ethanol, methanol, aluminum chloride and sodium carbonate were obtained from El-Gomhoreya Chemical Company, Cairo, Egypt. Meanwhile, Folin-Ciocalteu phenol reagent and 2,2-diphenyl-2-picrylhydrazyl radical (DPPH) were purchased from Sigma–Aldrich Inc. (St Louis, MO, USA). While, Commercial kits used for determining the activity of alanine aminotransferase (ALT); aspartate aminotransferase (AST); alkaline phosphatases (ALP); malondialdehyde (MDA) and superoxide dismutase (SOD) were purchased from Biodiagnostic Company Dokki, Egypt.

Animals:

Thirty six (36) male albino Wistar rats weighing an average 150±10 g were obtained from the Animal Experimental Unit in Food Technology Research Institute, Agricultural Research Center.

Preparation of dried hydroethanolic extract of savory leaves:

Savory leaves were washed in water several times to remove any adhering parts, dried in oven under vacuum, then ground well. Ground savory was mixed with 80% ethanol (1:100 w/v) and stored in dark bottle for 48h/4°C. The mixtures were filtered through filter paper (Whatman No. 1). Hydroethanolic solution was evaporated in rotary

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evaporator at 50°C and dried savory extract was collected. Different savory extracts were prepared from the dried leaves using different concentrations (25 and 50 ppm) for biological experiments (*El-Hadidy et al., 2018*).

DPPH radical scavenging activity:

Electron-donating ability of savory leaves extract was determined by implying DPPH radical-scavenging assay as described by (*Brand-Williams et al., 1995*). Aliquots 1 ml of sample extract was mixed with 1 ml of 0.2 mM DPPH in methanol. The control sample contained all the reagents except the extract. The reaction mixture was shaken well and allowed to react for 30 min at room temperature. The remaining DPPH free radical was determined by absorbance (A) measurement at 517 nm against methanol blanks.

The percentage scavenging effect was calculated from the decreased in absorbance against control according to the following equation: Scavenging activity (%) = $[(A_{\text{control}} - A_{\text{sample}}) / A_{\text{control}}] \times 100$

Determination of total phenolic content (TPC):

The total phenolic content of the extract was determined colorimetrically, using the Folin-Ciocalteu method, as described by (*Singleton et al., 1999*). Aliquots of 0.5 ml of each extract were added to 0.5 ml of Folin-Ciocalteu reagent, followed by addition of 0.5 ml of an aqueous solution (7.5%) of sodium carbonate. The mixture was stirred and allowed to stand for 30 min. With absorbance at 765 nm, while blank sample consisting of water and reagents was used as a reference.

The results were expressed as milligrams of gallic acid equivalents per ml extract (mg GAE/g) by reference to the gallic acid calibration curve.

Assay of total flavonoid content (TFC):

The total flavonoids content was determined according to the method of **(Mohdaly et al., 2012)**. A 100 µL aliquot of 2% aluminum chloride (AlCl₃) ethanolic solution was added to 100 µL of the extract, then mixed well. After keeping for 1 h at room temperature, the absorbance (A) at 420 nm was measured. A yellow color indicates the presence of flavonoids. The total flavonoids content was expressed as milligram quercetin equivalent (QE).

Savory ethanolic extract as antimicrobial and antibiofilm agent:

Antimicrobial activity

For examining the effect of the savory dried ethanolic extract (crude extract 30 mg/ml) on growth of some pathogenic bacteria, include Gram negative (*Campylobacter jejuni*; *Salmonella typhimurium* and *E.coli* 0157) and Gram positive (*Staphylococcus aureus*; *Bacillus cereus*; *Enterococcus faecalis*; *Listeria monocytogenes* and *Bacillus subtilis*) the 96-well plate was applied. The plates were incubated at 37 °C for 24 h. Absorption (A) was measured at 600 nm **(Ceruso et al., 2020)**.

Bacterial strains used in this study included strains (*Bacillus cereus* DSMZ 345, *Escherichia coli* O157:H7 ATCC 51659, *Salmonella typhimurium* ATCC 14028, *Staphylococcus aureus* ATCC 6528, *Campylobacter jejuni* ATCC 33250, *Enterococcus faecalis*, *Listeria monocytogenes* ATCC 19115 and *Bacillus subtilis*), were obtained from the Egyptian Microbial Culture Collection (EMCC) at the Microbial Resource Center (Cairo, MIRCEN), Faculty of Agriculture, Ain Shams University, Cairo, Egypt.

