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Irvingia gabonensis Seeds Fractions Implicated in Gastric Microstructure Alteration on Adult Wistar Rats

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Abstract | **Original Research Article**

Irvingia gabonensis is an African tree with edible yellow fruit resembling mangoes valued for its oil rich seed,fresh fruit, fuel, fibre, medicine,and hardy green termite resistant wood.this study was undertaken to investigate the effects of *Irvingia gabonensis* seed extract on the gastric microstructure of adult wistar rats. Thirty-five adult albino rats were used for this experiment. The rats were divided into seven different groups with 5 rats in each group. Group a (control group) was treated with distilled water. Group B1, B2, C1, C2, D1 and D2 (experimental groups) was treated with 0.85ml, 2.55ml, 0.80ml, 2.70ml, 0.90 ml and 2.55ml/kg body weight of *Irvingia gabonensis* seeds extract respectively, once daily for 21 days. The weight of the animals was taken once a week and the data were analyzed using descriptive statistical tool the doses for the extract were calculated from the lethal dose (LD_{50}) experiment conducted for dose determination. At the end of the administration, the stomachs were harvested from each group after animal sacrifice, processed and stained using hematoxylin and eosin staining technique. Histological findings revealed that group a showed normal stomach features, group B1and B2 treated with 0.85ml and 2.55ml of the raw ground extract in water showed inflammation of the mucous secreting cells and mildly erythematous mucosa. Group C1 and C2 administered 0.80ml and 2.70ml of the ethanolic extract showed a variant from the normal mucosa revealing a granular mucosa, Group D1 and D2 administered 0.90ml and 2.55ml showed no histological changes as compared to the control group which showed normal appearance of the stomach. It can safely be concluded that raw *Irvingia gabonensis* has a dose dependent damaging effect on the mucous secreting cells of the body and fundus of stomach
Key words: *Irvingia gabonensis*, Stomach, Wistar Rat, Hematoxylin.

INTRODUCTION

Thickening agents or thickeners are substances which, when added to an aqueous mixture, increase its viscosity without substantially modifying its other properties, such as taste and aroma. When added to a mixture, it helps to increase stability by absorbing moisture and improve suspension of added ingredients [1]. Thickening agents are often used as food additives and in cosmetics and personal hygiene products. Food thickening can be important for people facing medical issues with chewing or swallowing, as foods with a thicker consistency can reduce the chances of choking, or of inhalation of liquids or food particles, which can lead to aspiration pneumonia [2]. Most plant seeds have shown to have this property when pulverized. Their seed flour has gelatinous properties when mixed with water and it imparts a gummy texture when used in preparing soup. This contribute to its desirable property necessary for the eating of garri, pounded yam and a major food delicacies in Africa especially in west Africa [3].

Edible seeds such as Mucunasloaeneri (okobo), Brachystergianigerica (achi), *Irvingiagabonensis* (ogbono) are used in soup, stew or sauce additive for flavouring and thickening in most part of Nigeria especially in the South Eastern and Southern region[4]. These seed serves as a source of nutrients, carbohydrate, fiber and it is also medicinal. These include softening of stool and lower risk of colon cancer[5], promotion of early satiety and normal laxation [6], moderation of post-prandial blood glucose responses and improved insulin sensitivity[7, 6], reduction in total and low density lipoprotein (LDL)-cholesterol [7, 8],

regulationofappetiteand enhancement of sodium and fluid balance[9]. They are also used to treat constipation and prevent development of diverticulosis and diverticulitis[10].

Irvingiagabonensis is a tall and large deciduous tree, with the ability of attaining a height of between 30 to 40m [11]. It is usually an emergent tree species that grows high above the canopy in primary and secondary forest [12]. Generally, it has a large dense, compact and rounded crown. Based on the identified crown variation [13], classified it into two varieties Var. gabonensis and Var. excels (wombolu).

The leaves have high leaf dry weight [14] and are between 5-15cm long and 2.5 -6cm wide. They are elliptic to slightly obovate with one margin a little more rounded than the other, acute or shortly acuminate, cunnate or slightly rounded at the base [15]. They are leathery dark green in color and glossy with 5-10 pairs of irregular lateral nerves with the lower ones running out nearly to the margin and the upper ones looped with the veins, forming a close network between them [16]. Leaves stalk are stout and about 6mm long. Young leaves of bush mango are usually pale green and sometimes pink. Field and nursery observation have shown that some trees of *Irvingiagabonensis* have red leaves [17].

Flowers, fruits and seeds have shown variation in the number of floral flushes with most trees flowering and fruiting once every year [18]. However, some trees of the species fruit once in two years. The flower are yellowish to greenish white in colour and usually in slender clustered or in small panicles among the leaves or in branchlets and younger branches [16]. Flower stalks are slender and about 6mm long and its petals are bent right back and fall off quickly [16]. Flowering in *Irvingiagabonensis* tree commence 10-12 years after planting [19]. Fruit maturity in *Irvingiagabonensis* tree is between July- August (early fruiting) and August-September (late fruiting). In appearance, its fruit resembles that of a small cultivated mango, hence the common name; bush or wild mango [15]. The fruit is a drupe, broadly ellipsoid and flattened. It is generally green but becomes yellowish when ripe, although the color of the matured fruit has been reported to vary from tree to tree. The fruit has a yellowish, broad, fleshy and fibrous pulp (mesocarp) surrounding a large hard stone (endocarp). The pulp of *var. gabonensis* is scantily, sweetish and edible and usually eaten fresh by animals, rural and urban dwellers in west and central Africa. The fruit pulp of Var. excelsa is bitter and not edible.*Irvingagabonensis* is a one seeded fruit. The large fruit stone (seed is covered by a hard seed coat, which when cracked open reveals white cotyledon seed kernel) is wrapped in a brown testa. The oily seed kernel called 'ogbono' 'apon' or dika nut is the part of *Irvingiagabonensis* with the greatest commercial potential. Its seed is recalcitrant and cannot be stored successfully in the seed gene bank.

