The Pharma Innovation Journal 2023; 12(5): 2104-2108

www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277-7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2023; 12(5): 2104-2108

www.thepharmajournal.com
Received: 01-03-2023
Accepted: 10-04-2023

Anurag Semwal
Department of Aquaculture, College of Fisheries, Govind Ballabh Pant University of Agriculture and Technology (GBPUA&T), Pantnagar, Uttarakhand, India

Avdhesh Kumar
Department of Aquaculture, College of Fisheries, Govind Ballabh Pant University of Agriculture and Technology (GBPUA&T), Pantnagar, Uttarakhand, India

Corresponding Author:
Anurag Semwal
Department of Aquaculture, College of Fisheries, Govind Ballabh Pant University of Agriculture and Technology (GBPUA&T), Pantnagar, Uttarakhand, India

Identification of functional groups in *Butea monosperma* methanolic leaf extract through Fourier Transform Infrared Spectroscopy (FT-IR)

Anurag Semwal and Avdhesh Kumar

Abstract
The present study aims to identify the functional groups in *Butea monosperma* methanolic leaf extract through Fourier Transform Infrared Spectroscopy (FT-IR). In the current study, eleven functional groups were identified in *B. monosperma* methanolic leaf extract. The FTIR analysis of methanolic leaf extracts of *B. monosperma* confirmed the presence of alcohol, alkane, aldehydes, α, β-unsaturated ketone, alkyl aryl ether, anhydride, 1,4-disubstituted or 1,2,3,4-tetrasubstituted and halo compound, which show a major peak. Interpretation of FTIR results revealed the presence of myo Inositol, clionasterol and α-Tocopherol. The presence of myo Inositol, clionasterol and α-Tocopherol provides anti-pathogenic properties of the *B. monosperma* plant.

Keywords: *Butea monosperma*, FT-IR analysis, Functional group, Chemical bond, Extract

1. Introduction
Over the years, aquaculture has developed significantly in terms of productivity. However, due to intensive fish farming methods, infectious diseases are the major issue in fish farms resulting in significant economical losses. Many countries have strict laws and regulations on chemicals, antibiotics, hormones and synthetic pharmaceuticals used in aquaculture for the safety of consumers and promoting organic/natural therapeutics to minimize health-related issues. A wide range of medicinal herbs/plants exhibits immunostimulant, antimicrobial and growth-promoting properties. Numerous studies have been conducted on the benefits of phytochemicals and their potential role as feed additives in the aquaculture industry (Semwal *et al.*, 2022) [1].

In India, "Mrgayurveda," a subdiscipline of Ayurveda, focuses on animal life and the use of herbal medicines to treat animal diseases (Chakraborty and Hancz, 2011) [2]. Phytotherapy is a medical practice that focuses more on traditional approaches rather than modern medication. This biodegradable and environmental-friendly application is known as phytotherapy or more often commonly called herbalism (Semwal *et al.*, 2023) [3]. Medicinal plants are the major component of folk medicine formulations. These plants are used directly in the form of decoctions, juices or pastes and delivered either orally or topically (Ahmed *et al.*, 2012; Kumar *et al.*, 2019; Kumar *et al.*, 2022; Kumar *et al.*, 2021; Sharma *et al.*, 2019; Sharma *et al.*, 2022; Kumar *et al.*, 2022) [4, 5, 6, 7, 8, 9, 10]. Fourier transforms infrared spectroscopy (FT-IR) is a high-spectral resolution analytical technique to recognize the chemical bond, functional group and structure of compounds (Shah *et al.*, 2019) [11].

Butea monosperma commonly known as "Flame of forest" and bastard teak is one of several medicinal plants used from prehistoric times for a variety of diseases. *B. monosperma* is a member of the Fabaceae family, this deciduous tree is a medium-sized one (12 to 15 metres tall). Since many years ago, almost all of the components of *B. monosperma* have been used in medicine and other fields. It is considered a good source of gum, resin, food, fibre, dye and traditionally being used for the treatment of stomach disorders, asymmetrical menstrual flow, colds, coughs and sore throats. The plant has great promise as an antidote and in the treatment of scorpion sting and snake bite symptoms (Burli and Khade, 2007) [12]. The other genus of *Butea* includes *Butea frondosa*, *Butea parviflora and Butea superba* widely distributed throughout India (Khare, 2007) [13]. The aim of the present study is the identification of a functional group in the *B. monosperma* methanolic leaf extract by FT-IR analysis.

