INTERNATIONAL JOURNAL OF
PHARMACEUTICAL RESEARCH AND BIO-SCIENCE

FORMULATION AND AVAILABILITY OF SOME ANTI-OBESITY MATERIALS

ESMAIL M. RAMADAN, GALAL M. ABD ELGHANI, DINA N. NADY

Faculty of Pharmacy, Mansoura University, Pharmaceutical Department, Mansoura, Egypt.

Accepted Date: 04/05/2016; Published Date: 27/06/2016

Abstract: Obesity is a disease in which excess body fat has accumulated to the extent that it may have adverse effect on health. It is commonly caused by a combination of excessive food energy intake and lack of physical activity or may be due to genetic susceptibility. Some natural by-products in the form of tablets were selected as anti-obesity agents including (Sugarcane bagasse, Wheat straw, Pomegranate peel and Treated shrimp shell). Ashes were collected, washed, treated, dried, powdered and characterized for their moisture and glucose content. Flow properties of powders, particle size analysis, flowability, bulk and tap density, bulk and tap volume as well as compressibility were also characterized. Powders of different ashes were compressed into tablets and the mechanical and physical properties of these tablets were evaluated. Tablets were also tested for their in vitro availability in different pH media including distilled water, 0.1M HCl (pH 1.2) and phosphate buffer (pH 7.2). The results revealed that the release of glucose was pH independent in case of sugarcane bagasse and wheat straw tablets, while it was pH dependent in the case of pomegranate peel and treated shrimp shell tablets. The fastest dissolution rate was obtained by sugarcane bagasse tablets, while wheat straw and treated shrimp shell tablets show the slowest one. Bioavailability study was evaluated in rats by feeding each group on one type of diet represented in normal rat diet mixed with equivalent to 10% of sugarcane bagasse wet granulated powder (F_1), wheat straw wet granulated powder (F_2), 10% commercial bran capsule contents and plain normal rat diet. Weight of individual rat was recorded each three days for two months and the percent loss or gain in weight was calculated. Results indicate that maximum percent loss in weight was achieved by sugarcane bagasse wet granulated powder (23.92%) followed by wheat straw wet granulated powder (14.62%), however commercial bran capsule provides least percent increase in weight (13.36%), while the increase in weight of control group was (25.16%). Analyzing serum glucose, triglycerides and total cholesterol indicates that maximum reduction in all the tested parameters was obtained by sugarcane bagasse wet granulated powder followed by wheat straw wet granulated powder and finally commercial bran compared to the control group. Serum glucose was (66.57, 80.20, 87.25 & 100.75 mg/dl), triglycerides level was (97.5, 106.25, 119.63 & 127.76 mg/dl), total cholesterol level was (55.9, 63.5, 70.75 & 72 mg/dl) for sugarcane bagasse, wheat straw, commercial bran and control groups respectively.

Keywords: Obesity, Sugarcane bagasse, Wheat straw, tablets, in vitro release, in vivo study

Corresponding Author: MR. GALAL M. ABD ELGHANI

Access Online On:

www.ijprbs.com

How to Cite This Article:

Galal M. Abd Elghani, IJPRBS, 2016; Volume 5(3): 49-78

PAPER-QR CODE

INTRODUCTION

Obesity is a medical condition in which excess body fat has accumulated to the extent that it may be hazard for health [1].It is measured by body mass index (BMI), the simple index of weight for height calculated by dividing person's weight in kilograms by the square of his height in meters (kg/m2) [2]. Peoples are classified according to their BMI into: <18.5 Underweight, 18.5-24.9 Normal range, 25-29.9 Overweight, 30-34.9 Obesity I, 35-39.9 Obesity II and ≥40 Obesity III [3]. The main and direct reason for obesity is the high dietary caloric intake, which felt to be due to an easily accessible and fast diet [4]. Such types of diet are characterized by, high energy and low dietary fiber content, which produces lower satiety than low-energy dense foods [5], combined with lack in physical exercise due to increasing reliance on cars and automatic manufacturing [6,7].

A limited number of cases may be related to genetic factor or psychiatric illness [8]. Also, it was found that some medications may be associated with weight gain for example: atypical antipsychotics (clozapine), beta adrenergic blockers (propranolol), lithium, sodium valproate, sulphonylureas(including chlorpropamide, glibenclamide, glimepiride and glipizid), thiazolidinediones (pioglitazone)and tricyclic antidepressants(amitriptyline) [9]. Some other causes may lead to obesity like insufficient sleep, endocrine disturbance, environmental pollution that interferes with lipid metabolism and pregnancy at later age, which may cause susceptibility to obesity in children [10].

Many health problems caused by obesity such as diabetes, cardiovascular diseases and non-alcoholic fatty acid diseases [1,11],increased risk of developing atrial fibrillation [12] and cardiological problems such as (angina, myocardial infarction, congestive heart failure, high blood pressure and abnormal cholesterol level) [1]. In addition, obesity is considered to be a reason for increased risk of leukaemia [13], breast cancer [14-16], gallbladder cancer [17], ovarian cancer [18], pancreas cancer [19], prostate cancer [20], colon cancer [21,22], gastroesophagus reflux disease and its related complications including, adenocarcinoma [23,24] and renal cancer [21]. Different approaches for obesity management were followed such as combination of diet programs and physical exercise [25].

Medications are another pathway to overcome obesity. They employ their role in weight reduction through different mechanisms including appetite suppression, alternation of food absorption or consumption [26-28] for example sibutramine, fenfluramine and dexfenfluramine, rimonabant, phentermine, lorcaserin and diethyl propion, but unfortunately most of them have dangerous adverse effects on health that results in their withdrawal from market. Only orlistat is safe but with limited anti-obesity effect. In severe cases, bariatric surgery may be necessary, although it has different risks [29].

Sugarcane is the largest crop in the world cultivated mainly in tropical and subtropical regions and used as the major source of sucrose production [30], which results in producing large amounts of sugarcane bagasse as by-product. It may represents a potential source of inexpensive pharmaceutical excipients due to containing cellulose as a major component [31], while wheat straw is a fibrous by-product that is usually obtained after using wheat grain in different food industries. Both by-products are rich in cellulose, where it represents about33.29 &33.7- 40%for sugarcane bagasse and wheat straw respectively [32,33]. In addition, pomegranate peel represents about 67% of the whole fruit [34] and consists of about 26.22% cellulose and 10.88% hemicellulose [35]. Cellulose is a water insoluble polymer, with a digestive enzymes resistance in the small intestine. However, it can be fermented to certain degree in the large intestine producing short chain fatty acids [36]. In addition, upon contact with water, cellulose can absorb five times its own weight by capillary action [32]. Due to its indigestibility and its high swallowing capacity, cellulose was suggested to decrease human caloric intake that shares in obesity management process. Shrimp shells are very rich source of chitin, a homo-polymer of N-acetyl D-glucosamine residues linked by 1-4 bonds, the most natural abundant polysaccharide after cellulose that also shows anti-obesity effect [37].

Materials and methods:

Materials:

Raw materials: all raw materials were fresh and supplied from local market. Sugarcane bagasse was obtained from sugarcane stalks after crushing, wheat straw was obtained from wheat plant, pomegranate peel was obtained from pomegranate fruits and shrimp shell was obtained from shrimps. Commercial bran capsules of 400 mg were purchased from Arab Company for Pharmaceuticals & Medicinal Plants (MEPACO-MEDIFOOD), Enshas El Raml-Sharkeya, Egypt. Other materials: Gelatin, Talc, starch, Ethyl acetate, Sulfuric acid, D-Glucose, Sodium dibasic orthophosphate, Potassium dihydrogen orthophosphate and hydrochloric acid were obtained from ADWIC, EL NASR pharmaceuticals and chemicals co., Cairo, Egypt. Emcompress was purchased from Mendell co., England and anthrone from Oxford laboratory, Mumbai, India. Cholesterol, CHOD-POD, and triglycerides, GPO-POD, Enzymatic colorimetric kits, SPINREACT, Glucose-Liquizyme, GOD-PAP Enzymatic colorimetric kits, SPECTRUM. All materials and reagents were of analytical grade

Equipment:

Electric mill, ELECTRA, EK- 260, Japan, Electromagnetic sieve shaker (RP200N, Spain), A series of sieves of particle size (106 – 150 µm, 150 – 250 µm , 250 – 315 µm, 315 – 425 µm, 425 – 600 µm and 600 µm – 1.7 mm), Hot air oven, Heraeus G S model B 5042, West Germany, Electric balance, No. 06805, 1/4 AMP, U.S.A, Granulate flow tester G. D. T. 43449, Single punch tablet

machine, Tablet Hardness tester, Roche friabilator, Tablet disintegrator (Erweka –Apparatebau, G. m. b. H, Germany), Micrometer, Mitutoyo Corporation, Japan, PH-meter, Beckman instrument Fullerton, CA 92634, Eight Jars, Abotta, rotating paddle, tablet dissolution test apparatus, USA, Spectrophotometer UV-VIS DOUBLE BEAM PC UVD- 2950, LABOMED, INC, U.S.A.

