

Short Communication

The effect of aqueous leaf extract of fluted pumpkin on some hematological parameters and liver enzymes in 2,4-dinitrophenylhydrazine- induced anemic rats

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Anemia constitutes a serious health problem in many tropical countries including Nigeria because of the prevalence of malaria and other parasitic infections which possibly leads to decrease of hemoglobin. Fluted pumpkin has been reported to be very good in building the constituents of the blood and also replacing them. This study was designed to investigate the effects of the aqueous leaf extract of fluted pumpkin on some hematological parameters and liver enzymes; alanine aminotransferase (ALT) and aspartate aminotransferase (AST) in 2, 4-dinitrophenylhydrazine-induced anemia in experimental rat. Twelve Wister albino male rats were obtained from a nearby farm and separated into four groups of three rats each for the study. Rats in Groups 1 and 2 were injected 40 mg/kg 2, 4-dinitrophenylhydrazine for eight days to induce anemia, while rats in Groups 3 and 4 were fed with grower's mash and water *ad libitum*. All analysis was done using the standard methods. The result of this study shows that, oral administration of 50 mg/kg of aqueous leaf extract of fluted pumpkin to the rats in induced treated and normal treated groups (Groups 1 and 3), increased the hematological parameters under investigation while the rest remained significantly unchanged. Low level of ALT and AST was observed in rats in group 1 (induced treated group) suggesting a hepatoprotective property of the leaf extract which also indicate that the extract had no effect on the liver of the rats at the concentration used. The result of this research indicate that 50 mg/kg aqueous leaf extract of fluted pumpkin could elevate the packed cell volume, red blood cells and hemoglobin concentration in the rats induced with 2, 4-dinitrophenylhydrazine. Hence, oral administration of the extract could cure hemolytic anemia. The leaf extract also regulated the liver enzymes (ALT and AST) of the rats induced with 2, 4-dinitrophenylhydrazine. It can be concluded that aqueous leaf extract of fluted pumpkin is a potential blood booster and has hepatoprotective property.

Key words: Fluted pumpkin, hematological parameters, liver enzymes, 2, dinitrophenylhydrazine, anemia.

INTRODUCTION

Pumpkin as defined by the English dictionary is a large orange fruit or a round large fruit with a thick orange skinned rind, dry flesh and many seeds, cooked and eaten as a vegetable or in sweet dishes. It is known in

many different countries because of its value as most of the parts can be eaten and are rich in nutrients. It is easily grown and one plant in the garden can supply pumpkins and green leaves throughout the year. It grows

best in loose, rich soil, mainly in any old rubbish heap. There are many different varieties of pumpkin whose botanical name is 'Telfairia' which belong to the tribe, Joliffieae (Akoroda, 1990).

Fluted pumpkin is a dicotyledonous vegetable that develops long vine-like stems with trifoliolate leaves and edible large fleshy fruits which can be 5 cm high. The leaves are simple dark green if properly grown on a suitable soil. It thrives in well-drained soils and is usually cultivated in garden and family farms around homes. The dark green leaves can be 18 cm wide and 35 cm long (Iweala and Onyechi, 2009), some of its native names include 'Ugu' as known by Ibos, 'Umee' in Efik and "Umeke" in Edo (Akoroda, 1990).

It is a common tropical green leafy vegetable native to many African countries especially Eastern Nigeria (Burkett, 1968). In ethno medicine, the fresh leaves are used in treatment of malaria and convulsion (Alada, 2000; Gbile, 1986). There are various methods by which fluted pumpkin leaves can be prepared as meal and drinks. It has been observed by the users to be a vegetable that is outstanding in health nourishing. It is used in cooking different variety of food, preparing salad, stew and soup. Some local users reported that it can be used as local or native blood tonic while some suggested that it can be used as a cure for anemia precisely anemia due to blood loss (personal communication).

Anemia constitutes a serious health problem in many tropical countries including Nigeria because of the prevalence of malaria and other parasitic infections which possibly leads to decrease of hemoglobin. Over millions of people have anemia in the sense that any disease that can lead to blood shortage or loss is mostly coupled with anemia. The aim of this study, therefore, is to evaluate the effect of aqueous leaf extract of fluted pumpkin on some hematological parameters and liver enzymes in 2, 4-dinitrophenylhydrazine induced anemic rats and the effect of aqueous at unveiling the possibility of curing anemia through the use of vegetable, hence the choice of fluted pumpkin.

