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Formulation of low glycaemic index biscuits suitable for diabetics from unripe banana, okra, and stevia leaves / jujube fruit

Wilfred Damndja Ngaha*, William Tchabo, Rosane Soh Matsinkou, Lola Kangue Nyame and Edith Nig Fombang

Abstract

Biscuits are the largest category of consumed snacks among bakery products, but their simple sugar and fat high content associate to low fiber content make them unsuitable for diabetics. This study aimed to formulate biscuits with low glycaemic index from the blend of unripe banana, okra and stevia leaves or jujube fruit for diabetics. To achieve this goal, unripe banana, fresh okra, stevia leaves and jujube fruits were purchased from a main market in Ngaoundere and processed into flours. Proximate composition of the flours was determined, and then, Design Expert software was used to formulate two different blends, Banana/Okra/Stevia (BOS) and Banana/Okra/Jujube (BOJ). According to the results of preliminary sensory tests done in the laboratory, three biscuits with stevia (BOS1, BOS2, BOS3), three biscuits with jujube (BOJ1, BOJ2, BOJ3), and a witness biscuit with table sugar as sweetener were formulated. After the physicochemical characterisation of formulated biscuits, animal experimentation was carried out in order to evaluate the postprandial evolution of blood glucose in rats after having ingested the biscuits, and the Glycaemic Index (GI) of produced biscuits. A hedonic test with 30 consumers was done, parameters assessed including colour, texture, taste, flavour, and overall acceptability. The results obtained show that unripe banana is mostly made of complex carbohydrates, while okra and stevia are good sources of fibers, confirming their importance in diabetic's diet. All the biscuits produced with stevia and jujube did not raise the blood glucose compared to the witness biscuit made with table sugar. Biscuits produced with stevia leaves had a lower GI than those produced with jujube fruit among the biscuits produced with stevia leaves, BOS2 had the lowest GI, then the lowest increase of postprandial blood glucose in rats, and was the most appreciated by the consumers in terms of appearance, flavour, taste and overall acceptability. Therefore, BOS2 could be recommended as snack for diabetics.

Keywords Snacks, Diabetics, Glycaemic index, Banana, Okra, Stevia, Jujube

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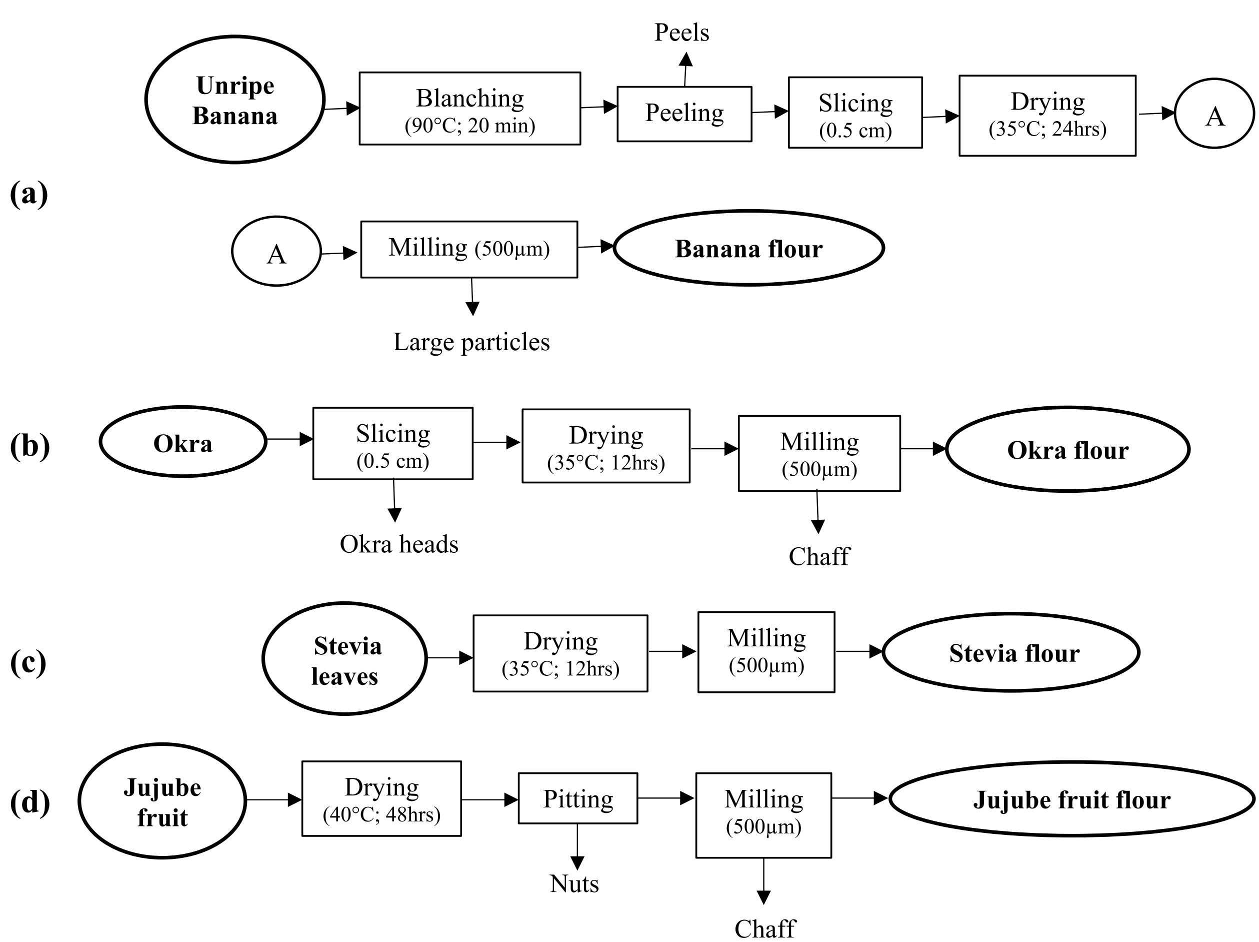
Graphical Abstract



