Biochemistry

PROXIMATE AND PHYTOCHEMISTRY OF COSTUS AFER PLANT FROM NIGER DELTA STATE AND ITS EFFECT ON HAEMATOLOGICAL PARAMETERS

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ABSTRACT:

Background

Costus afer therapeutically importance in southern part of Nigeria cannot be over emphasized; it is valued in this part of Nigeria for its anti-arthritic, anti-inflammatory and anti-diabetic properties2. The use of herbs and plants to support treatment of diabetes following the observed inadequacy in some therapeutic drugs is gradually been practiced worldwide now. Aim: The aim of this study was to determine the phytochemistry and proximate composition of costus afer. Materials and method: Following the preparation of costus afer stem extract the plant was screened for phytochemicals and proximate composition. Results: The result of proximate and phytochemical screening of costus afer palnt indicated the presence of alkaloid, flavonoids, tannins, steroids and cardiac glycosides, carbohydrate, protein, lipid, ash, fiber and moisture. WBC count, neutrophil count, lymphocyte count, monocyte count, eosinophil count, basophil count, RBC count, HGB, HCT, MCV, MCH, MCHC, RDW.SD, PLT, MPV and PDW of untreated diabetic induced animals were lower compared to treated animals and the controls, the reduction was only significant for RBC and RDW.SD while PCT of untreated animals was increased.

Introduction

Costus afer Ker-Gawl (Costaceae) plant is found mostly in West African countries such as Nigeria, Ghana, Cameroon etc., this plant belongs to the stout species, perennial and rhizomatous herbs of the genus Costus1.   Costus afer therapeutically importance in southern part of Nigeria cannot be over emphasized; it is valued in this part of Nigeria for its anti-arthritic, anti-inflammatory and anti-diabetic properties2. The use of herbs and plants to support treatment of diabetes following the observed inadequacy in some therapeutic drugs is gradually been practiced worldwide now. Some of the side effect of these diabetic drugs includes hypoglycemia, gastrointestinal problems3, inability to control adequately hyperlipidemia associated with the disease4. Alkaloids, glycoside, terpens, flavonoids has been found in plants which are claimed to be effective in the treatment and management of diabetes5. The aim of this study was to determine the phytochemistry and proximate composition of costus afer.

Materials and methods
Collection of plant, identification and preparation of extract

Fresh samples of Costus afer (stem) were obtained from a small swampy forest area in Banga Camp, along Ayama Road, Obogoro-Atissa Community in Yenagoa Local Government Area, Bayelsa State, Nigeria. The plant stem sample was identified at the herbarium of the Plant Science and Biotechnology Department, University of Port Harcourt, Rivers State, Nigeria by Dr. Ekeke Ekezie. The method of Emeh et al., 2014 was adopted in the preparation of the aqueous extract of Costus afer 6.

Phytochemistal estimation

A.O.A.C method was used to determine proximate composition of the stem extract8, phytochemical screening for alkaloid, flavonoids, tannins, anthraquinnones, steroids, saponin and cardiac glycosides9-12.

Determination of hematological parameters

Packed cell volume (PCV), white blood count (WBC), red blood count (RBC), haemoglobin (Hb) concentration, platelet count, eosinophils (Eos.), basophils (Bas.), lymphocytes (Lym.), monocytes (Mon.), neutrophils (Neu.), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC) and mean corpuscular volume (MCV) were determined using standard method13   to ascertain the effects of the Costus afer extracton haematological parameters.

Analysis of results

Values were reported as Mean ± standard error of mean (SEM) while one way ANOVA was used to test for differences between treatment groups. The results were considered significant at p-values of less than 0.05, that is, at 95% confidence level (p < 0.05).

