



## **Anti Oxidative Activity of Extracts from *Fenugreek Seed*:**

### **A review**

**Hazir<sup>1</sup>, Sonika Rani<sup>2</sup>, Luxmi Yeasmin<sup>3</sup>, Payal Saxena<sup>4</sup>**

1. Research Scholar, Krishna Pharmacy college Bijnor.
2. Research Scholar, Department of Pharmacology, school of Pharmacy and Research, Dev Bhoomi Uttarakhand University (Dehradun).
3. Assistant Professor, Department of Pharmacology, school of Pharmacy and Research, Dev Bhoomi Uttarakhand University (Dehradun).
4. Assistant Professor, Department of Pharmacology, school of Pharmacy and Research, Dev Bhoomi Uttarakhand University (Dehradun).

**Corresponding Author:** Luxmi Yeasmin

### **Abstract**

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Fenugreek is an annual plant in family Fabaceae, with leaves consisting of three small obovate to Oblong leaflets. It's cultivated world wide as a semiarid crop. Its Seed and leaves are common ingredients in dishes from the Indian subcontinent and used as a culinary ingredient since ancient times. Fenugreek (TFG) is a nutrient food rich in beneficial phytochemical. In this study three types of solvent extract of Fenugreek seeds were used to examine the effect of extraction solvent on total phenolics content (TPC), (DPPH) and (FRAP). Herbs and spices have been extensively used as food additives for natural antioxidant. It can be applied for preservation of Lipid peroxidation in biological system. Fenugreek is an important spice its dried seed have wide application in food as a flavoring additive as well as in medicines. Seed extraction of Fenugreek were prepared by extraction method with different solvent such as Methanol, ethanol dichloromethane, acetone, hexane and ethyl acetate. The antioxidant property of plant material is due to the presence of many active phytochemical including vitamins, flavonoids, terpenoids, carotenoids, coumarins, lignin, salicylic acid, plant sterol. Evaluating from different parameters were



in agreement with each other and reveal that all extracts of Fenugreek exhibit antioxidant activity, these findings suggest that fenugreek extract could act as a potent source of antioxidant.

**Keywords :** Herbal plants, Fenugreek, Anti Oxidative, antioxidant

## **Introduction**

The plant fenugreek (*Trigonella foenum-graecum*) is a member of the Fabaceae family. Although it is originally from the Mediterranean, it is now widely planted throughout the world, especially in China, Egypt, and India. For generations, fenugreek seeds have been used to cure a wide range of illnesses in traditional medicine.[1]

Fenugreek seeds, sometimes referred to as "methi" in Ayurvedic medicine, are used to cure a variety of ailments such as skin irritation, respiratory infections, and digestive issues. Fenugreek seeds are used in traditional Chinese medicine to strengthen the kidneys and reduce discomfort and swelling.[2]

Traditional uses of fenugreek seeds include galactagogue therapy, which stimulates nursing mothers' milk supply. Modern studies have demonstrated that fenugreek seeds can boost nursing women's milk supply, supporting this practice.[3]

Fenugreek seeds are utilised in food, particularly in Indian and Middle Eastern cuisines, in addition to their therapeutic benefits. They have a little bitter flavour and are frequently used to flavour breads, curries, and stews.[4]



Numerous bioactive substances, including as alkaloids, flavonoids, and saponins, are found in fenugreek seeds. These substances give the plant its many therapeutic benefits, such as lowering blood sugar levels, reducing inflammation, and enhancing digestion.[5]

Overall, fenugreek seeds have a long history of traditional use for therapeutic and culinary purposes, and contemporary research is consistent with their therapeutic potential. To completely comprehend their modes of action and choose the best dosages and formulations for therapeutic usage, more research is required.[6]

### **Definition of antioxidants and oxidative stress**

In the body, free radicals can cause harm that antioxidants can mitigate or neutralise. Free radicals are very reactive chemicals that can lead to oxidative stress, a state in which there is an imbalance between the body's capacity to neutralise free radicals through antioxidant systems and the generation of free radicals.[7]

Oxidative stress can harm DNA, proteins, and cells, which can result in a variety of health issues. Numerous chronic illnesses, including cancer, cardiovascular disease, diabetes, and neurodegenerative diseases like Alzheimer's and Parkinson's, have been linked to it.[8]

