

Research article

PHYTOCHEMICAL SCREENING AND STUDY OF ANTIOXIDANT ACTIVITY OF FENUGREEK SEED

SHINDE PUJA M, SANCHETI VP, PRAJAPATI ASHISH D

S.N.D COLLEGE OF PHARMACY, YEOLA

Corresponding Author: SHINDE PUJA M

ABSTRACT:

Trigonellafoenum-graecum (Family- Fabaeace) plant is eaten in India since long. It is also known as Methi and used in Ayurvedic medicines for the treatment of wounds, abscesses, arthritis, bronchitis, and digestive disorders. The seeds extracts were also subjected to preliminary Phytochemical screening. The data obtained in present study will serve as valuable tool for identification, authentication and detection of adulterants, standardization and quality control of the drug. TLC study of the alcoholic extract obtained from the seeds was carried out and seven compounds were separated. A comprehensive overview of the pharmacognostic, phytochemical analysis of the seed extract and the literature survey carried out for the anticancer properties of fenugreek seeds. These observations would be of immense value in the botanical identification and standardization of the drug in crude form and would help distinguish the drug from its other species. Phytochemical standardization parameters such as moisture content, total ash, water soluble and acid insoluble ash, alcohol soluble and water soluble extractives were determined. Preliminary identification of phytoconstituents was performed.

Spices and herbs possess antioxidant activity and can be applied for preservation of lipid peroxidation in biological systems. Crude extracts of fenugreek were prepared by soxhelt extraction method with different solvents such as methanol, ethanol, dichloromethane, acetone, hexane and ethyl acetate. Extracts were subjected for the measurement of total phenolic content (TPC) by Folin-Ciocalteu method as well as flavonoid content, chelating activity, reducing power and antioxidant/radical scavenging activity [1,1-diphenyl-2-picryl-hydrazyl (DPPH^o) free radical scavenging activity]. Results from different parameters were in agreement with each other. The results reveal that all extracts of the fenugreek exhibit antioxidant activity. These findings suggest that the fenugreek extracts could act as potent source of antioxidants.

KEY WORDS:Trigonellafoenum-graecum , Antioxidant activity,

INTRODUCTION

Trigonellafoenum-graecum (Family Fabaeace) is called methika in Ayurveda and used as medicine for the treatment of wounds, abscesses, arthritis, bronchitis and digestive disorders etc since oldest time. It is also eaten in winters as to improve immunity and protects heart, brain and other vital organs of body through its medicinal properties. In traditional Chinese Medicine it is also used for kidney problems and conditions affecting the male reproductive tract. The recent researches have proved it beneficial for Atherosclerosis, Constipation, Diabetes, High cholesterol and Hyper-triglyceridemia. The seeds of fenugreek contain alkaloids, flavonoids, saponins, amino acids, tannins and some

steroidal glycosides, proteins etc. Standardization of fenugreek seeds is done for the establishment of quality and identity profile of the drug for the purpose of safety monitoring and overall quality assurance of the industrially as well as commercially important drug . Since there is no report in literature regarding the standardization parameters of fenugreek seeds. Therefore, in the present investigation an attempt has been made to standardize fenugreek seeds by using macroscopy and microscopical characters, powder microscopy, fluorescence analysis, physio-chemical values, and phytochemical screening.

Fenugreek, *Trigonella foenum-graecum* L. is an annual crop from the family Leguminosae. The seed of this plant grown in South Asia, has been known to have health potential with the ability to lower blood glucose and cholesterol levels, and hence in the prevention and treatment of diabetes and coronary heart diseases. The species name "foenum-graecum" means "Greek hay" indicating its use as a forage crop in the past. Fenugreek is believed to be native to the Mediterranean region^[1], but now is grown as a spice in most parts of the world. It is reported as a cultivated crop in parts of Europe, northern Africa, west and south Asia, Argentina, Canada, United States of America (USA) and Australia.^[1-4] India is the leading fenugreek producing country in the world.^[3] Fenugreek has been used for centuries in folk medicine to heal ailments ranging from indigestion to baldness. Fenugreek is regarded as the oldest known medicinal plant in recorded history . Fenugreek seeds, which are described in the Greek and Latin Pharmacopoeias, are said to have anti-diabetic activity and hypocholesterolaemic effects. In addition, fenugreek has been reported to possess a curative gastric anti-ulcer action, anti-bacterial, anti-helminthic, anti-fertility effects and anti-nociceptive effects. Fenugreek seed contains both saponin and galactomannan polysaccharides which could be of use as natural antibacterial compounds. In spite of numerous medicinal uses attributed to this plant, there is very less pharmacognostical report on the microscopical and other physicochemical standards required for the quality control of the crude drugs. Hence the present investigation includes macroscopical evaluation, determination of physicochemical constants, preliminary phytochemical screening and a review on anticancer properties of *Trigonella foenum-graecum*.^[5]



