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Possible Functional Groups in Different Leaf Extracts of the Potential Ethnomedicinal Plant Aristolochia Assamica from Eastern Himalayan Region: An Analysis through FTIR

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Abstract: Fourier Transform Infrared Spectroscopy (FTIR) is an emerging spectroscopic tool to detect and analysis of the functional groups of a chemical compound. Infrared rays are approached in this technique to vibrate the molecules at specific frequency point against their absorbance. Medicinal plants are giant repositories of secondary metabolites famous for pharmacological activities. Aristolochia Assamica D. Borah & T. V. Do, sp. nova is a notable ethnomedicinal plant from the North Eastern region of India of which different leaf extracts were endeavoured through FTIR analysis in the present study. Among the four distinct extracts based on polarity, aqueous and methanol extract yielded highest percentage of extraction. All the extracts possessed similarities in the frequency ranges absorbed by the phytoconstituents resulting the possible existence of alcohol, alkanes, alkenes, alkynes, aldehyde, aromatics, amines, nitro compound, ethers, halogen, sulphur, phosphorous, and fluorine compoundswhich may pave the wayfor further pharmacological researches.

Keywords: Aristolochia, Extract, FTIR, Functional groups, Phytoconstituent

Introduction:

Wide array of phytocompounds including the phenolics, terpenoids, tannins, flavonoids, saponin etc. secondary metabolites embracing the medicinal plants providing a framework against diseases. The curative significance of these phytocompounds or acting as natural antioxidants, antimicrobial, anti-tumour agents earned global fame over synthetic drugs (Sharifi-Rad et al. 2019). The phytoconstituents act through modulating pathways involved in disease progression (Paul et al. 2024). Molecular characterization of these phytocompounds hence become apparent in medicinal plant research.

Insights into the structure of the phytochemical compounds can be interpreted through Fourier Transform Infrared Spectroscopy (FTIR) analysis. FTIR is a significant and prevalent spectroscopic technique to be used not only for qualitative characterization of compounds but also influences in standardization and authentication of the plants medicinal value and quality of crude extracts (He et al. 2024). Since, different molecules possess

uniqueness in vibration pattern after inducing radiation and absorbs infrared rays at attributed frequency ranges. These absorption frequencies correlates to certain functional groups generally present in the phytochemical compounds (Altemimiet al. 2017).

Aristolochia L. is a widespread genus of Aristolochiaceae family including overall554 accepted species (POWO). The species of Aristolochia has been utilized for traditional healthcare practices in Asian and European countries for centuries. Besides the renowned phytocompounds Aristolochic acids and Aristolactams isolated from the genus Aristolochia; it is a dwelling place to numerous bioactive compounds which may responsible for its pharmacological properties such as antioxidant, antiprotozoal, antimicrobial, antihistaminic, anti-inflammatory, anti-cancer, antidiabetic, anti-fertility, anti-venom, anticholinergic, anti-pruritic etc. activities(Borah et al. 2021, Guillen et al. 2025). Aristolochiaassamica D. Borah & T.V.Do, is stated to be a novel species occurred throughout the state of Assam and Arunachal Pradesh of North East India. It is a climber with cordate leaves as shown in Plate 1, stem slender with purplish younger branches, flowers appear in cymes, often creamy and tubular. Ethnomedicinal study conducted for this species uncovered the use of leaves, stem, roots, and rhizome in multitudinous therapeutic uses like juice of leaves are used in treating fever, pneumonia, stem juice in diabetes and high blood pressure, dried roots, or rhizome extract in stomach disorders by ethnic people of Assam. GC-MS analysis indicated the presence of several phytoconstituents comprising Acetic acid, 2-(2-t-butyl-4-methyl-6-oxo [1,3] dioxan-4-yl)-1-phenyl-ethyl ester, 2-Octylcyclopropene-1-heptanol, 11-Methyldodecanol, 6,10,14,18,22-Tetracosapentaen-2-ol,3-bromo-2,6,10,15,19,23-hexamethyl-, (all-E)-, cis-Z-.alpha.- Bisabolene epoxide, Levomenthol, Eudesma-5,11(13)-dien-8,12-olide,Cyclohexanol, 5-methyl-2-(1-methylethyl)-, [1S-(1alpha,2alpha,5beta)]-,3-Hexadecyne, alpha-Ylangene, 4-Tetradecyne, Neoisolongifolene, 8-bromo-, 1-Undecyne, Benzenepropanoic acid, 3,5-bis(1,1-dimethylethyl)-4-hydroxy-, methyl ester, etc. (Borah & Sarma, 2022). FTIR analysis in this study helps in the validation and verification of the functional groups of known and unknown phytochemical compounds present in the plant.