By scavenging free radicals and keeping them from wreaking havoc on cells, antioxidants can help to lessen the negative effects of oxidative stress. Antioxidants include, among others, vitamin C, vitamin E.[9]

Additionally, the body makes its own antioxidants, such glutathione, which can aid in preventing oxidative stress. The body's natural antioxidant defences may fall short in situations where there are too many free radicals to shield cells and tissues from damage.[10]

Consuming foods high in antioxidants, such as fruits, vegetables, nuts, and whole grains, can help the body acquire the nutrients it needs to keep the balance of its antioxidants and defend



against oxidative stress. The decrease of oxidative stress and promotion of general health and wellbeing can also be achieved through regular exercise and stress-reduction measures.[11]

### **Significance of studying the anti-oxidative activity of fenugreek seed extracts**

1. Health benefits: For ages, fenugreek seeds have been used in traditional medicine to treat a variety of illnesses, including as diabetes, gastrointestinal problems, and inflammation. Antioxidants and other bioactive chemicals found in abundance in the seeds may contribute to their potential therapeutic benefits.[12]
2. Chronic illness prevention: Numerous chronic diseases, including as cancer, cardiovascular disease, and neurological disorders, have been related to oxidative stress. Researchers can learn more about fenugreek seed extracts' ability to prevent or lower the incidence of various diseases by examining their anti-oxidative activities.[13]
3. Food and supplement sector: Fenugreek seed extracts are employed as a source of natural antioxidants in the food and supplement industry. Manufacturers can improve their goods to provide the greatest possible health advantages by taking into account the anti-oxidative activity of these extracts.[14]
4. Environmental applications: It has been demonstrated that fenugreek seed extracts have antioxidant and metal-chelating capabilities that may help lessen the impacts of pollution. These qualities may have potential applications in environmental rehabilitation.[15]

### **Anti-Oxidative Properties of Fenugreek Seed Extracts**

Natural antioxidants, which are substances that shield cells from damage brought on by free radicals, are abundant in fenugreek seeds. Free radicals are unsteady molecules that can harm DNA, proteins, and cells, causing oxidative stress and a number of chronic illnesses. A number



of antioxidant substances found in fenugreek seeds combine to scavenge free radicals and lessen oxidative stress.[16]

One of the major categories of antioxidants included in fenugreek seeds are phenolic acids. It has been demonstrated that several substances, including gallic acid, protocatechuic acid, and caffeic acid, exhibit strong antioxidant action. They function by providing electrons to free radicals in order to neutralise them and stop them from causing cell damage. Additionally, phenolic acids have been demonstrated to be anti-inflammatory and anti-cancer.[17]

Another type of antioxidant found in fenugreek seeds are flavonoids. These substances, which include apigenin, luteolin, and quercetin, have been demonstrated to have antioxidant action and may aid in the prevention of chronic diseases. Free radicals are neutralised by flavonoids, which also lessen oxidative stress. Additionally, they have been demonstrated to possess anti-inflammatory and anti-cancer properties.[18]

Another class of substances with antioxidant effects that can be found in fenugreek seeds are called saponins. It has been demonstrated that these substances can neutralise free radicals and lessen oxidative stress. Additionally, they have been demonstrated to have cholesterol-lowering and anti-inflammatory properties.[19,20]

**The vitamins with antioxidant properties found in fenugreek seeds:**

<b>Vitamin</b>	<b>Function</b>
<b>Vitamin C</b>	<b>Donates electrons to neutralize free radicals, helps regenerate other antioxidants such as vitamin E</b>
<b>Vitamin E</b>	<b>Donates electrons to neutralize free radicals, protects cell membranes and lipids from oxidative damage</b>



<b>Beta-carotene</b>	<b>Converted to vitamin A in the body, donates electrons to neutralize free radicals, protects cell membranes and lipids from oxidative damage</b>
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### **Mechanisms of antioxidant activity**

There are a number of antioxidant chemicals present in fenugreek seed extracts, including polyphenols, flavonoids, and alkaloids, which are thought to be responsible for the antioxidant activity of the extracts. Free radicals and reactive oxygen species (ROS), which can lead to oxidative stress and damage to cells and tissues, can be neutralised by these substances.

