

Phytochemical Screening, Ultra Violet and FT- IR Spectroscopy of Ethanolic Extract of *Terminalia Bellirica* Seed

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Abstract: *Terminalia bellirica* is an important medicinal plant. The present work deals with phytochemical screening, UV and FT-IR spectroscopy of ethanolic seed extract of this plant. In phytochemical screening the extract shows the presence of flavonoids, glycosides, phytosterols, terpenoids, phenolic compound, carbohydrates, proteins, tannins, gum and mucilage, alkaloids, saponins, anthoquinone, fixed oil and fats. The UV and FT-IR spectroscopy of ethanolic seed extract of this plant shows the presence of carbonyl group (ketone), α - β unsaturated amide and lactam, aromatic nature of compound, sulfur compound, nitro compound, flavones, fistin, quercetin, NaQSA (Sodium Salts of Quercetin 5' Sulfonic Acid), myricetin, chalcones and anthocyanin types of flavonoids. The above mention bioactive compound are mainly contributed in medicinal utility of the plant.

Keywords: *Terminalia bellirica*, phytochemical screening, ultra violet spectroscopy, FT-IR spectroscopy flavonoids, chromophoric groups

1. INTRODUCTION

Terminalia bellirica (Family : Combretaceae) is widely distributed throughout the world. It is a large deciduous tree found throughout Bangladesh, in hilly areas. The tree takes a height of over 30 meters, while the bark is brownish grey in color [1] (Figure 1). The seeds of *Terminalia bellirica* are drupe, globose or ovoid, slightly 5 ridged, the kernels are sweet, but narcotic, 1.5 to 2.5 cm in diameter, one seeded [2] . They blossom in the month of May[3] (Figure 2). The tree also yields a good-quality firewood and charcoal [4] . In Bangladesh, *T.bellirica* is known as a Bahera is a rejuvenative and laxative[5]. It proves beneficial for hair, throat and eyes [6] .The seeds of *Terminalia bellirica* is used in various eye ailments, such as myopia, corneal opacity, pterigium, and immature cataract, chronic and acute infective conditions[7] .The seeds also helps in loss of appetite, flatulence, thirst, piles and worms. It prevents ageing, imparts longevity, boosts immunity, improves mental faculties and enhances the body resistance against diseases' [5-7] .

The aim of current study was to analysis the ethanolic extract of *Terminalia bellirica* seed by UV& FT-IR along with phytochemical screening to get knowledge about the functional groups present in various secondary metabolities in this important medicinal plant. This will serve the knowledge about the justification of medicinal uses of seeds of this plant.



Fig1. *T. bellirica* tree.



Fig2. *T. bellirica* seeds.

2. MATERIALS AND METHODS

2.1. Collection and Identification of the Plant Sample

Fully matured fresh seeds of *T. bellirica* were Collected from local area of Rajshahi district, Bangladesh in the month of April 2016 and identified by the taxonomist of Bangladesh National Herbarium, Dhaka, where a voucher specimen (No.=4393) has been deposited.

2.2. Plant Materials Preparation

The matured seeds of plant were washed to remove dirt and it was air-dried. Then it was oven-dried at reduced temperature less than 45⁰C to make it suitable for grinding purpose. The screened (20 mesh) powder was then stored in air-tight container with marking for identification and kept in cool, dark, and dry place for future use.

2.3. Solvents and Chemicals

Analytical or laboratory grade solvents and chemicals were used in these experiments. All solvents and reagents used in the experiments were procured from E. Merck (Germany), BDH (England).

2.4. Preparation of Ethanolic Seed Extract

In extraction the powdered seed materials (120 g) is submerged in suitable solvents of increasing polarity as ethanol subsequently in an air-tight separating funnel for 5 days at room temperature with occasionally shaking and stirring. The major portion of the extractable compounds of the plant material will be dissolved in the solvent during this same time and hence extracted as solution. Then these extracts were dried by using a rotary evaporator to get ethanol extract (2.0 g). The extract thus obtained was then subjected to preliminary phytochemical screening for identification of various plant constituents by methods suggested by standard methods[7-11]. To find out the flavonoids, chemical and functional groups of phytochemicals present in the extract, spectral studies were carried out by Ultra-Violet and Infra-Red Spectroscopy[5,12-13,19-21].