Plate 1: Habit of A. assamica

Materials and Methods:

Fresh leaves of A. assamica were collected from Biswanath district, Assam, India in August, 2021 after taking proper permission from the concerned officials. It was identified by Dr. Dipankar Borah by the holotype, taxonomistand author of these new species currently working as an assistant professor in Kaliabor College, Assam. Herbarium was deposited to the Botanical Survey of India, Eastern Regional Centre, Shillong with accession no. 98880 for future reference. The leaves were then washed and dried under shade for two weeks. Dried leaves were then grinded and coarse powders were obtained (Plate 2). Extraction of the powdered materials (10g) were performed using different polar solvents; water, methanol, acetone, and petroleum ether through Soxhlet apparatus. The yield percentage of each extract was determined using the following formula (Uchenna et al. 2015):
Percentage yield of extract (%) = (Weight of the dry extract in g/Weight of the powdered material) × 100

FTIR analysis were performed using PerkinElmer Spectrometer. Each extract of A. assamica were prepared using KBr pellets for FTIR analysis to obtain exact absorbance. Absorption was taken at 450-4000 cm^{-1} infrared radiation. Standard FTIR frequency ranges were considered to analysis the obtained functional groups based on the stretching and bending patterns of the molecules.

Results and Discussion:

Extraction yield: The extractive yield of A. assamica leaves through different solvents showed polarity dependent outcomes. The aqueous and methanol extracts attained the highest and same percentage (28.2%), acetone extract yielded 15.4% and the least yield was noted for petroleum ether extract (3%).Solvents with higher polarity yield more extraction by dissolving the polar molecules than the non-polar one (Tripathi et al. 2025). In this study, water and methanol being the polar solvents resulted greater extraction

which is also line with the previous studies conducted by Chigayoet al. 2016 for ethno medicinal plant Kirkia wilmsii extracts.

Plate 2: Coarse Powder of A. Assamica dried leaves

FTIR analysis: The infrared spectra exhibited different peaks of certain range for each extract may possess different chemical compounds though similarity in the ranges were observed.

Aqueous extract: The spectrums depicted the presence of phytoconstituents respective to absorption peaks corresponding to diverse functional groups from the region 3260.88 cm^{-1} to 510.98 cm^{-1} (Figure3). As designed in the Table 1, the first peak observed at 3260.88 cm^{-1} is a strong indication to the presence of -OH and -NH_2may abundant in phenolic compounds, flavonoids, tannins, and alkaloids, known for their free radical scavenging, anti-inflammatory, and antimicrobial activities (Yahia et al. 2019). The peak number two ascribed to C=N stretching which is key structural features to alkaloids often associated with antioxidant and enzyme inhibition activities (Oliveira et al. 2016).The band at 1388.06 cm^{-1} is indicative of NO_2 symmetric stretching and methyl bending, suggesting to the possible presence of nitro compounds, alkanes, and alkenes featuredto alkaloids, terpenoids, and phenolic acids. The fourth peak at 1033.38 cm^{-1} portrayed the occurrence of alcohols, ethers, and phosphorous or sulphur containing compounds. These types of bonds are often found in glycosides, flavonoid derivatives, and organophosphates which play crucial role in pharmacological activities (Ullah et al. 2020). The presence of halogenated phytochemicals at absorption band at 510.98 cm^{-1} can influence pharmacological properties such as lipophilicity and antibacterial activity (Faleyeet al. 2024).

