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Spectral analysis of fresh and processed shoots of an edible bamboo *Dendrocalamus hamiltonii* (Nees & Arn.)

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Abstract
Objectives: Bamboo shoots are reported to have great nutritional and therapeutic value. Processing and preservation bring about physical and biochemical changes which alter the nutritional and therapeutic values of the shoots. The aim of the present study was to determine the chemical profile of fresh and processed shoots of *Dendrocalamus hamiltonii* by various spectral analysis.
Methods: Spectral analysis was done by different analytical methods such as UV-VIS, FT-IR, NMR and GC-MS.
Results: In NMR analysis, aqueous extract of shoots showed a number of peaks in between δ 0.7 to 10 indicating the increase in alcoholic and acids content after processing. In GC-MS analysis, 110 compounds were detected in the ethanol extract of fresh and processed shoots with different pharmacological properties. The highest number of compounds was detected in the extract of fermented shoots.
Conclusions: The results of this study may act as biochemical markers for processed shoots in the food and pharmaceutical industry.

Keywords: *Dendrocalamus hamiltonii*, NMR, GC-MS, Spectral analysis

1. Introduction
Bamboo is one of the most valuable plant taxa worldwide because of its innumerable uses including food and medicine. The fresh juvenile bamboo shoot has been used from ancient times as food and medicine and has gained worldwide reputation for various functional merits as a dietary supplement, food antioxidant, cosmetics ingredient and as a nutraceutical component for preventing diseases. Due to its nutritive value and presence of bioactive compounds, the shoots are gaining importance for its health benefits and are emerging as a potential ingredient for modern functional foods and nutraceuticals [13]. But fresh juvenile bamboo shoots are seasonal, highly perishable and have high content of cyanogenic glycoside. Processing is a necessary to remove anti-nutrients, acridity and make the shoots palatable, increase the shelf life and for value addition. Analysis of nutritive value of bamboo shoots has been performed by several researchers [8, 3, 5, 4, 9, 12, 15, 6]. The young shoots are rich in proteins, carbohydrates, amino acids, vitamins and minerals and could help in solving nutritional deficiency of rural poor [16, 17, 11]. Apart from being nutritious, the shoots are a rich source of bioactive compounds which have been shown to be effective against several diseases. However, bamboo shoots have a very short shelf life of 3-4 days and the anti-nutrient cyanogenic glycoside needs to be removed before consumption. Fermentation, boiling, soaking, canning and salting are some common traditional processing methods which significantly reduce the amount of cyanogens and also improve the shelf life of shoots. Despite improving the palatability and shelf life, food processing procedures are also recognized as one of the major factors leading to changes of natural phytochemicals present in foods [10, 1]. A number of reactions take place during processing, either in series or in parallel. The final quality of food may be the results of many interacting and complex reactions rather than a single elementary step. Some high molecular weight phytochemicals may break down into a number of smaller compounds with low molecular weight thereby increasing and/or decreasing the total nutrient content or some new combinations of phytochemicals might be formed that further affect the nutritional and therapeutic value in the processed when compared to unprocessed foods. The processing of shoots is being done conventionally since ancient times by soaking, drying, boiling and fermentation, which bring a lot of changes to the shoots. However, all the changes are rarely visible or detected through conventional methods and detailed scientific validation of traditional processing methods in terms of food quality and safety has not been attempted [2]. A critical issue will be how to detect untargeted compounds

and determine their identity in foods, for which the development of advanced analytical techniques is expected to play a crucial role [18]. By analyzing the raw materials used as ingredients in food products, it is often possible to predict their behavior during processing so that appropriate processing method can be selected or altered to produce a final product with desired properties [14]. Moreover, the effectiveness of the bioactive compounds in preventing diseases depends on preserving the stability, bioactivity and bioavailability of the active ingredients [7]. Hence, the objective of the present study was to identify the phytochemical constituents at the macro- and micro- levels of fresh and processed shoots of *Dendrocalamus hamiltonii* with the aid of UV-VIS, FT-IR, NMR and GC-MS spectral investigation. The study will provide valid scientific data for developing a suitable improved system for processing and preservation of this perishable commodity for rural entrepreneurship.

2. Materials and Methods

2.1 Collection and preparation of Sample

Fresh and tender shoots of *Dendrocalamus hamiltonii (D. hamiltonii)* were collected in July 2015 from Shillong, Meghalaya (India). The shoots were transported from Shillong to Department of Botany, Panjab University, Chandigarh (India) by air. In laboratory, the shoots were defoliated and washed. The unwanted parts were removed, and the soft edible portion was chopped into small pieces and subjected to different processing methods (boiling, salting, fermentation, drying). Fresh, boiled (15 minutes), brine treated (boiled for 10 minutes and preserved in 5% brine solution for 1 month) and fermented (2 months) shoots were taken separately and dried at 40 ^{0}C. Thereafter, the shoots were pulverized to fine powder using mortar and pestle.

2.2 Extraction of plant material

10 g of each sample of bamboo shoot powder was taken and soaked in 100 ml each of distilled water (for aqueous extract) and ethanol in a conical flask, plugged with cotton wool and then kept on a rotary shaker at 120 rpm for 24 h. Extracts were then filtered and dried using a hot air oven at low temperature. Dried extracts were collected and stored in aliquots at 4 ^{0}C for further analysis.

2.3 Ultraviolet visible absorption (UV)

The aqueous extract of fresh and processed shoots of *D. hamiltonii* was analyzed in UV-Visible range between 200-800 nm using UV-Visible Spectrophotometer (Perkin Elmer). This method is useful for analyzing organic compounds viz. ketones, dienes etc.

2.4 Fourier Transform Infrared spectroscopy (FT-IR)

Fourier Transform Infrared spectroscopy is one of the powerful analytical techniques which offer the possibility of chemical identification. The technique is based on the simple fact that chemical substance shows selective absorption in infrared region. After absorption of IR radiations, the molecules vibrate, giving rise to absorption spectrum. It is an excellent method for the qualitative analysis because except optical isomers, the spectrum of compound is unique. The IR spectra of aqueous extract of fresh and processed shoots were scanned on FT-IR Spectrophotometer (Perkin Elmer) over the frequency range from 4000-400 cm-1. UV-VIS and FT-IR analysis was performed at National Institute of Pharmaceutical Education and Research (NIPER), Mohali (India).

2.5 Nuclear Magnetic Resonance spectroscopy (NMR)

Nuclear Magnetic Resonance spectroscopy is a powerful technique for identifying and quantifying components in complex mixtures such as plant extracts and provides a fast and highly reproducible means for identity, purity, strength and composition verification; thereby providing product assessment and quality control analysis. Metabolite fingerprinting by NMR is a fast, convenient, and effective tool for discriminating between groups of related samples and it identifies the most important regions of the spectrum for further analysis. ^{1}H NMR spectra were recorded on a NMR-400 MHz and chemical shifts were recorded as δ values.

