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Effects of Aqueous Extract of *Triplochiton scleroxylon* on White Blood Cell Differentials in Alloxan-Induced Diabetic Rabbits

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Abstract: Studies were conducted on the effects of aqueous extract of *Triplochiton scleroxylon* on white blood cell differentials in alloxan-induced diabetic rabbits. At least 100 mL of the aqueous extract was administered orally to test rabbits of New Zealand strain weighing between 1200 to 1680g for a period of 28 days. Clean drinking troughs were used in the administration of water and aqueous extract to the control and test rabbits respectively. Blood was collected for analyses intravenously through the larger vein at the back of the ear of rabbits. Hematological Swelab autocounter 920^{E+} (UK) was used in the determination of white blood cell differentials; neutrophils, lymphocytes, basophils and eosinophils. Analyses of results showed that the aqueous extract of the bark of *Triplochiton scleroxylon* did not have significant effects ($p>0.05$) on white blood cell differentials in alloxan-induced diabetic rabbits studied. Plasma glucose concentration decreased significantly ($p<0.05$) on the 18th, 24th and 28th days of administration of aqueous bark extract of this plant. *Triplochiton scleroxylon* has hypoglycemic and anti-diabetic properties and the aqueous bark extract may not predispose to white blood cell dysfunctions.

Key words: *Triplochiton scleroxylon*, alloxan-induced, white blood cell

Introduction

Diabetes mellitus is one of the diseases of man that has defied any sustained cure. Its age long existence and increasing complications have provoked a lot of search world wide for a cure. The disease is clinically treated by the use of insulin, sulphonylureas and biguanides. However, these treatments are only palliatives and therefore lack the ability to produce a cure. Besides, the complications arising from the use of these drugs are enormous. Consequently, the need for a search for a new drug without life-threatening side effects cannot be over-emphasized (Marrif *et al*., 1995; Liaquat *et al*., 1994; Kako *et al*., 1996).

The use of plant by man for the treatment of diseases is an age-long practice (Dalziel, 1956; Sofowora, 1984). *Triplochiton scleroxylon* is one of the over 30 Nigerian herbs with hypoglycemic properties and therefore used by some Nigerian diabetics to treat their ailments (Prohp *et al*., 2006b). This plant belongs to the family of tropical medicinal plants widely distributed in tropical West Africa from Guinea to Cameroon along waterways and on abandoned farms in the transition zone between the humid evergreen and semi deciduous forests (Russel *et al*., 1997; Richter and Dallwitz, 2000).

This study was designed to investigate the possible effects of the aqueous extract of *Triplochiton scleroxylon* on white blood cell differentials in alloxan-induced diabetic rabbits with the view of its proper classification as an anti-diabetic antidote. It is a part of the earlier study on red blood cell and associated parameters in alloxan-induced diabetic rabbits (Prohp *et al*., 2006b).

Materials and Methods

Animals: Male and female rabbits of the New Zealand strain weighing between 1200 and 1680g were used. They were maintained under standard animal house conditions and allowed free access to food (growers mash) and water for 2 weeks of acclimatization.

Chemical: Glucose oxidase kit was obtained from Randox laboratories United Kingdom. Sodium fluoride from BDH chemicals Ltd (Poole, Dorset, U.K.) and alloxan was supplied by Sigma. All other chemicals were of analar grade. Lahor Public Health and Research Center, Benin City, Edo State Nigeria was used in the analyses of the hematological parameters.

Medicinal plants: The barks of Obeche (*Triplochiton scleroxylon*) were purchased from medicinal plant dealers at Oyingbo market, Lagos, Lagos State, Nigeria and identified by experts in Botany department, Ambrose Alli University, Ekpoma, Edo State, Nigeria.

