Journal of Pharmacology and Toxicology

ISSN 1816-496X

Academic Journals Inc.

www.academicjournals.com

OPEN ACCESS Journal of Pharmacology and Toxicology

ISSN 1816-496X
DOI: 10.3923/jpt.2017.161.169

Research Article
Antidiabetic Activity and Safety Used of Nutrasucre, a Food Complement, in Alloxan Induced Diabetic Rats

[1]Pierre Manda, [2]Nomane Bernard Goze, [3]Arthur Stephane Gnagne, [4]Alain Didier Abounan, [1]Aholia Jean Baptiste Adepo and [1]Djédjé Sebastien Dano

[1]Laboratory of Toxicology, Research-Training Unit of Pharmaceutical and Biological Sciences, University Felix Houphouêt Boigny, P. O. Box V 34 Abidjan, Côte.d'ivoire
[2]Laboratory of Physiology, Pharmacology and Pharmacopeia, Research-Training Unit of Sciences of nature, University Nangui Abrogoua, 02 P. O. Box 801, Abidjan 02, Côte.d'ivoire
[3]Laboratory Djeka Pharmaco, P. O. Box 683 Agboville, Côte.d'ivoire
[4]Laboratory of Anathomopathological, Research-Training Unit of Medical Sciences, University Felix Houphouët Boigny, Abidjan, Côte.d'ivoire

Abstract

Background and Objective: Type 2 diabetes is one of the major public health problem arround the world and particularly in Côte d'Ivoire. The relation ship between sugar consumption and type 2 diabetes treatment remained critical to be discovered. Nutrasucre, a food complement containing brown sugar and extracts of medicinal plants used for the treatment of type 2 diabetes in Côte d'Ivoire. This study aimed nutrasucre's aptitude to reduce glycemia and its safety used may provide a likely profit by supporting sugar consumption in type 2 diabetes treatment. **Materials and Methods:** Thirty five diabetics rats received nutrasucre solution at 1.43, 2.86 and 5.71 g kg^{-1} b.w., respectively. Blood withdrawals were performed at the end of treatment. Biochemical parameters were assessed: Glycemia, urea, creatinine, transaminases (ALAT, ASAT), triglycerides, total cholesterol, HDL cholesterol and LDL cholesterol. Acute and subacute toxicity were carried out using OECD Guidelines 423 and 407, respectively. Kidneys, heart and liver tissues were collected and subjected to microscopical analysis. One-way analysis of variance (ANOVA 1) and multiple comparisons of Tukey-Kramer *post-hoc* test were being used. **Results:** Nutrasucre possessed antihyperglycemic effect comparable to glibenclamide, a reference antihyperglycemic drug and increased triglyceride, total cholesterol, HDL and LDH cholesterols levels. This increase was significant for triglycerides ($p<0.05$) compared to diabetic controls receiving only the 0.9% NaCl solution. Nutrasucre also decreased ASAT and ALAT serum levels at the three levels tested. The median acute toxicity LD_{50} value of nutrasucre was higher than 5000 mg kg^{-1} b.w. and was classified as non toxic in the Globally Harmonized System of Classification and Labelling of Chemicals (GHS). No signs of subacute toxicity were recorded during the 28 observation days. Finally, the repeated administration of nutrasucre did not affect various vital organs. **Conclusion:** It is concluded that nutrasucre was antihyperglycemic, nontoxic and does not affect vital organs such as kidneys, liver and heart. It reduces serum ASAT, ALAT and total, LDH and HDL levels and contains hypolipidic substances. It can be advised as antidiabetic food complement.

Key words: Nutrasucre, food complement, antihyperglycemic activity, diabetic, biochemical parameters, acute toxicity, subacute toxicity

Citation: Pierre Manda, Nomane Bernard Goze, Arthur Stephane Gnagne, Alain Didier Abounan, Aholia Jean Baptiste Adepo and Djédjé Sebastien Dano, 2017. Antidiabetic activity and safety used of nutrasucre, a food complement, in alloxan induced diabetic rats. J. Pharmacol. Toxicol., 12: 161-169.

