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Anti-inflammatory and antioxidant potential of *Artemisia dracunculus* L. aqueous extract against acetic acid induced ulcerative colitis in male Wistar rats

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Ulcerative colitis (UC) is one of the two types of inflammatory bowel disease (IBD) which is increasing worldover due to modern life style. Patients with UC are prone to develop colorectal cancer. While the disease severity decides the treatment option, researchers look towards herbal medicines with anti-inflammatory properties for minimal or nil side effects. *Artemisia dracunculus* L., commonly called Tarragon, is a medicinal herb used in traditional Asian medicine mainly in Iran, India, Pakistan and Azerbaijan due to its special compounds. In this study, we tried to elucidate the effects of aqueous extract of tarragon on acetic acid induced ulcerative colitis (UC) in rats. Male Wistar rats were grouped into four groups of eight each viz., control; experimental control (UC was induced via luminal instillation of 4% acetic acid); and UC induced + aqueous tarragon extract (100 mg/kg) or prednisolone (2 mg/kg) orally for ten consecutive days. Tissue specimens were collected after the experimental period for evaluation of caspase-3 and cyclooxygenase-2 (COX-2) expression by immunohistochemistry. Real-time PCR was used to monitor the levels of IL-1, IL-6 and TNF-α in colonic homogenates. Moreover, the levels of myeloperoxidase, nitric oxide and total antioxidant capacity were measured in colonic homogenates. The results showed that both treatment regimens could similarly reduce the severity of disease symptoms. Treatment with aqueous extract of tarragon caused a better improvement (P <0.05) in the levels of myeloperoxidase enzyme, and total antioxidant capacity of colonic homogenates compared to prednisolone. Nevertheless, the levels of the expression of caspase-3, and COX-2 and TNF-α were reduced in UC rats received prednisolone more than UC rats received aqueous extract of tarragon. The was no statistical difference in the levels of nitric oxide, IL-1 and IL-6 between UC rats received tarragon extract or prednisolone. Overall, these findings suggest that the aqueous extract of tarragon is a promising strategy to control ulcerative colitis. Aqueous extract can also be used as an anti-inflammatory and immune system stimulant in conditions where the immune system is damaged.

Keywords: Caspase-3, Colorectal cancer, Cyclooxygenase-2, Disease activity index, Estragon, Inflammatory bowel disease (IBD), Tumor necrosis factor; Pro-inflammatory cytokines, Tarragon

Inflammatory bowel disease (IBD) is an inflammatory disease of the intestinal tissue that typically observed in two main types of ulcerative colitis (UC) and Crohn's disease[1]. The prevalence of inflammatory bowel disease (UC) is remarkably high, with an estimated 10,000 in every 25 patients in the West, and it is increasing in other parts of the world as well due to modern lifestyle[2]. Immunological functions, inflammation and genetic factors play a significant role in UC development. Based on empirical and theoretical evidence, this disease begins slowly and recurs gradually, resulting in a series of intestinal immune system disorders[3]. Clinical symptoms of UC are diarrhea, abdominal pain, and constipation. UC mainly affects the colon and the rectum, inflammatory response alter mucosal barrier function and intestinal integrity leading to apoptosis[2,4]. Patients with UC are more vulnerable to develop colorectal cancer[2]. Although the UC pathogenesis is not yet well understood, it is widely acknowledged that an imbalance between pro-inflammatory cytokines, such as TNF-α, and anti-inflammatory cytokines, such as IL-1 and IL-6, plays a pivotal role in the modulation of inflammation[5].

The development of experimental animal models was a breakthrough in IBD research[6], that contributed to the better design of clinical studies about specific components involved in IBD pathogenesis and immunological processes[7]. The first treatment of IBD is administering common steroid and nonsteroidal anti-inflammatory drugs (NSAIDs) to patients[8]. These drugs can cause side effects such as nausea, vomiting,

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hepatic and renal disorders, and thinning skin[9]. Moreover, all patients do not necessarily respond well to these therapeutic protocols[10]. Therefore, there is a need for less risky and more secure strategies for treating IBD[11]. Kayal & Shah[12] who reviewed various treatment strategies of UC recently, have observed that the treatment options vary with the disease severity. They have suggested to ensure informed decision making incorporating medication risks and the therapeutic benefits in patient discussions[12].

Considerable attention is paid to herbal medicines with anti-inflammatory properties due to their fewer side effects[10]. Additionally, most herbal extracts have strong antioxidant properties. Oxidative stress is an imbalance between oxidant and antioxidant factors, and has a possible effect on the development of UC[13].

