Evaluation of Phytochemical compounds from Jackfruit Leaves (Artocarpus heterophyllus, Lam.) and its GCMS Analysis

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ABSTRACT

Jackfruit is the most ancient indigenous plant present in the Western Ghats of India and in home gardens of countries like Sri Lanka, Bangladesh, Burma, Malaysia and Brazil. Because of the presence of nutrients in high amount in different parts of the plant, it attracted attention of many researchers. The phytochemical properties of methanol extract of leaves of jackfruit *Artocarpus heterophyllus* Lamk was carried out in the present study. From the phytochemical analysis the presence of bioactive compounds such as alkaloids, aminoacid, carbohydrates, flavonoids, phenol, protein and saponin were confirmed in the sample extract. The GCMS analysis of methanol extract of jackfruit leaves revealed the presence of 30 secondary metabolites. The overall results revelaed that the leaves of the jackfruit is rich in nutrients as well as phytochemicals and secondary metabolites which can be further used to treat various ailments and infection control especially in human health isssues.

Keywords: ailments, alkaloids, jackfruit, phytochemicals, tannins.

INTRODUCTION

Medicinal plants contain organic compounds with specific physiological action on the human body and these bioactive compounds include

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phytochemicals such as tannins, alkaloids, carbohydrates, terpenoids, steroids and flavonoids present in there various parts of the plant tissues (Mann, 1978; Edoga *et al.*, 2005). All these phytochemicals with different chemical classes have an inhibitory effects on all the microbial actions (Cowan, 1999).

Phytochemicals are bioactive compounds in plants which help the body to react to free radicals and The oxygen species. protective role of phytochemicals has been associated with their antioxidant activity. The excess production of reactive oxygen species and reactive nitrogen species (oxidants) in humans cause an imbalance and oxidative damage to large biomolecules such as lipids, DNA and proteins. There effects lead to the pathogenesis and chronic diseases such as Cardio Vascular Disease (CVD), some cancers, ageing and diabetes.

Among different types of medicinal plants, Jackfruit is found to have relatively high phytochemical Composition. Jackfruit has been found to contain high level of proteins, starch, calcium and thiamine (Burkill, 1997). Jackfruit tree is a species of the mulberry family. This tree is native to the tropical and subtropical regions of the world, especially southeast Asia. Popularly known for its large sweet tasty and yellow fruit, this tree has many beneficial parts which include it leaves, seeds, root and latex. The leaf has many therapeutic properties like control of diabetes, antioxidant and anti-aging properties. It has distinctive flavour and fragrance and it is known be rich in vitamin. Jackfruit leaves have several therapeutic effects and work when taken orally on an empty stomach. It helps to improve the glucose tolerance in normal and type-2 diabetes patients. Previous research in jackfruit had more

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focus towards phytochemical analysis on roots, fruit and seeds. So the present study is aimed to analyse the phytochemical constituents of methanol extract of leaves of jackfruit *Artocarpus heterophyllus* Lamk.

MATERIALS AND METHODS

Collection of Sample

The jackfruit leaf samples were collected from K.Vallundanpattu, Thanjavur (DT), Tamil nadu, India.

Phytochemical analysis

The phytochemicals in the methanol extract of leaves of *Artocarpus heterophyllus* Lamk. were determined qualitatively and quantitatively using standard methods (Bhad *et al.*, 2021). To identify the presence of phytochemicals such as alkaloids, carbohydrates, glycosides, saponins, phenolic, compounds and tannins, the following methods were used.

Detection of Alkaloids

Alkaloids were detected using Hager's test (Wagner *et al.*, 1996). Initially,50 mg of solvent-free extract was taken, stired well with few mL of diluted hydrochloric acid and then filtered . To few mL of filtrate, added 2 mL of Hager's reagent (saturated aqueous solution of picric acid). A prominent yellow precipitate indicates the test as positive.

Detection of carbohydrates

Detection of carbohydrates carried out with Molish's test (Ramakrishnan *et al.*, 1994). 100 mg of solvent free extract was taken and dissolved in 5 mL of distilled water and then filtered. To 2 mL of filtrate, added two drops of alcoholic solution and shaked well, then added 1 mL of concentrated sulphuric acid slowly along the sides of the test tube and allowed to stand. Formation of a violet ring indicates the presence of carbohydrates.

Detection of Glycosides

Detection of Glycosides was by using Borntrager's test (Evans, 1997). Fifty mg of the solvent free extract was taken and hydrolysed with concentrated hydrochloric acid for 2 h on a water bath and filtered. To 2 mL of filtrate, added 3 mL of chloroform and shaken well. Chloroform layer gets separated. Then added 10 % ammonia solution to it. Pink colour indicates the presence of glycosides.