Table 1. Possible functional groups in A. Assamica aqueous extract

Peak Number	X (cm^{-1})	Y (%T)	Functional Group	Type of Vibration	Possible Compound
1	3260.88	62.30	-OH, -NH, ≡C-H	O-H stretching, N-H stretching	Alcohol, Amines, Alkynes, Aldehyde
2	1582.46	42.36	C=C, C=N, NH	C=C stretching, N-H bending	Unsaturated aliphatic, Aromatics, Amines, Amino Acid
3	1388.06	46.91	NO_2, CH_3, CH_2	NO_2 symmetric stretching, -CH_3 or -CH_2 bending	Nitro compound, Alkanes, Alkenes
4	1033.38	42.65	C-O-C, C-OH, S=O, P=O, C-F	C-O, P=O, S=O, and C-F stretching	Ethers, Alcohol, Sulphur, Phosphorous and Fluorine compounds
5	510.98	34.27	C-halogen, Aromatic rings	C-X (halogen) stretching	Aromatic compounds, Halogen compounds

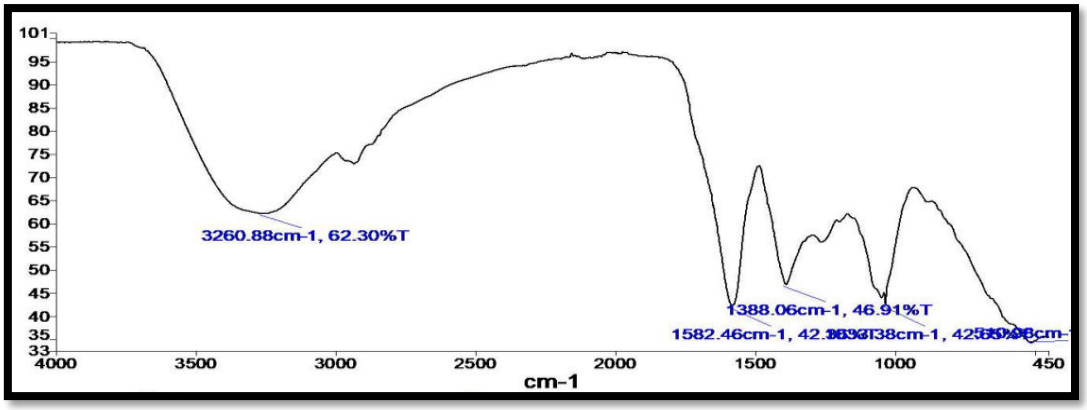

Figure 3. FTIR graph of A. Assamica aqueous extract

Methanol extract: Alike the aqueous extract a broad peak at 3360.74 cm^{-1} assigned the hydroxyl functional group as shown in the Table 2 and Figure 4, which strongly suggests the presence of phenolic compounds such as flavonoids, tannins, and phenolic acids. The occurrence of terpenoids or fatty acid derivatives, components of essential oils possess antioxidant, anti-inflammatory, antimicrobial, and anticancer properties depicted by the third and fourth peaks correlated to methyl groups (Mohammed et al. 2024). The band at 1593.69 cm^{-1} may be assigned to unsaturated aliphatic and amines, characteristic of flavonoids and tannins, reported to has neuroprotective, antidiabetic, cardiovascular protectivity (Pizzi 2021). Nitrogen-containing secondary metabolites like alkaloids or terpenoids which perform analgesic, antibacterial, anti-tumour, anti-malarial, anticancer, insecticidal etc. properties may be indicated through the band observed at 1375.58 cm^{-1} may

result from bending vibrations of CH_2 or CH_3 groups and NO_2 symmetric stretching (Wang et al. 2024).

Table 2. Possible functional groups in A. Assamica methanol extract

Peak Number	X (cm^{-1})	Y (%T)	Functional Group	Type of Vibration	Possible Compound
1	3360.74	83.29	-OH, -NH, ≡C-H	O-H, N-H, ≡C-H stretching	Alcohol, Amines, Alkynes, Aldehyde
2	2919.25	71.02	-CH, $-CH_2-$, $-CH_3$	-CH stretching	Aliphatic groups
3	2850.13	78.74	-CHO, $-CH_2-$, $-CH_3$	-CH stretching	Aldehydes, Aliphatic groups
4	1593.69	78.85	C=C, C=N, NH	C=C, C=N stretching, N-H bending	Unsaturated aliphatic, Aromatics, Amines
5	1375.58	68.13	NO_2, CH_3, CH_2	CH_3 bending, NO_2 stretching	Nitro compound, Alkanes, Alkenes
6	1234.52	71.58	C-O-C, C-OH, S=O, P=O, C-F	C-O, P=O, S=O, Si-O stretching	Ethers, Alcohol, Sulphur, Phosphorous and Fluorine compounds
7	1126.63	66.11	C-O-C, C-OH, S=O, P=O, C-F	C-O, P=O, S=O, Si-O stretching	Ethers, Alcohol, Sulphur, Phosphorous and Fluorine compounds
8	1033.31	58.22	Si-O and P-O	C-O, P=O, S=O, Si-O stretching	Phosphorous compounds
9	889.36	74.09	=C-H, -NH	=C-H out of plane bending	Alkenes and Aromatic compounds, Aliphatic compounds