Introduction

Biscuits are the largest category of consumed snack foods among bakery products without no geographic, social or age Boundaries (Chauhan et al. 2016), due to its ready-to-eat nature, good nutritional quality, availability and affordability (Misra et al. 2014). However, its high content in simple sugar and fat, as well as its low content in fiber make it unsuitable for people suffering from nutrition-related diseases (Hauner et al. 2012), such as diabetics who have hyperglycaemia triggered by the consumption of high glycaemic index foods, thus raising their blood sugar. It therefore appears crucial to find suitable food ingredients that could allow the production of biscuits recommended for diabetics.

Indeed, foods with a low glycaemic index, rich in fibers, and with non-nutritive sweeteners are recommended to diabetics to stabilize their blood glucose levels (Gray & Threlkeld 2000). Fibers improve glucose tolerance control, often decrease insulin requirements, promote satiety and tend to lower serum cholesterol and triglyceride values (Augustin et al. 2015), what is benefit for diabetics. Researches have shown that bananas, especially *Musa acuminata,* possess anti-diabetic potentials (Vijay et al. 2022; Kumar et al. 2021). Its low glycaemic index and technological properties make it exploitable in various bakery products snacks suitable for diabetics (Adedayo et al. 2016; Erukainure et al. 2014). While, Okra (*Abelmoschus esculentus)* which has been traditionally used in Africa as an alternative of ailments to regulate blood sugar (Basharat et al. 2021) could also serve as ingredient for diabetic snacks owing its high fibers content (Gemede et al. 2015).

Futhermore, the rheological properties of okra due to its high content in gums are highly sought by food industry (Dantas et al. 2021). Whilst among natural sweeteners, Stevia (*Stevia rebaudiana)* and jujube fruit *(Ziziphus* *mauritiana)* have grown in popularity, leading to their use as substitute for sugar in the formulation of snacks (Gasmalla et al. 2014). Therefore, from the above, this study seeks to formulate biscuits with low glycaemic index from the blend of unripe banana, okra and stevia or jujube fruit which could be serve as snack for diabetics.

Methods

Sampling of raw materials, flours processing and characterization

Unripe banana (*Musa acuminata)*, fresh okra (*Abelmoschus esculentus)*, stevia leaves (*Stevia rebaudiana)* and jujube fruits (*Ziziphus mauritiana)* were purchased from a local market of Ngaoundere in Adamawa region of Cameroon. Unripe banana, okra, stevia and jujube fruit flours were produced as describe in Fig. 1 and stored at 4 °C for further utilization. Moisture, ashes, total sugars, simple sugars and crude fibers contents of flours of unripe banana, mature okra fruit, stevia leaves and dry jujube fruit were determined as reported by Tedom et al. (Tedom et al. 2019).

Biscuits formulation

A ternary mixing plan with constraints carried out with Design Expert software was made of 2 different blends, Banana / Okra / Stevia (BOS) and Banana / Okra / Jujube (BOJ). The proportion of banana flour varied from 70 to 85%, okra flour from 10 to 15%, stevia from 5 to 10%, and that of jujube from 10 to 20%. A preliminary test was done to select samples for the continuation of the work, according to the taste and the level of sweetness in mouth. From the conditions above stated, six flours were produced, three from the blend BOS called BOS1 (75:15:10), BOS2 (82.5:10:7.5), BOS3 (85:10:5), and three from the blend BOJ named BOJ1 (70:10:20), BOJ2 (75:10:15), and BOJ3 (75:15:10). Thereafter, these flours formulation were used to produce plain biscuits according to Davidson I (Davidson 2019) as depicted in Fig. 2. A control biscuit (T) was also produced, replacing stevia and jujube by table sugar, in order to be used as positive control in animal experimentation.