Detection of saponins

Detection of saponins was carried out by using Frothing test (Kokate, 1999). Diluted 50 mg of solvent free extract with distilled water and made up to 20 mL. A 2cm layer of foam indicates the presence of saponins.

Detection of Protein

Protein was detected with Biuret test (Gahan, 1984). Dissolved 100 mg of solvent free extract in 10 mL of distilled water and filtered through Whatman No. 1 filter paper. To 2 mL of filtrate, added one drop of 2 % copper sulphate solution. To this added 1 mL of ethanol (95%) followed by excess of potassium hydroxide pellet. Pink colour in the ethanol layer indicates the presence of proteins.

Detection of Amino Acids

Amino Acids were detected with the help of Ninhydrin test (Yasuma and Ichikawa, 1953). Added two drops of ninhydrin solution (10 mg of ninhydrin in 200 mL of acetone) to 2 mL of aqueous filtrate. A characteristic purple colour indicated the presence of amino acids.

Detection of phytosterols

Detection of phytosterols was carried out using Libermann- Burchard test (Finar, 1986). Dissolved 50 mg of solvent free extract in 2 mL of acetic anhydride. To this, added two drops of concentrated sulphuric acid slowly along the sides of the test tube. An array of colour changes showed the presence of phytosterols.

Detection of Fixed Oil and Fats

Saponification test (Kokate, 1999) conducted for the Detection of Fixed Oil and Fats. Added a few drops of 0.5 N alcoholic potassium hydroxide solution to a small quantity of extract along with a drop of phenolphthalein. Then, heated the mixture on water bath for 2 h. Formation of soap or partial neutralization of alkali indicated the presence of fixed oil and fats.

Detection of Phenolic compounds

Phenolic compounds were detected with Lead acetate test. Dissolved 50 mg of solvent free extract in distilled water, and to this, added 3 mL of 10 % lead acetate solution. A bulky white precipitate indicated the presence of phenolic compounds.

Detection of Flavonoid Glycosides

Flavonoid Glycosides were detected following Alkaline reagent test (Raaman, 2006). Dissolved 50 mg of solvent free extract in 5 mL of alcohol and added few fragments of magnesium ribbon and concentrated hydrochloric acid (dropwise). If pink to crimson colour develops, it indicated the presence of flavonol glycosides.

GC-MS Analysis

The GC analysis of the extract was carried out(Muthuelvam, 2019) in Heber Analytical Instrumentation Facility, Bishop Heber College, Tiruchirappalli, Tamil Nadu, India. Column Oven Temperature: 50.0°C. Injection Temperature: 250.00°C. The Injection Mode is split. Injection Volume:1.00. Flow Control Mode Linear Velocity. Pressure:68.1 kPa. Total Flow:16.2 mL/min. Linear Velocity:39.7 cm/sec. Pure Flow:3.0 mL/min. Split Ratio:10.0. EquilibriumnTime:0.5 min [[GC-2010]. Ion Source Temperature:200.00°C. Interface Temperature:250.00°C [GC Program] [GCMS-QP2020].

RESULTS

Bioactive compounds

From the results of phytochemical analysis, it was observed that important medicinal phytochemicals such as alkaloids, aminoacid, carbohydrates, flavonoids, phenol, protein and saponin were present in sample extract. The phytochemicals found in solvents are tabulated in Table 1&2.

Table:1 Qualitative phytochemical analysis of Jackfruit leaf sample

S. No	Phytochemical	Methanol			
	compounds	solvent			
1.	Alkaloids	+			
2.	Aminoacids	+			
3.	Carbohydrates	+			
4.	Fixed oils and Fats	-			
5.	Flavonoids	+			
6.	Glycoside	-			
7.	Phytosterols	+			
8.	Protein	-			
9.	Saponin	+			
10.	Phenols	+			

Table:2 Quantitative phytochemical analysis of Jackfruit leaf sample

S. No	Phytochemical	Quantity			
	compounds	(mg/g)			
1.	Alkaloids	2.16±0.12			
2.	Aminoacid	1.23±0.33			
3.	Carbohydrate	3.00±0.32			
4.	Flavonoids	1.26±0.45			
5.	Phenols	1.00±0.6			
6.	Protein	3.20±0.11			
7.	Saponin	2.20±0.00			