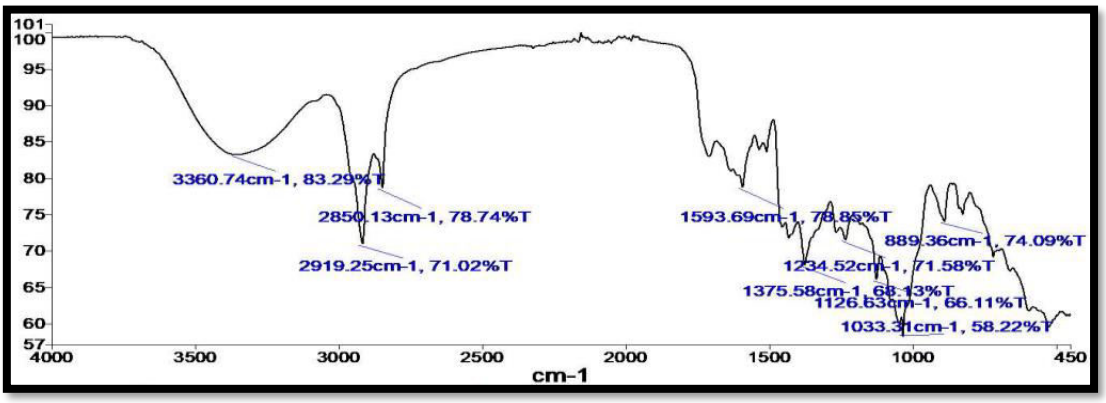

Figure 4. FTIR graph of A. Assamica methanol extract

Acetone extract: The spectrum of acetone extract as presented in the Table 3 and Figure 5 showed broad peak at 3363.65 cm^{-1} assigned for alcohol, amines, alkynes, and aldehydes pertaining to phenolic compounds, such as flavonoids, tannins, and alkaloids. Other strong absorption bands at 2919.08 cm^{-1} and 2850.13 cm^{-1} correspond to asymmetric and symmetric stretching of aliphatic -CH_2 and -CH_3 groups, which are associated with terpenoids of antioxidant and antimicrobial properties (Siddiqui et al. 2024). A prominent peak at 1707.24 cm^{-1} is characteristic of carbonyl stretching, indicating the presence of aldehydes, ketones, carboxylic acids, esters, or even quinones, which are often biologically active components in flavonoids, coumarins, or essential oils. Nitrogen containing secondary metabolites may be suggesting for the peak at 1375.24 cm^{-1} associated with symmetric NO_2 stretching. Bands at 1233.75 cm^{-1} and 1125.98 cm^{-1} represent sulphur, phosphorous and fluorine compounds, which are indicative of organosulfur and organo phosphorus phytoconstituents known for acetyl cholinesterase inhibition property (Ore et al. 2023).

Table 3 Possible functional groups in A. Assamica acetone extract

Peak Number	X (cm^{-1})	Y (%T)	Functional Group	Type of Vibration	Possible Compound
1	3363.65	83.87	-OH, -NH, ≡C-H	O-H, N-H, ≡C-H stretching	Alcohol, Amines, Alkynes, Aldehyde
2	2919.08	75.06	-CH, -CH_2-, -CH_3	C-H stretching	Aliphatic groups
3	1707.24	79.95	C=O	C=O stretching	Acid halides, aldehydes, carboxylic acids, ketones, quinines
4	1453.03	73.82	NO_2, CH_3, CH_2	-NO_2, CH_3 or CH_2 bending	Nitro compound, Alkanes, Alkenes
5	1375.24	72.73	NO_2, CH_3, CH_2	-NO_2, CH_3 or CH_2 bending	Nitro compound, Alkanes, Alkenes