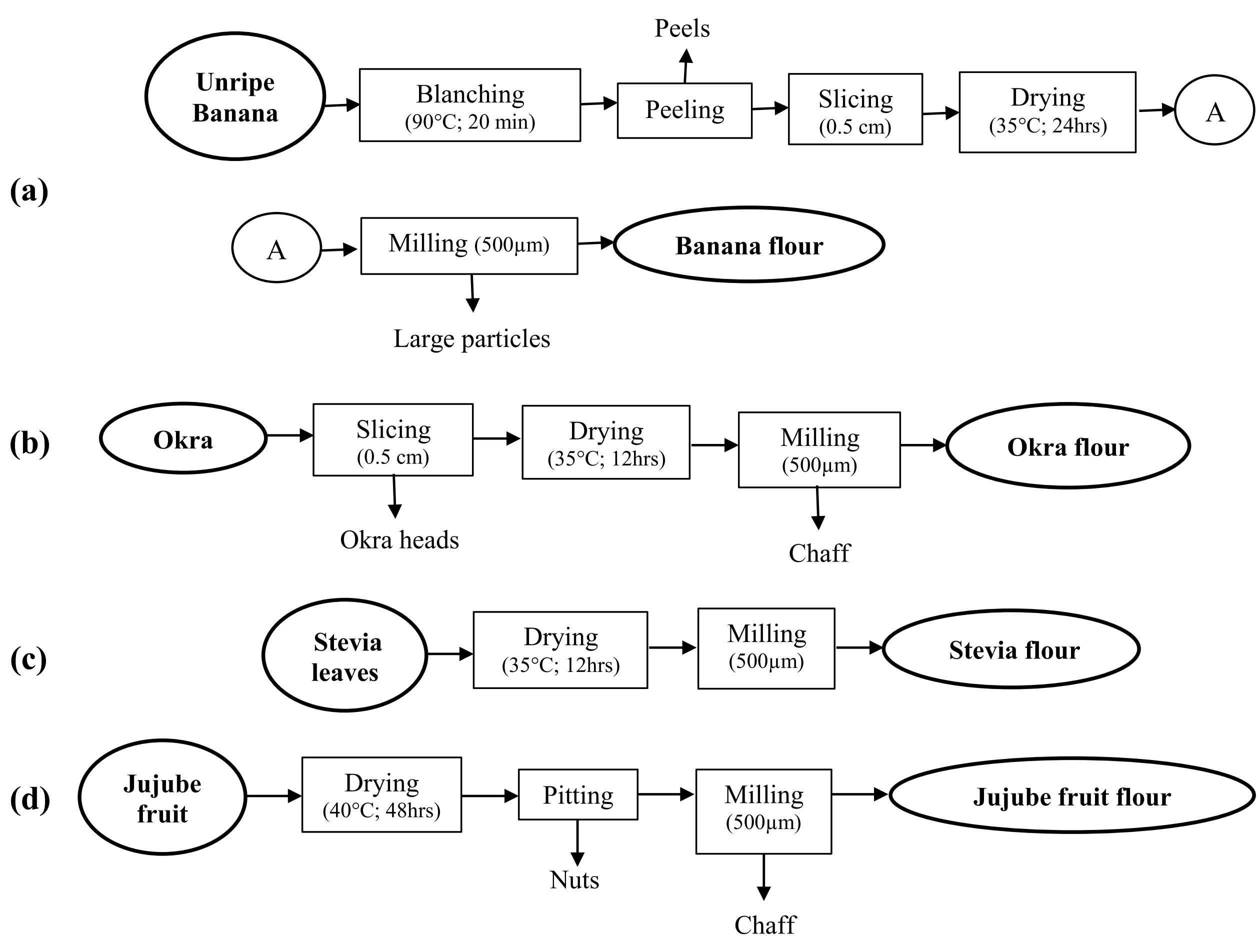


Fig. 1 Process flow diagrams of flours of unripe banana (**a**), okra (**b**), stevia (**c**) and jujube (**d**)

