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Phytochemical screening and GC–MS analysis of *Tamarindus indica* L. fruit shell extract

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Abstract
The present study was undertaken to investigate the phytochemical constituents of *Tamarindus indica* L. fruit shell through qualitative phytochemical screening and gas chromatography–mass spectrometry (GC-MS) analysis. Fruit shell extracts prepared using water, ethanol, chloroform and petroleum ether were screened for different classes of phytochemicals. The results revealed solvent-dependent variations in the detected constituents. Alkaloids and flavonoids were present in all four extracts. Coumarins and glycosides were detected in the aqueous, ethanolic and chloroform extracts, whereas phenols and saponins were detected only in the aqueous extract. Phlobatannins and terpenoids were detected in the ethanolic extract while quinones were present in the water, ethanolic and petroleum ether extracts. Tannins were not detected in any of the tested extracts. GC-MS analysis of the fruit shell water extract resulted in the reported identification of nine chemical constituents through comparison of their mass spectra with the National Institute of Standards and Technology (NIST) library. The reported compounds included 2-furaldehyde, 3-hexen-2-one, 4-hydroxy-4-methylpentan-2-one, 4-hydroxymethyl-2-furaldehyde, 1-octadecyne and fatty-acid-related compounds. Overall, the findings demonstrate that *T. indica* fruit shell contains diverse phytochemical constituents whose detection varies with the extraction solvent. The results provide preliminary chemical information that may support further investigations into the composition and potential applications of tamarind fruit shell.

Keywords: *Tamarindus indica* L., fruit shell, phytochemical screening, GC-MS

Introduction

Plants are important sources of natural bioactive compounds, particularly secondary metabolites such as alkaloids, flavonoids, phenolics, tannins, saponins and terpenoids. These compounds play important roles in plant defence and have attracted considerable scientific interest because of their structural diversity and potential nutritional, medicinal and biological applications. Phytochemical investigation combined with modern analytical techniques provides valuable information about the chemical composition of plant materials and supports their scientific characterization (Harborne, 1998; Wink, 2013; Amir et. al., 2019, Mukherjee et. al., 2024) [2, 17, 24].

Tamarindus indica L. (Fabaceae), commonly known as tamarind, is a widely distributed tropical tree valued for its edible fruits, traditional uses and economic importance. Different plant parts, including leaves, pulp, seeds, bark and fruit peel, have been investigated for their phytochemical composition and biological properties (Bhadoriya et. al., 2011; Menezes et. al., 2016; Komakech et. al., 2019) [3, 11, 16]. Previous studies have reported diverse phytochemical constituents in tamarind leaves and other plant parts, demonstrating that their chemical profiles may vary according to the plant tissue and extraction solvent (Sharma et. al., 2021; Mehdi et. al., 2021) [15, 20]. Furthermore, the utilization of tamarind processing by-products has gained attention because of their potential industrial and environmental applications (Mansingh et. al., 2021) [13].

Qualitative phytochemical screening is a preliminary approach for detecting major groups of secondary metabolites in plant extracts. However, it does not establish the identity of individual chemical constituents. Therefore, gas chromatography–mass spectrometry (GCMS) is frequently employed as an analytical technique for separating and tentatively identifying volatile and suitably semi-volatile compounds through their chromatographic characteristics and mass spectra. Previous investigations have demonstrated the applicability of GCMS for characterizing chemical constituents in tamarind leaves, peel and seed extracts (Sharma et. al., 2021; Mehdi *et al*., 2021; Tavanappanavar et. al., 2024) [15, 20, 22]. Similarly, pharmacognostical and physicochemical investigations have emphasized the importance of systematic chemical characterization of tamarind-derived materials (Amir et. al., 2019) [2].

Although previous studies have investigated various aspects of *Tamarindus indica* L. phytochemical composition of fruit shell extracts may vary with geographical origin, extraction solvent, and analytical conditions. Therefore, the present investigation, titled “Phytochemical Screening and GCMS Analysis of *Tamarindus indica* L. Fruit Shell Extract,” was undertaken to evaluate the qualitative phytochemical constituents of fruit shell extracts prepared with different solvents and to characterize their detectable chemical compounds using GCMS analysis. The study aims to provide additional information on the chemical composition

of tamarind fruit shell and to offer a scientific basis for subsequent investigations into its potential applications.