The stem and bark of the two species of *Irvingiagabonensis* differ, the bole of *I .gabonensis* is usually straight, slender, fluted or cylindrical while the bole of *I. excelsa* is slightly buttressed which could sometimes reach a height of 6m [20]. The bark of bush mango tree is greyish in color and fissured, smooth or very slightly scaly, its slash is thick, light brown, granular with a narrow pale-yellow layer on the underside and brittle [16]. The bark of the species contains tannin and dyes [15] and is known to be used for medicinal purpose. The heart wood of *Irvingiagabonensis* is pale brown or orange-yellow in colour, but on exposure, the color usually fades to grey brown sometimes with dark grey streaks [21]. Texture of wood is fine to medium, while the grain which is very hard and fine is straight to interlock without luster [22]. The wood is also described as heavy, durable, hard and difficult to cut which limit its usefulness [22].

Fig-1: Irvingia*gabonensis* seed Source (https://www.google.com.ng)

Taxonomic classification

Kingdom- Plantae, Phylum– Angiosperm, Class - Eudicots, Order-Rutales Family -Irvingiaceae, Genus - IrvingiaSpecies - *Irvingiagabonensis*[23].

The genus name of *Irvingiagabonensis* commemorates Irving (1816-1855) a scot botanist. The Irvingiod group was placed as a subfamily within the Simaroubacene, despite the lack of quassin and other related bitter principles [22]. It is now generally recognized that Irvingiaceae is a family in itself containing the genus Irvingia[22].The names given to *Irvingiagabonensis varies* based on region and location. English: bush mango, wild mango, African wild mango, dika nut.Spanish: manguier du Gabon.

Chemical constituent

Irvingia species seeds are the most valuable products of the tree and have the most industrial potential. The crude fat content is 62.5% [23] proving them to be very good oil seeds. The seeds also contain ash, moisture, protein, fat, fiber, carbohydrate, predominant mineral is magnesium. Fruits of both Irvingia species possessed all five phytochemicals (alkaloids, flavonoids, saponins, tannins and glucosides). However, whilst both species had the same amounts of flavonoids and glycosides, *Irvingiawombolu* contain relatively higher amounts of alkaloids, saponins and tannins than *Irvingiagabonensis*. *Irvingiawombolu* may be the preferred choice of domestication would be based on phytochemicals. In like manner, *Irvingiagabonensis*may be the preferred choice for domestication if taste, weight and size of fruits are the parameters of interest[24]. Water absorption, oil emulsion capacities are relatively high while foaming capacity and least gelation concentration are low. Presence of lipid (dika fat) and its polymeric gum components have been used by pharmaceutical scientist as excipients in various formulations.

Uses of the irvingia gabonensis seeds

Processing of *Irvingiagabonensis* seed involve a number of steps. The seed is first split into two halves with a cutlass or the seed coat is cracked open with a stone. The kernel is used as a thickening agent in stew and soup. This soup in Nigeria is popularly known as "ogbono soup". This thickening property is due to its mucilaginous polysaccharides in the kernel which become more viscous during cooking. Fat extracted from the kernels can be used for food applications, such as in margarine or cooking oil, and is also suitable for soap, cosmetics and pharmaceuticals. Flour can be produced from the kernels [25]. The kernel can also be processed into a stiff cake to produce dika cake, Gabon chocolate or dika bread.

Gross anatomy of the stomach

The stomach is a J-shaped muscular expanded structure. The stomach lies in the right and left upper quadrant and left hypochondrium. It is part of the alimentary tract and it is between the oesophagus and the small intestine. It is specialized for the accumulation of ingested food, which it chemically and mechanically prepares for digestion and passage into the duodenum [26]. The stomach is situated in the upper part of the abdomen, extending from beneath the left costal margin region into the epigastric and umbilical regions. Much of the stomach lies under cover of the lower ribs. It has two openings, the cardiacand pyloric orifices; two curvatures, the greater and lesser curvatures and two surfaces, an anteriorand a posterior surface [27].

The shape and position of the stomach vary markedly depending on the degree of distention and the body habitus and even in the same individual as a result of diaphragmatic movements during respiration, stomach's content and position of the person. It has four parts, the cardia, fundus, body and pyloric part. The cardia part surrounds the cardial orifice, the superior opening or inlet of the stomach; it lies at the eleven thoracic vertebrate. The fundus dilated superior part is related to the left dome of the diaphragm and lies posterior to the left 6^{th} rib in the plane of the mid-clavicular line[28].The body part is between the fundus and the pyloric antrum. The pyloric part is a funneled shaped outflow region of the stomach and leads to the terminal part which is marked by a thickening of the circular layer of smooth muscles that controls the discharge of the stomach content into the duodenum. The stomach is quite mobile and is fixed above at the esophagogastric junction and below at the gastroduodenal junction. Externally, the stomach is covered completely by peritoneum, except where the blood vessels run along its curvatures, and a small bare area posterior to the cardiac orifice. The peritoneum is reflected at the lesser curvature forming the lesser omentum that extends to the liver and at the greater curvature to become the greater omentum (a double layer of fatty peritoneumsuspended from the greatercurvature)[29].

