Physico-chemical, Fatty Acid Profile and Mineral Nutrients of *Diplocyclos palmatus* (L.) C. Jeffery (Shivlingi) Seeds

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Abstract –

Diplocyclos palmatus (L.) C. JeffreySyn. *Bryonia laciniosa* ((L.) is commonly known inChhattisgarh as 'Shivlingi' due to its seed resemblance to 'Shivling', which is abstract or symbolic representation of the Hindu God 'Lord Shiva'. Traditionally *Diplocyclos palmatus (*L.) C. Jeffery has been used for treatment of various ailment since generations. These seeds have been found to be rich in oil content, high calorific value and high protein value. Proximate analysis of full fat seed reveals Moisture – 4.15%, Ash - 3.81%, Volatile Matter-80.34%, Fixed carbon-11.70% and Ultimate analysis reveals N – 2.31%, C- 60.03%, H
– 8.759%, S - 0.16%. Atomic absorption analysis offull fat seed powder of seeds indicated the presenceof Na^{+}, K^{+}, Ca^{++}, Mg^{++}, Fe^{+++}, Co^{++}, Ni^{++}, $Cu^{++,}$ Zn^{++}, Pb^{++} cations. Fatty acid composition of seed oil of *Diplocyclos palmatus* (L.) C. Jeffery by GC-FID reveals presence of Palmitic Acid (C16:0) - 28.78%, γ -Linolenic Acid (C18:3n6) - 17%, Linoleic Acid (C18:2n6c) - 16.47%, Oleic Acid (C18:1n9c) - 16.26%, Stearic Acid (C18:0) – 8.20%, Behenic Acid (C22:0) – 4.50%, Nervoinic Acid (C24:1n9) –2.97%. Results reveal that *Dipliocyclos palmatus* (L.) C. Jeffery may be a rich source of nutritional and medicinal properties due to their phytochemicalconstituents.

Key Words: Physico-chemical, Proximate-Ultimate Analysis, Atomic Absorption Spectroscopy, Fatty Acid Composition, *Diplocyclos palmatus* Seed.

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I. INTRODUCTION:

Diplocyclos palmatus (L.) C. Jeffrey Syn *Bryoniopsis laciniosa* (L.) [1] which belongs to the family Cucurbitaceae is a herb that is distributed throughout India and is known as Shivlingi [2], Indian bryony, Lollipop climber, Vaduballi, Gargumaru etc. The seeds of *Diplocyclos palmatus* (L.) C. Jeffery are known as 'Shivlingi' because the upper surface of seeds has a marking and morphology [3], which is abstract or symbolic representation of the Hindu God 'Lord Shiva'. It is an annual climber with white red fruits and is reported to be of high in medicinal value [4] including treatment for infertility. The WHO has estimated that the infertility affects millions of people of reproductive age worldwide, between 48 million couples and 186 million individuals live with infertility globally. [5,6,7]

Medicinal Properties

D. palmatus seed is studied and known to have various medicinal properties, such as: Fertility booster, Uterine tonic, Aphrodisiac, Spermatogenic. As Anodyne: Anti-diabetic, Anti-fungal, Antihyperlipidemic, Anti-inflammatory, Antimicrobial, Antioxidant, Antipyretic and Carminative. [8] The whole plant has been used to treat several diseases, such as asthama [9], fever [10], inflammations [11,12] and various skin conditions [13-15]. Bryonin [16,17], non-ionoc glucomannan [18], goniothalamin [19] and punicic acid [20] are the main active constituents of the

plants. Therefore, this study investigated the chemical composition, physical properties and nutritional value of *D. palmatus* seeds.

II. METHODS AND MATERIAL REQUIRED:

A. Seed selection and sampling

Diplocyclos palmatus (L.) C. Jeffery seeds were collected in November to December month from Eastern Chhattisgarh. Remove the seeds from fruit pulp. Dry the seeds in shade at room temperature, crushed it by using mortar-pestle and store in airtight bottles at 4°C. After crushing of seeds Extraction and Proximate – Ultimate analysis performed from original and defatted seed.

B. Extraction

Oil from the shade dried seeds powder of *D. palmatus* was extracted by using Soxhlet Apparatus.The seeds of *D. palmatus* were exhausted with petroleum ether (Boiling point 60-80°C). [21-23] A brown colour oil is obtained in 29% yield. The chemicals used are AR grade and Sigma chemical company.

C. Proximate and Ultimate Analysis

Proximate chemical composition of the seed and physico-chemical characteristics of the extracted oil were determined by standard methods. Calorie contents were determined using Julius and Peter`s Bomb calorimeter.

