

Phytochemical, physicochemical, and fatty acids composition analyses of four species of *Ficus* genus from the arid zone of Rajasthan

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Abstract

Background: With the advancement of the oilseed economy, there is a great deal of interest in using unusual and underexploited sources of seed oils. The seed oils of different species of *Ficus* genus are well known for their nutritional and medicinal potential.

Aims: The aim of the present research work is to investigate the phytochemical, physicochemical, and fatty acids composition of the seed oil from four different species of *Ficus* genus from the arid zone of Rajasthan with a view to analyze their nutritional and therapeutic potential.

Materials and Methods: The seed oils of *Ficus benghalensis*, *Ficus benjamina*, *Ficus carica*, and *Ficus racemosa* were extracted by soxhlet extraction technique using petroleum ether as a solvent. The fatty acid methyl esters were obtained by transesterification process and their fatty acid profiles were studied by gas chromatography-mass spectrometry technique.

Results: The phytochemical analysis showed carbohydrate, flavonoids, alkaloids, terpenoids, and tannin in the seed oil of these *Ficus* species. The study suggested the presence of unsaturated fatty acids in *F. benjamina* (71.72%), *F. racemosa* (70.44%), *F. carica* (59.00%), and *F. benghalensis* (55.32%).

Conclusion: The major amount of polyunsaturated fatty acids in *F. benjamina* (60.81%) and alpha linolenic acid in *F. carica* (12.74%) indicates their nutritional and medicinal values.

Keywords: Fatty acids analyses, *Ficus* genus, phytochemical composition

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INTRODUCTION

The genus *Ficus* belonging to the family *Moraceae* is classified among the biggest genera of dicotyledons with approx. Eight hundred species of woody trees including shrubs found globally in the tropic and sub-tropics areas.^[1] They are also known as fig trees. About 500 *Ficus* species are found mainly in Asia and Australia. The genus consists of many varieties with considerable genetic diversity and excellent pharmaceutical activities that are of terrific trade value.^[2] The *Ficus* species are widely known for their use as traditional

medicines. Its species have been recognized as a major source of phenolic acid and flavonoids which lead them to be useful in the treatment of oxidative stress disorders.^[3] Plant extracts obtained from *Ficus* species have been found to be useful in the treatment of stomach pain, cancer, hemorrhoids, gastric ulcer, diarrhea, various other inflammation, and long-term oxidative stress.^[4] *Ficus* species are also biologically active as they showed anti-cancer, anti-diabetic, and anti-inflammatory properties.^[5] In the present study, seeds from

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four plant species; *Ficus benghalensis*, *Ficus benjamina*, *Ficus carica*, and *Ficus racemosa* belonging to *Moraceae* family were investigated for their phytochemical, physicochemical properties and fatty acid compositions using chemical, chromatographic, and spectroscopic techniques. The increased intake of saturated fats is allied with cardiovascular disease, which is the leading global cause of 30% of mortality in modern society.^[6] Currently, omega-3 (ω -3) and omega-6 (ω -6) polyunsaturated fatty acids (PUFAs) are highly desired ingredients in oil with unique dietary and functional characteristics. To enhance and sustain human health over the entire lifespan, PUFA is considered essential active mediators, but only the cardiac effect has been widely stated. In recent times, omega-3 fatty acids have been shown to perform a valuable role in the treatment of severe human diseases, particularly obesity and type 2 diabetes mellitus, in addition to this PUFA are also attributed with a reduced rate of stroke, atherosclerosis, and cardiac problems. Omega-3-Alpha-linolenic fatty acids (ALA) are required for normal health, especially for the brain development and function.^[7] Nicolas B *et al.*, has suggested that ALA has anti-inflammatory and other potential beneficial properties therefore it may reduce stroke risk, size, and/or consequences.^[8] Many countries in their dietary guidelines promoted intake of PUFA^[9] such as Dietary Guidelines for Americans,^[10] American Heart Association,^[11] and Brazilian Society of Cardiology.^[12] Recently, in economically advanced countries oil rich in unsaturated fatty acids are usually recommended in contrast to saturated fats.^[13] With a view to figure out the nutritional value and therapeutic potential of the four *Ficus* plant species, in the current research, their seed oils were extracted and their phytochemical, physicochemical, and fatty acid compositions were analyzed.

MATERIALS AND METHODS

Sample collection

In the present study *F. benghalensis*, *F. benjamina*, *F. carica*, and *F. racemosa* seeds were collected from the Jodhpur district of Rajasthan (India). The fresh seeds samples were dried in shade and grounded to fine powder. One-half of the seeds were used for phytochemical analysis and the rest for extraction of oil from seeds.

Preliminary phytochemical analyses

Extraction

Twenty gram of powdered seeds were dissolved in 100 ml of methanol. Methanolic extract of seeds was obtained using shaker system for about 48 h. The seed extract of all the four plant species was tested for the presence of carbohydrates, proteins, amino acids, steroids, glycosides, flavonoids, alkaloids, tannins, saponins, terpenoids, and resins. The phytochemical analysis of the methanolic extracts is carried out using standard methods shown in Table 1.^[14-16]

Physico-chemical analyses

The rest halves of the grounded seeds were weighed.