2. Materials and Methods

2.1 Collection of *Butea monosperma* plant leafs

The fresh leafs of *B. monosperma* were collected from the Medicinal Plants Research and Development Centre (MRDC), Haldi, Pantnagar, Udham Singh Nagar, Uttarakhand and authentication was done by Dr. D. S. Rawat Head, Department of Biological Science, Govind Ballabh Pant University of Agriculture and Technology, Pantnagar. The leaves were properly cleaned with tap water two or three times before being soaked in distilled water for 30 minutes to remove the dust. They then underwent a seven-day process of drying and grinding in the shade (Fig 1 & 2).

Fig 1: Dried leafs of *Butea monosperma* after seven days of drying.

Fig 2: Powder form of *Butea monosperma* leafs after grinding.

2.2 Preparation of methanolic extract of *B. monosperma* leafs

To provide the plant extract, after air drying in the shade, dry leaves (100 g) were grinded using an electric grinder (Pars Khazar brand) then methanol (85%, 1 Litre) was added. For a proper mixing of the sample, the methanol and leaf powder mixture was shaken for 48 hours (Fig 3). The mixture was then filtered using Whatman filter paper No. 1 before the remaining supernatant was centrifuged at 2460 rpm for 10 minutes. The supernatant was kept at 4 °C till further use. After that, the supernatant was then evaporated at 40 °C in the rotary evaporator, so that all the supernatant was evaporated. The remaining part in the rotary evaporator was an extract of *B. monosperma*. Collect the extract and kept at -20 °C till further use (Farsani *et al.*, 2019) [14] (Fig 4).

Fig 3: Electric shaker used to properly mix *Butea monosperma* leaf powder with methanol.

Fig 4: Methanolic leaf extract of *Butea monosperma*.

2.3 FT-IR spectrum analysis of *B. monosperma* methanolic leaf extract

The Fourier Transform Infrared Spectrophotometer (FT-IR) is likely the most effective spectrometer for identifying the different functional groups or chemical bonds that are present in the photochemical. The feature of the chemical bond visible in the annotated spectrum is the wavelength of light absorbed. By analysing the infrared absorption spectrum, the chemical bonds or functional groups can be identified. FT-IR analysis was performed using a methanolic leaf extract of *B. monosperma*. The dried extract powder was dissolved in potassium bromide in an amount of 5 mg (KBr) (Fig 5). The mixture of leaf extract and KBr was thoroughly combined in a mortar before being compressed at 6 bars for two minutes to form a thin disc of KBr. The disc was then placed in a sample cup. The leaf extracts of *B. monosperma* were loaded into a Pekin Elmer infrared spectroscope with a frequency range of 400 to 4000 cm^{-1} (Shah *et al.*, 2019) [11].