Fig1..Seed for fenugreek

History:

For the last few decades, phytochemistry (study of plants) has been making rapid progress and herbal products are becoming popular. There has been dramatic rise in the sale of herbal products like *Allium sativum*, *Spirulina*, *Trigonella Foenum Graecum*, and *Silybum marianum*. Herbal medicine has produced number of distinguished researchers and due to its accessibility to traditions it is still practiced even by lay practitioners. Ayurveda, the ancient healing system of India, flourished in the Vedic era in India. According to historical facts, the classical texts of Ayurveda, *Charaka Samhita* and *Sushruta Samhita* were written around 1000 B.C. The Ayurvedic *Materia Medica* includes 600 medicinal plants along with therapeutics. Herbs like turmeric, fenugreek, ginger, garlic and holy basil are integral part of Ayurvedic formulations. The formulations incorporate single herb or more than two herbs (polyherbal formulations).^[4]

Origin and centre of diversity of fenugreek

Fenugreek is a diverse species. Authors have widely debated the probable ancestry of *Trigonella foenum-graecum* (L.), although the divergent schools of opinion identify three probable centers of origin for the plant (Acharya et al. 2008). Vavilov (1951, 1926) suggested that fenugreek originated from the Mediterranean region. However, according to Fazli and Hardman (1968), and De Candolle (1964) fenugreek has an Asian/Indian center of origin. Dangi et al. (2004) also proposed that the species originated in Turkey.^[4] Collections of fenugreek from different countries have been made for the purposes of taxonomic investigation and characterization. Results of these studies have revealed other probable centers of diversity for fenugreek; e.g., Serpukhova (1934) proposed that Yemen and Abyssinia are centers of diversity for fenugreek, while Moschini (1958) suggested that Sicily, Tuscany and Morocco are centers of diversity for fenugreek. In another study, Yemen, the Transcaucasia region of Eurasia, Africa, Afghanistan, the China-Iran region, and India also have been proposed as diversity centers for fenugreek (Furry 1950).^[5]

Cultivation and Harvesting:

Fenugreek is now cultivated in all habitable continents of the world. Some of these continents have a long history of use, while other continents only started cultivating the crop during the past 2-3 decades. Asia is positioned in 1st place among continents in terms of fenugreek production and acreage with India leading in fenugreek seed production,^[4,5] producing about 90% of the world fenugreek grown (Acharya et al. 2008, 2007). Among other Asian countries; Iran, Israel, China and Pakistan also have high levels of production. Asia is followed by the continent of Africa in terms of fenugreek production and acreage as well as richness in genetically distinct fenugreek germplasm.^[5]

Crop becomes ready for harvest in about 120-150 days. At the time of ripening or maturity, leaves and pods become yellowish and leaves start falling. Timely harvesting is very important for this crop as late harvest leads to seed losses due to pod bursting, while in early harvest, the grains remain immature and small. Harvesting should be done early in the morning. After harvest, plants should be dried in threshing yard and threshed by

trampling under the feet of bullocks. Seeds should be separated and cleaned by winnowing.^[6]

Preparation and Storage:

Dried seeds should be lightly roasted before using (don't overdo it though, or they will become bitter). After roasting, they are easily ground. A small amount will complement many other spices, but too much can be overpowering.^[6,7] If the seeds are required as part of a curry paste they can be soaked overnight to swell and soften, and be easily mixed with the other ingredients.^[7]