Antibiofilm effect

Inhibitory effects of savory dried ethanolic extracts on bacterial biofilm formation were determined *in vitro* using the commonly used 96-well polystyrene microtiter plate's method. After incubation at 37 °C for 24 h, content of the microtiter plates was

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poured off, and the wells were washed three times with phosphate-buffered saline (PBS, pH 7.2). The remaining adhered bacteria were fixed with 250 μ L of methanol per well. After 15 min, microliter plates were emptied and air-dried. The microtiter plates were stained with 250 μ L per well of 1% crystal violet used for Gram staining for 5 min. The excess of stain was rinsed off by placing the microliter plates under running tap water. After drying the microtiter plates, the dye bound to the adherent cells was extracted with 250 μ L of 33% (v/v) glacial acetic acid per well. The absorbance (A) of each was measured at 570 nm using a spectrophotometer as described and modified by (**Elhariry et al., 2014**).

Biological experiment design:

The experiment was conducted on thirty six male Albino Wistar rats; they were housed in special cages under controlled conditions. The animals were fed on basal diet according to AIN-93 guidelines (**Reeves et al., 1993**) and were provided with water *ad libitum* during the experimental period.

The rats were randomly divided into six groups with six rats in each group. Group one was reserved as normal control (NC). Groups from two to six, rats were administrated intraperitoneal (IP) injection with repeated dose of 2 ml/kg body weight by mixture of (1:1 v/v CCl₄/paraffin oil) according to (**Malgorzata et al., 2009**). Group two kept as injury control (IC); each rat in group three and four received basal diets in which starch was replaced with 1 and 2 % dried savory leaves. While, group five and six received its weight corresponding dosage from dried savory leaves extract at 25 and 50 ppm administered oral by gavage in a dosage of one ml/100 g body weight per day for successive 28 days as reported by (**Rouanet et al., 2010**).

The blood samples were taken then serum was separated and kept at -18°C until the biochemical analyses; the liver was taken immediately after anatomy for histopathological examination. The experiment was carried out in accordance with the Agricultural Research Center guidelines for animal experimentations; the experimental protocol was approved by the Ethical Committee at Agricultural Research Center.

Biochemical investigation:

Liver biomarker enzymes:

The activities of serum alanine aminotransferase (ALT) and aspartate aminotransferase (AST) were determined according to *(Reitman and Frankel, 1957)*. While, the activity of alkaline phosphatase (ALP) was determined according to *(GSCC, 1972)*.

Oxidative stress parameter:

The extent of lipid peroxidation in the liver was determined according to *(Ohkawa et al., 1979)*. Superoxide dismutase (SOD) was assayed according to *(Kakkar et al., 1984)*.

Histopathological Examination:

The liver of the different groups of rats was excised after sacrificed and kept in 10% buffered formalin then sectioned and embedded in paraffin. Slides were stained with hematoxylin and eosin according to *(Banchroft et al., 1996)*.

Statistical analysis:

Descriptive values of data were expressed as the Mean±SE and they were applied to the (ANOVA); followed by Duncan's test. In all cases $p < 0.05$ was used as the criterion of statistical significance by SAS program *(SAS, 2003)*.

Results and Discussion

Antioxidant power of dried savory leaves extract:

From the results presented in **Table (1)**, it could be noticed that the savory plant extract is a good source of total phenols and flavonoids and had a great free radical scavenging activity. These results are in agreement with those of (**Pländer et al., 2012**) who found that flavonoids of savory plant ranges from 1.37 - 7.09 % which enhances the antioxidant activity. Also, (**Vabkova and Neugebauerova, 2012**) found great content of total phenolic and flavonoids in fresh savory plant in the first harvest.