Methods and Material

Collection and Preparation of Plant Material

The fruit shells of *Tamarindus indica* L. were collected from the local area of Nanded district, Maharashtra, India. The collected plant material was authenticated by Dr. Dinesh Vijigiri, Head, Department of Botany, Shri Renuka Devi Mahavidyalaya, Mahur, Taluka Mahur, District Nanded, Maharashtra. The fruit shells were thoroughly cleaned to remove adhering dust and other foreign particles and then shade-dried under suitable conditions. The dried material was ground into a fine powder using a mortar and pestle. The powdered fruit shell material was stored in airtight containers at 4°C until further use for solvent extraction, qualitative phytochemical screening, and gas chromatography mass spectrometry (GC-MS) analysis.

Preparation of Fruit Shell Extracts by Soxhlet Extraction

The powdered fruit shell material of *Tamarindus indica* L. was subjected to Soxhlet extraction using four different solvents: water, ethanol, chloroform and petroleum ether. Briefly, 50 g of powdered fruit shell material was extracted separately with each solvent using a Soxhlet apparatus for 9 hours. Following extraction, the resulting extracts were concentrated using a rotary evaporator. The concentrated extracts were stored at 4°C until further use for qualitative phytochemical screening and gas chromatography–mass spectrometry (GC-MS) analysis.

Qualitative Phytochemical Screening.

Qualitative phytochemical screening was performed to investigate the presence of selected phytochemical constituents in the fruit shell extracts of *Tamarindus indica* L. The water, ethanol, chloroform, and petroleum ether extracts were examined for alkaloids, saponins, glycosides, flavonoids, phenols, and tannins using standard qualitative phytochemical tests, following the methods described below

Qualitative Phytochemical Analysis

Qualitative phytochemical screening of the water, ethanol, chloroform and petroleum ether extracts of *Tamarindus indica* L. fruit shell was performed to determine the presence of selected phytochemical constituents using standard qualitative methods.

Test for Phenolic Compounds: The extract (500 mg) was dissolved in 5 mL of distilled water, and a few drops of neutral 1% aqueous or alcoholic ferric chloride solution were added. The development of green, purple, blue, or black coloration indicated the presence of phenolic compounds (Harborne, 1973) [9].

Test for Flavonoids: To 10 mL of the aqueous filtrate, 5 mL of dilute ammonia solution was added, followed by a few drops of concentrated sulphuric acid. The appearance of yellow coloration, which disappeared on standing, indicated the presence of flavonoids (Harborne, 1973) [9].

Test for Tannins: Approximately 0.5 g of the powdered fruit shell material was boiled in 20 mL of distilled water and filtered. A few drops of 0.1% ferric chloride solution were added to the filtrate. The development of bluish-black or brownish-green coloration indicated the presence of tannins (Trease and Evans, 1989) [23].

Test for Alkaloids: Approximately 0.5–0.6 g of the powdered fruit shell material was mixed with 8 mL of 1% hydrochloric acid, gently warmed, and filtered. Two millilitres of the filtrate were treated separately with Mayer's reagent and Dragendorff's reagent. The formation of a precipitate or turbidity indicated the presence of alkaloids (Harborne, 1973) [9].

Test for Glycosides: Cyanogenic glycosides were qualitatively examined using the sodium picrate paper test. Fresh plant material (100 mg) was immediately ground and mixed with 1 mL of distilled water in a screw-capped tube. Yellow sodium picrate paper was suspended inside the tube without touching the liquid, and the tube was sealed. A change in the paper colour from yellow to reddish-brown indicated the possible presence of cyanogenic compounds, associated with the release of hydrogen cyanide (Bradbury et. al., 1999) [4].