Specific mechanisms of antioxidant action include the following:

1. Free radical scavenging: Fenugreek seed extract antioxidants can give electrons to unstable free radicals, stabilising them and stopping them from interacting with other molecules.[21]
2. Metal chelation: Some of the antioxidants in fenugreek seed extracts can bind to metal ions like iron and copper and stop them from catalysing the production of ROS.[22]
3. Enzyme inhibition: Xanthine oxidase and NADPH oxidase are two ROS-producing enzymes that can have their activity inhibited by certain antioxidant chemicals found in fenugreek seed extracts.[23]
4. Upregulation of endogenous antioxidant systems: Fenugreek seed extracts have been found to boost the body's natural defence against oxidative stress by increasing the expression of antioxidant enzymes such superoxide dismutase (SOD), catalase, and glutathione peroxidase.[24,25]

### **Antioxidant properties and prevention of chronic diseases**



The phrase "antioxidant properties and prevention of chronic diseases" alludes to the potential health advantages of utilising fenugreek seed extracts. Free radicals are unstable chemicals that can harm cells and play a role in the onset of chronic diseases including cancer, diabetes, and heart disease. Antioxidants are substances that shield cells from this oxidative stress. Since ancient times, fenugreek seeds have been utilised in Ayurvedic medicine for their wide range of health advantages, including their antioxidant capabilities.[26]

The potential health advantages of fenugreek seed extracts, notably in the prevention of chronic diseases, have drawn more attention in recent years. The antioxidant capacities of fenugreek seed extracts and their potential value in the prevention or treatment of chronic illnesses have been the subject of numerous investigations. With a focus on their antioxidant qualities and potential role in preventing chronic diseases, this topic seeks to give a summary of the most recent research on the health benefits of fenugreek seed extracts.[27]

### **Anti-inflammatory effects**

Since ancient times, fenugreek seeds have been used for their numerous health advantages, including their anti-inflammatory effects. The body naturally responds to damage or infection with inflammation, but persistent inflammation can cause a variety of health issues, including as autoimmune illnesses, heart disease, and cancer.[28]

By inhibiting the formation of pro-inflammatory molecules and boosting the production of anti-inflammatory molecules, fenugreek seed extracts may help to reduce inflammation in the body, according to studies. These effects could be brought on by the flavonoids and alkaloids found in fenugreek seeds.[29]

Fenugreek seed extracts have also been linked to improved blood sugar regulation, lowered cholesterol levels, and increased milk production in nursing mothers, according to research.



These advantages might result from fenugreek seeds' anti-inflammatory and antioxidant characteristics.[30]

### **Potential as a treatment for diabetes and metabolic syndrome**

Diabetes and metabolic syndrome are two illnesses that are strongly related to one another and are spreading more and more around the globe. High blood sugar levels are a hallmark of diabetes, which can be caused by either an inability to produce insulin (Type 1 diabetes) or a resistance to insulin (Type 2 diabetes). A group of illnesses known as metabolic syndrome raises the risk of diabetes, heart disease, and stroke. High blood pressure, high blood sugar, excess body fat around the waist, and abnormal cholesterol levels are some of these problems.[31]

Researchers have looked into the possibility of using fenugreek seed extracts to treat metabolic syndrome, diabetes, and assist control blood sugar levels. Fenugreek seed extracts may assist persons with Type 1 and Type 2 diabetes lower blood sugar levels, according to several clinical studies. This could be as a result of the presence of soluble fibre, which slows down the body's absorption of sugars and carbohydrates, as well as chemicals like trigonelline and 4-hydroxyisoleucine, which could trigger the release of insulin.[32]

Fenugreek seed extracts have been investigated for their ability to assist manage various components of metabolic syndrome in addition to blood sugar regulation. According to research, fenugreek seed extracts, for instance, may help lower blood pressure and enhance cholesterol levels, both of which are crucial risk factors for heart disease and stroke development.[33]

Fenugreek seed extracts appear promising as a potential all-natural therapeutic option for diabetes and metabolic syndrome, even if additional research is required. However, before taking any supplements or making significant dietary or pharmaceutical changes, people with these problems should speak with their healthcare practitioners.