Moisture content

Weighed amount of fresh grounded seeds samples were taken then it was placed in a hot air oven for about an hour for 1000C and the dry weight of the seeds sample become constant.

Table 1: Phytochemical analysis of *Ficus* species

Phytochemical constituents	<i>Ficus benghalensis</i>	<i>Ficus benjamina</i>	<i>Ficus Carica</i>	<i>Ficus racemosa</i>
Carbohydrate	+	+	+	+
Amino acids	-	-	-	-
Starch	+	+	+	+
Flavonoid	+	+	+	+
Alkaloids	+	+	+	+
Tannin	+	+	+	+
Saponins	-	+	-	-
Terpenoids	+	+	+	+
Glycosides	-	+	-	-
Steroids	-	-	-	-

+: Presence of compound; -: Absence of Compound

$$\text{Moisture \%} = \frac{\text{weight of fresh seeds} - \text{weight of dried seeds}}{\text{weight of fresh seeds}} \times 100$$

Extraction of oils

Oils were extracted using the soxhlet apparatus and petroleum ether (40–600C) as a solvent. The rotary evaporator was used to evaporate the remaining solvent in vacuum. The standard methods of American Oil Chemist's Society (AOCS) were employed to find out the analytical values of oil seeds.^[17] Fatty acids methyl esters (FAMES) of the seed oils were prepared using the transesterification process.

Refractive indexes

Abbe's refractometer was used to determine (Refractive index [R. I] value) of the oils. The Kjeldahl method was adopted to evaluate protein content from defatted seeds.

Saponification value

One gram weighed oil samples were reacted with 50 ml of 4% alcoholic KOH solution then refluxed it for about 3–4 h. Further, titrate it with 0.5N NaOH solution. Phenolphthalein is used as an indicator. A blank determination was also performed simultaneously.

Iodine value

Wiz's method is adopted to elucidate the iodine values of the seed oils. In a round bottom flask, 1 g of oil samples + 25 ml Wiz's solution + 25 ml carbon tetrachloride were reacted. Then, it was kept in dark for about 30–40 min. Thereafter, 20 ml of 15% KOH + 100 ml of water were added to this mixture. The iodine was liberated which then made to titrate against 0.1N Hypo solution. One percent of starch solution used as an indicator. Along with this, a blank titration was also performed.

Analytical techniques

Thin layer chromatography

The thin layer chromatography was performed using silica gel G as an adsorbent material. Hexane-diethyl ether-acetic acid in the ratio of (80:20:1) was used as a mobile phase. The sample spots were observed under an iodine chamber.

Gas chromatography-mass spectrometry

Thermo scientific TSQ 8000 Gas chromatography-mass spectrophotometer used for analyses of FAMES of the *Ficus* species. The carrier gas was helium with a flow rate of 1 mL/min. The injector and detector temperature were maintained at 250°C and 260°C, respectively. The initial temperature of the oven was 80°C and

raised up to 200°C then further increased to 250°C with increase rate of 8°C–10°C/min. A capillary column of polysilphenylene-siloxane was used.

RESULTS AND DISCUSSION

The results of the qualitative screening of *Ficus* species in methanolic extract of seeds are shown in Table 1. The results indicated the presence of biologically active chemical components in the seeds of all the four *Ficus* species. The phytochemical compounds such as carbohydrates, starch, flavonoid, alkaloid, tannin, and terpenoids are found in the methanolic extract of all the four species. Amino acids and steroids were completely absent in all the *Ficus* species. Saponin was found only in *F. benjamina* and *F. racemosa*. The oil percentages in the *Ficus* species were ranging from 12.34% to 17.23%. *F. benghalensis* showed the highest percentage of protein (10.62%). *F. carica* showed the highest percentage of moisture (9.71%). All the four *Ficus* species were found to have high saponification values ranging 190.77–203.24. The Iodine values for *F. benghalensis*, *F. benjamina*, *F. carica*, and *F. racemosa* were found 89.67, 76.92, 99.07, and 106.34, respectively. The R. I values for the *Ficus* species seeds oils were found in between 1.47 and 1.48. The Gas chromatography–mass spectrometry analysis of fatty acids in the oils from seeds of *Ficus* species have been recorded in Figures 1a, b and 2a, b. The identification of saturated and unsaturated fatty acid in all the *Ficus* species was performed by comparing its retention time with that of standards lipids. Different types of unsaturated and saturated acids were found in these seed oils. Among the unsaturated fatty acids, linoleic acid was reported in the highest amount (55.42%) in *F. benjamina* and Alpha-Linolenic acid (ω -3) was observed in moderate amount (12.74%) only in *F. carica*. The unsaturated fatty acids were reported in significant amounts in all the above-mentioned *Ficus* species varied from 55.32% to 71.72%. The PUFA was found in the major amount in *F. benjamina* (60.81%) given in Table 2. From the preliminary qualitative phytochemical screening of *Ficus* species seeds extracts, it was found that their methanolic extract possessed maximum quantity of secondary metabolites. Akanni *et al.* mentioned that seed oils with high saponification values are important from