GCMS analysis Analysis of Methanolic extracts

By the GC-MS Analysis of the Methanolic leaf extracts 30 secondary metabolites were identified, namely: 1-Heptanamine, N,Ndimethyl-,4-(1-aminoethyl)-3,3-dimethyl-2azetidinone, hexadecanoic acid, methylester, 9, 12, 15-Octadecatrienoic acid, methyl eter, (Z,Z,Z)-, phytol, Cyclohexasiloxane, dodecamethyl-, silikonfett, Cycloheptasiloxane, tetradecamethyl-, Heptasiloxane, hexadecamethyl-, silicone grease, siliconfett, 1, 1, 1, 3, 5, 7, 9, 9, 9-, nonamethyl pentasiloxane, 3-Ethoxy-1, 1,15,5,5-hexamethyl-3-(trimethyliloxy) triiloxane, siliconeGrease, siliconfett, 2,2,4,4,6,6,8,8,10,10,12,12,14,14,16, 16,18,18,20,20-icosamethyl Cyclodecasiloxane, Cyclononasiloxane, Octadecamethyl-, cyclonona Siloxane, octademethyl-, Cyclononasiloxane, Octadecamethyl-, Carbamazepine-10, 11dihydrodiol,2TMS derivative, silikonfett, 1,1,1,3,5,7,9,9,9nonamethylpentasiloxane, Benzenediol, 2,5-bis silikonfett, 1,4-(1,1)dimethylethyl)-,3-isopropoxy-1,1,1,5,5,5 hexamethyl -3-(trimethylsiloxy)trisil oxane, Ethyl 4,4,6,6,8,8-hexamethyl-11-oxo-3,5,7,9,12-pentaoxa -4,6,8-trisilatetra decan-1-oate, Ethyl 4,4,6,6,8,8hexamethyl-11-oxo-3,5,7,9,12-pentaoxa-4,6,8triilatetradecan-1-oate, Ethylhomovanillate, TMS derivative,heptasiloxane,1,1,3,3,5,5,7,7,9 ,9,11,11 ,13,13-tetradecamethyl-,silikonfett,Ethyl 4,4,6,6,8,hexamethyl-11-oxo-3,5,7,9,12-pentaoxa-4,6,8trisilatetradecan-1-oate,pentasiloxane, 1,1,3,3,5 ,5,7,7,9,9- decamethyl.(Table.3, Fig.1).

DISCUSSION

Plant products in diet was more important to exert protection against infections and maintenance of health. Epidemiological studies indicate that population consuming high levels of plant derived foods have low incidence rate of various diseases. Phytochemicals are naturally occurring biochemical compounds that plants developed, in order to protect themselves from oxidation, insect disease and other hazards in their environment. These phytochemicals give their characteristic colour, flavour, smell and texture. Phytochemicals present in the various parts of plants are responsible for protective effects and it is because of the presence of caretenoids, alkaloids, minerals, vitamins and polyphenols.

Preclinical studies have shown that jackfruit possesses antioxidant, anti-inflammatory, antibacterial, anticariogenic, antifungal, antineoplastic, hypoglycemic, wound healing effects. Phytochemical studies have shown that jackfruit contains useful compounds like the flavonoids, sterols and prenylflavones which may have been responsible for the various pharmacological properties. (Baliga 2011).

Earlier studies on the phytochemical analysis of *Artocarpus heterophyllus* leaves also showed the presence of terpenoid, saponins, flavanoids, tannins, alkaloids, phenol and cardiac glycosides in different type of extracts (Prasad 2014). Parts of the jackfruit plant such as stems, roots, leaves and fruit have medicinal properties. Specifically, jackfruit leaves were reported to contain sapogenins,cycloartenone, cycloartenol, β - sitosterol, and tannin (Trina and Fitmawati, 2014).

Phytochemical tests results by Badriyah (2017) showed that jackfruit and moringa leaves contain various types of secondary metabolites such as phenolics, saponins, flavonoids, tannins, triterpenoids and alkaloids. Jackfruit roots, bark and leaves have relatively high composition of phytochemicals and high antioxidant properties. The distribution of the phytochemicals, also vary in the different tissues. The different parts of the plant can therefore be used as natural antioxidants.

