Seed Oil Extraction and Phytochemical Analysis of *Putranjiva roxburghii* Wall. (Putranjivaceae)

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ABSTRACT

Medication using herbs have been played a significant role in human civilization. The present investigation deals with the extraction of oil from *Putranjiva roxburghii* seeds using Soxhlet Extraction Method and profile chemical components by GC-MS method. Seed oil was analyzed by GC-MS, revealed the presence of seven bioactive compounds. 9-Octadecenoic acid and Ethyl Oleate were identified as major compounds. Identified compounds exhibit following bioactivity such as Antibacterial, Antioxidant, Antiandrogenic, Antidiabetes, Antiarthritic, Antieczemic, Antiacne, Alphareductase inhibitor, Autoimmune, Cancer, Inflammatory diseases, high blood pressure facilitate wound healing, Nematicide, Anticoronary, Hemolytic, 5-Alpha reductase inhibitor, Hypocholesterolemic, obesity, Insectifuge, Pesticide, Lubricant, Flavor and Plasticizer etc. Hence, *P. roxburghii* seed oil recommended as the potent bioactive reservoir.

Key Words: Putranjivaceae, *Putranjiva roxburghii*, Soxhlet extraction method, Seed oil, GC-MS analysis, Chhattisgarh.

INTRODUCTION

Plants have been used for medication from ancient time. Rural population for centuries have availed medicinal plants [1]. Among various plant parts, seeds are also valuable for therapeutic use. Proteins, carbohydrates and lipids are the major components that are present in the seeds. They may be as wax, fat or oil [2], [3]. Among these, oil is indispensable for seed germination because it doubles the supply of energy needed for the germination process when compared to proteins and carbohydrates (Salunkhe *et. al.*, 1992).

Oils can be derived from any part of the plant and have been used both as culinary and as therapeutic. The refined oils extracted from plants are safe for oral consumption and their general biology is well categorized due to wide use in food materials. It is also very important for cosmetics due to the presence of triglycerides containing fatty acids and fatty acid derivatives (Burnett *et. al*., 2017). The percentage of chemical constituents in seed oil plant depends on its locality and genetics. Its nutritional content also varies according to its oil type (Baud *et. al*., 2010). On the other hand, Vegetable Oil and Hydrogenated Vegetable Oil also play an important role in cosmetics (Belsito *et. al*., 2011). The lipid content was determined by extraction using solvents. It can also be determined by non-solvent wet extraction methods and by instrumental methods that rely on the physical and chemical properties of lipids.

The oil extracted from the seeds of *Putranjiva roxbugrhii* Wall. contains isopropyl and 2-butylisothio-cyanates as the main constituents and 2-methy-butyl isothiocyanate as a minor component. The oil of *P. roxburghii* seed is not commercially available. The seeds yield fatty oil, useful heating and cooking (Krishnamurthi *et. al*., 1948). Nag *et. al*., (1995) reported that oil can be used for the production of factice and Tripathi *et. al*., (2007) reported its usage for herbal preservative for peanuts during storage.

Fig.1 Seeds of *Putranjiva roxburghii*

Fig.2 *Putranjiva roxburghii* Wall.

MATERIALS AND METHODS

Collection and Authentication

The plant was collected from Kunkuri in Jashpur district of Chhattisgarh, India. Identification and authentication was done by Dr. S. John Britto, Director and Head of The Rapinat Herbarium and Centre for Molecular Systematics St. Joseph's College (Autonomous) Tiruchirappalli, India. The voucher specimen was deposited at the centre with accession number RHT67530. **(Fig.1 and Fig.2)**

Extraction of Oil

The air-dried seeds were powdered and thoroughly extracted using Soxhlet extraction to yield oil. Powdered sample was mixed with sodium sulphate in the ratio 4:1. Sodium sulphate absorbs the water or moisture content present in the sample. The solvent used in this experiment was hexane with boiling point 50ºC-70°C. 150 ml of Hexane was taken in 250 ml in a round bottomed flask and was kept for heating. The evaporation and extraction of oil took few hours. Then round-bottomed flask was removed and it was placed in the pre-heated water bath. After complete evaporation of the solvents oil was collected and fat content was calculated.

$$\text{Fat content} = \frac{\text{Difference between the weight of flask before addition of sample and after concentration of sample}}{\text{Original weight of sample}}$$

Gas Chromatography-Mass Spectrometry (GC-MS) Analysis

Gas Chromatography- Mass Spectrometry (GC-MS) is an analytical method of combination of Gas-Chromatography (GC) and Mass Spectrometry (MS). It provides a multidimensional compound identification and quantification of the sample (Skoog *et. al.*, 2007). Helium or un-reactive gas such as nitrogen act as mobile phase usually and microscopic layer of liquid or polymer inside glass or metal tube, as stationary phase is known as column. Mass Spectrometry (MS) is the detector for GC. GC separates volatile and semi-volatile compounds with great resolution, but it is incapable to identify them. MS can give information related to compound's structure which leads to its identification but cannot separate them (Hussain *et. al.*, 2014).