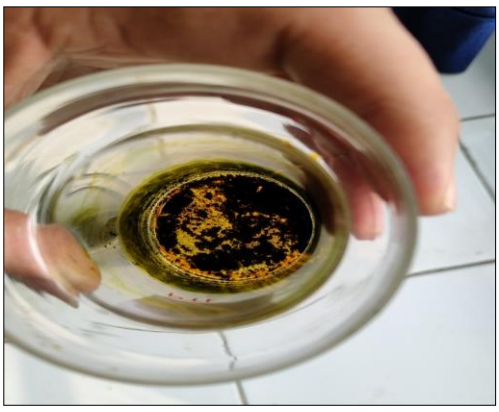
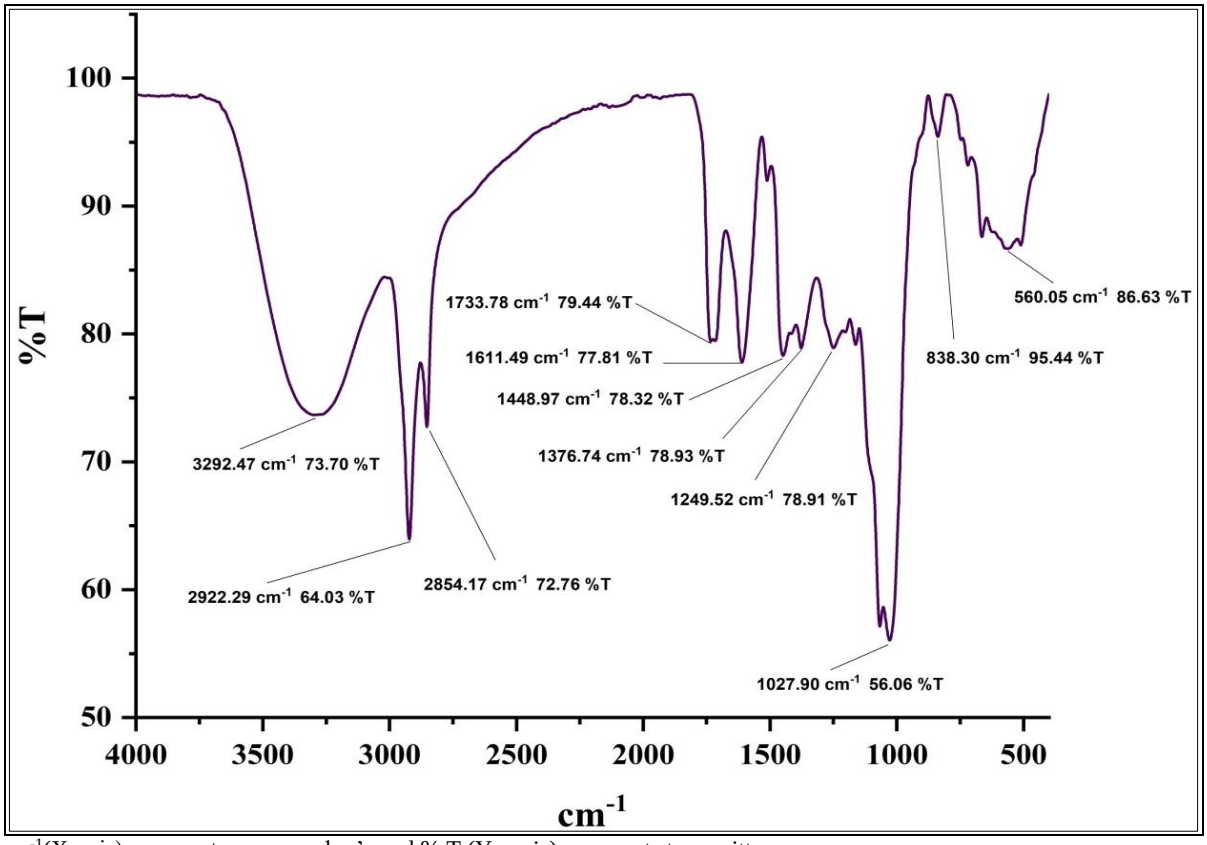


Fig 5: *Butea monosperma* leafs dried extract powder for FT-IR analysis.

3.1 Results

The *B. monosperma* leaf methanolic extract was exposed to the FTIR analysis and showed peak at different wavenumber's (Fig 6). The present results show the presence of 11 functional groups and chemical bonds recognized from the *B. monosperma* leaf extract in (Table 1). The FTIR analysis of methanol leaf extracts of *B. monosperma* confirmed the presence of alcohol, alkane, aldehydes, α,β-unsaturated ketone, alkyl aryl ether, anhydride, 1,4-disubstituted or 1,2,3,4-tetrasubstituted and halo compound, which show a major peak. The FTIR analysis of leaf extract gives a strong broad peak obtained at 3292.47 cm^{-1}, which indicates the presence of alcohol (O-H stretching). The medium instance peak was obtained at 2922.29 cm^{-1}, which indicates the presence of alkane (C-H stretching). The medium peak was recognized at 2854.17 cm^{-1}, which is assigned to the alkane (C-H stretching). The strong peak was obtained at 1733.78 cm^{-1}, which indicates the presence of aldehydes (C=O stretching). The strong peak was identified at 1611.49 cm^{-1}, which is assigned to α,β-unsaturated ketone (C=C stretching). The medium peak was obtained at 1448.97 cm^{-1}, which indicates the presence of alkane (C-H stretching).



cm^{-1}(X-axis) represents wavenumber's and % T (Y –axis) represents transmittance

Fig 6: FTIR Spectrum analysis of *Butea monosperma* leaf extract.