### **Extraction Methods and Analysis of Fenugreek Seed Extracts**





Many different extraction techniques, each with their own benefits and drawbacks, can be used to generate fenugreek seed extract. Several typical extraction techniques include:

1. **Soxhlet extraction:** This technique entails separating the required component from the plant material using a Soxhlet equipment. A thimble containing the plant material is placed in a chamber that also contains a solvent. Heat is applied to the solvent, which is then allowed to condense, evaporate, and drop back into the thimble. Up till the necessary amount of substance is extracted, the procedure is done several times. Soxhlet extraction is effective and can remove a variety of substances, although it can take a long time and use a lot of solvent.[34]
2. **Maceration:** In this procedure, the plant material is soaked in a solvent for a predetermined amount of time. The remaining plant matter is then filtered out of the solvent, and the resultant extract is collected. This approach is straightforward and affordable, but it could not be as effective as other approaches and might require a longer extraction period.[35]
3. **Ultrasonic extraction:** With this technique, the plant material's cell walls are destroyed by ultrasonic vibrations, allowing the required chemicals to escape into the solvent. Comparatively speaking, this procedure uses less solvent and is quick and effective. However, it could need specialised tools and might not be appropriate for chemicals that are sensitive to heat.[36]
4. **Supercritical fluid extraction:** In this technique, the necessary chemicals are extracted utilising supercritical fluids, such as carbon dioxide. Although it needs specialised equipment and might be costly, this procedure is effective and environmentally beneficial.[37]



The active compounds present in the fenugreek seed extract can be identified and measured using a variety of techniques, including FTIR (Fourier transform infrared spectroscopy), GC-MS (Gas chromatography-mass spectrometry), and HPLC (High-performance liquid chromatography).[38]

### **Methods for measuring antioxidant activity**

There are numerous ways to evaluate antioxidant activity, some of which include:

1. ORAC (Oxygen Radical Absorbance Capacity): This technique assesses the peroxy radical-scavenging capacity of antioxidants. A fluorescent probe and a peroxy radical generator are combined with the sample to assess the rate of fluorescence degradation. Based on the degree of fluorescence decay inhibition, the ORAC value is determined.[39,40]
2. FRAP (Ferric Reducing Antioxidant Power): This technique assesses the antioxidants' capacity to convert  $Fe^{3+}$  to  $Fe^{2+}$ . After a predetermined amount of time, the sample is combined with a solution of  $Fe^{3+}$  and a coloured probe, and the solution's absorbance is assessed. Based on the level of absorbance reduction, the FRAP value is determined.[41]
3. DPPH (2,2-diphenyl-1-picrylhydrazyl): This technique assesses an antioxidant's capacity to neutralise DPPH radicals. The sample is combined with a DPPH radical solution, and after a predetermined amount of time, the solution's absorbance is assessed. Based on the level of absorbance reduction, the DPPH value is determined.[42]
4. ABTS: This technique gauges an antioxidant's capacity to scavenge ABTS radicals (2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid)). The sample is combined with an ABTS radical solution, and after a predetermined amount of time, the solution's absorbance is assessed. Based on the level of absorbance reduction, the ABTS value is determined.[43]



5. TPC (Total Phenolic Content): This technique counts all phenolic substances in a sample. The sample is combined with a Folin-Ciocalteu reagent and basic solution solution, and after a predetermined amount of time, the solution's absorbance is measured. The degree of absorbance is used to compute the TPC value.[44]

### **Factors Affecting Antioxidant Activity of Fenugreek Seed Extracts**

Variations in plant growth and seed makeup can have an impact on the antioxidant activity of fenugreek seed extracts. The following are a few of the variables that may affect the antioxidant activity of fenugreek seed extracts:

1. Plant genotype: Due to changes in the makeup and concentrations of bioactive substances including phenolic acids, flavonoids, and saponins, different genotypes of fenugreek plants can differ in their antioxidant potential.[45]
2. Growing conditions: The content and concentration of bioactive chemicals in the seeds of fenugreek plants can be influenced by environmental factors such as temperature, light, and soil nutrients. For instance, it has been noted that fenugreek seeds contain more phenolic acids when exposed to high light intensity.[46]
3. Seed maturity: Depending on the stage of seed development, fenugreek seeds' antioxidant activity may differ. Fully grown seeds have a higher antioxidant capacity than immature seeds, according to studies.[47]
4. Seed processing: Grinding, soaking, and roasting fenugreek seeds can change the composition and stability of bioactive chemicals, which can have an impact on the antioxidant activity. For instance, it has been demonstrated that toasting fenugreek seeds increases their antioxidant activity by raising their phenolic acid and flavonoid levels.[48]



5. Extraction technique: The antioxidant activity of fenugreek seed extracts might also be impacted by the selection of the extraction technique. The types and concentrations of bioactive chemicals that can be extracted from seeds using various extraction techniques, including as solvent extraction, microwave-assisted extraction, and supercritical fluid extraction, can have an impact on the antioxidant activity.[49]