Method:

1. Preparation of different ashes powders:

Ashes including sugarcane bagasse, wheat straw and pomegranate peel were collected, washed with tap water several times to remove dust then washed with distilled water. Using scissor, the ashes were cut into small pieces then exposed to sunlight for 10 days for complete drying. Ashes were milled using electrical mill, the obtained powder was dried in a hot air oven at 40 ºC for complete drying.

2. Preparation of shrimp shell powder:

Shrimp shell waste materials were collected, scraped free of loose tissue, washed with cold water and dried in sun for 3 days then demineralization by suspending shells in 4% HCl at room temperature in the ratio of 1:14 (w/v), after 36 hours the shells were rinsed with water to remove acid and calcium chloride. Then, deproteination process was carried out by treating the demineralized shells with 5% NaOH at 90 °C for 24 hours with a solid to solvent to solid ratio of 1:12 (w/v). The residue was collected and washed for neutrality in running tap water followed by distilled water. Then, it was dried in sunlight for three days. The dries residue was milled using electrical mill into powder and placed in a foil plates, placed in hot air oven at 40 °C for complete drying [38]. Then, different powders were stored in air tight, amber glass containers for further studies.

Powder characteristics

1- Loss on drying (LOD):

Weights of the ashes were recorded initially and then dried in a hot air oven at 40ºC to constant weight. The water mass (or moisture) was the difference between the weights of the wet and dry samples [38].

$$\text{Moisture powder percent} = \frac{Wet\ powder\ weight - Dry\ powder\ weight}{Wet\ powder\ weight} \text{ X } 100$$

2- Particle size analysis:

Ashes were analyzed for particle size using electromagnetic sieve shaker. A series of sieves of particle size range (106 – 150 µm, 150 – 250 µm, 250 – 315 µm, 315 – 425 µm, 425 – 600 µm and 600 µm – 1.7 mm) were used in order to obtain a suitable particle size range for tablets compression. Sieves were arranged in a descending order according to the particle size range, about 2.5 kg of each powder was placed individually as amounts of 100 g at the top sieve and sieved for 10 minutes and then the particles of each size range were removed and filled in a separate airtight, amber glass container.

3. Compressibility of powder:

The compressibility index and Hausner ratio are determined by measuring both the bulk volume (V_0) and tapped volume (V_f) of a powder using the following equation [39].

$$\text{Compressibility index} = \frac{V_0 - V_f}{V_0} \text{ X } 100$$

$$\text{Hausner ratio} = \frac{V_0}{V_f}$$

Only 15 g of powder were placed in 100 ml volumetric cylinder, then the bulk volume (V_0) was determined, the cylinder was tapped several times until the final volume of powder (tapped volume, V_f) is constant and then measured.

The compressibility index and Hausner ratio are calculated also using measured values of bulk density (ρ_{bulk}) and tapped density (ρ_{tapped}) **[39].** Bulk density (ρ_{bulk}) = weight of powder/ bulk volume, while tapped density (ρ_{tapped}) = weight of powder/ tapped volume. The compressibility index and Hausner ratio were measured for each size range of each powder individually.

$$\text{Compressibility index} = \frac{\rho_{tapped} - \rho_{bulk}}{\rho_{tapped}} \text{ X } 100$$

$$\text{Hausner ratio} = \frac{\rho_{tapped}}{\rho_{bulk}}$$

4. Angle of repose

The used method was to fix the height of the funnel through which the powder flow, while the diameter of the powder cone may be allowed to vary as the pile forms [39]. Where amount of powder was placed in the funnel of apparatus then allowed to flow until a cone of constant height was obtained. A circle of the obtained cone base was drawn then radius was measured. Angle of repose was measured for each size range of each powder individually. Results are the

average of three determinations and the angle of repose is calculated according to the following equation (Tables 2-7).

$$\text{Tan } (\alpha) = \frac{\text{Height}}{0.5 \text{ base}}$$

Preparation of tablets containing different ashes:

Flowability and compressibility results of different particle size ranges illustrate that particle size range of choice was (150 – 250 µm) in case of sugarcane bagasse, pomegranate peel and treated shrimp shell powders, while was (106 – 150 µm) in the case of wheat straw powder.

Ingredients included in each formula vary from one ash to another according to the characteristics of the powder particles, binding force between them and their disintegration. Powders of each formula were geometrically mixed and lubricated, then compressed into tablets by direct compression using a single-punch flat faced tablet machine. This method was suitable only for pomegranate peel and treated shrimp shell powders.

To prepare acceptable tablets for other ashes wet granulation of sugarcane bagasse powder with 10% gelatin solution, and wheat straw powder using 12.5% gelatin solution as the granulating liquid. Gelatin solution was prepared by dispersing 10 g of gelatin powder in 90 g distilled water. Let for 1 hour for complete soaking of gelatin powder to obtain slurry of 10 % w/w gelatin solution (to prepare 12.5% w/w gelatin solution same procedure was applied by dispersing 12.5 g gelatin in 87.5 g distilled water). The obtained slurry was added to the ash powder in a mortar and mix until a suitable paste was obtained. Press this paste through a sieve no. 12 to form granules. Spreading the wet granules on foil sheets, place in hot air oven at 37º C for complete drying. The obtained dried granules were removed from sheets, milled in an electrical mill. The wet granulated powders passed through a series of sieve to obtain size ranges (106-150 and 150 - 250 µm) in the case of wheat straw and sugarcane bagasse respectively, in order to obtain powder particles of suitable size range that can be easily compressed into tablets (Table 8).

The tablet weight was adjusted to be 600 mg. The compression force was adjusted to produce tablets with hardness greater than 4 kg/cm2. The prepared tablets were stored away, protected from moisture in airtight amber glass containers and left for 24 hour for elastic recovery before their evaluation. The tablet formulations are listed in Table 9.

1. Calibration curve of glucose in different pH media

Stock solutions were prepared by dissolving 1000 mg glucose in 100 ml distilled water, 0.1M HCl (pH 1.2) or phosphate buffer (pH 7.2), pipette out 10 ml from these solutions, then dilute to 100 ml using the same previous media. Pipette out 10 ml again then dilute to 100 ml as before

to obtain a concentration of 100 µg/ml. Serial dilutions were prepared from these stock solutions to obtain concentrations of 10, 13.33, 16.66, 20, 23.33 and 26.66 µg/ml. The calibration curve for glucose was constructed by plotting the measured absorbance at (λmax= 620 nm) of the glucose versus the corresponding concentrations in different selected media. Results are the average of three determinations and illustrated in Figures 1-3

2. Determination of average glucose content

Ten tablets were randomly selected, weighed and the average tablet weight was determined. The tablets were ground in a mortar and pestle. Certain amount of powder equivalent to the average tablet weight of each ash was extracted with 100 ml of distilled water in a 250 ml closed flask for twenty-four hours, shaking frequently during first six hours and then allow to stand for eighteen hours [40], filter using Millipore filter 0.45µm. Then, 0.4 ml suitably- diluted sample was mixed with 0.1 ml freshly prepared 2% anthrone in ethyl acetate in a 10 ml glass test tube. One ml of 95-97% sulphuric acid was added into each sample slowly then shake the test tube well. The reaction tubes were cooled for 10 minutes to room temperature and then assayed spectrophotometrically at 620 nm [41].

3. Evaluation of tablets:

The prepared tablets were evaluated for their uniformity of weight, thickness, hardness, friability percent, content uniformity, disintegration time and dissolution rate.

3. 1. Uniformity of tablets weight:

Twenty tablets were selected randomly, dedusted and weighted carefully. The mean weight and standard deviation were calculated [39].

3. 2. Thickness of tablets

The thickness of each weighed tablet was determined by means of a micrometer [39]. The mean thickness and standard deviation values were calculated.

3. Hardness test:

The test was performed using Tablet Hardness tester (Erweka Apparatebau, T. B. 24, G.m.b.H, Germany). Place the tablet between the jaws, taking in account, each measurement orient the tablet in the same way with respect to the direction of application of weight. Enough weight was applied to cause tablet breakage.

Results were taken only from tablets, which split cleanly into two halves without any sign of lamination. Carry out the measurement of six tablets, taking care that all fragments of tablets

have been removed before each determination [39]. The mean hardness and standard deviation values are calculated (Table 11).

3. 4. Friability test:

The test was performed on twenty tablets using Roche friabilator (Erweka –Apparatebau, G.m.b.H, Germany). The tablets were carefully dedusted prior to testing. Accurately weigh the tablets sample, and then place them in the drum. Rotate the drum at a rate of 25 revolutions per minute for 4 minutes, and remove the tablets. Remove any loose dust from the tablets as before, and accurately weigh [39] (Table 11).

The percentage friability was calculated using the following equation

$$\% F = \frac{W_1 - W_2}{W_1} X\ 100$$

Where: % F represents the percent of friability, W1 and W2 are the initial and final weights of tablets, respectively.

3. 5. Content uniformity of glucose:

Ten tablets were randomly selected. Each of the ten tablets was assayed individually for its glucose content by crushing in a clean mortar and pestle, then complete as mentioned under determination of average glucose content.

