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Platelet response to the ethanolic extract of *Pennisetum subangustum* in the albino Wistar rat

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Abstract

Pennisetum subangustum, locally known as "fuur" in some populations in the Jos Plateau is used to stop bleeding in the event of an injury. The leaves of the plant were marshed and soxhlet extracted with ethanol for 72 hours. The extract of *P. subangustum* was then administered to albino Wistar rats. Platelet count, bleeding time and clotting time, were determined before and after the administration of the extract. Results show that the platelet count was reduced; bleeding time and clotting time prolonged. A precise mechanism of the haemostatic role of *P. subangustum* is yet to be established, and this is planned for in the next phase of the investigations. Any beneficial role ascribed to it for arresting bleeding is not supported by this study.

Keywords: *Pennisetum subangustum*; Haemostasis; Platelet; Bleeding.

Introduction

Pennisetum subangustum is a hairy, soft, erect annual plant. It is a common weed in dry land rice ecosystem in the tropical region. On the Jos Plateau area of Nigeria, some local population use the plant (locally called 'Fuur') to stop bleeding after a cut sustained from farm implements. So strong is the belief of the people in this plant that its use is preferred to orthodox medical intervention, following a trauma.

The Word Health Organization (WHO) consultative group defined a medicinal plant as one which in one or more of its organs contain substances that can be used for therapeutic purposes, or which are precursors of useful drugs (Sofowora 1982). The problem with the medicinal claims for some plants has been the insufficient scientific data on the plants (Akpata 1979; Briejer *et al.*, 2001). *Pennisetum subangustum* belongs to this inadequately studied group.

The investigations of physiological and pharmacological processes depend, to a large extent on the use of laboratory animals (Basketter 1994). Apart from determining the physiological mechanisms in the animal, the use of animals serves as a pool from which human processes could be extrapolated. Coagulation studies are of great importance considering the role of blood in life (Aballi and de-Lamerans, 1962). Platelets are the blood cells involved in coagulation (Jackson and Nemerson, 1980, Williams and Levine,

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1982). In our view, if *Pennisetum subangustum* can effectively achieve coagulation as claimed, then it could have some effect on platelet population and or functions (Burstein *et al.*, 1981).

The aim of the study therefore, is to assess the influence of *P. subangustum* on platelet population and functions.

Experimental

The rats used in this study were purchased from the animal house, University of Jos. Twenty (20) male albino Wistar rats weighing between 130g and180g, divided into two groups of ten (10) each were used. One group was control while the other, test. The plant, *Pennisetum subangustum*, was collected from the University of Jos environment, and was identified by a plant taxonomist, Mr. I. A. Kareem of the Federal College of Forestry, Jos.

60g of the fresh leaves of *P. subangustum* was mashed using a mortar, and Soxhlet extracted in 250ml of absolute ethanol for 72 hours. The ethanol solvent was evaporated using hot plate, leaving a paste weighing 4.10g. The extract was kept in a refrigerator until when required. A stock concentration of 100mg/ml of the extract was prepared; from the stock a dose of 1mg/kg body weight was obtained by serially diluting down and used subsequently for this work.

Platelet studies. The platelet count, bleeding time, and clotting time was determined using the method of Dacie and Lewis (1984). For platelet count, 0.19ml of Boar's fluid was taken in a test tube. Using a rat restrainer, blood was taken from each rat by tail – end amputation and sucked up to the 0.01m mark of white blood cell pipette. This was introduced into the Boar's fluid and mixed thoroughly. The mixture was gently dropped onto the counting chamber, covered with a cover slip, and using 'x 40' objective lens, platelets were counted. For determination of bleeding time, the base of each rat tail was cleansed, and pricked with a sterile lancet. A stop clock was started immediately. Blood was blotted every 15seconds using Whatman filter paper until bleeding ceased. Time taken for the blood to stop is the bleeding time. Clotting time was determined by a cut on the distal part of each rat tail using a sterile scissors. The blood was placed on a grease-free glass slide. A stop clock was started immediately. A needle was passed through the blood on the glass slide every 15 seconds until a thread – like structure was seen. The time taken to form the thread – like structure is taken as the clotting time.

Results and Discussion

The bleeding time (BT) before the administration of the extract were 2 minutes 15 seconds in the control and 2minutes 75seconds in the test group respectively. After the administration of the extract, BT in the test group was 3 minutes 25seconds. The control was 2minutes 45seconds. These are shown in Table 1.

Results of the clotting time (CT) are shown in Table 2. Baseline readings were 1minute 35seconds, and 1minute 50seconds in the control and the test groups respectively. After the administration of the extract, the readings in the corresponding groups were 1minute 30seconds and 1minute 45seconds respectively.

The platelet counts (PC) are shown in Table 3. The baseline PC in the control and test groups are 30,000/mm^3 and 18,000/mm^3 respectively. After the extract the PC are 36,000/mm^3 and 26,000/mm^3 respectively.

Table 1. Mean bleeding time in rat following the administration of *Pennisetum subangustum*.

Group	Baseline reading	Second reading	Average
Control	2min 15sec	2min 45sec	2min 3sec
Test	2min 75sec	3min 25sec	3min 0sec

Table 2. Mean clotting time in rat following the administration of *Pennisetum subangustum*.

Group	Baseline reading	Second reading	Average
Control	1min 35sec	1min 30sec	1min 33sec
Test	1min 50sec	1min 45sec	1min 75sec

Table 3. Mean platelet count in rat following the administration of *Pennisetum subangustum*

Group	Baseline count	Second count	Average
Control	30, 000 / mm^3	36, 000 / mm^3	33, 000 / mm^3
Test	18, 000 / mm^3	26, 000 / mm^3	22, 000 / mm^3

P. subangustum has not been much reported on. The platelet count is reduced in rats treated with *P. subangustum* when compared with control ($P<0.05$). The bleeding time and clotting time are prolonged. Blood coagulation requires that platelet be in sufficient size, number and function. In experimental animals, it has been reported that the bone marrow has a narrow reserve of platelets (Burstein *et al.*, 1981). Prolonged bleeding time and clotting time, in the presence of a reduced platelet count is implicative of a general platelet insufficiency to form haemostatic plugs. *P. subangustum* might be interfering with the activation of serine proteases, synthesis of thromboxane A2 or be a heparin-dependent inhibitor of thrombin. These are necessary steps in haemostasis (Jackson and Nemerson, 1980, Tollefson and Blank, 1981).

Pennisetum subangustum, contrary to claims of it being able to arrest bleeding, impairs haemostasis. "Fuur", as it is known in the local population on the plateau should be discouraged from being used as a procoagulant. We intend to analyze the chemical constitution of *P. subangustum*, and to be able to define it, precise haemostatic role in the next stage of our study.

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