Corresponding Author: Pierre Manda, Laboratory of Toxicology, Research-Training Unit of Pharmaceutical and Biological Sciences, University Felix Houphouêt Boigny, P. O. Box V 34 Abidjan, Côte.d'ivoire Tel: 00225 05 69 87 26

Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Nutrasucre is a food complement that has been renewed interest in phytotherapy, especially for the treatment of chronic diseases, such as diabetes in Côte d'Ivoire. Diabetes is a chronic metabolic disease that affects 5-7% of the world population[1]. This high prevalence is due to demographic growth, aging of the world population, unbalanced diet, obesity and a sedentary life style[2]. Modern antidiabetic drugs present many adverse effects and can be very costly. In the face of such problems, traditional medicine provides hope for the impoverished populations of Africa. The addition of scientific proof to the existing ethnobotanical evidence of the therapeutic efficiency of plant-derived drugs would improve consumer confidence, contributing to the popularization of their use[3]. A study reported that 25% of medical prescriptions contain substances derived from plants so that the herbal remedies received a great attention as alternative to synthetic pharmaceutical products, leading to the increase in their demand[4,5]. In accordance with the World Health Organization (WHO) recommendations[3], an Improved Traditional Medicine (ITM), nutrasucre, was evaluated. This product consists of a mixture of brown sugar and plant extracts and is sold by pharmacies and supermarkets in Abidjan. There are present no studies on the use of nutrasucre in diabetes. The aim of this study was to evaluate the therapeutic efficacy and safety of nutrasucre.

MATERIALS AND METHODS

Materials

Test substance: The test substance was nutrasucre. It is comprised of bioactive ingredients (flavonoids, glycoproteins, terpenes and polysaccharides) and excipients (zinc gluconate, magnesium stearate, calcium carbonate and cholecalciferol), all mixed in commercial brown sugar. This product is sold in pharmacies and supermarkets.

Animals: The experiments were carried out in February 2016 and were performed on 12-16 weeks-old male and female normoglycemic and alloxan-induced diabetic albino Wistar rats. They were maintained in a 12 h dark/light cycle at a temperature of 20-22°C in the animal house of the Laboratory of Toxicology of the Pharmaceutical and Biological Sciences of the University Félix Houphouet Boigny (Abidjan, Côte d'Ivoire). They were fed standard pellets and given water *ad libitum*. They were acclimatized to their cages for 2 weeks prior to experiments. All experimental protocols were performed in full compliance with the European Council Legislation 87/609/EEC for the protection of experimental animals.

Drugs and chemicals: The following drugs were used: Alloxan (Sigma, USA), anhydrous glucose (Sigma, USA), commercial brown sugar (Côte d'Ivoire), formol (Sanofis, France) and ether (VWR International-Geldenaakfebaan 464-B-3001 Leuven-Belgium). All chemicals regents were analytical grade.

Methods

Preparation of nutrasucre solutions: The product was prepared by the promoter and delivered to the laboratory in its commercial form. The sample used for the study was from lot 06/SUC/015 (date of production: 02/2015, expiration: 02/2017). A measuring instrument in the bottle (a white plastic spoon) is used by the consumer. The average weight of one spoonful of sugar weight was estimated to be 10.43±1.02 g (n = 10). Three doses were specified: 100, 200 and 400 g, corresponding to 1.43, 2.86 and 5.71 g kg^{-1} b.wt., for an adult of 70 kg, respectively.

Oral glucose tolerance test of rats to assess the antihyperglycemic activity of nutrasucre: The method described by Keita *et al.*[6] was followed, with slight modifications. Thirty-six rats were deprived of food for at least 18 h prior to the experiments. The normoglycemic rats were randomized into six groups of six rats each. All animals received the test solutions by oral gavage. Group 1 (negative control) received saline solution (0.9% sodium chloride) at 2 mL kg^{-1} b.wt. Group 2 received only commercial brown sugar at a dose of 5.71 g kg^{-1} b.wt., Group 3 received glibenclamide (antidiabetic reference drug) at 5 mg kg^{-1} b.wt. Groups 4, 5 and 6 were given nutrasucre at doses of 1.43, 2.86 and 5.71 g kg^{-1} b.wt., respectively. Thirty minutes after oral administration of the substances, all animals received an anhydrous glucose solution at 10 g kg^{-1} b.wt., by oral gavage. Blood withdrawals were performed by nicking the tails with a sharp razor, after disinfection with alcohol, every 30 min, for 4 h. TheGlycemia was directly determined using a One Call Plus glucometer (USA).The results are expressed in mg dL^{-1} blood.