The medium peak was obtained at 1376.74 cm^{-1}, which indicates the presence of alcohol (O-H stretching). The strong peak was obtained at 1249.52 cm^{-1}, which indicates the presence of alkyl aryl ether (C-O stretching). The strong peak was obtained at 1027.90 cm^{-1}, which indicates the presence of anhydride (CO-O-CO stretching). The strong peak was recognized at 838.30 cm^{-1}, which indicates the presence of 1,4-disubstituted or 1,2,3,4-tetrasubstituted (C-H bending). The strong peak was recognized at 560.05 cm^{-1}, which indicates the presence of a halo compound (C-Br stretching).

Table 1: FTIR spectral wavenumber's values, absorption range, chemical bonds and functional groups obtained from the *B. monosperma* methanolic leaf extract.

Wavenumber's cm^{-1} (test sample)	Absorbtion range cm^{-1} (Reference number)	Chemical bond	Functional group
3292.47	3550-3200	O-H stretching	Alcohol
2922.29	3000-2840	C-H stretching	Alkane
2854.17	3000-2840	C-H stretching	Alkane
1733.78	1740-1720	C=O stretching	Aldehyde
1611.49	1620-1610	C=C stretching	α,β-unsaturated ketone
1448.97	1450	C-H bending	Alkane
1376.74	1420-1330	O-H bending	Alcohol
1249.52	1275-1200	C-O stretching	Alkyl aryl ether
1027.90	1050-1040	CO-O-CO stretching	Anhydride
838.30	810 ± 20	C-H bending	1,4-disubstituted Or 1,2,3,4-tetrasubstituted
560.05	690-515	C-Br stretching	halo compound

4. Discussion

The present study identified the functional group and chemical bond in *B. monosperma* leaf extract through Fourier Transform Infrared Spectroscopy (FT-IR). To recognize the chemical constituents, functional groups and elucidate the chemical structure as bioactive constituents for the treatment of various diseases. Jain *et al.,* (2016) [15] reported the identification of functional groups in *Mentha spicata* plant extract by FT-IR and reported that the alcohol group present in the extract. Similar results were observed by Pharmawati and Wrasiati, (2020) [16] who reported that the functional groups of alkanes are responsible for the medicinal properties of *Enhalus acoroides* chloroform and ethanol extract. Research studies by Kamble and Gaikwad, (2016) [17] depict that the aldehydes and ketones were present in the extract of *Embelia ribes* leaves. The presence of C-O bonding structures was responsible for the presence of the ether group. Wang *et al.,* (2017) [18] reported that 1,2,3,4-tetrasubstituted is a anisucoumaramide, a bioactive coumarin from *Clausena anisumolens.* Dhivya and Kalaichelvi, (2017) [19] reported that the anhydrides and halo compounds present in the extract of *Micrococca mercurialis* (L.) Benth.

5. Conclusion

Interpretation of FTIR results revealed the presence of myo Inositol, clionasterol and α-Tocopherol. The presence of myo Inositol, clionasterol and α-Tocopherol provides anti-pathogenic properties of the *B. monosperma* plant. The interest in using plants as functional feed additives in aquaculture has increased tremendously over the past year and is likely to continue growing as international antimicrobial usage rules tighten and antimicrobial resistance is acknowledged as a global health issue. The pre-clinical and toxicological study is advised to formulate the best administration dosages in aquaculture. Moreover, research into their mode of action, plant component stability in aquatic conditions, digestibility and in vivo and in vitro toxicity testing are required for safe use.

6. Acknowledgements

The authors are thankful to the Central Instrumentation Facility of Lovely Professional University and the Dean, College of Fisheries, Pantnagar for providing the necessary facilities for the study.

