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## Research Article Effect of Fenugreek (*Trigonella foenum-graecum* L.) Seeds and its Aqueous Extract on Gastric-Acidity and Blood pH of Rats

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### Abstract

**Background and Objective:** Fenugreek seeds are supposed to be of practical importance as a source of phytochemicals that are of relevance to the pharmaceutical industry. The objective of the present investigation was to explore the utility of Fenugreek Seeds Powder (FSP) and its aqueous extract FSPE as a natural anti-acidity, anti-osteoporosis instrument in rats. **Materials and Methods:** Forty male rats weighing 190±10 g were randomly divided into five groups, including the negative control group and groups that were fed on 5 and 10% of FSP or with 2 mL of its aqueous extract. Rats were sacrificed and serum was collected for biochemical analysis. **Results:** Results revealed a significant increase in final weight and body weight gain in the fourth group compared with the other groups. The positive control group recorded the lowest level of bicarbonate and there was a significant increase in the bicarbonate levels in rats that received aqueous extract compared to the negative control. It was also noted that stomach acidity and blood pH were affected positively by FSP and FSPE treatment and there was a significant difference between the positive group and the other groups. Rats fed on a diet supplemented with 10% fenugreek seeds powder presented the highest mean value of serum calcium. The addition of FSP and FSPE to the rat's diet reduced blood malondialdehyde and increased total antioxidant capacity and maintained the gastric layers and bones histology intact. **Conclusion:** Results confirm a protective role of Trigonella against gastric ulcer and blood pH, which can be attributed to the probable effect of dietary fibre and antioxidant compounds.

Key words: Fenugreek seeds, gastric hyperacidity, aqueous extract, anti-osteoporosis, antioxidative indices, saponins, mucilaginous fibre

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

#### INTRODUCTION

Food plays an important role in health and there is a growing common opinion that promotes a lifestyle where food is used as a form of medicine<sup>1</sup>. The prevalence of complementary and alternative medicine in the worldwide population is increasing. Some may attribute it to the expensive costs of conventional medicine that undeveloped countries can hardly afford. A variety of medicinal plants and their extracts (leaves, seed, peel or core) have been reported for their significant activity in animal models against induced diseases. Currently, about 80% of the world population depends on plant-derived drugs.

In this respect, fenugreek seeds have been experimentally reported to have several therapeutic advantages and it was found that the daily dietary intake of fenugreek seeds might produce essential nutritive value, in addition to a high level of efficiency and safety in obesity treatment<sup>2</sup>. Many pharmacological activities have been investigated to clarify the therapeutic properties of fenugreek and its main metabolites<sup>3</sup>. Some studies have been carried out on extracts from different plant parts (seeds, leaves) or pure phytochemicals (saponins, steroids, alkaloids) to validate traditional uses of fenugreek as an herbal remedy.

Fenugreek (Trigonella foenum-graecum L.) is an annual herb that belongs to the family Leguminosae. The seeds of Fenugreek are commonly used in India and oriental countries as a spice in food preparations due to their strong flavour and aroma<sup>4</sup>. The seeds are also used as herbal medicine in many parts of the world for their carminative, tonic and aphrodisiac effects. Fenugreek seeds contain 45-60% carbohydrates, mainly mucilaginous fibre; 20-30% proteins high in lysine and tryptophan; 5-10% fixed oils, pyridine-type alkaloids, mainly trigonelline (0.2-0.36%), choline (0.5%), gentianine and carpaine; the flavonoids apigenin, luteolin, orientin, guercetin, vitexin and isovitexin; free amino acids, such as 4-hydroxyisoleucine (0.09%); arginine, histidine and lysine; calcium and iron; saponins (0.6-1.7%); glycosides yielding steroidal sapogenins on hydrolysis and sitosterol; vitamins A, B<sub>1</sub>, C and nicotinic acid and 0.015% volatile oils, which are thought to account for many of its presumed therapeutic effects⁵.