Plant Protection:

It is pest-free, but susceptible to Cercospora leaf spot, a fungus disease^[6]. Fenugreek appears very resistant to attacks by insects and animal enemies. The seeds of fenugreek can be store for more than 10 years without any treatment. The peculiar smell of the fenugreek plants and seeds may be a possible factor for their resistance to the attack of insects.^[8]

MAIN CONTENT OF REVIEW:

PLANT PROFILE:



Fig.: Trigonella Foenumgraecum

Domain	:	Eukarya
Kingdom	:	Plantae
Division	:	Magnoliophyta (or Anthophyta)
Class	:	Magnoliopsida
Order	:	Fabales(or Leguminales)
Family	:	Fabaceae
Sub-family	:	Trifoliae

Genus : **Trigonella**
Sub-genus : **Foenum-
graecum**
Species : **Trigonellafoenu
m-graecum**

Morphology:

1. **Appearance:** Solid- rhomboidal seeds,3 to 5 mm long,2 mm thick. Hard, pebble-like.
2. **Colour:** Yellowish brown-light brown
3. **Odour:** characteristic spicy
4. **Taste:** Bitter and mucilaginous

PHARMACOGNOSTICAL STUDIES:

1. Macroscopic and Microscopic characteristics of fenugreek:

Macroscopical characters:

The morphological studies were carried out for shape, size, color, odor and taste and fracture identification of the fenugreek seed.

Microscopic studies:

The transverse sections of leaf and seed were prepared by using sharp razor then sections were treated with few amount of chloral hydrate. Best section was selected and mounted glycerin temporarily and observed under light microscope. For powder microscopy powder of seed was taken on glass slide and observed under light microscop.

Macroscopical evaluation:

Seeds

The macroscopical characters of seeds are -Solid-rhomboidal, pebble like shape, 3-5cm long, 2mm thick, plain surface, yellow, bitter mucilaginous taste and have characteristic odor.

Microscopical characters:

Transverse section



Fig 3. : Transverse section of seed**Chemical constituents:**

Alkaloides: Trimethylamine, Neurin, Trigonelline,Choline, Gentianine, Carpaine and Betain.

Amino acids: Isoleucine, 4-Hydroxyisoleucine,Histidine, Leucine, lysine, L-tryptophan, Argenine.^[8]

Saponins: Graecunins, fenugrin B, fenugreekine,trigofenosides A-G.

Steroidal saponogens: Yamogenin, diosgenin,smilagenin, sarsasapogenin, tigogenin, neotigogenin,gitogenin, neogitogenin, yuccagenin, saponaretin.

Flavonoids: Quercetin, rutin, vetixinisovetixin.

Fibers: Gum, neutral detergent. fiber

Othe: Coumarin, lipids, vitamins, minerals. 28% mucilage; 22 % proteins; 5 % of a stronger-swelling, bitter fixed oil.^[9]

PRELIMINARY PHYTOCHEMICAL INVESTIGATION: The plant material thus collected is subjected for following tests:

Sr.NO	TEST	OBSERVATION	INFERENCE
1.	Test for carbohydrates: Molisch'stest(General test):To 2-3 ml aqueous extract,add few drops of alpha-naphthol solution in alcohol,shake&addconcentratrd H ₂ SO ₄ from side of the test tube.	Violet ring is formed at junction of two liquid.	Carbohydrates are present.
2.	Test for proteins: Biuret test (General test): To3 ml T.S.add 4%NaOH & few drops of 1% CuSO ₄ Solution.	No violet or pink colour appears.	Proteins are absent.
3.	Test for Amino acids: Ninhydrin test(General test)Heat 3 ml T.S.and 3 drops 5% Ninhydrin solution in boiling water bath for 10 min.	Purple or bluish colour appears.	Amino acids are present.
4.	Test for fats and oils: Place a thick section of drug on glass slide.Add a drop of sudan red3 reagent. After 2 min. wash with 50% alcohol mount in	No oil globules are appear.	Fats and oils are absent.

