Research Journal of Applied Sciences 2 (1): 114-116, 2007
Medwell Journals, 2007

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.±0.3) and (0.5-25±0.1) for the seeds and seed coats, respectively. The following lipids were identified; phosholipids, glycolipids, sterols, fatty acids and neutralipids. These lipid classes were isolated and identified using different organic extract ants 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, glycohpid, neutral lipid, lecithin, raw material

INTRODUCTION

Fluted pumpkin seed (*Telfaria occidental is*) is a dioecious perennial vine plant. It is of the family cucurbitacae 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 in cultivated in many parts of West Africa. It a drought tolerant vine plant, favoured by rainfall of sufficient amount[1,3,4]. Fluted pumpkin grow best a cower a litude 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 from, 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 coals (epicarps) using clean knife. The seeds and seed coats were each divided into two portions, for fresh and dry processing. The seeds and seed coal for dry processing were air-dried for 72 h. Both fresh seeds, seed coats and dried seeds seed were reduced into small sizes with electric hopper, then each part ground into powder using electric blender. Each powdered seeds and seed coals (fresh and dried) were sieved using 2 mm-steel sieve, then stored in a 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 mh petroleum-ether in a 80xhlet 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 was weighed percentage weight calculated.

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

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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: galacial acetic acid: water (65:25:8:4). Other cipid 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 cipid 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 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 signification 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 fatly acids. The presence of these cipid 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 haemolcytic-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 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±01
,,	Fresh	Folch *et al.*	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.

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

Parts of seeds	Conditions	Eluating solvent system used	RF values obtained	Class of lipid identified by comparison with standard lipid RF values
Seed	Dry	Petroleum-ether: diethy/ ether	0.14, 0.24	Monoglyceride (o.14) and
Seed	Wet	galacial acetic acid	0.29, 0.45,085 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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