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MASS SPECTROMETRY ANALYSIS OF VOLATILE CONSTITUTENTS OF JACK FRUIT POWDER

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Abstract:
Jackfruit *(Artocarpus heterophyllus)* powder was investigated in terms of its volatile chemical composition through Gas Chromatography – Mass Spectrometry (Scion 436-GC Bruker model coupled with a Triple quadruple mass spectrophotometer) and NIST-MS library. Twenty seven compounds were identified from jackfruit powder. The major jackfruit volatile compounds with significant relative percentage were: 9,12-Octadecadienoic acid (Z,Z)-, Lup-20(29)-en-3-one, 9,19-Cyclo-9β-lanostane-3β,25-diol, 5-Hydroxymethylfurfural and n-Hexadecanoic acid. Major of the compounds belongs to fatty acids, steroids and terpernoids which have various pharmacological activities such as anti-inflammatory, anti allergic, antioxidant, antidiabetic, anti-microbial and many more. This study is an important data for nutraceutical and pharmaceutical industries, which use multiple bioactive compounds for the formulation of drugs and other functional foods.
Keywords: Jackfruit, GC-MS, steroids, fatty acids

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INTRODUCTION:

The plant chemical composition may be divided into three categories. Firstly, primary metabolites compounds like nucleic acids, amino acids and sugars which occur in all cells. Secondly, the high-molecular-weight polymeric likes cellulose, lignin and protein which form the cellular structures. Finally, secondary metabolites compounds those are characteristic of a limited range of species. Mostly, drugs are obtained from pure and best behaviour derivation of secondary metabolite natural products [1].

The *Artocarpus heterophyllus* Lam tree belongs to Moraceae family and commonly known as jackfruit. Jackfruit a wild plant found throughout the tropics, bears the largest known edible fruit (up to 35 kg) [2]. It is cultivated in many parts of the tropics particularly in Southeast Asia i.e., Africa, Australia, Brazil, China, Indonesia, India, Malaysia, Philippines, Srilanka and Thailand. Jackfruit is used in several ways. Young fruits and seeds are used as vegetables. The pulp of ripe is eaten fresh and used in fruit salads. A significant amount of peel (approx. 2,714 – 11,800 kg per tree per year) is discarded as agricultural wastes [3].

The root, leaves, bark, pulp and seeds of jackfruit have been the subject of study for research carried out till date. Literature proves the antibacterial and antioxidant activity of leaf extract [4], polyphenols and antitumor activity of seeds [5] and carotenoid composition in the kernel [6]. However, its peel is almost completely neglected. Therefore, the peel emerges as a potential subject of study. The aim of the present study was to investigate the nutritional and biological potential of the powder prepared by shell of *A. heterophyllus.*

The gross composition of jackfruit, its vitamin content [7-9], water-soluble sugars [10 - 11], starch [12], free sugars and fatty acids [13] have been documented. To date, there is very little information on the mass spectrometry analysis of the jackfruit powder. Hence, this study is an effort to unravel the potential phytochemical composition through GC-MS and its structure and functional activity through clinical literature evidence.

MATERIALS AND METHODS:

Sample Collection and Preparation

The jackfruit powder (*Artocarpus heterophyllus*) was collected from the local market available at Thanjavur, Tamil Nadu. Around 25 g jackfruit powder was soaked in 30 ml of ethanol overnight and then filtered through filter paper. The filtrate is then concentrated through nitrogen gas flushing up to 1ml. The concentrate was again filtered in the Whatmann No.41 filter paper along with 2 g Sodium sulfate to remove the sediments and traces of moisture in the filtrate.