Tarragon, also known as estragon, is a perennial herbaceous plant from genus *Artemisia* and the family *Asteraceae*. This species is endemic to Central and North Asia, the Caucasus, and the Far East. The alluvial valleys of Russia and the western regions of North America are the main origin of tarragon[10]. Anti-inflammatory and antioxidant activities of tarragon (*Artemisia dracunculus* L.) have already been proven[12]. Tarragon extract can cleanse free radicals[14]. This perennial herb is commonly used in traditional medicine[15,16], and its non-toxicity has also been proven[17]. It has also been shown that tarragon can present analgesic effects[18]. In traditional medicine, tarragon is used to treat cough, cold, fever, pain and menstrual problems. In Iranian traditional medicine, it is also used to treat seizures, coagulation disorders and hyperlipidemia. There are also reports about the antimicrobial and hepatoprotective effects of tarragon[19]. Froushani *et al.*[10] have reported the immunomodulatory properties of this plant. Only limited studies are available on the effects of tarragon on ulcerative colitis. Therefore, in the present study, we tried to assess the anti-inflammatory effects of aqueous extract of tarragon on the animal model of UC.

Materials and Methods

Preparation of plant extract

The new aerial parts of tarragon (*Artemisia dracunculus* L.) were gathered from Urmia, Iran, by an herbalist of the Faculty of Science (Roghaiyeh Zeinali), Urmia University (Herbarium code:512). The collected samples were carefully washed, cut into small pieces, and dried in the shade. A routine percolation method was used to prepare aqueous extract of the dried and milled plant. In the following, the extract was dried by evaporation at 42°C. Then, the extract was kept at –20°C away from light[10].

Animals

Thirty-two male Wistar rats weighing 100-200 g collected from Laboratory Animal Reproduction Center, Faculty of Science Urmia university, Urmia, Iran were reared in wire-bottomed cages under controlled condition (12 h light/dark cycles and 21-25°C) fed with standard laboratory animal pellet diet as well as tap water, *ad libitum*. After a week of accommodation, the rats were randomly divided into four groups of eight each. Group I (control) did not receive any agents during experiment period; Gr. II (experimental control) received luminal instillation of 4% acetic acid to induce ulcerative collis; and Gr. III & IV UC rats received tarragon extract (100 mg/kg P.O.)[10] and prednisolone (2 mg/kg P.O.), respectively. After 10 days, the rats were sacrificed and samples were prepared from the distal part of their intestine for microscopic evaluations.

Induction of UC with acetic acid

After 24 h of food restriction, the rats were gently anesthetized with ether and a baby catheter was inserted into their rectum to inject 1-2 mL solution of 4% acetic acid to the rectum. The rats were kept diagonally for 90 s to both help acetic acid absorption and prevent its removal of the rectum[20,21].

Evaluation of disease symptoms

As shown in Table 1, all animals were daily examined for changes in body weight, stool consistency and blood in the stool. The disease activity index (DAI) was equal to the sum of daily scores obtained from Table 1. At the end of the study, the experimental groups were compared in terms of the daily and total mean DAI[10].

Homogenization of tissue samples

A 10 cm sample of the rectum was taken from each rat and thoroughly washed with normal saline. Then normal cold saline was added to samples as ten times their volume. After homogenization, samples were centrifuged at 12000 g for 10 min at 4°C[22].

Table 1 — Scoring system for evaluation of UC rats

Score	Stool consistency	Blood feces	Weight loss
0	Normal	Negative	Negative
1	Soft	Red	1-9%
2	Very Soft	Dark Red	10-19%
3	Diarrhea	Black	<20%

[The disease activity index (DAI) was expressed as the sum of scores of all criteria]

Measurement of myeloperoxidase (MPO) activity in tissue samples

According to the protocol, 10 µL of homogenized colon tissue were mixed with 110 µL of TMB solution (2.9 mM TMB in 14.5% DMSO plus 150 µM sodium sulfate buffer at pH 5.4) and 80 µL of 0.75 µM hydrogen peroxide. Then, samples were incubated at 37°C for 5 min. Next, for termination of the reaction, 50 µL of 2 mM sulfuric acid was added to each chamber and the absorbance was determined by an Elisa Reader at 450 nm. Finally, a standard curve was drawn to estimate MPO activity in tissue samples[23].

