Saghaei *et al.*, ***Journal of Medicinal Herbs***, Vol. 13, No. 1: 9-17, 2022
doi: 10.30495/MEDHERB.2022.691251

Journal of Medicinal Herbs
journal homepage:**www.jhd.iaushk.ac.ir**

Gastroprotective effects of ethanolic extract of *Satureja bachtiarica* against ethanol-induced gastric ulcers in rats

Firoozeh Saghaei[*1], **Abdolrasoul Namjou**[2]

[1]*Department of Pharmacology, Faculty of Veterinary Medicine, Shahrekord Branch, Islamic Azad University, Shahrekord, Iran;*
***Email:** *F.saghaei@iaushk.ac.ir*

[2]*;Department of Pathology, Shahrekord Branch, Islamic Azad University, Shahrekord, Iran ;*

ARTICLE INFO

Type: *Original Research*
Topic: *Medicinal Plants*
Received *December13th2021*
Accepted *April 18th2022*

Key words:
- ✓ *Satureja bachtiarica*
- ✓ *Gastric ulcers*
- ✓ *Malonedialdehyde*
- ✓ *Superoxidedismutase*
- ✓ *Rat*

ABSTRACT

Background & Aim: Free radicals play an important role in the development of some diseases, including gastrointestinal ulcers. Current gastric ulcer drugs have side effects, so the tendency to use herbs and replace them with synthetic drugs has increased. In this regard, plants of the *Satureja* family have been studied in terms of healing and antioxidant properties. The aim of this study was to evaluate the protective effects of *Satureja bachtiarica* ethanolic extract on ethanol-induced gastric mucosal ulcers in rats.

Experimental: Thirty rats were divided into 5 equal groups. Group 1 received 200 mg / kg of ethanolic extract of *Satureja bachtiarica* (EESB), group 2 received 400 mg / kg EESB, group 3 received 20 mg / kg omeprazole, group 4 received 1 mL of CMC and group 5 received 1 mL of normal saline. One hour after treatment, each animal received absolute ethanol (1ml / rat) orally by gavage. The other 30 mice were treated with oral ethanol after induction of gastric ulcer as described above, but all treatments were once a day for 14 days. At the end of both tests, ulcer index, histopathologic exams, Malonedialdehyde (MDA) and Superoxide dismutase (SOD) were measured.

Results: Ethanolic extract of *Satureja bachtiarica* (400 mg/kg) and omeprazole had a significant effect on reducing ulcer index and increasing ulcer inhibition. It significantly preserved the gastric wall mucosa and reduced the formation of ethanol-induced gastric lesions. Also in this study a significant increase in superoxide dismutase (SOD) activity and a significant decrease in Malonedialdehyde (MDA) in gastric tissue were observed.

Recommended applications/industries: The anti-ulcer effect of this plant seems to be due to the reduction of oxidative stress, which is probably due to the presence of thymol and carvacrol in the plant extract. This plant may be considered officially in the treatment and prevention of stomach ulcers in the future.

1. Introduction

Peptic ulcer is the most common gastrointestinal disorder caused by an imbalance between the invasive and defensive factors of the gastric mucosa. Invasive agents include acid, pepsin, Helicobacter pylori, nonsteroidal anti-inflammatory drugs (NSAIDs), ethanol, and oxidative stress, and defense factors that protect the gastrointestinal mucosa include bicarbonate, mucus secretion, blood flow, and cell regeneration. Internal factors that may be involved in protecting the gastrointestinal tract include prostaglandins (PGE2), somatostatin, nitric oxide (NO) and sulfhydryl compounds. (Kountouras *et al.*, 2001; Demir *et al.*, 2003).

In the pathology of the gastric mucosa, increasing of free radicals and reducing antioxidants and blood flow to the gastric mucosa play an important role (Al Asmari *et al*., 2014). Ethanol consumption increases oxidative stress and lipid peroxidation, hydroxyl radical production and DNA damage, leading to lesions and erosion of the gastric mucosa. Ethanol-induced ulcers are also associated with decreased mucosal microcirculation and increased apoptosis (Hernandez-Munoz *et al*., 2000; Amanvermez *et al*., 2008).

Common treatments for gastric ulcers include H2-receptor blockers, proton pump inhibitors, antacids, anticholinergics and antibiotics, and mucosal protectors such as sucralfate and bismuth (Bighetti *et al*., 2005). These drugs have limited efficacy and severe side effects and need to be treated safely and effectively. Herbal remedies are traditionally and modernly used to treat many ailments including digestive disorders such as gastric ulcers (Xie *et al*., 2013). This is an important reason to choose and study the anti-ulcer effects of herbs that have traditionally been used to treat stomach diseases.

Satureja bachtiarica is a native plant of Iran that is distributed in the western, central and southwestern regions of the country (Ahmadi *et al*., 2009).