3. 6. Disintegration of tablets

The test was performed on six tablets using Erweka Tablet disintegrator (Erweka-Apparatebau, ZT3, Germany). Place one tablet in each of the 6 tubes of the basket and operate the apparatus using distilled water as a disintegrating medium, maintained at 37±2 ºC, as an immersion fluid [39]. The disintegration time was taken to be the time that tablets fragmented into granules that can pass through the mesh screen. The mean disintegration time and the standard deviation values were calculated. Results are the average of three determinations and listed in Table 11.

3. 7. In vitro drug release:

The in vitro release of glucose from the prepared tablets were determined according to the II-paddle dissolution method [39] at a rotating speed of 100 rpm in 500 ml distilled water, 0.1M HCl (pH 1.2) and phosphate buffer (pH 7.2) at 37 ± 0.5 ºC using dissolution tester (Eight Jars, Abotta, rotating paddle, tablet dissolution test apparatus, USA). One tablet in each cell was placed. At appropriate time intervals of 10, 20, 30, 40, 50, 60, 90, 120 and 180 minutes, 5 ml samples were withdrawn from the dissolution medium. Each sample was replaced with an

equivalent volume of the fresh dissolution medium at the same temperature in order to maintain the volume in the cell constant. The withdrawn samples were filtered using Millipore filter (0.45µm) and analyzed spectrophotometrically for their glucose content directly by measuring the absorbance at 620 nm using anthrone sulphuric assay [41].

Statistical analysis was carried out using one-way analysis of variance (ANOVA) test followed by Tukey Kramer test for comparison [42]. Results are the average of three determinations and results are illustrated in Figures 4-10

In-Vivo Study on the Anti-Obesity Activity of Some Selected Ashes Formulations The protocol of this study complies with ethical principles and guidelines for the care and the use of laboratory animal adopted by the "Research Ethical Committee", Faculty of pharmacy, Mansoura University.

Experimental animals:

Thirty two female three months old rats weighing 145 – 178 gram were obtained from the Experimental Animal House, Faculty of Pharmacy, Mansoura University. Rats were housed eight per cage at 30 °C on a 12/12 h light/dark cycle and had free access to standard food and water for seven days for their adaptation.

Methodology:

Rats were divided into four groups of initial weights of (171.38±3.5, 162.63±2.2, 157.13±1.64 & 149.63±3.38 g) for G1, G2, G3 & G4 respectively. Rats were fed on high fat diet containing 16% crude protein, 4o% fat and 2.24% fibers and minerals for 21 days to induce obesity [43]. Rats weight after feeding high fat diet were (247.63±5.66, 233.5±4.21, 215.63±5.68, 205.25±5.92 g) for G1, G2, G3 & G4 respectively. Then rats in each group were fed on the following diets:

Group 1(G1): Rats were fed on normal rat diet containing 12.5 % F1 equivalent to 10% sugarcane bagasse wet granulated powder. Group 2 (G2): Rats were fed on normal rat diet containing 12.5 % F2 equivalent to 10% wheat straw wet granulated powder. Group 3 (G3): Rats were fed on normal rat diet containing 10 % commercial bran capsule contents. Group 4 (G4): Rats were fed on normal rat diet containing 0 % ashes as a control group.

The formulations F1, F2 and commercial bran capsule contents were added to diet according to R. H. Mahmoud & W. A. Elnour, 2013 [44]. Rats were weighed individually and weights were recorded each three days for two months and recorded. Percent loss/increase in weight in experimental groups was calculated according to the following equation. Mean percent loss/increase in rat weight ± standard deviation was also calculated (Figures 11 & 12).

$$\text{Percent loss/increase in rat weight} = \frac{\text{Initial rat weight} - \text{Final rat weight}}{\text{Initial rat weight}} \text{ X } 100$$

Comparison between percent loss/increase in weight in tested and control groups was carried out using unpaired student t test. At the end of experiment, four rats from each group were selected randomly and blood samples (2 ml) were withdrawn from eyes of rats. Plasma was separated by centrifugation at 5000 rpm for 10 minutes. Immediately analyzed for their serum glucose (SG), triglycerides (TG) and total cholesterol (TCHOL) by a colorimetric analytical method using specific analytical kits according to manufacturer instructions. Food intake of each group was also recorded. Results are illustrated in Table 12

Result and Discussion

1. Loss on drying:

Table 1 reveals that drying periods were ranged between 19 & 41 h and the moisture content was ranged from 1.18 to 11.31 %. The results illustrate that treated shrimp shell powder has shortest drying period (19 h), while pomegranate peel powder has the longest one (41h). This is may be attributed to moisture content of both, where it was (1.18 & 11.31 %) for treated shrimp shell and pomegranate peel powders respectively, where the moisture content is directly proportional to time required for complete drying.

Table 1: Drying period and moisture content of different ashes powders

Type of powder	Drying period (Hour)	Moisture content (Percent)
Sugarcane bagasse	27	4.59
Wheat straw	22	2.81
Treated shrimp shell	19	1.18
Pomegranate peel	41	11.31

2. Particle size analysis:

The obtained size ranges in all powders, where subjected for further powder technology studies to select the most suitable size range in each powder to be used in preparation of tablets.

3. Compressibility index and Hausner ratio:

As illustrated in Tables 2-5 compressibility index of different size ranges were (33 – 36.93%), (35.50-38.80%), (24-27.53%), (14.29-24.87%), while Hausner ratio were (1.49-1.59), (1.52-1.63), (1.31-1.38), (1.13-1.17) & (1.17-1.33) for sugarcane bagasse, wheat straw, treated shrimp shell and pomegranate peel powders respectively. It was found that best compressibility index and Hausner ratio values were obtained by the size range of 150-250 µm in all powders except wheat straw, where best results were achieved by the size range of 106-150 µm. So these size ranges may be used for tableting process.

4. Angle of repose

Tables 2-5 show that angle of repose of different tested size ranges were (56.66-64.91º), (64.23-68.21º), (43.71-47.98º) & (34.36-43.82º) for sugarcane bagasse, wheat straw, treated shrimp shell and pomegranate peel powders respectively. Results obtained by angle of repose test were similar to that of compressibility index and Hausner ratio, where best values were obtained by the size range of 150-250 µm in case of all powders except wheat straw, which show the least angle of repose value by the size range of 106-150 µm. As a result, these size ranges were selected for tableting process.

Table 2: Physical characteristics of sugarcane bagasse powder

Particle size range	Compressibility index (Percent)	Hausner ratio	Angle of repose (Degree)
Less than 106µm	36.93	1.59	64.91
106-150µm	36.40	1.57	60.07
150-250µm	33.00	1.49	56.66
250-315µm	34.07	1.52	58.71
315-425µm	34.87	1.54	59.29
425-600µm	34.87	1.54	59.29
600µm-1.7mm	35.80	1.56	59.83

Table 3: Physical characteristics of wheat straw powder

Particle size range	Compressibility index (Percent)	Hausner ratio	Angle of repose (Degree)
Less than 106µm	38.80	1.63	68.21
106-150µm	35.50	1.52	64.23
150-250µm	36.93	1.59	65.00
250-315µm	37.80	1.61	66.34
315-425µm	37.80	1.61	66.34
425-600µm	38.07	1.61	66.78
600µm-1.7mm	38.33	1.62	67.04

Table 4: Physical characteristics of treated shrimp shell powder

Particle size range	Compressibility index (Percent)	Hausner ratio	Angle of repose (Degree)
Less than 106µm	27.53	1.38	47.98
106-150µm	25.27	1.33	45.67
150-250µm	24.00	1.31	43.71
250-315µm	24.47	1.32	44.95
315-425µm	25.07	1.33	45.36
425-600µm	25.07	1.33	45.36
600µm-1.7mm	27.13	1.37	47.11

Table 5: Physical characteristics of pomegranate peel powder

Particle size range	Compressibility index (Percent)	Hausner ratio	Angle of repose (Degree)
Less than 106µm	24.87	1.33	43.82
106-150µm	20.00	1.25	38.11

150-250µm	14.29	1.17	34.36
250-315µm	14.80	1.17	34.42
315-425µm	14.67	1.17	34.59
425-600µm	15.00	1.18	34.96
600µm-1.7mm	15.27	1.18	35.36

Table 6 illustrates physical characteristics of the selected size ranges of each powder to be compressed into tablets and indicates that compressibility index was ranged from 14.29 to 35.5 and Hausner ratio lies between 1.17 and 1.52. Pomegranate peel powder has smallest compressibility index and Hausner ratio (14.29 & 1.17) respectively, while wheat straw powder has the largest values of both (35.5 & 1.52) respectively. The results also indicate that angle of repose of different ashes powders lies between 34.36 and 64.23º. Pomegranate peel powder has the smallest angle of repose 34.36º, while wheat straw powder has the largest one 64.23º. Results of angle of repose are also in agreement with those of Compressibility index and Hausner ratio according to the scale of flowability Table 7 [39]. Physical characteristics including compressibility index, Hausner ratio and angle of repose are considered as an indicator for the flow behavior of different powders. These parameters in the case of pomegranate peel powder were 14.29 %, 1.17 & 34.36˚ respectively, where it shows good flow properties but with treated shrimp shell powder. These parameters have the value of 24%, 1.31 & 43.71˚ respectively, that indicate powder with passable flowability. On the other hand, the values of the previous parameters were (33%, 1.49 & 56.66˚) in the case of sugarcane bagasse powder and were (35.5%, 1.52 & 64.23˚) for wheat straw powder that results in very poor flow properties of both powders. Both good and passable flow characteristics enable powders to be compressed directly into tablets, while powders that show very poor flow characteristics will require further treatment in order to enhance their flow characteristics to enable their tableting process.