Testing of antihyperglycemic activity of nutrasucre in diabetic rats

Induction of experimental diabetes: The method described by Nagappa et a*l.*[7] was followed, with slight modifications. Normoglycemic rats of both sexes were deprived of food for

at least 18 h and diabetes was induced by intraperitoneal administration of alloxan at 300 mg kg^{-1} (diluted in 0.9% sodium chloride). Anhydrous glucose (5%) was administered to rats at 10 g kg^{-1} b.wt., to stabilize them and reduce the oxidant effect of alloxan. Glycemia was recorded after one week and only rats with glycemia higher than 200 mg dL^{-1} were selected for further experiments.

Antihyperglycemic activity of nutrasucre in diabetic rats: Thirty-five diabetic rats were divided into five lots of seven rats. Group 7 (negative control group) received a 0.9% saline solution at 2 mL kg^{-1} b.wt. Group 8 (positive control lot) received glibenclamide (antidiabetic reference drug) at 5 mg kg^{-1} b.w. Groups 9-11 received the nutrasucre solution at doses of 1.43, 2.86 and 5.71 g kg^{-1} b.wt., respectively.

A single daily dose of the products was administered over a period of 28 days. Blood withdrawals were performed by nicking the tails with a sharp razor, after disinfection with alcohol, at the end of treatment, to determine the glycemia. The reading of glycemia was directly determined using a One Call Plus glucometer (USA). The result was expressed in mg dL^{-1} blood.

Toxicity study of nutrasucre in nondiabetic rats

Acute toxicity: The experiment was performed according to OECD Guideline 423[8]. The median acute LD_{50} toxicity value of nutrasucre was determined in rats using the limit test to 5000 mg kg^{-1} b.wt.

Subacute toxicity: Subacute toxicity was assessed according to OECD Guideline 407[9], which consists of the daily administration of nutrasucre via the oral route to 4 groups of rats with one dose per group for 28 days. Fifty nondiabetic rats were randomly divided into 4 groups of 10 animals, consisting of three test groups and a control group. Each group included five males and five females. An additional satellite group of 10 rats in the group treated with the highest dose (5.71 g kg^{-1} b.wt.) was included to observe the reversibility, persistence, or late appearance of toxic effects at least 14 days after stopping the treatment.

Group 12 (nondiabetic rats and the control group), received a 0.9% saline solution at 2 mL kg^{-1} b.wt. Groups 13, 14 and 15 (nondiabetic rats) received the nutrasucre solution at doses of 1.43, 2.86 and 5.71 g kg^{-1} b.wt., respectively.

Blood samples were taken from each rat at the end (D28) of the experiment for biochemical analysis. The blood samples were collected in dry tubes and/or tubes containing the anticoagulant, EDTA. The blood was centrifuged at 3000×g at 4°C for 5 min. The resulting serum was stored in wells at -20°C until biochemical analysis. The following parameters were assessed: Blood glucose, urea, creatinine, transaminases (ALAT, ASAT), triglycerides, total cholesterol, HDL cholesterol and LDL cholesterol. The animals were sacrificed at the end of the experiment (D28) by ether overdose. The kidneys, heart and liver were removed and stored in a 10% formol solution for pathological examination.

Clinical signs and ponderal evolution: We followed weight changes and clinical signs of toxicity for each dose during the 28 day period of the experiment. The clinical signs of toxicity that were assessed were: Behavioral anomalies (apathy, excitation or excessive grooming), skin and mucous membrane anomalies, signs of partial or generalized problems of the nervous system (coma, tremors or convulsions), signs of digestive troubles (refusal of food or drink, changes in the volume or aspect of the feces), signs of corrosive or vascular effects (oral, nasal or anal bleeding) and mortality.

The animals were observed during the first 4 h after administration of the product and on the 2nd and 28th days.

Bio-statistics: All assays were repeated 3 times. Comparisons between groups versus controls were made using one way ANOVA[10] test and values of $p<0.05$ were considered statistically significant using GraphPad Prism 5.01 (San Diego, California, USA) software. The analysis was completed by multiple comparisons of the average values of the different parameters using the Tukey Kramer[11] *post-hoc* test, if significant differences were revealed between the tested averages.

RESULTS

Effect of nutrasucre on oral glucose overload in rats: The results of the experiments to test the antihyperglycemic effect of nutrasucre on oral glucose overload in rats are presented in Table 1. Oral administration of anhydrous glucose (10 g kg^{-1}) and commercial brown sugar (5.71 g kg^{-1}) led to a significant increase ($p<0.05$) in glycemia after 60 min, which reached in the saline control (group 1) animals and in animals of group 2 (commercial brown sugar control group). From 60-240 min after the administration of anhydrous glucose, glycemia significantly decreased ($p<0.05$), in group 1 (saline control group) and in group 2.