	glycerin observe under microscope.		
5.	Test for steroid: Liebaermann-Burchard reaction : mix 2 ml extract with chloroform. Add 1-2 ml acetic anhydride and 2 drops conc. H ₂ SO ₄ from the side of the test tube.	Chloroform layer appears red and acid layer shows greenish yellow fluorescence.	Steroid are present.
6.	Test for cardia glycoside: (keller-killiani test):to 2 ml extract ,add glacial acetic acid ,1 drop 5%FeCl ₃ and conc. H ₂ SO ₄ .	Reddish brown colour appears at junction of the two liquid layers and upper layers appears bluish green.	Cardiac glycosides are present.
7.	Test for saponin glycoside Foam test: shake the drug extract or dry powder vigorously with water.	Persistent foam observed.	Saponin glycoside are present.
8.	Test for Alkaloides: Wagner test 2-3 ml filtrate with few drops wagners reagent.	Reddish brown ppt.	Alkaloids are present.
9.	Test for tannin: 5% FeCl ₃ solution: alcoholic extract,add few drops of 5%FeCl ₃ solution.	Deep blue-black colour.	Tannins are present.

Table1.Preliminary phytochemical Investigation

Evaluation of antioxidant activity:**In vitro antioxidant activity:**

i) DPPH radical scavenging activity: DPPH radical scavenging activity was measured using the method ,with some modifications,3 ml of reaction mixture containing 0.2 ml of DPPH,2.8 ml of test solution ,at various concentrations,(20,40,80,160,320g/ml)of the extract fractions was incubated at 37^oc 30 min. absorbance of the resulting solution was measured at 517 nm using Beckman model DU-40 spectrophotometer,The percentage inhibition of DPPH radical was calculated by comparing the results of the test with those of the control (not treated with extract)using the following equation:

Percentage inhibition=(1-absorbance of test /absorbance of control)x100

ii) Reductive ability: Reducing power of the test samples was determined on the basis of the ability of their antioxidant principles to form colored complex with potassium ferricyanide,TCA FeCl₃ it was measured by the method reported. 1 ml of different concentrations (25,50,100,200,400 g/ml) of the extract fractions were mixed with potassium ferricyanide (2.5 ml,1%)2.5 ml of phosphate buffer (pH6.6).The mixture was incubated at 50^oc for 20 min.2.5 ml TCA(10%)was added to it and centrifuged at 3000

rpm for 10 min. 2.5 ml supernatant was taken out to this 2.5 ml water 0.5 ml FeCl_3 (0.1%) were added absorbance was measured at 700 nm. Higher absorbance of the reaction mixture indicated higher reducing power.

Reagents required:

Sr.NO	Reagents	Preparation
1.	DPPH	100 Mm DPPH in 100 ml Methanol.
2.	Potassium ferricyanide	1 gram potassium ferricyanide in 100 ml of water.
3.	Phosphate buffer (pH 6.6)	By diluting 31.2gms $\text{NaH}_2\text{PO}_4 \cdot 2\text{H}_2\text{O}$ to 1000ml.
4.	TCA (10%)	10 ml TCA in 100 ml water.

Table 2: Reagents required for anti-oxidant activity

Antioxidant assay:

The antioxidant activity of the fenugreek sprouts was determined by two methods: namely β -carotene oxidation and 1,1-diphenyl-2-picrylhydrazyl (DPPH) inhibition. The β -carotene oxidation model system as described by Miller was followed with some modifications.³² The β -carotene solution was prepared by dissolving 10 mg of β -carotene in 50 ml of chloroform in an amber coloured flask to prevent light oxidation. One ml of this solution was pipetted to a flask covered with aluminium foil. Chloroform was then evaporated under vacuum at 40°C for 5 min. The β -carotene was then dissolved in 20 μ l of linolenic acid and 184 μ l of Tween 40 emulsifier. Then 50 ml of H_2O_2 solution (176 μ l H_2O_2 in 100 ml distilled water) was added and mixed thoroughly till the β -carotene was completely dissolved. Five ml of this prepared β -carotene solution was added to 100 μ l of the phenolic extracts. Control tubes contained 100 μ l of 95% ethanol. As soon as the emulsion was added the zero time absorbance was read at 470 nm. Subsequent absorbance readings were recorded after a 30 min incubation period in a 50°C water bath. The antioxidant protection factor (APF) was used to express antioxidant activity as a ratio of sample absorbance at 30 min to that of the control. The antioxidant activity of fenugreek sprouts was also determined by the 1,1-diphenyl-2-picrylhydrazyl (DPPH) assay as described by Cervato.³³ To 3ml of 60 μ M DPPH, 100 μ l of fenugreek sprout extract was added, mixed well and incubated at room temperature for 15 minutes. The absorbance was monitored at 517nm. The fenugreek sprout extract radical scavenging activity was compared with the activity of equivalent concentration of quercetin, a strong antioxidant standard.^[10]