Measurement of nitric oxide (NO) production in tissue samples

The Griess test was employed to measure the intensity of nitric oxide production in tissue samples. According to the procedure, 50 µL of Griess reagent (3% phosphoric acid, 0.1% naphthylethylenediamine and 0.1% sulfanilamide) were mixed with 50 µL of a homogenized tissue sample. The resulting mixture was incubated for 10 min in the dark at room temperature (25°C). The optical density of samples was measured by a standard microplate reader at 540 nm. Finally, a standard curve was generated for each set of samples assayed[24].

Measurement of total antioxidant capacity (TAC)

The TAC of biomolecules in different samples was determined by the ferric reducing ability of plasma (FRAP) based on the single-electron transfer mechanism by the reaction-caused color change at 593 nm. The numerical value of TAC can be estimated from the standard curve. In practice, 20 µL of the homogenized colon sample (as the standard) to 1 mL of the working solution was added and vortexed. In the following, the optical density (OD) of samples was read at times zero and 4 min at 593 nm, compared to the blank solution (working solution). Then optical density values were inserted into the TAC formula to obtain numerical values of TAC[25].

Assessment of Malondialdehyde (MDA)

The level of MDA in the colonic homogenates was followed similarly to the procedure described earlier[26]. In brief, 5 mL reaction buffer (0.25M HCl, 0.37% thiobarbituric acid, and 15% trichloroacetic acid, 1:1:1 ratio) was mixed to 200 µL of gut homogenate, and warmed at 95°C about 1 h. Thereupon, the admixture was centrifuged at 4000 g for 15 min. Finally, a microplate reader at a wavelength of 540 nm was used to monitor the absorbance of the admixture. Values were presented as nM of MDA/mg protein.

Expression of IL-6, IL-1 and TNF-α

The expression level of IL-6, IL-1 and TNF-α were measured through real-time PCR. To extract total RNA using Trizol, 1000 µL of Trizol was added to the sample and the resulting mixture was homogenized. Then, 200 µL of chloroform and the resulting mixture was vortexed for 4-5 min, incubated on ice for 15 min, and centrifuged at 1200 g for 5 min. The liquid portion was added to 500 µL of iso-propanol in an Eppendorf tube, and then it was incubated on ice for 5-10 min and centrifuged at 1200 g for 5 min. The supernatant was separated and washed with 500 µL of 70% ethanol, and the resulting mixture was centrifuged at 7500 g for 10 min and then air-dried. Depending on the extracted RNA volume, DEPC-treated water was added to the mixture and RNA sediments were dissolved by pipetting. To synthesize cDNA, total RNA extract was mixed with other components of the kit in an RNase-free tube (according to the manufacturer's protocol) in a vortex, and then the resulting mixture was incubated at 25°C for 10 min and at 47°C for 60 min. The reactions were stopped by heating the mixture at 85°C for 5 min following by cooling on ice or at 4°C. Each thermal cycler program included initial step of template denaturation at 95°C for 15 s to 2 min, 45 cycles of denaturation at 95°C for 30 s, primer annealing at 50-60°C for 30 s, and primer extension at 72°C for s, and final extension at 75°C for approximately 1-2 min[28]. The designed primers were used according to Table 2.

Measurement of cyclooxygenase 2 (COX2) and caspase 3

The level of COX2 and caspase 3 in tissue samples was examined through the immunohistochemistry (IHC) staining method[29]. For this purpose, tissue sections of 5 µm in thickness were prepared and put in

Table 2 — List of primer sequences used for Real-time PCR analysis

Gene	Forward primer	Reverse primer
IL-1	5'-AAGACAAGCCTGTGTTGCTGAAGG	5'-TCCCAGAAGAAAATGAGGTCGGTC
TNF-α	5'-AAATGGGCTCCCTCTCATCAGTTC	5'-TCTGCTTGGTGGTTTGCTACGAC
IL-6	5'-TCCTACCCCAACTTCCAATGCTC	5'-TTGGATGGTCTTGGTCCTTAGCC
GAPDH	5'-GTATTGGGCGCCTGGTCACC	5'-CGCTCCTGGAAGATGGTGATGG