2.6 Gas chromatography and mass Spectroscopy (GC-MS)

GC-MS analysis of ethanolic extract of fresh and processed shoots was carried out on a GC clarus 500 Perkin Elmer system comprising a AOC-20i auto sampler and gas chromatograph interfaced to a mass spectrometer instrument employing the following conditions: column Elite-1 fused silica capillary column (30 × 0.25 mm ID ×1EM df, composed of 100% Dimethyl poly siloxane), operating in electron impact mode at 70 eV; helium (99.999%) was used as carrier gas at a constant flow of 1ml/min and an injection volume of 0.5 EI was employed (split ratio of 10:1) injector temperature 250 °C; ion-source temperature 280 °C. The oven temperature was programmed from 110 °C (isothermal for 2 min), with an increase of 10 °C/min, to 200 °C/min, then 5 °C/min to 280 °C/min, ending with a 9 min isothermal at 280 °C. Mass spectra were taken at 70 eV; a scan interval of 0.5 s and fragments from 40 to 550 Da. ^{1}H NMR and GC-MS analysis was carried out in Sophisticated Analytical Instrumentation facility (SAIF), Panjab University Chandigarh, India.

3. Results

3.1 Ultraviolet visible absorption (UV)

UV-Visible spectra of aqueous extract of fresh and processed shoots are shown in Fig. 1. The UV spectrum of fresh shoots showed absorption maxima at 495.50, 275.50, 489.50 and 241.00 nm. The fermented shoots showed absorption maxima at 778.00, 756.50, 741.00, 644.00, 537.50, 493.00, 254.50, 721.00, 546.50, 510.00, 485.00 and 239.50 nm. The brine treated shoots showed absorption maxima at 738.50, 693.50, 643.50, 589.00, 573.00, 568.50, 557.50, 537.00, 496.50, 272.00, 721.50, 690.00, 647.00, 571.00, 546.00, 509.50, 455.00 and 243.50 nm while, boiled shoots showed absorption maxima at 785.00, 498.50, 277.00, 645.50, 536.50, 496.50 and 241.50 nm.

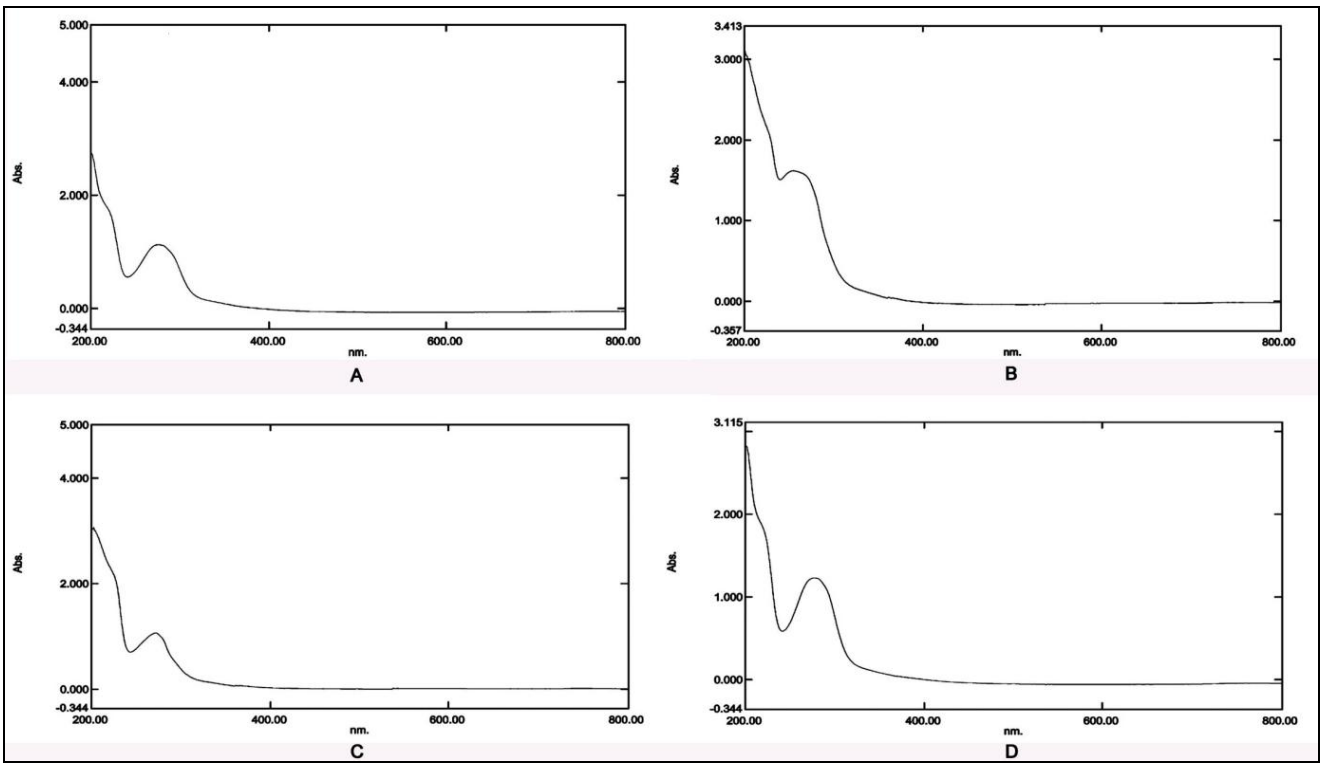

Fig 1: UV-VIS spectra of aqueous extract of fresh (A), fermented (B), brine treated (C) and boiled (D) shoots of *D. hamiltonii*

3.2 FT-IR spectroscopy

FT-IR spectra of aqueous extract of fresh and processed shoots are shown in Fig. 2. The mid infrared, approximately 4000–400 cm^{-1} was used to study the fundamental vibrations and associated rotational vibrational spectrum. Interpretation of aqueous extract of fresh and processed shoots is given in Table 1.