The main chemical constituents responsible for the anti-inflammatory activity are alkaloids, saponins and flavonoids<sup>6</sup>. Fenugreek seeds administration and its extracts significantly decreased plasma cholesterol, triglyceride and Low-Density Lipoprotein (LDL)-cholesterol. However, High-density lipoprotein (HDL)-cholesterol level was found to be

constant and not affected<sup>7</sup>. Administration of aqueous extract of fenugreek seeds increased antioxidant levels and prevented a further rise in lipid peroxidation. Histopathological studies related to the rat liver and brain revealed the protective role of the seeds extract against ethanol-induced toxicity<sup>8</sup>. A study performed by Abdel-Latif<sup>9</sup> showed an increase in the bodyweight of rabbits supplemented with fenugreek in alloxan monohydrate induced diabetic rabbits. Plasma glucose level was reduced by the oral administration of fenugreek seeds powder not only in diabetic rabbits but also in non-diabetic rabbits. The hypoglycemic effect is due to the soluble fibre, which decreases the rate of gastric emptying, thereby delaying the absorption of glucose from the small intestine<sup>10</sup>.

Dietary fibre from fenugreek blunts glucose and cholesterol after a meal and regulates the production of cholesterol in the liver. The mechanisms for these effects have not been fully elucidated. Fenugreek seeds contain 45.4% dietary fibre (32% insoluble and 13.3% soluble) and the gum is composed of galactose and mannose. The latter compounds are associated with reduced glycemia and cholesterolemia. Fenugreek's hypoglycemic effect has been especially documented in humans and animals with type 1 and type 2 diabetes mellitus. In addition, this dietary fibre has the potential for widespread use in the food industry because its galactomannan component has emulsifying and stabilizing properties<sup>11</sup>. Gastric acid has an important pathophysiological role in humans. Numerous methods have been evaluated over the years in an attempt to measure gastric acid and stomach acidity, to study the role of gastric acid in gastrointestinal diseases in humans and to evaluate the effects of acid-suppressing drugs<sup>12</sup>.

The present study was designed to evaluate the protective effects of fenugreek seeds and their aqueous extract on gastric ulcer, stomach and blood pH, bone pain, kidney and liver failure in hyperacidity rats.

#### **MATERIALS AND METHODS**

**Place and study duration:** This research was conducted during the period extending from September, 2019-February, 2020 in the Department of Food Science and Human Nutrition, Qassim University, KSA.

**Reagents and chemicals:** All chemicals were purchased from Sigma-Aldrich (St. Louis, MO) Chemical Co. The standard reagent kits were from (bio-Merieux Laboratory Reagents and Products, France.

#### J. Biol. Sci., 21 (2): 70-79, 2021



Fig. 1: Flow chart of experimental design

**Plant materials:** Dried fenugreek seeds (*Trigonella foenumgraecum* L.) were obtained from a local market in the Qassim region, KSA and subjected to grinding to obtain a fine powder. White bread was obtained from a local market, roasted at 100°C and ground to powder.

**Animals:** Forty male Wistar albino rats  $(190\pm10 \text{ g})$  were obtained from the College of Pharmacy, University of King Saud and Riyadh, Saudi Arabia. The animals were housed in clean poly acrylate plastic cages  $(38\times23\times10 \text{ cm})$  and allowed to acclimatize to the laboratory environment for one week.

**Basal diet:** The basal diet obtained from the Faculty of Pharmacy, King Saud University, KSA, including casein 14%, corn oil 10%, salt mixture 4%, vitamin mixture 1%, cellulose 5%, DL-Methionine 0.7%, choline bitartrate 0.3% and starch 65%. The compositions of salt and vitamin mixtures were applied according to Kumar<sup>13</sup>.

**Preparation of fenugreek aqueous extract:** About 25 g of fenugreek-powdered seeds were extracted with 500 mL boiling distilled water for 5 min. The heated decoction was taken and allowed to cool for 30 min, at room temperature and filtrated twice. The filtrate was lyophilized and stored in a refrigerator. The rat dose of aqueous extract was 2.0 mL kg<sup>-1</sup> b.wt. orally administered by stomach tube.

