Phytochemical Screening and Gc-Ms Analysis of *Garudan Samba* Traditional Rice Variety

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Abstract— *The Garudan Samba, an important Traditional variety of Tamil Nadu was investigated for its phytochemical screening and GCMS study. The brown rice was extracted using ethanol. The results obtained after GCMS studies were confirmed by spectral analysis. The analysis of the ethanol extract showed the presence of n-Hexadecanoic acid, 9,12-Octadecadienoic acid (Z,Z)-, and Oleic acid, as major constituents of the total compounds (76.33%), which have the capacity to prevent many health related disorders. It also contained the medicinally important compounds like Caryophyllene, Ethyl Oleate, Squalene, γ-Tocopherol, Lup-20(29)-en-3-ol, acetate, (3β)-, and phyto sterols like Campesterol, Stigmasterol and β-Sitosterol. Hence, Garudan samba may be considered as one of the important Traditional variety with high level of medicinally important phytoconstituents.*

Keywords— Garudan Samba, Phytochemical Screening, GC-MS.

I. INTRODUCTION

Traditional medicinal plants are often cheaper, locally available and easily consumable, possessing medicinal properties. Nowadays, traditional medicinal practices form an integral part of complementary or alternative medicine. Although their efficacy and mechanisms of action have not been tested scientifically in most cases, these simple medicinal preparations often mediate beneficial responses due to their active chemical constituents (Park and Pezzutto, 2002).

Rice is a major staple food for half of the world's population, providing 80 % of daily calorie intake (Khush, 2005). Carbohydrates, proteins, fatty acids and micronutrients (vitamins and trace minerals) are the major nutrient components in rice. Traditional rice in India are reported to harbor vast amount of medicinal properties and aroma (Das and Oudhia, 2003). These traditional varieties are potent sources for various agronomic traits as well as sources of many bioactive non nutrient components (Crozier *et al*., 2009).

Traditional colored rice varieties are known to be rich in dietary fibre, resistant starch, minerals, carotenoids, flavanoids and polyphenols and consumption of grains of these pigmented rice varieties help in improving human health (Hanhineva *et al*., 2010). The bioactive phytochemicals and micronutrient components from these traditional rice varieties are used as dietary supplements and expected to play a major role in attenuating the incidence of non-communicable diseases viz., cardiovascular diseases, diabetes, cancer and stroke (Vichapong *et al*., 2010).

Garudan samba is a traditional rice variety, white in colour and grown in southern part of Tamil Nadu. The grain has the appearance like the neck of the Brahmin kite (*Garudan*), with a white patch. It is also called "*Kaadai Kazhuththaan*". It is suitable for making a specialty dish called '*Puttu*' of South India. In the present study, the ethanol extract of *Garudan Samba* was evaluated for GCMS analysis.

II. MATERIAL AND METHOD

The *Garudan samba* rice variety was collected from the village of Kuruvadippatti, Thanjavur, Tamil Nadu, India. The paddy was shelled and the brown rice obtained was pulverized and used for phytochemical screening and GC-MS study.

Qualitative Phyto-chemical screening was performed by the standard procedures. (Trease and Evans, 1989). The phytochemicals namely, Alkaloids, Cardiac glycosides, Flavonoids, Phyto sterols, Saponins , Tannin and Terpenoids were tested.

III. PREPARATION OF SAMPLE FOR GC-MS ANALYSIS

Twenty grams of the powdered brown rice sample was soaked in ethanol at 1:2 ratios for 12 h. The extract was then filtered through filter paper and concentrated to 1ml by flushing nitrogen gas into the solution. The extract was then filtered through Whatmann filter paper No. 41 along with 2 g sodium sulfate to remove the sediments and traces of water in the filtrate. 2µl was employed for GCMS analysis (Merlin *et al.,* 2009). The ethanol was selected as an extraction solvent as most of the phytochemicals in *Garudan samba* were found to be polar in nature.

The GC – MS analysis was carried out on a GC Clarus 500 Perkin Elmer, Carrier gas: 1ml per min, Split: 10:1, Detector: Mass detector: Turbomass gold-Perkin Elmer, Software: Turbomass 5.2, Sample injected: 2µl, Column: Elite-5MS (5% Diphenyl / 95% Dimethyl poly siloxane), L × I.D. 30 m × 0.25 mm, df 0.25 µm, Oven temperature Programme: 110° C with 2 min hold ,Up to 200° C at the rate of 10 ° C/min without hold, Up to 280 ° C at the rate of 5° C / min with 9 min hold, Injector temperature 250° C, Total GC running time 36 min, Inlet line temperature 200°C, Source temperature 200°C Electron energy:70 eV, Mass scan (m/z): 45-450,Solvent Delay: 0-2 min, Total MS running time: 36 min (Srinivasan *et al* ., 2014) .