Table 1: Interpretation of FT-IR of aqueous extract of fresh and processed shoots of *D. hamiltonii*

Wavelength (cm^{-1})	Interpretation
Fresh shoots	
3350.43	Alcohol, phenol (O-H stretch)
1629.12	1º amines (N-H bend)
1514.96	Nitro compounds (N-O stretch)
1399.03	Aromatics (C-C stretch)
1307.51	Esters (C-O stretch)
1220.38	Carboxylic acid (C-O stretch)
1157.57	Alcohol (C-O stretch)
1075.60	Aliphatic amines (C-C stretch)
794.92	Alkyl halides (C-Cl stretch)
Fermented shoots	
3302.51	Alcohol, phenol (O-H stretch)
2126.58	Alkynes (C≡C stretch)
1638.07	1º amines (N-H stretch)
Brine treated shoots	
3362.34	Alcohol, phenol (O-H stretch)
2926.88	Alkanes (C-H stretch)
2138.01	Alkynes (C≡C stretch)
1638.36	1ºamines (N-H stretch)
1083.62	Aliphatic amines (C-N stretch)
Boiled shoots	
3389.99	Alcohol, phenol (O-H stretch)
2930.14	Alkanes (C-H stretch)
1633.92	1ºamines (N-H stretch)
1515.34	Nitro compounds (N-O stretch)
1402.78	Aromatics (C-C stretch)
1308.67	Esters (C-O stretch)
1237.81	Carboxylic acid (C-O stretch)
1158.57	Alcohol (C-O stretch)
1075.6	Aliphatic amines (C-N stretch)
795	Alkyl halides (C-Cl stretch)

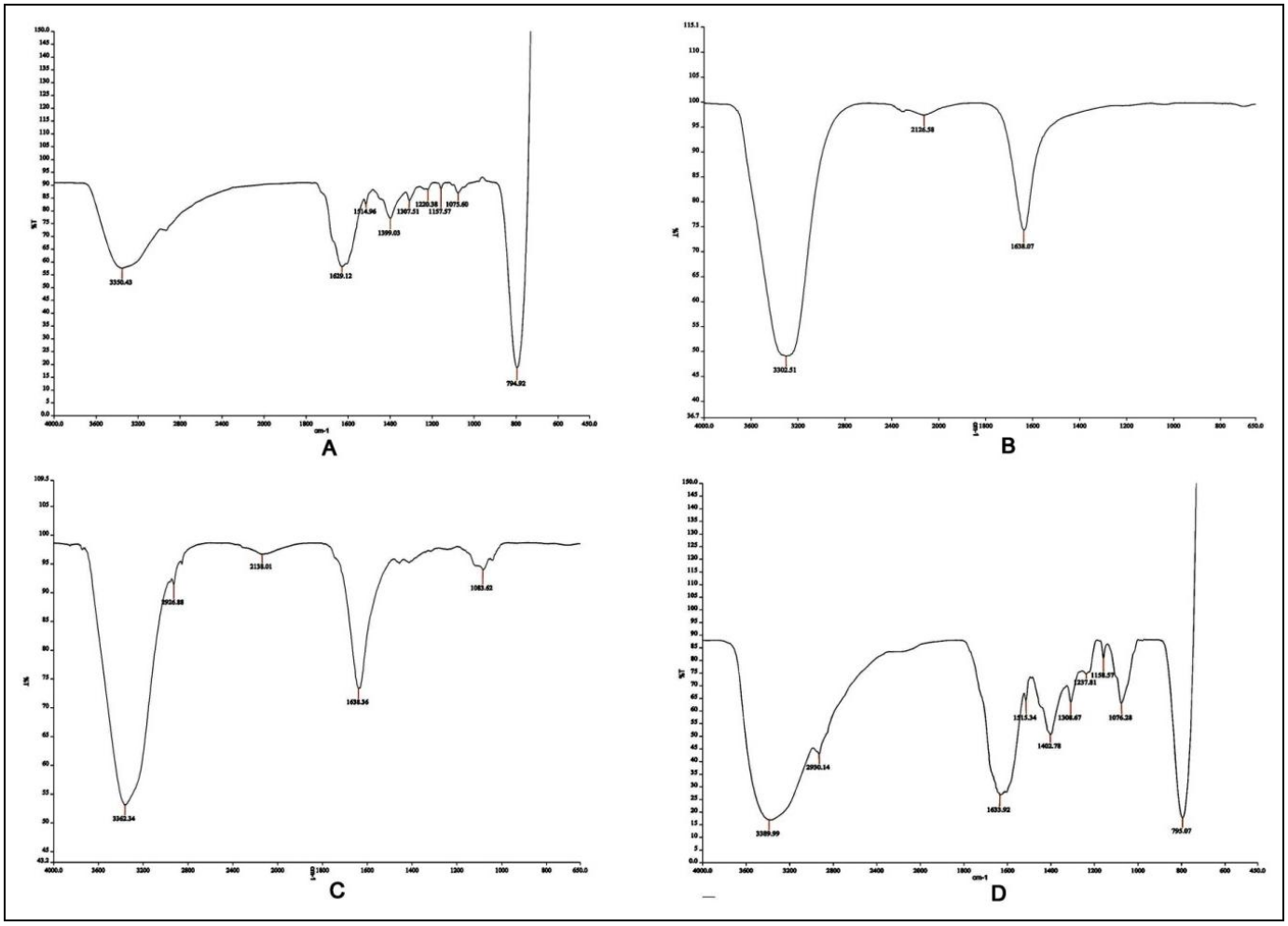

Fig 2: FT-IR spectra of the aqueous extract of fresh (A), fermented (B), brine treated (C) and boiled (D) shoots of *D. hamiltonii*

3.3 NMR spectroscopy

^{1}H NMR spectra of aqueous extract of fresh and processed shoots was recorded on a NMR-400 MHz and chemical shifts were recorded as δ values. The result graph was compared with the reference chart and possible functional groups present in the shoots were determined. ^{1}H NMR analysis of the fresh and processed shoots showed a number of peaks in between δ 0.7 to 10. Each chemical shift value corresponds to a set of protons in a particular environment. The intensity of each signal signifies the number of protons of each type. The chemical shift values of the various signals and their functional groups present in fresh and processed shoots are given in Table 2, Fig.3.

Table 2: ^{1}H NMR data and their assignment from aqueous extract of fresh and processed shoots of *D. hamiltonii*

Sr. No.	Chemical shift (ppm)	Nature of proton			
		Fresh shoots	Fermented shoots	Brine treated shoots	Boiled shoots
1	0.7	-	O-H	-	O-H
2	0.8	-	O-H	O-H	O-H
3	0.9	-	RCH_3	RCH_3	RCH_3
4	1-5	RNH_2	RNH_2	RNH_2	RNH_2
5	1.3	R_2CH_2	R_2CH_2	R_2CH_2	R_2CH_2
6	1.8	HC-NHR	-	-	HC-NHR
7	1.9	-	HC-NHR	HC-NHR	Ar-C-H
8	2.4	-	Ar-C-H	Ar-C-H	-
9	2-2.6	-	HC-COOH	HC-COOH	HC-COOH
10	2.7	-	HC-C=O	-	HC-C=O
11	2.8	-	-	ROH	-
12	3-4	HC-Cl	HC-Cl	HC-Cl	HC-Cl
13	3.4-4	HC-OH	HC-OH	HC-OH	HC-OH
14	4-4.5	HC-F	HC-F	HC-F	HC-F
15	6-8.5	Ar-H	Ar-H	Ar-H	Ar-H
16	7-7.5	C=C-H	C=C-H	C=C-H	C=C-H
17	9-10	RCHO	-	RCHO	-