Studies have shown that *Satureja bachtiarica* has beneficial effects such as anti-flatulence, appetizing, anti-diarrhea, treatment of indigestion, relieving weakness and stomach cramp and intestinal fermentation, relieving cough and dyspnea (Memarzadeh *et al*., 2015). It is also useful for relieving visceral pain (Saghaei, 2018).Various studies have shown that, the essential oils of *Satureja bachtiarica* aerial parts contain thymol (44.5%), gamma-terpinene (23.9%), p-cymene (7.3%), beta-caryophyllene (5.3%), and borneol (4.2%) (Sefidkon and Jamzad, 2000). The essential oil of pre-flowering *Satureja bachtiarica* contained 20% carvacrol and 19% thymol, and that of flowering *Satureja bachtiarica* contained approximately 26% carvacrol and 5% thymol (Sefidkon *et al*., 2007). In measuring the antioxidant activity of methanolic extract of two samples, *Satureja bachtiarica*, wild and cultured, showed that the wild sample has a greater free radical scavenging effect and antioxidant power, which is probably due to higher amounts of phenolic compounds and flavonoids (Jafari *et al*., 2016). The present research aimed at exploring possible effects of *Satureja bachtiarica* on prevention of gastric ulcer that is probably related to its antioxidant effects.

2. Materials and Methods

2.1. Drugs and chemicals

Ethanol was purchased from Merck Co. Carboxymethyl Cellulose (CMC), Potassium chloride, Buthanol, 1,1,3,3 tetraetoxy propane purchased from Sigma Aldrich Co (Dorset, England), and omeprazole purchased from Abidi Pharmaceutical Co (Iran), Superoxide Dismutase Eliza kit (ZellBio GmbH (Germany)**)** purchased from Novin Navand Salamat Company.

2.2. Plant collection and extraction preparation

The aerial part of *Satureja bachtiarica* was collected from the mountains of *Chaharmahal and Bakhtiari* province (Iran). It was then approved as *Satureja bachtiarica* by Chaharmahal and Bakhtiari Agricultural and Natural Resources Research and Education Center (herbarium number: 885-D). The plants were dried at room temperature in the shade and then powdered .The powder is soaked in absolute ethanol for 72 hours. It was then filtered with Whatman No. 1 filter paper. A rotary device was then used to remove residual ethanol and percentage yield was determined (Uroko *et al*., 2020).

2.3. Experimental Animals

60 male Wistar rats (200-250 g weight and 8 week age) were obtained from animal house of Pastor Research Institute (Tehran).

All animals were kept under standard conditions at 23 to 35 ° C in the 12 hour light / 12 hour dark cycle and were freely allowed access to standard mouse food and water. During the experiment, the animals were kept in separate cages with mesh floors to prevent coprophagy.

2.4. Gastroprotective studies

2.4.1. Experiment 1: prevention groups

30 rats were divided into 5 equal groups (6 rats per group), after 24h of fasting (water was accessible expect for the last 2 hours) all the rats were orally administered medication as below:

Group 1 received 200mg/kg ehtanolic extract of *Satureja bachtiarica* (EESB), Group 2 received

400mg/kg EESB (Doses were determined based on a pilot experiment), Group 3 received 20mg/kg omeprazole, Group 4 received 1 mL CMC and Group 5 received 1 ml normal saline. One hour after the treatment, each animal received orally, absolute ethanol (1mL/rat) through gavages.

2.4.2. Experiment 2: therapeutics groups

The other 30 rats also divided into 5 equal groups (n=6) and after induction of gastric ulcer by oral ethanol, they were treated according to the method described above, but all treatments were once a day for 14 days, because omeprazole is prescribed to treat stomach ulcers for at least 14 days (Ghamarian, 2010). In both experiments, all rats were anesthetized with ketamine and xylazin 1 hour after the last administration. The rats' abdomens were opened and the stomachs were removed, opened along greater curvature and washed with cold saline solution.

2.5. Determination of ulcer Index

For a more accurate assessment, the stomachs were expanded between two glass plates. The length of gastric hemorrhagic lesions (ulcers) was then measured under a dissecting microscope. The maximum length of each lesion (mm) for each stomach was expressed as the ulcer Index (UI), and the inhibition percentage was calculated by the following formula:

Inhibition (%) = $[(uI_{control}\text{-}uI_{treatment})/uI_{control}]\times 100\%$

(Vemula *et al*., 2012).

2.6. Histopathologic studies

All stomachs were fixed in 10% neutral formalin for histopathology examinations. They were paraffin - embedded and micro-sectioned at a minimal thickness of 4µm and then stained with hematoxylin and eosin for histopathology examinations.

2.7. Superoxide dismutase (SOD) analysis

SOD measurements were performed by an Eliza kit. The ZellBio GmbH assay kit uses superoxide anion to convert hydrogen peroxide and oxygen under enzymatic reaction conditions. Finally, it produces a chromogenic product that is measured by colorimetry at 420 nm.

$$2O2\text{-} + 2H^{+} \rightarrow O_2 + H_2O_2$$

The following steps were performed according to the kit method: The sample was cut and weighed. A certain amount of PBS (100mM, pH =7.4) was added for homogenization. The sample was melted and kept at 2-8°C and a certain amount of PBS (PH = 7.4) was added to it and then the sample (~ 100 mg of tissue per mL of PBS buffer) was completely homogenized by hand or homogenizer. The homogenized mixture is centrifuged for approximately 20 minutes (at 4000-6000 RPM). The supernatant is then carefully collected and part of it is used for examination and the rest is frozen for later use. It is well mixed and the well absorption rate is read at 0 and 2 minutes with a microplate reader / ELISA at 420 nm. SOD activity in unknown samples was calculated based on the following formula:

SOD activity (U/mL) = $(V_p\text{-}V_c)/V_p \times 60$

$V_p = OD_{sample\ 2\ min} - OD_{blank\ 2\ min}$

$V_c = OD_{sampl\ 0\ min} - OD_{blank\ 0\ min}$

2.8. Malondialdehyde (MDA) analysis

Gastric mucosal MDA assay was performed by thiobarbituric acid method. First, the parts of frozen stomachs were defrosted and halogenated in 10mL of 100g/L KCl. The homogenate (0.5 mL) was added to a solution containing 0.2mL of 80g/L sodium lauryl sulfate, 1.5 mL of 200 g/L acetic acid, 1.5 mL of 8 g/l 2-thiobarbiturates and 0.3 ml distilled water. The mixture was incubated at 98° for one hour and 5ml of n-butanol: pyridine (15:1) was added upon cooling. The mixture was shake for one minute and centrifuged for 30 minutes at 4000 round mp. The absorbance of the supernatant was measured at 532 nm. The standard curve was obtained by using 1, 1, 3, 3-tetraethoxy propane (Albayrak *et al*., 2015).

2.9. Statistical analysis

The obtained data was statistically evaluated by one-way ANOVA followed by Duncan's test used to identify statistical differences between means and presented as mean ± standard deviation (S.D.) at significant level of $P \leq 0.05$.

3. Results and discussion

In this study, the protective and therapeutic effect of *Satureja bachtiarica* extract on ethanol-induced gastric ulcer and its antioxidant activity were investigated**.** Previous studies have demonstrated gastro-protective and ulcer healing properties of many medicinal plants (Rozza and Pellizzon, 2012). Substances with

antioxidant properties, such as phenolic compounds, appear to protect the stomach from ethanol-induced gastric damage (Stahl-Biskup and Saez, 2003).

3.1. Effect of EESB on ethanol - induced ulcer

Healthy rats did not show ulcers or lesions in their stomachs (Figure 1a). Rats receiving ethanol showed bleeding lesions in the stomach wall (Figure 1b). Rats treated with EESB and omeprazole showed reduction in lesions size and ulcers (Figure 1c, d, and e). Administration of CMC did not show any healing in lesions (Figure 1f).

Absolute ethanol caused an ulcer index (UI) of (3.468±0.37) while pretreatment with EESB (200mg/kg and 400 mg/kg) and omeprazole (20mg/kg) caused reduction in ulcer index respectively. In treatment groups, EESB and omeprazole caused a significant decrease in ulcer index compared to ethanol group (Table 1).

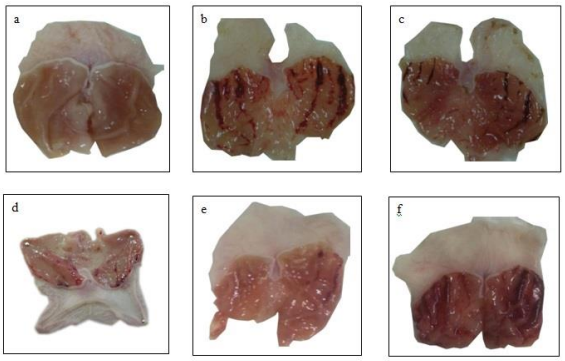

Figure 1: Macroscopic investigation of stomach tissue in different groups.
a) Normal stomach, **b)** ethanol induced ulcer, **c)** *Satureja bachtiarica* extract treatment (200 mg/kg), **d)** *Satureja bachtiarica* extract treatment (400 mg/kg), **e)** omeprazol treatment (20 mg/kg) **f)** CMC treatment.

Table 1: Effect of ethanolic extract of *Satureja bachtiarica* (EESB), omeprazole (OM P) and carboxy methyl cellulose (CMC) on Ulcer index (UI) in ethanol induced peptic ulcer rats.

Groups	UI in pretreatment groups	UI in treatment groups	Inhibition in pretreatment groups (%)	Inhibition in treatment groups (%)
EESB 200mg/kg	1.613±0.28^{b}	2.002 ± 0.33 ^{b}	36.590±5.5^{c}	52.054±3.2^{c}
EESB 400mg/kg	0.888±0.15^{bc}	1.069 ± 0.40 ^{c}	66.680±3.7^{b}	82.820±7.1^{c}
OMP 20 mg/kg	0.333±0.18^{c}	0.733 ± 0.06 ^{c}	45.770±4.1^{a}	82.820±6.5^{a}
Control	0.333±0.13^{c}	3.047±0.65 ^{a}	81.220±4.3^{a}	0
CMC 1mL	3.420±0.40^{a}	3.468 ± 0.37 ^{a}	0	1.000±2.09

All values are expressed as mean ± SD. Means with different superscripts are significantly different (P≤ 0.05).