The spectrum obtained in GCMS compounds were compared with the spectrum of known components using NIST library.

IV. RESULTS AND DISCUSSIONS

Screening of *Garudan Samba* brown rice extract for the presence of phytochemicals showed the presence of Alkaloids, Cardiac glycosides, Flavonoids, Phyto sterols, Saponins, Tannin and Terpenoids compounds and the results presented in Table 1.

TABLE 1
PHYTOCHEMICAL SCREENING OF GARUDAN SAMBA BROWN RICE

Phytochemicals	Ethanol extract of *Garudan Samba* Brown Rice
Alkaloids	+
Cardiac Glycosides	+
Flavanoids	+
Phytosterols	+
Saponins	+
Tannins	+
Terpenoids	+

4.1 GC – MS studies

GC–MS chromatogram of the ethanol extract of *Garudan Samba* brown rice extract (Fig. 1) clearly shows 23 peaks, indicating the presence of 23 phytochemical compounds. The identification of the phytochemical compounds was based on the Retention Time (RT), Molecular formula, Molecular weight. The % of Peak area for each compounds was calculated assuming total eluted compounds as cent percent. The details of eluted compounds and their biological activities were given in Table 2.

TABLE 2
COMPOUNDS IDENTIFIED IN THE TRADITIONAL VARIETY GARUDAN SAMBA BROWN RICE

Sr. no.	RT	Name of the compound	Molecular Formula	Molecular Weight	Peak Area %	Biological Activity
1.	4.00	D-Mannopyranose	$C_6H_{12}O_6$	180	0.01	To prevent recurrent urinary tract infections
2.	5.08	5-Hydroxymethylfurfural	$C_6H_6O_3$	126	0.24	Treatment of sickle cell disease
3.	6.22	4-Hydroxy-3-methylacetophenone	$C_9H_{10}O_2$	150	0.16	Antimicrobial Activity
4.	6.74	Phenol, 2-methoxy-4-(1-propenyl)-, (Z)-	$C_{10}H_{12}O_2$	164	0.93	Ingredients of soaps, shampoos and detergents, bath tissue, and topical cosmetic applications.
5.	7.72	Caryophyllene	$C_{15}H_{24}$	204	0.30	Antiinflammatory Activity
6.	8.14	Sucrose	$C_{12}H_{22}O_{11}$	342	0.35	Preservative
7.	8.74	Phenol, 2-methoxy-4-(2-propenyl)-, acetate	$C_{12}H_{14}O_3$	206	0.43	Used in perfumes, flavorings, and essential oils. It is also as a local antiseptic and anaesthetic
8.	9.33	Ethanone, 1-(3,4-dimethoxyphenyl)-	$C_{10}H_{12}O_3$	180	0.16	Precursor to resins**,** raw material for the synthesis of some pharmaceuticals
9.	10.10	Ethyl α-d-glucopyranoside	$C_8H_{16}O_6$	208	1.35	Antituberculous Activity, Antioxidant, Alpha amylase inhibitory activity, Hypolipidimic activity, Anticonvulsant.
10.	11.57	Tetradecanoic acid	$C_{14}H_{28}O_2$	228	0.27	Used in cosmetic and topical medicinal preparations which gives good absorption through the skin.
11.	13.63	Hexadecanoic acid, methyl ester	$C_{17}H_{34}O_2$	270	0.34	Antioxidant, Hypocholesterolemic, Nematicide, Pesticide, Lubricant, Antiandrogenic, Flavor, Hemolytic ,5-Alpha reductase inhibitor.
12.	14.27	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	256	19.07	Used to produce soaps, cosmetics, and release agents
13.	14.59	Hexadecanoic acid, ethyl ester	$C_{18}H_{36}O_2$	284	2.04	Antioxidant, Flavor, Hypocholesterolemic, Nematicide, Pesticide, Lubricant, Antiandrogenic, Hemolytic, 5-Alpha reductase inhibitor .
14.	16.76	9,12-Octadecadienoic acid (Z,Z)-	$C_{18}H_{32}O_2$	280	43.60	Hypocholesterolemic, Nematicide, Antiarthritic, Hepatoprotective,5-Alpha reductase inhibitor, Antihistaminic,Anticoronary,Insectifuge,Antieczemic, Antiacne, Antiandrogenic, Hypocholesterolemic.
15.	16.86	Oleic Acid	$C_{18}H_{34}O_2$	282	13.66	Anticancer Activity
16.	17.05	Ethyl Oleate	$C_{20}H_{38}O_2$	310	6.28	Pharmaceutical drug preparations involving lipophilic substances such as steroids, lubricant and a plasticizer and food additive
17.	22.07	Hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl)ethyl ester	$C_{19}H_{38}O_4$	330	1.87	Hemolytic, pesticide, flavor, antioxidant
18.	26.34	Squalene	$C_{30}H_{50}$	410	0.58	Antibacterial, Antioxidant, Pesticide, Antitumor, Cancer preventive, Immunostimulant, Chemo preventive, Lipoxygenase-inhibitor.
19.	29.90	γ-Tocopherol	$C_{28}H_{48}O_2$	416	0.30	As a Food Additive.
20.	32.27	Lup-20(29)-en-3-ol, acetate, (3β)-	$C_{32}H_{52}O_2$	468	0.66	Antiprotozoal, antimicrobial, antiinflammatory, antitumor and chemopreventive properties.
21.	33.65	Campesterol	$C_{28}H_{48}O$	400	1.83	Anti-oxidant and hypocholesterolemic.
22.	34.25	Stigmasterol	$C_{29}H_{48}O$	412	1.75	Antihepatotoxic, Anti-oxidant, Hypocholesterolemic, Anti-inflammatory, Estrogenic, Antiviral
23.	35.99	β-Sitosterol	$C_{29}H_{50}O$	414	0.05	Anti-oxidant, Analgesic, Anti-inflammatory and Hypocholesterolemic .