Development of GC-MS Method and Interpretation of Compounds

The chemical composition of jackfruit powder was investigated through Gas Chromatography Mass Spectrometry/ Mass Spectrometry Electron Ionization (GC-MS/EI) mode. The GC-MS/MS is a Scion 436-GC Bruker model coupled with a Triple quadruple mass spectrophotometer with fused silica capillary column BR-5MS (5% Diphenyl / 95% Dimethyl poly siloxane) and Length : 30m; Internal diameter: 0.25mm; Thickness: 0.25µm. Helium gas (99.999%) was used as the carrier gas at a constant flow rate of 1 ml/min and an injection volume of 2 µl was employed (split ratio of 10:1). The column oven temperature program was as follows: 80°C hold for 2 min, Up to 160°C at the rate of 20°C/min-No hold, Up to 280°C at the rate of 5°C / min-No hold, Up to 300°C at the rate of 20°C/min-10 min hold, Injector temperature 280°C and total GC running time was 41 min [14]. This last increase was to clean the column from any residues. The mass spectrometer was operated in the positive electron ionization (EI) mode with ionization energy of 70eV. The solvent delay was 0-3.0 min. A scan interval of 0.5 seconds and fragments from m/z 50 to 500 Da was programmed. The inlet temperature was set at 280°C, source temperature 250°C. The relative percentage amount of each component was calculated by comparing its average peak area to the total areas. Software adopted to handle mass spectra and chromatograms was MS Work station 8. The NIST Version 2.0 library database of National Institute Standard