The finding of macroscopic examination shows that absolute ethanol can produce gastric ulcer and can increase the ulcer index. The use of EESB and omeprazole for treatment or before treatment leads to a significant reduction in the severity and number of lesions caused by ethanol. The EESB-treated group (400 mg/kg) showed mild mucosal damage and the omeprazole-treated group showed normal mucosa. These results were similar with that of antiulcerogenic activity of *Bauhinia holophylla* hydroalcoholic extract (Rozza *et al*., 2015) and effects of *Pithecellobium Jiringa* ethanol extract against ethanol-induced gastric mucosal injuries by (Ibrahim *et al*., 2012) and protective effect of M. pruriens on ethanol-induced gastric ulcer in rats (Golbabapour *et al*., 2013).

3.2. Histopathologic findings

Ethanol administration causes tissue necrosis, edema, inflammation, and cellular infiltration under the mucosa (Figure 2a). In the groups treated with EESB showed mild necrosis to the surface epithelium and minor edema of submucosal layer (Figure 2b,c) and omeprazole showed mild superficial epithelial abnormalities, but no deep mucosal damage and approximate improvement of the mucosal layer was observed (Figure 2d). CMC as an omeprazole carrier, had no mucosal protection and showed severe superficial epithelial dysfunction, necrotic lesions that penetrated deep into the mucosa, extensive submucosal edema, and leukocyte infiltration (Figure 2e).

The use of alcohol to cause stomach ulcers and oxidative damage has been confirmed by other research. Ethanol causes hemorrhagic erosion and gastric ulcer after oral administration (Shetty *et al*., 2000). In addition, ethanol causes gastric ulcers by producing free radicals, oxidative function and lipid peroxidation (Ali Khan *et al*., 2013). Ethanol affects mucosal blood flow, damages capillary endothelium and releases arachidonate metabolites, leukoteriene C4

and D4 and platelet activating factor (PAF) (Goel and Bhattacharya, 1991).

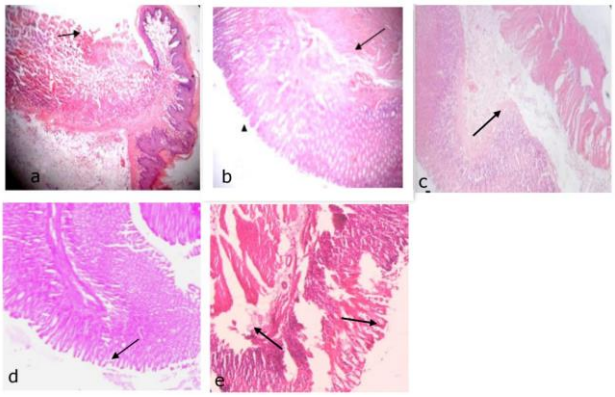

Figure 2: Microscopic investigation of stomach tissue in different groups:
a) Rats receiving only absolute ethanol. **b,c)** Rats pre-treated with EESB (200 and 400 mg/kg). **d)** Rats pre-treated with omeprazole. **e)** Rats pre-treated with CMC. (H&E stain 10×).

Omeprazole is widely used to treat diseases related to gastric acid secretion (Li *et al*., 2004). Omeprazole is not only a proton pump inhibitor, but also acts as an anti-inflammatory, antioxidant and stimulant of gastric mucosal secretion (Moghadamtousi *et al*., 2014). Kobayashi explained that omeprazole protects against gastric lesions caused by the 48.80 compound in rats. Its protective effect is through anti-inflammatory and antioxidant effects (through non-protein sulfhydryl content) and increase the activity of myeloperoxidase and xanthine oxidase (XO) and lipid peroxide (LPO) in gastric mucosa (Kobayashi *et al*., 2002). In ethanol-induced gastric ulcers, omeprazole protects gastric tissue at doses that do not inhibit acid secretion (Maity *et al*., 2003; Kushima *et al*., 2005).

Other studies have shown that omeprazole has a protective and therapeutic effect on gastric ulcer through its antioxidant and ROS-removing properties (Wandall, 1992; Saghaei *et al*., 2012). Oxidative stress plays a main role in the pathogenesis of various diseases such as gastric ulcer (Ibrahim *et al*., 2012). Antioxidants protect cells from damage caused by oxidative stress and strengthen the immune system of mice (Abdulla *et al*., 2010).

ROS causes tissue damage, and antioxidant protection systems such as GSH, Catalase, GPX, and SOD reduce their levels. These defense systems protect the stomach tissue from damage caused by ethanol-induced ROS production.

3.3. SOD Activity

Comparison of SOD levels in the treatment and pre-treatment groups showed that only in the two groups receiving EESB (400 mg / kg) and omeprazole a significant increase in SOD was observed, although it was not significant in comparison between the treatment and pre-treatment groups (P≤0.05) (Table 2).

Table 2. Effect of ethanolic extract of *Satureja bachtiarica* (EESB), omeprazole (OM P) and carboxy methyl cellulose (CMC) on superoxide dismutase (SOD) activity (u/mg protein) and malondialdehyde (MDA) in gastric mucosa (nmol/mg protein) in ethanol induced peptic ulcer rats.

