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CHARACTERIZATION OF CRUDE STARCH EXTRACT OF WILD CASSAVA TUBER (*Ampelocissus africana*)

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ABSTRACT

Ampelocissus africana (wild cassava tuber starch) is a starch-rich tropical plant that has been used as a part of diet for people for several years, particularly during times of famine. Its stalk and foliage are used to treat many ailments. In this study, the tuber starch extracted from wild cassava tuber (Ampelocissus africana) was evaluated. The iodine test indicated a starch content of 10.12%. The isolated starch was observed to be odorless and tasteless, with a faint ash-off white color. SEM analysis revealed that the starch granular morphology was globular and oval. Other found properties are: ash (2.5%), moisture content (69.00%), fat (1.5%), fibre (5.5%), crude protein (4.46%), and accessible carbohydrate (84.54 %). These findings revealed that wild cassava tuber starch is a good source of carbohydrates, primary starch, which has a wide range of household and commercial applications.

Keywords: Wild cassava starch, proximate analysis, extraction, carbohydrate, morphology.

INTRODUCTION

Binders, matrix formers, drug release modifiers, film coating formers, thickening agents or viscosity promoters, stabilizers, suspending agents, gelling agents, and bioadhesives are all examples of polymeric materials. According to Vishwanath (2017), in their molecular structure, polymers contain reactive functional groups such as hydroxyl, amino acid, and carboxylic acid groups, which are fundamental for chemical modification.

(Kaushik *et al.,* 2016)) reported that the two main elements in any pharmaceutical composition would be active components and additives. Excipients facilitate in the production of oral dosage forms while also improving their physicochemical qualities. As additives, polymers are essential in any dosage form. Biopolymers should be suitable, non-toxic, stable, and economically significant effect on the drug delivery system (Kaushik *et al.* (2016)). Polymers are divided into three categories: natural polymers, semi-synthetic polymers, and synthetic polymers, according to the (Kaushik et al. 2016) . In the oral drug delivery system, natural polymers are commonly utilized as rate-controlling, taste-masking, test-protecting, and stabilizing agents.(Kaushik *et al.* 2016). Certain polymers were employed to offer uniform drug distribution, decreasing the probability of dosage and improving the efficiency of the drug administration via localisation to the targeted site. Due to a slew of issues with medication conveyance systems and synthetic polymer side effects, businesses are increasingly turning to biopolymers. Natural polymers are biocompatible and have no or few negative effects because they are polysaccharides.

Polysaccharides are biopolymers that are known for their biocompatibility and biodegradability, making them among the most important natural resources for polymeric drug delivery (Carbinatto et al. 2012).

One of the most widespread polymers is starch, which can be obtained from a multiple sources. According to Liu *et al*, (2008) polysaccharides are divided into two types: linear and branched-chain polysaccharides (i.e. amylose and amylopectin). Polysaccharides have various reactive functional groups in their molecular structure, such as hydroxyl, amino, and carboxylic acid groups, indicating that functionalization is possible. Starch was the main polymer that was present in leaves, flowers, fruits, seeds, various types of stems, and roots, and is the most plentiful carbohydrate resource in flora. Smith (2001) believed that starch is used by plants as a source of carbon and energy. The biochemical chain responsible for starch synthesis involves glucose molecules produced in plant cells by photosynthesis (Alcázar-alay, Angela, and Meireles 2015).

Starch occurs as a fine powder or angular, irregular masses readily reducible to powder. Most pure starch would be white, tasteless and odorless powder that would be insoluble in cold water or alcohol. Starch chemically contains two different polysaccharides: amylase (*B-amylose*) and amylopectin (*α-amylose*) in the proportion of 1:2. Amylose would be water soluble, while amylopectin would be water insoluble but swells in water and its responsible for the gelatinizing property of the starch.

Bordoloi *et al.*, (2019) stated that depending upon the source that would be the type of plant, starch generally contains 20 to 25% amylase and 75 to 80% percent amylopectin by weight.