Test for Saponins: Approximately 2 g of the powdered fruit shell material was boiled in 20 mL of distilled water in a water bath and filtered. Ten millilitres of the filtrate were mixed with 5 mL of distilled water and shaken vigorously to produce froth. Three drops of olive oil were then added, and the mixture was shaken again. The formation of an emulsion indicated the presence of saponins (Trease and Evans, 1989) [23].

Test for Terpenoids: 5 mL of the aqueous extract were mixed with 2 mL of chloroform, followed by the careful addition of 3 mL of concentrated sulphuric acid to form a separate layer. The development of reddish-brown coloration at the interface indicated the presence of terpenoids.

Test for Phlobatannins: 2 mL of the aqueous extract were treated with dilute hydrochloric acid. The formation of a red precipitate indicated the presence of phlobatannins.

Test for Coumarins: 1mL of the extract was mixed with 1 mL of 10% sodium hydroxide solution. The development of yellow coloration indicated the presence of coumarins.

Test for Quinones: 1 ML of the extract was treated with 1 mL of concentrated sulphuric acid. The development of red coloration was considered a positive indication of quinones.

GC-MS analysis

Degradation of the dyes by the isolates was confirmed by GCMS analysis. The samples were prepared by extracting the products in Di-Chloro Methane (DCM). The decolorized broth was centrifuged at 10,000 RPM for 20min. The supernatant was separated out and was collected in a separating funnel. An equal amount of dichloromethane (DCM) was added to the separating funnel possessing the supernatant. The funnel was shaken energetically to extract the products in DCM for 20 min. The separating funnel was

left undisturbed for the separation of two phases like the solvent phase and the liquid phase. The separated solvent was left out from the funnel. The products were extracted in the solvent. These extracted products were concentrated in a plastic vial by evaporation of the solvent at room temperature (RT). This was then analyzed by Gas chromatography and Mass spectroscopy (GCMS). The GC-MS analysis of the metabolites was done using a Shimadzu 2010 MS Engine, equipped with integrated gas chromatograph having HP1 column (0.25 mm id, 60m long, non-polar). The carrier gas used was Helium, having a flow rate of 1mlmin $^{-1}$. The injector temperature was maintained at 280 ^{0}C and was kept constant for 2min and with 10 ^{0}Cmin $^{-1}$ rate elevatedup to 200 ^{0}C. The compounds were identified on the basis of mass spectra and using the NIST library (Shertate R. and Thorat P. R., 2012) [21].

Result

Qualitative analysis

The results of qualitative phytochemical characterization of *Tamarindus indica* L. (Fruit shell) extract of water solvent showed the presence of alkaloid, coumarins, flavonoids, glycoside, phenols, quinone and saponins. Whereas, in ethanol solvent showed the presence of alkaloid, coumarins, flavonoids, glycoside, phenols, Phlobatanin, quinone and terpenoids. While in the chloroform extract alkaloids, coumarins, flavonoids, glycosides and Phlobatanin are found. In the petroleum ether extract alkaloids, flavonoids and quinones are shown in Table 1.

The results of qualitative phytochemical characterization of *Tamarindus indica* L. (Fruit shell) has good source of bioactive molecules *Viz.*, alkaloids, coumarins, flavonoids, glycosides, phenols, Phlobatanin and quinone.

Table 1: Qualitative Phytochemical Characterization of *Tamarindus indica* L. (Fruit shell) in Different Solvents.

Phytoconstituents	Solvents			
	Water	Ethanol	Chloroform	Petroleum ether
Alkaloids	+	+	+	+
Coumarins	+	+	+	-
Flavonoids	+	+	+	+
Glycoside	+	+	+	-
Phenols	+	+	-	-
Phlobatanin	-	+	+	-
Quinone	+	+	-	+
Saponins	+	-	-	-
Tannins	-	-	-	-
Terpenoids	-	+	-	-

(Where: - Absent, + Present)

The results of the preliminary phytochemical studies in different parts of the plant confirmed the presence of more bioactive compounds such as alkaloids, flavonoids, phenols, saponins, steroids, tannins and terpenoids in the water extract of different medicinal plant *Tamarindus indica* L. (Fruit shell). Hence, further GCMS, analyses were carried out in water extract to assess the active compounds with bioactive properties.