Savory hydroethanolic extracts as antimicrobial agent:

The effect of savory ethanolic extract (SEE) on the growth of some bacteria from Gram-positive (**G+ve**) and Gram-negative (**G-ve**) bacteria was studied using the microtiter plate method. In the presence of the SEE, there were significant increases in the growth of Gram-positive bacteria except for *Listeria monocytogenes* (**Table 2**). Moreover, all studied Gram-negative bacteria showed significant increase in their growth when the SEE was added to the culture medium at concentration of 30 mg/ mL (**Table 2**). The present result indicated that the SEE prepared in the current study did not have inhibitory effect on the growth of the planktonic bacterial cells of the studied strains. This finding was in agreement with those mentioned by (**Popovici et al., 2019**) who mentioned that SEE did not showed antimicrobial activity agent against G-ve (*E. coli* and *P. aeruginosa*) or G+ve (*S. aureus* and *B. cereus*). Moreover, (**Akin et al., 2022**) found the same finding as reported in their study that SEE had no antimicrobial activity agent for G-ve (*E. coli*). These results may be because the study was conducted on the dried savory leaves without the essential oil.

Antibiofilm effect of dried savory leaves extract:

The ability of all tested microbial strains to form biofilm was also estimated using the standard method for studying biofilm formation in the absence and presence of prepared savory ethanolic extract. In the absence of savory ethanolic extract, all tested bacterial strains were strong biofilm producers (+++) except for *Listeria monocytogenes*, *Staphylococcus aureus* and *Enterococcus faecalis* (moderate biofilm producer, (++)), on the otherhand both of *B. subtilis* and *B. cereus* (non-biofilm producers, (0)). Where adding savory ethanolic extract to the culture medium it could be noticed a significant decrease in the ability of tested bacterial strains to form biofilm except for *Listeria monocytogenes* and *Staphylococcus aureus* which became strong biofilm producer, *B. subtilis* and *B. cereus* still non-biofilm producers (Table 3). The results obtained in the present study indicated that the savory ethanolic extract has an antibiofilm effect against the studied strains. This effect may be due to the content of savory ethanolic extract different bioactive compounds such as phenolices and isoflavones as reported by (Huttunen *et al.*, 2011). These components are exerted a dual extract on biofilm formation either stimulating or inhibiting, depending on the concentration (Plyuta *et al.*, 2013). At higher concentrations the biofilm formation was suppressed, throughout reducing the adhesion of bacterial cells to the surfaces.

Effects of summer savory leaves treatments on hepatic functions parameters:

The liver is a major target organ for toxicity from hazardous xenobiotics most ingested orally. The present study was focused on investigating the role of dried savory leaves and extract against CCl₄ induced hepatotoxicity and to find its possible mode of action in hepatoprotection.

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Rats subjected to CCl₄ developed significant hepatocellular damage as evident from the serum activities of ALT, AST and ALP compared to normal values, and which have been used as reliable markers of hepatotoxicity (**Table 4**). Supplemented diet with savory plant leaves powder (1 and 2%) and administration extract (25 and 50 ppm) exhibited a significant reduction in the activities of ALT, AST and ALP as compared with liver injury control rats group. This was also confirmed by (**Boroja et al., 2018**) results who demonstrated that the level of biochemical parameters of intoxicated rats liver decreased compared to the injury control when using 100 mg / kg body weight of the savory extracts.

Effects of summer savory treatment on oxidative stress biomarkers:

Increased lipid peroxidation is generally believed to be an important underlying cause of the initiation of oxidative stress related to various tissue injury, cell death, and further progression of many acute and chronic diseases (**Halliwell and Gutteridge, 1999**).

The results in **Table (5)** depicted that changes in the levels of serum malondialdehyde (MDA) as an indication of lipid oxidation in rats groups. Supplementation with savory plant leaves powder and administration extract decreased the levels of intoxicated rat's serum MDA comparable to liver injury control group. The significant reduction in the levels of MDA confirms that savory plant leaves powder and its extract could effectively protect against free radicals induced by CCl₄. In similarity with our results, that was reported that administration of *Savory hortensis* extracts effectively reduced stress conditions and enhanced fish health status (**Rudiansyah et al., 2022**).

Superoxide dismutase (SOD) is the major antioxidant defense systems against hydrogen peroxide and hydroperoxides. Changes in (SOD) activity of different groups has been shown in **Table (5)**, rats treated with CCl₄ alone significantly lowered the serum superoxide dismutase (SOD) activity. While, supplementation by savory plant leaves powder and administration extract improved the SOD activity in the serum. Such beneficial effects of savory plant have been reported by (**Boroja et al., 2018**)who demonstrated that the savory extract increasing the SOD activity level in liver tissues.