In contrast, pretreatment of rats with glibenclamide at 5 mg kg^{-1} blocked hyperglycemia at 60 min after the administration of glucose, which significantly dropped by 180 min. This hypoglycemia lasted until 240 min comparable

Table 1: Effect of nutrasucre on oral glucose overload

			Glycemia (mg dL^{-1}) (%)*						
Groups	Description	Dose	$T_{0\ min}$	$T_{30\ min}$	$T_{60\ min}$	$T_{90\ min}$	$T_{120\ min}$	$T_{180\ min}$	$T_{240\ min}$
Group 1	0.9% NaCl control	2 mL kg^{-1}	44.7±0.42	55.3±0.76 (19.16)	118±1.63 (62.11)	111±1.42 (59.72)	105±2.06 (57.42)	73±3.30 (38.76)	48.5±1.43 (7.83)
Group 2	Brown sugar control	5.71 g kg^{-1}	80.5±347	151±5.67 (46.68)	179±7.09 (71.78)	164±8.79 (50.91)	125±6.76 (35.6)	90.3±9.23 (10.85)	84.2±2.02 (4.39)
Group 3	Glibenclamide	5 mg kg^{-1}	43.8±3.89	85.2±1.14 (48.59)	61.8±0.60 (29.12)	49.5±0.76 (11.51)	40.3±0.56 (-8.68)	31.5±0.85 (-39.04)	45±0.77 (-8)
Group 4	Nutrasucre	1.43 g kg^{-1}	54.8±1.17	108±1.39 (49.25)	133±2.19 (58.79)	121±1.8 (54.71)	108±1.54 (49.25)	82.8±3.02 (33.81)	76.8±4.15 (28.64)
Group 5	Nutrasucre	2.86 g kg^{-1}	65.5±3.52	117±4.98 (44.01)	152±6.11 (56.9)	126±4.32 48.01)	105±1.38 (37.61)	69.5±4.33 (5.75)	68.5±4.31 (4.37)
Group 6	Nutrasucre	5.71 g kg^{-1}	78.5±2.88	134±3.78 (41.41)	121±3.48 (35.12)	112±4.65 (29.91)	116±3.39 (32.32)	60.5±1.73 (-22.92)	65.2±2.14 (-20.39)

*Percentage increase relative to T_0; 0.9% NaCl: Control group treated witha 9% sodium chloride solution. n = 6 for each group, There was a significant decrease in glycemia in groups 4, 5 and 6 treated with nutrasucre relative to the NaCl control group, Values are given as mean standard error followed on average

Table 2: Effect of nutrasucre in alloxan-induced diabetic rats

			Glycemia (mg dL^{-1})	
Groups	Description	Dose	D_0	D_{28} (% of reduction)
Group 7	0.9% NaCl	2 mL kg^{-1}	96.16±12.03	94±12.75
Group 8	Glibenclamide	5 mg kg^{-1}	305±13.54	94.2±13.80 (69.11)
Group 9	Nutrasucre	1.43 g kg^{-1}	339±12.15	151±12.80 (55.45)
Group 10	Nutrasucre	2.86 g kg^{-1}	342.17±13.81	138±14.41 (59.49)
Group 11	Nutrasucre	5.714 g kg^{-1}	345.32±15.74	109±13.5 (68.40)

D: day, 0.9% NaCl: Control group treated with a 0.9% sodium chloride solution. n = 7 for each group. The anti hyperglycemic effect of nutrasucre was dose dependent and significant relative to the 0.9% NaCl control group. Values are given as mean standard error followed on average

to that of the saline control group (group 1). Treatment of the rats with nutrasucre (groups 4, 5 and 6) resulted in a significantly lower level of glycemia than control group 2 (brown sugar) at 60 min, which continued out to 180 min. This effect was dose dependent. This hypoglycemia was then constant until 240 min. The average glycemia of animals treated with the three doses of nutrasucre were significantly lower than the average glycemia of the animals treated with commercial brown sugar only (group 2).

Effect of nutrasucre in alloxan induced diabetic rats

Alteration of the glycemia in diabetic rats treated by nutrasucre: The results of experiments to test the effect of nutrasucre on the glycemia of diabetic rats are presented in Table 2. In the absence of treatment, the initial glycemia in control rats which received the 0.9% sodium chloride solution (group 7) remained constant until the end of treatment. Daily oral administration of glibenclamide at 5 mg kg^{-1} to the alloxan-induced diabetic rats (group 8) during the 28 days of the experiment significantly reduced ($p<0.05$) glycemia.