1) Proximate Analysis – The proximate analysis determines only the Fix Carbon, Volatile matter, Moisture, Ash percentage, Crude Protein, Fiber etc.[24, 25]

2) Ultimate Analysis – The ultimate analysis determines all seed components elements like C, H,N, S. [24, 25]

D. Total fat and fatty acid analysis: [30]

Fatty acids were analysed by AOAC (2001. 996.06) methods. Using 0.5 M methonolic KOH to form fatty acid methyl esters (FAME) the isolated fat was trans-esterified. Fatty acids were analysed by Gas Chromatograph (7890B of Agilent Technologies) equipped with flame ionization detector and Agilent

- DBFFAP column (nitroterephthalic-acid-modified polyethylene glycol (PEG) of high polarity for the analysis of volatile fatty acids). At initial temperature of 100°C for 5 min the temperature of the column was maintained, raised to 240°C at the rate of 4°C /min. As carrier gas Nitrogen was used at a column flow rate of 1.0 ml/min. At 280°C

RESULT AND DISCUSSION**: Table 1: Characteristics of Seed and Oil of *Diplocyclos palmatus* (L.) C. Jeffery [26-28]**

Sl No	Sample	Properties	Value
1	Seed	Moisture % in seeds	4.15%
2		Oil %	29.00%
3		Protein %	14.43%
4		Ash %	3.81%
5		VM %	80.34%
6		FC %	11.70%
7		Calorie content calorie/gm	6004.37 cal/gm
8	Oil	Colour	Brown
9		Total Fat	145 ml
10		Refractive index	1.48
11		Specific gravity oil at 25^{O}C	0.884gm/ml
12		Acid Value	3.3
13		Saponification Value	204.3
14		Ester Value	201
15		Free Fatty Acid	1.6
16		Iodine Value	170
17		Unsaponifiable Matter	1.20%

A. Physico-chemical properties.

The physico-chemical properties of *D. palmatus* seed oil in Table 1 shows the qualitative property of seed oil. The moisture content of seeds was 4.15% which is low and therefore beneficial for prolonging the shelf life of the seeds. The seeds contained significant amounts of volatile matter - 80.31%, protein - 14.43%, ash - 3.81%. The refractive index of oils depends on their molecular weight, fatty acid chain length, degree of unsaturation, and degree of conjugation. The refractive index of
D. palmatus is 1.48. The refractive index is positively related to iodine value, which is a measure of the degree of unsaturation of oils and gives an idea of their oxidative stability. The iodine value 170.2 of I_2/100gm of oil, characteristic of the presence of unsaturation.

D. palmatus seeds oil had a saponification value 204.3. This value is due to high content of medium chain fatty acids (i.e., C16 and C18). The concentration of free fatty acids and the acid value of *D. palmatus* is 1.6 and 3.3 mg of KOH/g of oil. These low values were a result of lower hydrolysis of triglycerides and signified that the oil could have long shelf life. Image of *D. palmatus* is given in Fig.1

Fig.1: Image of *Diplocyclos palmatus* (L.) C. Jeffery (Shivlingi) seeds

B. Ultimate Analysis of seed and seed ash

The Ultimate analysis was carried for both original and defatted *D. palmatus* seeds using standard method. The result of these analysis is tabulated in Table 2.

Table 2: Ultimate analysis of *D. palmatus* seeds (%)

	Carbon	Hydrogen	Nitrogen	Sulphur
Original seeds	60.03	10.439	2.31	0.16
Defatted seeds	50.76	9.917	3.58	0.23

C. Atomic Absorption Spectroscopy

The ash of the seeds was subjected to analysis [29] of cations present in them using flame photometer for Na, K, Ca ions and atomic absorption spectrophotometer for other cations. Working condition for determination of cations analysis of seed ash are tabulated in Tables 3.