and Technology (NIST) having more than 62,000 patterns was used for identifying the chemical components. The spectrum of the unknown component was compared with the spectrum of the known components stored in the NIST library. The name, molecular weight and structure of the components of the test materials were ascertained. The GC-MS/MS was performed by Food Safety & Quality Testing Laboratory, Institute of crop processing technology, Thanjavur.

RESULTS AND DISCUSSION:

The *ethanol* extract of jackfruit powder was selected for GC-MS/MS analysis due to its ability to dissolve the polar and semi polar bioactive components. The phytochemical composition

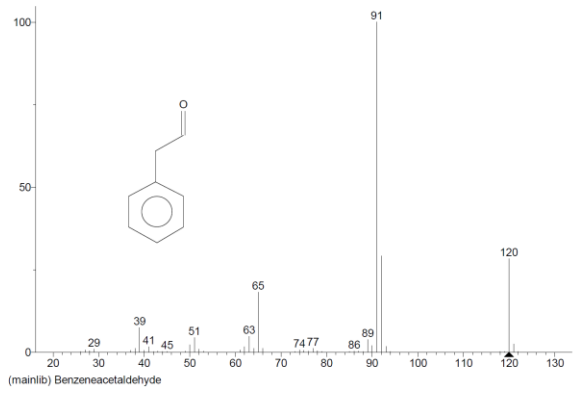
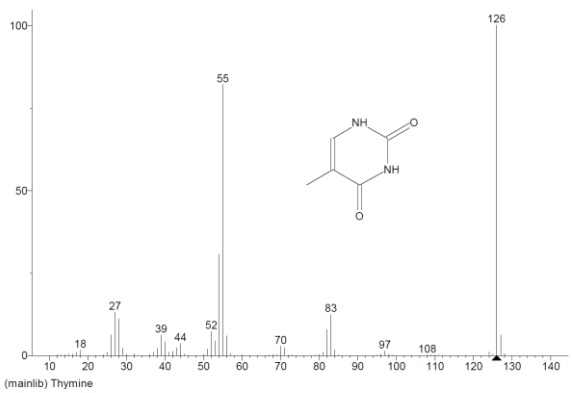
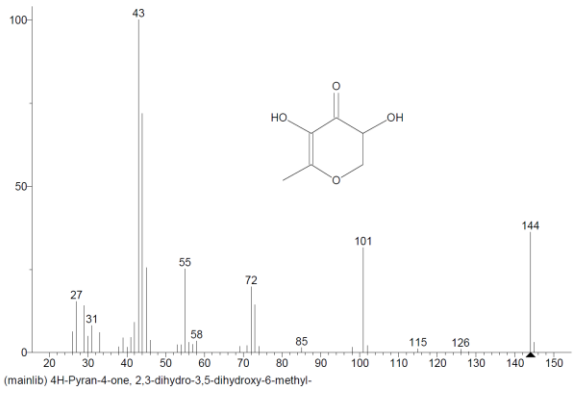
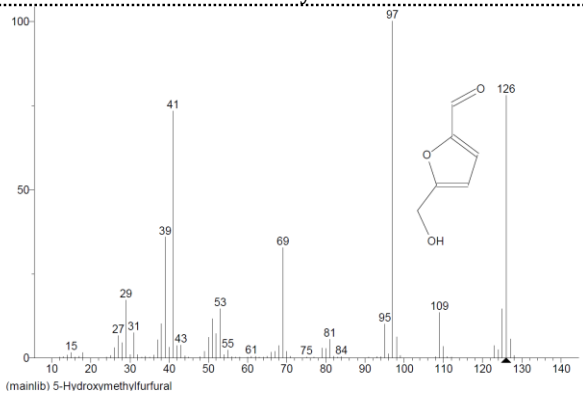
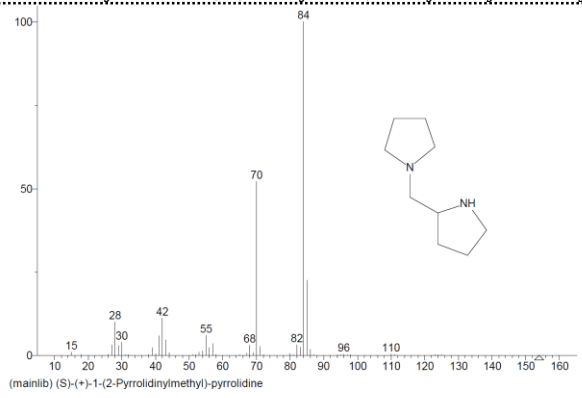
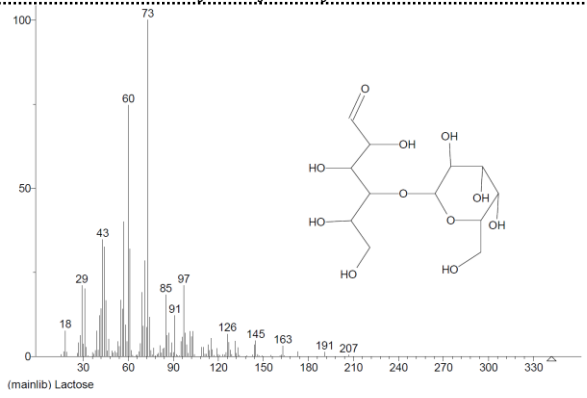
analysis by mass spectrometry is considered as a proper way to convey the pharmacological importance and therapeutic nature of the plant species. It is very clear from the present study, the ethanolic extract of the jackfruit powder contain terpenes, fatty acid, alcohols and steroids. These compounds were clinically proven for its various pharmacological activities such as anti-inflammatory, anti allergic, antioxidant, antidiabetic, anti-microbial and many more. The list of compounds identified through GC-MS/MS was presented in Table 1 and the mass spectra with structure of the chemicals were presented in Table 2.