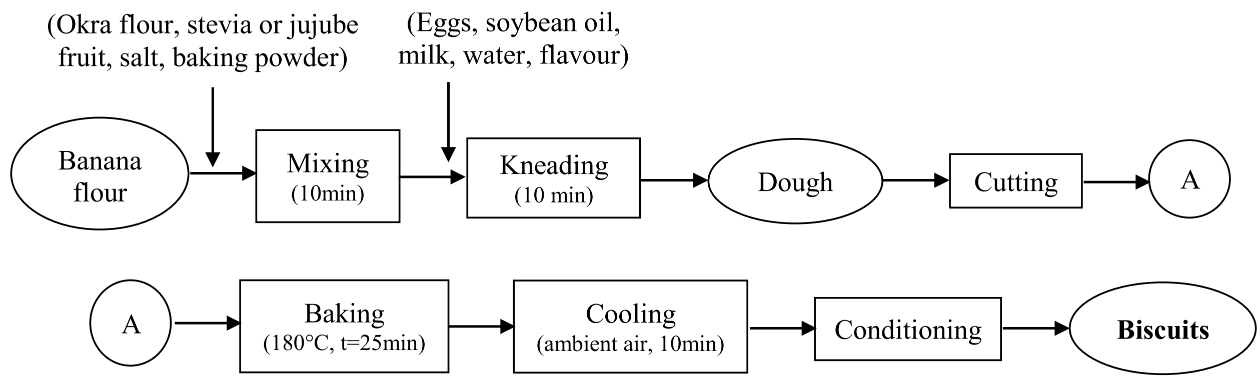


Fig. 2 Process flow diagram for biscuit formulation

Characterisation of biscuits

Moisture, ash, total sugars, simple sugars, proteins, lipids and dietary fibers contents of biscuits were determined as described by Tedom et al. (Tedom et al. 2019).

In vivo analysis

To evaluate the effect of the consumption of biscuits on plasma blood glucose level, an in vivo analysis was performed according to Susiloningsih & Nilasari (Susiloningsih & Nilasari 2018), in the animal section of the Laboratory of Food Biophysics Biochemistry and Nutrition (LABBAN) of the Department of Food Sciences and Nutrition in the National School of Agro-Industrial Sciences (ENSAI) located in the University of Ngaoundere. In brief, 27 "Wistar" rats aged from 10 to 12 weeks, with a body weight between 155 and 315 g were divided in nine batches of three rats each one. The rats were acclimated for 14 days during which they were fed once a day with a standard normal diet (Amin & Nagy 2009). Afterwards the glucose tolerance test (GTT) was carried out after 16 hours of fast. Once their fasting blood plasma glucose was first taken, batch 1 was fed with normal diet (5 g/100 g), batch 2 with a solution of table sugar at 10% (5 ml/100 g), batch 3 with control biscuit (5 g/100 g), batches 4, 5 and 6 with BOJ1, BOJ2 and BOJ3 biscuits (5 g/100 g) respectively, and batches 7, 8 and 9 with BOS1, BOS2 and BOS3 biscuits (5 g/100 g) respectively. Their postprandial blood plasma glucose level was recorded for 2 h every 30 min (Susiloningsih & Nilasari 2018). The experiment was repeated three times, and the Glycaemic Index (GI) determined (Eq. 1) using the incremental area under blood glucose response curve (iAUBGR) calculated (Eq. 2) (Wolever et al. 1991).

Where A, B, C, D, and E correspond to Blood Glucose Level at 0, 30, 60, 90, 120 minutes respectively. It is important to notice that animals did not received no illicit substance and were not sacrificed to carry out the experiment.

$$\mathrm{GI} = \frac{\text{iAUBGR curve after biscuit}}{\text{iAUBGR curve after glucose}} \text{ x } 100 \qquad (1)$$

$$\begin{aligned}\mathrm{iAUBGR} =& (\mathrm{At}/2) + \mathrm{At} + (\mathrm{B}-\mathrm{A})\mathrm{t}/2 + \mathrm{Bt} \\ &+ (\mathrm{C}-\mathrm{B})\mathrm{t}/2 + \mathrm{Ct} + (\mathrm{D}-\mathrm{C})\mathrm{t}/2 \\ &+ \mathrm{Dt} + (\mathrm{E}-\mathrm{D})\mathrm{t}/2 + \mathrm{Et}\end{aligned} \qquad (2)$$

Hedonic test of biscuits

The sensory test was performed in the LABBAN of ENSAI. The produced biscuits were subjected to sensory assessment within the 24 hours after production. Thirty (30) consumers consisted of students of Food Science and Nutrition Department of ENSAI, which are familiar with biscuit (have consumed it from time to time.) were involved in the assessment. It was ensured that all the consumers were neither ill nor allergic to baked products and an oral informed consent was obtained from them. Biscuits were served on a transparent plate. The samples were presented in blocks on the same plate, coded with random numbers. During the sensory evaluation, the consumers were asked to rinse their mouths with table water between samples and to assess the next sample after an interval of 4 min. A sensory evaluation sheet was provided to each consumers, and they scored the product based on colour, appearance, texture, flavour, taste and overall acceptability, on a nine-point hedonic scale from 1 = dislike extremely to 9 = like extremely (Tsikritzi et al. 2014). It was instructed to the consumers to rate colour before tasting each product.

Statistical analysis

Results are presented as means and standard deviation of three determinations. The data was subjected to analysis of variance (ANOVA) and means difference were assess using the least significant difference test at the 5% level of significance performed with Statgraphics centurion XVI.I.