Groups	SOD pretreatment groups (u/mg protein)	SOD treatment groups (u/mg protein)	MDA pretreatment groups (nmol/mg protein)	MDA Treatment groups (nmol/mg protein)
EESB 200mg/kg	3.991 ± 0.58^{bcd}	2.177 ± 0.4^{bc}	$3.790\pm0/42^{ab}$	0.776 ± 0.48^{ab}
EESB 400mg/kg	4.462 ± 0.16^{abc}	3.202 ± 0.39^{a}	3.167 ± 0.31^{bc}	0.347 ± 0.056^{b}
OMP 20 mg/kg	5.032 ± 0.95^{ab}	3.201 ± 0.29^{a}	2.526 ± 0.26^{c}	0.351 ± 0.45^{b}
Control	3.153 ± 0.65^{cde}	2.292 ± 0.38^{c}	4.118 ± 0.059^{a}	1.036 ± 0.37^{a}
CMC 1mL	2.952 ± 0.73^{de}	1.366 ± 0.28^{bc}	3.846 ± 0.74^{ab}	1.052 ± 0.42^{a}

All values are expressed as mean ± SD. Means with different superscripts are significantly different (P≤0.05).

SOD is an important antioxidant enzyme that breaks down superoxide anions into hydrogen and oxygen peroxides. (Devi *et al*., 2007). Therefore, it protects the stomach against oxidative stress caused by various stimuli in cells and tissues (Shin *et al*., 2013). The increase in SOD levels in the groups receiving the extract and omeprazole indicates an antioxidant mechanism that underlies its protective function. This indicates that the plant extract has the ability to prevent lipid peroxidation and can be a therapeutic drug to inhibit free radicals. The decrease in SOD activity in gastric tissue (control group) is probably due to increased production of reactive oxygen species (Ibrahim *et al*., 2012). Other scholars have also

reported results supporting this finding: SOD and catalase activity was decreased in gastric tissue of mice with gastric ulcers induced by indomethacin, hydrochloric acid and ethanol. (Izzo *et al*., 1994. Brzozowski *et al*., 1998; Alvarez-Suarez *et al*., 2011). In the present study, after administration of EESB (400 mg / kg) and omeprazole (20 mg / kg) in both treatment and pretreatment groups, SOD activity increased and gastric ulcer healed, possibly related to the active ingredients of *Satureja bachtiarica* extract (Carvacrol and thymol).

3.4. MDA activity

In the pretreatment groups, ethanol administration alone (control group) greatly increased the amount of MDA in the gastric tissue. In other treatment groups, the amount of MDA decreased, which was statistically significant only in the EESB (400 mg / kg) and omeprazole groups ($P<0.05$). The observed effects on MDA in these two groups was equivalent. In the treatment groups after 14 days, the amount of MDA in the control group and the groups receiving CMC and EESB (200 mg / kg) were equivalent and not statistically different, but in the two groups receiving EESB (400 mg / kg) and omeprazole there was a significant decrease in MDA is seen ($P\leq0.05$) (Table 2).

MDA is the most important product of lipid peroxidation (Johansen *et al*., 2005; Ibrahim *et al*., 2012). Increased MDA levels lead to increased free radicals. These radicals increase cell lipid peroxidation, causing cell degranulation, which leads to the destruction of the structure and function of the cell membrane (Michiels *et al*., 1994; Ali Khan *et al*., 2013). In addition, a strong correlation has been reported between the levels of lipid peroxidation of gastric mucosa and stress-induced gastric ulcer (Tandon *et al*., 2004).

In the present study, a significant decrease in MDA levels of homogenized gastric tissue of rats treated with omeprazole or plant extract was observed, possibly due to the antioxidant activity of carvacrol or thymol as well as omeprazole. The results of other researchers support this claim, Mehmet Guvenc and et al. found that thymol and carvacrol reduced MDA levels in the testes, liver and kidneys (Guvenc *et al*., 2018). Increased SOD activity and decreased MDA levels have been observed in the treatment of gastric ulcer with *Pithecellobium jiringa* extract. This plant contains polyphenolic compounds that have antioxidant effects (Ibrahim *et al*., 2012).

Methanolic extract of *Tabernaemontana divaricata* (L.) R. Br flower has been associated with a protective effect on rat gastric tissue and a decrease in malondialdehyde (MDA) concentration, which indicates antioxidant activity and reduced lipid peroxidation (Ali khan *et al*., 2013). Karanjin (a flavonoid from *Pongamia pinnata*) has shown that it can inhibit oxidative stress by normalizing lipid peroxidation (MDA) and levels of antioxidant enzymes (such as catalase, peroxidase, and superoxide dismutase) (Vismaya *et al*., 2011). Methanolic Extracts of *Satureja bachtiarica* have high antioxidative activity that ascribable to the oxygenated compounds such as carvacrol and thymol (Rabiei *et al*., 2020; Sefidkon *et al*., 2007).