Histopathological examination:

Liver injury was evaluated by histopathological alterations finding in **Micrograph (1)** to **(6)**. Microscopic examinations of liver for the group one were given in (**Micrograph 1**).The pictures in the plates revealed the normal histological structure of hepatic lobules, normal central vein, portal triad and hepatocytes.

Oncontrary, liver of rats from group two exhibited severe histopathological changes described by fibroplasia in the portal triad, oval cells proliferation and portal infiltration with inflammatory cells, as well as focal hepatocellular necrosis associated with inflammatory cells infiltration (**Micrograph 2**).

However, liver of rats from group three revealed slight activation of Kupffer cells, slight proliferation of oval cells in the portal triad. (**Micrograph 3**).

Moreover, improved alteration in hepatocytes was noticed in liver of rats from group four, examined sections showed slight activation of Kupffer cells, congestion of hepatoportal blood vessel and slight proliferation of oval cells in the portal triad (**Micrograph 4**).

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Some examined sections from group five revealed slight activation of Kupffer cells and apparent normal portal triad, whereas, other sections showed slight proliferation of oval cells associated with inflammatory cells infiltration in the portal triad (**Micrograph 5**).

Moreover, some sections from group six showed slight activation of Kupffer cells, whereas, other sections from this group described fibroplasia in the portal triad, hyperplasia of biliary epithelium associated with formation of newly formed bile ductules (**Micrograph 6**).

The literature studies for histological changes for savory leaves as hepatoprotective agents was fewer. Therefore, these findings supported the hepatoprotective effect of incorporated diet with savory plant leaf which retained to near normal histological structure.

Conclusion

Based on the above results, it could be concluded that summer savory (*Satureja hortensis*) leaves exhibited a potential antioxidant and antibiofilm properties. Also, the attenuating effect of *S. hortensis* against CCl₄-induced oxidative damage in hepatic tissues by improving serum liver function, oxidative biomarkers and restore the histological alteration of the liver.

Table (1):
Antioxidants contents of dried savory leaves
extract

| Parameters | (On dry weight basis) |
|-----------------------------------|-----------------------|
| Total Phenols mg GAE/g extract | 26.04 ± 0.251 |
| Total Flavonoids mg QE /g extract | 4.297 ± 0.059 |
| Scavenging activity % | 98.01 ± 0.015 |

Data are mean ± SE, n=3

Table (2):
Effect of dried savory leaves extract on growth of some pathogenic bacteria

| | Bacterial Strain | NoE | Se | Growth of some pathogenic bacteria | |
|--------|------------------------|------------|------------|------------------------------------|-------------------------------|
| | | | | Control | SamplesCrude extract 30 mg/ml |
| (G-ve) | Campylobacter jejuni | 0.26±0.001 | 0.56±0.002 | +++ | +++ |
| | Salmonella typhimurium | 0.01±0.002 | 0.58±0.003 | 0 | +++ |
| | E.coli 0157 | 0.03±0.001 | 0.52±0.003 | + | +++ |
| (G+ve) | Staphylococcus aureus | 0.03±0.002 | 0.51±0.002 | + | +++ |
| | Bacillus cereus | 0.24±0.001 | 0.39±0.001 | +++ | +++ |
| | Enterococcus faecalis | 0.28±0.002 | 0.50±0.002 | +++ | +++ |
| | listeria monocytogenes | 0.50±0.002 | 0.48±0.002 | +++ | +++ |
| | Bacillus subtilis | 0.22±0.001 | 1.39±0.007 | +++ | +++ |

Data are mean ± SE, n=3.

Strains were classified as no growth producer (0), weak growth producer (+), moderate growth producer (++) or strong growth producer (+++); n = 3.

NoE: Growth in the absence of savory extract, Se: Growth in the presence of savory extract.