6	1233.75	71.68	C-O-C, C-OH, S=O, P=O, C-F	C-O, P=O, S=O stretching	Ethers, Alcohol, Sulphur, Phosphorous and Fluorine compounds
7	1125.98	62.94	C-O-C, C-OH, S=O, P=O, C-F	C-O, P=O, S=O stretching	Ethers, Alcohol, Sulphur, Phosphorous and Fluorine compounds
8	1033.11	60.97	Si-O and P-O	Si-O, P-O stretching	Organosilicon, Phosphorous compounds
9	899.82	69.59	=C-H, -NH	=C-H, -NH bending	Alkenes and Aromatic compounds, Aliphatic compounds

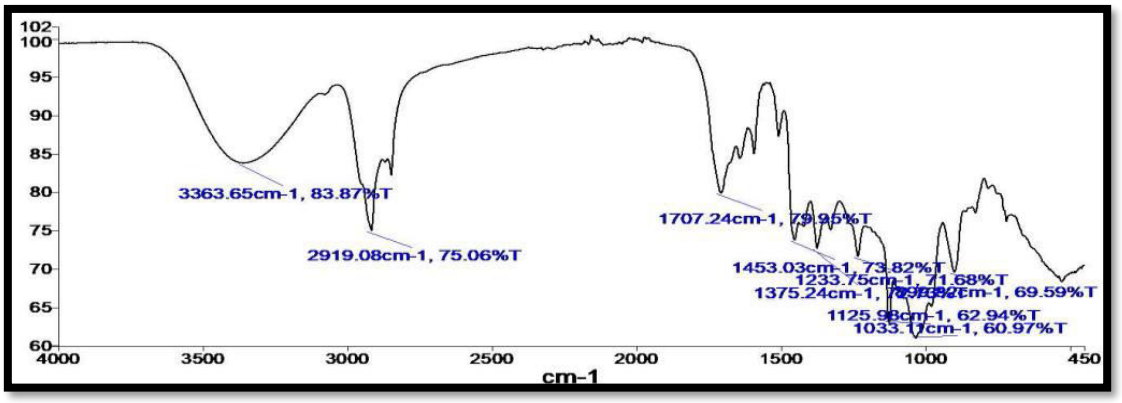

Figure 5 FTIR graph of A. Assamica acetone extract

Petroleum ether extract: A total of ten absorption peaks (3331.43-839.02 cm^{-1}) were counted for the petroleum ether extract represented in the Table 4 and Figure 6. The first peak denoted the -OH and $-NH_3$ groups, indicative of phenolic compounds such as flavonoids, tannins, and alkaloids potent to antiviral, anti-inflammatory, anticancer and antibacterial effects (Rahman et al. 2022). Absorption peaks at 2923.73 cm^{-1} and 2853.55 cm^{-1} correspond to asymmetric and symmetric stretching of $-CH_3$ and $-CH_2$ groups, also suggesting the presence of aliphatic chains found in bioactive terpenoids. A strong band at 1740.20 cm^{-1} represents C=O stretching, characteristic of carbonyl compounds such as carboxylic acids, esters, and acid halides, often seen in coumarins revealed to have anti-inflammatory, anti-coagulative, analgesic, antimicrobial, anti-HIV etc. properties (Wu et al. 2009).

Table 4 Possible functional groups in A. Assamica petroleum ether extract

Peak Number	X (cm^{-1})	Y (%T)	Functional Group	Type of Vibration	Possible Compound
1	3331.43	88.46	-OH, -NH, ≡C-H	O-H, N-H, ≡C-H stretching	Alcohol, Amines, Alkynes, Aldehyde
2	2923.73	67.90	-CH, $-CH_2-$, $-CH_3$	C-H stretching	Aliphatic groups
3	2853.55	77.35	-CHO, -CH2-, -CH3	C-H stretching	Aldehydes, Aliphatic groups
4	1740.20	82.05	C=O	C=O stretching	Acid halides, Carboxylic acids, Esters
5	1536.31	81.92	NO_2, CH_3, CH_2	NO_2 stretching, CH_3 or CH_2 bending	Nitro compound, Alkanes, Alkenes
6	1432.51	73.89	C-O-C, C-OH, S=O, P=O, C-F	C-O, S=O, P=O stretching	Ethers, Alcohol, Sulphur, Phosphorous and Fluorine compounds
7	1376.67	71.85	NO_2, CH_3, CH_2	NO_2 stretching (symmetric), CH_3 or CH_2 bending	Nitro compound, Alkanes, Alkenes
8	1033.37	66.93	Si-O and P-O	Si-O, P-O stretching	Phosphorous compounds, Organosilicon
9	889.05	76.54	=C-H, -NH	=C-H, -NH bending	Alkenes and Aromatic compounds, Aliphatic compounds
10	839.02	79.32	C-halogen	C-X stretching	Aromatic compounds