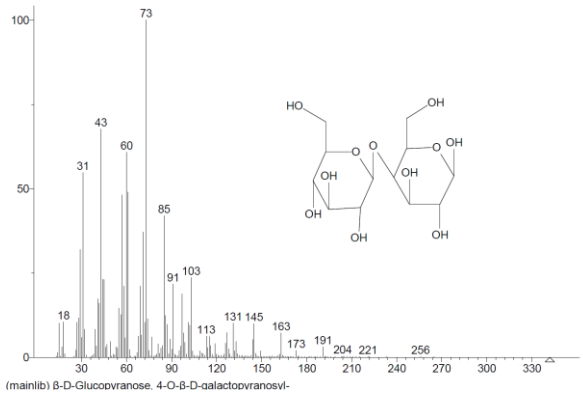
Table 1: Compounds identified through GC-MS/MS with their molecular weight and formulae

No.	RT	Name of the compound	Molecular Formulae	Molecular Weight	Peak Area %
1.	6.05	Benzeneacetaldehyde	C_8H_8O	120	1.22
2.	6.72	Thymine	$C_5H_6N_2O_2$	126	4.47
3.	7.90	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl-	$C_6H_8O_4$	144	2.56
4.	9.27	5-Hydroxymethylfurfural	$C_6H_6O_3$	126	9.91
5.	10.88	(S)-(+)-1-(2-Pyrrolidinylmethyl)-pyrrolidine	$C_9H_{18}N_2$	154	5.52
6.	13.14	Lactose	$C_{12}H_{22}O_{11}$	342	1.03
7.	15.67	β-D-Glucopyranose, 4-O-β-D-galactopyranosyl-	$C_{12}H_{22}O_{11}$	342	0.30
8.	16.74	17-Octadecynoic acid	$C_{18}H_{32}O_2$	280	0.11
9.	20.76	(9Z)-hexadec-9-enoic acid	$C_{16}H_{30}O_2$	254	0.37
10.	21.22	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	256	11.22
11.	24.22	9,12-Octadecadienoic acid (Z,Z)-	$C_{18}H_{32}O_2$	280	7.22
12.	24.33	9,12,15-Octadecatrienoic acid, (Z,Z,Z)-	$C_{18}H_{30}O_2$	278	5.39
13.	26.66	Cholestan-3-ol, 2-methylene-, (3β,5α)-	$C_{28}H_{48}O$	400	0.27
14.	28.54	Obtusifoliol	$C_{30}H_{50}O$	426	0.80
15.	29.29	9,19-Cyclolanost-24-en-3-ol, acetate, (3β)-	$C_{32}H_{52}O_2$	468	1.49
16.	30.31	Glycerol β-palmitate	$C_{19}H_{38}O_4$	330	1.64
17.	31.99	9,19-Cycloergost-24(28)-en-3-ol, 4,14-dimethyl-, (3β,4α,5α)-	$C_{30}H_{50}O$	426	0.01
18.	32.96	9,12-Octadecadienoic acid (Z,Z)-, 2-hydroxy-1-(hydroxymethyl)ethyl ester	$C_{21}H_{38}O_4$	354	1.70
19.	33.07	8,11,14-Eicosatrienoic acid, (Z,Z,Z)-	$C_{20}H_{34}O_2$	306	1.45
20.	34.72	Squalene	$C_{30}H_{50}$	410	7.51
21.	35.98	Androstan-3-one, 17-hydroxy-1,17-dimethyl-, (1α,5α,17β)-	$C_{21}H_{34}O_2$	318	1.08
22.	41.90	Campesterol	$C_{28}H_{48}O$	400	0.48
23.	42.60	Stigmasterol	$C_{29}H_{48}O$	412	0.17
24.	43.23	Lanosterol	$C_{30}H_{50}O$	426	8.58
25.	44.36	γ-Sitosterol	$C_{29}H_{50}O$	414	4.85
26.	46.21	Lup-20(29)-en-3-one	$C_{30}H_{48}O$	424	11.06
27.	46.85	9,19-Cyclo-9β-lanostane-3β,25-diol	$C_{30}H_{52}O_2$	444	9.59

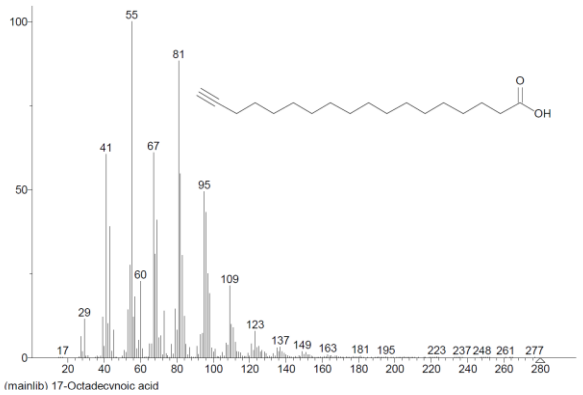
Note: RT- Retention Time, MW- Molecular Weight, MF – Molecular Formulae

Table 2: Mass spectra and structure of the compounds of Jackfruit identified through GC-MS/MS

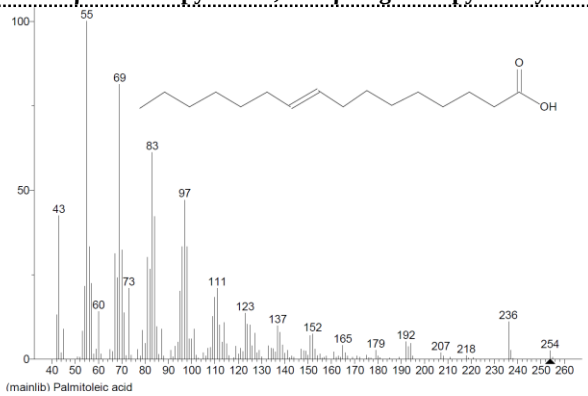
	
Benzene acetaldehyde	**Thymine**
	
4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl-	**5-Hydroxymethylfurfural**
	