Carvacrol reduces interleukin (IL) -1β, IL-4, IL-8 and malondialdehyde (MDA) (Carvalho *et al*., 2020). Oral administration of carvacrol for 21 days in male wistar rats with hepatotoxicity and oxidative damage induced by D-Galactosamine (D-GalN) decreased the activity of the superoxide dismutase and glutathione peroxidase. Levels of other antioxidants (vitamin C, vitamin E and reduced glutathione) also returned to normal in plasma, red blood cells, liver and kidneys (Aristatile *et al*., 2009).

In another study, carvacrol and thymol had antiproliferative effects on human uterine cancer cells. This effect was dose dependent (Mastelic *et al*., 2008). Ismaeili studied the antioxidant activity of thymol in ethanol. The results showed it has good antioxidant properties and can be used as a natural antioxidant (Esmaeili and Khodadadi, 2011).

Hashemipour and Colleague studied the effect of thymol and carvacrol on the activity of antioxidant enzymes in broiler chickens and showed that this plant product increases the activity of superoxide dismutase and glutathione peroxidase and reduces malondialdehyde levels in thigh muscles, serum and liver (Hashemipour *et al*., 2013)

4. Conclusion

Our findings showed that ethanolic extract of *Satureja bachtiarica* have gastro-protective effect by enhancing the production of antioxidant enzyme such as SOD and decreasing of MDA level after using ethanol for inducing peptic ulcer. These effects may be

related to carvacrol and thymol compounds in *Satureja bachtiarica* extract.

5. Acknowledgements

This study was suppurated by Islamic Azad University of Shahrekord. The authors are thankful to this support.

6. References

Abdulla, M.A., Ahmed, K.A.A., Al-Bayaty, F.H. and Masood, Y. 2010. Gastroprotective effect of *Phyllanthus niruri* leaf extract against ethanol-induced gastric mucosal injury in rats. *African Journal of Pharmacy and Pharmacology*, 4(5):226-230.

Ahmadi, S., Sefidkon, F., Babakhanlo, P., Asgari, F., Khademi, K. and Karimifar, M. 2009. Comparing essential oil composition of *Satureja bachtiarica Bunge* before and full flowering stages in field and provenance. *Iranian Journal of Medicinal and Aromatic Plants*, 25(2):159-169.

Al Asmari, A., Arshaduddin, M., Elfaki, I., Kadasah, S., Al Robayan, A. and Al Asmary, S. 2014. Aripiprazole an atypical antipsychotic protects against ethanol induced gastric ulcers in rats. *International journal of clinical and experimental medicine*,7(8):2031-44.

Albayrak, A., Alp, H.H. and Suleyman, H. 2015. Investigation of antiulcer and antioxidant activity of moclobemide in rats. *The Eurasian Journal of Medicine*, 47(1):32-40.

Ali Khan, M.S., Mat Jais, A.M. and Afreen, A. 2013. Prostaglandin analogous and antioxidant activity mediated gastroprotective action of *Tabernaemontana divaricata* (L.) R. Br. flower methanolic extract against chemically induced gastric ulcers in rats. *BioMed Research International*, 2013: Article ID 185476, 18 pages.

Alvarez-Suarez, J.M., Dekanski, D., Ristic, S., Radonjic, N.V., Petronijevic, N.D. and Giampieri, F. 2011. Strawberry polyphenols attenuate ethanol-induced gastric lesions in rats by activation of antioxidant enzymes and attenuation of MDA increase, *Plos One*: 6(10), e25878.

Amanvermez, R., Tuncel, O. K., Demir, S., Kefeli, M., Bek, Y. and Celik, C. 2008. Protective effects of cysteine, methionine and vitamin C on the stomach in chronically alcohol treated rats. *Journal of Applied Toxicology*, 28(5): 591-598.

Aristatile, B., Al-Numair, K.S., Veeramani, C. and Pugalendi, K. V. 2009. Effect of carvacrol on hepatic marker enzymes and antioxidant status in d-galactosamine-induced hepatotoxicity in rats. *Fundamental and Clinical Pharmacology*, 23(6): 757-765.

Bighetti, A.E., Antonio, M.A., Kohn, L.K., Rehder, V.L.G., Foglio, M.A., Possenti, A., Vilela, L. and Carvalho, J.E. 2005. Antiulcerogenic activity of a crude hydroalcoholic extract and coumarin isolated from Mikania laevigata Schultz Bip. *Phytomedicine*, 12(1-2): 72-79.

Brzozowski, T., Konturek, S., Kwiecien, S., Pajdo, R., Drozdowicz, D. and Sliwowski, Z. 1998. SU-840, a novel synthetic flavonoid derivative of sophoradin, with potent gastroprotective and ulcer healing activity. *Journal of Physiology and Pharmacology*, 49(1):83-98.

Carvalho, F. O., Silva, E. R., Gomes, I. A., Santana, H., S. R., Santos, D. N., Souza, G. P. O. et al., 2020. Anti-inflammatory and antioxidant activity of carvacrol in the respiratory system: *A systematic review and meta-analysis. Phytotherapy Research*, 34 (9): 2214-2229.

Demir, S., Yilmaz, M., Koseoglu, M., Akalin, N., Aslan, D. and Aydin, A. 2003. Role of free radicals in peptic ulcer and gastritis. *The Turkish Journal of Gastroenterology*, 14(1):39-43.