7. References

1. Semwal A, Khati A, Kumar A, Yadav MK, Arya D. A review on antimicrobial peptides (AMPs). *Pharma innov*ation. 2022;11(4):1139-1146.
2. Chakraborty SB, Hancz C. Application of phytochemicals as immunostimulant, antipathogenic and antistress agents in finfish culture. Reviews in Aquaculture. 2011;3(3):103-119. https://doi.org/10.1111/j.1753-5131.2011.01048.x
3. Semwal A, Kumar A, Kumar N. A review on pathogenicity of *Aeromonas hydrophila* and their mitigation through medicinal herbs in aquaculture. Heliyon. 2023;9:e14088. DOI: https://doi.org/10.1016/j.heliyon.2023.e14088
4. Ahmed F, Siddaraju NS, Harish M, Urooj A. Effect of *Butea monosperma* Lam. leaves and bark extracts on blood glucose in streptozotocin-induced severely diabetic rats. Pharmacognosy Research. 2012;4(1):33-36. https://doi.org/10.4103/0974-8490.91032
5. Kumar N, Sharma JG, Singh SP, Singh A, Harikrishna V, Chakrabarti R. Validation of growth enhancing, immunostimulatory and disease resistance properties of *Achyranthes aspera* in *Labeo rohita* fry in pond conditions. Heliyon. 2019;5(2):e01246. https://doi.org/10.1016/j.heliyon.2019.e01246
6. Kumar N, Sharma JG, Mittal P, Chakrabarti R. Effect of leaves and seeds of *Achyranthes aspera* as feed supplements on the immunological and stress parameters and related-gene expressions of Asian catfish (*Clarias batrachus*). Veterinary Research Communications. 2022;47:99-109. https://doi.org/10.1007/s11259-022-09932-5
7. Kumar N, Sharma JG, Kumar G, Shrivastav AK, Tiwari N, Begam A, *et al*. Evaluation of nutritional value of prickly chaff flower (*Achyranthes aspera)* as fish feed ingredient. Indian Journal of Animal Sciences. 2021;91(3):239-244. https://doi.org/10.56093/ijans.v91i3.114152
8. Sharma JG, Kumar N, Singh SP, Singh A, Harikrishna V, Chakrabarti R. Evaluation of immunostimulatory properties of prickly chaff flower *Achyranthes aspera* in rohu *Labeo rohita* fry in pond conditions. Aquaculture. 2019;505:183-189. https://doi.org/10.1016/j.aquaculture.2019.02.065
9. Sharma JG, Kumar N, Mittal P, Chakrabarti R. Evaluation of UV-B protective properties of leaves and seeds of *Achyranthes aspera* in Asian catfish, *Clarias batrachus* (Linn.). Photochemical & Photobiological Sciences. 2022;21:1341-1356. https://doi.org/10.1007/s43630-022-00222-2
10. Kumar G, Sharma JG, Goswami RK, Shrivastav AK, Tocher DR, Kumar N, *et al*. Freshwater macrophytes: Potential source of minerals and fatty acids for fish, poultry and livestock. Frontiers in Nutrition. 2022;9:869425. https://doi.org/10.3389/fnut.2022.869425
11. Shah TK, Kumar A, Tandel R, Bhat RAH, Sarma D. Identification of functional groups in *Thymus linearis* ethanol leaf extract through fourier transform infrared spectroscopy (FTIR). Journal of Pharmacognosy and Phytochemistry. 2019;8(4):3013-3015.
12. Burli DA, Khade AB. A comprehensive review on *Butea monosperma* (Lam.) Kuntze. Pharmacognosy Reviews. 2007;1(2):333-337.
13. Khare CP. Indian Medicinal Plants: An Illustrated Dictionary. Springer, New York, 2007.
14. Farsani MN, Hoseinifar SH, Rashidian G, Farsani HG, Ashouri G, Van Doan H. Dietary effects of *Coriandrum sativum* extract on growth performance, physiological and innate immune responses and resistance of rainbow trout (*Oncorhynchus mykiss*) against *Yersinia ruckeri*. Fish & shellfish immunology. 2019;91:233-240.
15. Jain PK, Soni A, Jain P, Bhawsar J. Phytochemical analysis of *Mentha spicata* plant extract using UV-VIS, FTIR and GC/MS technique. Journal of Chemical and Pharmaceutical Research. 2016;8(2):1-6.
16. Pharmawati M, Wrasiati LP. Phytochemical screening and FTIR spectroscopy on crude extract from *Enhalus acoroides* leaves. Malaysian journal of analytical sciences. 2020;24(1):70-77.
17. Kamble V, Gaikwad N. Fourier Transform infrared spectroscopy spectroscopic studies in *Embelia ribes*

burm. F.: a vulnerable medicinal plant. Asian Journal of Pharmaceutical and Clinical Research. 2016;9(3);41-47.
18. Wang YS, Li BT, Liu SX, Wen ZQ, Yang JH, Zhang HB, *et al*. Anisucoumaramide, a Bioactive Coumarin from Clausena anisum-olens. Journal of Natural Products. 2017;80(4):798-804.
19. Dhivya K, Kalaichelvi K. Screening of phytoconstituents, UV-VIS Spectrum and FTIR analysis of *Micrococca mercurialis* (L.) Benth. International Journal of Herbal Medicine. 2017;5(6):40-44.