Present trends towards technologies and processes that increase the use of residues make starchy vegetal biomass an important alternative material in various applications due to starch's versatility, low cost and ease of use especially when its physicochemical properties was altered (Alcázar-Alay & Meireles, 2015). Starch was increasingly used in many industrial applications and as a renewable energy resource. Starch can be modified to enhance its positive attributes and eliminate deficiencies in its native characteristics (Alcázar-Alay & Meireles, 2015).

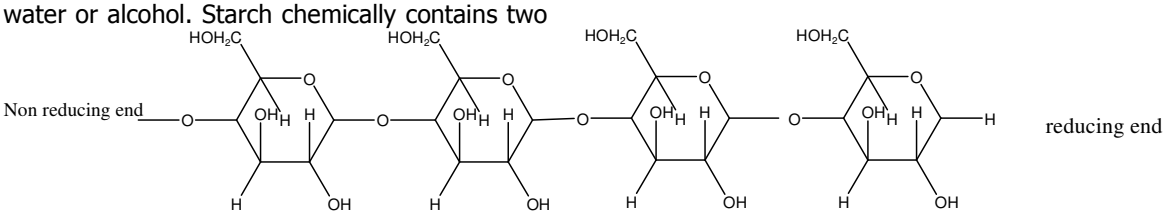

FIG.1: Chemical Structure of Amylase (Bordoloi *et al.*, 2019)

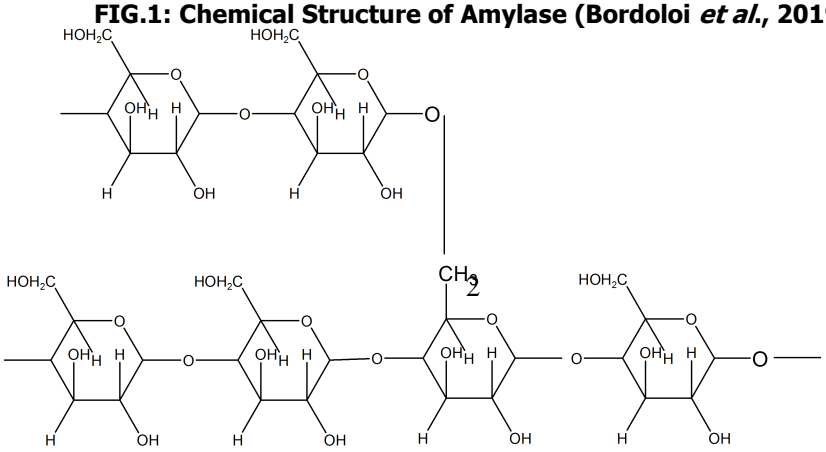

FIG. 2: Chemical Structure of Amylopectin (Bordoloi *et al.*, 2019).

Cereals and seeds (such as corn, maize, wheat, rice, sorghum, barley, or peas) as well as tubers or roots (such as potato or cassava) of plants contain starch. Corn produced the majority of the starch produced around the world, although other types of starch such as cassava, sweet potato, potato, and wheat starches were also produced in substantial quantities (Mohammed, 2017).

In Nigeria's Niger state, wild cassava (Figure 1) is abundant. During the famine, it was used as a source of food, especially for the people leaving in the villages. Its stem and foliage are often utilized for therapeutic uses and can also provide a source of revenue for the plant's owners. Its prevalence in Guinea, Sudan, and the Sahel savanna region was attributed to its capacity to withstand extreme heat and drought. For its accessibility, non-toxicity, and biodegradability, wild cassava tuber starch (Figure 2) can be used in oral solid dosage preparations for optimal medication (Bordoloi, Kalita, and Shil 2019).

This research was aim to characterized the crude starch extract of wild cassava tuber starch (Ampelocissus Africana) which can be used for food and effective drug delivery.