Treatment of alloxan-induced diabetic rats with nutrasucre (group 9, 10 and 11), for 28 days, significantly decreased ($p<0.05$) the glycemia. This decrease in glycemia of the rats that received nutrasucre was dose dependent and although significant, was less than that of the rats treated with glibenclamide (group 8).

Effect of nutrasucre on the weight gain of alloxan-induced diabetic rats: Animals of control group (group 7) that received the 0.9% sodium chloride solution showed a small weight gain by the end of the experiment (Table 3). Animals treated with glibenclamide or nutrasucre showed weight gain.

Effect of nutrasucre on biochemical parameters in rats

Action of nutrasucre on lipid levels in alloxan-induced diabetic rats: Treatment of diabetic rats with nutrasucre at concentrations of 1.43 and 2.86 g kg^{-1} for 28 days increased triglyceride, total cholesterol, HDL and LDH cholesterols levels. This increase was significant for triglycerides ($p<0.05$) compared to the diabetic controls receiving only the 0.9% NaCl solution. The dose of 5.71 g kg^{-1} (group 11) resulted slight reduction of lipidic parameters relative to the two control groups (group 7 and 8). All results are presented in Table 4.

Effect of nutrasucre on hepatic enzymes in alloxan induced diabetic rats: Nutrasucre treatment of alloxan-induced diabetic rats significantly reduced ($p<0.05$) ASAT levels. This reduction was dose dependent (Table 4). However, nutrasucre had an opposite effect on ALAT levels.

Table 3: Ponderal evolution of alloxan-induced diabetic rats treated for 28 days

			Weight gain (g)		
Groups	Description	Dose	D_0	D_{28}	change
Group7	0.9% NaCl	2 mL kg^{-1}	148.38±17.57	145.6±12.3	+ 2.78
Group 8	Glibenclamide	5 mg kg^{-1}	125.53±22	130.28±22.52	+4.75
Group 9	Nutrasucre	1.43 g kg^{-1}	131.68±28	140.4±34.16	+8.72
Group10	Nutrasucre	2.86 g kg^{-1}	144.54±17	158.9±19.40	+14.36
Group11	Nutrasucre	5.71 g kg^{-1}	146.62±17	154.28±19.84	+7.66

D: day, 0.9% NaCl: Control group treated witha 0.9% sodium chloride solution, animals in groups 9, 10 and 11 treated with nutrasucre exhibited normal weight gain. Values are given as mean standard error followed on average

Table 4: Effect of nutrasucre on biochemical parameters of diabetic rats after treatment for 28 days

Groups	Description	Urea (mg dL^{-1})	Creatinine (mg dL^{-1})	ASAT (UI L^{-1})	ALAT (UI L^{-1})	Triglyceride (mg dL^{-1})	T Cholest (mg dL^{-1})	HDL (UI L^{-1})	LDH (UI L^{-1})
Group 7	Distilled water control	0.28±0.03	8.91±1.06	160±24.2	74±6.1 3	0.14±0.01	0.3±0.05	0.48±0.04	3046±245
Group 8	Glibenclamide	0.29±0.01	10.3±0.26	164±13	72±6.11	0.14±0.04	0.27±0.04	0.53±0.05	3362±345
Group 9	Nutrasucre (1.43 g kg^{-1})	0.36±0.18	7.98±0.6	161±3	56±2	0.27±0.03	0.33±0.03	0.55±0.05	3073±441
Group 10	Nutrasucre (2.86 g kg^{-1})	0.58±0.15	9.99±1.08	147±6.36	79.3±16.9	0.23±0.09	0.37±0.06	0.55±0.01	3169±126
Group 11	Nutrasucre (5.71 g kg^{-1})	0.42±0.02	8.50±0.39	145±6.33	82.7±6.27	0.12±0.02	0.26±0.03	0.44±0.03	2989±443

ASAT and ALAT: Hepatic enzymes, T Choles: Total cholesterol, HDL and LDH cholesterols. Distilled water control: Diabetic rats having received distilled water, n = 7 for each group. Values are given as mean standard error followed on average