Experimental design: After a period of adaption (7 days), rats were randomly classified into five groups (n = 8). The first group fed on standard rat chow and intended as a negative control (-C), while other groups were fed on a basal diet supplemented with 15% white bread for two weeks to induce the gastric ulcer. The experimental groups were subjected to the following treatments given in Fig. 1: group (2) continued on supplemented diet and named as a positive control (+C). The third group was fed on the supplemented diet and 5% seeds powder (5% FSP). The fourth group was fed on a supplemented diet and 10% seeds powder (10% FSP). The fifth group was fed on a supplemented diet and received orally 2.0 g kg<sup>-1</sup> b.wt. of seeds aqueous extract (FSPE). The guidelines of the Saudi National Committee of Bio and Med Ethics for laboratory animal care was adopted for handling the experimental animals and ethical approval was also obtained from the Research Ethics Committee at Qassim University, KSA.

**Biological evaluation:** At the end of the experiment period (6 weeks), rats were fasted overnight anaesthetized by diethyl ether, bled and sacrificed. Blood samples were collected and centrifuged at 3000 rpm for 10 min. Serum was collected and stored at -20°C until analysed.

#### Chemical's analysis

**Calcium content:** Calcium was determined according to Robertson and Marshall<sup>14</sup>.

**Phosphorus content:** Phosphorus was determined in serum according to the colorimetric methods as described by Tubino<sup>15</sup>.

**pH value:** pH value of the blood and stomach were determined by direct immersion of pH electrode in blood and gastric fluid at room temperature (25°C) using the digital pH meter model (3020 Dunmou, Jenway, Essex, UK) according to the method described by Ghosh<sup>12</sup>.

**Bicarbonate (HCO<sub>3</sub><sup>-</sup>):** Bicarbonate was determined in serum according to Rifai<sup>16</sup>.

**Fenugreek seeds chemical analysis:** Proximate analysis was carried out according to the method of Hooda and Jood<sup>17</sup>. The total Polyphenolic Content of fenugreek seeds was determined using a spectrophotometer (Secomam, France) based on the method by Singleton<sup>18</sup>. The total flavonoid content of fenugreek seeds was determined by the aluminium chloride colorimetric method of Seasotiya<sup>19</sup>. Total Antioxidant Capacity (TAC) was determined in blood serum according to the method described by Koracevvic<sup>20</sup>.

Lipid peroxides were determined in serum as malondialdehyde MDA according to Namıduru<sup>21</sup>. The results of the MDA measurements were given as nmol mL<sup>-1</sup>.

**Histopathological examinations:** Autopsy samples were taken from the stomach of rats in different groups and fixed in 10% formal saline for 24 hrs. Washing was done in tap water then serial dilutions of alcohol (methyl, ethyl and absolute ethyl) were used for dehydration. Specimens were cleared in xylene and embedded in paraffin at 56°C in a hot air oven for 24 hrs. Paraffin bees wax tissue blocks were prepared for sectioning at 4 microns' thickness by sledge microtome. The obtained tissue sections were collected on glass slides, deparaffinized and stained by hematoxylin and eosin stain for routine examination through the light electric microscope<sup>22</sup>.

**Statistical analysis:** Data was presented as Mean±standard error of the mean. Analysis was performed using the one-way ANOVA test along with the Duncan test will be performed using the SPSS software (Chicago, IL, USA). All data will be subjected to statistical analysis according to the procedure reported by Steel<sup>23</sup>.