Table 6: Physical characteristics of the selected size range of different ashes powders

Type of powder	Compressibility index (Percent)	Hausner ratio	Angle of repose (Degree)
Sugarcane bagasse	33	1.49	56.66
Wheat straw	35.5	1.52	64.23
Treated shrimp shell	24	1.31	43.71
Pomegranate peel	14.29	1.17	34.36

Table 7: Scale of flowability

Flow characteristics	Compressibility index (Percent)	Hausner ratio	Angle of repose (Degree)
Excellent	1-10	1.00-1.11	25-30
Good	11-15	1.12-1.18	31-35
Fair	16-20	1.19-1.25	36-40
Passable	21-25	1.26-1.34	41-45
Poor	26-31	1.35-1.45	46-55
Very poor	32-37	1.46-1.59	56-65
Very, very poor	>38	>1.60	>66

Wet granulation of both sugarcane bagasse and wheat straw powders using gelatin solution as a granulating liquid (10% and 12.5% gelatin solution respectively) provide powders with excellent and passable flow characteristics respectively. The compressibility index, Hausner ratio and angle of repose were (8.69%, 1.095& 25.77º) and (23.33, 1.30 & 41.19) for sugarcane bagasse and wheat straw wet granulated powders respectively (Table 8).

Table 8: Physical characteristics of the wet granulated powder prepared using gelatin solution

Type of granules	Percent of gelatin solution	Compressibility index (Percent)	Hausner ratio	Angle of repose (Degree)
Sugarcane bagasse	10	8.69	1.095	25.77
Wheat straw	10	26.33	1.36	46.19
Wheat straw	12.5	23.33	1.30	41.19

Formulation and preparation of tablets containing different ashes powders

All the prepared tablets were elegant in appearance. Pomegranate peel and treated shrimp shell tablets were easy prepared by direct compression without any manufacturing problems, while both sugarcane bagasse and wheat straw tablets required wet granulation of powders before compression. (Table 9)

Table 9: Formulation of tablets containing different ashes

Formula code Ingredients (percent)	F_1	F_2	F_3	F_4	F_5	F_6	F_7	F_8
Sugarcane bagasse wet granulated powder	80							
Wheat straw wet granulated powder		80						
Pomegranate peel powder			80	80	80			
Treated shrimp shell powder						80	80	80
Talc powder	1	1	1	1	1	1	1	1
Starch powder				5	10		5	10
Emecompress powder	Complete to 100%							

1. Calibration curve for glucose in different media

Figures (1- 3) show the relationship between the absorbance and the concentration of glucose at the corresponding λmax = 620 nm in distilled water, 0.1 M HCl (pH 1.2) and phosphate buffer (pH 7.2). The calibration curves of glucose in all pervious media were linear and the correlation coefficient (r2) was higher than 0.9950. This indicates that glucose obeys Beer's low at the tested concentrations range.

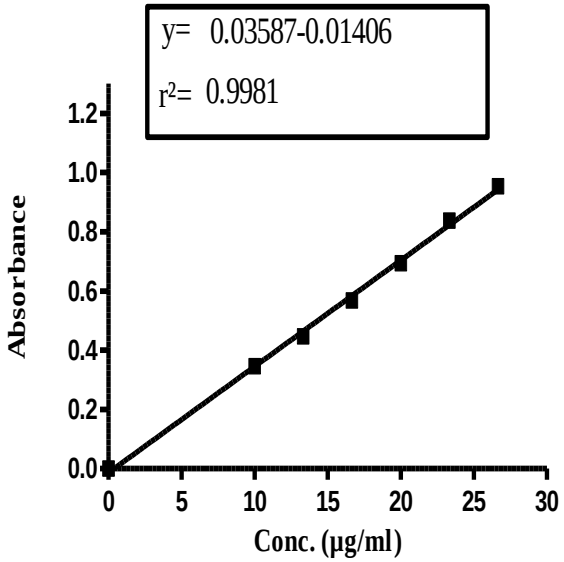

Figure 1: Calibration curve of glucose in distilled water

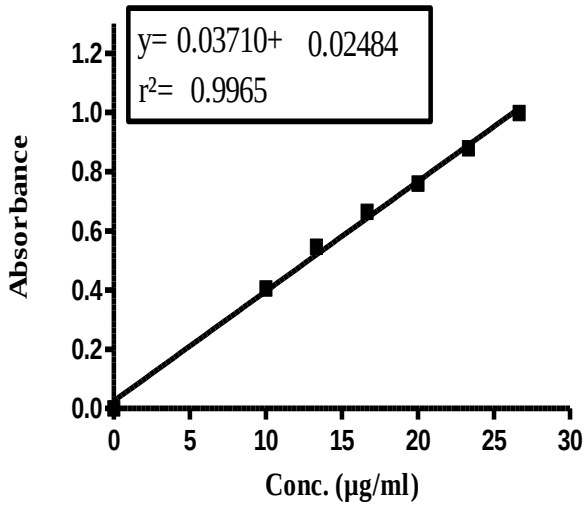

Figure 2: Calibration curve of glucose in 0.1M HCl (pH 1.2)

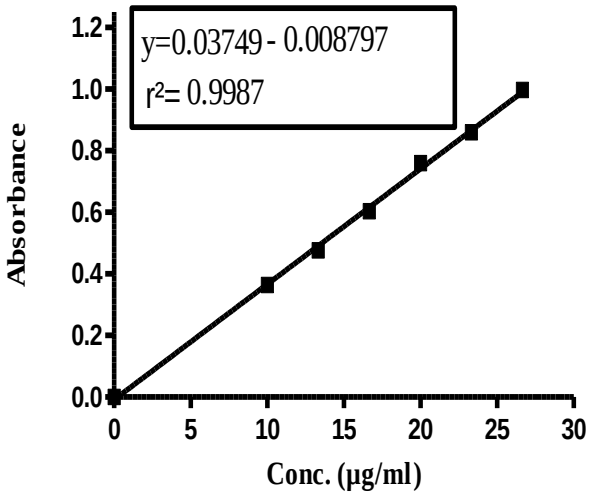

Figure 3: Calibration curve of glucose in phosphate buffer (pH 7.2)

2. Determination of the average glucose content

Table 10 shows the tablets that were varied in their glucose content from 11.145 to 129.822 mg. It was found that pomegranate peel tablets containing highest glucose content (129.822 mg), while sugarcane bagasse tablets contain the least one (11.145 mg).

Table 10: Average glucose content in tablets containing different ashes

Parameter Formula code	Average glucose content per tablet (mg)±SD
F_1	11.145±0.51
F_2	17.481±0.74
F_3	120.72±1.16
F_4	123.89±1.06
F_5	129.822±0.9
F_6	11.08±0.36
F_7	12.10±0.57
F_8	13.963±0.71

3. Evaluation of the prepared tablets

The prepared tablets were evaluated for their uniformity of weight, thickness, hardness, friability percent, glucose content uniformity, disintegration time and dissolution rate.

3. 1. Uniformity of tablets weight

As summarized in Table 11, the weight of tablets ranged from 542.5 to 601.1 mg for different formulations with acceptable percent deviation values (± 4.49%) indicating weight uniformity for all the prepared tablets and comply with the pharmacopoeial specifications.

3. 2. Thickness

Table 11 revealed that the thickness of tablets varied between 3.12 to 3.59 mm for different tablets with low standard deviation values ranged from (± 0.01-0.02) indicating uniformity of thickness for the all prepared tablets.

3. 3. Hardness

Results represented in Table 11 show that the hardness of different formulations was found to be within the range of 6 and 15 kg/cm2. Sugarcane bagasse tablets were the hardest one, while pomegranate peel tablets were of the least hardness. This is may be attributed to compressibility and binding properties of the powder.

3. 4. Friability test

Table 11 illustrates that all the prepared tablets have an acceptable friability percent, where friability percent lies between 0.22 and 0.68%, which are within the B.P specifications (not more than 1%). The pomegranate peels tablets had the smallest friability percent 0.22%, while sugarcane bagasse tablets had the largest one (0.68%). These results may be attributed to sticky nature of pomegranate peel powder [45] that provides high binding force between particles.