3. RESULTS AND DISCUSSIONS

3.1. Phytochemical Screening

The ethanolic extract of *T. bellirica* seed shows the presence of alkaloids, flavonoids, glycosides, phytosterols, terpenoids, phenolic compound, carbohydrates, fixed oil and fats, proteins, tannins, gum and mucilage. The results are presented in Table 1.

Table 1. Phytochemical Screening of ethanolic extract of Terminalia bellirica Seed

Sl.No	Plant constitution test/ Reagents	Results	Sl.No	Plant constitution test/ Reagents	Results
1.	Alkaloids		8.	Types of carbohydrates	
	• Mayer's reagents	+		• Glucose	-
	• Wagner's reagents	+		• Fructose	-
	• Hager's reagents	+		• Galactose	-
2.	Carbohydrates			• Lactose	+
	• Molisch's test	+		• Starch	+
	• Benedict's reagents	+	9.	Glycosides	
	• Fehling solution	+		• Keller killiani test	+
3.	Terpenoids	+	10.	Phytosterols	
	• Salkowski test	+		• Liebermann's test	+
4.	Fixed oil & fats		11.	Saponins	
	• Spot test	+		• Foam test	-
5.	Phenolic compounds		12.	Tannins	+
	Ferric chloride solution	+		Lead acetate solution	
6.	Proteins	+	13.	Amino acids	
	Xanthoprotic test	+		Ninhydrine reagents	+
	Biuret test	+	14.	Flavonoids	
7.	Gums & Mucilages			Con H ₂ SO ₄ + Magnesium ribbon	+
	Alcoholic precipitation	+	15.	Anthraquinones	
	Molisch's test	+		Borntrager's test	-

3.2. UV Spectroscopy

The UV spectrum of ethanolic seed extract of *T. bellirica* were recorded in the ranges of 273-292 nm. The UV spectrum shows weak absorption bands at 292.28 nm is due to aromatic nature of compounds, and aldehydes. These weak band indicates Flavone & Fisetin types of flavonoids. The absorption band at 289.66 nm & 288.80 nm is due to 3° amine, & polyene (β -carotene) which indicates Quercetin. The characteristic broad band at 287.80 nm indicates the presence of amide group (protein). There is a band at 285.60 nm reveals the presence of amino group (Aniline). The characteristic band at 284.20 nm, and 283.42 nm is due to Ketones, aldehydes group. These characteristic band indicates Flavone & Fisetin types of flavonoids. The band at 282.2 nm 281.84 nm and 281.10 nm shows the presence of aldehydes group. The sharp band at 280.26 nm, 278.20 nm and 277.84 nm is due to Ketones group. The band at 274.82 nm and 273.30 nm shows the presence of alkene group.

UV spectroscopy shows the presence of three types of flavonoids viz. flavone, fisetin, quercetin, (Figure 3 & Table 2)