Ethical approval
This manuscript contains studies with animals and sensory evaluation with human participants. Therefore, ethical clearance (N° 01−/2023) was obtained from the ethical review committee of the Department of Food Science and Nutrition, National School of Agro-industrial Sciences, The University of Ngaoundere, Cameroon.

Results and discussion
Proximate composition of flours
Table 1 presents the proximate composition of flours produced. Moisture content of flours was noted to ranges from 2.3 to 6.6%, with jujube having the highest moisture content contrary to stevia which have the lowest moisture content. This result shown that the flours produced can have a stable and long shelf life, knowing that food products with moisture lower than 14% can resist microbial growth and contribute to best storage (Mahloko et al. 2019). With respect of the total ash, okra and stevia flours were found to have high content (1%), which suggests that these two food matrices are rich in minerals than banana and jujube, being given that ash content gives information on the mineral composition of food products (Tedom et al. 2019). Besides, the total ash of banana (0.9%) and jujube (0.7%) were also within the expected range (0.3–1.4%) of ash content in dietary flours. The Crude fibers content in okra, stevia and banana flours is respectively of 23.0, 16.8 and 7.4%, confirming that they are good sources of dietary fibers (Tufaro et al. 2022; Abou-Arab et al. 2009) and making them "high-fiber foods" with values above 6% of dry matter (Ndife et al. 2011). Therefore, theses flours could be suitable in food formulation for diabetics, due to the positive effects of fibers in the body such as improving glucose tolerance control, often decreasing insulin requirements, promoting satiety and in addition tending to lower serum cholesterol and triglyceride values (Augustin et al. 2015). Furthermore, the total sugar content of all the powder were noted to represents more than 50% of dry matter, with the highest value for jujube (82.7%) and the lowest for okra (65.1%). However, apart for jujube, the simple sugar content was found to be low in all the others matrices (3.6% for stevia, 4.5% for okra and 5.7% for banana), meaning that stevia, okra and unripe banana flours are principally made of polysaccharides such as starches and fibers. The results obtain shown that theses floors are suitable for the formulation of biscuits for diabetics owing their low content in simple sugar which are slowly digested, thus did not rapidly increase glucose blood level. Though jujube recorded a high content in simple sugars (17.8%), it is mostly composed of fructose which are simple sugars, therefore, it could lead to the reduction of postprandial glucose and insulin responses (Dornas et al. 2015). Thereby, jujube could be considered as useful ingredients for diabetics' food formulation.

Table 1 Proximate composition of flours produced (g/100 g dry matter)

Samples	Banana flour	Okra flour	Stevia flour	Jujube flour
Moisture	5.0 ± 0.2^{b}	5.2 ± 0.3^{b}	2.3 ± 0.1^{a}	6.6 ± 0.3^{c}
Total Ash	0.9 ± 0.2^{b}	1.0 ± 0.1^{b}	1.0 ± 0.1^{b}	0.7 ± 0.1^{a}
Simple sugar	5.7 ± 0.4^{c}	$4.5 \pm 0.4^{b,c}$	3.6 ± 0.6^{a}	17.8 ± 0.4^{d}
Total sugar	77.0 ± 0.3^{c}	67.3 ± 0.5^{b}	65.1 ± 0.4^{a}	82.7 ± 0.2^{d}
Crude fibers	7.4 ± 0.4^{b}	23.0 ± 0.7^{d}	16.8 ± 0.7^{c}	2.0 ± 0.1^{a}

Values are mean ± standard deviation

Values in the same followed by different superscript row are significantly different ($p < 0.05$)

Table 2 Proximate composition of biscuits (g/100 g of dry matter)