#### RESULTS

**Chemical composition, antioxidant contents and activity of fenugreek seeds:** The data of Table 1 shows the chemical compositions of fenugreek seeds. Fenugreek Seeds (FS)

Table 1: Chemical composition and antioxidant components of fenugreek seeds

Parameters	Fenugreek seeds
Moisture (%)	9.63±0.39
Protein (%)	25.36±1.29
Fat (%)	5.45±0.37
Ash (%)	3.38±0.29
Total carbohydrate (%)	54.78±1.21
Total fiber (%)	46.44±1.1
Vitamin C (mg/100 g)	45.00±1
Polyphenol (mg/100 g)	251.71±1.29
Flavonoids (mg/100 g)	398.70±1.21
lron (mg/100 g)	35.50±1.2
Calcium (mg/100 g)	177.00±2.3
Phosphorus (mg/100 g)	299.00±3

Table 2: Mean values±SE of bicarbonate, stomach pH and blood pH for the experimental rats

Groups			Bicarbonate	
	Blood pH	Stomach pH	(mmol L <sup>-1</sup> )	
Group 1 (-C)	7.10±0.14ª	4.20±0.34ª	26.20±0.37ª	
Group 2 (+C)	$7.01 \pm 0.11^{b}$	$3.50 \pm 0.41^{b}$	23.56±0.31 <sup>b</sup>	
Group 3 (5% FSP)	$7.07 \pm 0.07^{a}$	3.92±0.31ª	26.44±0.51ª	
Group 4 (10% FSP)	7.15±0.15ª	3.94±0.21ª	26.15±0.40ª	
Group 5 (2% FSPE)	$7.21 \pm 0.06^{\circ}$	3.98±0.26ª	26.38±0.58ª	
Significance	*	*	**	

-C: Negative control, <sup>+</sup>C: Positive control, FSP: Fenugreek seeds powder, FSPE: Fenugreek seed powder extract. Means in the same column within each classification bearing different letters are significantly different (p<0.05 or 0.01). NS: Not significant, \*Significant (p<0.05), \*\*Significant (p<0.01)

contents of protein, dietary fibre, fat, ash and moisture were 25.36, 46.44, 5.52, 3.4 and 9.7%, respectively. The polyphenol, flavonoids contents are also presented in Table 1.

**Final weight, body weight gain and feed intake:** Data presented in Fig. 2 illustrates the impact of treatments on initial body weight, Final Body Weight (FW) and Body Weight Gain (BWG) of the experimental groups. It was noticed that only FW and BWG were significantly (p<0.05) affected by the treatments. However, the other parameters were not statistically different. It worth noting that the use of 5 and 10% FSP gave the best values of FW (229.6 and 286 g, respectively) and BWG (135 and 118.6 g), respectively compared to the other groups.

Results in Fig. 2 confirmed that increasing fenugreek seed level-up to 10% (group 4)-was associated with a significant decrease in body weight gain (119 g) in comparison to the lower FSP level (5%). The lower level of FS (5%) resulted in a marked increase (p<0.05) in body weight gain (135 g). This may be due to the activity of fenugreek seeds as anti-hyperlipidaemia and anti-obesity agent. This means that consuming higher levels of fenugreek seeds will result in lowering the end body weight gain.

**Bicarbonate, stomach and blood pH:** The mean values of serum bicarbonate, stomach and blood pH of the experimental groups were presented in Table 2. As for



Fig. 2: Mean values of feed intake, final weight and body weight gain for the experimental rats



Fig. 3: Mean values of calcium and phosphorous in the blood of experimental rats



Fig. 4: Mean values of MDA and TAC in blood serum of experimental rats

bicarbonate values, the positive control group was affected by acidosis and recorded the lowest mean value of bicarbonate (23.6 mmol L<sup>-1</sup>), when Fenugreek seeds powder and aqueous extract were added there was a significant increase in bicarbonate levels that were almost similar to the levels recorded for negative control.

The stomach pH was affected by FSP and its aqueous extract as manifested by a significant difference between the positive control group and the other groups. The stomach pH in the negative control group recorded the highest mean value (4.2); these results indicated the ability of FSP to help to equate the stomach acidity. Based on these results, we can conclude that phenolic compounds in fenugreek seeds aqueous infusions are major performed on the stomach and intestinal mucin discharge.