Measurement of Antioxidant Properties

Reducing Power Ability (RPA):

The reducing power of fenugreek extracts was quantified by the method described previously with minor modification.^[7] Fenugreek extract (0, 1.0, 2.0, 3.0, 5.0, 7.0, 9.0, 11.0 mg) in 1 ml of 80% methanol were mixed with phosphate buffer (5.0 ml, 2.0 M, pH 6.6)

and potassium ferricyanide (5.0 ml, 1.0%) the mixtures were incubated at 50 °C for 20 min. A portion (5.0 ml) of trichloroacetic acid (10%) was added and the mixture was

centrifuged at 3000 rpm for 10 min. The upper layer of the solution (5.0 ml) was mixed with distilled water (5.0 ml) and ferric chloride (1.0 ml, 0.1%), and then absorbance of the pink color mixture was measured spectrophotometrically at 700 nm. Increased absorbance of the mixture indicates increased reducing power. The experiment was conducted in triplicate and results were averaged.^[10]

Free Radical Scavenging (FRS) Activity:

Free radical scavenging capacity of fenugreek extracts was determined according to the previously reported procedure using the stable 2, 2-diphenyl-1-picrylhydrazyl radical (DPPH^o).^[6-7] Briefly, a freshly prepared DPPH^o solution in ethanol (0.5 ml) was added to 3 ml of diluted each fenugreek extract to start the radical antioxidant reaction. The final concentration was 100 µM for DPPH^o. The decrease in absorbance was measured at different intervals, i.e. 0, 0.5, 1, 2, 5, 10 and 15 min. up to 50% at 517 nm. The remaining concentration of DPPH^o in the reaction mixture was calculated from a standard calibration curve. The absorbance measured at 5 min of the antioxidant-DPPH^o radical reaction was used to compare the DPPH^o radical scavenging capacity of each fenugreek extract. % of DPPH remaining = $\frac{[DPPH]_T}{[DPPH]_{T=0}} \times 100$ Where T is the time interval.^[11]

Antioxidant capacity:

The antioxidant capacity of the fenugreek extracts were analyzed by using the free radical scavenging (DPPH^o) and the ferric reducing antioxidant power (FRAP) methods. The DPPH^o test is the oldest indirect method for determining the antioxidant activity, which is based on the ability of the stable free radical 2, 2-diphenyl-1-picrylhydrazyl to react with hydrogen donors including phenols.^[7] Radical scavengers may directly react and quench with peroxide radicals to terminate the peroxidation chain reaction and improve the quality and stability of food product. The stable DPPH radical has been used to evaluate antioxidants for their radical quenching capacity and to better understand their antioxidant mechanism(s) each fenugreek extract was evaluated for radical scavenging activity against DPPH^o. The decrease in absorbance of DPPH radical is caused by antioxidant through the reaction between antioxidant molecule and radical results in the scavenging of the radical by hydrogen donation. As (p<0.05) decrease in the concentration of DPPH^o due to scavenging activity of fenugreek extract.^[12]

USES:

1. Ancient use of fenugreek:

Historical uses of fenugreek have been reported by many authors. In the tomb of the Egyptian Pharaoh, Tutankhamun (1333 BC to 1324 B.c.), seeds of fenugreek were found. The Egyptians also used the leaves of fenugreek as one of the components of holy smoke in fumigation and embalming rites (Fazli and Hardman, 1968). Yoshikawa et al. (1997) mentioned that fenugreek was used as an aid to induce labor during childbirth and delivery in ancient Rome.^[8]