the industrial point of view,^[18] as they played crucial role in soap and cosmetic industries which further favors the industrial potential of these *Ficus* species seed oils. Researchers have reported that oils having saturated fatty acids in majority showed low iodine values, while oils having unsaturated fatty acids in majority have high iodine values.^[19] The significantly higher iodine values of these *Ficus* species account for their good percentage of unsaturated fatty acids. Similar to the study of Emine Nakilcioglu *tas F. carica* was found to have a higher proportion of Unsaturated Fatty Acids (UFA) than Saturated Fatty Acids (SFA).^[20] Emine in his research also suggested that fig seeds have a crucial role in human health due to their rich nutritional content. On contrary to the findings of Lahcen *et al.*^[21] and Ozge *et al.*^[22] in *F. carica* seed oil we have found higher percentage of linoleic acid (18:2) than linolenic acid (18:3), but the presence of Alpha-linolenic acid (ω -3) was in fare agreement with both of them.

CONCLUSION

On account of high saponification value, all these four species of *Ficus* have industrial significance. The higher amount of unsaturated fatty acids and polyunsaturated fatty acids in *F. benjamina*, *F. racemosa*, and the presence of Alpha-linolenic acid (ω -3) in *F. carica* made them valuable for human nutrition and medicinal point of view. The presence of phytochemical bioactive compounds which include flavonoids, terpenoids, alkaloids, and tannin in all these species furnishes their importance from nutritional and pharmaceutical perspectives. This research provides a base for further analyses of *Ficus* species for their phytochemical and nutritional characteristics.

Table 2: Cumulative fatty acid composition

Name of <i>ficus</i> species	Total SFA (%)	Total UFA (%)	PUFA (%)
<i>Ficus benghalensis</i>	44.68	55.32	39.91
<i>Ficus benjamina</i>	28.27	71.72	60.81
<i>Ficus carica</i>	41.00	59.00	34.91
<i>Ficus racemosa</i>	17.45	70.44	40.13

SFA: Saturated fatty acid, UFA: Unsaturated fatty acid, PUFA: Polyunsaturated fatty acid

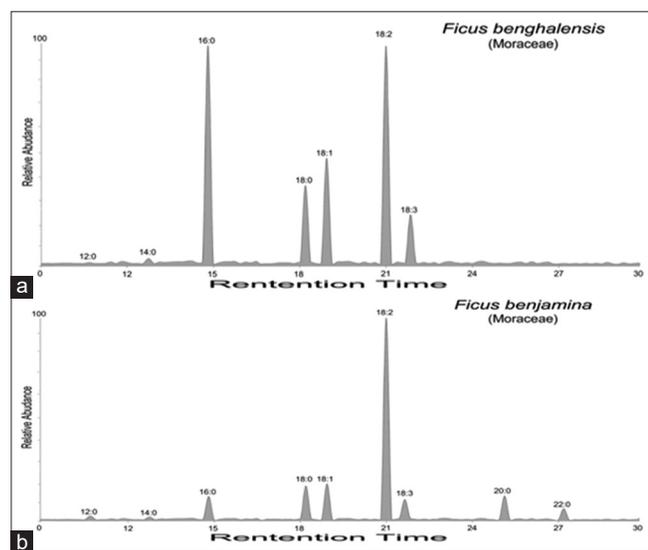


Figure 1: Gas chromatography–mass spectrometry graphs for (a) *Ficus benghalensis* (b) *Ficus benjamina*

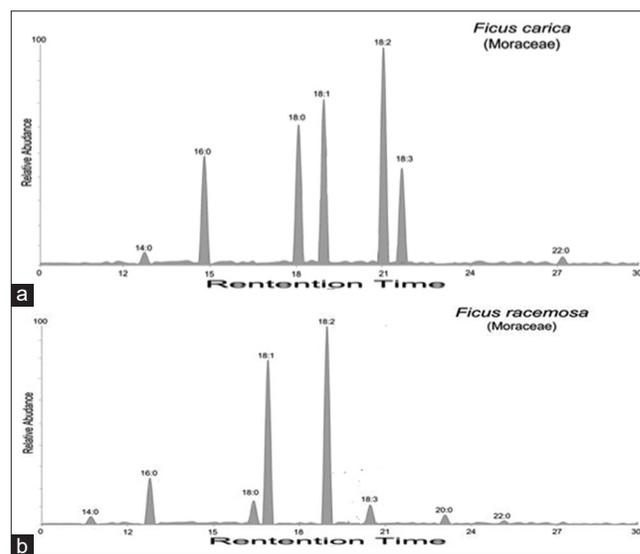


Figure 2: Gas chromatography–mass spectrometry graphs for (a) *Ficus carica* and (b) *Ficus racemosa*

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Conflicts of interest

There are no conflicts of interest.

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