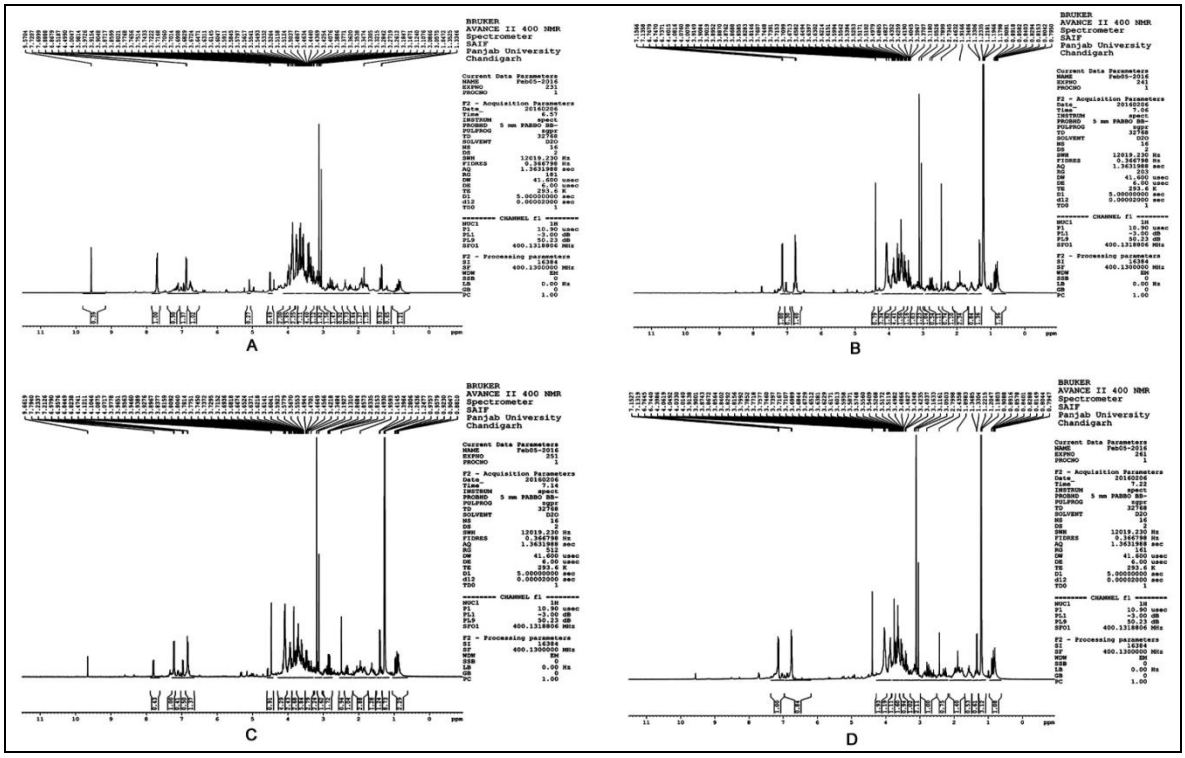

Fig 3: [1]H NMR spectra of aqueous extract of fresh (A), fermented (B), brine treated (C) and boiled (D) shoots of *D. hamiltonii*

3.4 GC-MS spectroscopy

GC-MS analysis has found a variety of analytical uses, including quality control analysis in both the pharmaceutical and food product industries. Applications of this technique include drug detection, fire investigation, environmental analysis, and identification of unknown samples. The detail tabulation of the GC-MS analysis of ethanol extract of fresh and processed shoots of *D. hamiltonii* is given in Table 3.

Table 3: Phytochemicals identified in the ethanolic extract of fresh and processed shoots of *D. hamiltonii* by GC-MS

S. No.	RT	Peak Area (%)	Molecular formula	Name of the compound
Fresh shoots				
1	10.65	2.52	$C_{11}H_{24}O$	1-Undecanol
2	13.44	3.62	$C^{16}H^{32}O$	Hexadecen-1-ol,Trans-9-
3	14.19	8.63	$C_{10}H_9ClO_3$	Propanoic acid,3-chloro-,4-formylphenyl ester
4	14.27	2.91	$C_7H_6O_2$	Benzaldehyde, 3-hydroxy-
5	14.57	2.12	$C_7H_{10}N_2$	3-Pyridinamine, N,N-dimethyl-
6	14.66	1.39	$C_{16}H_{27}N_3O_2$	5-Aminovaleramide,N-methyl-N-[4-(l-pyrrolidinyl)- 2-butynyl]-N'-acetyl-
7	14.74	1.20	$C_{14}H_{42}O_7Si_7$	Cycloheptasiloxane, tetradecamethyl-
8	15.41	4.95	$C_{14}H_{22}O_2$	2-(1,1,3,3-Tetramethylbutyl)-1,4-benzenediol
9	15.91	2.49	$C_{13}H_{26}$	Cyclotridecane
10	18.14	1.17	$C_{17}H_{36}O$	n-Heptadecanol-1
11	26.14	2.43	$C_{27}H_{54}O_4Si_2$	1-Monolinoleoylglycerol trimethylsilyl ether
12	26.26	0.69	$C_{27}H_{52}O_4Si_2$	9,12,15-Octadecatrienoic acid, 2,3-bis[(trimethylsilyl) oxy]propyl ester, (Z,Z,Z)-
13	26.86	25.02	$C_{22}H_{43}NO$	13-Docosenamide,(Z)-
14	28.49	1.07	$C_{27}H_{42}O_4$	Propanoic acid, 2-(3-acetoxy-4,4,14-trimethylandrost -8-en-17-yl)-
15	33.99	2.93	$C_{30}H_{50}O_6$	Olean-12-ene-3,15,16,21,22,28-hexol
16	35.97	2.40	$C_{28}H_{38}O_{10}$	3-Desoxo-3,16-dihydroxy-12-desoxyphorbol 3,13, 16, 20-tetraacetate
17	36.05	4.43	$C_{29}H_{50}O$	ç-Sitosterol
18	36.19	3.94	$C_{32}H_{50}O_6$	Dodecanoic acid, 1a,2,5,5a,6,9,10,10a-octa hydro-5,5a-dihydroxy-4-(h ydroxy methyl)-1,1,7,9-tetramethyl-11-oxo-1H-2,8a- methano cyclopenta[a] cyclopropa [e] cyclodecen-6-yl ester
19	36.84	9.18	$C_{27}H_{36}O_{10}$	4H-Cyclopropa[5',6']benz [1',2':7,8]azuleno [5,6]oxire n-4-one, 8,8a-bis(acetyloxy)-2a-[(acetyloxy)methyl]-1, 1a,1b, 1c,2a,3,3a,6a, 6b,7,8,8a-dodecahydro-6b-0 hydroxy-3a- methoxy-1,1,5,7-tetramethyl-
20	38.49	4.97	$C_{27}H_{46}O_5$	Cholestan-26-oic acid, 3,7,12-trihydroxy-
Fermented shoots				
21	5.53	1.99	$C_6H_{12}O_2$	2-Pentanone, 4-hydroxy-4-methyl-
22	10.05	1.14	$C_{10}H_{30}O_5Si_5$	Cyclopentasiloxane, decamethyl-
23	10.66	3.48	$C_{16}H_{34}O$	1-Hexadecanol
24	13.45	2.89	$C_{14}H_{28}$	Cyclotetradecane
25	13.99	2.62	$C_7H_8O_2$	Benzenemethanol, 4-hydroxy-