3. 5. Disintegration time:

Sugarcane bagasse and wheat straw containing tablets show acceptable disintegration time (11.40±0.32 & 17±0.06) minutes respectively (Table 11), pomegranate peel and treated shrimp shell tablets (F3 & F6) require 42.81& 60.22 minutes respectively, which is a relatively long disintegration time. This long disintegration time may be attributed to sticky nature of pomegranate peel [45] and adhesive property of chitin (treated shrimp shell) [46], so addition of starch as a disintegrant was suggested. When 5% of starch was added, the disintegration time of the prepared tablets (F4 & F7) was moderately enhanced (33.24 & 45.32 minutes)

respectively but this disintegration time still unacceptable, so starch percent up to 10% was used (F5 & F8). Both F5& F8 have acceptable disintegration time (20.65 & 29.95 minutes) respectively.

Sugarcane bagasse tablets were found to have the most rapid disintegration time 11.40 minutes. This may be attributed to swelling mechanism of α-Cellulose, the major component (33.29 ± 1.43%) in sugarcane bagasse [32], where upon contact with water, it can absorb water five times its own weight by capillary action and as a result the adhesiveness of other tablet ingredients was overcome resulting in tablet disintegration. In addition, α-cellulose obtained from sugarcane bagasse has a superior disintegrant action over cornstarch and microcrystalline cellulose providing tablets with a very fast dissolution rate. These results were in agreement with that reported by Bakre Lateef Gbenga et al., 2014 [47], who extract α-Cellulose from sugarcane bagasse and use it as a tablet disintegrant.

3. 6. Glucose content uniformity

Glucose content was found to be ranged from 99.76 to 102.43 % as shown in Table 11.

Table 11: Physical properties of tablets containing different ashes

Parameter Formula code	Uniformity of weight (g) ± SD	Hardness (kg/cm^2) ± SD	Friability (%)	Thickness (mm) ± SD	Disintegration time (Minutes) ± SD	Glucose content (%) ± SD
F_1	0.5763 ±0.02	15.00 ±0.00	0.68	3.37 ±0.02	11.40 ±0.32	99.76 ± 2.65
F_2	0.5820 ±0.001	14.58 ±0.41	0.25	3.59 ±0.01	17.00 ±0.06	102.43 ±2.42
F_3	0.5492 ±0.01	6.45 ±0.25	0.22	3.36 ±0.02	42.81 ±0.26	100.32 ±1.97
F_4	0.5478 ±0.003	6.25 ±0.24	0.22	3.36 ±0.02	33.24 ±0.17	101.16 ±1.48
F_5	0.5425 ±0.004	6.00 ±0.24	0.23	3.36 ±0.01	20.65 ±0.38	102.16 ±1.64
F_6	0.6006 ±0.004	15.00 ±0.22	0.49	3.12 ±0.02	60.22 ±0.06	100.12 ±2.23
F_7	0.6003 ±001	15.00 ± 0.23	0.49	3.13 ±0.02	45.32 ± 0.05	101.32 ±1.97
F_8	0.6011 ±0.01	14.73 ±0.22	0.50	3.13 ±0.02	29.95 ±0.04	100.20 ±3.33

3. 7. Dissolution rate

3. 7. 1. Effect of pH of the dissolution medium on the glucose release profile from tablets containing different ashes

Figure 4 illustrates the percent glucose released from treated shrimp shell tablets indicating rapid release of glucose after 10 minutes followed by a relatively moderate release during the first hour, while delayed release of glucose was obtained in the second hour. There was no significant difference ($P > 0.05$) between percent glucose release from tablets in both distilled water and phosphate buffer (pH 7.2), where chitin (treated shrimp shell) is insoluble in water and neutral pH media [48]. On the other hand, treated shrimp shell tablets dissolution profile was significantly higher ($P < 0.05$) in 0.1 M HCl (pH 1.2) compared to that of both in distilled water and phosphate buffer (pH 7.2). About 99.98 % of glucose was released after 60 minutes in the case of 0.1 M HCl (pH 1.2) compared to 81.31 and 80.84 % only in both distilled water and phosphate buffer (pH 7.2) respectively.

This may be attributed to higher swelling capacity of chitin (treated shrimp shell) in acidic dissolution medium providing a higher percent drug release. These results were in agreement with that reported by T. R. Arunraj et al., 2014 [49], who formulate doxorubicin, the chemotherapeutic agent used in the treatment of liver cancer in chitin nanogel form for site-specific delivery.

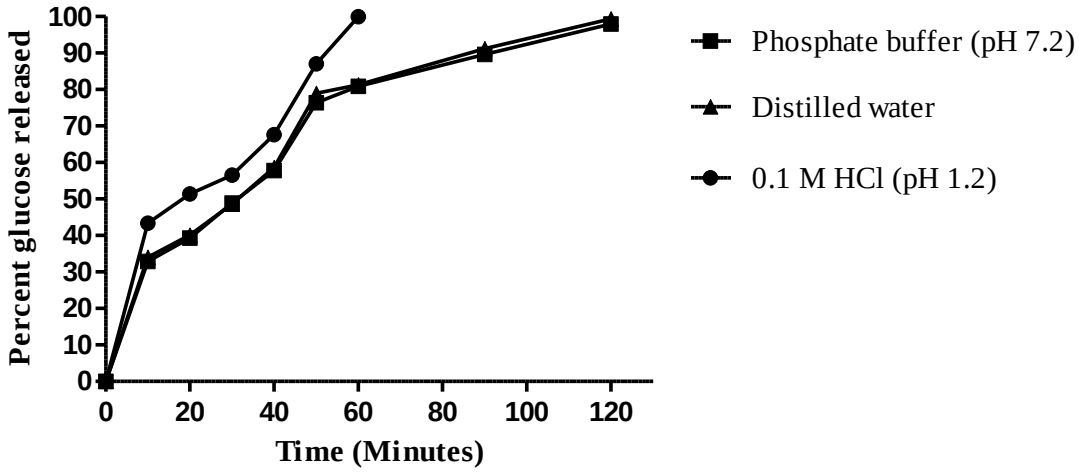

Figure 4: Percent glucose released from treated shrimp shell tablets in different pH media

Figure 5 illustrates the dissolution profile of glucose released from pomegranate peel tablets, where glucose was released from these tablets in a steady state with respect to time. There was no significant difference ($P > 0.05$) in the glucose release profile in distilled water and phosphate buffer (pH 7.2) as dissolution media. However, glucose was released from pomegranate peel tablets in 0.1M HCl (pH 1.2) more slowly with a significant difference at ($P < 0.05$) compared to that released in both distilled water and phosphate buffer (pH 7.2). After 50 minutes about 95.95 & 92.61% of glucose was released from pomegranate peel tablets in phosphate buffer (pH 7.2) and distilled water respectively compared to only 80.25% in 0.1 M HCl (pH 1.2). This can be rationalized by the presence of a high phenolic content in pomegranate peel represented in hydroxyl benzoic acids (gallic acid and ellagic acid), hydroxyl cinnamic acids (caffeic acid, chlorogenic acid & p-Coumaric acid) and carboxylic acids (qunic acid) [50], that retard tablet dissolution in acidic medium.

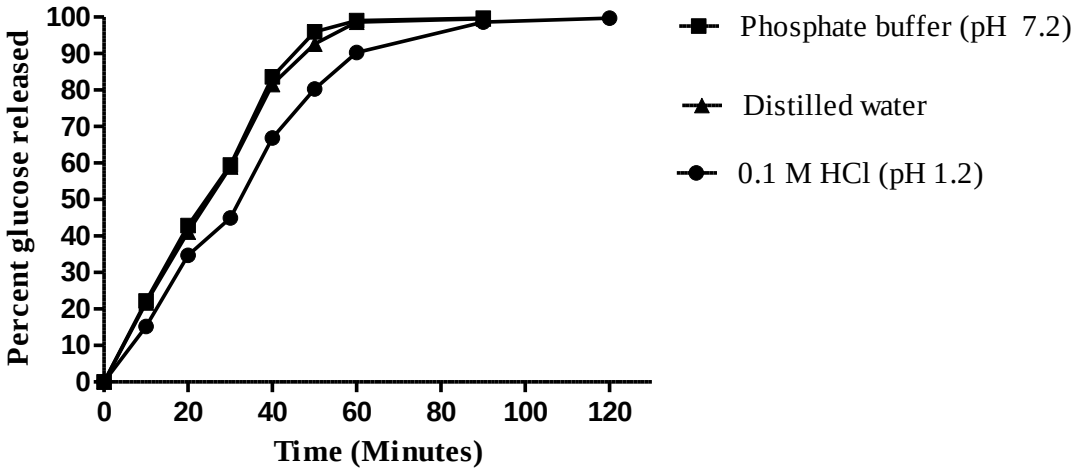

Figure 5: Percent glucose released from pomegranate peel tablets in different pH media

pH of dissolution medium has no effect on dissolution profile of sugarcane bagasse and wheat straw tablets as illustrated in Figures 6 & 7. Statistically, there is no significant difference ($P > 0.05$) in the glucose release in distilled water, 0.1 M HCl (pH 1.2) and phosphate buffer (pH 7.2). This may be attributed to the pH independence of the wettability and the swelling capacity of cellulose which is the main component in both. Also these results may be attributed to the absence of any strong functional group carrying charge that positively or negatively affected by pH of the dissolution medium, where water uptake and wettability of cellulose and its uncharged derivatives are not affected by pH of the dissolution medium, where non-ionic polymers are not sensitive for pH of swelling medium [51]. These results were in agreement with those obtained by Peter Dios et al., 2015 [52], who studied the wettability of low substituted hydroxyl propyl cellulose in both distilled water and 0.1 M HCl. In addition, when

Ramona-Daniela et al., 2015 [53], prepared a based bacterial cellulose and gelatin matrix and studied the properties of the produced hydrogel upon contact with aqueous medium, he found that in acidic environment, based bacterial cellulose and gelatin hydrogels showed slightly higher swelling capacity compared with the basic environment. These results were attributed to the polyelectrolyte property of gelatin, which contains amino groups in its structure, which are protonated in acidic media, causing electrostatic repulsions leading to an increase of swelling properties. In basic medium, amino groups are deprotonated, the electrostatic repulsions are reduced, leading to a decrease of swelling, while when S. Sultana el al., 2012 [54], studied the swelling properties of acrylamide/carboxy methyl cellulose (CMC) co-polymer he found that swelling ratio increases with the increase in pH of the swelling medium. The lower swelling property in the more acidic region may be due to the protonation of the carboxylic group of CMC. These results also indicate that when cellulose is combined with other charged material, the wettability and swelling capacity of the product is pH dependent according to the charge of the added material.