### **Extraction conditions**

The antioxidant activity of fenugreek seed extracts can be considerably impacted by the extraction conditions, including the kind of solvent, temperature, and duration. Among the things to take into account are:

1. Solvent kind The bioactive chemicals derived from fenugreek seeds might vary in composition and concentration, which can impact their antioxidant activity. For the extraction of fenugreek seeds, common solvents include methanol, ethanol, water, and their mixtures. The antioxidant activity of fenugreek seeds may be impacted by the types and concentrations of bioactive chemicals that may be extracted using various solvents.[50]
2. Temperature: The antioxidant activity of fenugreek seed extracts can also be influenced by the temperature of extraction. Heat-sensitive molecules can degrade at high temperatures, which also decreases the antioxidant activity in general. The release of bound or conjugated bioactive molecules, however, may be the reason why some studies have found greater antioxidant activity at higher temperatures.[51]
3. Extraction time: The length of time it takes to extract bioactive components from fenugreek seeds can have an impact on both their amount and quality. Although longer extraction durations can improve extraction efficiency, they also run the risk of degrading labile chemicals. In order to maximise the yield of bioactive chemicals and sustain their antioxidant activity, the ideal extraction duration should be identified.[52]



4. Extraction technique: Different extraction techniques, such as supercritical fluid extraction, ultrasound-assisted extraction, and microwave-assisted extraction, can extract various kinds and concentrations of bioactive chemicals from fenugreek seeds, which can alter the antioxidant activity.[53]

### **Conclusion**

Fenugreek seeds are known to contain high levels of antioxidants such as flavonoids, alkaloids, and saponins, which contribute to their potential health benefits. Several studies have investigated the anti-oxidative activity of fenugreek seed extracts, both in vitro and in vivo. In vitro studies have shown that fenugreek seed extracts have the ability to scavenge free radicals and inhibit lipid peroxidation, which can help to prevent oxidative stress and cellular damage. In addition, these extracts have been found to increase the activity of antioxidant enzymes such as superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GPx), further enhancing their antioxidant effects. In animal studies, fenugreek seed extracts have been found to reduce oxidative stress and inflammation, as well as improve markers of liver and kidney function. These effects have been attributed to the antioxidant activity of fenugreek seeds, which may help to protect against the development of chronic diseases such as diabetes, cardiovascular disease, and cancer. Overall, the available evidence suggests that fenugreek seed extracts have significant anti-oxidative activity, which may contribute to their potential health benefits. However, further research is needed to determine the optimal dose and duration of supplementation, as well as the potential interactions with other dietary factors and medications.

### **Future directions for research**

Although there has been some research on the anti-oxidative activity of fenugreek seed extracts, there are still many areas that require further investigation. Some potential future directions for research on this topic include:



1. **Clinical trials:** While some animal studies have shown promising results, more clinical trials are needed to determine the effectiveness of fenugreek seed extracts in humans. These studies could explore the potential health benefits of fenugreek seed extracts in preventing or treating various diseases, as well as the optimal dose and duration of supplementation.
2. **Mechanisms of action:** Although the antioxidant activity of fenugreek seed extracts has been established, the exact mechanisms by which they exert their effects are not fully understood. Future research could explore the molecular pathways involved in their anti-oxidative activity, as well as their potential interactions with other dietary factors and medications.
3. **Extraction methods:** There are several methods for extracting fenugreek seed compounds, each of which may yield different results. Further research is needed to determine the most effective extraction methods for obtaining the maximum amount of antioxidant compounds from fenugreek seeds.
4. **Formulation and delivery:** Fenugreek seed extracts can be delivered in various forms, such as capsules, powders, and teas. Future research could investigate the most effective formulations and delivery methods for maximizing the bioavailability and antioxidant activity of these extracts.
5. **Safety and toxicity:** While fenugreek seed extracts are generally considered safe, more research is needed to determine their potential toxicity and adverse effects, particularly when used in high doses or in combination with other supplements or medications.

### **Acknowledgement**

The conception and writing of this article were greatly assisted by all of the above authors.

### **Conflict of interest**



The Authors declare no conflict of interest.

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**Anti Oxidative Activity of Extracts from *Fenugreek Seed*:**

**A review**



*Section A-Research paper*