Table (3):

Antibiofilm effect of dried hydroethanolic savory leaves extract

| | Bacterial Strain | Blank | NoE | Se | Biofilm formation | |
|--------|------------------------|-------|------------|-------------|-------------------|------------------------------|
| | | | | | Control | SamplesCrude Extract30 mg/ml |
| (G+ve) | Campylobacter jejuni | 0.03 | 0.25±0.001 | 0.02±0.004 | +++ | 0 |
| | Salmonella typhimurium | 0.03 | 1.36±0.005 | 0.19±0.012 | +++ | +++ |
| | E.coli 0157 | 0.03 | 1.99±2.719 | 0.04±0.024 | +++ | + |
| (G+tv) | Staphylococcus aureus | 0.04 | 0.12±0.000 | 0.88±0.048 | ++ | +++ |
| | Bacillus cereus | 0.04 | 0.01±0.001 | 0.002±0.001 | 0 | 0 |
| | Enterococcus faecalis | 0.04 | 0.10±0.001 | 0.03±0.001 | ++ | 0 |
| | Listeria monocytogenes | 0.04 | 0.11±0.001 | 0.17±0.001 | ++ | +++ |
| | Bacillus subtilis | 0.04 | 0.02±0.001 | 0.04±0.002 | 0 | 0 |

Data are mean ± SE, n=3.

Strains were classified as no biofilm producer (0), weak biofilm producer (+), moderate biofilm producer (++) or strong biofilm producer (+++); n = 3.

Blank: medium only, NoE: Biofilm in the absence of savory extract, Se: Biofilm in the presence of savory extract.

Table (4):

Effect of dried savory leaves and its hydroethanolic extracts on serum hepatic parameters

| Groups | Parameters | | |
|--------|---------------------------|--------------------------|---------------------------|
| | AST (IU/L) activity | ALT (IU/L) activity | ALP (IU/L) activity |
| 1 | 41.80 ^d ±1.07 | 20.20 ^c ±0.86 | 84.20 ^c ±1.36 |
| 2 | 112.60 ^a ±1.36 | 88.20 ^a ±1.07 | 241.40 ^a ±2.79 |
| 3 | 71.40 ^b ±1.36 | 45.00 ^b ±1.52 | 134.80 ^b ±2.69 |
| 4 | 65.60 ^c ±1.29 | 43.20 ^b ±1.28 | 128.40 ^b ±2.42 |
| 5 | 68.60 ^{cb} ±1.03 | 43.60 ^b ±0.93 | 132.40 ^b ±1.21 |
| 6 | 65.00 ^c ±1.14 | 41.80 ^b ±1.07 | 128.40 ^b ±2.42 |

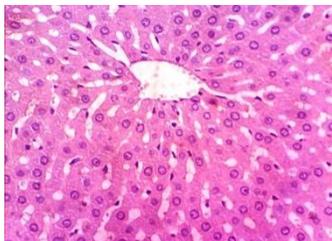
Data are mean ± SE, n=5, Different uppercase letters in the same column represent statistically significant data at 5%. Whereas; 1: normal control group; 2: Injury control group; 3: CCl4 + Dried savory leaves 1% group; 4: CCl4 + Dried savory leaves 2% group; 5: CCl4 + Savory leaves extract 25 ppm group; 6: CCl4 + Savory leaves extract 50 ppm group.

Table (5):

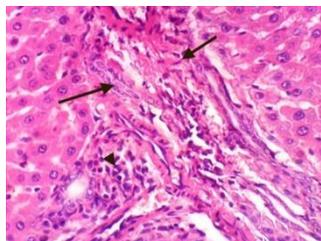
Oxidative stress biomarkers of different rats groups fed on dried savory leaves and its dried hydroethanolic extracts

| Parameters Groups | SOD (mg/dl) activity | MDA (nmol/ml) levels |
|----------------------|---------------------------|--------------------------|
| 1 | 35.46 ^a ±0.86 | 4.62 ^f ±0.20 |
| 2 | 10.64 ^c ±0.27 | 24.72 ^a ±1.03 |
| 3 | 32.23 ^b ±0.92 | 15.97 ^b ±0.39 |
| 4 | 32.61 ^b ±1.21 | 12.45 ^c ±0.19 |
| 5 | 33.57 ^{ab} ±1.21 | 9.77 ^d ±0.13 |
| 6 | 31.30 ^b ±0.75 | 7.80 ^e ±0.34 |

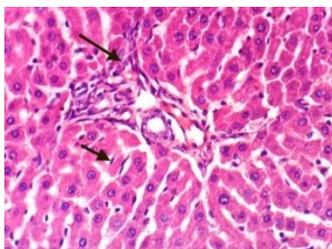
Data are mean ± SE, n=5, Different uppercase letters in the same column represent statistically significant data at 5%. Whereas; 1: normal control group; 2: injury control group; 3: CCl₄ + Dried savory leaves 1% group; 4: CCl₄ + dried savory leaves 2% group; 5: CCl₄ + Dried savory leaves extract 25 ppm group; 6: CCl₄ + Dried savory leaves extract 50 ppm group.