Table 5: Ponderal evolution in non-diabetic rats

			Weight increase (g)		
Groups	Description	Dose	D_0	D_{28}	Change
Group 12	0.9% NaCl	2 mL kg^{-1}	130.58±15.91	154.55±14.84	+23.97
Group 13	Nutrasucre	1.43 g kg^{-1}	113.43±18.45	128.98±17.93	+15.55
Group 14	Nutrasucre	2.86 g kg^{-1}	128.01±11.29	144.16±15.55	+16.15
Group 15	Nutrasucre	5.71 g kg^{-1}	124.16±23.65	133.61±27.13	+9.45

D: Day, 0.9% NaCl: Control group treated with a 0.9% sodium chloride solution, n = 10. Values are given as mean standard error followed on average

Effect of nutrasucre on renal function in alloxan-induced diabetic rats: Treatment of diabetic rats with the three doses of nutrasucre significantly reduced ($p<0.05$) average creatinine levels while no modification was observed in creatinine levels for the control group receiving the 0.9% sodium chloride solution and those treated with glibenclamide. In contrast, oral administration of nutrasucre caused a slight increase in urea levels in alloxan-induced diabetic rats relative to the control group that received the 0.9% sodium chloride solution and those treated with the glibenclamide. The results are presented in Table 4.

Histology of rat organs in alloxan-induced diabetic ratstreated with nutrasucre: Histological sections of various organs (kidney and liver) of the diabetic rats treated with various doses of nutrasucre were examined. The organs of the diabetic rats of group 8 treated with glibenclamide did not show any distinguishing features. The histology of the hepatic tissue of the diabetic rats treated with nutrasucre at 5.71 g kg^{-1} (group 11) also did not shown any particularities. However, renal tissues from the same animals showed atrophy of the proximal and distal circumvented tubes and the glomeruli and vascular congestion in the interstitium.

Acute toxicity: No acute toxicity after oral administration of nutrasucre was observed to the limiting dose of 5000 mg kg^{-1}. All animals survived to the end of the 14 day observation period, implying that the LD_{50} is higher than 5000 mg kg^{-1}.

Subacute toxicity

Effect Effect of nutrasucre on weight gain: No particular clinical signs of subacute toxicity was observed during the 28 days of treatment. All animals of the control group and the three nutrasucre treated groups experienced normal weight gain (Table 5).

Effect of nutrasucre on rat biochemical parameters

Effect of nutrasucre on the lipidic profile: Oral administration of nutrasucre at a concentration of 2.86 g kg^{-1} did not significantly modify average triglyceride levels relative to the control group. In contrast, treatment of the rats with nutrasucre at concentrations of 1.43 or 5.71 g kg^{-1} significantly

Table 6: Effect of nutrasucre on biochemical parameters of non-diabetic rats after treatment for 28 days

Groups	Description	Urea (mg dL^{-1})	Creatinine (mg dL^{-1})	ASAT (UI L^{-1})	ALAT (UI L^{-1})	Triglyceride (mg dL^{-1})	T Cholest (mg dL^{-1})	HDL (UI L^{-1})	LDH (UI L^{-1})
Group 12	Distilled water control	0.32±0.06	8.87±0.57	179±12.7	116±2	0.1±0.009	0.25±0.06	0.4±0.07	2744±129
Group 13	Nutrasucre (1.43 g kg^{-1})	0.29±0.04	8.81±0.37	171±4.67	103±4.67	0.22±0.07	0.26±0.09	0.46±0.01	3017±285
Group 14	Nutrasucre (2.86 g kg^{-1})	0.34±0.01	8.64±0.31	160±5.77	101±8.82	0.11±0.01	0.32±0.03	0.41±0.08	2681±230
Group15	Nutrasucre (5.71 g kg^{-1})	0.34±0.01	7.42±0.06	184±44	102±2.6	0.28±0.18	0.27±0.07	0.49±0.07	3320±548

ASAT and ALAT: Hepatic enzymes, T Cholest: Total cholesterol consisting of HDL and LDH cholesterols, n: 10 for each group. Values are given as mean standard error followed on average

increased ($p<0.05$) triglyceride levels (Table 6), where as total and HDL cholesterol levels did not significantly increase relative to those of the control group (Table 6).

Effect of nutrasucre on hepatic enzymes: Treatment of rats with nutrasucre significantly reduced ($p<0.05$) average ALAT levels at all three tested concentrations with the levels of these enzymes. Treatment with the two lowest concentrations of nutrasucre significantly reduced ($p<0.05$) ASAT levels, but the highest concentration increased the level of this enzyme (Table 6).