an oven at 60°C for 25 min. The samples were deparaffinized in xylene and then dehydrated using different degrees of ethanol. For antigen retrieval, tissue slides were placed in 10 mM sodium citrate buffer at pH 7.2. In the next step, the production level of the intended proteins was examined using special kits. Based on the manufacturer's protocol (Kit of Pars Tous, Mashad, Iran), after fixing the tissue slides following the antigen retrieval process, they were put in a microwave at 720 mV for 3 min and then at 180 mV for 10 min. The slides were incubated with oxygenated water for 5 min to suppress peroxidase activity. In the next step, tissue sections were washed with a buffer before incubating them with droplets of a super blocking solution to block avidin and biotin. After washing the tissue sections with phosphate-buffered saline (PBS), they were re-incubated with the primary antibody for 18 h at 4°C. Once again, the tissue sections were washed with PBS and incubated at room temperature with secondary antibody for 10 min. Further, the tissue slides were washed with PBS and incubated with a chromogen called diaminobenzidine (DAB) for 15 min. The samples were rewashed and immersed in a hematoxylin solution for 10 s. Finally, the tissue sections were washed with distilled water and covered with a lamella. The level of COX2 and caspase3 in positive cells were estimated at one mm^2 of the colon tissue.

Results

Following instillation of acetic acid into the rectum, the symptoms of UC were observed in affected rats (Fig. 1A). The statistical analysis of DAI indicated that treatment with prednisolone led to a significant regression in the cumulative disease score from the fourth day after acetic acid instillation into the colonic lumen of the rats. This period was started from day sixth after induction of disease for UC rats received tarragon aqueous extract (Fig. 1A). Nevertheless, at the end of the investigation, there was no significant difference between the average mean cumulative disease score of UC rats received tarragon aqueous extract or prednisolone (Fig. 1B). Therefore, both tarragon aqueous extract and prednisolone were equally effective in reducing DAI (Fig. 1).

The evaluation of MPO activity showed that the induction of UC significantly increased the MPO level. Oral administration of tarragon aqueous extract to the UC rats reduced the MPO activity, reaching to the MPO activity in healthy rats. Prednisolone did not perform much successfully and failed to return the MPO activity to the normal level (Fig. 2A). As expected, the induction of UC significantly increased NO production in homogenized tissue samples. Both tarragon aqueous extract and prednisolone reduced NO production in tissue samples almost equally (Fig. 2B).

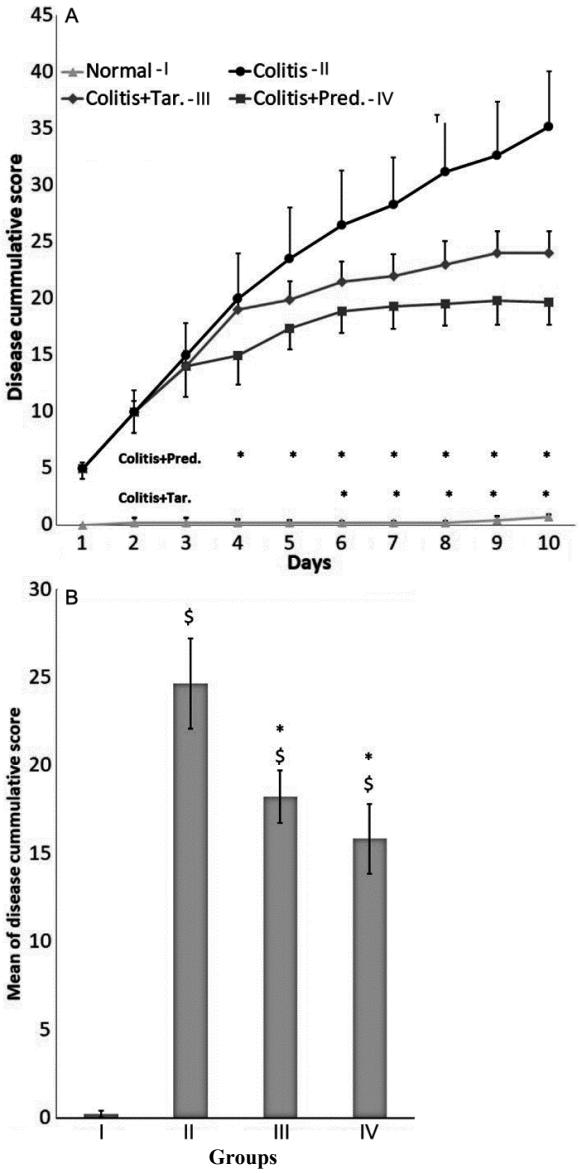

Fig. 1 — Assessment of clinical schema of ulcerative colitis (UC) rats. Acetic acid-induced UC rats received aqueous extract of tarragon or prednisolone as detailed under Materials and Methods. (A) Mean cumulative disease score; and (B) Average mean cumulative disease score. [Findings are presented as mean ±S.D. P <0.05 *vs.* healthy rats; *P <0.05 *vs.* PBS-treated UC rats. Gr. I, Normal; Gr. II, Colitis; Gr. III, Colitis+Tar; and Gr. IV, Colitis+Pred. Tar., Aqueous extract of tarragon; Pred., Prednisolone]