Calcium (Ca), phosphorous (P), total antioxidant capacity (TAC) and malondialdehyde (MDA): The data of Fig. 3 is showing concentrations of calcium, phosphorous. The level of calcium in the negative control was  $10.33\pm0.26$  mg dL<sup>-1</sup>, which was significantly higher than the positive control  $(8.83\pm0.19 \text{ mg dL}^{-1})$ . On the other hand, FSP and FSE increased significantly the levels of phosphorus and calcium in all experiment groups. Results of the antioxidative indices (TAC and MDA) are shown in Fig. 4. It was found that the mean value of TAC was 1.14 nmol dL<sup>-1</sup> for the negative control, which decreased significantly in the positive control (0.99 nmol dL<sup>-1</sup>). There was a significant increase in levels of TAC in the third group (5% FSP), fourth group (10% FSP) and fifth group (2 mL FSPE). Concerning MDA, it was noticed that the level of MDA in the positive control was 4.36 nmol  $dL^{-1}$ , which is considered the highest mean value of MDA compared to the negative control, which recorded the lowest value (3.72 nmol dL<sup>-1</sup>). Moreover, the evaluation of the aqueous extract of fenugreek is completely limited and no data have been reported before to explain the scavenging capacity of extracts infusion. The preponderance of research accessible in the literature stated mostly antioxidant activity of the plant's essential oil or methanolic extracts.

#### J. Biol. Sci., 21 (2): 70-79, 2021



#### Fig. 5(a-g): Hematoxylin-eosin (H/E) stained sections of rat stomach

(a-b) Negative control group, showing the normal histological structure of gastric layers (mucosa, submucosa and mucosa), (H and EX200), (c-d) Positive control group showing focal necrosis of gastric mucosa and submucosal oedema associated with inflammatory cells infiltration, focal desquamation of liming epithelium of gastric mucosa, Inflammatory cells infiltration in lamina propria (H and EX200), (e) Rats treated with 5% FSP showing slight submucosal oedema, showed a few leucocytic cells infiltration in lamina propria associated with submucosal oedema (H and EX200), (f) Rats treated with 10% FSP showing submucosal inflammatory cells infiltration, few leucocytic cells infiltration in lamina Propria (H and EX200) and (g) Rats treated with 2 mL of FSPE showing no histopathological changes and normal structure of gastric mucosa (H and EX200)

**Histopathological examination:** In the present study, the Negative control group revealed the normal histological structure of gastric layers (mucosa, submucosa and mucosa) (Fig. 5a-b), while, the positive group revealed focal necrosis of gastric mucosa and submucosal oedema with inflammatory cells infiltration (Fig. 5c-d). Slight sub-mucosal oedema was observed in the third group (Fig. 5e), moreover, the fourth group showed sub-mucosal inflammatory cells infiltration (Fig. 5f). However, group 5 revealed no histopathological changes (Fig. 5g). The obtained results showed that the positive control group recorded mucosal necrosis; however, no fenugreek treated groups (seeds and aqueous extract) showed any mucosal necrosis.

Microscopically, bones of the negative group revealed no histopathological changes (Fig. 6a); meanwhile, the positive group showed cracks in the cortical bone (Fig. 6b). Examined sections from the 5% FSP group showed cracks in the cortical bone (Fig. 6c). On contrary, the bone of rats from the fourth and fifth groups showed no histopathological changes (Fig. 6d-e).

These studies confirmed the possible protective mechanism of fenugreek seed extract for gastric ulcers through its antioxidant also anti-inflammatory activity and adhesive property.