26	15.06	1.22	$C_{14}H_{22}O$	Phenol, 2,5-bis(1,1-dimethylethyl)-
27	15.93	2.75	$C_{16}H_{32}O$	Hexadecen-1-ol, trans-9-
28	18.16	1.52	$C_{21}H_{42}$	10-Heneicosene (c,t)
29	19.99	1.35	$C_{20}H_{30}O_4$	Phthalic acid, butyl 4-octyl ester
30	20.20	3.74	$C_{18}H_{36}O_2$	Hexadecanoic acid, ethyl ester
31	21.78	3.01	$C_{20}H_{36}O_2$	9,12-Octadecadienoic acid, ethyl ester
32	23.62	11.20	$C_{22}H_{43}NO$	13-Docosenamide, (Z)-
33	25.02	1.02	$C_{27}H_{54}O_4Si_2$	1-Monolinoleoylglycerol trimethylsilyl ether
34	26.05	1.86	$C_{27}H_{42}O_4$	Propanoic acid, 2-(3-acetoxy-4,4,14-trimethylandrost-8-en-17-yl)-
35	26.05	1.86	$C_{28}H_{40}O_{10}$	9-Desoxo-9-x-acetoxy-3,8,12-tri-O-acetylingol
36	26.75	0.94	$C_{27}H_{46}0_5$	Cholestan-26-oic acid, 3,7,12-trihydroxy-
37	27.26	0.64	$C_{27}H_{36}O_8$	(22R)-21-Acetoxy-6à,11á-dihydroxy-16à,17à-propy lmethylenedioxypregna-1,4-diene-3,20-dione
38	28.76	1.14	$C_{24}H_{31}FO_6$	Betamethasone acetate
39	30.20	0.66	$C^{27}H^{52}O^4Si^2$	9,12,15-Octadecatrienoic acid, 2,3-bis[(trimethylsilyl)oxy]propyl ester, (Z,Z,Z)-
40	31.04	0.77	$C_{28}H_{38}O_{10}$	3-Desoxo-3,16-dihydroxy-12-desoxyphorbol 3,13,16,20-tetraacetate
41	33.78	2.43	$C_{28}H_{38}O_{12}$	5H-Cyclopropa[3,4]benz[1,2-e]azulen-5-one, 2,9,9a-tris(acetyloxy)-3-[(acetyloxy)methyl]-1,1a,1b, 2,3,4, 4a,7a,7b,8,9,9a-dodecahydro-3,4a,7b-trihydro xy-1,1,6,8-tetramethyl-
42	33.85	2.04	$C_{30}H_{50}O_6$	Olean-12-ene-3,15,16,21,22,28-hexol
43	33.93	1.23	$C_{18}H_{24}MoO_4$	Molybdenum, dicarbonylbis(ü-4-2-methylene cycloheptanone)-
44	35.42	0.45	$C_{29}H_{56}N_2O_{10}Si_3$	D-Glucopyranosiduronic acid, 3-(5-ethylhexahydro-1,3-dimethyl-2,4,6-trioxo-5-pyrimidinyl)-1-methy lbutyl 2,3,4-tris-O-(trimethylsilyl)-, methyl ester
45	35.60	1.21	$C_{34}H_{50}O_5$	1',1'-Dicarboethoxy-1á,2á-dihydro-3'H-cycloprop[1, 2]cholesta-1,4,6-trien-3-one
46	35.77	6.30	$C_{29}H_{50}O$	ç-Sitosterol
47	35.84	3.58	$C_{29}H_{50}O$	á-Sitosterol
48	36.02	8.33	$C_{28}H_{46}O_2$	3Beta,5-epoxy-a-homo-5beta-cholest-4-en-3alpha-ol
49	36.50	2.63	$C_{25}H_{39}NO_5$	Acetic acid, 17-acetoxy-3-hydroxyimino-4,4,13-trimethyl-hexad ecahydro cyclopenta [a] phenanthren-10-ylmethyl ester
50	36.88	3.48	$C_{30}H_{40}O_6$	2,4,6-Decatrienoic acid, 1a,2,5,5a,6,9,10,10a-octahydro-5,5a-dihydroxy-4-(h ydroxymethyl)-1,1,7,9-tetramethyl-11-oxo-1H-2,8a- methano cyclopenta[a] cyclopropa[e] cyclodecen-6-yl ester
51	37.84	0.83	$C_{27}H_{54}O_4Si_2$	1-Monolinoleoylglycerol trimethylsilyl ether
52	38.13	1.12	$C_{16}H_{50}O_7Si_8$	Octasiloxane
53	38.54	11.60	$C_{29}H_{48}O$	Stigmast-4-en-3-one
54	39.23	1.26	$C_{28}H_{38}O_{10}$	4a,7a-Epoxy-5H-cyclopenta [a]cyclopropa [f]cyclo undecen-4(1H)-one, 2,7,10,11-tetrakis (acetyloxy)-1a,2,3,6,7,10,11,11a-oc tahydro-1,1,3,6,9-pentamethyl
55	40.77	1.48	$C_{36}H_{76}O_5Si_5$	Silane,[[(3á,5à,11á,20S)-pregnane-3,11,17,20,21 -pentayl]p entakis(oxy)]pentakis[trimethyl-
Brine treated shoots				
56	5.87	2.64	$C_{20}H_{22}N_2O_2$	1,16-Cyclocorynan-17-oic acid, 19,20-didehydro-, methyl ester, (16S,19E)-
57	6.00	1.79	$C_{10}H_{19}N_3O_4$	Glycine, N-(N-glycyl-L-leucyl)-
58	6.09	1.42	$C_{30}H_{38}O_{11}$	Carda-16,20(22)-dienolide, 3-[(6-deoxy-3,4-O-methyl enehexopyranos-2-ulos-1-yl)oxy]-5,11,14-trihydroxy-12-oxo-
59	6.24	1.70	$C_{52}H_{80}O_2$	Phenol, 2-methoxy-6-(3,7,11,15,19,23,27,31,35-nona methyl-2,6,10,14,18,22,26,30,34-hexa triacontanonaenyl)-
60	6.42	2.08	$C_{32}H_{58}N_2O_6Si_3$	Pregn-4-ene-3,11,20-trione,6,17,21-tri[(trimethylsilyl) oxy]-, 3,20-bis(O-methyloxime), (6á)-
61	6.61	1.44	$C_{53}H_{72}N_8O_9$	L-Lysine, N6-acetyl-N2-[N-[N-[N-(N2-acetyl-N,N,N2-trimeth yl-L-asparaginyl)-N-methyl-L-phenylalanyl]-N-meth yl-L-phenylalanyl]-N,1-dimethyl-L-tryptophyl]-N2, N6-dimethyl-, methyl ester
62	6.83	2.50	$C_{15}H_{14}N_2O_2$	Pyrrolidine-2,5-dione,1-(1-naphthylaminometh yl)-
63	6.97	2.79	$C_{37}H_{24}Cl_2N_6O_4$	2,2-Bis[4-[[4-chloro-6-(3-ethynylphenoxy)-1,3,5-tri azin-2-yl]oxy]phenyl]propane
64	7.01	2.84	$C_4H_8Cl_2O_4S_2$	3,3-Dichloro-2,4-dithiahexane 2,2,4,4-tetroxide
65	7.64	1.75	$C_{11}H_{21}N$	2-Cyclohexylpiperidine
66	7.73	2.80	$C_8H_{15}NO_2$	4-Propionyloxypiperidine
67	7.80	1.32	$C_2H_2Cl_2O$	Dichloroacetaldehyde
68	7.84	2.73	$C_{16}H_{21}N_3O_2S_2$	N-Morpholinomethylidene-3-morpholino-2-(2-thien yl)thioacrylamide
69	8.17	3.61	$C_{32}H_{48}O_6$	2-(16-Acetoxy-11-hydroxy-4,8,10,14-tetramethyl-3- oxohexadecahydrocyclopenta[a]phenanthren-17-ylid ene)-6-methyl-hept-5-enoic acid, methyl ester
70	8.26	3.73	$C_{30}H_{61}NO_5Si_3$	Prost-13-en-1-oic acid, 9-(methoxyimino)-11,15-bis [(trimethylsilyl)oxy]-, trimethylsilyl ester
71	8.42	4.22	$C_{27}H_{38}O_9$	3,9-Epoxypregnan-14-ol-20-one, 3,11,18-triacetoxy-
72	8.47	2.29	$C_{36}H_{48}O_8$	2,4,6,8-Tetradecatetraenoic acid, 9a-(acetyloxy)-1a, 1b, 4,4a,5,7a,7b,8,9,9a-decahydro- 4a,7b-dihydroxy-3-(hydroxymethyl)-1,1,6,8-tetramet hyl-5-oxo-1H-cyclo propa[3,4]benz[1,2-e]azulen-9-yl ester
73	8.82	2.51	$C_{19}H_{38}O_3$	7 Octadecanoic acid, 4-hydroxy-, methyl ester
74	8.96	2.49	$C_6H_{13}NO$	Oxazolidine, 2-ethyl-2-methyl-
75	9.31	2.22	$C_{25}H_{24}Cl_3N_3O_2$	7-Chloro-3-[2,4-dichlorophenyl]-1,3,4,10-tetrahydro -10-hydroxy-2-methyl-1-[1-piperidinylimino]-9(2H)- acridinone
76	9.69	1.71	$C_{16}H_{24}O_2$	1,3-Dioxolane, 2-heptyl-4-phenyl-
77	10.05	1.83	$C_{23}H_{26}N_2O_4$	N-(1-Hydroxy-4-oxo-1-phenylperhydroquinolizin-3-yl) carbamic acid, benzyl ester
78	10.14	4.41	$C_{26}H_{54}$	Octadecane, 3-ethyl-5-(2-ethylbutyl)-
79	10.35	2.42	$C_{24}H_{32}O_6$	Butanoic acid, 1a,2,5,5a,6,9,10,10a-octahydro-5a-hydroxy-4-(hydr oxymethyl)-1,1,7,9-tetramethyl-6,11-dioxo-1H-2,8a –methanocyclo penta[a] cyclopropa [e]cyclodecen-5-y l ester