The antero-superior surface of the stomach is in contact with the diaphragm, gastric surface of the spleen, left and quadrate lobes of the liver, the anterior abdominal wall, and the transverse colon—when the stomach is empty. The postero-inferior surface (stomach bed) is formed by the posterior wall of the omental bursa and retroperitoneal structures between it and the posterior abdominal wall. Superiorly, the stomach bed includes part of the diaphragm (left crus), the spleen, the left suprarenal gland, and upper pole of the left kidney. Inferiorly, the stomach bed includes the body and tail of pancreas, transverse mesocolon, left colic flexure, the splenic artery, and, in some people, the transverse colon [30].

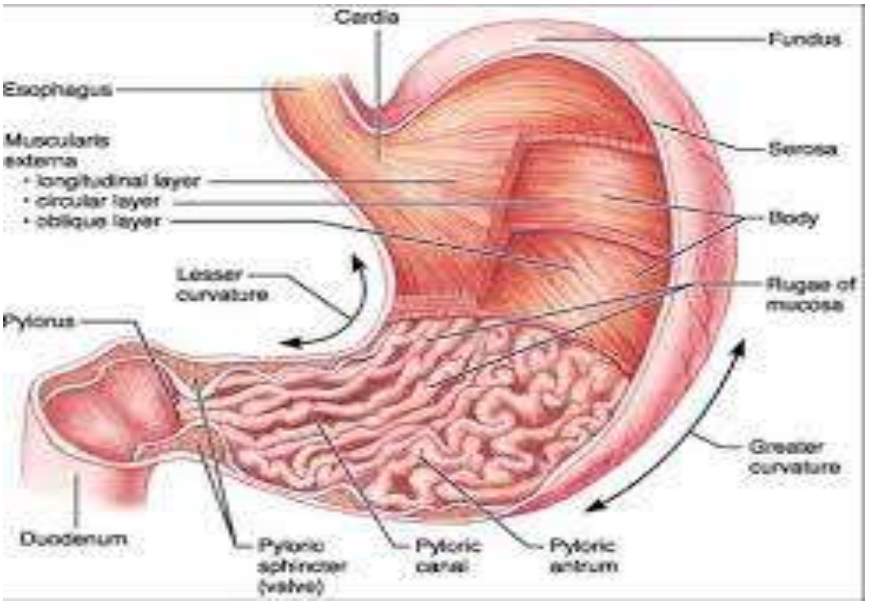

Fig-2: Gross Anatomy of the stomach (http: //www.google.com.ng)

Histology of the stomach

The surface of the stomach is covered by a simple columnar epithelium. The three histological region of the stomach include: the cardia, fundus and body, and the pylorus. The luminal surface of the stomach is pitted with numerous tiny opening called gastric pits. These are formed by the luminal epithelium that invaginates the underlying connective tissue lamina propriaof the mucosa. The tubular gastric glands are located below the luminal epithelium and open directly into the gastric pits to deliver their secretions into the stomach lumen. The gastric glands descend through the lamina propria to the muscularis mucosae. Below the mucosa of the stomach is the dense connective tissue submucosa containing large blood vessels and nerves. The thick muscular wall of the stomach, the muscularisexterna, exhibits three muscle layers instead of the two [31].

The fundus and the body comprise about two thirds of the stomach and have identical histology. As a result, the stomach has only three distinct histologic regions. The fundus and body form the major portions of stomach. Their mucosae consist of different cell types and deep gastric glandsthat produce most of the gastric secretionsor juices for digestion. Also, all stomach regions exhibit rugae, the longitudinal folds of the mucosa and submucosa. These folds are temporary and disappear when the stomach is distended with fluid or solid material [32]. There are works that have been done on this subject by different authors [6-9, 11, 13-15, 17-19, 22-25, 42, 43].

Statement of the problem

Irvingiagabonensis are semi-wild trees that grow abundantly in Nigeria and other tropical countries. While the seedsoil of this plant is used industrially [33], reports suggest that its nutritional components would add food value to nutritionally deficient diet [34-36]. Other reports suggest that the oil has pharmaceutical properties [37-40]. The properties of *Irvingiagabonensis* seeds to act as an emulsifier and add viscosity to liquids may act in the body, and this soluble fiber-like property may cause effects such as satiety and digestion. There is evident increase in the use of *Irvingiagabonensis* in food preparation, by pharmaceutical industries and it has therefore become necessary to investigate the effect of *Irvingiagabonensis* seeds. Thus this study was to ascertain the effect of the ingestion of seed extract of *Irvingiagabonensis* on the microstructure of the stomach.

Aim of the study

This study was aimed at determining the effects of *Irvingiagabonensis* seed extract on the gastric microstructure of adult albino wistar rats and to investigate the effect of *Irvingiagabonensis* seed extract on the body weight of the rats.

MATERIALS AND METHODS

Research design- This study was experimental

Data collection

Thirty-five (35) adult Albino Wistar rats were obtained from the Animal House, Faculty of Basic Medical Sciences, University of Uyo, Uyo. They were housed in cages with adequate space to encourage free movement, saw dust were spread on the bottom of the cages to provide bedding and were replaced with clean ones weekly. The animals were allowed twelve hours of light and twelve hours dark cycle at a room temperature. They were fed with standard rat pelletized diet (Vital Feed Growers, Grand Cereals Nigeria Ltd) and water. The animals were acclimatized for two weeks and after that divided into seven groups (Groups A, B1, B2, C1, C2, D1, D2) containing five rats each. They were identified by different colour markings on

their tails and head. All the animals were handled and cared for in accordance and in compliance with applicable guidelines and standard for the care and use of laboratory animals.