2. Fenugreek as a functional food:

Fenugreek is recognized as the principal source of soluble dietary fiber in the plant. Dietary fiber is known to have the potential to reduce risk of cardiovascular disease and to protect against some cancers through the reduction of low density lipoprotein (LDL) and total cholesterol. Fenugreek seeds are most commonly used in everyday life as a spice and a seasoning in soups and curries. Galactomannan (mucilage or gum) in fenugreek acts as a thickener or stabilizer in foods such as soups, sauces and ice-cream^[9,11]. The alkaloid, trigonelline that has been extracted from fenugreek contributes to its distinctive odour. Trigonelline can be used in the manufacture of imitation maple syrup and artificial flavoring for licorice, vanilla, rum and butterscotch.^[9]

3. Fenugreek as a traditional medicine:

Fenugreek seeds and leaves have been used as part of traditional medicinal purposes.^[9] Fenugreek contains phytochemicals such as steroids, flavonoids and alkaloids, which have been identified, isolated and extracted by the pharmaceutical industry to serve as raw materials for the manufacture of hormonal and therapeutic drugs.^[10] In ancient Rome, it was used as an aid to induce labour during childbirth and delivery and in China, it was used as a tonic and treatment for weakness and edema (tissue swelling due to excess lymph fluid) of the legs.^[9] Polysaccharides form the mucilage (galactomannan) present in the plant, and are finding wider applications in the food, pharmaceutical, cosmetics, paint and paper industries.^[1-13]

4. Fenugreek as a forage crop:

During the ancient Greek period, fenugreek was cultivated as a forage crop. The high forage value of fenugreek is attributed to its rich content of protein, vitamins, and amino acids along with its good digestibility in cattle. The seeds contain diosgenin, a growth and reproduction hormone. In another study^[11], fenugreek seed was supplemented into a dairy cattle diet and was reported to significantly improve the fatty acid profile in the milk produced, an increase in the polyunsaturated fatty acid (i.e. linoleic, linolenic and conjugated linolenic acids) concentrations was observed. The study also found that the fenugreek fed cattle had a 4% reduction in blood cholesterol concentration as well as a 19 % decrease in milk cholesterol levels compared to controls, potentially extending health benefits to human consumers of the milk.

5. Fenugreek as animal food:

Although fenugreek is mostly known as a spice crop, the species name *foenum-graecum* refers to “Greek hay” supporting its use as a forage crop in early years (Acharya et al. 2008). It is used as green fodder and hay for cattle in India and Turkey (Petropoulos 2002). In Japan, it is used as silage. Petropoulos (2002) reviewed fenugreek as an alternative to alfalfa or forage peas. Fenugreek seeds are also used as feed for lactating cattle as it increases the flow of milk (Duke 1981; Hidvegi et al. 1984). Its ability to provide high quality forage at all stages of growth has made fenugreek a desirable forage (Acharya et al. 2008).^[12]

6. Agricultural and others uses:

As a legume, fenugreek has the ability to fix atmospheric nitrogen in the soil by harboring nitrogen-binding bacteria in its roots. The crop requires a minimal amount of nitrogen fertilizer for its growth, and reduces the need for nitrogen fertilizers for subsequent crops (Acharya et al. 2010b). Fenugreek is considered a dryland crop thus water requirement of this crop is low. Use of fenugreek in arid and semi-arid environments, and in regions with limited water supply can reduce the cost of irrigation, reduce the potential for eutrophication of surface water and limit contamination of groundwater sources (Acharya et al. 2008; Basu 2006). Fenugreek seed and leaf extracts were reported to have widespread antimicrobial activity against both gram positive and gram negative bacteria (Bhatti et al. 1996).^[11-12]

The aroma and flavor of fenugreek are attributed to volatile constituents it comprises. Fenugreek seed contains 0.02–0.05% volatile compounds (Petropoulos 2002). The major components in this group are heptanoic acid, n-hexanol, dihydroactinolide, dihydrobenzofuran, tetradecane, a-muurolene, b-elemene and pentadecane (Leela and Shafeekh 2008).