Samples	BOJ1	BOJ2	BOJ3	BOS1	BOS2	BOS3	T
Moisture	2.8 ± 0.2^{c}	2.7 ± 0.7^{abc}	2.6 ± 0.2^{a}	2.5 ± 0.7^{ab}	2.6 ± 0.04^{bc}	2.7 ± 0.1^{bc}	3.2 ± 0.1^{c}
Ash	2.6 ± 0.4^{a}	2.5 ± 0.3^{a}	2.7 ± 0.2^{a}	2.6 ± 0.3^{a}	2.4 ± 0.2^{a}	2.6 ± 0.5^{a}	2.6 ± 0.3^{a}
Simple sugar	25.2 ± 0.7^{b}	23.8 ± 0.6^{b}	22.9 ± 1.1^{b}	11.7 ± 2.1^{a}	11.2 ± 0.3^{a}	10.5 ± 1.4^{a}	38.9 ± 4.2^{c}
Total sugar	71.8 ± 0.3^{a}	72.6 ± 0.7^{ab}	73.2 ± 0.4^{b}	72.1 ± 0.2^{ab}	72.6 ± 0.4^{ab}	72.8 ± 1.2^{ab}	73.0 ± 0.5^{b}
Crude fibers	11.2 ± 0.2^{a}	11.5 ± 0.2^{a}	11.5 ± 0.4^{a}	13.6 ± 0.4^{c}	14.2 ± 0.3^{bc}	13.0 ± 0.3^{b}	11.7 ± 0.5^{a}
Lipids	$14.0 \pm 0.5^{a\,b}$	14.7 ± 0.3^{ab}	14.7 ± 0.3^{b}	14.3 ± 0.3^{ab}	14.5 ± 0^{ab}	13.8 ± 0.8^{a}	13.0 ± 0.5^{ab}
Proteins	12.2 ± 0.2^{a}	12.3 ± 0.2^{a}	12.3 ± 0.3^{a}	12.4 ± 0.7^{a}	13.0 ± 0.1^{a}	12.7 ± 0.6^{a}	12.3 ± 0.3^{a}
Energy (Kcal)	393.7 ± 1.8^{a}	394.1 ± 1.4^{a}	393.1 ± 0.8^{a}	393.5 ± 1.4^{a}	394.3 ± 0.8^{a}	393.6 ± 1.9^{a}	393.5 ± 1.4^{a}

Values are mean ± standard deviation

B Banana, *O* Okra, *J* Jujube, *S* Stevia; *T* Control biscuit

Values followed by different superscript in the same row are significantly different ($p < 0.05$)

Proximate composition of biscuits

The proximate composition and energy value of biscuits produced are presented in Table 2, and there is no significant difference between moisture, ash, total sugars, proteins, lipids content and energy value of BOJ and BOS biscuits. According to the results recorded, simple sugars content in BOJ biscuits is higher than the one of BOS biscuits, while BOS biscuits are richer in fibers. Moisture content ranging from 2.5 to 2.8% is an interesting characteristic of formulated biscuits because this value is expected not to be above 5% as the lower the moisture content of biscuits the longer its shelf life (Thivani et al. 2016). Concerning ash content, the values obtained in this work (2.4–2.6%) were higher than those proposed for normal biscuits (1.01%), and this reflect that the formulated biscuits are good sources of mineral elements which are responsible for preventing many diseases (Saja et al. 2021). Moreover, the fibers content of all the produced biscuit was found to be high in all the formulated due mainly to the presence of okra flour as raw material, but this content is highest in BOS biscuits because in addition to okra, as shown in Table 1, stevia is another good source of dietary fibers. High content of dietary fibers in formulated biscuits in general and in BOS biscuits in particular is benefit for diabetics according to the role of fibers in the body (Augustin et al. 2015). It is well known that diabetes patients need food energy that does not rapidly raise their blood glucose level in order to cover their basal metabolic rate, metabolic response to food, energy cost of physical activities, and growth of new tissue (Gray & Threlkeld 2000). Hence, the produced biscuit may be suitable as snack for diabetes due to their energy values which is that recommended by USDA (2018).

In vivo analysis in rats

Figure 3 depicting the postprandial evolution of blood glucose level in rats after ingested the biscuits, showed a fast increase in blood glucose during the first 30 min and then remained steady up to 120 min. The rats fed with only sugar was found to express the highest increase in postprandial blood glucose level, followed by the batch fed with control biscuit (T), then the one fed with normal diet. Furthermore, Rats fed either BOJ or BOS biscuits were noted to have slow rise in blood sugar. While, those fed with BOS2 resulted to the lowest increase of blood glucose.