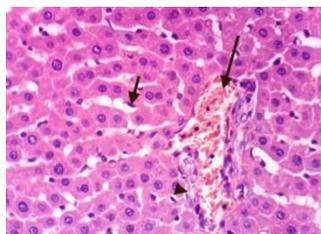
Micrograph (1):
Liver of normal control rat (H & E, 400X).



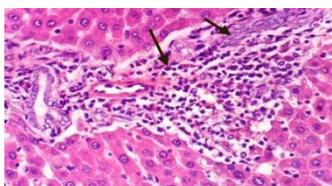
Micrograph (2):
Liver of CCl₄-treated rat (H & E, 400X).



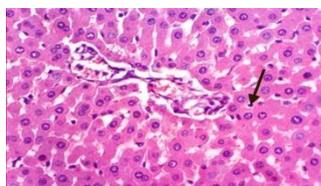
Micrograph (3):
Liver of rat CCl₄-treated and fed
on dried savory leaves 1%
(H & E, 400X).



Micrograph (4):
Liver of rat CCl₄-treated
and fed on dried savory leaves 2%
(H & E, 400X).



Micrograph (5):
Liver of rat CCl₄-treated
and administrated dried savory
leaves extract 25 ppm (H & E, 400X)



Micrograph (6):
Liver of rat CCl₄-treated
and administrated dried savory
leaves extract 50 ppm (H & E, 400X).

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الأنشطة المضادة للأوكسدة، المضادة للأغشية الحيوية والبيولوجية

للسافورى الصيفى

سمر حسن محمود محمد¹ ، إيهاب صلاح عشوش¹ ، هشام

محسن الحريرى¹ ، إسحق مراد الحديدى²

¹ قسم علوم الأغذية - كلية الزراعة - جامعة عين شمس - القاهرة - مصر

² معهد بحوث تكنولوجيا الأغذية - مركز البحوث الزراعية - الجيزة - مصر

الملخص العربى

كان لأمراض الكبد في جميع أنحاء العالم اهتماماً أكبر بأساليب الوقاية، يمكن أن يكون النظام الغذائي المخطط فعالاً ووقائياً. لذلك، هدفت هذه الدراسة إلى فحص الدور المضاد للأوكسدة والوقائى للكبد للأوراق السافورى المجففة (1 و 2٪) والمستخلص الإيثانولى (25 و 50 جزء في المليون) ضد الضرر الكبدى المستحدث برابع كلوريد الكربون (CCl₄) في الجرذان . كما تم دراسة تأثير مستخلص أوراق السافورى على تكوين الأغشية الحيوية لبعض السلالات الميكروبية. أظهرت المجموعة المستحثة للضرر الكبدى زيادة معنوية في متغيرات الكبد في الدم (AST ، ALT و ALP) ومستويات المالنوالدهيد (MDA) . في حين، إنخفاض نشاط (SOD). علاوة على ذلك ، سجلت المجموعة المستحثة للضرر الكبدى تغيرات نسيجية عديدة وشديدة بسبب تلف الخلايا الكبدية بواسطة CCl₄. أدت المعاملة بالأوراق المجففة للسافورى ومستخلصاتها إلى تحسن كبير في وظائف الكبد ومعايير الإجهاد التأكسدي. أيضاً، توقف التغيير النسيجي في الكبد. تتمتع كل من البكتيريا الموجبة والسالبة لجرام بالقدرة على تكوين غشاء حيوي بدرجات متفاوتة. يحتوي المستخلص الإيثانولى للسافورى على تأثير مضاد لتكوين الأغشية الحيوية ضد سلالات البكتيريا تحت الدراسة . أيضاً، أظهر مستخلص أوراق السافورى نشاطاً في خلب الشقوق الحرة نظراً لاحتوائها على المركبات الفينولية والفلافونويد، والمسؤولة عن خصائصها المضادة للأوكسدة والمضادات لتكوين الأغشية الحيوي ووقاية الكبد. يمكن الاستنتاج أن أوراق السافورى المجففة ومستخلصاتها يمكن استخدامها كمكونات في الأطعمة الوظيفية لوقاية الكبد