Benefits of Fenugreek:

1. 25 - 100 grams of fenugreek seeds eaten daily can diminish reactive hyperglycemia in diabetic patients.^[10]
2. Fenugreek leaves and seeds help in blood formation. They are good for preventing anemia and rundown conditions.
3. Including fenugreek seed in lactating mothers increases the flow of milk.
4. A paste of the fresh fenugreek leaves, applied on the face prevents pimples, blackheads, dryness of the face and early appearance of wrinkles.
5. For removal of dandruff in hair.^[11-13]
6. If you add half a teaspoon of fenugreek seeds to the lentil and rice mixture while soaking, dosas will be more-crisp.^[14]

CONCLUSION:

Trigonella foenum-graecum (fenugreek) is an important culinary and therapeutic plant in many cultures. Fenugreek seeds have been widely studied for their reputed antidiabetic, hypocholesterolaemic, antifertility and hypolipidemic effects. Properties of fenugreek that have been reported but which have received less attention include anticancer, antibacterial, antihelminthic, anti-cholinergic and anti-inflammatory effects. The present research was focused on the seeds of *Trigonella foenum-graecum* extracts (using different extracts), for the pharmacognostic studies such as fluorescence analysis, ash value, extractive value, loss by drying etc. This comparative and multidisciplinary approach to the study of *Trigonella foenum-graecum* does help in understanding its identification taxonomical determination, and medicinal importance in depth. The adulterants in drugs obtain from *Trigonella foenum-graecum* can be identified by this investigation. Adulterants if any can be easily identified using these parameters. In conclusion, the pharmacognostic investigations on physicochemical characteristics and fluorescence analysis shows that authentic botanical of this crude drug prevents adulteration,

substitution and has a crucial role in standardization of crude drugs. This study also indicates that the fenugreek seeds can be an effective treatment for the cancerous conditions and that the indigenous medicinal plants can be used successfully as an alternative treatment in the management of deadly disease such as cancer.

ACKNOWLEDGEMENT

It is said that 'accomplishment must be created to those who had put up the foundation of the particular chore. Here, I pay tribute to **my parents** for lifting up till this phase of life. I thank them for their love, trust, patience and support of course for bearing all kind of stresses, they could, to make me what I am .I owe everything to them and for being my constant companion the strongest source of motivation and inspiration.

I wish to express my deep sense of gratitude and indebtedness to my esteemed guide **Mrs. Sancheti .V.P.** and also thankful to **Mr. Kashid G.A.Sir** for their continuous perseverance, motivation, untiring efforts and encouragement throughout the course.

I am grateful to **Mr.Pawar S.C.** for providing necessary guidance to carry out this work.

I am very thankful to my colleagues **Ashish,Sagar, &Vaibhav** for their contribution and support for my work.

REFERENCES:

1. Anonymous. Quality Control Methods for Medicinal Plant Materials.WHO, Pharma, 1992; 4th edition, 492-559.
2. Ansari SH. Essentials of Pharmacognosy, Birla Publication Pvt. Ltd, 2005; 1th edition, 357-384.
3. Ayurvedic Pharmacopoeia. Ministry of Health and Family Welfare, Govt. of India, New Delhi, 1996; 1, 43.
4. Farnsworth NR. Biological and Phytochemical Screening of Plants, J. Pharm Science, 1966; 55, 225-276.
5. Harborne JB. Phytochemical Methods: A Guide to Modern Techniques of Plant Analysis. Chapman and Hall, London, 1998; 3th edn, 32-38.
6. Sharma R. An evaluation of hypocholesterolemic factor of fenugreek seeds (*T foenumgraecum*) in rats.Nutr Rep Int 1986;33:669- 677.
7. Fenugreek. Available from: <http://www.fenugreek-seeds.com/>
8. Mukherjee PK. Evaluation of Indian Traditional Medicines, Journal of Drug Information, 1996; 35, 631-640.
9. Alkofahi, A., Batshoun, R., Owais, W. and Najib, N. 1996. Biological activity of some Jordanian medicinal plant extracts. Fitoterapia LXVII, 435-42.
10. Mathur, Jaya, Khatri, Pankaj, Samanta, Kartick Chandra. Pharmacognostic and phytochemical investigations of AMARANTHUS SPINOSUS (LINN.)Leaves. International Journal of Pharmacy and Pharmaceutical Sciences, 2010; 2, 121-124.
11. Bhatia, A.L., K. Manda, S. Patni, A.L. Sharma. Prophylactic Action of Linseed (*Linumcatissimum*) Oil Against Cyclophosphamide-Induced Oxidative Stress in Mice Brain - J Med Food, 2006. 9 (2): 261-264.
12. Al-Habori, M. and Raman, A. 2002. Pharmacological Properties in Fenugreek - The genus *Trigonella*(1st edition) by G.A. Petropoulos (ed.), Taylor and Francis, London and New York, 10: 163-182.
13. Tiran, D. 2003. The use of fenugreek for breast feeding women. Complement Ther. Nurs. Midwifery, 9(3): 155-156.