MATERIALS AND METHODS

MATERIALS

The materials used include: pH meter. jenway 3510. jenway of china, ft-ir cary630. Agilet technologiest of USA, electronic weighing balance. Shimazu/aw320. Shimazu of japan, SEM. Phenompro. Phenomworld thermo fisher of schwisland, Air oven. Dhg9023a. gallenkam of united kingdom, centrifuge machine Tdl-4. Pro-research centurion of USA and sieve. Sethi standard `sieve of chia. Also, the chemical used were of pure grade.

METHODS

SAMPLE COLLECTION

The wild cassava tuber was collected at Unguwan Makarantan Garo Rijau Local Government Area of Niger State, Nigeria and was taken to plant taxonomist for identification.

Figure 3: wild cassava tuber (*Ampelocissus Africana* Tuber)

EXTRACTION OF WILD CASSAVA STARCH

The procedure described by Narayana and Re- (2002) was adopted. The tuber of wild cassava was skinned, chopped into pieces, and measured using an electronic weighing scale. To prevent oxidation, the wild cassava cubes were soaked overnight in 10 dm^3 of 0.075% sodium metabisulphite solution. To acquire the starch sediments, the mixture was milled, mixed, then filtered through double-fold clean muslin that was allowed to settle fully. The starch was isolated from the water and non-water soluble components after decanting the water and centrifuging the suspension at 4000 rpm for 10 minutes using a table-top centrifuge. The centrifuged pure starch was air-dried at 40 °C in a hot air oven for 24 hours, powdered with a grinder, then sieved through Seth standard sieves. After that, the flour was sealed in an airtight container and stored at room temperature for further analysis.

CHARACTERIZATION OF NATIVE AND MODIFIED STARCHES

PHYSICAL APPEARANCE

The moisture content of a sample is defined as the mass of water in the sample expressed as a The colour, odour and texture of the native wild cassava starch were virtually examined.

SCANNING ELECTRON MICROSCOPY (SEM)

The analysis was conducted by mounting the starch sample on metal stubs and coating it with gold-palladium alloy (15 nm thickness) using the quorum Q150TES vacuum coating device to investigate the microstructure of the starch. The sample was examined using a scanning electron microscope at a 15 kV acceleration potential.

FOURIER TRANSFORMED INFRARED RADIATION (FTIR) ANALYSIS

The starches was analyzed by Fourier transformed infrared radiation (FTIR) in transmission mode. Transmission spectral was recorded using at least 64 scans with $8cm^{-1}$ resolution in the spectral range 4000- 400 cm^{1}.

PROXIMATE ANALYSIS

Determination of moisture content

This was done according to the association of official analytical chemist (AOAC) (2010) method.

percentage of the dry mass, usually heating at 105 ^{O}C. It was calculated according to the equation (1) below;

moisture content, w = $\frac{mw}{md} X\ 100 - - - - - - equ\ (1)$

where MW = is mass of water

MD = is dry mass of the sample

Determination of Ash Content

Starch sample 1.5 g was measured into a known-weight crucible. The sample in the crucible was placed in a furnace. The crucible was then taken out of the furnace after ashing, kept at room temperature, and weighed (AOAC) (2010) method.. The sample's quantity/content was determined using the equation (2) below.;

$$\%\ of\ the\ ash\ in\ the\ sample = \frac{weight\ of\ the\ ash}{weight\ of\ the\ sample} x100 \quad - - - - - - equ\ (2)$$

Determination of Fibre Content

It was determined as illustrated by association of official analytical chemist (AOAC) (2010). Acid-base digestion was used to quantify the crude protein. The decanted residue in the solvent phase was taken and mixed with 20 cm^3 of 10 % H_2SO_4 solution and distilled water, then heated for 30 minutes and filtered. The residue was poured into a crucible and dried in the oven. The weight of the dry ash was weighed, and the % fibred was estimated using equation (3) below.

$$\%\ of\ the\ crude\ fibre = \frac{weight\ of\ the\ rsidue}{weight\ of\ the\ sample} x100 \quad - - - - - - equ\ (3)$$