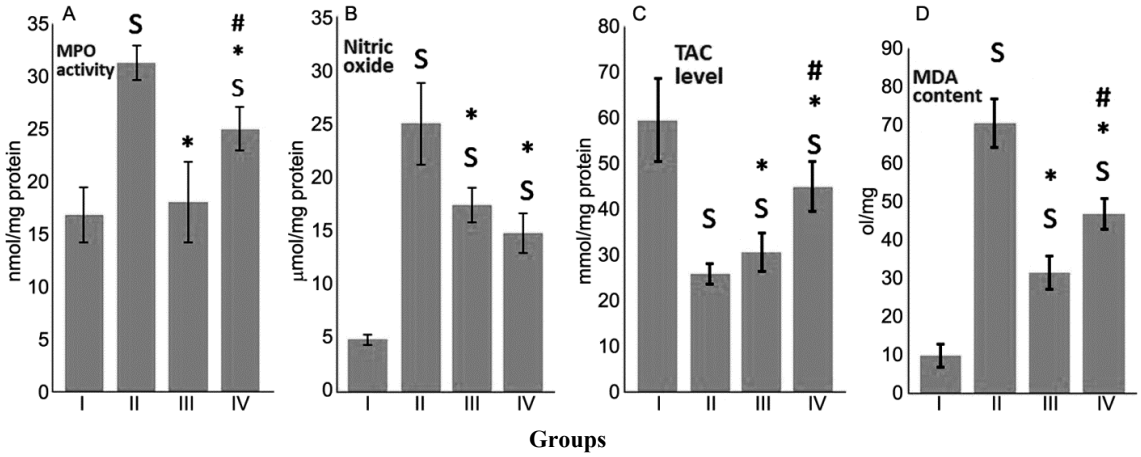

Fig. 2 — Biochemical changes in the gut homogenate of UC rats. (A) MPO activity; (B) Nitric oxide level; (C) TAC level; and (D) MDA content. Gr. I, Normal; Gr. II, Colitis; Gr. III, Colitis+Tar; and Gr. IV, Colitis+Pred. [Data are reported as mean ±S.D. (P <0.05 *vs.* healthy rats; *P <0.05 *vs.* PBS-treated UC rats; P <0.05 *vs.* Tarragon treated UC rats; P <0.05 *vs.* Prednisolone treated rats). Tarr., aqueous extract of tarragon; Pred., Prednisolone]

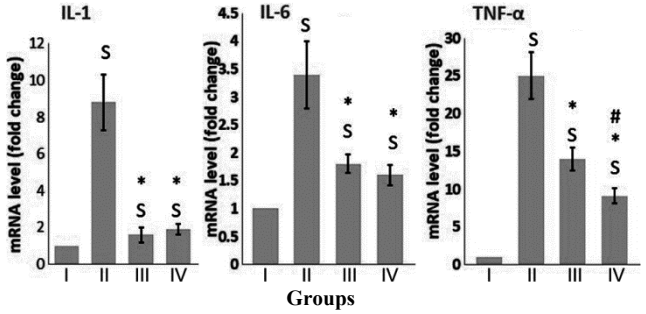

Fig. 3 — Effect of therapeutic regimens on the production of inflammatory cytokines in the colonic homogenate of experimental rats. Gr. I, Normal; Gr. II, Colitis; Gr. III, Colitis+Tar; and Gr. IV, Colitis+Pred. [The values are shown as mean ±S.D. (P <0.05 *vs.* healthy rats; *P <0.05 *vs.* PBS-treated UC rats; & P <0.05 *vs.* Tarragon treated UC rats; P <0.05 *vs.* Prednisolone treated rats). Tarr., aqueous extract of tarragon; Pred., Prednisolone]

The results also indicated that TAC significantly reduced in affected rats compared to healthy ones. Although both tarragon aqueous extract and prednisolone could increase TAC levels, tarragon aqueous extract could significantly increase the TAC levels more than prednisolone.