80	10.47	3.11	$C_{36}H_{69}NO_6Si_3$	Glycine, N-[(3à,5á,7à,12à)-24-oxo-3,7,12-tris [(trimethylsilyl) oxy]cholan-24-yl]-, methyl ester
81	10.51	1.82	$C_{23}H_{37}NO_3Si_2$	Dihydromorphine, di(trimehylsilyl) ether
82	10.58	2.26	$C_{16}H_{21}N_3O_2S_2$	N-Morpholinomethylidene-3-morpholino-2-(2-thien yl)thioacrylamide
83	10.73	3.91	$C_{12}H_{36}O_6Si_6$	Cyclohexasiloxane, dodecamethyl-
84	11.15	1.21	$C_{33}H_{55}ClO_3$	Acetic acid, 17-(4-chloro-5-methoxy-1,5-dimethyl hexyl)-4,4,10,1 3,14-pentamethyl-2,3,4,5,6,7, 10,11, 12,13,14,15,16, 17-tetradecahydro-1-phenanthryl-
85	11.27	1.73	$C_{26}H_{54}$	Octadecane, 3-ethyl-5-(2-ethylbutyl)-
86	12.20	2.10	$C_{24}H_{40}O_5$	Trihydroxycholanic acid
87	12.40	2.18	$C_{22}H_{34}O_5$	Benzenedodecanoic acid, 3-methoxy-2-(methoxycarbonyl)-, methyl ester
88	12.40	2.18	$C_{35}H_{70}O_3$	1,3-Dioxane, 5-(hexadecyloxy)-2-pentadecyl-, trans-
89	12.96	4.24	$C_{16}H_{50}O_7Si_8$	Octasiloxane, 1,1,3,3,5,5,7,7,9,9,11, 11,13, 13,15, 15-hexadecamethy l-
Boiled shoots				
90	10.65	3.08	$C_{11}H_{24}O$	1-Undecanol
91	13.45	3.22	$C_{13}H_{26}$	Cyclotridecane
92	15.92	2.97	$C_{16}H_{32}O$	Hexadecen-1-ol, trans-9-
93	18.15	2.55	$C_{22}H_{44}O_2$	1-Heneicosyl formate
94	19.99	1.33	$C_{20}H_{29}ClO_4$	Phthalic acid, butyl 8-chlorooctyl ester
95	21.77	1.82	$C_{21}H_{38}O_2$	n-Propyl 9,12-octadecadienoate
96	25.28	3.00	$C_{28}H_{43}NO_6$	(5á)Pregnane-3,20á-diol, 14à,18à-[4-methyl-3-oxo-(1-oxa-4-azabutane-1,4-diy l)]-, diacetate
97	26.70	1.07	$C_{27}H_{52}O_4Si_2$	9,12,15-Octadecatrienoic acid, 2,3-bis[(trimethylsilyl) oxy]propyl ester, (Z,Z,Z)-
98	26.88	28.48	$C_{22}H_{43}NO$	13-Docosenamide, (Z)-
99	27.87	0.92	$C_{35}H_{70}O_3$	1,3-Dioxane, 5-(hexadecyloxy)-2-pentadecyl-, trans-
100	30.71	0.75	$C_{27}H_{54}O_4Si_2$	1-Monolinoleoylglycerol trimethylsilyl ether
101	30.79	1.22	$C_{25}H_{34}O_7$	(22S)-6à,11á,21-Trihydroxy-16à,17à-propylmethyle nedioxypregna-1,4-diene-3,20-dione
102	30.88	0.96	$C_{12}H_{38}O_5Si_6$	Hexasiloxane, 1,1,3,3,5,5,7,7,9,9,11,11-dodecamethyl-
103	31.98	2.12	$C_{27}H_{40}O_4$	Spirost-8-en-11-one, 3-hydroxy-
104	32.27	1.66	$C_{36}H_{69}NO_6Si_3$	Glycine, N-[(3à,5á,7à,12à)-24-oxo-3,7,12-tris[(trimethylsilyl)oxy]cholan-24-yl]-, methyl ester
105	33.49	2.17	$C_{27}H_{36}O_{10}$	4H-Cyclopropa [5',6']benz[1',2':7,8] azuleno [5,6-b]ox iren-4-one, 8,8a-bis(acetyloxy)-2a-[(acetyloxy)methyl]-1,1a,1b,1c,2a,3,3a, 6a,6b, 7,8,8a-dodecahydro-6b-hydroxy-3a- methoxy -1,1,5,7-tetramethyl-
106	33.56	3.12	$C_{35}H_{48}O_3$	25-Norisopropyl-9,19-cyclolanostan-22-en-24-one,3-acetoxy-24-phenyl-4,4,14-trimethyl-
107	33.64	4.33	$C_{26}H_{44}O_5$	Ethyl iso-allocholate
108	35.54	9.21	$C_{29}H_{50}O$	ç-Sitosterol
109	36.79	0.88	$C_{36}H_{58}O_6$	Hexadecanoic acid,1a,2,5,5a,6,9,10,10a-octahydro-5,5a-dihydroxy-4-(hydroxymethyl) -1,1,7,9-tetramethyl-11-oxo-1H-2,8a-methano cyclopenta[a] cyclopropa[e] cyclodecen-6-yl ester
110	38.55	4.30	$C_{23}H_{32}O_6$	Hydrocortisone acetate