(S)-(+)-1-(2-Pyrrolidinylmethyl)-pyrrolidine	**Lactose**



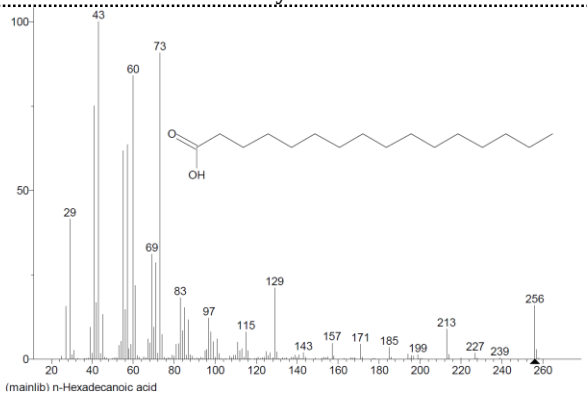
β-D-Glucopyranose, 4-O-β-D-galactopyranosyl-



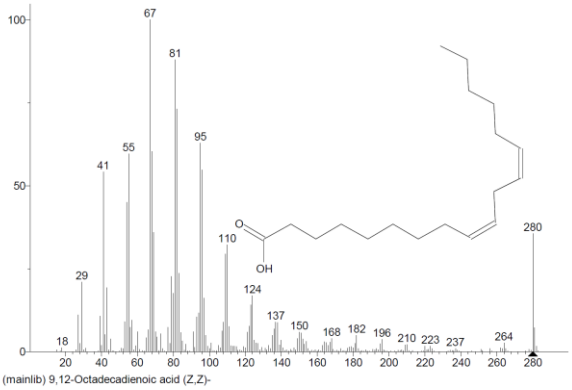
17-Octadecynoic acid



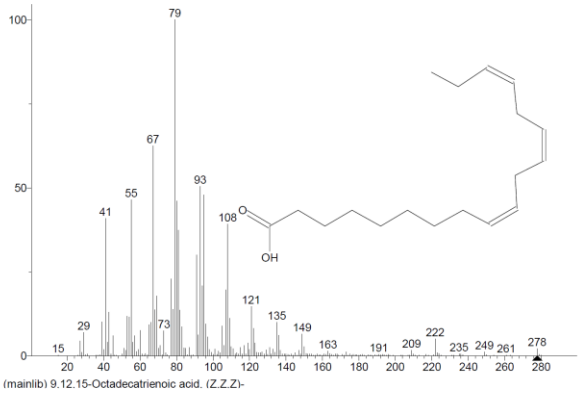
Palmitoleic acid



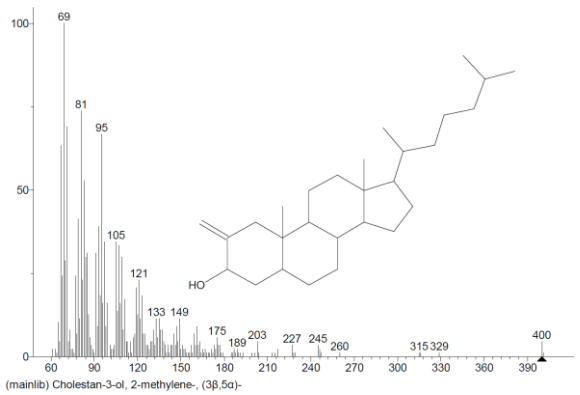
n-Hexadecanoic acid



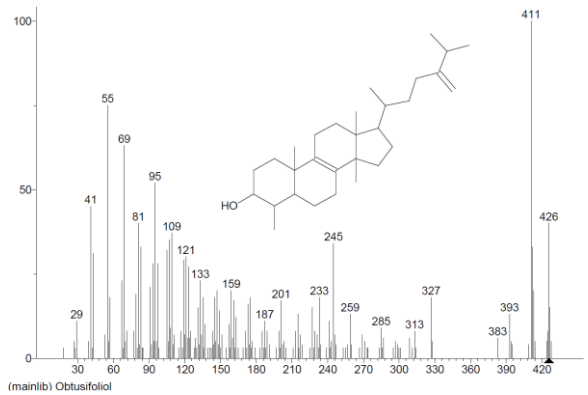