Accordingly, there is no significant difference in total antioxidant capacity between UC rats treated with tarragon or treated with prednisolone (Fig. 3C). As shown in the 3D figure, there was a significant increase in the amount of MDA in the gut tissues of UC rats compared to healthy cases. Treatment with aqueous extract of tarragon caused a more profound decrease in the MDA content in the gut homogenate compared to UC rats treated with Prednisolone.

In this study, IL-6, IL-1, and TNF-α expression were assessed via real time PCR (Fig. 4). The results demonstrated that the expression of inflammatory cytokine genes in intestinal tissue significantly increased in the UC rats. Both treatments with tarragon aqueous extract and prednisolone could similarly suppress the expression of IL-6 and IL-1 compared to UC rats without treatment (Fig. 4). In addition, the data showed that treatment with prednisolone resulted in a significantly greater reduction in TNF-α levels than treatment with tarragon aqueous extract (Fig. 4). The results of IHC staining for measurement of caspase-3 in the gut tissue samples showed that the induction of UC sharply increased the caspase-3 level, as logically expected. Although both herbal extract and prednisolone reduced the level of caspase 3, prednisolone is much more successful in this regard than extract (Fig. 4 A-E). The results also indicated that the Cox-2 level increased in the UC rats. Both tarragon aqueous extract and prednisolone significantly reduced the Cox-2 level. However, this reduction was more prominent in UC rats received prednisolone compared to UC rats treated with tarragon aqueous extract (Fig. 4 F-J).

Discussion

The present investigation evaluated *A. dracunculus* extract as a novel treatment for the animal model of ulcerative colliits (UC). Findings indicated that the oral administration of aqueous extract of tarragon markedly decreased the clinical score of rats with UC. The immunomodulatory benefits of the aqueous extract of tarragon have been documented previously. Accordingly, the aqueous extract of *Artemisia*