Effect of nutrasucre on renal function: There were no There were nosignificant differences between the creatinine values of the 1.43 and 2.86 g kg^{-1} level of nutrasucre groups and the control group. In contrast, treatment with the highest dose of nutrasucre reduced creatinine levels. There were no significant differences between the average urea levels of the three treated groups and the sodium chloride control group (Table 6).

Histology of the nondiabetic rat organs treated by the nutrasucre: Histological sections of the various organs (kidney and liver) of non diabetic rats treated with various doses of nutrasucre were examined. The organs of the control group (group 12) did not show any distinguishing characteristics. The histology of the hepatic tissue of the rats treated with nutrasucre at 5.71 g kg^{-1} (group 15) also did not shown any particularities. However, renal tissues from the same animals showed atrophy of the proximal and distal circumvented tubes and the glomeruli and vascular congestion in the interstitium.

DISCUSSION

This study showed that the nutritional complement nutrasucre has antihyperglycemic activity. Indeed, nutrasucre, like the antihyperglycemic drug glibenclamide, slowed the increase in glycemia in rats by 30 min after administration of a glucose bolus, preventing it from reaching the peak levels obtained in the saline control and the other groups 60 min after administration. Furthermore, the 2.86 and 5.71 g kg^{-1} doses of nutrasucre returned glycemia to below the initial value after 280 and 180 min, respectively. For comparison, hypoglycemia was obtained at 120 min in the glibenclamide group. Comparison of the glycemia of the animals of group 2, which received brown sugar at a dose of 5.71 g kg^{-1}, to those which received 5.71 g kg^{-1} nutrasucre highlights the antihyperglycemic properties of the elements mixed with brown sugar to make nutrasucre.

We also evaluated the antihyperglycemic potential of nutrasucre in alloxan-induced diabetic rats, a well-known experimental diabetes model[12]. Alloxan produces free radicals in the rats, which can cause necrosis of pancreatic β-islet cells, preventing insulin secretion and inducing a significant increase ($p<0.05$) in serum glucose concentrations[13]. This hyperglycemia is due to poor glucose utilization by tissues or an increase in neoglucogenenis[14].

Daily administration of nutrasucre to the diabetic rats for 28 days reduced their glycemia to a similar extent as glybenclamide, demonstrating the antihyperglycemic effect of nutrasucre, despite the presence of brown sugar. A similar effect has been shown in another study on the treatment of alloxan-induced diabetic rats with plant extracts, such as *Musanga cecropioides* and *Berberis aristata* or improved traditional medicines[15]. Nutrasucre contains several bioactive compounds including flavonoids, glycoproteins, terpenes and polysaccharides. The anti hyperglycemic activity of nutrasucre is likely due to flavonoids. These results are in accordance with those obtained in several studies showing that flavonoids from plants, such as *Eugenia jambolana*, *Cassia auriculata* L. and *Teucrium polium*, stimulated and promoted there generation of pancreatic β-islet cells[16-19].

Daily oral administration of nutrasucre for 28 days induced significant weight gain in the animals, comparable to that observed with glibenclamide. Many studies have shown weight loss in diabetic rats[1,20,21]. This effect is likely due to continuous lipolysis caused by the lack of insulin. In this study, lipolysis was corrected by the administration of nutrasucre.

Analysis of the lipid profiles of the diabetic rats showed that treatment with 5.71 g kg^{-1} nutrasucre substantially

reduced triglyceride, total cholesterol, HDL-cholesterol and LDH-cholesterol levels of the rats, whereas triglyceride levels increased by the end of treatment in rats treated with 1.43 and 2.86 g kg^{-1} nutrasucre. The lipidic profile of diabetic rats treated with the three doses of nutrasucre were not very different from those of the saline control and glibenclamide groups except for triglycerides, at doses 1.43 and 2.86 g kg^{-1}. Lipid assessment is of crucial importance in the treatment of cardiovascular diseases and the management of diabetic patients[22]. Cardiovascular complications associated with diabetes are believed to be due to disturbances in lipidic metabolism[23]. Thus, decreasing lipid levels in diabetes reduces the risk of cardiovascular complications[24,25]. The highest dose of nutrasucre (5.71 g kg^{-1}), which led to a significant improvement of the lipid profile in diabetic rats comparable to those treated by glibenclamide, could potentially prevent these cardiovascular complications in diabetics.