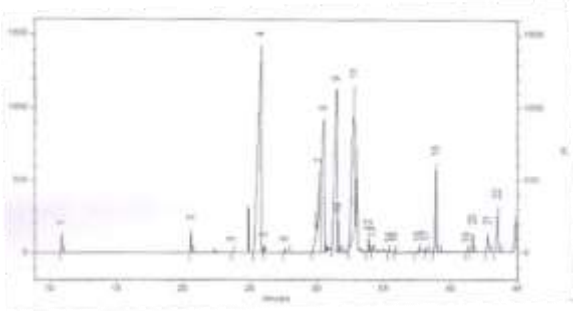
Name	Retention Time	Area	Area %
Capric Acid Methyl Ester (C10:0)	10.936	3579940	0.55%
Myristic Acid Methyl Ester (C14:0)	20.619	5570406	0.86%
cis-10-Pentadecenoic Acid Methyl Ester (C15:1)	23.673	326150	0.05%
Palmitic Acid Methyl Ester (C16:0)	25.875	186077143	28.7
Palmitoleic Acid Methyl Ester (C16:1)	26.107	948798	
Heptadecanoic Acid Methyl Ester (C17:0)	27.614	1068370	
Stearic Acid Methyl Ester (C18:0)	30.246	5302057	
Oleic Acid Methyl Ester (C18:1n9c)	30.591	10512	
Linoleic Acid Methyl Ester (C18:2n6c)	31.529	10	
Linolelaidic Acid Methyl Ester (C18:2n6t)	31.618		
γ-Linolenic Acid Methyl Ester (C18:3n6)	32.853		
α-Linolenic Acid Methyl Ester (C18:3n3)	33		
Arachidic Acid Methyl Ester (C20:0)			
cis-11-Eicosenoic Acid Methyl Ester (C20:1n9)			
cis-11,14-Eicosadinoic Acid Met Ester (C20:2)			
cis-5,8,11,14,17-Eicosapenta Acid Methyl Ester			
Heneicosanoic Acid Met (C21:0)			
Behenic Acid Methyl			
cis-13,16-Docos Methyl Es			
c4,7,10,13,16,1 Acid			
Lignocer			
Nerv			

Table 3: Working condition for determination of cations analysis of seed ash.

S. N.	Test Parameter	Measurement Unit	Test Method	Test Result
I.	**Chemical Testing**			
	Metals			
1	Sodium (as Na)	mg/kg	Flame Photometer	423.08
2	Potassium (as K)	mg/kg	Flame Photometer	7730.76
3	Calcium (as Ca)	mg/kg	Flame Photometer	5423.08
4	Magnesium (as Mg)	mg/kg	AAS	3807.69

5	Iron (as Fe)	mg/kg	AAS	62.5
6	Cobalt (as Co)	mg/kg	AAS	0.96
7	Nickel (as Ni)	mg/kg	AAS	7.69
8	Copper (as Cu)	mg/kg	AAS	9.61
9	Zinc (as Zn)	mg/kg	AAS	48.08
10	Lead (as Pb)	mg/kg	AAS	Absent

D. *palmatus* seeds also contained significant amounts of minerals. The most abundant was K^{+} followed by Ca^{++}, Mg^{++}, Na^{+}, Fe^{+++}, Zn^{++}, Cu^{++}, Ni^{++}, Co^{++}. By Ultimate analysis Carbon is in highest % followed by Hydrogen, Nitrogen, Sulphur. These elements except Lead and Nickel are required in very small amounts (in micrograms) by the human body and animal. However, lead and nickel cause toxic effects.



Fatty acid composition of seed oil of *D. palmatus* by GC-FID reveals quantitatively presence of highest % of Palmitic Acid - 28.78% followed by γ-Linolenic Acid – 17%, Linoleic Acid - 16.47%, Oleic Acid -16.26%, Stearic Acid - 8.20%, Behenic Acid - 4.50%, Nervoinic Acid - 2.97%, Lignoceric Acid -1.04%, α- Linolenic Acid - 0.52%. Palmitic Acid is most abundant saturated fatty acid in our human body because 20-30% of the phospholipid of each cell membrane are made of palmitic acid to enable its proper functioning. γ-Linolenic Acid (GLA) for eczema, asthma, arthritis, high blood pressure, nerve pain related to diabetes. Oleic Acid is most commonly used for preventing heart disease and reducing cholesterol. Stearic Acid is used for softens and smooths the skin's surface while also helping to maintain the skin barrier, Behenic Acid - to give hair conditioner and moisturizers their smoothing properties, Nervoinic Acid - regulate the function of brain cell membranes and have a neuroprotective effect. *D. palmatus* seeds are rich in Saturated Fatty Acid Palmitic, Stearic, Behenoic acid (SFA- 41.48%) as well as Unsaturated Fatty acid, Oleic acid, Nervonic acid (MUFA-19.23 %) and Linoleic acid, Omega fatty acid such as α-Linolenic acid and γ-Linolenic acid (PUFA-33.99%).

III. CONCLUSION:

The phytochemical study on the chemical composition, physicochemical properties, and nutritional value of *Diplocyclos palmatus* (L.) C. Jeffery seeds suggests that these seeds could be considered as an alternative source of oil, protein, and micronutrients. The oil 29% is higher compared to the G. R. Dave *et. al* (12%) [31] and G. Gowirkumar *et.al* (23%) [32] and protein % is less than that 14.43%. The oil % is less than palm seed oil. The oil obtained from the *D. palmatus* seeds have the potential phytochemical constituents and could be used as food supplement and pharmaceutical industries.

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