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Short Communication

Biochemical observations in wistar rats fed with the histological dye extracted from *Sorghum bicolor*

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The biochemical effects of the histological dye extracted from *Sorghum bicolor* on Wistar rats were studied. The rats were fed with different doses of the aqueous extracts, 10, 100, 1000, 1600, 2900 and 5000 mg/kg for 24 h and then sacrificed through cervical dislocation. Thereafter, the sera were analyzed for random blood sugar, AST, ALT, sodium, potassium, chloride, bicarbonate, urea and bilirubin, and compared with the control rats. The serum values of random blood sugar, AST, sodium, potassium, bicarbonate and urea increased progressively with increase in concentration of the extract administered to the rats. The value of chloride also increased but it was within the reference range. The bilirubin levels did not change remaining at <17 mmol/l, except in rats that died during the period of administration of the dye where the bilirubin level reached between the range of 17 - 21 mmol/l. However, serum ALT decreased with increase in concentration of the extract administered to the rats.

Key words: *Sorghum bicolor*, ALT, AST, bilirubin, random blood sugar.

INRODUCTION

Sorghums are widely distributed throughout the tropics, subtropics, and warm temperate areas of the world. It is known under a variety of names such as Kaffir corn in South Africa, guinea corn in West Africa, Karan dafi in Northern Nigeria, and Oka pupa in the South Western part of Nigeria (Dalziel, 1948). It is an annual or short - term perennial plant coarse, erect, with much variability in growth characteristics; culm solid or sometimes with spaces in pith, 0.6 - 5 m tall, depending on variety and growing conditions, 5 to over 30 mm in diameter. Their leaves are broad and coarse, similar in shape to those of corn but shorter and wider. Though, sorghum is used largely for forage in the US, it is very important in the world's human diet, with over 300 million people dependent on it (Bukantis, 1980). The seeds are used as food in brewing local beer, the corn malt and cornmeal is fermented to make a sour mash, the pith is eaten and sweet culms chewed (Watt and Brayer-Brandwijk, 1962). Sorghum has been reported to be an anti-abortive, demulcent, diuretic and intoxicant. In India, the plant is considered anti-helminthic and insecticide, and in South Africa, in combination with Erigeron canadense it is used for eczema (Watt and Brayer-Brandwijk, 1962). The natives of Curacao drink the leaf decoction for measles, grinding the seeds with those of the calabash tree for lung ailments (Morton (1981). A study conducted on albino rats supported the traditional use of *Sorghum bicolor* as a remedy for anaemia (Ogwumike, 2002). Crude ethanolic extract of *Sorghum bicolor* has been used as a stain for collagen and muscle fibers (Avwioro et al., 2006). In their study, the alkaline mixtures did not stain any tissue used but the acidic and natural alcoholic mixtures stained collagen fibers, muscles and red blood cells in shades of pinkish-yellow. Elution of the stain from stained sections confirmed the dye to be apigenindin.

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Table 1. Biochemical analysis.

Group	R.B.S mmoL/L	Urea mmol/L	Na^+ mmoL/L	K^+ mmol/L	Cl^- mmoL/L	HCO_3 mmoL/L	AST 1µ/L	ALT 1µ/L	Total bilirubin mmol/L
Control	2.63	2.66	110.2	3.03	100.4	21.2	10.5	17.5	<17
10 mg	2.82	2.68	116.3	3.21	100.4	22.3	10.5	17.5	<17
100 mg	3.92	3.37	120.2	3.21	101.3	22.4	12.4	16.5	<17
1000 mg	3.94	3.45	121.1	3.33	101.3	23.4	14.3	20.5	<17
1,600 mg	5.30	4.75	129.3	3.72	103.2	23.5	16.5	12.5	<17
2,900 mg	7.84	6.35	135.3	4.31	104.4	27.2	17.3	11.5	<22
5000 mg	10.7	7.00	141.2	5.20	104.5	32.3	18.5	10.5	<22

Key:
R.B.S. -Random blood sugar
Na^+ -Sodium
K^+ -Potassium
Cl^- -Chloride
HCO_3 -Bicarbonate
AST -Aspartate aminotransferase
ALT -Alanine aminotransferase

Sorghum contains hydrocyanic acid and the alkaloid hordenine. Sometimes plants accumulate toxic levels of nitrate (Morton, 1981). HCN is destroyed when fodder is ensile or cured as hay. In Africa, the dye extracted from *Sorghum bicolor* is used in coloring leather, clothes, and calabashes and as a body pigment (Cobley and Steele, 1976). Studies have shown that the dye extracted from *Sorghum bicolor* to be a flavonoid described as apigenindin. Paracelsus introduced the modern idea that: "All substances are poisonous, there is none which is not a poison. The right differentiates a poison from a remedy". He defined toxicity as the capacity to cause injury. In order for a substance to cause injury, he stated that it must be present at its site of action at an effective dose. All substances at low enough levels are not toxic. Most substances at high enough levels are toxic. The aim of this work was to determine the biochemical changes in Wistar rats fed with the histological dye extracted from *Sorghum bicolor.*(Table 1*)*

