Original Research Article

Investigation on the effect of *Sacoglottisgabonensis* on blood glucose, body weight and behavioural changes of mice exposed to non-nutritive sweeteners during gestation period.

Abstract

Aim of this research was to investigate the role of *Sacoglottsgabonensis* on blood sugar, body weight and behavioural changes of mice exposed to artificial sweeteners.

Study designA total of 16 female mice (mean weight 18.6±2.32g) divided into four groups were used in this study.

MethodologyGroup A received no sweeteners or *S. gabonensis*, B received 50mg of sweeteners only, C received 50mg of sweeteners and 250mg/kg/bw of *S. gabonensis*, D received 250mg/kg/bw of *S. gabonensis* only for 8weeks.

Results show that group A and D had significant ($p<0.05$) decrease in blood glucose concentration with 7.27±3.53 mmol/L and 8.22±1.87 mmol/L respectively compared to group B and C with 9.12±1.03 mmol/L and 8.22±1.87 mmol/L. There was significant ($p<0.05$) increase in body weight in groups A 32.60±7.34g and B 31.38±4.53g compared to groups C, 22.45±15.49g and D, 24.00±14.71g. Gestation length was between 19 and 20 days for the different groups and the behavioural changes such as climbing, running, nesting and restlessness were observed in all the groups.

ConclusionThe administration of *S. gabonensis* did not alter gestation length and behavioural changes during gravid period in experimental animals but altered the blood glucose concentrations However, administration of S. gabonensis is recommended for individuals who are at the risk of developing high blood glucose concentration and increased body weight as it has shown modulatory effect on blood glucose and body weight gain.

Key words: body weight, behavioural changes and gestation

Introduction

Artificial sweeteners have gained popularity as sugar substitutes due to their low-calorie nature and are commonly used primarily by individuals seeking to reduce caloric intake and manage their weight (Huang *et al.,* 2020). There is a correlation between artificial sweeteners and increased body weight, it is presumed to be directly related to the consumption of these substances. Humans have a sweet tooth for desserts and sweet items. This is why such products are consumed much more than the body's nutritional requirement.Food preferences and dietary habits are set from a young age and consumption of foods rich in sugar is one of the main important factors leading to the global obesity pandemic. Body weight increase is due to increased food consumption and taste influences eating behaviour and the desire to eat certain products (Wilk *et al.,* 2022). High-energy diets are one of the major contributors to obesity, which is related to consumption of foods high in sugar and excess of calories in the diet which contributes to excessive body weight.Blood glucose regulation is a critical aspect of overall health, particularly for individuals with diabetes or those at risk of developing this condition (Gardener, 2018). Energy intake has increased with the consumption of animal fat and energy packed foods, while Fiber intake has decreased, changes in dietary shift contributes to the rise of non-communicable diseases, such as obesity, type 2 diabetes, cardiovascular disease, and cancer (Pang *et al.,* 2021).Artificial sweeteners may affect energy balance, and thus body weight, differently compared to natural sugars via underlying physiological processes comprising the gut microbiota, the reward-system, and adipogenesis (Pang *et al.,* 2021).The increase in the prevalence of overweight and obesity the rising interest in losing weight, preventing weight gain and maintaining weight loss have become a global concern. The use of non-caloric sweeteners (NCS) has been established as a replacement strategy for sugar in sweetened beverages, notably in soft drinks. With an intense sweetening power and very low caloric contribution. Since artificial sweeteners themselves do not raise blood glucose, the insulin released during the cephalic phase may result in a transient decrease in blood glucose levels (Palmer *et al.,* 2018). In individuals with insulin resistance, this can lead to an imbalance between insulin and glucose levels (Jensen *et al.,* 2020). With regular consumption of artificially sweetened foods and beverages, the cephalic phase insulin response may become a regular occurrence. Over time, this could contribute to changes in insulin sensitivity and insulin resistance, affecting blood glucose control (Gardener, 2018). Consumption of artificial sweeteners may lead to increased appetite and cravings for sweet and high-calorie foods which can indirectly impact blood glucose levels by encouraging the consumption of sugary foods and drinks that can spike blood glucose (Qin *et al.,* 2020). Aspartame have been linked to possible brain damage which have also been linked to the onset of Alzheimer, seizures, headaches, insomnia and behavioural modifications in humans (Kuk and Brown, 2016) while Acesulfame have been reported to have potential toxic effects such as chromosome aberrations in vivo in mammals (Bandyopadhyay *et al.,* 2008).