4. Discussion

The ultraviolet spectroscopy method is very much useful for identification of unsaturated bonds present in a plant component, which can be used to distinguish between conjugated and non conjugated system. Compounds containing halogen, ether, amino and hydroxy groups absorb light in the shorter wavelength from 190 to 380 nm due to $n \rightarrow \sigma^*$ transition. The compounds with unsaturated centers in the molecule such as alkenyl, carbonyl, imino, and azo groups absorb light in the longer wavelength from 380 to 900 nm due to $n \rightarrow \pi^*$ transition. The results of UV-VIS spectrum provided the evidence that most of the compounds in the processed bamboo shoots are with unsaturated centers in their molecules. The FT-IR spectra were used to identify and detect the characteristic peaks and functional groups of the active components based on the peak value in the region of infrared radiation. The more intense bands in fresh shoots occurring at 3350 cm^{-1}, 1629 cm^{-1}, 1514 cm^{-1}, 1399 cm^{-1}, 1307 cm^{-1}, 1220 cm^{-1}, 1157 cm^{-1}, 1075 cm^{-1}, 794 cm^{-1} corresponding to O-H/N-H/N-O,C-C, C-O and C-Cl stretching/bending vibrations respectively indicate the presence of alcohols, phenols, amino acids, nitro compounds, aromatics, esters, carboxylic acids, aliphatic amines and alkyl halides. The strong absorption band observed around 3302 – 3389 cm^{-1} may be due to the presence of bonded O-H stretching of alcohols and phenols. The very strong absorption at 3350.43 cm^{-1} in the extract of fresh shoots, 3302.51 cm^{-1} in fermented shoots, 3362.34 cm^{-1} in brine treated shoots and 3389.99 cm^{-1} in boiled shoots was observed. The more intense bands in extract of fermented shoots occurring at 3302 cm^{-1}, 2126 cm^{-1} and 1638 cm^{-1} corresponding to O-H, C≡C and N-H stretching/bending vibrations respectively indicate the presence of alcohols/phenols, alkynes and primary amines. In brine treated shoots, the very strong absorption was observed at 3362 cm^{-1}, 2926 cm^{-1}, 2138 cm^{-1}, 1638 cm^{-1} and 1083 cm^{-1} corresponding to O-H, C-H, C≡C, N-H and C-N stretching/bending vibrations respectively indicate the presence of alcohols/phenols, alkanes, alkynes, primary amines and aliphatic amines. In the case of boiled shoots the more intense bands occurring at 3389 cm^{-1}, 2930 cm^{-1}, 1633 cm^{-1}, 1515 cm^{-1}, 1402 cm^{-1}, 1308 cm^{-1}, 1237 cm^{-1}, 1158 cm^{-1}, 1076 cm^{-1} and 795 cm^{-1} corresponding to O-H, C-H, N-H, N-O, C-C, C-O, C-N, C-Cl stretching/bending vibrations respectively indicate the presence of alcohols/phenols, alkanes, primary amines, nitro compounds, aromatics, esters, carboxylic acids, alcohols, aliphatic amines and alkyl halides. These results provided the evidence that fresh and processed shoots of *D. hamiltonii* have high content of alcohols/phenols and amino acids.