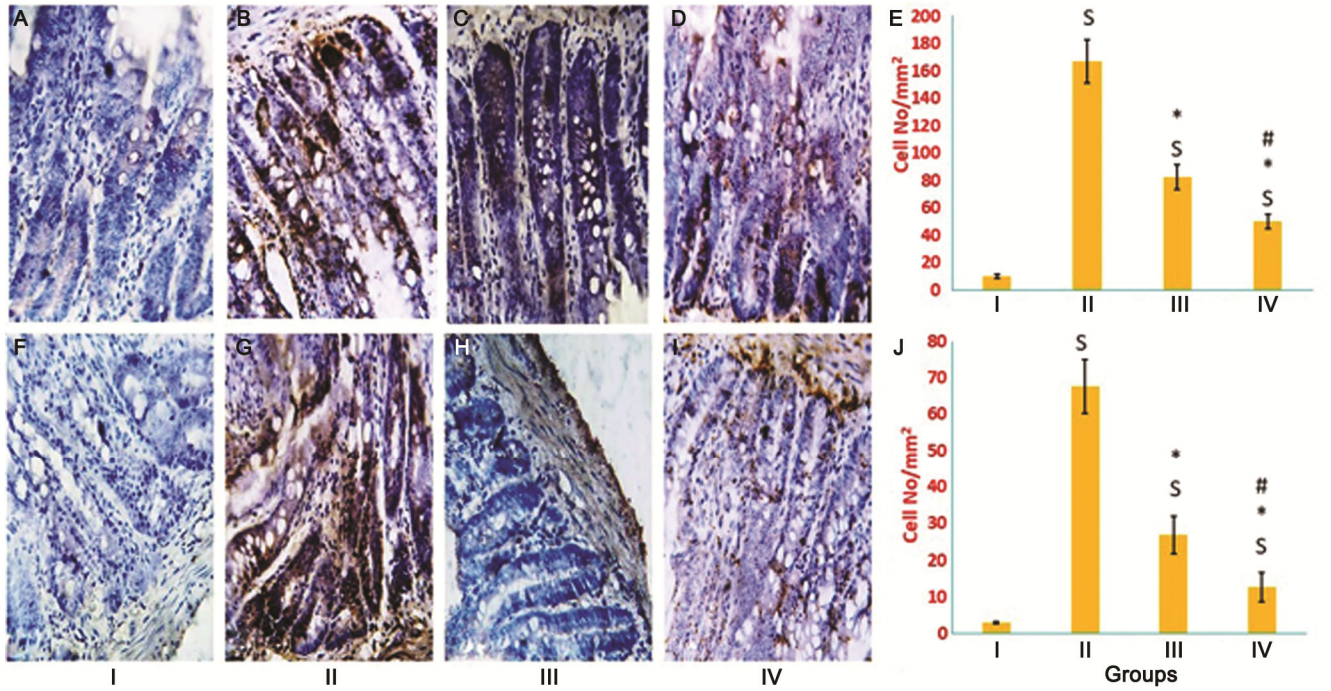

Fig. 4 — Evaluation of caspase-3 and COX-2 expression by immunohistochemical staining of colonic specimens. Representative sections illustrated the immunohistochemical staining ofcaspase-3(A-D) and COX-2 (F-I) in the colonic specimens (400X). The count ofcaspase-3(D) and COX-2 (J) positive cells were determined inone mm^2 of the colonic tissues. Gr. I, Normal; Gr. II, Colitis; Gr. III, Colitis+Tar; and Gr. IV, Colitis+Pred. [The values were shown as mean ±S.D. (*P* <0.05 *vs.* healthy rats; **P* <0.05 *vs.* PBS-treated UC rats; *P* <0.05 *vs.* Tarragon treated UC rats; *P* 0.05 *vs.* Prednisolone - treated rats). Tarr., aqueous extract of tarragon; Pred., Prednisolone]

dracunculus can inhibits the production of inflammatory cytokines (IL-17 and IFN-γ) and promote anti-inflammatory macrophages in mice immunized with sheep red blood cells[15]. IL-17 and IFN-γ, along with macrophages with inflammatory phenotype, play an essential role in the formation and spread of ulcerative colitis[30]. Previous findings have shown that estragole, methyl-eugenol, coumarins, tannins, alkaloids,flavonoids, thymol, vitamins,and phenolic carbonic acids are the main bioactive compounds of tarragon[31,32]. The Committee on Herbal Medicinal Products (HMPC) warned that Stragol and Methyl Eugenol may be carcinogenic and genotoxic. A novel study reported that the aqueous extract of tarragon, similar to what we used in this study, is safe because it does not contain detectable amounts of these two potentially harmful compounds[10]. On the other hand, Prednisolone is a common glucocorticoid that up-regulates the expression of anti-inflammatory proteins, and simultaneously down-regulates the expression of pro-inflammatory proteins[33]. Stool status is one of the most important indicators for assessing the severity and progression of UC. Stool tests provide valuable information about consistency, the existence of blood, epithelial cell debris, mucous volume, and more.

Based on clinical studies, the stool contained some blood and lacked consistency in rats afflicted with acute UC. Moreover, watery stools can be attributed to improper absorption of water due to inflammation and destruction of the colon epithelium, which disrupted food absorption and excretion[34]. The study results indicated that stool consistency was higher in rats of the control group, and no bloody stool was pointed out in their samples. By contrast, watery stool and bloody stool were revealed in samples taken from affected rats in the other three groups. Administration of tarragon aqueous extract and prednisolone improved these conditions in some cases. However, since the colon mucosa remained unrepaired, the disease symptoms persisted in some other cases. The findings demonstrated that both tarragon aqueous extract and prednisolone were effective in reducing DAI.

Acetic acid destroys tissue barriers by acidifying the environment, and thus the bacterial flora of the intestine leaks into the underlying layers and causes inflammation. Then immune cells, such as neutrophils, infiltrate into the mucosa, and macrophages accumulate at the site, which is an essential symptom of UC[35]. In IBD, under condition of tissue damage, innate immune cells produce high

levels of proteases and reactive oxygen species (ROS), such as anion superoxide and hydrogen peroxide, and active nitrogen species, such as NO. These mediators play an important role in eliminating pathogens. The intestinal mucosa is equipped with an antioxidant system to resist oxidative and nitrative damages[36]. Unfortunately, this antioxidant system is disturbed due to the excessive production of radicals or their production in unfavorable conditions during IBD[37]. These factors cause lipid peroxidation, increase mucosal permeability and blood vessels, increases the entry of neutrophils, and exacerbate inflammation[38]. Therefore, the use of antioxidants can be useful in the treatment of IBD[4]. Previous studies have shown that the methanolic extract obtained from tarragon leaves exhibits strong antioxidant properties and can scavenge oxygen free radicals[19]. Tarragon aqueous extract is an herbal extract with antioxidant properties, whereas prednisolone cannot directly suppress free radicals. The findings revealed that this property led to the elimination of free radicals and reduced TAC to a healthy level in the groups treated with herbal extract. Since prednisolone lacks this property, TAC in the groups treated with prednisolone was the same as that of those in affected groups that received no treatment.