Determination of Crude Protein

The Kjeldahl method was used to calculate protein contents according to the association of official analytical chemist (AOAC) (2010). Prior to the determination of protein, nitrogen was evaluated. Sample of 0.5g was weighed. Then 20cm^3 of concentrated H_2SO_4 was added to the sample and shake to obtained the homogeneous mixture. volumetric flask of 50cm^3 capacity was filled with distilled water to the mark and the solution was then poured into the digestion block to be digested. After digestion, 10 cm^3 of the solution was combined with 20 cm^3 of NaOHaq and 30 cm^3 of distilled water to make a total volume of 60cm^3. 20cm^3 of boric acid indicator was measured and placed in a separate 250cm^3 beaker. The prepared sample was transferred to the Macrokjelder equipment and distilled. When the vapor passed through the condenser, the lipids condensed, fell into a separate container of boric acid indicator, set in a lower part of the Macrokjelder apparatus. The procedure was repeated until the color of the boric acid indicator changed from pink to green. The green-colored sample was titrated with 0.01 M H_2SO_4 until it turned pink again, and the tittered crude lipid value, which was nitrogen, was recorded. The percentage nitrogen was determined, and protein content was estimated by multiplying the percentage nitrogen by a protein conversion factor as reported by association of official analytical chemist (AOAC) (2010).

Determination of Crude Lipid

Crude lipid was extracted with n-hexane: a weighed sample (2g) of was suspended in n-hexane and allowed to stay overnight and then the oily portion was decanted and heated to evaporate the solvent and the oil content was determined according to the following equation:

$$\%\ of\ the\ crude\ lipid = \frac{weight\ of\ the\ oil}{weight\ of\ the\ sample} x100 \quad - - - - - - equ\ (4)$$

Determination of Total Carbohydrates

The carbohydrates content was determined as illustrated by Association of Official Analytical Chemist (AOAC) (2010), as follows; it was estimated according to equation (8) below.

Total carbohydrates= 100 – (moisture +%protein +%fats +% crude fibre) ----------equ (5)

RESULTS AND DISCUSSION

Visual examination

The extracted starch was found to be fine, light ash-colored powder with no flavour or smell.

Physicochemical analysis

A screening study on native starch with an iodine solution revealed that, the solution turned blue-black. Additionally, granular microstructure was examined using a scanning electron microscope, with the results displayed in the tables and figures below.

Figure 1: wild cassava flesh

Figure 2: wild cassava (*Ampelocissus africana*) tuber starch

TABLE 1: Preliminary Results

S/no	analysis	Result
1	Yield	10.12
2	Texture	Fine
3	Odour	odourless
4	Taste	tasteless
5	Colour	Light ash colour
6	Iodine test	++
7	Granular morphology	Some spherical while some oval shape
8	pH	7.51

FT-IR of a native wild cassava tuber starch (*Ampelocissus africana*) is shown in figure 3.2.1. above. The spectra produced from this study indicate the presence of these three major absorption peaks, which includes: O-H stretching, C-H stretching and C-O stretching vibration.

Spectra of the selected excipients displayed their characteristic absorption vibrations in the region between 3600 and 2700 cm−1 because of the C–H and O–H stretching vibrations, with a strong broad band at 3430 cm−1 in the case of CS, due to the additional stretching vibrations from N–H bonds (Pramod et al. 2015). The characteristic strong absorbance peaks of PCL between 3000 and 2800 cm−1 and at 1728 cm−1 due to aliphatic CH_2 and carbonyl group (C=O) stretching, respectively, were noted in the spectra of pure PCL and combination forms (Aminu *et a*l. 2021).

SEM analysis revealed that the starch granular morphology was globular and oval in shape. The SEM analysis showed that the particles of TCS and FLB-loaded NG were mostly spherical (Aminu 2019).