GC-MS analysis of fruit shell extract of *Tamarindus indica* L.

Characterizations of Active Principles from Bioactive Extracts:

The chemical constituents present in the aqueous extract of *Tamarindus indica* L. fruit shell were characterized using gas chromatography–mass spectrometry (GC–MS). The GC–MS chromatogram was examined to determine the retention behaviour and mass spectra of the detected compounds. The molecular ions and characteristic fragmentation patterns were analysed to obtain information about the molecular masses and structural characteristics of the detected constituents. The mass spectra of the individual peaks were compared with the reference spectra available in the National Institute of Standards and Technology (NIST) library for tentative identification of the compounds. The GC–MS chromatogram and corresponding mass spectra of the detected constituents are presented in the figure.

GC-MS Analysis and Identification of Compounds present in Plant Extracts

Gas Chromatography-Mass Spectroscopy (GC-MS) is a powerful technique employed for many applications which has very high specificity and sensitivity. The combination of a principle separation technique Gas Chromatography with the best identification technique Mass Spectroscopy (MS) made Gas Chromatography-Mass Spectroscopy (GC-MS) an ideal for the qualitative and quantitative analysis for volatile and semi-volatile compounds (Hussein *et al.*, 2016) [10].

Interpretation of GC-MS mass spectrum after GC-MS analysis of different medicinal plant extracts achieved using the database of National Institute Standard and Technology (NIST) having more than 60,000 patterns stored in the system library. The spectrum of the unknown phyto component was further compared with the spectrum of the known phyto components which was stored in the NIST library. The name of phyto component, molecular weight and molecular formula of the components of in the medicinal plants were then ascertained.

Table 2: GCMS Analysis of *Tamarindus indica* L. (Fruit Shell)

Sr. No.	Compounds	Molecular Formula	Molecular Weight
1.	7-hydroxyl-oct-3-ene-2-carboxylic acid	$C_{10}H_{14}O_3$	182
2.	1-Octadecyne	$C_{18}H_{34}$	250
3.	Malonic ester	$C_{18}H_{12}O_4$	172
4.	6 Doeicosenoic acid	$C_{22}H_{42}O_2$	338

5.	Octandecanoic acid (Stearic acid)	$C_{16}H_{36}O_2$	284
6.	2-Furaldehyde	$C_5H_4O_2$	96
7.	3-Hexen-2-one	$C_6H_{10}O$	98
8.	4-hydroxyl-4-methyl pentan-2-one	$C_6H_{12}O_2$	116
9.	4-hydroxylmethyl-2-furaldehyde	$C_6H_6O_3$	126

GC-MS chromatogram of the water leaf extract of *Tamarindus indica* L. (Fruit Shell) showed 09 major peaks (Figure 1) and have been identified after comparison of the mass spectra with NIST library (Table 2), indicating the presence of various phyto components. Nine compounds were found in the water leaf extract of *Tamarindus indica* L. (Fruit Shell) as depicted in Table 2 *Viz.* – 7-hydroxyl-oct-3-ene-2-carboxylic acid, 1-Octadecyne, Malonic ester, 6 Doeicosenoic acid, 6 Doeicosenoic acid, Octandecanoic acid (Stearic acid), 2-Furaldehyde, 3-Hexen-2-one, 4-hydroxyl-4-methyl pentan-2-one and 4-hydroxylmethyl-2-furaldehyde.