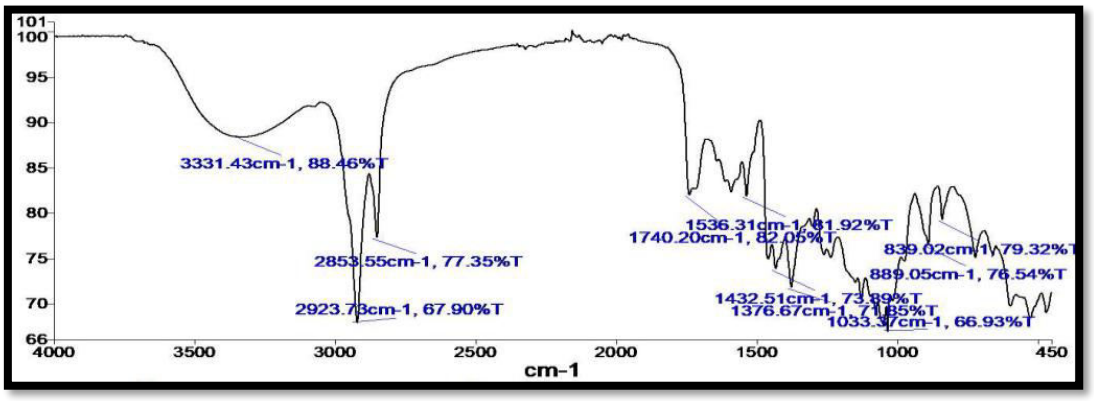

Figure 6. FTIR graph of A. Assamica petroleum ether extract

Conclusion:

The FTIR spectrum of four different extracts of A. Assamica revealed prominent absorption peaks corresponding to several functional groups. These vibrations indicate the presence of diverse classes of secondary metabolites such as phenols, flavonoids, alkaloids, and other bioactive phytoconstituents of biological activities. Furthermore, the structural diversity revealed by the FTIR spectrum emphasize the potentialities of this plant as a source of therapeutic agents for applications in nutraceuticals, phytopharmaceutical formulations, and new days pharmacological inventions.

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Conflict of interest: The author declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

References:

1. Altemimi, A., Lakhssassi, N., Baharlouei, A., Watson, D. G., and Lightfoot, D. A. (2017).Plants (Basel). Phytochemicals: extraction, isolation, and identification of bioactive compounds from plant extracts. Volume 6 Number 4.
2. Borah, P. J., Borah, D., Das, U., Das, T. J., and Sarma, R. (2021). Notulae Scientia Biologicae. A review on ethno pharmacological utility, traditional knowledge and phytochemistry of Aristolochia species in Assam, India. Volume13Number3.
3. Chigayo, K., Mojapelo, P. E. L., Mnyakeni-Moleele, S., and Misihairabgwi, J. M. (2016). Asian Pacific Journal of Tropical Biomedicine. Phytochemical and antioxidant properties of different solvent extracts of Kirkia wilmsii tubers. Volume6 Number12: Page.1037-1043.