MATERIALS AND METHODS

Male and female albino rats weighing between 190 and 230 g were obtained from the animal house of the Department of Biological Science, Ahmadu Bello University, Zaria, Nigeria. They were kept in rat cages with free access to water and dry rat pellet feeds. The leaf sheaths of *Sorghum bicolor* plant were collected by cutting with knife. Small bundles of the leaf sheaths of the sorghum plant were collected in the month of March 2007. They were taken immediately to the herbarium of the Department of Biological Sciences, Ahmadu Bello University, Zaria, Nigeria. The leaves were washed in water, air dried at room temperature (35 - 38°C) for 21 days, after which they were milled with the use of ceramic mortar and pestles. They were sieved with a sieve of diameter 0.6mm to remove coarse and hard materials, leaving a fine smooth powder. The fine smooth powder material was kept in a cool dry place prior to extraction. 100 g of the powdered plant material was measured into a conical flask and completely soaked in 500 ml of 70% ethanol with intermittent agitation for 24 h. The mixture was filtered with a no. One Whatmann filter paper. 100 ml of 70% ethanol was added to the sediment, mixed and then filtered again. The process was repeated until filtrate was near colourless. The filtrate was distilled under reduced pressure by a vacuum pump and then concentrated by evaporation with the aid of a rectangular water bath to produce a semi - solid concentrate. It was then evaporated to dryness at 75°C (Avwioro et al., 2006). The weight of the dry extract was 10.66 g. A stock concentration was prepared in which 1 g of the dry extract was dissolved in 20 ml of normal saline and then administered to the rats accordingly. Three animals served as the control as these received 0.5 ml of normal saline. They were all placed under investigation for 24 h after which the observed result was recorded. Final investigation was carried out with six groups; each consisting of four rats (2 males and 2 females) and specific doses of 10, 100, 1000, 1600, 2900 and 5000 mg/kg body weight respectively were administered. The rats were sacrificed by cervical dislocation after 24 h of administration of the *S. bicolor* extract and blood collected from the heart into oxalate and lithium heparin bottles. Chloride was determined by the Rose (1936) method. Sodium, potassium and bicarbonate were determined according to the methods described by Korzum and Miller (1996) and Segal (1955) respectively. Urea determination was by the Kazmierczack (1996) method. Blood glucose was determined by the method of Gochman and Schmitz (1972). Aspartate aminotransferase, alanine aminotransferase (Tekum and Timothy, 1988) and bilirubin (Cheesbrough, 2005) were also determined.

RESULTS

Average values were taken in each group. Animal death was observed at levels above 1600mg probably as a result of congestion. The values of random blood sugar (RBS), urea, sodium (Na^+), potassium (K^+) and aspartate aminotransferase (AST) increased progressively when the quantity of *S. bicolor* extract administered increased but were within the normal range except for values above 1600 mg/kg where the animals died. Bicarbonate (HCO^3), chloride (Cl^-), bilirubin and alanine aminotransferase (ALT) levels also remained normal when compared with the control.

DISCUSSION

Sorghum bicolor has been used extensively as forage (Bukantis, 1980), food (Watt and Brayer-Brandwijk, 1962) and medicines for several ailments (Watt and Brayer-Brandwijk, 1962; Morton, 1981). It has also been used as a dye for clothes, leather and calabashes (Cobley and Steele, 1976) and as a histological dye for collagen fibres, muscles and red blood cells (Avwioro et al., 2006). Since no serious medical complaints have been reported in the use of *S. bicolor*, it is assumed that its traditional use is harmless, but it is often necessary for scientists to determine the toxicity of dyes before their use can generally be accepted, because certain dyes were found harmful to human tissues and their use had to be discontinued (Sewekow, 1988). All substances are toxic, and what differentiates a poison from remedy is the dose that is capable of causing injury to human health. Dyes are coloured, ionizing, aromatic organic compounds. They are chemicals, and like all chemicals, they are similar in their reactions to other chemicals, and distinctly different from others. This includes the possibility that they are toxic. They may be carcinogenic or mutagenic, or harmful to health in some other ways (Avwioro et al., 2006). The biochemical parameters determined in this experiment were substantially normal when administered in low doses, although in some cases, there were progressive increases relative to the doses of extract given. The bilirubin level was normal (<17), an indication that there was no red blood cell lysis. The alanine aminotransferase (ALT) level was also normal, an indication that the liver cells and striated muscles were not damaged (Bain, 2003), even in high doses of the extract. High levels of carbohydrates, chloride, potassium, sodium, protein and other minerals have been reported in *S. bicolor* (Miller 1958; Wu Leung et al., 1972). This probably accounts for the increase in sugar, potassium, sodium, chloride and bicarbonate ions in the sera of the animals, which increased when the extract administered to the rats increased. However, there was a slight increase in the value of the cardiac enzyme-aspartate aminotransferase (AST) in high doses of the extract as a result of congestion of the heart arising from the high doses administered to the rats.

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