The Effects Of Neem (Azadirachta Indica) Leaves Extracts, On Some Haematological Indices Of Wistar Rats

*Ndodo N.D 1, Anuka J.A2, Esomonu U.G3, Onu J.E4, Okolo R.U 1 And Onwuchekwa C5.

Department of Human Anatomy1, Physiology5 College of Health Sciences, Usman Danfodiyo University, Sokoto, Nigeria.

Department of Clinical Pharmacology2, Faculty of Pharmaceutical Sciences, Ahmadu Bello University, Zaria, Nigeria.

Department of Human Anatomy3, Faculty of Medicine, Bayero University, Kano.

Department of Veterinary Anatomy4, Usman Danfodiyo University, Sokoto, Nigeria.

ABSTRACT : The effects of water and methanol extracts of Neem (Azadirachta Indica) leaves were studied in male Wister rats following treatment with 20 % w/w and 30% w/w equivalent of water and methanol extracts of Neem leaves incorporated into rat diet and administered orally for 90 days. The control group had normal diet and water ad libitum. The result showed that PCV of the control group was significantly lower than that of the treated groups at p<0.001. 30% w/w equivalent of neem leaves (C2) significantly increased the PCV (p<0.001) of treated rats. This is consistent with the little or no toxicity observed in Neem extracts. There was no significant variation between the mean TLC of the control and the neem treated groups at P > 0.05. The findings suggest relative safety of the Neem extracts.

KEY WORDS: Neem leaves, PCV, TLC, Haematological indices

I. INTRODUCTION

Azadirachta indica A. Just popularly called Neem is a member of the mahogany family, called Meliaceae. Neem trees are broad-leaved evergreens that grow up to 30m tall and 2.5m in girths. Azadirachta indica is native to India and Burma. Brigadier- General Sir Fredrick Guggisberg introduced neem to West Africa. (National Research Council, 1992). Neem was introduced to Nigeria in 1928 in former Bornu Province and later to Sokoto, Katsina, Kano in 1930 for afforestation purposes. The immunomodulatory activities of neem have been reported (Labadie et al, 1989). The antimicrobial, anti-inflammatory and antipyretic properties have also received attention (Okpanyi and Ezeukwu, 1981). Lai et al, (1986) observed the anti-fertility effects of the neem oil. Riar et al (1990) recorded the potential spermicidal effect, while Upadhayay et al. (1990, 1992), highlighted the immuno-contraceptive properties of neem. Neem oil acted as a spermicidal agent and inhibited sperm motility (Riar et al, 1990; Sharma et al, 1996) Neem-oil has been shown to have anti-fertility activity and to stimulate cell mediated immune response. Immunocontraceptive properties, anti-implantational as well as abortificient effects of neem are well documented (Upadhay et al, 1992, Riar et al 1984, Sinha et al, 1984a, b).

A fraction of Neem oil called Nim-76 was said to evoke spermicidal activity which makes it suitable for use as pre-coital antifertility formulation for human use, which has undergone phase one clinical trials (Riar et al, 1991, Sai Ram , et al, 2000). Mukherjee and Talwar, (1996) reported that the neem seed extracts, could completely abrogate pregnancy in rodents at an early post implantation stage. The treatment did not have any residual effects on future fertility of the animals. The effects of the extracts on the implantation were due to a transient increase in cytokines Y-interferon and TNF (Tumour Necrosis Factor) after the treatment. This current study evaluated the effects of the Water and methanol extracts of neem leaves on the haematological indices of wistar rats.
II. MATERIALS AND METHODS

Extraction of Neem Leaves

Water Extraction

1000g of powdered Neem leaves were dissolved in distilled water in a soxhlet apparatus; the liquid extract was further concentrated in a rotary evaporator to yield a solid aqueous extract weighing 30g.

Methanol Extraction

Using Soxhlet extractor, 1000g of powdered Neem leaves were extracted in methanol to yield 20g of dry, concentrated extract with the aid of rotary evaporator.