Preparation of extract

1kg of *Irvingiagabonensis*seeds was obtained from a recognized farm produce merchant atItam market in Uyo Local Government Area of AkwaIbom state. The seeds were extracted using the soxhlet apparatus. The seeds were divided into three parts (400g, 400g, and 200g). The first parts were extracted using ethanol and the second part using hexane. The third part was pulverized using a grinder and stored in a container prior to administration. The extract was administered to the animals based on their body weight.

Determination of median lethal dose (ld_{50}) using lorke's method

Lorke's method was calculated as geometrical mean of maximum dose producing 0% mortality and minimum dose producing 100% mortality $LD_{50}=\sqrt{ab}$ [41].

The acute toxicity of *Irvingiagabonensis* seed extract was determined through intra-peritoneal administration of the ethanolic extract of *Irvingiagabonensis.*18 mice were used, and they were divided into 6 groups (3 mice in each group). Each group received 500, 400, 300, 200, 100, 50mg/kg of the ethanolic extract dissolved in 20% tween 80. All experimental animals were observed for physical signs of toxicology such as convulsion, gasping, writhing, death within 24 hours. There was no death recorded after 24 hours (0% lethal)

Experimental protocol

The 35 rats dividedinto seven different groups received dosages as follows: Group A was given distilled water, Group B1 was given 0.85mls of asolution formed by dissolving 2grams of ground *Irvingiagabonensis* seeds in 100mls of water (low dosage), Group B2 was given 2.55mls of asolution formed by dissolving grams of ground *Irvingiagabonensis* seeds in 100mls of water (high dosage). Group C1 was given 0.80mls of ethanolicextract of ground *Irvingiagabonensis* seeds (low dosage), Group C2 was given 2.70mls of ethanolic extract of ground *Irvingiagabonensis* seeds (high dosage), Group D1 was given 0.90mls of N-Hexane extract of ground *Irvingiagabonensis* seeds (low dosage), Group D2 was given 2.55mls of N-Hexane extract of ground *Irvingiagabonensis* seeds (high dosage).

The dosages were calculated and administered based on theaverage body weight of animal in each group using the formula:

$$\frac{\text{Body Weight}\times\text{dose (10\% of } LD_{50})}{1000 \;\; 10\text{mg/ml(stock solution)}}$$

The dosages of *Irvingiagabonensis* were given to the animals once daily for 21 days. During the experimental period, the body weight of the rats was measured after every 6 days. At the end of 21 days, the rats were sacrificed using chloroforms inhalation method, after which the stomach were harvested and rinsed in normal saline to remove excess blood before fixing in 10% buffered formalin in labeled sterile bottles for one week.

Tissue processing

After 21 days of administration, the animals were starved for 24 hrs in order to empty bowels before sacrifice. The rats were anaesthetized with cholorofoam soaked in cotton wool in the dessicator. The stomach of each rat were removed and fixed in 10% buffered formalin in a well labeled tissue bottles. This is to prevent autolytic changes and also harden the tissue for trimming.

Dehydration: The tissues were then transferred to ascending grades of alcohol; two changes of 95% alcohol for a period of 50 minutes and two changes of absolute alcohol for a period of 50minutes. This was done to remove water in the tissue which is not miscible with paraffin wax.

Clearing: the tissues were cleared using xylene at two changes of 50 minutes each. This was done to remove alcohol from the tissue and to prepare the intracellular and intercellular spaces for proper infiltration.

*Infiltration***:** the tissues were impregnated with paraffin wax in an oven at 55^0c - 65^0c. the wax penetrate the tissue thereby enabling the intracellular spaces of the stomach tissues to be filled with paraffin wax earlier cleared by xylene and enable embedding to take place.

Embedding: Tissues were then oriented in a supporting medium (paraffin wax), and then allowed to solidify, melted paraffin wax was poured into metal block that were greased with glycerine to prevent the wax from adhering to the molds.

Mounting and Sectioning: The embedded liver tissues were mounted on wooden blocks for proper sectioning. The paraffin embedded stomach tissue was sectioned using a rotary microtome at 3.5-5.0um thickness into a hot water bath. This is to create a conventional current to enable the ribbons to spread, and then pick up by slides smeared with egg albumen, which serve as gum and for proper orientation of the tissue on the slides.

Staining technique

The staining method used was the Haematoxylin and Eosin staining technique.

Solutions required: Haematoxylin, eosin, xylene, alcohol (70%, 95%, and absolute alcohol), 1% acid alcohol, DPX (moutant)

Procedure involved

Dried slides of the stomach were dewaxed in two changes of xylene for five minutes each. It was then hydrated in descending grade of alcohol (from absolute to 70% alcohol) for one minute each.

Sections were then washed in running tap water for ten minutes
Tissues were then stained in haematoxylin for 10 minutes

Sections were then washed in running tap water and differentiated in 1% acid alcohol briefly, washed in water and blue. Slides were then counter stained in eosin for 3 minutes, this was done to provide contrast background for the staining of tissue components (as haematoxylin stains nuclei and eosin stain the cytoplasm).

The sections are then washed and dehydrated in ascending grades of alcohol, cleared in xylene and mounted in DPX; cover slipped and allowed to dry. The slides were subsequently viewed under the light microscope and photomicrographs were taken.

Statistical analysis

Data were analyzed using descriptive statistical tool, primer, version 3.0. Microsoft excel package was used for graph

RESULTS AND DISCUSSIONS

Table-1: Comparison of Changes in Average Body Weight of Albino Rats in Grams.