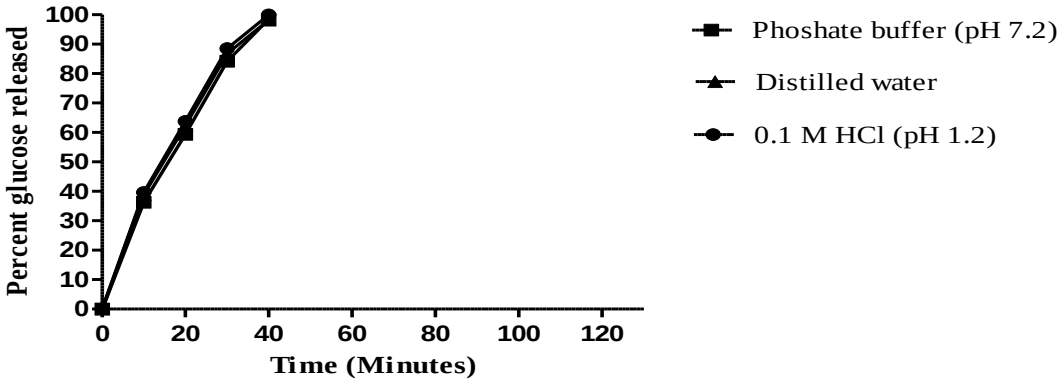

Figure 6: Percent glucose released from sugarcane bagasse tablets in different pH media

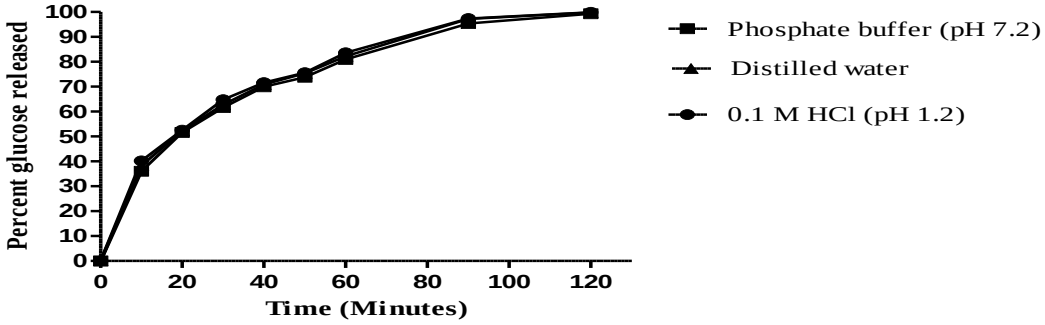

Figure 7: Percent glucose released from wheat straw tablets in different pH media

3. 7. 2. Effect of different ashes on the release of glucose

Figure 8 illustrates the dissolution profile of glucose released from different tablets in phosphate buffer (pH 7.2). Glucose was released completely from sugarcane bagasse tablets after 40 minutes, exhibiting a rapid release rate of glucose, which may be explained by their rapid disintegration. While after 60 minutes about 99.02, 80.96 and 80.84% of glucose were released from pomegranate peel, wheat straw and treated shrimp shell tablets respectively. Pomegranate peel tablets show relatively rapid release of their glucose content. Wheat straw and treated shrimp shell tablets show relatively delayed glucose release, which may be attributed to their relatively long disintegration time. Similar dissolution pattern was obtained in distilled water as illustrated in Figure 9.

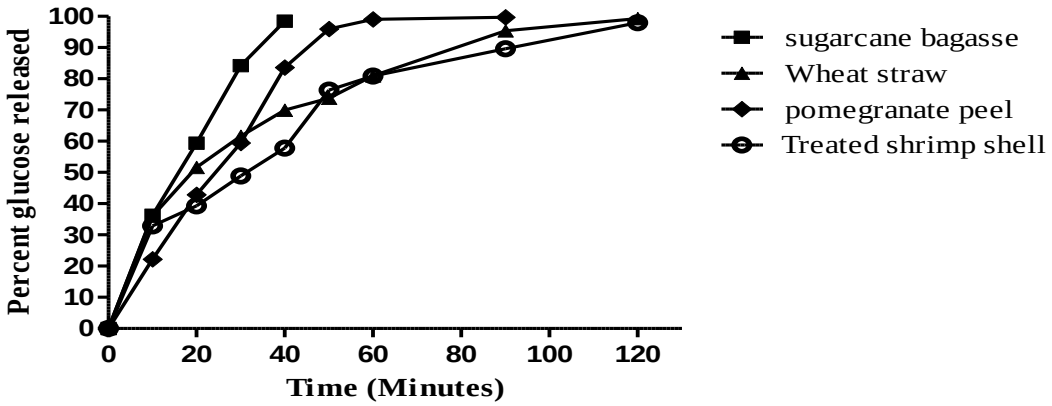

Figure 8: Percent glucose released from tablets containing different ashes in phosphate buffer (pH 7.2)

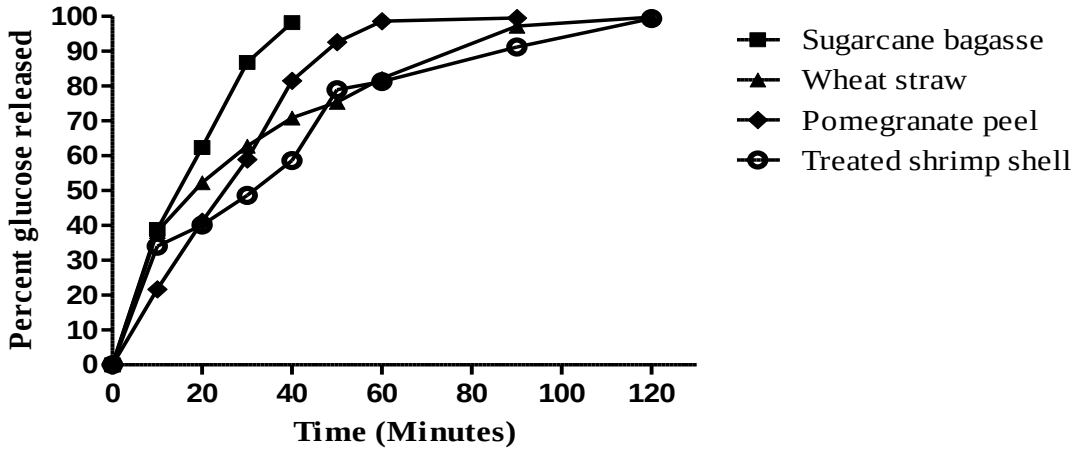

Figure 9: Percent glucose released from tablets containing different ashes in distilled water

3. 7. 3. Effect of concentration of the granulating fluid

Figure 10 illustrates dissolution profiles of both sugarcane bagasse and wheat straw tablets indicating initially high rate of glucose release (within the first 10 minutes) in both tablets. After 40 minutes, the dissolution rate gradually decreased in the case of wheat straw tablets, where about 98.20 and 70.87% of glucose was released from sugarcane bagasse and wheat straw tablets respectively.

Wheat straw tablets reach the maximum release of glucose after 120 minutes. Statistically, the release of glucose from sugarcane bagasse tablets was significantly higher ($P < 0.05$) compared with that of wheat straw tablets. This can be explained on the bases of that both tablets contain nearly similar concentration of cellulose as a main component about (33.29 and 33.7- 40%) for sugarcane bagasse and wheat straw respectively [32, 33]. The use of higher concentration of granulating fluid (12.5%) in the case of wheat straw tablets compared to (10%) in the case of sugarcane bagasse tablets, which results in increasing bonds between tablet components, which results in delaying the dissolution of wheat straw tablets.