Animal Treatment Procedure.

Twenty-five male Wistar rats of proven fertility weighing between 200g-250g, were used. They were housed in a Perspex cage with stainless steel-mesh tops, kept in the animal house of the Human Anatomy department, ABU. The rats had free access to food and water and were maintained in standard environmental conditions. The rats were fed growers mash and provided with water ad libitum. The rats were divided into 5 groups, A, B₁, B₂, C₁ and C₂. Group A is the control group while groups B to C were treatment groups.

Group A: this is the control group consisting of 5 male rats. This group received normal diet and water. Group B: this group was treated orally with Water extract of Neem leaves. B₁ received 20% w/w equivalent of water extract of Neem leaves mixed with the feed; for 12 weeks while B₂ received 30% w/w equivalent of the aqueous extract mixed with the feed for 12 weeks. Group C: This group was treated orally with methanol extract of Neem leaves. C₁ group received 20% w/w equivalent of methanol extract of Neem leaves mixed feed for 12 weeks. C₂ group: received 30% w/w equivalent of methanol extract of Neem leaves mixed with feed for 12 weeks. Each group consisted of 5 wistar rats.

Estimation of Packed Cell Volume (PCV) and White blood Cell (WBC).

PCV was determined by the microhaematocrit method. In this method blood samples collected by means of tail nipping were collected in heparinized capillary tubes and sealed off by rotating the blood- free ends in a flame avoiding charring the blood or bending the tubes. The sealed tubes were then spun in a centrifuge for about 15 minutes at 3,000 r.p.m. The haematocrit then read off directly using the microhaematocrit reader provided. WBC counts were estimated using the new improved Neubauer counter haemocytometer as described by Ganong (1995).

Statistical analysis

All data were subjected to one-way analysis of variance. Treatment means were separated using Tukey-Kramer’s Multiple Comparison Test, where significant differences were detected.

III. RESULTS

Haematological Parameters

Packed Cell Volume (PCV)

Figure 1 shows the effect of water and Methanol extracts of Neem leaves on PCV of Wistar rats. PCV of the control group was significantly lower than that of the treated groups at p<0.001, but with that of C2 at p<0.05.
Fig. 1. Shows the effect of neem leaves extracts on PCV of wister. One error bar shows SEM.

**Total Leucocytes Count (TLC)**

Figure 2 shows that there was no significant variation between the mean TLC of the control and the neem treated groups at $P > 0.05$.

Figure 2. Shows the effect of the water and methanol extracts of Neem leaves on TLC of wister rats, following oral administration for 90 days.
IV. DISCUSSION

This study was designed to ascertain whether water and methanol extracts of Neem leaves had effects on the haematological indices of male Wistar rats. In view of the abundance of Neem in the northern region of Nigeria, and the near massive dependence of the local populace on this same plant, it is then expedient that the role of the Nigerian breed of the Neem be subjected to evaluation.

30% w/w equivalent of neem leaves (C2) significantly increased the PCV (p<0.001) of rats. This finding underscores the little or no toxicity observed in Neem extracts. This result is in variance with Abubakar (1997) who observed decrease in PCV with increase in dose of Neem leaves extract. However,

TLC was not affected. This study revealed that in all groups, that there was continuous weekly body weight gain. The mean weekly body weight increased all through the study except for 9th and 11th week where the control outweighed the treated groups at P<0.05 (data not shown). Nutritional status did not show any change as revealed by the haemoglobin content to some extent. This suggests that the extracts might not have had a negative effect on the rat’s food consumption as well as their nutrient utilization. However, the decrease in the 9th and 11th week may suggest that prolonged administration of the extract could affect food consumption. The above finding is in agreement with the results of Katsuri et al, (1997).

V. CONCLUSION

The finding suggests relative safety of the Neem extracts. It also underscored the huge dependence of the populace on neem for many years.

REFERENCE