Results

The qualitative analysis of costus afer shows the presence of the following phytochemicals: alkaloid, flavonoids, tannins, steroids and cardiac glycosides, the result of this analysis is shown in tables 1 while proximate analysis of costus afer indicated the presence of carbohydrate, protein, lipid, ash, fiber and moisture in their various compositions as shown in tables 2. WBC count, neutrophil count, lymphocyte count, monocyte count, eosinophil count, basophil count, RBC count, HGB, HCT, MCV, MCH, MCHC, RDW.SD, PLT, MPV and PDW of untreated diabetic induced animals were lower compared to treated animals and the controls, the reduction was only significant for RBC and RDW.SD while PCT of untreated animals was increased table 3-table 20.

Table 1: Qualitative phytochemical compositions of costus afer stem aqueous extract

Phytochemical	Test	Result
Alkaloid	Dragendorff's	+ve
	Mayers	+ve
	Hagers	-ve

Flavonoids	Shinoda	+ve
	Alkali	-ve
Tannin	Ferric chloride	+ve
	Phlobatanins	-ve
Anthraquinones	Free anthraquinones	-ve
	Combine anthraquinones	-ve
Saponin	Frothing	-ve
Cardenolide	Keller killani	+ve
	Kedde	+ve
Triterpenoids/steroid	Lieberman	+ve
	Salwoski	+ve

Table 2: Proximate compositions of costus afer stem aqueous extract

Component	(%) Concentration
Carbohydrate	38.5 ± 1.17
Protein	3.38 ± 0.46
Lipid	1.3 ± 0.02
Ash	3 ± 0.3
Fiber	33 ± 0.86
Moisture	20.7 ± 1.12

Table 3: Mean (±SD) of hematological values of the experimental animals

	GRP 1	GRP 2	GRP 3	GRP 4	GRP 5
WBC (10^9/L)	7.46±1.53	4.42±0.57	5.01±1.02	6.51±1.50	6.51±1.72
Neutrophil (10^9/L)	29.70±7.40	17.90±13.58	15.87±8.98	20.73±7.82	14.67±5.51
Lymphocyte (10^9/L)	43.67±9.07	45.77±31.96	35.30±32.87	52.00±36.47	37.33±13.65
Monocyte (10^9/L)	9.00±2.17	7.27±2.95	4.50±2.52	3.97±2.45	3.70±1.40
Eosinophil (10^9/L)	3.07±0.42^d	1.90±2.02	1.47±0.60	0.30±0.20^a	0.47±0.55
Basophil (10^9/L)	1.70±0.62^{be}	0.47±0.31^a	0.67±0.47	0.70±0.10	0.47±0.38^a
RBC (10^6/L)	4.79±0.56	4.52±1.11	5.35±0.09	6.28±1.15	5.32±2.52
HGB (g/dl)	12.54±2.08	9.20±1.80	10.85±0.05	12.20±1.56	12.27±0.15
HCT (%)	45.26±5.98^b	27.30±7.00^a	33.40±2.90	40.23±8.29	40.05±1.15
MCV (10^{-15}L)	80.11±8.06^{bcde}	60.30±0.70^a	62.40±4.40^a	63.93±1.37^a	59.15±2.35^a
MCH (10^{-12})	28.50±2.51^{bcde}	20.60±1.10^a	20.30±0.40^a	19.57±1.01^a	18.10±0.00^a
MCHC (g/dL)	33.20±0.78	34.20±2.20	32.70±3.00	30.63±2.20	30.60±1.25

Table 3: Continuation Mean (±SD) of hematological values of the experimental animals

	GRP 1	GRP 2	GRP 3	GRP 4	GRP 5
RDW-CV (%)	0.16±0.00	0.15±0.01	0.17±0.00	0.18±0.02	0.17±0.01
RDW-SD (%)	47.8±4.74	39.30±2.50	44.50±4.10	47.10±6.80	41.90±3.50
PLT (K/UL)	405.33±36.30^e	520.00±144.00	447.00±31.00^e	546.33±83.76	740.00±90.00^{ac}
MPV (10^{-15})	7.27±0.86	6.80±0.10	6.75±0.05	6.73±0.40	6.30±0.00
PDW (10^{-15})	15.60±0.20^{de}	15.50±0.00^e	15.40±0.00	15.30±0.10^a	15.15±0.05^{ab}
PCT (μg/L)	0.25±0.04^e	0.36±0.10	0.30±0.02	0.37±0.08	0.47±0.06^a

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Superscript "d" shows significant difference, (p<0.05) when Group 4 is compared with other groups.
Superscript "e" shows significant difference, (p<0.05) when Group 5 is compared with other groups.