Groups	Subgroups	Week 1	Week 2	Week 3
A		155	177	209
B	B_1	168	178	193
	B_2	167	177	197
C	C_1	157	173	173
	C_2	177	195	214
D	D_1	178	175	201
	D_2	171	184	203

Keys
A = control group given distilled water
B1 = rats given 0.85mls of asolution formed by dissolving 2grams of ground *Irvingiagabonensis*seeds in 100mls of water (low dosage).
B2 = rats given 2.55mls of asolution formed by dissolving 4grams of ground *Irvingiagabonensis*seeds in 100mls of water (high dosage).
C1 = rats given 0.80mls of ethanolic extract of ground *Irvingiagabonensis* seeds (low dosage).
C2 = rats given 2.70mls of ethanolic extract of ground *Irvingiagabonensis* seeds (High dosage).
D1 = rats given 0.90mls of N-Hexane extract of ground *Irvingiagabonensis*eeds (low dosage).
D2 = rats given 2.55mls of N-Hexane extract of ground *Irvingiagabonensis*seeds (high dosage).

Table-2: Comparison of differences in initial and final Average Body Weight of Albino Rats in Grams

Groups	Subgroups	Initial weight	Final weight	Difference
A		155	209	54
B	B_1	168	193	25
	B_2	167	197	30
C	C_1	157	173	16
	C_2	177	214	37
D	D_1	178	201	23
	D_2	171	203	32

Keys:
A = control group given distilled water
B1 = rats given 0.85mls of asolution formed by dissolving 2grams of ground *Irvingiagabonensis*seeds in 100mls of water (low dosage).
B2 = rats given 2.55mls of asolution formed by dissolving 4grams of ground *Irvingiagabonensis*seeds in 100mls of water (high dosage).
C1 = rats given 0.80mls of ethanolic extract of ground *Irvingiagabonensis* seeds (lowdosage).
C2 = rats given 2.70mls of ethanolic extract of ground *Irvingiagabonensis* seeds (highdosage).

D1 = rats given 0.90mls of N-Hexane extract of ground *Irvingiagabonensis*seeds
(low dosage).
D2 = rats given 2.55mls of N-Hexane extract of ground *Irvingiagabonensis*seeds
(high dosage).

Hematoxylin and Eosin (H&E) Method for General Demonstration of Stomach

Hematoxylin and Eosin stain (H&E) is used routinely with all tissue specimens to reveal underlying tissue structures and condition. It stains clearly all cell structures including the cytoplasm, nucleus, organelles and extra-cellular components. The Hematoxylin and Eosin stain method uses two dyes, hematoxylin and eosin andthis combination is used as the dyes stain different tissue component.

In the demonstration of the fundus and body of the stomach, the simple columnar epithelium whichis specialized for secretion could be identified along with the gastric mucosa containing gastric pits, which are surface invaginations and also serve as the ducts of the underlying intrinsic gastric glands. Cell types that could be seen in the fundus and body include:

- Mucus-secreting cells: These cells form the surface epithelium and extend inward to line the gastric pits. Nuclei are basal, and the supranuclear cytoplasm containing mucinogen granules appears clear or vacuolated with H & E stain. Mucous neck cells occur in the junctionalregion of the gastric pits and glands
- Parietal cells: These pyramidal or spherical cells appear wedged in between other cells of the gastric glands. They are characterized by their finely granular acidophilic cytoplasm due to an abundance of mitochondria, and by their central, spherical nucleus
- Chief (zymogen) cells: These cells are involved in the secretion of enzymes pepsinogen (pepsin in the active state). As is characteristic of cells involved in protein synthesis and secretion, these cells contain basophilic cytoplasm, particularly at their base due to the extensive development of rough endoplasmic reticulum.

The mucosa is identified with the luminal surface mucous secreting cells, the gastric pits and the cells lining those Parietal cells are particularly prominent, and chief cells and mucous neck cells are present. The muscularismucosae, forms a boundary between the mucosa and submucosa, and it containthe loose connective tissue which surrounds the gastric pits and the blood vessels in the submucosa. The cell bodies and nerve fibers of the Meissner's plexus are found in the submucosa. The muscularisexterna is found smooth muscle oriented in several different planes. A serosa covers the external surface of the gland. The myenteric plexus (Auerbach's plexus) is located between the external and adjacent inner layers of smooth muscle.

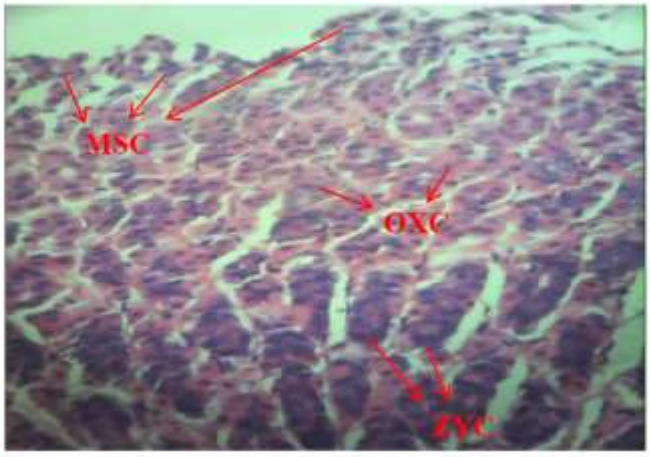

Plate I (H & E, X100)