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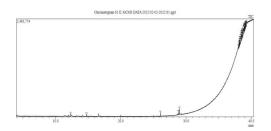
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Peak	Retention time	Start time	End time	M/Z	Area	Area %	Height	Height %	A/h	Mark	Name
1	12.32	12.285	12.405	TIC	237257	2.41	71275	2.05	3.33		1-heptanamine, n,n- dimethyl-
2	14.759	14.715	14.845	TIC	221882	2.25	66410	1.91	3.34		4-(1-aminoethyl)-3,3- dimethyl-2-azetidinone
3	26.067	26.025	26.11	TIC	195658	1.98	96455	2.78	2.03		Hexadecanoic acid, methyl ester
4	28.825	28.78	28.87	TIC	168747	1.71	79236	2.28	2.13		9,12,15-octadecatrienoic acid, methyl ester, (z,z,z)-
5	29.001	28.955	29.055	TIC	365252	3.7	160072	4.61	2.28		Phytol
6	38.045	38.025	38.065	TIC	166227	1.69	75845	2.18	2.19	V	Cyclohexasiloxane, dodecamethyl-
7	38.085	38.065	38.105	TIC	176453	1.79	77490	2.23	2.28	V	Silikonfett
8	38.13	38.105	38.155	TIC	278496	2.82	106985	3.08	2.6	V	Cycloheptasiloxane, tetradecamethyl-
9	38.18	38.155	38.205	TIC	295422	3	104088	3	2.84	V	Heptasiloxane, hexadecamethyl-
10	38.215	38.205	38.23	TIC	153158	1.55	108451	3.12	1.41	V	Silicone grease, siliconfett
11	38.24	38.23	38.26	TIC	191925	1.95	108431	3.12	1.77	V	1,1,1,3,5,7,9,9,9- nonamethylpentasiloxane
12	38.27	38.26	38.28	TIC	142156	1.44	123096	3.54	1.15	V	3-ethoxy-1,1,1,5,5,5- hexamethyl-3- (trimethylsiloxy)trisiloxane
13	38.29	38.28	38.305	TIC	178783	1.81	124373	3.58	1.44	V	Silicone grease, siliconfett
14	38.36	38.305	38.44	TIC	111861 9	11.3 4	149233	4.3	7.5	V	2,2,4,4,6,6,8,8,10,10,12,12,14,1 4,16,16,18,18,20,20- icosamethylcyclodecasiloxan e #
15	38.455	38.44	38.51	TIC	585171	5.93	148928	4.29	3.93	V	Cyclononasiloxane, octadecamethyl-
16	38.52	38.51	38.535	TIC	215967	2.19	148626	4.28	1.45	V	Cyclononasiloxane, octadecamethyl-

Table:3 Secondary metabolites identified from GCMS analysis of methanolic leaf extracts of A.heterophyllus

17	38.547	38.535	38.56	TIC	223216	2.26	159512	4.59	1.4	V	Cyclononasiloxane, octadecamethyl-
18	38.585	38.56	38.63	TIC	618386	6.27	151535	4.36	4.08	V	Carbamazepine-10,11- dihydrodiol,2tms derivative
19	38.645	38.63	38.67	TIC	351757	3.57	154591	4.45	2.28	V	Silikonfett
20	38.68	38.67	38.815	TIC	122523 5	12.4 3	147691	4.25	8.3	V	1,1,1,3,5,7,9,9,9- nonamethylpentasiloxane
21	38.83	38.815	38.845	TIC	250868	2.54	147785	4.25	1.7	V	Silikonfett
22	38.905	38.845	38.925	TIC	614613	6.23	134780	3.88	4.56	V	1,4-benzenediol, 2,5-bis(1,1- dimethylethyl)-
23	38.935	38.925	38.95	TIC	184386	1.87	129399	3.73	1.42	V	3-isopropoxy-1,1,1,5,5,5- hexamethyl-3- (trimethylsiloxy)trisiloxane
24	38.965	38.95	38.985	TIC	263987	2.68	131952	3.8	2	V	Ethyl 4,4,6,6,8,8-hexamethyl- 11-oxo-3,5,7,9,12-pentaoxa- 4,6,8-trisilatetradecan-1-oate
25	38.995	38.985	39.03	TIC	322939	3.27	129492	3.73	2.49	V	Ethyl 4,4,6,6,8,8-hexamethyl- 11-oxo-3,5,7,9,12-pentaoxa- 4,6,8-trisilatetradecan-1-oate
26	39.05	39.03	39.105	TIC	459835	4.66	109770	3.16	4.19	V	Ethyl homovanillate, tms derivative
27	39.119	39.105	39.135	TIC	166846	1.69	102031	2.94	1.64	V	Heptasiloxane, 1,1,3,3,5,5,7,7,9,9,11,11,13,13- tetradecamethyl-
28	39.175	39.135	39.19	TIC	266206	2.7	83627	2.41	3.18	V	Silikonfett
29	39.214	39.19	39.225	TIC	136943	1.39	72915	2.1	1.88	V	Ethyl 4,4,6,6,8,8-hexamethyl- 11-oxo-3,5,7,9,12-pentaoxa- 4,6,8-trisilatetradecan-1-oate
30	39.24	39.225	39.25	TIC	84449	0.86	69198	1.99	1.22	V	Pentasiloxane, 1,1,3,3,5,5,7,7,9,9-decamethyl-

Fig:1 GCMS Chromatogram of Leaf extract of A.heterophyllus



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