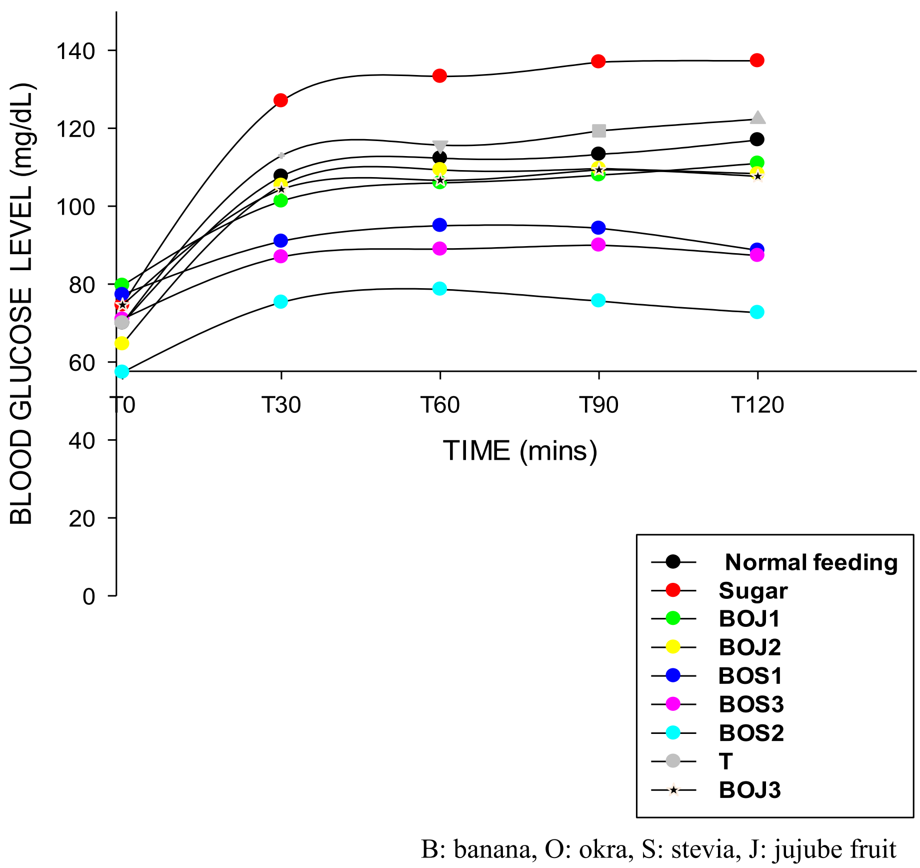


Fig. 3 Curve of postprandial blood glucose level evolution in rats after feeding

The magnitude and time of the peak plasma glucose concentration depend on a variety of factors, including the timing, quantity, and mainly the composition of the meal (Association AD Postprandial 2001). Moreover, ADA (Association AD Postprandial 2001) stated that the content in carbohydrate, the rate of gastric emptying, digestion within the lumen of the small intestine, and the rate of absorption into the portal vein may also influence the glucose absorption. Hence, at T0 the fasting blood plasma glucose in each batch rats were low (57–80 mg/dL). This might be due to the fasting state and the fact that the glucose stores are depleted, thus causing the blood plasma level to decrease (Rui 2014). In fact, fasting induce the reduction of the variability in basal blood glucose (Jensen et al. 2013). As mentioned by ADA (Association AD Postprandial 2001), the rising in glucose concentrations 10 min after the beginning of meal could be attributed to the absorption of dietary carbohydrates.

Furthermore, the sharp increase in plasma glucose level at T30 noted in the batch of rats fed just with table sugar could be associated to their consumption of sucrose (glucose + fructose) principally made of, as simple sugars, such as glucose, are rapidly absorbed into the bloodstream, thereby leading to a swift increase in blood sugar (Holesh et al. 2022). Whereas, the slow increase in blood plasma glucose level at T30 in the batch of rats that were fed only with BOS2 formulation, could be attributed to the effect of its high fiber and starch contents which are complex carbohydrates who need to be digested in glucose before to be absorbed by the body, thus delaying the blood glucose absorption (Rolfes et al. 2014). In addition to it, soluble fiber traps nutrients and delays their transit through the gastrointestinal tract which result in slowing down glucose absorptions, thus preventing glucose spikes (Rolfes et al. 2014). Similarly finding has been reported, stipulating that banana starch resistant towards α-amylase enzyme may reduce the absorption of amylose (Falcomer et al. 2019). In another hand, the highest peak of blood plasma glucose was found at T60. This increment could be ascribed to the digestibility carbohydrate which act the same manner like glucose uptake as reported by Alongi et al. (Alongi et al. 2019)

Glycaemic index of biscuits

The outcomes presented in Table 3 shown that BOS biscuits have lower glycaemic index (GI) than control biscuit made with table sugar records. The lowest GI was found for BOS2 biscuit. This could be due its highest content of okra which is a rich source of fibers (Tufaro et al. 2022) that are known to lower the GI of foods. Alongi et al. (Alongi et al. 2019) have associated the reduction of GI of biscuits made with apple pomace as functional ingredient to its dietary fibers content. Moreover, the viscosity of fiber can have a large influence on foods GI ranking as viscous soluble fibers like those of okra can transform the contents of the intestine into a gel-like /gooey matter. The gooey consistency of this matter slows down the enzymatic activity on starch and so slows down the rate of digestion, thus reduces the rate of rise in blood glucose level (Céspedes et al. 2010). Hence, based on the GI classification index (GI of food is low from 0 to 55, medium from 56 to 69, and high between 70 and 100), the BOS2 may be considered as the best formulated biscuit to regard to the GI of food. Indeed, low GI reflect the slow break down of carbohydrates during digestion, thus a delay in the blood sugar release into the bloodstream allowing a constant blood sugar level (Roze et al. 2021). Besides, the high GI recorded in BOJ formulation might be due to their high content in jujube fruits mainly made of sucrose and the glucose, which their quickly release in the bloodstream lead to high spike in blood sugar levels compared to complex carbs (Holesh et al. 2022).