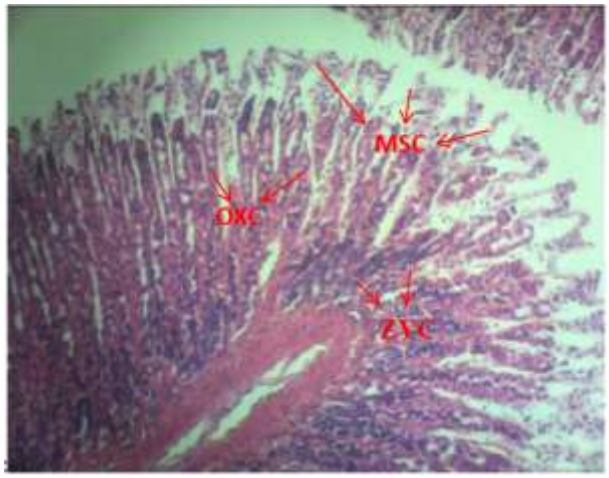

Plate II (H & E, X400)

Plates I and II. Photomicrograph of cross sections of the body and fundus of the stomach of adult male albino *Wistar* rats given distilled water. Group A, Sections revealed intact mucous-secreting-cells (MSC) that cover the luminal surface of the stomach and line the gastric pits. Both the oxyntic (OXC) and zymogenic (ZYC) cells are also intact. Normal cellular architecture of the gastric pits (GP), etc. were observed. Inference: There was no evidence of cellular abnormality.

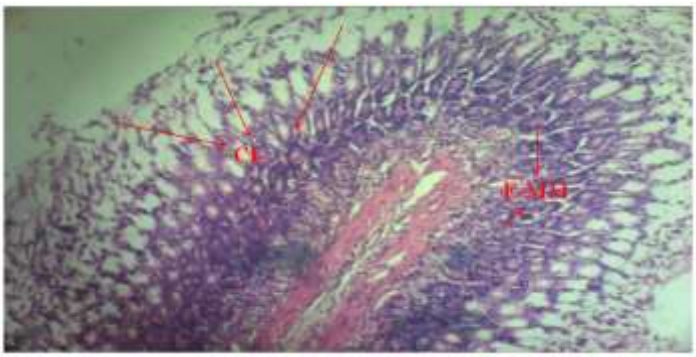

Plate III. (H & E, X100)

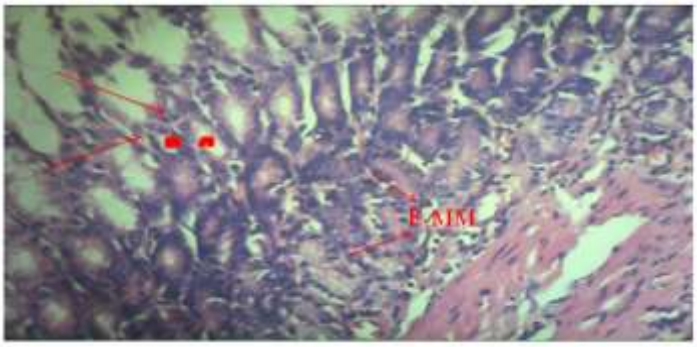

Plate IV (H & E, X400)

Plates III and IV: Photomicrograph of cross sections of the body and fundus of the stomach of adult male albino *Wistar* rats given 0.85mls of asolution formed by dissolving 2grams of ground *Irvingiagabonensis* seeds in 100mls of water (low dosage). Group B1, Sections revealed mild to moderate inflammation of the mucous-secreting cells (I-MSC) and diffuse mildly erythematous mucosa (E-MM).Inference: There was inflammation of the stomach lining (gastritis).

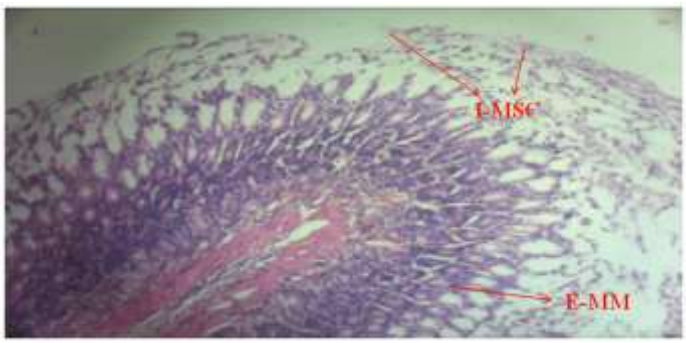

Plate V. (H & E, X100)

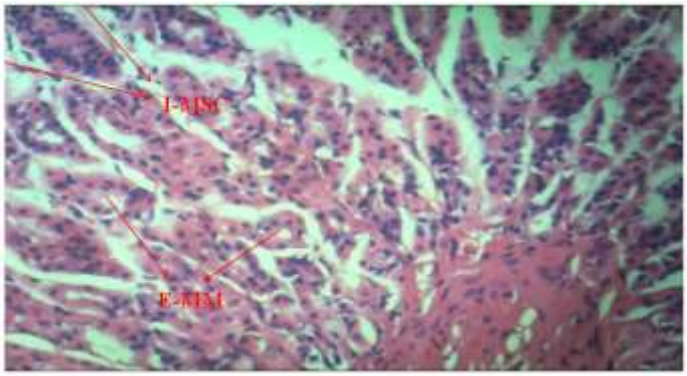

Plate VI. (H & E, X400)

PlatesV and VI: Photomicrograph of cross sections of the body and fundus of the stomach of adult male albino *Wistar* rats given 2.55mls of asolution formed by dissolving 4grams of ground *Irvingiagabonensis*seed in 100mls of water (high dosage). Group B2, Sections revealed moderate inflammation of the mucous-secreting cells (I-MSC) and diffuse erythematous mucosa (E-MM).Inference: There was inflammation of the stomach lining (gastritis).