4. Faleye, O. S., Boya, B. R., Lee, J., Choi, I., and Lee, J. (2024). Pharmacological Reviews. Halogenated antimicrobial agents to combat drug-resistant pathogens. Volume76:Page.90-141.
5. Guillen, A. P., Bautista, E., Jimenez, J. M., and Douterlungne, D. (2025). Fitoterapia. Aristolochia species (Aristolochiaceae) from the Americas, a review of their traditional medicinal uses, phytochemistry, and pharmacological properties. Volume 183.
6. He, G., Zhu, X., Shen, T., and Wang, Y. (2024). Infrared Physics & Technology.FT-IR spectroscopy coupled with HPLC for qualitative and quantitative analysis of different parts of Gentiana rigescens Franch. Volume136.
7. Mohammed, H. A., Sulaiman, G. M., Khan, R. A., Al-Saffar, A. Z., Mohsin, M. H., Albukhaty, S., and Ismail, A. (2024). Process Biochemistry.Essential oils pharmacological activity: Chemical markers, biogenesis, plant sources, and commercial products. Volume 144: Page. 112-132.
8. Oliveira, R. N., Mancini, M. C., Oliveira, F. C. S., Passos, T. M., Quilty, B., Thire, R. M. S. M., and McGuinness, G. B. (2016). Revista Materia.FTIR analysis and quantification of phenols and flavonoids of five commercially available plants extracts used in wound healing. Volume 11743: Page. 767-779.
9. Ore, O. T., Adeola, A. O., Bayode, A. A., Adedipe, D. T., and Nomngongo, P. N. (2023). Environmental Chemistry and Ecotoxicology. Organophosphate pesticide residues in environmental and biological matrices: Occurrence, distribution and potential remedial approaches. Volume 5: Page. 9-23.
10. Paul, J. K., Azmal, M., Haque, ANM., S., N., B., Talukder, O. F., Meem, M., and Ghosh, A. (2024). Advances in Redox Research. Phytochemical-mediated modulation of signaling pathways: A promising avenue for drug discovery. Volume 13. 100113.
11. Pizzi, A. (2021). Sustainable Chemistry and Pharmacy.Tannins medical / pharmacological and related applications: A critical review. Volume 22.
12. Rahman, M. M., Rahaman, M. S., Islam, M. R., Rahman, F., Mithi, F. M., Alqahtani, T., Almikhlafi, M. A., Alghamdi, S. Q., Alruwaili, A. S., Hossain, M. S., Ahmed, M., Das, R., Emran, T. B., and Uddin, M. S. (2022).Molecules. Role of phenolic compounds in human disease: Current knowledge and future prospects.Volume 27.
13. Sharifi-Rad, J., Ozleyen, A., Tumer, T. B., Adetunji, C. O., Omari, N. E., Balahbib, A., Taheri, Y., Bouyahya, A., Martorell, M., Martins, N., and Cho, W. C. (2019). Biomolecules. Natural products and synthetic analogs as a source of antitumor drugs. Volume 9Number 11.
14. Siddiqui, T., Khan, M. U., Sharma, V., and Gupta, K. (2024). Phytomedicine Plus.Terpenoids in essential oils: Chemistry, classification, and potential impact on human health and industry. Volume 4.

15. Tripathi, S., Singh, S., Mishra, N., and Mishra, N. (2025). Current Research in Nutrition and Food Science. The impact of solvent polarity on the phenolic and antioxidant capacity of green coffee beans (Robusta species) extracts. Volume 132.
16. Uchenna, E. F., Adaeze, O. A., and Steve, A. C. (2015). Journal of Agricultural Science.Phytochemical and antimicrobial properties of the aqueous ethanolic extract of Saccharum officinarum (Sugarcane) bark. Volume 7, Number 10.
17. Ullah, A., Munir, S., Badshah, S. L., Khan, N., Ghani, L., Poulson, B. G., Emwas, A. H., and Jaremko, M. (2020). Molecules. Important flavonoids and their role as a therapeutic agent. Volume25.
18. Wang, X., Xin, J., Sun, L., Sun, Y., Xu, Y., Zhao, F., Niu, C., and Liu, S. (2024). Molecules. Exploring the biomedical potential of Terpenoid Alkaloids: Sources, structures, and activities. Volume 29 Number 9.
19. Wu, L., Wang, X., Xu, W., Farzaneh, F., and Xu, R. (2009). Current Medicinal Chemistry.The structure and pharmacological functions of coumarins and their derivatives. Volume 16 Number 32: Page. 4236-4260.
20. Yahia, Y., Bagues, M., Zaghdoud, C., Al-Amri, S. M., Nagaz, K., and Guerfel M. (2019). South African Journal of Botany. Phenolic profile, antioxidant capacity and antimicrobial activity of Calligonumarich L., desert endemic plant in Tunisia. Volume 124: Page. 414-419.