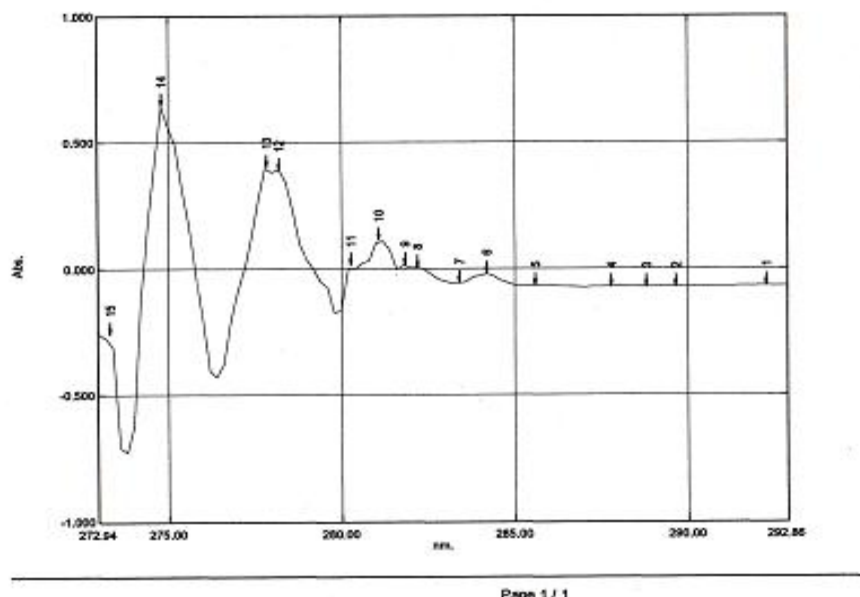


Fig3. UV Spectrum of ethanolic extract of *T. bellirica* seed.

Table2. UV spectroscopy of ethanolic extract of *T. bellirica* seed

Sl. No	Wavelength (nm)	Abs.	Chromophoric group	Types of flavonoids
1.	292.28	0.068	Aldehyde(-CHO)	Flavone &Fistein
2.	289.66	0.070	3°amine, Polyene(β -Carotain.)	Quercetin
3.	288.80	0.071	3°amine, Polyene(β -Carotain.)	Quercetin
4.	287.80	0.070	Amide group (protein).	
5.	285.60	0.066	Amino group (Aniline)	
6.	284.20	0.021	Ketone(=C=O)&aldehyde(CHO)	Flavone &Fistein
7.	283.42	0.055	Ketone(=C=O)&aldehyde(CHO)	Flavone &Fistein
8.	282.20	0.005	Aldehyde(-CHO) group.	Flavone &Fistein
9.	281.84	0.015	Aldehyde(-CHO) group.	Flavone &Fistein
10.	281.10	0.115	Aldehyde(-CHO) group.	Flavone &Fistein
11.	280.26	0.017	Ketones(=C=O) group.	Flavone &Fistein
12.	278.20	0.393	Ketones(=C=O) group.	Flavone &Fistein
13.	277.84	0.402	Ketones(=C=O) group.	Flavone &Fistein
14.	274.82	0.646	Alkenegroup (Naphthalene).	-
15.	273.30	0.255	Alkenegroup.(Naphthalene).	-

3.3. FT-IR Spectroscopy

The FT-IR spectrum of ethanolic extract of *T. bellirica* seed shows the peak at 628.01cm^{-1} , indicates the presence of alkyne, C-H bending vibrations amides and quercetin. The sharp peak at 879.86cm^{-1} is due to aromatic substitution, C-H bending vibrations, and gem disubstituted olefinic group. This peak again confirms the presence of quercetin. The very sharp peak at 1046.93cm^{-1} shows the presence of sulfur compound, S=O stretching vibrations, thiocarbonyl group, sulfoxides and NaQSA [Sodium Salts of Quercetin 5' Sulfonic Acid]. The presence of sulfur compound, thiocarbonyl group and NaQSA [Sodium Salts of Quercetin 5' Sulfonic Acid] further supported by the strong peak at 1087.98cm^{-1} Sulfur compound prominently active against microbes. The FT-IR spectrum shows the peak at 1274.19cm^{-1} indicates the presence of C-N Stretch, & indicates the functional group Aliphatic amine. The peak at 1326.88cm^{-1} indicates aromatic nature of compound, sulphonamides, gem dimethyl group, nitro compound and myricetin type of flavonoids. The characteristic peak at 1381.59cm^{-1} again confirms the presence of C-CH₃ bending, nitro/sulfur compound, gem dimethyl group and myricetin. The FT-IR spectrum shows the peak at 1451.31cm^{-1} & 1658.37cm^{-1} indicates the presence of C-H bend, -C=C- Stretch & indicates the functional group Alkanes.& Alkenes. Appearance of peak at 2893.93cm^{-1} and 2974.87cm^{-1} reveals the presence of C-H stretching vibrations

and aldehydes , & alkanes. There is a clear hump at 3360.49cm^{-1} is corresponding to 1° , 2° amines, Amides and N-H stretching vibrations. FT-IR spectrum of *T. bellirica* reveals the presence of Three type of flavonoids viz. quercetin, NaQSA [Sodium Salts of Quercetin 5' Sulfonic Acid], myricetin. (Figure 4 & Table 3).