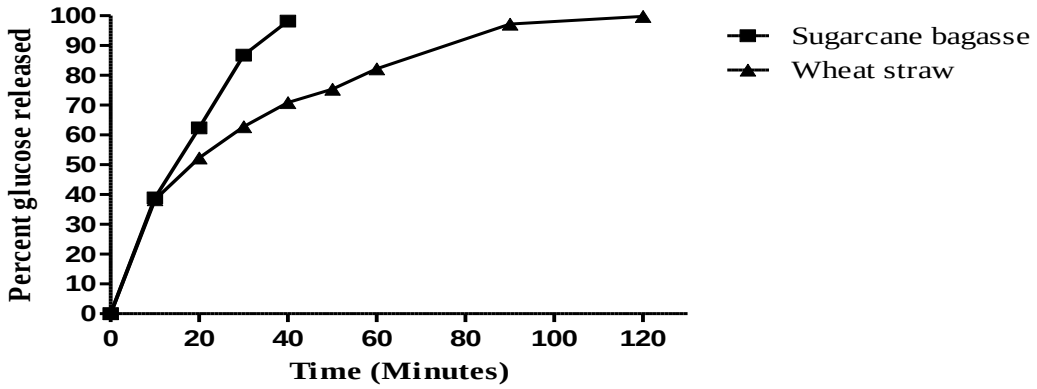

Figure 10: Effect of granulating liquid on percent glucose released from sugarcane bagasse and wheat straw tablets in distilled water

In-Vivo Study of Some Selected Ashes Formulations as anti-obesity agents

Figures11-12 illustrate that the addition of both F1& F2 equivalent to 10 % sugarcane bagasse and wheat straw wet granulated powders to the normal rat diet results in a marked decrease in body weight of rats by about 23.92 & 14.62 % respectively. While addition of 10% commercial bran to the normal rat diet hinders the normal weight gain rate compared to control group fed on normal rat diet containing 0 % ashes, where rats of G3 gain weight by about only 13.36 % compared with 25.16% body weight gain in rats of G4. At the first 21 days of experiment, a

significant reduction in weight of rats in groups G1 and G2 was obtained followed by a relatively very slow rate of weight reduction.

On the other hand, rats in G3 and G4 gain weight, which was also significant in the first 21 days of experiment and then becomes relatively slow. Percent loss in weight achieved by F1 was a significantly higher ($P< 0.05$) compared with that obtained by F2. This is may be attributed to the average glucose content of both ashes, where the previously determined total glucose content of sugarcane bagasse and wheat straw wet granulated powders were 11.145 & 17.481 mg/tablet respectively. As glucose is the first, direct and simplest source of caloric intake, weight loss is inversely proportional to the average glucose concentration in ashes.

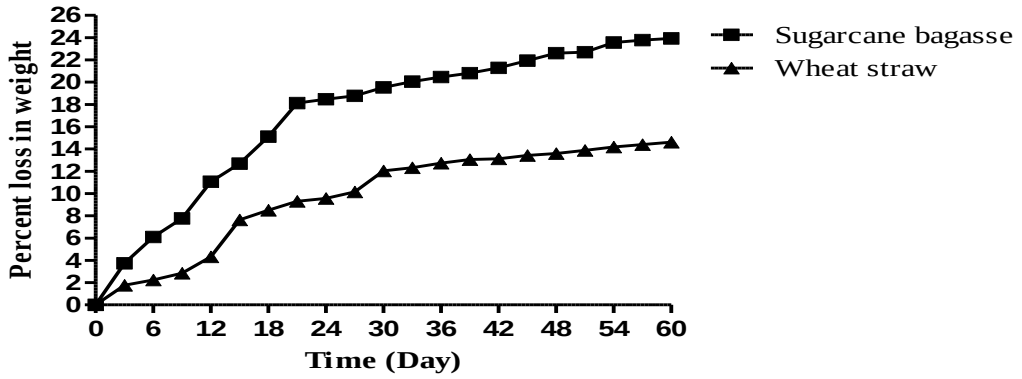

Figure 11: Mean percent loss in weight in sugarcane bagasse and wheat straw groups

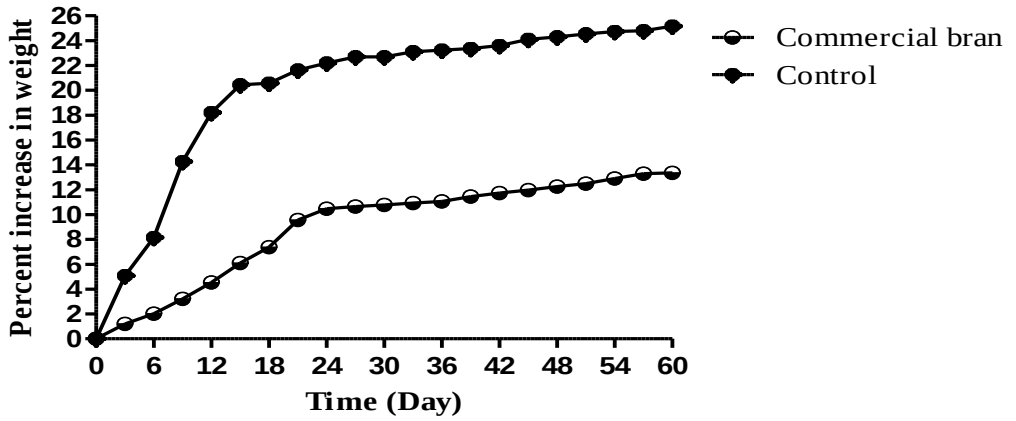

Figure 12: Mean percent increase in weight in commercial bran and control group

Indicates that significant reduction in serum triglycerides was obtained in G1 and G2 compared to control group, which explained by their reduction in weight, while G3 shows a limited reduction that was not significant (P> 0.05) compared to G4. Statistically there was no significant difference (P> 0.05) in serum triglycerides in G1 compared with G2. Total cholesterol level of G1 was significantly higher (P< 0.05) compared with G2. These results were in agreement with that obtained by J. Atangwho et al., 2012 [55], who studied the lipid profile and serum glucose level of obese rats treated with Vernonia amygdalina. Results obtained by serum glucose analysis were found to be similar to that obtained by total cholesterol analysis. When food intake during the experiment was calculated, it was found that amounts of food consumed by G1 and G2 were (5.5 & 6.2 kg/2 months) respectively indicating that G1 was lower than G2. On the other hand, food intake value in G3 and G4 were (7.7 &12.13 kg/2 months) respectively. Food intake of G4 was relatively higher than that of other groups. These results may explain the loss in weight of both G1 and G2 and the limited increase in weight in G3 compared with the increase in weight in G4, where the increase in food intake is followed by increase in body weight.

Table 12: Serum triglycerides, total cholesterol and glucose and food intake of rats treated with normal diet containing selected ashes

Parameter Group Code	Serum triglyceride (mg/dl) ± SD	Total cholesterol (mg/dl) ± SD	Serum glucose (mg/dl) ± SD	Food intake (kg/2 months)
G_1	97.5±5.7	55.9±7.37	66.57±6.06	5.5
G_2	106.25±5.98	63.5±8.33	80.20±7.69	6.2
G_3	119.63±6.15	70.75±6.18	87.25±5.0	7.7
G_4	127.76±4.82	72±8.98	100.75±6.12	12.13

Conclusion:

The results of this study have established that pomegranate peel and treated shrimp shell powders have acceptable flow characteristics that enable their compression into tablets, while sugarcane bagasse and wheat straw powders have very poor flow characteristics that they must undergo wet granulation before tableting.

Sugarcane bagasse tablets show very rapid disintegration and dissolution with minimum average glucose content.

Sugarcane bagasse and wheat straw can be used in obesity management, where they exhibit reduction in weight over commercial bran.

REFERENCES:

1. Haslam DW and James WP: Obesity. Lancet 2005; 366 (9492): 1197–1209.
2. World Health Organization 2015.
3. World Health Organization 2000.
4. Drewnowski A and Specter SE: Poverty and Obesity: The Role of Energy Density and Energy Costs. Am. J. Clin. Nutr 2004; 79 (1): 6–16.
5. CS Byme, ES Chambers, DJ Morrison and G Frost: The Role of Short Chain Fatty Acids in Appetite Regulation and Energy Homeostasis. International Journal of Obesity 2015; 39: 1331-1338.
6. Nestle M and Jacobson MF: Halting the obesity epidemic: A public health policy approach. Public Health Rep. 2000; 115 (1): 12–24.
7. James WP: The fundamental drivers of the obesity epidemic: Obes Rev. 2008; 1: 6–13.
8. Bleich S, Cutler D, Murray C and Adams A: Why is the developed world obese?.Annu Rev Public Health 2008; 29: 273–295.
9. Leslie WS, Hankey CR and Lean ME: Weight gain as an adverse effect of some commonly prescribed drugs: a systematic review. QJM 2007; 100 (7): 395-404.
10. Keith SW, Redden DT, Katzmarzyk PT, et al.: Putative Contributors to the Secular Increase in Obesity: Exploring The Roads Less Traveled. Int J Obes (Lond) 2006; 30 (11): 1585–1594.
11. Bray GA: Medical consequences of obesity. J. Clin. Endocrinol.Metab. 2004; 89 (6): 2583–2589.
12. Wanahita N, Messerli FH, Bangalore S, Gami AS, Somers VK and Steinberg JS: Atrial fibrillation and obesity--results of a meta-analysis. Am Heart J2008; 155(2): 310-315.
13. Larsson SC and Wolk A: Overweight and obesity and incidence of leukemia: a meta-analysis of cohort studies. Int J Cancer 2008; 122 (6): 1418-1421.
14. Connolly BS, Barnett C, Vogt KN, Li T, Stone J and Boyd NF: A meta-analysis of published literature on waist-to-hip ratio and risk of breast cancer. Nutr Cancer 2002; 44 (2): 127-138.
15. Harvie M, Hooper L and Howell AH: Central obesity and breast cancer risk: a systematic review. Obesity Reviews 2003; 4 (3): 157-173.
16. Guh DP, Zhang W, Bansback N, Amarsi Z, Birmingham CL and Anis AH: The incidence of co-morbidities related to obesity and overweight: a systematic review and meta-analysis. BMC Public Health 2009; 9:88.