The increase in serum urea concentrations in diabetic rats treated with the three doses of nutrasucre may be due to protein degradation caused by proteolysis induced by the alloxan or food, which can be degraded into amino acids and then urea. Serum urea concentrations are generally high in diabetics, due to the activation of ureagenesis, a liver-specific function[26]. Urea is the ultimate product of free amino acid catabolism and serum free amino acid levels are high in diabetics[27]. In contrast, serum creatinine concentrations were close to those of the control groups that received distilled water or glibenclamide, showing that kidney function was good.

Transaminases are enzymes with significant intracellular metabolic activity. An increase in their activity reflects a cellular lesion, in particular hepatic, cardiac, renal, or muscular[28]. The reduction in ASAT and ALAT serum levels in animals treated with the three doses of nutrasucre suggest protection of the liver, heart and muscles. This result is supported by the results from the histological sections of these organs, which did not reveal any particular lesions in the liver, heart, or kidneys in this study. Nutrasucre may thus have a beneficial effect on these organs in diabetics.

The acute toxicity study, performed using the limit test, showed that the LD_{50} of nutrasucre is higher than 5000 mg kg^{-1}. According to OECD protocol 423[8], nutrasucre can be placed in the "non-classified category" of products considered to be non toxic in the Globally Harmonized System (HGS) of classification and labelling of chemicals.

No signs of subacute toxicity were recorded during 28 days of observation for the three doses of nutrasucre studied. No mortality was recorded during the experimentation. Moreover, the animals showed normal weight gain, a sign of the nontoxicity of nutrasucre.

Nutrasucre also had little effect on most of the biochemical parameters studied. The lipid profile did not vary greatly, suggesting that it would not have a harmful effect on the mobilization of free fatty acids.

Urea and creatinine values remained with in normal values and did deviate much from those of the control group receiving physiological saline, indicating no effect on renal filtration[29]. In addition, the transaminases ASAT and ALAT were also not altered during the study, showing that there was no liver or muscle damage during the 28 days of treatment with nutrasucre. The histological sections did not reveal any particular characteristics in the various organs. Overall, these results show that nutrasucre is not toxic to vital organs, such as liver and kidneys.

Can nutrasucre, a food additive consisting mostly of brown sugar and plant extracts, be taken by people with type 2 diabetes? It was formerly believed that sucrose (Table sugar) and other sugars contributed to the appearance of diabetes and that people who developed this disease had to avoid sugar. This arose from the idea that sugars were digested and absorbed more quickly than other carbohydrates and would thus worsen hyperglycemia. Sucrose and fructose induce a glycemic reaction that is less marked than for equal quantities of many starches. Sugars can be part of a healthy diet for people with type 1 or type 2 diabetes. The Canadian Association of Diabetes (CAD) does not recommend avoiding sugars. In its clinical practice guidelines for 2013 for the prevention and treatment of diabetes in Canada, the CAD states that sucrose can represent up to 10% of total daily caloric intake (i.e. 50-65 g day^{-1} for people who consume 2,000-2,600 kcal day^{-1}), because there is no reason to believe that this has a harmful effect on glycemic control or lipid assessment in people with type 1 or type 2 diabetes[30,31]. Sucrose consumption that represents more than 10% of total daily caloric intake can increase glycemia and triglyceride levels in some individuals[32,33]. This principle could apply to nutrasucre which could be indicated in the prevention and treatment of type 2 diabetes.

CONCLUSION

Nutrasucre is a food complement with antihyperglycemic properties which is non toxic as it does not affect vital organs such as the kidneys, liver and heart. Indeed, it may protect these organs. It reduces serum ASAT, ALAT and total, LDH and HDL levels and contains hypolipidic substances. Nutrasucre can thus bean important element in a program for the

prevention and treatment of diabetes. It will be necessary to reduce the ratio of sugar to plant extracts to better reinforce the beneficial activity of this food complement.

ACKNOWLEDGMENT

Authors would like to thankful to all other members of Laboratory of Toxicology and Laboratory of Anatomical Pathology (University Felix Houphouët Boigny), for their encouragement, direct technical assistance as well as indirect assistance during these investigations.

SIGNIFICANCE STATEMENTS

This study discovers the possible antidiabetic activity of the nutrasucre, a food complement, in alloxan induced diabetic rats that can be beneficial for diabetics. This study will help the researcher to uncover the critical areas of the treatment of type 2 diabetes by consuming nutrasucre that many researchers were not able to explore. Thus a new theory on this food complement and its possibility to heal type 2 diabete may be arrived at new natural antidiabetic drugs for the population.

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