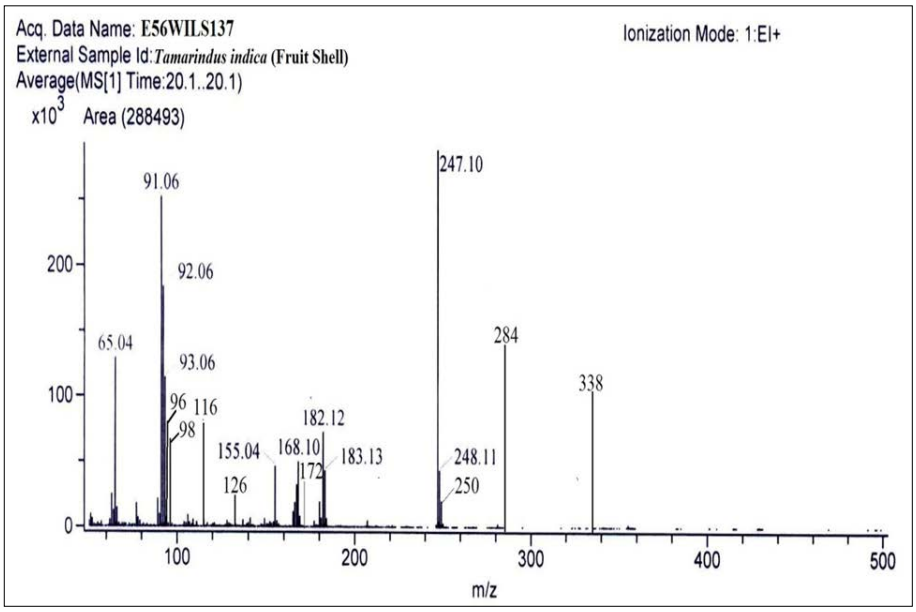


Fig 1: GCMS Analysis of *Tamarindus indica* L. (Fruit Shell)

Discussion

Phytochemical Screening and GC–MS Analysis of *Tamarindus indica* L.

Previous investigations have demonstrated that *Tamarindus indica* L. contains a wide range of phytochemical constituents, including alkaloids, flavonoids, phenolic compounds, tannins, terpenoids, saponins, quinones and glycosides. Gomathi et. al., (2017) [8] reported the occurrence of several of these phytochemical groups in the aqueous fruit shell extract, suggesting that tamarind shell represents a potential source of naturally occurring bioactive compounds. Similarly, Roy et. al., (2020) [18] identified alkaloids, glycosides, saponins, tannins and phenolic compounds in hydromethanolic extracts of tamarind leaves, fruit pulp and seed coats. Their findings also demonstrated that the extraction solvent influenced the recovery of phytochemicals with hydromethanolic extracts exhibiting comparatively greater phytochemical diversity than aqueous extracts. Senthilkumar et. al., (2021) [19] further confirmed the presence of carbohydrates, phenols, tannins, flavonoids, alkaloids, terpenoids and anthocyanins in methanolic and ethanolic extracts, although saponins, steroids and glycosides were not detected. In addition, Mahm, (2021) reported 38 chemical constituents in the aqueous leaf extract of *T. indica*, including 3-O-Methyl-D-glucose, myo-inositol derivatives, propane derivatives, 2-pentanone derivatives, and different nitrogen-containing compounds. Collectively, these studies establish the phytochemical richness of different parts of tamarind and demonstrate the influence of plant material and extraction solvents on the composition of the resulting extracts.

Further investigations have provided evidence of considerable variation in the phytochemical composition of different tamarind plant parts. Ebabhi et. al., (2025) [5] identified saponins, reducing sugars, terpenoids, cardiac glycosides, steroids, flavonoids and phenolic compounds in the investigated plant extract. Similarly, Marques et. al., (2026) [14] observed a broader range of phytochemical constituents in tamarind seed extracts, including flavonoids, tannins, terpenoids, saponins, amino acids and cardiac glycosides, whereas the peel extracts exhibited a comparatively restricted profile, predominantly characterized by reducing sugars. These observations suggest that phytochemical distribution varies considerably among plant organs and that the choice of extraction solvent and plant material plays an important role in determining the diversity of compounds detected.