9,12-Octadecadienoic acid (Z,Z)-



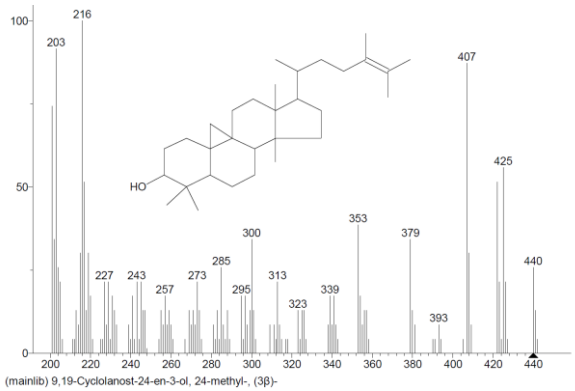
9,12,15-Octadecatrienoic acid, (Z,Z,Z)-



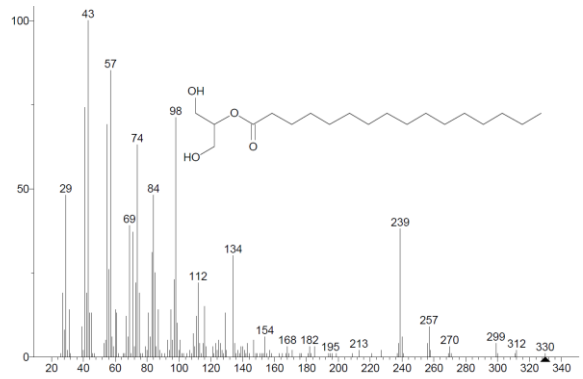
Cholestan-3-ol, 2-methylene-, (3β,5α)-



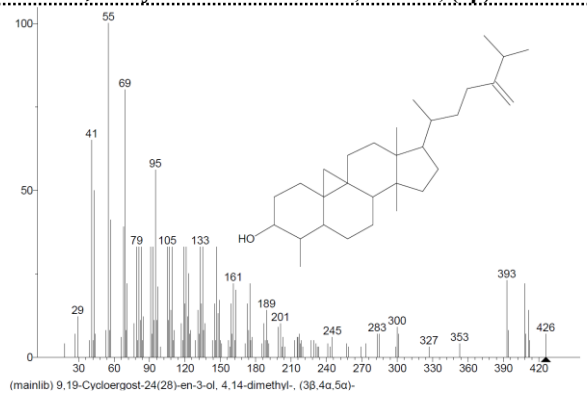
Obtusifoliol



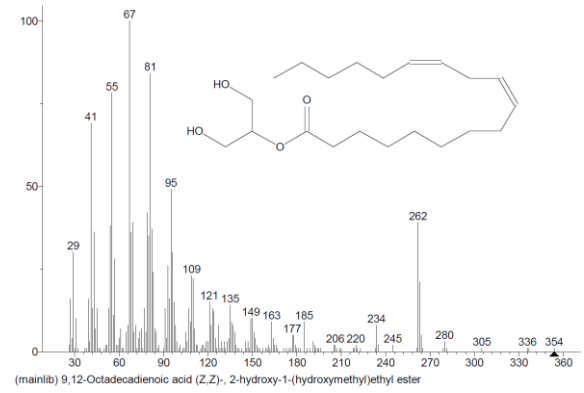
9,19-Cyclolanost-24-en-3-ol, acetate, (3β)-