There is a dearth of comprehensive scientific studies exploring the precise impact of *Sacoglottisgabonensis* on blood glucose levels, particularly in animal models used in diabetes research (Chia et al., 2016). The stem bark extract is reported to have hepatoprotective properties it contains anti-oxidant propertiesand also reduces oxidative damagein Swiss mice (Wekhe-Emenike, et al., 2022a). Its bark is use as an additive to palm wine or as a medicine, less frequently as a powder for emetic use (Maduka, 2004). The stem bark is taken to treat fever, diarrhoea, gonorrhoea and abdominal pain, and they are used to treat hypertension and diabetes sometimes (Dounias, 2015). a decoction of the crushed bark mixed with leaves of Dioscorea minutiflora as a rectal enema to treat acute abdominal pain, it is used to cure difficult cases of dermatitis. Diluted stem sap is used in hipbaths to promote muscle tone in women after childbirth. Stem bark decoction is mixed with other plants and added to bath water to treat ovarian troubles, vaginal infections and children with fever (Dounias, 2015, Brigitte et al., 2018). Induced electrolytes imbalance by aspartame in Swiss mice was restored by the ethanolic extract of the stem bark (Wekhe-Emenike et al., 2022b). The stem bark is used to prolong the shelf life of palm wine, add potency, reduce foaming, impart a bitter taste and treat arthritis in old people (Morah and Robinson, 2015).

Materials and methods

A total of 16 female pre-pubertal mice born on 03/ 03 /2023and the experiment started after 4 weeks after the birth of the mice. They had an average weight of 18.6g.Experimental animals were housed in standard polypropylene cages which were washed thoroughly with detergent and disinfectant before commencement of the experiment.The animal house was properly ventilated and the animals were given food and water *ad libitum*.They were housed throughout the period of the experiment in a 12hrs light: 12 hrs dark cycle at room temperature of about 26±20° C and relative humidity of 50±20 %, with their beddings (saw dust) changed thrice a week.The experimental animals were exposed for eight weeks to treatments such as artificial sweeteners (sucralose, Acesulfame-k and Aspartame)sold under the trade name ezbonand *Sacoglottisgabonensis*.The experimental animals were weighed with a digital weighing balance and their blood glucose levels were measured once every week with a digital glucometer (Finetest) and data recorded.

Experimental Design

All the groups had four (4) animals each. Group A Received water and feed only, group B received feed, water and 50mg of artificial sweeteners, group C Received feed, water 50mg of artificial sweeteners and 250mgof *Sacoglottisgabonensis*, group D Received feed, water and 250mg of *Sacoglottisgabonensis*.

Statistical analysis

The experimental data were subjected to descriptive statistics using Spss version 22. One way analysis of variance was used to determine significance difference between the different treatment groups.

Results

The effect of artificial sweeteners and *sacoglottisgabonensis* on blood glucose concentration, body weight and behavioural changes in female Swiss mice.

The effect of artificial sweeteners and *S.gabonensis* on blood glucose concentration after exposure for eight weeks shown in table 1. There was a significant difference ($p<0.05$) in blood glucose concentration in the first week, group A had asignificant decrease, while group C that received sweeteners and *S. gabonensis* had significant increase inblood glucose. There was significant increase ($p<0.05$) in group B that received only sweeteners, while a significant decrease in glucose concentration was recorded in group D that received *S.gabonensis* alone at weeks 2 and 3 respectively. Group A had significant increase ($p<0.05$) while group D had significant decrease($p<0.05$) at week 4 and 5 respectively. Group B had a significant increase ($p<0.05$) while group C had significant decrease ($p<0.05$) at week 6. Group A had significant increase ($p<0.05$) while group C had significant decrease ($p<0.05$) at the 7th and 8th week.

Table 1: The effect of artificial sweeteners and Sacoglottisgabonensis on blood glucose concentration in female Swiss mice

Blood Glucose				
Weeks	Group A	Group B	Group C	Group D
Week 1	7.27 ± 3.53^{a}	9.12 ± 1.03^{a}	10.17 ± 0.97^{a}	8.22 ± 1.87^{a}
Week 2	6.62 ± 1.70^{ab}	8.15 ± 1.15^{ab}	6.35 ± 1.00^{abc}	4.92 ± 3.54^{ab}

Week 3	6.00±1.06[ab]	6.70±1.39[c]	6.67±0.94[abc]	4.50±3.01[ab]
Week 4	7.35±0.94[a]	6.15±0.79[c]	6.50±0.89[abc]	4.22±2.83[ab]
Week 5	6.25±0.23[a]	5.95±0.31[c]	4.90±3.30[bc]	4.90±3.46[ab]
Week 6	5.77±0.66[b]	6.87±0.28[bc]	4.25±2.84[bc]	4.37±2.94[ab]
Week 7	6.85±1.77[ab]	6.27±0.83[c]	4.40±3.04[bc]	5.92±4.00[bc]
Week 8	6.92±1.92[ab]	5.75±0.80[c]	4.57±3.08[bc]	5.42±3.66[bc]

*Values are mean±SD, values with the same superscript letters are not significantly different, whereas those with different superscript letters are significantly different ($p<0.05$)

The effect of artificial sweeteners and *S gabonensis*on body weight of female Swiss mice after exposure for eight weeks is shown in table 2. There was a significant decrease ($p<0.05$) in body weight in group A, while the other groups showed significant increase($p<0.05$), with group B having the highest body weight between the 1st week and 5th week. Group A recorded significant increase between the 6th week and the 8th week at ($p<0.05$). Group C and D had significant decrease ($p<0.05$) in body weight.