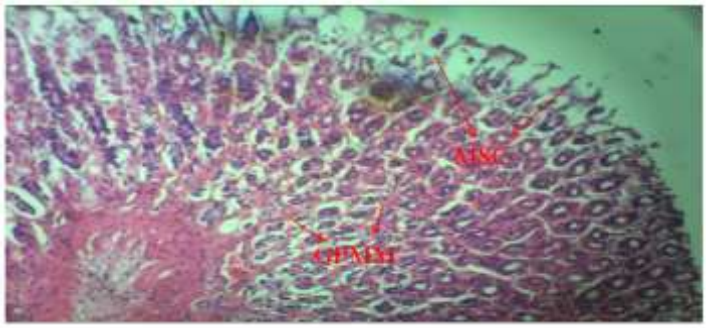

Plate VII. (H and E, X100)

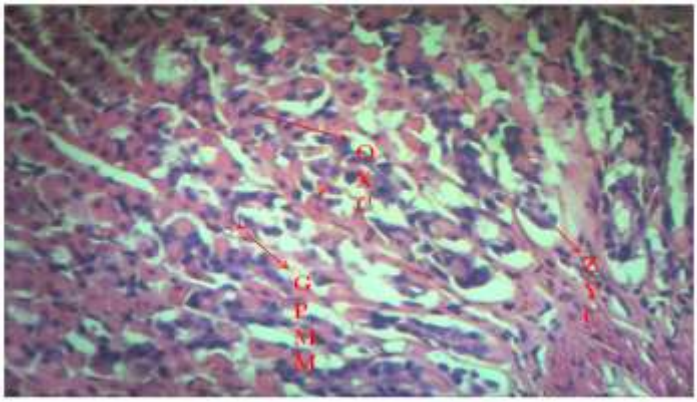

Plate VIII. (H & E, X400)

PlatesVII and VIII. Photomicrographof cross sections of the body and fundus of the stomach of adult male albino *Wistar* rats given 0.80mls of ethanolic extract of ground *Irvingiagabonensis* seeds (low dosage). Group C1, Sections revealed intact mucous-secreting-cells (MSC), and granular pattern mucosa (GPMM). The oxyntic (OXC) and zymogenic (ZYC) cells are also intact. Inference: This is a variant of normal mucosa. There was no evidence of cellular abnormality.

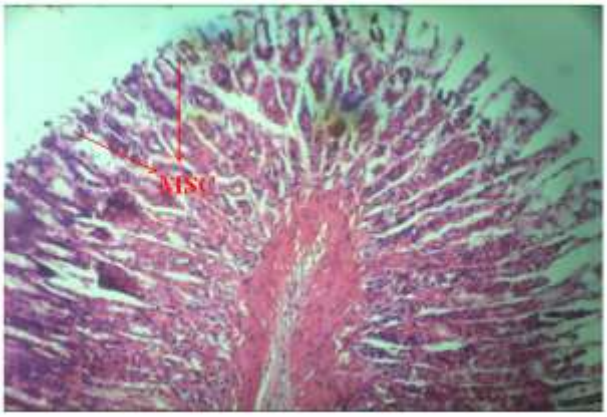

Plate IX. (H and E, X100)

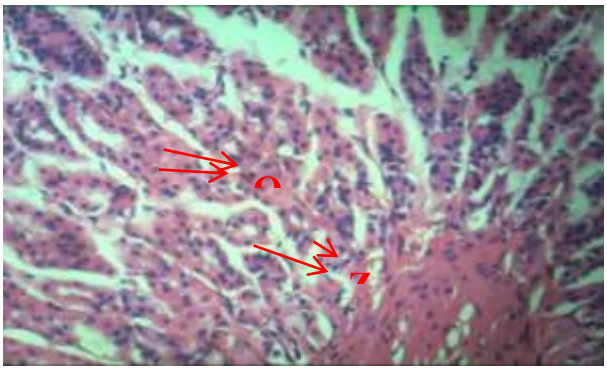

Plate X. (H and E, X400)

Plates IX and X: Photomicrograph of cross sections of the body and fundus of the stomach of adult male albino *Wistar* rats given 2.70mls of ethanolic extract of ground *Irvingiagabonensis* seeds (high dosage). Group C2, Sections revealed intact mucous-secreting-cells (MSC). The oxyntic (OXC) and zymogenic (ZYC) cells are also intact. Inference: There was no evidence of cellular abnormality.

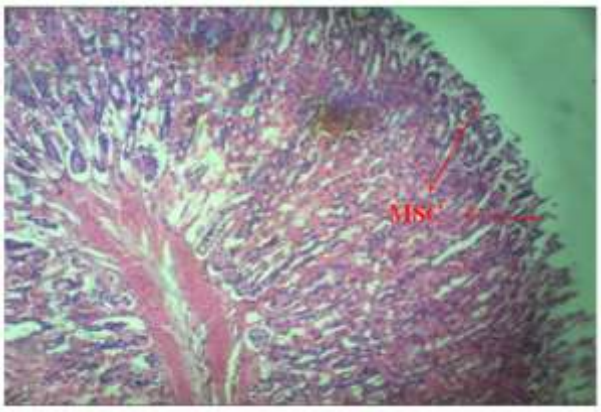

Plate XI. (H and E, X100)