#### DISCUSSION

The ulceration of gastric mucosa causes dangerous problems to human health all over the globe. Several natural products and current synthetic medicines have been used to heal peptic ulcer illness but ideal medicine has not been identified and investigation of novel antiulcer medicines has survived a range of active studies<sup>24</sup>. This research investigated the chemical composition, antioxidant activity of fenugreek



#### Fig. 6(a-e): Representative photomicrograph of experimental rat bone

(a) Negative group showing normal histological architectures, (H and EX100), (b) Positive group showing cracks in the cortical bone (H and EX100), (c) Rats fed on basal diet supplement with 5% FSP showing cracks in the cortical bone, (H and EX100), (d) Rats fed on basal diet supplement with 5% FSP showing cracks in the cortical bone, (H and EX100), (d) Rats fed on basal diet supplement with 5% FSP showing cracks in the cortical bone, (H and EX100), (d) Rats fed on basal diet supplement with 5% FSP showing cracks in the cortical bone, (H and EX100) and (e) Rats treated by 2 mL FSPE showing normal histological architectures. H and E stain magnification × 100

seeds and their effects on the gastric ulcer, blood pH and lipid profile in Wistar rats. The chemical composition and antioxidant compounds in the seeds of the fenugreek used in the study were very similar to the results<sup>25</sup>, who found that the moisture, protein, fat, ash and fibre and gums contents of fenugreek seeds were 10.36, 25.4, 5.60, 3.10 and 56.30%, respectively.

The improvement of the final weight and body weight gain may be related to the constituents of the fenugreek seeds that are a rich source of vitamins, minerals and antioxidants, which help protect the body's cells from damage caused by unstable molecules known as free radicals. Fenugreek is traditionally used to promote digestion, improve appetite and support respiratory health. The high-fibre content may also support healthy bowel function and its lecithin content promotes fat metabolism<sup>11</sup>. The decrease in body weight gain means that consuming higher levels of fenugreek seeds will result in a lowering of body weight at the end of the experiment. This may be due to the activity of fenugreek seeds as an anti-hyperlipidemic and anti-obesity agent. The efficacy of fenugreek seed as an anti-hyperlipidemic and anti-obesity agent on obese mice was examined<sup>26,27</sup> and they found that fenugreek seed lowered serum total cholesterol and triglyceride. In fact, Muraki et al.28, concluded that fenugreek seed Dose-dependently increased the excretion of cholesterol and total bile acids into the faeces. It was suggested that fenugreek inhibited lipid accumulation in the liver by increasing the lipid excretion in the faeces.

The generality of herbs is used after wetness in water and then this water extract is possessed for disease recovery, but a few herbs are taken as a powder. The metals from these may be extracted in the stomach after the action of acid. Therefore, these acid extracts may supply information associated with metals available from these herbs for biological research. This study showed that the fenugreek seed and its aqueous extract have an effective acid-induced gastric ulcer in Wistar rats. The gastroprotective effect of the extract may be related to an antacid effect or cytoprotective properties of the extract.

Reduction in the serum level of bicarbonate (HCO<sub>3</sub>) resulted in a reduction of blood pH. The bulk of the HOCO<sub>3</sub> is generated in the proximal convoluted tubule as a result of NH<sub>3</sub> production and its excretion in urine as NH<sub>4</sub><sup>29</sup>. Our work was consistent with the results<sup>30</sup>, which explained the gastroprotective effect of fenugreek enriched fraction against indomethacin-induced gastric ulcer, which is achieved by increasing mucin secretion. The fenugreek seeds extract could protect intestinal mucosa against gastric damage by its antioxidant properties and reducing gastric juice acidity<sup>31</sup>. The improvement in acidity is due to the containment of the fenugreek seeds and their aqueous extract on flavonoids, especially coumarins, which has an alkaline effect. The treatment of acute acidosis by alkali therapy is regularly shown to support and maintain the plasma pH to greater than 7.2 using methoxy coumarins nucleus which changes plasma pH from acid to basic as from obtained results anti-inflammatory activity by hind paw Edema methods which exhibited an 87% of oedema inhibition<sup>32</sup>. Based on these results, we can conclude that phenolic compounds in fenugreek seeds aqueous infusions are major performed on the stomach and intestinal mucin discharge.

