

Isolation and Identification of Lipids from Different Parts of Fluted Pumpkin Seeds

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Abstract: Lipid content and composition of different parts of fluted pumpkin seeds, were investigated. The percentage yield of oil extracts from the samples were $(29.5-37. \pm 0.3)$ and $(0.5-25 \pm 0.1)$ for the seeds and seed coats, respectively. The following lipids were identified; phospholipids, glycolipids, sterols, fatty acids and neutral lipids. These lipid classes were isolated and identified using different organic extractants and thin-layer chromatography, respectively. The various RF values of the sample were matrix matched with the standard RF values of known lipids.

Key words: Fluted pumpkin seed, phospholipid, glycolipid, neutral lipid, lecithin, raw material

INTRODUCTION

Fluted pumpkin seed (*Telfaria occidentalis*) is a dioecious perennial vine plant. It is of the family cucurbitaceae and a species of *telfaria occidentalis* hook^[1]. Fluted pumpkin is a vegetable crop and one of the local vegetable in the tropics^[2]. Fluted pumpkin is cultivated in many parts of West Africa. It is a drought tolerant vine plant, favoured by rainfall of sufficient amount^[1,3,4]. Fluted pumpkin grows best at a low altitude under medium to higher rainfall, best on sand, well-drained soil^[4]. The fluted pumpkin seeds, leaves and roots are edible, use as herbal medicine and soup thickeners^[3,5]. The seeds of fluted pumpkin are known to contain 47% oil, 31% crude protein, substantial quantities of vitamins, minerals, carbohydrate^[2,5].

We, therefore, report the isolation and identification of lipids from different parts of fluted pumpkin seeds.

MATERIALS AND METHODS

Sample collection: Fluted pumpkin pods were harvested from University of Nigeria, NSUKKA, demonstration farm, Enugu State. The pods were identified by DR. E.U Onuigbo of crop science department, University of Nigeria NSUKKA.

Sample preparation and analysis: The seeds were manually harvested from the pods, washed clean. The

seeds were separated into seeds (endocarps) and seed coats (epicarps) using a clean knife. The seeds and seed coats were each divided into two portions, for fresh and dry processing. The seeds and seed coat for dry processing were air-dried for 72 h. Both fresh seeds, seed coats and dried seeds were reduced into small sizes with an electric hopper, then each part ground into powder using an electric blender. Each powdered seed and seed coats (fresh and dried) were sieved using a 2 mm-steel sieve, then stored in labeled plastic containers until required for analysis.

Oil extraction: Powdered seed and seed coat samples (fresh and dried), 40 g each was extracted with 100 mL petroleum-ether in a Soxhlet apparatus for 21^{1/2} h. The petroleum-ether extract of each sample was concentrated using a rotary evaporator at 45°C, in a hot air-circulating oven to get an oil extract for each sample. The weighed percentage weight was calculated.

Folch et al. extraction: Ten grammes of each powdered seed and seed coat samples (fresh and dried) were transferred into different Conical flasks, homogenized for five minutes using 30 mL Chloroform-methanol (2:1) volume/volume. Each homogenized mixture was filtered using glass-funnel fitted with Whatman No 40 fluted paper. Each residue was rehomogenized twice. Each residue again rehomogenized with 30 mL chloroform and 30 mL methanol for ten minutes and filtered. The combined filtrates of each extract were dehydrated by

adding ten grammes of anhydrous sodium carbonate, stirred for ten minutes each, filtered, each extract was weighed^[6]. Thin layer chromatography was used for the identification of lipid classes from the oil extracts from different parts of fluted pumpkin seeds. Each class of lipids were separated using different solvent mixture and each spot developed using iodine vapour;

Neutral lipid fraction: Petroleum ether: diethyl ether: glacial acetic acid (90: 10:1). Polar lipids; chloroform: methanol: glacial acetic acid: water (65:25:8:4). Other lipid fractions; n-heptane: diethyl ether: glacial acetic acid (80: 20:1) RF values were calculated from the developed spots and matrix with standard RF values of known lipid compounds.