MATERIALS AND METHODS

Sample collection and preparation

Fresh leaves of fluted pumpkin was obtained from a nearby farm in Mubi, Adamawa State, washed and weighed on daily basis for each of the experiment.

Extraction technique

About 50 g of fluted pumpkin leaves was chopped into smaller bits and ground with mortar and pestle. 100 ml of distilled water was

poured into the ground leaves to obtain the extract which is 50 mg/ml. The aqueous extract was obtained raw using a sieve to avoid the entrance of particles. 50 mg/kg of the extract was administered to the rats according to the weight through oral gavage.

Experimental design

Twelve albino male rats weighing between 150 to 220 g were obtained from local breeders within Adamawa State University, Mubi. The rats were kept in the laboratory for one week for acclimatization before the on-set of the experiment. The rats were distributed into four groups of three rats each and were treated as follows:

Group 1 (Induced treated)

The rats in this group were labeled R₁, R₂ and R₃ and were also made anemic by daily oral administration of 40 mg/kg of 2,4-dinitro phenyl hydrazine for eight days. The rats were also fed with grower's mash and water *ad libitum*. They were treated with 50 mg/kg of the freshly prepared leaf extract for five days after being confirmed anemic.

Group 2 (Induced control)

The rats in this group; R₄, R₅ and R₆, were made anemic by daily oral administration of 2,4-dinitro phenyl hydrazine (PHZ) at 40 mg/kg for eight days during which they were fed with grower's mash and water *ad libitum* with no other treatment.

Group 3 (Control treated)

The rats in this group; R₇, R₈ and R₉ were fed normally with grower's mash and water and were given 50 mg/kg of the freshly prepared leaf extract each day for complete five days.

Group 4 (Normal control)

The rats in this group; R₁₀, R₁₁ and R₁₂ were fed with growers mash and water *ad libitum* throughout the period of experiment, with no other treatment.

Analysis of hematological parameters

Packed cell volume (PCV), white blood cell (WBC) and red blood cell (RBC) count

The packed cell volume, white blood cell count and red blood cell count were obtained by the method described by Hoffbrand and Moss (2011).

Hemoglobin concentration determination

The hemoglobin concentration was determined using acid haematin method also known as Sahli's method (Hoffbrand and Moses, 2011).

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Table 1. Effect of the aqueous leaf extract on some hematological parameters

Parameter	Group 1 (induced treated)	Group 2 (induced control)	Group3 (normal treated)	Group 4 (normal control)
PCV ₁ (%)	45.33±1.77	48.00±0.58	46.33±0.88	44.00±0.06
PCV ₂ (%)	38.00±0.18	39.00±0.58	49.00±1.55	48.67±3.72
PCV ₃ (%)	41.67±±.88	39.67±0.88	51.67±0.88	49.33±3.39
WBC ₁ (10 ³ /mm ³)	1.92±0.05	2.08±0.05	2.00±0.03	1.90±0.06
WBC ₂ (10 ³ /mm ³)	2.72±0.05	2.48±0.12	2.12±0.02	2.02±0.08
WBC ₃ (10 ³ /mm ³)	2.27±0.12	2.35±0.03	1.62±0.05	1.88±0.05
RBC ₁ (10 ⁷ /mm ³)	2.31±0.02	2.47±0.13	2.37±0.02	2.36±0.06
RBC ₂ (10 ⁷ /mm ³)	2.18±0.02	2.27±0.06	2.72±0.02	2.69±0.09
RBC ₃ (10 ⁷ /mm ³)	3.31±0.03	3.00±0.08	3.79±0.02	3.01±0.19
Hb ₁ (g/dl)	9.67±0.18	10.00±0.12	10.10±0.16	9.87±0.37
Hb ₂ (g/dl)	8.20±0.12	8.50±0.12	10.47±0.18	10.40±0.35
Hb ₃ (g/dl)	12.37±0.13	10.07±0.07	12.97±0.04	11.47±0.37

All values are in Mean ± SEM for n=3. PCV₁, PCV₂ and PCV₃ are the packed cell volume of the rats before inducing anemia, after inducing anemia and after treatment, respectively; WBC₁, WBC₂ and WBC₃ are white blood cell count of the rats before inducing anemia, after inducing anemia and after treatment, respectively; RBC₁, RBC₂ and RBC₃ are red blood cell count of the rats before inducing anemia, after inducing anemia and after treatment, respectively; Hb₁, Hb₂ and Hb₃ are hemoglobin concentration of the rats before inducing anemia, after inducing anemia and after treatment, respectively.