Diosgenin, a steroidal sapogenin and dietary intake in fenugreek seeds may have a positive effect on rat skeletal and bone minerals, especially calcium and phosphorus. Our results are consonant with the results<sup>33,34</sup>, who found that calcium and phosphorus content had a significant increase in rat's bone fed on trigonelline powder (*Trigonella foenum-greacum* L.).

Malondialdehyde has a very devastating effect in altering the structure and function of cell membranes<sup>35</sup>. The fenugreek seeds have the potential to prevent malondialdehyde formation and restore the total antioxidant capacity in experimental rats<sup>36</sup>. The fenugreek seeds have five active flavonoids: vitexin, tricine, naringenin, quercetin and tricine-7-0- $\beta$ -d-gluco-pyranoside. Flavonoids are important in scavenging free radicals in cells of the organism<sup>37</sup>. Moreover, the evaluation of the aqueous extract of fenugreek is completely limited and no data have been reported before to explain the scavenging capacity of infusion of extracts. The preponderance of research accessible in the literature stated mostly antioxidant activity of the plant's essential oil or methanolic extracts.

The present results showed that the gross and histopathological lesions following induction of gastric ulcer were significantly improved in the treatment groups compared with the positive control group. It is clear that the extract relatively preserved the gastric mucosa against acidity. Polysaccharide fractions of fenugreek seed, such as gel from galactomannan, cover the surface of the mucosa or form complexes between gel and mucus barrier. Such arrangements protect against agents introduced into the stomach or against endogenously formed acid and pepsin in the stomach. It is speculated that the polysaccharide composition of the gel and/or the flavonoids are responsible for the gastro-protective and anti-secretary activities of the seeds<sup>38</sup>. Gastro-mucosal damages induced by different experimental ulcer models have different mechanisms<sup>39</sup>. In previous studies, the effects of fenugreek seeds for treating gastric mucosa have been evaluated. Methods for inducing gastric injuries in these studies were oral administration of irritant materials such as aspegic (salicylic acid and acetic anhydride)<sup>40</sup>. The oral administration of fenugreek seeds extract could protect intestinal mucosa against experimental intestinal ischemia-reperfusion injuries in rats<sup>31</sup>. Aqueous extract of fenugreek seeds has an effective antioxidant activity and, therefore, its clinical treatment could attenuate the hardness of gastric ulcers and inhibit gastric mucosal injuries<sup>41</sup>. These studies confirmed the possible therapeutic results of fenugreek seed extract for gastric ulcers through its antioxidant also anti-inflammatory activity and adhesive property.

In the present study, it was found that in the treatment groups, the infiltration of inflammatory cells especially neutrophils decreased and that was consistent with the reduced gastric mucosal damages. Since these inflammatory cells are a potential source of oxygen metabolites and pro-inflammatory cytokines<sup>42,43</sup>, fenugreek improved the free radicals-induced gastric mucosal damage by decreasing the neutrophils population via its antioxidant and antiinflammatory actions in gastric ulcers in rats.

#### CONCLUSION

The present study demonstrated the potential therapeutic effect of fenugreek seed powder and aqueous extract against gastric ulcer via its antioxidant and antiinflammatory activity and mucilaginous property. fenugreek seed has an effective antioxidant activity and, therefore, its clinical administration can attenuate the severity of gastric ulcer and prevent gastric mucosal damages.

#### SIGNIFICANCE STATEMENT

This study discovered that fenugreek seed powder and aqueous extract have many beneficial health effects when regularly consumed. Fenugreek seed powder and aqueous extract showed significant ulcer protective effects by boosting the antioxidant potential of the gastric mucosa thereby relieving mucosal injury. Dietary intake of fenugreek seeds may have a positive effect on rat skeletal and bone minerals, especially calcium and phosphorus. This study will help the researchers to uncover the critical area of gastric ulcer and osteoporosis that many researchers will not be able to explore. Thus, a new theory on the role of fenugreek consumption and anti-ulcer and anti-osteoporosis may be arrived at. Thus, we suggest that fenugreek may have beneficial health effects on human subjects.

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