14. Broca, C., Breil, V., Cruciani-Guglielmacci, C., Manteghetti, M., Rouault, C., Derouet, M., Rizkalla, S., Pau, B., Petit, P., Ribes, G., Ktorza, A., Gross, R., Reach, G. and Taouis, M. 2004.
15. The insulinotropic agent 1D1101(4-hydro-xyisoleucine) activates insulin signaling in rat. *Am. J. Physiol. Endocrinol. Metab*, 287(3):E463-E471. Devi, B.A., Kamalakkannan, N. and Prince, P.S. 2003. Supplementation of fenugreek leaves to diabetic rats- Effect on carbohydrate metabolic enzymes in diabetic liver and kidney. *Phytother Res.*, 17(10): 1231-1233.
16. Suboh, S.M., Bilto, Y.Y. and Aburjai, T.A. 2004. Protective effects of selected medicinal plants against protein degradation, lipid peroxidation and deformability loss of oxidatively stressed human erythrocytes. *Phytother. Res.*, 18(4):280-284.
17. Devasena, T. and Menon, V.P. 2003. Fenugreek affects the activity of beta-glucuronidase and mucinase in the colon. *Phytother Res.*, 17(9): 1088-1091.
18. Tahiliani, P. and Kar, A. 2003b. Mitigation of thyroxine induced hyperglycaemia by two plant extracts. *Phytother. Res.*, 17(3): 294-296
19. Spice board of India. Cultivation practice. Fenugreek. Available from: http://www.indianspices.com/html/spices_spfarm_fenu.html
20. Mehrafarin A A, Rezazadeh SH, Naghdi BH, Noormohammadi GH, Zand E, Qaderi A. A review on biology, cultivation, biotechnology of fenugreek as a valuable medicinal plant and multipurpose. *Journal of Medicinal Plants*. 2011; 10(37): 1-19.
21. Indus extract. Fenugreek. Available from: <http://www.indusextracts.com/trigonella-foenium-dry-extract.htm>
22. Indian Herbal Pharmacopoeia. Indian Drugs Manufacturing Association Mumbai, Regional Research Laboratory Jammu Tawi, 1999; 1, 33.
23. Madhav, N.V. Satheesh, Kumer Upadhyaya. Phytochemical screening and standardization of poly herbal formulation of dyslipidemia. *International Journal of Pharmacy and Pharmaceutical Sciences*, 2011; 3, 235-238.
24. Fenugreek: Archeology, Properties, Constituents and Biochemicals. Available from: <http://www.mdidea.com/products/new/new00405.html>
25. Kokate CK, Purohit AP. and Gokhale SB. *A Text Book of Pharmacognosy*, 2005; 31, 130-132.
26. Beautiful. Available from: <http://www.beauti-full.com/ingredients.htm>
27. Herb and supplement encyclopedia. Fenugreek seeds. Available from: <http://www.florahealth.com/flora/home/canada/healthinformation/encyclopedias/fenugreekseed.asp>
28. Natural agro product. Fenugreek seeds. Available from: <http://naturalagroproducts.com/fenugreek.html>
29. Vortex health. Fenugreek. Available from: <http://www.vortexhealth.net/fenugreek.html>
30. Cidpusa foundation. Fenugreek seeds. Available from: <http://www.cidpusa.org/Fenugreek%20seed.html#FS-GENERAL>.