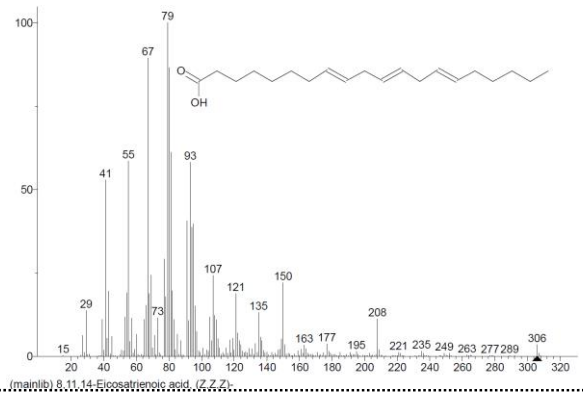
Glycerol β-palmitate



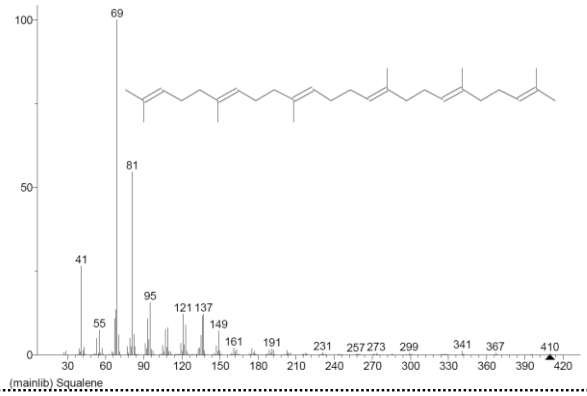
9,19-Cycloergost-24(28)-en-3-ol, 4,14-dimethyl-, (3β,4α,5α)-



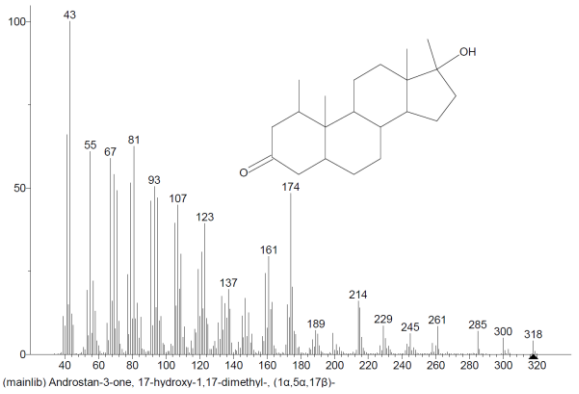
9,12-Octadecadienoic acid (Z,Z)-, 2-hydroxy-1-(hydroxymethyl)ethyl ester



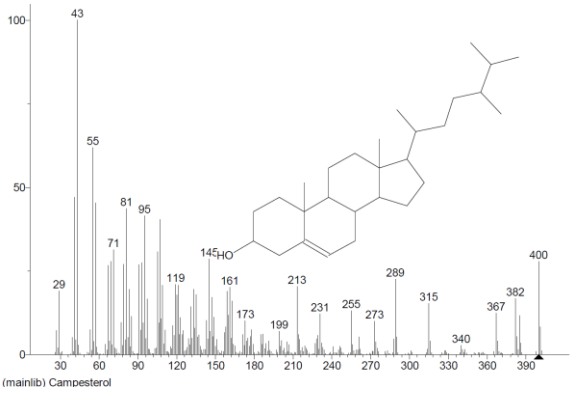
8,11,14-Eicosatrienoic acid, (Z,Z,Z)-



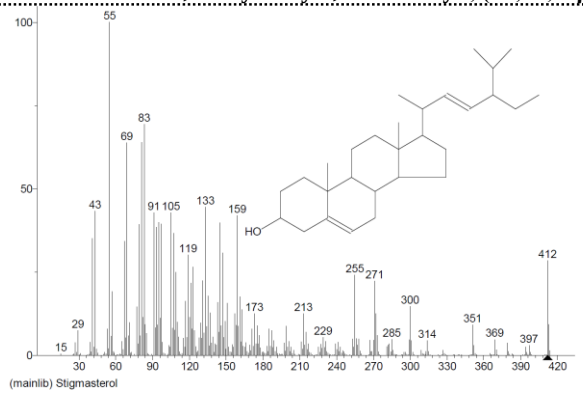
Squalene



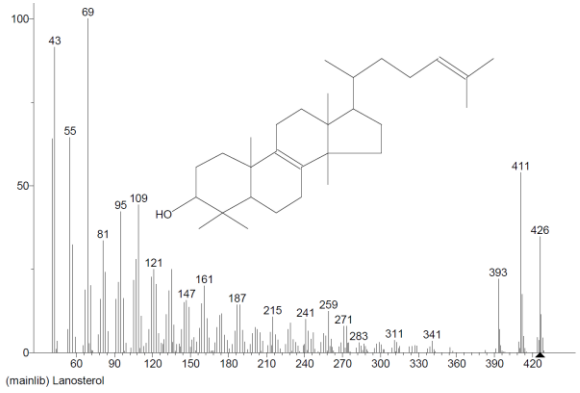
Androstan-3-one, 17-hydroxy-1,17-dimethyl-, (1α,5α,17β)-