Table 3 Glycaemic index of biscuits

Samples	Glycaemic index
BOJ1	79.4 ± 0.3^{c}
BOJ2	79.6 ± 1.1^{c}
BOJ3	78.6 ± 1.7^{c}
BOS1	65.7 ± 3.3^{b}
BOS2	54.0 ± 1.0^{a}
BOS3	63.9 ± 1.9^{b}
T	88.1 ± 3.4^{d}

Values are mean ± standard deviation

B Banana, *O* Okra, *S* Stevia, *J* Jujube, *T* control biscuit

Values followed by different superscript in the same row are significantly different ($p < 0.05$)

Glycaemic Index is also affected by the type of starch, ratio amylose/amylopectin and starch dimension, presence of lipids, proteins, food matrix and compounds able to inhibit the action of digestive enzymes (Lal et al. 2021). In particular, the presence of slowly digestible starch and resistant starch could reduce the GI of food product (Raigond et al. 2015). *Musa acuminata* is a good source of resistant starch due to its high in amylose to amylopectin content (Falcomer et al., 2019). Resistant starches have been championed for human health with respect to a number of key physiological responses due to their low susceptibility to hydrolytic enzyme (amylolytic) resulting from altered amylopectin structure (Bede & Zaixiang 2021).

Sensory analysis results

Figure 4 presents the results of sensory analysis of biscuits produced It appeared that in term of overall acceptability, biscuits made with stevia, especially the BOS2 was the

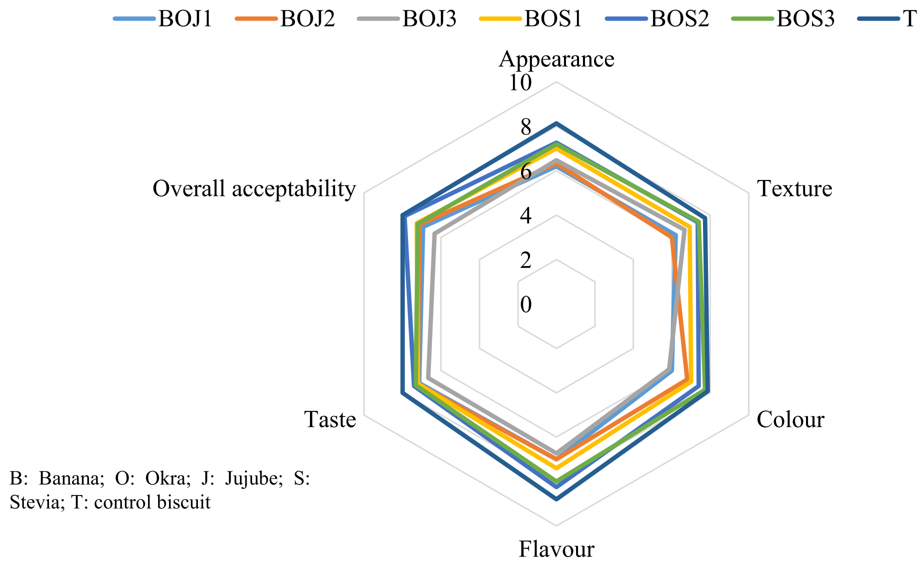


Fig. 4 Sensory results of formulated biscuits

most appreciated among the formulated biscuit with no difference than that of the control biscuit made with table sugar (T). Furthermore, that BOS biscuits were noted to have a higher sweeter taste compare to BOJ biscuits. This finding shown that stevia could be an effective sweetener than jujube. This stems could be ascribed to the contain steviol glycosides (mainly stevioside and rebaudioside) in stevia leaves, which is 200 to 300 times sweeter than sucrose (Brandle 2004), and the fructose responsible for the sweetness of jujube fruits (Hernández et al. 2016).

Conclusion

Biscuits produced with banana, okra and stevia, especially formulation BOS2 recorded the lowest glycaemic index, so they are more suitable for diabetic's diet than those formulated with jujube as sweetener. In term of perspective to this work, it will be interesting to determine if biscuit formulated with banana, okra and stevia has anti-hyperglycaemic and/or hypoglycaemic properties.

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Authors' contributions

This work was carried out in collaboration among all authors. Authors WDN and WT contributed to the conception and design of the work. They also interpreted the data, performed the statistical analysis and wrote the first draft of the manuscript. Author LKN participated in laboratory analysis, acquisition and analysis of data. Author RSM have substantively revised the manuscript, and Author ENF supervised the work from the design of the study to the write up and editing of the manuscript. All authors read and approved the final manuscript.

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Availability of data and materials

All data generated or analysed during this study are included in this published article.

Declarations

Ethics approval and consent to participate

The research in animals was conducted according to the International Council for Laboratory Animal Science (ICLAS) ethical guidelines.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

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