RESULTS AND DISCUSSION

Research on seed oil has gained great importance due of extensive demands for both human consumption and as well as industrial applications. Extracted seed oil showed 54% of fat content. GC-MS analysis of showed seven bioactive compounds such as Palmitic acid, Hexadecanoic acid, ethyl ester, 9-

Octadecenoic acid, Linoleic acid ethyl ester, Ethyl Oleate, Octadecanoic acid, ethyl ester and Bis(2-ethylhexyl) phthalate. 9-Octadecenoic acid and Ethyl Oleate were identified as major compounds. (**Table. 1 and Fig.3**)

9-Octadecenoic acid is also known as Oleic acid. It is a fatty acid, which naturally occurs in various animal and vegetable fats and oils. Normally it is an odourless and colorless oil, its commercial samples may be yellowish in color. Thomas *et. al.*, (2000) reported its chemical classification as a monounsaturated omega-9 fatty acid. The term "oleic" means related to, or derived from, olive oil which is mostly composed of oleic acid. Oleic acid is emitted by the decaying corpses of a number of insects, including bees. Oleic acid is used in foods in the form of triglycerides, which is a part of human diet as animal fat and vegetable oils. It is also used as emulsifying agent, emollient (Carrasco *et. al.*, 2005) or moisturizer, excipient in pharmaceuticals and solubilizing agent in aerosol product (Smolinske *et. al.*, 1992).

Ethyl oleate is a fatty acid ester, which is colorless to light yellow and can be formed by the condensation of oleic acid and ethanol. It is produced by the body during ethanol intoxication (Dan *et. al.*, 1997). It is used as a solvent in drug preparations (Ory *et. al.*, 1983), in addition its usefulness as lubricant, plasticizer and food additive. It has been identified as a primer pheromone in honeybees (Leoncini *et. al.*, 2004).

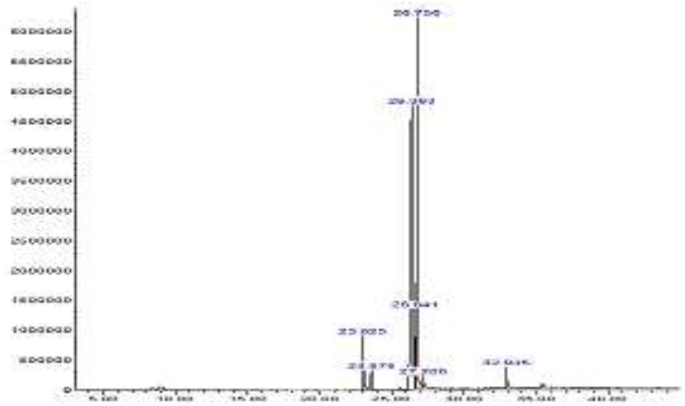
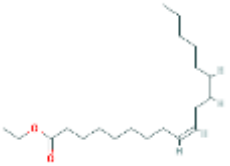
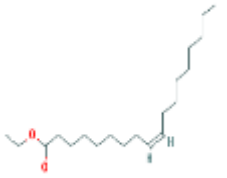
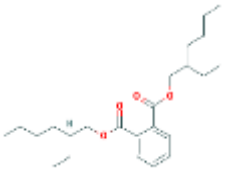


Fig 3: Gas Chromatogram of Seed oil

Table.1 GC-MS profile of Seed Oil

Name	Mol. Formula	Mol. Wt.	Rt	Area %	Structure	Activity
Palmitic acid	$C_{16}H_{32}O_2$	256	23.02	4.90		Antioxidant, Hypocholesterolemic, Nematicide, Pesticide, Lubricant, Antiandrogenic, Flavor, Hemolytic, 5-Alpha reductase inhibitor
Hexadecanoic acid, ethyl ester	$C_{18}H_{36}O_2$	284	23.6	1.32		Antioxidant, Hypocholesterolemic, Nematicide, Pesticide, Antiandrogenic flavor, Hemolytic, Alphareductase inhibitor

9-Octadecenoic acid	$C_{18}H_{34}O_2$	282	26.4	46.93		Cancer, Autoimmune and Inflammatory diseases, Antidiabetes, Obesity, High blood pressure facilitate wound healing
Linoleic acid ethyl ester	$C_{20}H_{36}O_2$	306	26.6	6.62		Hypocholesterolemic, Nematicide, Antiarthritic, Hepatoprotective Antiandrogenic, Hypocholesterolemic, 5-Alpha reductaseinhibitor Antihistaminic, Anticoronary, Insectifuge, Antieczemic, Antiacne
Ethyl Oleate	$C_{20}H_{38}O_2$	310	26.8	36.95		Antibacterial
Octadecanoic acid, ethyl ester	$C_{20}H_{40}O_2$	312	27.2	1.05		No activity recorded
Bis(2-ethylhexyl) phthalate	$C_{24}H_{38}O_4$	390	32.9	2.23		Plasticizer

CONCLUSION

GC-MS analysis *P. roxbugrhii* seed oil revealed the presence of identify the phyto constituents and they are anticancerous, antimicrobial, antioxidant, antiacne and various other properties provides a potential source of industrial application. Major compounds are 9-Octadecenoic acid and Ethyl Oleate and were reported for following properties such as Anticancer, Autoimmunity, Inflammatory diseases, Antidiabetes, Obesity, High blood pressure facilitate wound healing and Antibacterial. Hence, the biological values of *P. roxbugrhii* seed oil contain pharmacological active compounds that may enhance its use as a traditional drug. Based on these report further compound isolation can be done and novel therapeutic herbal medicine can be design.

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