17. Larsson SC and Wolk A: Obesity and the risk of gallbladder cancer: A meta-analysis. Br J Cancer, 2007; 96 (9): 1457-1461.
18. Olsen CM, Green AC, Whiteman DC, Sadeghi S, Kolahdooz F and Webb PM: Obesity and the risk of epithelial ovarian cancer: a systematic review and meta-analysis. Eur J Cancer 2007; 43 (4): 690- 709.
19. Berrington de Gonzalez A, Sweetland S and Spencer E: A meta-analysis of obesity and the risk of pancreatic cancer. Br J Cancer 2003; 89 (3): 519-523.
20. MacInnis RJ and English DR: Body size and composition and prostate cancer risk: Systematic review and meta-regression analysis. Cancer Causes Control 2006; 17 (8): 989-1003.
21. Bergstrom A, Pisani P, Tenet V, Wolk A and Adami HO: Overweight as an avoidable cause of cancer in Europe.Int J Cancer 2001; 91(3): 421-430.
22. Wolk A, Gridley G, Svensson M, Nyren O, McLaughlin JK, Fraumeni JF, et al.: A prospective study of obesity and cancer risk (Sweden). Cancer Causes Control 2001; 12 (1): 13-21.
23. Hampel H, Abraham NS and El-Serag HB: Meta-analysis: obesity and the risk for gastroesophageal reflux disease and its complications. Ann Intern Med 2005; 143 (3): 199-211.
24. Kubo A and Corley DA: Body mass index and adenocarcinomas of the esophagus or gastric cardia: a systematic review and meta-analysis. Cancer Epidemiol Biomarkers Prev. 2006; 15 (5): 872-878.
25. Lau DC, Douketis JD, Morrison KM, Hramiak IM and Sharma AM: Canadian clinical practice guidelines on the management and prevention of obesity in adults and children, CMAJ 2007; 176 (8): S1–13.
26. National Institute for Health and Clinical Excellence (NICE), Obesity: the prevention, identification, assessment and management of overweight and obesity in adults and children. London: NICE (2006).
27. Cooke D and Bloom S: The obesity pipeline: current strategies in the development of anti-obesity drugs. Nat. Rev. Drug Discov. 2006; 5: 919-931.
28. Sargent BJ and Moore NA: New central targets for the treatment of obesity. Br. J. Clin. Pharmacol.2009; 68: 852-860.
29. Kral JG and Naslund E: Surgical treatment of obesity.Nature ClinPract: Endocrinol.Metab.2007; 3: 574-583.
30. Food and Agricultural organization (2010).
31. Ohwoavworhua F, Ogah E and Kunle O: Preliminary investigation of physicochemical and functional properties of celluloses obtained from waste paper – a potential pharmaceutical excipient. J Raw Matr Res 2005; 2: 84-93.
32. P Sugumaran and S Seshadri: Evaluation of selected biomass for charcoal production. Journal of Scientific & Industrial Research 2009; 68: 719-723.
33. TehminaSaleem Khan and UmarahMubeen: Wheat Straw: A Pragmatic Overview. Current Research Journal of Biological Sciences 2012; 4 (6): 673-675.

34. Prakash, CVS and I Prakash. Int. j. Res. Chem. Environ 2011; 1: 1-8.
35. Alaa Ibrahim Attia Ibrahim et al: Utilization of Pomegranate Waste in The Production of Pan Bread, Zagazig University 2013.
36. James M Lattimer and Mark D Haub: Effects of Dietary Fiber and Its Components on Metabolic Health. Nutrients 2010; 2: 1266-1289.
37. Pradip Kumar Dutta, JoydeepDutta and V S Tripathi: Chitin and chitosan: Chemistry, properities and applications. Journal of Scientific & Industrial Research 2004; 63: 20-31.
38. Md. Monarul Islam, Shah Md. Masum, M. MahbuburRahman, Md. Ashraful Islam Molla, AA Shaikh and SK Roy: Preparation of chitosan from shrimp shell and investigation of its properties. International Journal of Basic & Applied Sciences 2011; 11: 116-129.
39. British Pharmacopeia (2015).
40. AG Patil, SP Koli , DA Patil and AV Phatak: Evaluation of Extraction Techniques with Various Solvents to Determine Extraction Efficiency of Selected Medical Plants. IJPSR 2012; 3 (8): 2607-2612.
41. Xue-Song Zhang, Rodolfo Garcia-Contreras and Thomas K Wood: Escherichia coli transcription factor YncC (McbR) regulates colonic acid and biofilm formation by repressing expression of periplasmic protein YbiM (McbA). International Society for Microbial Ecology 2008; 2: 615-631.
42. JanezDemˇsar: Statistical Comparisons of Classifiers over Multiple Data Sets. Journal of Machine Learning Research 2006; 7: 1–30.
43. SamyEleawa and Hussein F Sakr: Effect of Exercise and Orlistat Therapy in Rat Model of Obesity Induced with High Fat Diet. Med. J. Cairo Univ. 2013; 81 (2): 59-67.
44. RH Mahmoud and WA Elnour: Comparative evaluation of the efficacy of ginger and orlistat on obesity management, pancreatic lipase and liver peroxisomal catalase enzyme in male albino rats. European Review for Medical and Pharmacological Sciences 2013; 17: 75-83.
45. Nesrine M Hegazi, Amani N Hashim, NahlaAayoub, and Sahar A Hussien: Anti-Inflammatory Activity of Pomegranate Peel Extract. International Conference on Agriculture, Biology and Environmental Sciences 2014.
46. Judilynn N Solidum: An exploratory study on possible preparations that may be formulated from Tahong Shells collected from Cavite City, Philippines. International Journal of Chemical and Environmental Engineering 2010; 1 (2): 102-107.
47. BakreLateefGbenga and OdumalaKehinde Fatimah: Investigation of α-Cellulose Content of Sugarcane Scrappings and Bagasse as Tablet Disintegrant. Journal of Basic & Applied Sciences 2014; 10: 142-148.
48. VeeranGowdaKadajji and Guru V. Betageri: Water Soluble Polymers for Pharmaceutical Applications. Polymers Journal 2011; 3: 1972-2009.

49. TR Arunraj, N SanojRejinold, N Ashwin Kumar and R Jayakumar: Bio-responsive chitin-poly(L-lactic acid) composite nanogels for liver cancer. Colloids and Surfaces B: Biointerfaces 2014; 1: 394–402.
50. ChaturvedulaVenkataSaiPrakash and IndraPrakash: Bioactive Chemical Constituents from Pomegranate (Punicagranatum) Juice, Seed and Peel-A Review. International Journal of Research in Chemistry and Environment 2011; 1 (1): 1-18.
51. JuergenSiepmann, Ronald Siegel and Michael Rathbone: Fundamentals and Applications of Controlled Release Drug Delivery. Eds. Spinger, New York 2012; 75-106.
52. Peter Dios, TivadarPernecker, Sandor Nagy, Szilard Pal and Attila Devay: Influence of different types of low substituted hydroxyl propyl cellulose on tableting, disintegration, and floating behavior of floating drug delivery system. Saudi Pharmaceutical Journal 2015; 23: 658-666.
53. Ramona-Daniela pavaloiu, Anicuţastoica-Guzun and TănaseDobre: Swelling Studies of Composite Hydrogels Based on Bacterial Cellulose and Gelatin. U.P.B. Sci. Bull., Series B 2015; 77: 53-62.
54. S Sultana, MR Islam, NC Dafader and ME Haque: Preparation of Carboxy Methyl Celluose/Acrylamide co-polymer Hydrogel Using Gamma Radiation and Investigation of Its Swelling Behavior. Journal of Bangladesh Chemical Society 2012; 25 (2): 132-138.
55. J. Atangwho, Emmanuel E. Edet, Daniel E. Uti, Augustine U. Obi, Mohd. Z. Asmawi and Mariam Ahmad: Biochemical and histological impact of Vernonia amygdalina supplemented diet in obese rats. Saudi Journal of Biological Sciences 2012; 19: 385–392