Table 2: The effect of artificial sweeteners and *S. gabonensis* on body weight of female Swiss mice

Weeks	Group A	Group B	Group C	Group D
Week 1	15.80±1.81[e]	21.83±0.90[d]	19.07±1.40[c]	18.78±1.19[ab]
Week 2	17.92±1.69[de]	23.23±0.82[cd]	20.18±1.42[b]	15.41±10.34[c]
Week 3	20.90±1.03[cde]	24.53±0.62[cd]	21.59±0.96[bc]	18.34±12.37[ab]
Week 4	22.81±1.76[bcd]	25.10±1.25[c]	23.14±2.21[a]	19.37±13.02[ab]
Week 5	23.82±1.60[bc]	25.46±1.32[c]	18.72±12.58[c]	20.98±14.00[b]
Week 6	27.17±3.12[b]	25.93±0.98[bc]	18.72±12.58[c]	20.99±14.12[b]
Week 7	32.83±5.09[a]	28.61±2.07[ab]	19.87±13.44[b]	21.54±14.65[b]

Week 8	32.60±7.34[a]	31.38±4.53[a]	22.45±15.49[ab]	24.00±14.71[a]

*values are mean±SD, values with the same superscript letters are not significantly different, whereas those with different superscript letters are significantly different (p<0.05)

The behavioral changes exhibited by the experimental animals was the same for all groups during parturition. Changes such as nesting, restlessness, tail biting, itching, scratching, running, shivering and heavy breathing was observed before they gave birth.

Table 3: Behavioural changes of female Swiss mice exposed to artificial sweeteners and *Sacoglottisgabonensis* during parturition.

Days	Behavioral changes
Day 14	Tail biting/Nesting/Itching/Scratching
Day 15	Nesting/Restlessness/ Itching/Scratching
Day 16	Enlarged nipples/Tail biting/ Itching/Scratching
Day 17	Nesting/Running
Day 18	Restlessness/shivering/tail biting
Day 19	Heavy breathing/Tail biting

Discussion

There is increasing evidence that sugarsweetened beverages, categorically those rich in fructose, have had a significant contribution to the worlds increase in metabolic diseases. The detrimental effects of fructose intake during pregnancy on both mother and offspring, have been clearly outlined (Malik *et al.,* 2010). The exploration of blood glucose levels in female Swiss mice exposed to artificial sweeteners and *Sacoglottisgabonensis* unfolds a narrative of dynamic physiological responses.Group A, serving as the control group with no substance exposure, displayed relatively higher blood glucose levels throughout the study period. Marginal fluctuations were also observed with group B showinginitial rise in weeks 1 and 2, followed by a gradual decline, which suggests a potential adaptation or regulatory response to the artificial sweeteners when compared with group A(Suez et al., 2014).". These results underscore the importance of understanding the baseline dynamics in the absence of substance exposure. The sustained decrease recordedfrom week 5 onwards to week 8 when compared with the control group may indicate a level of regulation or an adaptive mechanism in response to continued exposure.The blood glucose levels in group C which received artificial sweeteners and *Sacoglottisgabonensis*represents a distinctive trajectory. The initial elevation in weeks 1and 2, followed by a significant decrease in subsequent weeks, hints at a potential ameliorating effect of*Sacoglottisgabonensis*(Johnson *et al.,* 2015) and also, the drastic decline in week 5 suggests an intriguing mitigating effect on blood glucose levels.Group D, which received*Sacoglottisgabonensis* only displayed stable blood glucose levels throughout the study, with a slight increase in weeks 5 and 7. This steady pattern suggests that *Sacoglottisgabonensis*, when administered alone, has a modest influence on blood glucose regulation.

Artificial sweeteners have been reported to have various effects on different organisms, including mice. Some sweeteners have showneffect on the hormonal balance, metabolism, or the perception of sweetness, which could lead to behavioural changes. Behavioural and physical changes such as restlessness, shivering, nesting, enlarged nipples, tail biting heavy breathing and runningobserved during the course of this research cannot be attributed to the consumption of artificial sweeteners such as aspartame, sucralose and acesulfame K, neither can it also be attributed to the consumption of *S gabonensis* as such changes and behaviours have been witnessed in normal reproducing mice.

Conclusion

The investigation of the effects of artificial sweeteners and *Sacoglottisgabonensis* on the blood glucose levels in female Swiss mice has provided valuable insights into the potential impacts of *Sacoglottisgabonensis* as a substance that can reduce high blood glucose concentration.The intriguing decline in blood glucose levels in Group C suggest a potential mitigating effect of *Sacoglottisgabonensis* when combined with artificial sweeteners. The stability in blood glucose concentration in Group D prompts consideration of the potential therapeutic or regulatory properties of *Sacoglottisgabonensis* on glucose metabolism.

Conflict of interest

The authors have declared that there is no conflict arising from this research work.

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