Devi, R.S., Narayan, S., Vani, G. and Devi, C.S.S. 2007. Gastroprotective effect of *Terminalia arjuna bark* on diclofenac sodium induced gastric ulcer. *Chemico-biological Interactions*,167(1):71-83.

Esmaeili, A. and Khodadadi, A. 2011. Antioxidant Activity of a solution of thymol in ethanol. *Zahedan Journal of Research in Medical Sciences*, 14(7): 14-18.

Ghamarian, A.R. 2010. Physicians Desk Reference Iran.vol. 1. *Ghamarian press.* pp. 588.

Goel, R.K. and Bhattacharya, S.K. 1991. Gastroduodenal mucosal defence and mucosal protective agents. *Indian Journal of Experimental Biology*, 29(8):701-714.

Golbabapour, S., Hajrezaie, M., Hassandarvish, P., Abdul Majid, N., Hadi, A.H., Nordin, N., et al. 2013. Acute toxicity and gastroprotective role of M. pruriens in ethanol-induced gastric mucosal injuries

in rats. *BioMed Research International*, 2013: Article ID 974185.

Guvenç, M., Cellat, M., Gokçek, I., Yavaş, I. and Ozsoy, S. Y. 2018. Effects of thymol and carvacrol on sperm quality and oxidant/antioxidant balance in rats. *Archives of Physiology and Biochemistry*, 125 (5): 396-403.

Hashemipour, H., Kermanshahi, H., Golian, A. and Veldkamp, T. 2013. Effect of thymol and carvacrol feed supplementation on performance, antioxidant enzyme activities, fatty acid composition, digestive enzyme activities, and immune response in broiler chickens. *Poultry Science*, 92(8): 2059-2069.

Hernandez-Munoz, R., Montiel-Ruiz, C. and Vazquez-Martinez, O. 2000. Gastric mucosal cell proliferation in ethanol-induced chronic mucosal injury is related to oxidative stress and lipid peroxidation in rats. *Laboratory Investigation*, 80(8):1161-1169.

Ibrahim, I. A. A., Qader, S.W., Abdulla, M.A., Nimir, A.R., Abdelwahab, S.I. and Al-Bayaty, F.H. 2012. Effects of *Pithecellobium jiringa* ethanol extract against ethanol-induced gastric mucosal injuries in Sprague-Dawley rats. *Molecules*, 17(3): 2796-811.

Izzo, A., Carlo, G.D., Mascolo, N. and Capasso, F. 1994. Antiulcer effect of flavonoids. Role of endogenous PAF. *Phytotherapy Research*,8(3):179-181.

Jafari, F., Ghavidel, F. and Zarshenas, M.M. 2016. A critical overview on the pharmacological and clinical aspects of popular Satureja species. *Journal of Acupuncture and Meridian Studies*, 9(3): 118-127.

Johansen, J.S., Harris, A.K., Rychly, D.J. and Ergul, A. 2005. Oxidative stress and the use of antioxidants in diabetes: linking basic science to clinical practice. *Cardiovascular Diabetology*, 4:5-15.

Kobayashi, T., Ohta, Y., Inui, K., Yoshino, J. and Nakazawa, S. 2002. Protective effect of omeprazole against acute gastric mucosal lesions induced by compound 48/80, a mast cell degranulator, in rats. *Pharmacological Research*, 46(1):75-84.

Kountouras, J., Chatzopoulos, D. and Zavos, C. 2001. Reactive oxygen metabolites and upper gastrointestinal diseases. *Hepato-gastroenterology*, 48(39):743-751.

Kushima, H., Hiruma-Lima, C.A., Santos, M.A., Viana, E., Coelho-Ferreira, M. and Brito, A.R.M.S. 2005. Gastroprotective activity of Pradosia huberi on experimentally induced gastric lesions in rodents: Role of endogenous sulphydryls and nitric oxide. *Journal of Ethnopharmacology*, 101(1-3):61-67.

Li, X.Q., Andersson, T.B., Ahlström, M., Weidolf, L. 2004. Comparison of inhibitory effects of the proton pump-inhibiting drugs omeprazole, esomeprazole, lansoprazole, pantoprazole, and rabeprazole on human cytochrome P450 activities. *Drug metabolism and disposition*, 32(8):821-827.

Maity, P., Biswas, K., Roy, S., Banerjee, R.K. and Bandyopadhyay, U. 2003. Smoking and the pathogenesis of gastroduodenal ulcer–recent mechanistic update. *Molecular and Cellular Biochemistry*, 253(1-2):329-38.

Mastelic, J., Jekovic, I., Blazevic, I., Poljak-Blazi, M., Borovic, S., Ivancic-Bace, I., et al. 2008. Comparative study on the antioxidant and biological activities of carvacrol, thymol, and eugenol derivatives. *Journal of Agricultural and Food Chemistry*, 56(11): 3989-3996.