The bands occurring at 1514 cm^{-1}, 1399 cm^{-1}, 1307 cm^{-1}, 1220 cm^{-1}, 1157 cm^{-1}, 1075 cm^{-1} and 794 cm^{-1} in fresh shoots and 1515 cm^{-1}, 1402 cm^{-1}, 1308 cm^{-1}, 1237 cm^{-1}, 1158 cm^{-1}, 1076 cm^{-1} and 795 cm^{-1} in boiled shoots indicate the presence of nitro compounds, aromatics, esters, carboxylic acids, alcohols, aliphatic amines and alkyl halides respectively. These bands were not observed in the aqueous extract of brine treated and fermented shoots. The absorption band observed around 2100-2260 cm^{-1} may be due to the

presence of C≡C stretching of alkynes. The aqueous extract of brine treated and fermented shoots showed absorption at 2138 cm^{-1} and 2126 cm^{-1} respectively. But there was no absorbance in between the region 2100-2260 cm^{-1} in the case of fresh and boiled shoots, indicated that no cyanide group was present in the aqueous extract of fresh and boiled shoots. ^{1}H NMR analysis of the shoots showed a number of peaks in between δ 0.7 to 10. Each chemical shift value corresponds to a set of protons in a particular environment. The intensity of each signal signifies the number of protons of each type. The range 0.7 to 0.8 ppm signifies the presence of hydroxy proton (-OH). The doublet at 0.9 ppm indicates the presence of primary aliphatic compounds. The range 1 to 5 ppm signifies the presence of amino acids while, the signal at 1.3 and 1.8 ppm is an indicative of secondary aliphatic compounds and amines respectively. The benzylic, hydroxylic and carbonyl compounds gave signals at 2.4, 2.8 and 2.7 ppm respectively. Other identified compounds were chlorides (3-4 ppm), alcohols (3.4-4 ppm), fluorides (4-4.5 ppm), aromatics (6-8.5 ppm), vinylic, conjugated (7-7.5 ppm) and aldehydes (9-10 ppm). The peak for aldehydes was not observed in the extract of fermented and boiled shoots. The hydroxy protons (-OH) were identified with the signal at the range of 3.4 to 4 ppm in the extract of fresh shoots but in processed forms, hydroxy protons (-OH) also gave the signals at 0.7 and 0.8 ppm. Similarly, the range 2 to 2.6 ppm which is an indicative of acids was not observed in the extract of fresh shoots but identified in all the processed forms. It showed that alcoholic and acid content increased after processing.

The results of FT-IR and NMR spectroscopy were further confirmed with the aid of GC-MS spectroscopy. Gas chromatography-mass spectrometry (GC-MS) is a method that combines the features of gas-liquid chromatography and mass spectrometry to identify different substances within a test sample. In the presence study, GC-MS data showed the highest presence of fatty acid amide namely 13-Docosenamide, (Z)- in the ethanol extract of fresh (25.02%) and boiled shoots (28.48%). 13-Docosenamide, (Z)- is used as antistatic agent. It is also used in the manufacturing of food packaging material, personal daily care products like perfumes, deodorant, lotions, moisturizers, talcum powder, soaps, toothpaste etc. Other main components present in the ethanol extract of fresh shoots were 4H-Cyclopropa [5',6'] benz [1',2':7,8] azuleno [5,6] oxiren-4-one,8,8a-bis(acetyloxy)-2a-[(acetyloxy) methyl]-1,1a,1b, 1c,2a,3,3a,6a, 6b,7,8,8a-dodecahydro-6b-0 hydroxy-3a- methoxy-1,1,5,7-tetramethyl- (9.18%), Propanoic acid, 3-chloro-, 4-formylphenyl ester (8.63%), 2-(1,1,3,3-Tetramethyl butyl)-1,4-benzenediol (4.84%) and ç-Sitosterol (4.43%). The ethanol extract of fermented shoots showed the highest presence of Stigmast-4-en-3-one (11.60%), followed by 13-Docosenamide, (Z)- (11.20%), 3Beta,5-epoxy-a-homo-5beta-cholest-4-en-3alpha-ol (8.33%) and ç-Sitosterol (6.30%). Stigmast-4-en-3-one is an alcohol having hypoglycaemic properties. The major bioactive compounds present in the ethanol extract of brine treated shoots were Octadecane, 3-ethyl-5- (2-ethylbutyl)- (4.41%) followed by 3,9-Epoxypregnan-14-ol-20-one,3,11,18-triacetoxy- (4.22%) and Octasiloxane, 1,1,3,3,5,5,7, 7,9,9,11,11,13, 13,15, 15-hexadecamethy l- (4.24%). Octadecane, 3-ethyl-5-(2-ethylbutyl)- is reported to have antifungal properties while, Octasiloxane, 1,1,3,3,5,5,7,7,9,9,11, 11,13, 13,15, 15-hexadecamethy l-, a volatile organic compound having antimicrobial properties. The compounds such as 9, 12, 15-Octadecatrienoic acid, 2,3-bis [(trimethylsilyl)oxy] propyl ester, (Z,Z,Z)-, 1-Monolinoleoylglycerol trimethylsilyl ether and ç-Sitosterol were detected in all kind of shoots but their concentration changed after processing. The level of 9, 12, 15-Octadecatrienoic acid, 2,3-bis [(trimethylsilyl)oxy] propyl ester, (Z,Z,Z)- (Linolenic acid ester) increased in boiled shoots while decreased after fermentation. The health beneficial properties of this compound include anti-inflammatory, hypocholesterolemic, antiarthritic, nematicide and hepatoprotective. 1-Monolinoleoylglycerol trimethylsilyl ether (steroid) having antioxidant, antimicrobial, anti-inflammatory and antiarthritic properties. The concentration of this compound decreased in processed shoots when compared to unprocessed shoots.

The phytosterol content increased after fermentation and it is well-established that high intakes of plant sterols can lower serum total and LDL cholesterol concentrations in humans. In all, 110 compounds were identified in the ethanol extract of fresh and processed shoots. The highest number of compounds (35) was identified in the ethanol extract of fermented shoots followed by brine treated shoots (34), boiled shoots (21) and fresh shoots (20). The results of GC-MS spectroscopy revealed that processing techniques bring a lot of changes in the chemical profile of bamboo shoots. Hence, the final quality of shoots may be the results of many interacting and complex reactions rather than a single elementary step. Some high molecular weight phytochemicals may break down into a number of smaller compounds with low molecular weight during processing thereby increasing and/or decreasing the total nutrient and bioactive content of shoots that further affect the nutritional and therapeutic value in the processed shoots when compared to unprocessed shoots.

5. Conclusion

Spectroscopic techniques have become a powerful analytical tool for the qualitative and quantitative analysis of biological materials. In this study, changes in the phytochemical constituents of shoots of *D. hamiltonii* after processing were determined using UV-VIS, FT-IR, NMR and GC-MS spectroscopic analysis. Through FT-IR analysis, functional groups such as alcohols, phenols, alkanes, amines, nitro compounds, aromatics, esters, carboxylic acids and alkyl halides were identified in the extract of fresh and boiled shoots while alcohols, phenols, amines and alkynes were identified in the extract of fermented and brine treated shoots. The ^{1}H NMR spectroscopic analysis indicated the increase in alcoholic and acids content in the aqueous extract of fermented and brine treated shoots. In GC-MS analysis 110 compounds were detected in the ethanol extract of fresh and processed shoots with different pharmacological properties. The highest number of compounds (35) was identified in the extract of fermented shoots followed by brine treated shoots (34). The possible presence of important functional groups in the fresh and processed shoots of *D. hamiltonii* as determined by FT-IR, ^{1}H NMR and GC-MS analysis has opened avenues for purification and detection of the compounds for exploitation of shoots for the food and pharmaceutical potentials.

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8. Reference

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