Table 1: Mean Plasma Glucose Concentration (mg 100 mLG^{1}) of alloxan-induced diabetic rabbits administered aqueous extract of *Triplochiton scleroxylon*

Time (days)	Non-diabetic control	Diabetic control	*Triplochiton scleroxylon*
0	90.00±2.04	172.67±7.32	164.32±4.62
1	84.45±3.26	176.52±2.57	168.91±4.58
6	71.25±5.60	177.33±7.23	163.67±3.72
12	92.18±4.12	190.25±2.53	161.35±3.50
18	74.36±6.32	180.00±5.30	*97.33±2.36
24	53.81±4.51	194.06±4.59	*99.25±3.26
28	72.12±5.35	179.58±2.35	*84.06±6.05

Values are mean plasma glucose concentration±S.E.M. of three separate readings from nine rabbits, *Values significantly different ($p<0.05$) from diabetic control

Table 2: Mean White Blood Cell Count (x 10^9 cells/L) of Alloxan-induced Diabetic Rabbits Administered Aqueous Extract of *Triplochiton scleroxylon*

Time (Days)	Non-Diabetic control	Diabetic control	Treated Diabetic (*Triplochiton scleroxylon*)
0	8.03±1.69	5.33±2.37	7.03±2.37
1	9.30±2.21	7.43±0.94	7.50±2.46
6	7.60±2.11	6.57±1.13	6.00±0.87
12	7.60±1.68	7.10±1.50	9.57±0.49
18	7.60±2.58	6.10±2.00	7.10±1.41
24	8.50±1.33	7.85±0.75	6.50±1.02
28	8.03±1.67	4.45±1.75	8.90±0.80

Values are mean white blood cell count±S.E.M of three separate determinations from nine rabbits, Values not significantly different ($p>0.05$) from diabetic control

Table 3: Mean Neutrophil Count (x 10^9 cells/L) of Alloxan-induced Diabetic Rabbits Administered Aqueous Extract of *Triplochiton scleroxylon*

Time (Days)	Non-Diabetic control	Diabetic control	Treated Diabetic (*Triplochiton scleroxylon)*
0	3.10±0.26	3.14±0.56	2.95±0.78
1	3.94±0.62	3.09±0.29	3.25±0.12
6	3.31±0.21	2.83±0.56	2.51±0.38
12	3.51±0.35	2.89±0.63	3.55±0.26
18	2.95±0.22	2.46±0.78	2.79±0.18
24	3.39±0.62	3.16±0.11	1.98±0.91
28	3.14±0.54	2.90±0.22	3.52±0.46

Values are mean neutrophil count±S.E.M of three separate determinations from nine rabbits, Values not significantly different ($p>0.05$) from diabetic control

Preparation and administration of plant extracts: Aqueous extract of this herb was prepared and administered to test rabbits according to the procedure of Onoagbe *et al*. (1999) and reported by Prohp *et al*., 2006a.

Administration of alloxan: Alloxan was administered to rabbits according to the procedure reported by Prohp *et al*., 2006b.

Collection of blood: Blood was obtained intravenously as reported by Prohp *et al*., 2006b.

Table 4: Mean Lymphocyte Count (x 10^9 cells/L) of Alloxan-induced Diabetic Rabbits Administered Aqueous Extract of *Triplochiton scleroxylon*

Time (Days)	Non-Diabetic control	Diabetic control	Treated Diabetic (*Triplochiton scleroxylon*)
0	4.64±0.55	4.21±0.29	3.76±1.13
1	4.90±0.61	3.97±0.60	3.88±1.25
6	3.87±0.35	3.41±0.50	3.20±0.45
12	4.66±0.54	3.85±0.79	5.67±0.23
18	3.97±0.42	3.34±0.92	3.93±0.27
24	4.66±0.36	4.30±0.61	3.68±0.56
28	4.45±0.64	4.10±0.41	4.84±0.31

Values are mean lymphocyte count±S.E.M of three separate determinations from nine rabbits, Values not significantly different ($p>0.05$) from diabetic control

Results

The results of the effects of the aqueous extract of *Triplochiton scleroxylon* on white blood cell differentials have been presented in Tables 1-6. Statistical analyses of results showed that the aqueous extract of this herb did not have any significant effect ($p>0.05$) on white blood cell differentials in alloxan-induced diabetic rabbits when compared with the non-diabetic controls. Plasma glucose concentration decreased significantly ($p<0.05$) on the 18th, 24th and 28th days of experiment as earlier reported by Prohp *et al*. (2006b).