Table 2. Effect of aqueous leaf extract of fluted pumpkin on the liver enzymes

Parameter	Group 1 (induced+extract)	Group 2 (induced control)	Group 3 (control treated)	Group 4 (normal control)
ALT(iu/l)	7.00±0.29	11.17±0.73	8.50±0.29	9.00±1.00
AST(iu/l)	22.17±0.44	28.33±0.88	47.33±0.34	30.00±1.48

All values are in Mean ± SEM for n=3; ALT stands for Alanine transaminase; AST stands for Aspartate transaminase.

Enzyme assays

The enzymes that were tested are Alanine aminotransferase (ALT) and Aspartate aminotransferase (AST). The test was done by the method described by Reitman and Frankel (1957) using Randox test kits.

Statistical analysis

Results were expressed as mean ± standard error of mean (SEM) for triplicate determination.

RESULTS

The results of the hematological parameters are presented in Table 1. The result shows that there was an increase in the hematological parameters of the rats as shown in Table 1. Daily intra peritoneal injection of 40 mg/kg 2, 4-dinitrophenyl hydrazine for eight days caused a decrease in the hematological parameters of the rats in group 1 and 2, while the packed cell volume dropped from 45.33 ± 1.77 and 48.00±0.58 to 38.00±0.18 and 39.00±0.58,

respectively. There was also a drop in the red blood cell count and hemoglobin concentration, whereas the white blood cell count increased.

The hematological parameters were raised after the oral administration of the aqueous leaf extract. The packed cell volume, red blood cell count and hemoglobin concentration was 38.00±0.18, 2.18±0.02 and 8.20±0.12 respectively, with those of the induced and treated rats raised to 41.67±0.88, 3.31±0.03 and 12.37±0.13 respectively, while that of the induced control group remained low.

Table 2 shows the result of the effect of the extract on the liver enzymes; ALT and AST. In the groups treated with the extract, low level of ALT was observed. The average result was 7.00±0.29 iu/l and 8.50±0.29 iu/l for groups 1 and 3, respectively. Low level of AST was observed in the rats in group 1(22.17±0.44) and high level in the rats in group 3 (47.33±0.34).

DISCUSSION

As shown in Table 1, the decrease in the hematological

parameters in Groups 1 and 2 could be as a result of breakdown of the red blood cells caused by the 2,4-dinitrophenylhydrazine. The increase in white blood cells could be due the defense mechanism against the entrance of a foreign material in the body system of the rats (2, 4-dinitrophenylhydrazine).

This study has shown that the aqueous leaf extract of fluted pumpkin caused an increase in packed cell volume, white blood cell count, red blood cell count and hemoglobin concentration in rats. The increase in hematological parameters investigated could be as a result of some constituents such as iron and some B complex vitamins which it possess as these serves as hematopoietic factors that influence directly on blood production in the bone marrow (Ganong, 2005). This study also agreed with the work of Salman et al. (2008), who reported that there was a significant increase in the hematological parameters of rats that were treated for two weeks with the aqueous leaf extract of fluted pumpkin. Some scientists have proposed the use of fluted pumpkin in treatment of anemia, following studies which reported that extracts of fluted pumpkin helps to maintain blood level in subjects given its extracts (Fiona and Latunde - Dada, 2011).

This research also shows that the leaf extract can serve as a cure for anemia as reported by some researchers. The increase in weight of the rats could be as a result of rich nutrients such as amino acids, fatty acids, mineral and vitamins (Fagbemi, 2007). Specified and suitable concentration of the aqueous extract leads to positive effects on the hematological parameters investigated.

As shown in Table 2, the lower level of ALT and AST in the rats induced with 2, 4-dinitrophenylhydrazine could be as a result of the hepatoprotective property of the plant on rats as reported (Oboh, 2005). AST of the normal rats was elevated than normal after treatment which showed that the concentration was high as regards to the health status of the rats, therefore, to some extent could be toxic to the rats. This effect attests to the observation of irregularity in the liver after a long term consumption of *Telfairia occidentalis* supplemented diet in rats (Iweala and Obidoa, 2009).

Conclusion

The result of this research indicated that 50 mg/kg aqueous leaf extract of fluted pumpkin could elevate the packed cell volume, red blood cells and hemoglobin concentration in the rats induced with 2, 4-dinitrophenylhydrazine. Hence, oral administration of the extract could cure hemolytic anemia. The leaf extract also regulated the liver enzymes (ALT and AST) of the rats induced with 2, 4-dinitrophenylhydrazine.

Conflict of interests

The authors did not declare any conflict of interest.

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