GC–MS investigations have further contributed to the identification of individual chemical constituents present in *Tamarindus indica*. Akter et. al., (2023) [1], while comparing sour and sweet tamarind extracts, identified several compounds, including cyclohexanamine, 2-furanethanol derivatives, 1-ethyl-2-hydroxymethylimidazole, hydrazinecarboxamide derivatives, fumaric acid esters, hydroxymethyl-substituted furan derivatives and 3-O-Methyl-D-glucose. Other constituents such as N-glycylglycine, hexadecenoic acid derivatives, methyl-substituted pyranones and cis-13-octadecenoic acid were

also detected. Fagbemi et. al., (2022) [6] reported 5-hydroxymethylfurfural (31.06%) as the predominant constituent in the methanolic extract, followed by 3-O-Methyl-D-glucose (16.31%) and 1,6-anhydro-β-D-glucopyranose (9.95%). Additional compounds included furan derivatives, pyranones, n-hexadecanoic acid, cis-vaccenic acid and several sugar derivatives. Similarly, Ghaly et. al., (2023) [7] identified bis(2-ethylhexyl) phthalate, 2,4-bis(1,1-dimethylethyl) phenol and 1,2-benzenedicarboxylic acid derivatives among the major constituents of a purified ethanolic extract, with reported relative percentages of 56.5%, 43.6%, and 22.4%, respectively.

Furthermore, Tavanappanavar et. al., (2024) [22] demonstrated the presence of diverse chemical constituents in chloroform and ethyl acetate extracts of *T. indica* peel through GC–MS analysis. The chloroform extract was characterized by compounds such as hexadecanoic acid, 9,12-octadecadienoic acid, (9Z)-octadecenoic acid, neophytadiene and 7,9-di-tert-butyl-1-oxaspiro (4,5) deca-6,9-diene-2,8-dione. In contrast, the ethyl acetate extract contained isovanillic acid, tetradecanoic acid, 9-octadecenoic acid, methyl palmitate, hexadecanoic acid and ethyl palmitate. The authors also reported considerable phytochemical concentrations in the ethyl acetate seed extract, particularly terpenoids, phenolics and flavonoids. These findings highlight the importance of solvent selection in extracting and characterizing different classes of phytochemicals.

Overall, the findings of these previous investigations demonstrate that *Tamarindus indica* L. is a rich source of diverse primary and secondary metabolites, including phenolic compounds, flavonoids, terpenoids, organic acids, fatty acids and carbohydrate derivatives. The differences in reported chemical profiles may be attributed to variations in plant parts, geographical origin, extraction procedures, solvent polarity and analytical conditions. These studies provide a valuable foundation for the present investigation on the phytochemical screening and GC–MS characterization of *Tamarindus indica* L. fruit shell extract. In particular, the available literature indicates that the fruit shell deserves further investigation as a potential source of bioactive chemical constituents and provides a basis for comparing its phytochemical composition with that of other tamarind plant parts.

Conclusion

The present investigation demonstrated the presence of diverse phytochemical constituents in *Tamarindus indica* L. fruit shell extracts prepared using water, ethanol, chloroform and petroleum ether. Qualitative screening revealed that alkaloids and flavonoids were detected in all four extracts, while the occurrence of coumarins, glycosides, phenols, phlobatannins, quinones, saponins and terpenoids varied according to the solvent used. The aqueous extract showed the presence of seven reported phytochemical groups, including phenols and saponins, whereas the ethanolic extract exhibited the broadest range of detected constituents. Tannins were not detected in any of the extracts under the experimental conditions employed. Furthermore, GC–MS analysis provided a preliminary profile of nine reported chemical constituents, including furan derivatives, a ketone, an alkyne and fatty-acid-related compounds. These findings indicate that the choice of extraction solvent influences the phytochemical profile obtained from tamarind fruit shell. The study contributes to the existing chemical characterization of *Tamarindus indica* L. and highlights the potential value of investigating fruit-processing residues as sources of plant-derived chemical constituents. However, further confirmation of the tentative GC–MS identifications, quantitative analysis and biological evaluation is necessary before drawing conclusions regarding the specific activities or practical applications of the identified compounds.

Conflict of interest

The authors declare that there are no conflicts of interest relevant to this article.

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