Discussion

Amongst most Africans and in other parts of the world, the use of herbs is steadily gaining acceptance as an alternative to orthodox medicine. This has become necessary as the only means of averting or evading some life-threatening complications arising from the use of most orthodox drugs. Some of these drugs by their expensive market value have become the exclusive reserves of the highly affluent only, leaving the under-privileged with no better option. Herbs are therefore widely cultivated in some parts of Nigeria and the world at large and are readily available and affordable as against the expensive orthodox drugs. In the United States of America, medicinal plants constitute about 25% of all new referred prescription dispensed from community pharmacies (Prohp *et al*., 2006a,b; Trease and Evans, 1989). According to Alberti and Zimmet (1998), an estimated 6,000 to 7,000 tonnes of herbs are used annually as ingredients in about 5,500 different herbal products. In the cities of Africa, there is the uncompromisingly uneven distribution of personnel between rural and urban areas. Consequently, medicinal herbs are used in higher proportions in the rural areas (Onoagbe *et al*., 1999; Prohp *et al*., 2006b). There is therefore strong need to conduct thorough scientific investigation on these locally available herbs with the view of authentication of their efficacies for classification as designated or specific antidotes.

Triplochiton scleroxylon known locally as Obeche in Nigeria is one of the over 30 medicinal plants in use in

Table 5: Mean Basophil Count (x 10^9 cells/L) of Alloxan-induced Diabetic Rabbits Administered Aqueous Extract of *Triplochiton scleroxylon*

Time (Days)	Non-Diabetic control	Diabetic control	Treated Diabetic (*Triplochiton scleroxylon*)
0	0.04±0.00	0.04±0.00	0.04±0.00
1	0.05±0.00	0.04±0.00	0.04±0.00
6	0.04±0.01	0.03±0.01	0.03±0.00
12	0.04±0.00	0.04±0.01	0.05±0.00
18	0.04±0.01	0.03±0.00	0.04±0.00
24	0.04±0.00	0.04±0.00	0.03±0.01
28	0.04±0.00	0.03±0.01	0.04±0.00

Values are mean basophil count±S.E.M of three separate determinations from nine rabbits, Values not significantly different (p>0.05) from diabetic control

Table 6: Mean Eosinophil Count (x 10^9 cells/L) of Alloxan-induced Diabetic Rabbits Administered Aqueous Extract of *Triplochiton scleroxylon*

Time (Days)	Non-Diabetic control	Diabetic control	Treated Diabetic (*Triplochiton scleroxylon*)
0	0.07±0.01	0.07±0.01	0.07±0.00
1	0.08±0.01	0.07±0.00	0.07±0.00
6	0.07±0.00	0.06±0.01	0.06±0.01
12	0.07±0.01	0.07±0.01	0.09±0.00
18	0.08±0.01	0.05±0.00	0.09±0.01
24	0.07±0.01	0.08±0.01	0.05±0.00
28	0.08±0.01	0.07±0.00	0.08±0.00

Values are mean eosinophil count±S.E.M of three separate determinations from nine rabbits, Values not significantly different (p>0.05) from diabetic control

some part of Nigeria for the medication of diabetes mellitus. The medicinal value of any plant is a function of the presence of active ingredients viz. alkaloids, glycosides, resins, volatile oils, gum and tannins found in large concentrations in storage organs like roots, seeds, bark and leaves (Dalziel, 1987).

In this study, aqueous extract of the bark of *Triplochiton scleroxylon* was administered through clean drinking troughs to alloxan-induced diabetic rabbits for a period of 28 days. Analyses of results showed that the aqueous extract of this plant did not have significant effect (p>0.05) on the white blood cell differentials (white blood cell, neutrophil, lymphocyte, basophil and eosinophil counts) when compared with the non-diabetic controls. Studies have shown that reduction in neutrophil and lymphocyte counts are indicative of susceptibility to infection and other physiological anomalies e.g leukaemia, arthritis and possible compromise of cellular and humoral mediated immunity respectively. Changes in the values of basophil and eosinophil counts are common in allergic reactions and bone marrow suppression (Bochner *et al*., 1991; Bochner *et al*., 1994). The use of this extract as a hypoglycemic and anti-diabetic antidote may therefore not stimulate or trigger any of the abnormalities associated with the reduction of white blood cell differentials. Further studies are continuing on the scientific authentication of the use of *Triplochiton scleroxylon* as an anti-diabetic herb. Experimental diabetes would be induced with streptozotocin instead of alloxan.

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