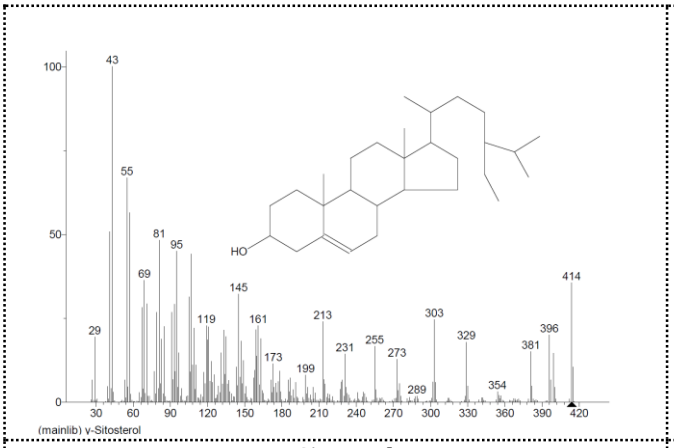
Campesterol



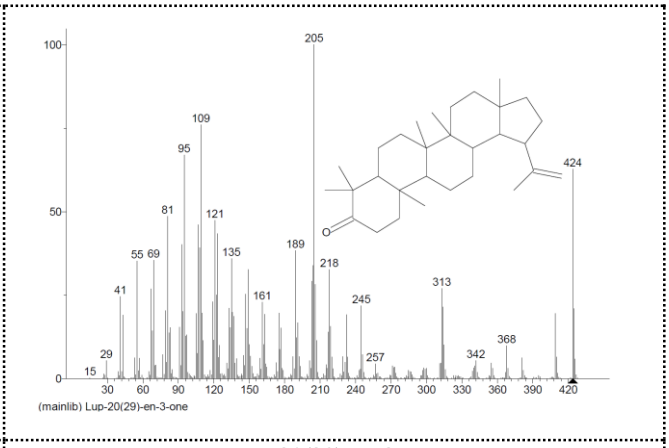
Stigmasterol



Lanosterol



γ-Sitosterol



Lup-20(29)-en-3-one

The identified Phytosterols namely Campesterol, Stigmasterol, Lanosterol, γ-Sitosterol and Cholestan-3-ol, 2-methylene-, (3β,5α)- have been reported for the treatment of diabetic mellitus by lowering fasting blood glucose levels by cortisol inhibition (Devaraj and Jialal 2006). Palmitic acid, linoleic acid, linolenic acid are essential fatty acids found in animals and plants which are primarily used to produce hormone like substances that regulate a wide range of functions, including blood pressure, blood clotting, blood lipid levels, the immune response, and the inflammation response to injury infection (Altieri et al. 2009). Squalene is mostly used in cosmetic industry. β -sitosterol positively influence a diabetic state by directly lowering fasting blood glucose levels by cortisol inhibition (McAnuff et al. 2005). Phytochemical constituents were also studied by Chowdhury et al. 1997 by Gas liquid chromatography and listed capric, myristic, lauric, palmitic, oleic, stearic, linoleic and arachidic acids as major compounds with varying proportions in different parts of the jackfruit.

CONCLUSION:

The ethanolic extract of jackfruit powder has been prepared and analyzed with Gas Chromotography Mass Spectrometry successfully. The results conclude that jackfruit powder contains a lot of potential phytochemicals especially the essential fatty acids and steroids. This study suggests that jackfruit powder has a lot of potential in food, cosmetics, pharmaceuticals and bio-nanotechnology industries. This work throws some light on and helps further research on jackfruit powder and may play a significant role in bio-industrial product development.

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