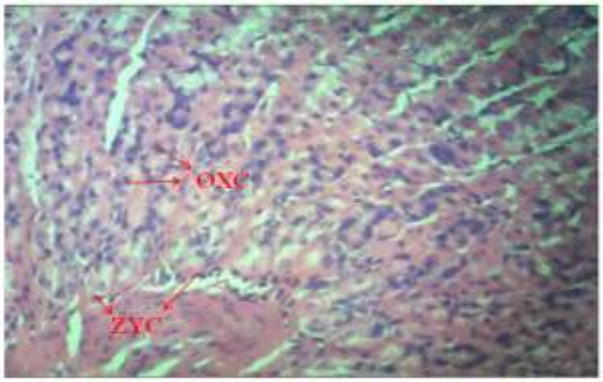

Plate XII. (H and E, X400)

Plates XI and XII: Photomicrograph of cross sections of the body and fundus of the stomach of adult male albino *Wistar* rats given 0.90mls of N-Hexane extract of ground *Irvingiagabonensis* seeds (low dosage).Group D1, Sections revealed intact mucous-secreting-cells (MSC). The oxyntic (OXC) and zymogenic (ZYC) cells are also intact. Inference: There was no evidence of cellular abnormality.

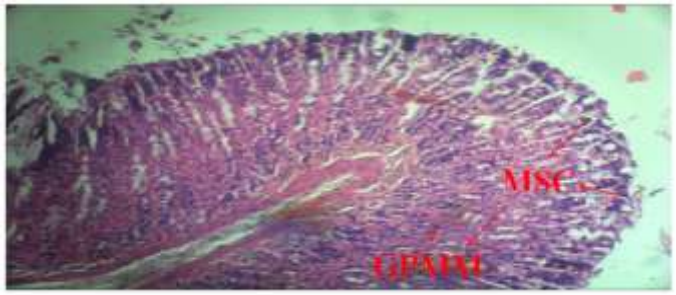

Plate XIII. (H and E, X100)

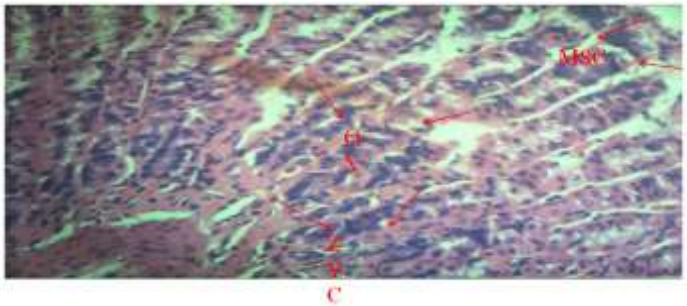

Plate XIV. (H and E, X400)

Plates XIII and XIV: Photomicrograph of cross sections of the body and fundus of the stomach of adult male albino *Wistar* rats given 2.55mls of N-Hexane extract of ground *Irvingiagabonensis* seeds (high dosage).Group D2, Sections revealed intact mucous-secreting-cells (MSC). The oxyntic (OXC) and zymogenic (ZYC) cells are also intact. Inference: There was no evidence of cellular abnormality.

Present day cultivation of *Irvingiagabonensis*h as being driven by its high demand as food components, but more recently by its demand as nutritional supplements. Majority of *Irvingiagabonensis* produced is desired for cooking in households and restaurants with ogbono soup featuring on the menus of most restaurants in Nigeria. The demand in southern Nigeria alone was estimated at 80,000 tons per year in 1997 [42].

From the photomicrograph of the stomach of the experimental animals, the following was observed; plate i and ii which serves as the control group which showed normal histological feature of a stomach indicating intact mucous-secreting-cells (MSC) that cover the luminal surface of the stomach and line the gastric pits. Both the oxyntic and zymogenic cells were also intact showing normal cellular architecture of the gastric pits. Plate's iii and iv revealed inflammation of the mucous secreting cells, presence of red irritating lining due to diffuse mildly erythematous mucosa.

The diseases of the stomach mucosa include gastritis and gastroparesis which involve the

inflammation of the gastric mucous cells which is usually accompanied by abdominal pains, nausea, vomiting, and occasionally, belching, bloating, loss of appetite and indigestion. This is the usual feeling after a meal of *Irvingiagabonensis* seed.

Histological result in for group C1 and C2 shows no cellular alteration, but a change in the mucosa pattern to granular. This finding is in agreement with studies done by [43, 6] that consumption of *irvingiagabonensis* seed extract delayed stomach emptying effect.

Our finding on body weight showed that *Irvingiagabonensis*had significant effect on the body weight of the experimental animals, as the animal increased in body weights throughout the period of administration. This result is contrary to that of Ngondi, *et al.* [44] that reported that *Irvingiagabonensis* significantly reduced the body weight of obese patient.

In this study, the cytoarchitectural integrity of the stomach of the animals was examined. The result obtained for the experimental group (treated with *Irvingiagabonensis*) was compared with that obtained from the control group (no treatment). The control group showed normal histological features of the stomach with normal areas of the fundus , mucosa and structural composition of the stomach. While that of experimental group showed remarkable differences with evidence of inflammation and change in pattern of mucous secreting cells. It was also observed to be dose dependent.

CONCLUSION

From the observations made in this research, it can hereby be concluded that *Irvingiagabonensis*in raw ground form/state as an inflammatory effect on the gastric microstructure with noticeable areas of inflamed mucous secreting cells and diffuse mildly erythematous mucosa which leads to gastritis

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Author's contributions

We write to state that all authors have contributed significantly, and that all authors are in agreement with the contents of the manuscript. 'Author A' (Christopher C. Mbadugha) designed the study and protocol, 'Author B' (Ekpedeme Paulinus Udoh) wrote the first draft of the manuscript and managed the literature search, 'Author C' (John Nwolim Paul) examined the manuscript for intellectual content, 'Author D' (Tarimobo Michael Otobo) managed the analyses of the study. All authors read and approved the final manuscript.

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