RESULTS AND DISCUSSION

Table 1 showed that the seeds (fresh and dried) contain more oil (29-37%±10.1) than the seed coats (fresh and dried) (0.5-25.20%±0.3) this may be due to the

Table 1: Oil extracts from different parts of fluted pumpkin seeds in percentage values

Parts of seeds	Condition of seed	Method of extraction used	% Yield
Seeds	Fresh	Soxhlet	34.05±0.10
Seed	Dried	Soxhlet	29.00±0.30
Seed water	Fresh	Soxhlet	12.50±0.20
„	Dried	Soxhlet	0.50±0.1
„	Fresh	Folch <i>et al.</i>	37.00±0.20
Seed	Dried	„	30.50±0.01
„	Fresh	„	25.02±0.02
Seed water	Dried	„	7.50±0.10

Values are means of four determinations±standard deviation.

facts that seeds are usually a storage organ of plants while the seed coats usually served as protective organ of plant seeds (endocarps). The fresh seeds and seed coats yielded more oil than the pried seeds and seed waters 7.5-37% and 0.5-29%, respectively. This significant difference between fresh seeds, seed coats and dried seeds, seed coats may be clue loss of volatile components of the oil in the seeds and seed coats during the drying exercise. Also the methods of oil extraction used yield a significant difference in oil quantity 7.50-37% and 0.5-34% for Folch *et al.* and soxhlet, respectively. This may be due to the affect of repeated extractions used in the Folch *et al.* method which increases separation efficiency and yield.

Table 2 showed the lipid classes identified. These include; neutral lipids glycolyids, phospholipids, sterols, sterylesters and free fatty acids. The presence of these lipid classes in different parts of fluted pumpkin seeds makes it a natural source of bioactive substances in diets and herbal medicine^[3,5]. Lecithin for instance, is derived from phospholipid; phosphatidyl choline is used as emulsifying agents in processed such as chocolate, candies, ice-creams^[7,8].

Lecithin when hydrolyzed by snake venom enzymes yields lysolecithin and lysocephalin which are used in herbal and orthodox medicine as haemolytic-action inducing substances^[7]. These bioactive substances present in fluted pumpkin seeds makes it a potential source of raw materials for food and pharmaceutical industries.

Further, work is still going in our Laboratory to quantify and characterized these bioactive substances identified in these oil extracts.

Table 2: Classes of lipid identified and their associated RF values

Parts of seeds	Conditions	Eluuting solvent system used	RF values obtained	Class of lipid identified by comparison with standard lipid RF values
Seed	Dry	Petroleum-ether: diethyl/ ether	0.14, 0.24	Monoglyceride (0.14) and
Seed	Wet	galacial acetic acid	0.29, 0.45, 0.85 and 0.97	diglyceride (0.24).
Seed coat dry	Dry	„ „ „	0.24, 0.29, 0.45, 0.85 and 0.97	Cholesterol (0.29), glyceroltristearate fatty acids (0.45) and cholesterol stearate (0.97).
Seed coat	Wet	„ „ „	0.24, 0.29, 0.45, 0.85 and 0.97	Same as above
Seed coat	Dry	Chloroform: methanol:	0.62	Phosphatidy/inositol (0.62)
Seed coat	Wet	„ „ „	0.62	„ „
Seed coat	Dry	„ „ „	0.62	„ „
Seed coat	Wet	„ „ „	0.62	„ „
Seed	Dry	n-heptane: diethyl/ether: galacial acetic acid	0.00, 0.23,	„ „
Seed	Wet	„ „ „	0.00, 0.23	Phospho lipidi (0.00) and sterol (0.23).
Seed	Dry	„ „ „	0.00, 0.23	same as above
Seed coat	Wet	„ „ „	0.00, 0.23	Same as above
Seed coat	Dry	„ „ „	0.00, 0.23	Same as above

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