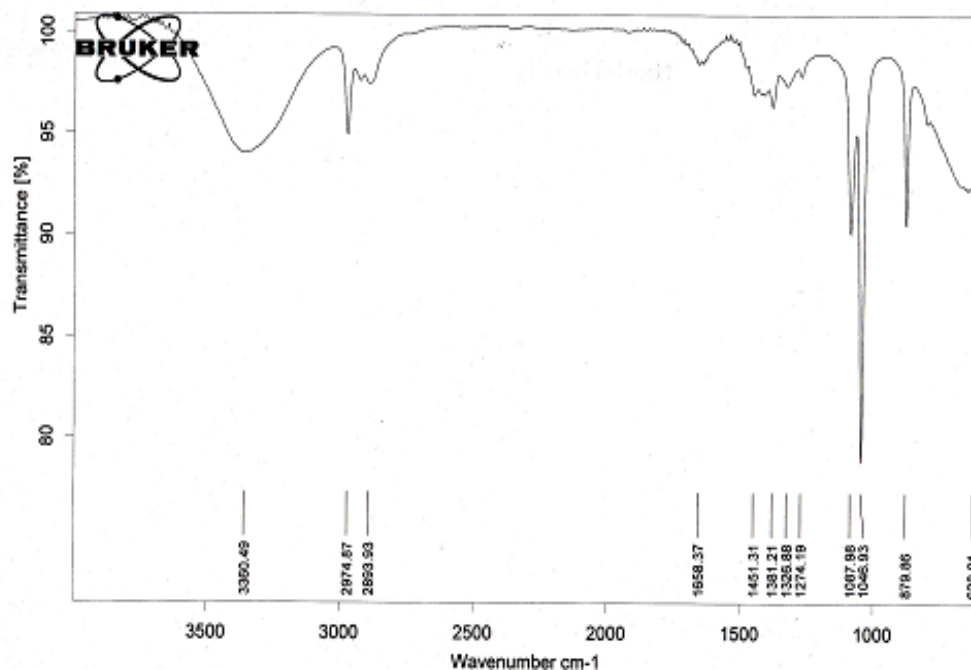


Fig4. FT-IR Spectrum of ethanolic extract of *T. bellirica* seed.

Table3. FT-IR spectroscopy of ethanolic extract of *T. bellirica* seed.

Sl. No.	Peak value (cm^{-1})	bonding type	Functional group	Types of Flavonoids
1.	628.01	C-Hbending	Alkyne	Quercetin
2.	879.86 Sharp	C-H bending vibration	Aromatic substitution, gem-distributed, olefinic group	Quercetin
3.	1046.93	S=O stretching vibration	Sulfur compounds, sulfoxides, Thiocarbonyl group	NaQSA
4.	1087.98	S=O stretching vibration	Sulfur compounds, Thio carbonyl group	NaQSA
5.	1274.19	C-N stretch	Aliphatic amine	
6.	1326.88 Strong	C-N stretch	Aromatic, sulphonamide, gem-dimethyl group & Nitro compounds.	Myricetin
7.	1381.59	C-CH ₃ bending	Nitro/Sulfurcompounds,gem-dimethyl group	Myricetin
8.	1451.31	C-H bend	Alkanes	
9.	1658.37	-C=C- Stretch	Alkenes	
10.	2893.93	C-H stretching vibration	Aldehyde	
11.	2974.87	C-H stretch	Alkanes	
12.	3360.49	N-H stretch	1° , 2° amines, Amides	

4. CONCLUSION

This investigation has gives preliminary information to determine the chemical composition of *T. bellirica* seeds. The presence of chromophoric group, functional group, flavonoids, alkaloids, glycosides, fixed oil and fats, phytosterols, terpenoids, phenolic compound, tannins is mainly contributed in medicinal utility of plant. The presence of these bioactive compounds in plant extract confirms the correct use of this plant in traditional medicinal system. It also holds for the production of novel drugs with isolation of specific compound.

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