Memarzadeh, S. M., Ghasemi Pirbalouti, A. and Adibnejad, M. 2015. Chemical composition and yield of essential oils from Bakhtiari savory (*Satureja bachtiarica* Bunge.) under different extraction methods. *Industrial Crops and Products*, 76: 809-816**.**

Michiels, C., Raes, M., Toussaint, O. and Remacle, J. 1994. Importance of Se-glutathione peroxidase, catalase, and Cu/Zn-SOD for cell survival against oxidative stress. *Free Radical Biology and Medicine*,17(3): 235-248.

Moghadamtousi, S.Z., Rouhollahi, E., Karimian, H., Fadaeinasab, M., Abdulla, M.A. and Abdol Kadir, H. 2014. Gastroprotective activity of *Annona muricata* leaves against ethanol-induced gastric injury in rats via Hsp70/Bax involvement. *Drug Design, Development and Therapy*, 8:2099-110.

Rabiei, Z., Shirchi, M., Rafieian-kopaei, M. and Asgharzadeh, S. 2020. Effects of pretreatment with *Satureja Bachtiarica* essential oil in preventing seizure in pentylenetetrazol-kindled mice. *Basic and Clinical Neuroscience*. In press.

Rozza, A.L., Cesar, D.A.S., Pieroni, L., Saldanha, L.L., Dokkedal, A.L. and De-Faria, F.M. 2015. Antiulcerogenic activity and toxicity of *Bauhinia holophylla* hydroalcoholic extract. *Evidence-Based Complementary and Alternative Medicine*, 2015, Article ID 439506.

Rozza, A.L. and Pellizzon, C.H. 2012. Essential oils from medicinal and aromatic plants: a review of the

gastroprotective and ulcer-healing activities. *Fundamental & clinical pharmacology*, 27(2013):51-63.

Saghaei, F., Karimi, I., Jouyban, A. and Samini, M. 2012. Effects of captopril on the cysteamine-induced duodenal ulcer in the rat. *Experimental and Toxicologic Pathology* , 64(4):373-377.

Saghaei, F., Motamedi, S. 2018. Analgesic and anti-inflammatory effect of hydroalcoholic extract of *Savory Bakhtiari* on mice. *Journal of Herbal Medicine*, 8(3): 179-184.

Sefidkon, F. and Jamzad, Z. 2000. Essential oil of *Satureja bachtiarica* Bunge. *Journal of Essential Oil Research*, 12(5): 545-546.

Sefidkon, F., Sadeghzadeh, L., Teymouri, M., Asgari, F. and Ahmadi, S. 2007. Antimicrobial effects of the essential oils of two Satureja species (*S. Khuzistanica Jamzad* and *S. bachtiarica* Bunge) in two harvesting time. *Iranian Journal of Medicinal and Aromatic Plants*, 23(2): 182-191.

Shetty, R., Kumar, V., Naidu, M. and Ratnakumar, K. 2000. Effect of *Ginkgo biloba* extract in ethanol induced gastric mucosal lesions in rats. *Indian Journal of Pharmacology*, 32: 313-317.

Stahl-Biskup, E. and Sáez, F. 2002. Thyme: the genus Thymus. Taylor and Francis Group, *CRC Press*, pp 293-317.

Shin, I.S., Jeon, W.Y., Shin, H.K., Cha, S.W. and Lee, M.Y. 2013. *Banhabaekchulchunma-tang*, a traditional herbal formula attenuates absolute ethanol-induced gastric injury by enhancing the antioxidant status. *BMC Complementary and Alternative Medicine*, 170.

Tandon, R., Khanna, H.D., Dorababu, M. and Goel, R.K. 2004. Oxidative stress and antioxidants status in peptic ulcer and gastric carcinoma. *Indian journal of physiology and pharmacology*, 48(1):115-118.

Uroko, R.I., Chukwu, C.N., Egba, S.I., Adamude, F.A. and Ajuzie, J.C. 2020. Combined ethanol extract of *Funtumia africana* and *Abutilon mauritianium* leaves improves the lipid profile and kidney function indices of benign prostatic hyperplasia in rats. *Acta Scientiarum Polonorum, Technologia Alimentaria*, 19(4): 395-404.

Vemula, S.K., Chawada, M.B., Thakur, K.S. and Vahalia, M.K. 2012. Antiulcer activity of Amlapitta Mishran suspension in rats: A pilot study. *Ancient Science of Life*, 32(2):112-115.

Vismaya Belagihally, S.M., Rajashekhar, S., Jayaram, V.B., Dharmesh, S.M. and Thirumakudalu, S.K. 2011. Gastroprotective Properties of Karanjin from Karanja (*Pongamia pinnata*) Seeds; Role as Antioxidant and H, K-ATPase Inhibitor. *Evidence-Based Complementary and Alternative Medicine*, Article ID 747246.

Wandall, J.H. 1992. Effects of omeprazole on neutrophil chemotaxis, super oxide production, degranulation, and translocation of cytochrome b-245. *Gut*, 33(5): 617-621.

Xie, J.H., Chen, Y.L., Wu, Q.H., et al,. 2013. Gastroprotective and anti-Helicobacter pylori potential of herbal formula HZJW: safety and efficacy assessment. *BMC complementary and alternative medicine*, 13: 119.