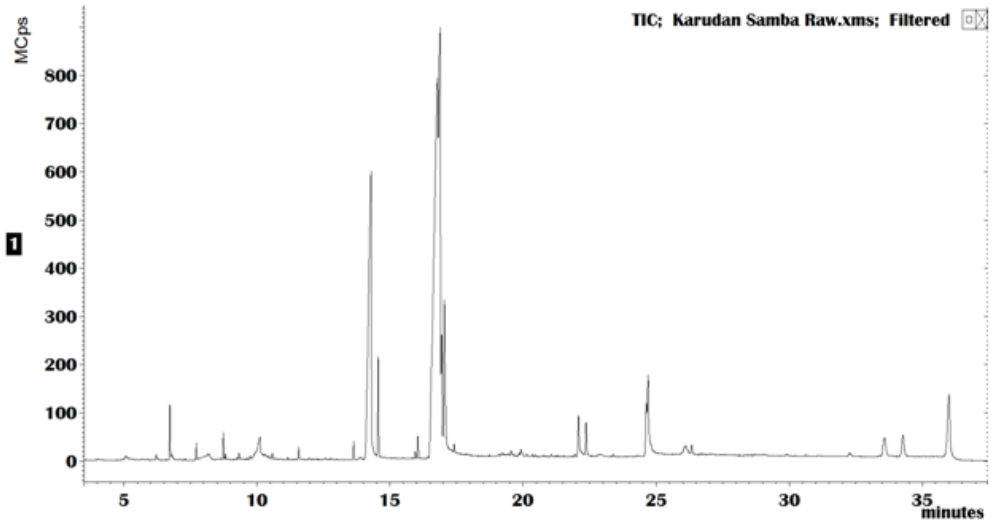

FIG.1.GC-MS CHROMATOGRAM OF ETHANOLIC EXTRACT OF *GARUDAN SAMBA* BROWN RICE

Among the various compounds identified in the extract, 9,12-Octadecadienoic acid (Z,Z)- showed the maximum peak area of 43.60%. This compound has the potential to act as Hypocholesterolemic, Antiarthritic, Hepatoprotective, 5-Alpha reductase inhibitor, Antihistaminic, Anticoronary and Antiandrogenic (Sudha *et al., 2013*). The compounds Oleic acid, Squalene possessing anticancer activity with the peak area of 13.66% and 0.58 % were also observed in the brown rice extract. The compounds like Campesterol (1.83%), Hexadecanoic acid, ethyl ester (2.04%) possessing antioxidant properties were also present. The anti-inflammatory compound Stigmasterol (1.75 %) and Ethyl Oleate (6.28%), used for steroid drug preparation were also observed in the brown rice extract. These phytochemicals in brown rice of *Garudan samba*, might be the reason that traditionally it is advised to consume this rice.

This indicates that the presence of above compounds may prevent many health related disorders such as Cancer, Heart and many other diseases. The presence of other minor phytochemicals also justifies the use of this rice for various ailments. The phytochemicals present in *Garudan samba* shows the potential to use this variety as a highly valuable variety, possessing various important medicinal properties.

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