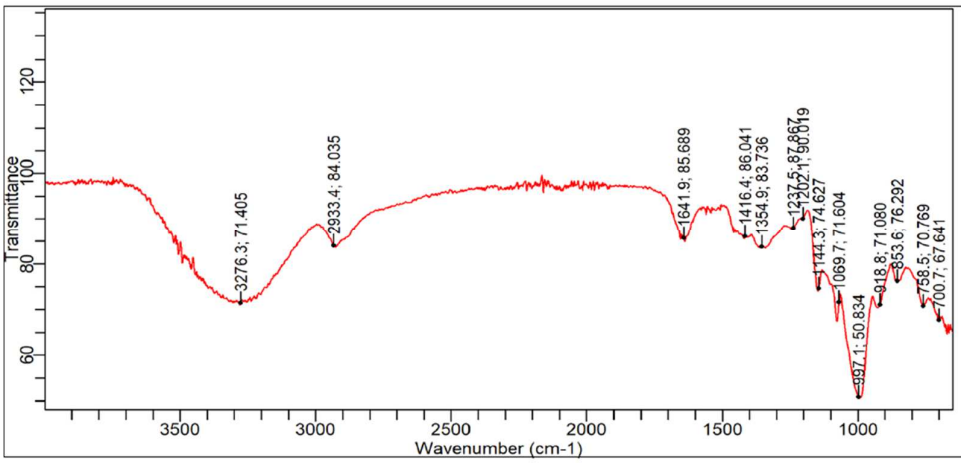

Figure 3: FT-IR analysis of wild cassava tuber starch

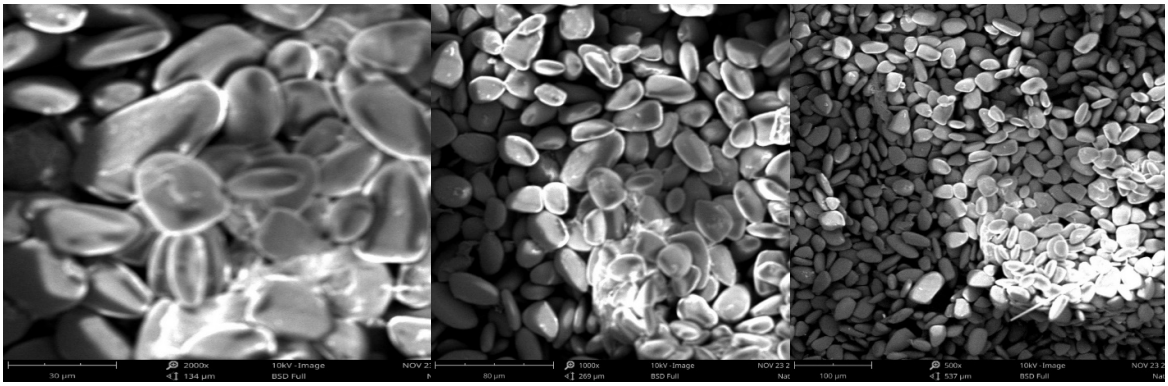

Figure 4: SEM analysis of (*Ampelocissus africana*) wild cassava tuber starch

Table 2: proximate analysis (Nwoko et al. 2016)**.**

parameters	**Result (%)**
Moisture content	69.00
Ash value	2.5
Lipid	1.5
fiber content	5.5
Crude protein	4.46
Carbohydrates	84.54

The proximate analysis of the *Ampelocissus africana* tuber starch from the table 3.3.1. above showed a positive result, even though the specie had a moisture content of 69.0%, is more suitable for long storage of it tuber than those with high moisture value found by (Nwoko et al. 2016). The content of carbohydrate, fibre, protein, and lipid was found to be 84.54%, 5.5%, 4.46% 1.5% respectively. Therefore the crude starch extracted from the (*Ampelocissus africana*) can serve as a source of food and it can be modified to be used as an excipient for effective drug delivery.

CONCLUSION

The percentage yield of the extracted starch was found to be 10.12 and it was a fine, light ash-colored powder with no flavour or smell. A screening study on native starch with an iodine solution revealed that the solution turned blue-black, and the FT-IR analysis confirm the present of starch. Both moisture and carbohydrates content was found to be 69.00 and 84.54 percent respectively, which indicate the possibility of making use of the starch as a source of food and also as an excipient for effective drug delivery.

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