MDA is one of the excellent predictors of lipid peroxidation[11]. The better results of tarragon in reducing MDA levels are also due to the direct antioxidant properties of tarragon. On the other hand, prednisolone is only able to indirectly reduce the level of malondialdehyde by reducing inflammatory mediators.

NO is the main framework of free nitrogen radicals produced by immune cells following inflammation. The results showed that the NO level increased after the induction of UC. However, there was no significant difference between the treated groups. The herbal extract and prednisolone managed to reduce NO to be the same as that of the control group.

After the recruitment of neutrophil into the acetic acid inflamed colon, the MPO enzyme produces oxygen free radicals and plays a meaningful role in inflammatory processes and oxidative stress. MPO discharged because of inflammation in peripheral matrices, the best way to investigate its activity in inflamed tissue is to examine the entire tissue. As a result, the MPO level is considered one of the indicators of acute inflammation[39]. The data obtained in the current study showed that the plant extract was better in reducing MPO than prednisolone, as MPO level in the animals treated with herbal extract reached that of the control group. Better performance of tarragon in MPO reduction can probably be attributed to its antioxidant and anti-inflammatory properties.

Colonic instillation of acetic acid promoted the production of potent inflammatory cytokines (like IL-1, IL-6 and TNF-α), which are known as the targets of curative interventions. These inflammatory cytokines play a significant role in inflammation as well as the activation of leukocytes, such as neutrophils[40]. A previous review demonstrated that aqueous extract of tarragon could modulate the immune system and production of pro-inflammatory cytokines in a multiple sclerosis mouse model[41]. Moreover, the treatment of UC with anti-TNFs effectively reduces inflammatory cell infiltration and negatively regulates IL-18, TNF, and INF-ϒ secretion. It has also been shown that although local TNF production in the UC increases TNBS, treatment of the disease with anti-TNFs can positively affect this process[42]. These cytokines trigger the activity of other inflammatory cytokines, accelerating the disease progression[43]. Interestingly, flavonoid metabolites, which find in tarragon extract, reduce TNF-α secretion in human THP-1 monocytes[44]. Various cells can produce IL-6 and apply pleiotropic effects on different organs. It has been observed that this inflammatory mediator is involved in causing various inflammatory diseases such as UCCrohn's disease[45]. The study results demonstrated that both tarragon aqueous extract and prednisolone almost equally suppressed the expression of IL-1 and IL-6. According to TNF-α, although the extract and prednisolone was successful in suppressing this gene, prednisolone was better than tarragon.

Caspase 3 protein interacts with caspase 8 and caspase 9 in cells. It is encoded by the CASP3 gene and activated by caspases 8, 9 and 10. Caspase 3 also activates caspase 6 and caspase 7. This enzyme is activated in the apoptotic cell both by death ligand like TF-super family and, mitochondrial pathways[46]. This study also indicates that the caspase level increased after induction of UC to the rats; In contrast, there was no increase in caspase expression in the control group. Although both plant extract and prednisolone reduced the caspase levels in intestinal tissue, prednisolone is more effective than tarragon in this regard. However, none of them managed to reduce the caspase level to reach that of the control group. Today, anti-inflammatory drugs such as

glucocorticoids and 5-aminosalicylates are prescribed to control UC. These drugs can inhibit both forms of cyclooxygenase (COX1 and COX2) involved in the spread of inflammation[47]. Targeting selectivity for COX2 isoenzyme can reduce some of the adverse effects of these medications like the risk of peptic ulceration[10]. The study results showed that the COX2 level increased after the induction of UC, but both tarragon aqueous extract and prednisolone reduced it. However, the expression level of COX-2 gene in UC rats treated with prednisolone decreased more than tarragon aqueous extract.

Conclusion

Results of this study has demonstrated that aqueous extract of the herb tarragon (*Artemisia dracunculus* L.) reduced the symptoms of ulcerative collis (UC) similar to prednisolone, and also improved myeloperoxidase enzyme level and antioxidant capacity of colon homogenate more than prednisolone. However, the expression levels of caspase, COX2, and TNF-α in UC rats receiving prednisolone decreased more than those in UC rats receiving tarragon aqueous extract. The findings indicate possible use of aqueous extract of tarragon in the treatment of inflammatory diseases either alone or in combination with other treatments.

Conflict of Interest

Authors declare no competing interests.

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