KEY: GRP 1= 2ml Extract; GRP 2= 1ml Extract; GRP 3= 0.50ml Extract; GRP 4= 0.25ml Extract; GRP 5= 0.10ml Extract.

Table 4: Effect of costus afer stem extract on white blood cell (WBC) count (K/µL) of alloxan-induced diabetic wistar albino rats

	GRP 1	GRP 2	GRP 3	GRP 4	GRP 5	GRP 6	GRP 7	GRP 8
Week 1	4.99± 3.76	0.52± 0.79	5.01± 1.02	3.39± 0.17	5.85± 3.90	4.40± 0.80	5.33± 1.06	5.85± 0.44
Week 2	6.84± 1.24^b	0.07± 0.02^{acdefgh}	4.71± 1.15^b	6.25± 0.28^b	6.05± 2.74^b	5.83± 1.48^b	5.50± 0.72^b	6.66± 1.38^b
Week 3	6.91± 1.16	0.56± 0.85^{fh}	5.24± 1.01^h	8.05± 0.09	7.42± 2.88	8.80± 4.92^b	5.80± 0.50^h	14.24± 5.38^{bcg}
Week 4	11.00± 7.45	0.62± 0.96	5.01± 1.02	9.45± 5.96	4.44± 2.45	7.43± 3.87	6.41± 2.32	12.53± 2.20

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Superscript "f" shows significant difference, (p<0.05) when Group 6 is compared with other groups.
Superscript "g" shows significant difference, (p<0.05) when Group 7 is compared with other groups.
Superscript "h" shows significant difference, (p<0.05) when Group 8 is compared with other groups.

KEY: GRP 1= Normal control; GRP 2= Diabetic control; GRP 3= Normal (non-diabetic) rats + 1ml Extract; GRP 4= Diabetic rats treated with Metformin; GRP 5= Diabetic rats treated with 0.25ml Extract; GRP 6= Diabetic rats treated with 0.50ml Extract; GRP 7= Diabetic rats treated with 1.0ml Extract; GRP 8= Diabetic rats treated with 2.0ml Extract.

Table 5: Effect of costus afer stem extract on neutrophil count (109/L) of alloxan-induced diabetic wistar albino rats

	GRP 1	GRP 2	GRP 3	GRP 4	GRP 5	GRP 6	GRP 7	GRP 8
Week 1	36.87± 1.62^{bcfgh}	11.43± 0.95^{ade}	17.83± 2.78^{ad}	42.00± 1.00^{bcfgh}	28.90± 15.30^b	16.30± 1.05^{ad}	18.17± 1.62^{ad}	20.77± 2.36^{ad}
Week 2	39.00± 5.23^{bcdefg}	10.63± 0.71^{ah}	13.70± 1.90^{ah}	18.30± 1.10^{ah}	7.33± 3.27^{agh}	7.80± 3.22^{agh}	20.43± 7.40^{aefh}	36.50± 4.88^{bcdefg}
Week 3	36.67± 1.30^{bcf}	10.00± 1.40^a	12.77± 0.59^a	27.50± 10.79^f	26.77± 6.54^f	6.10± 1.30^{ade}	18.93± 5.53	22.97± 12.99
Week 4	30.03± 12.74	12.40± 1.80	23.70± 7.95	49.63± 12.37	45.57± 24.25	33.45± 4.65	30.38± 2.66	35.90± 8.50

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Table 6: Effect of Costus afer stem extract on lymphocyte count (109/L) of alloxan-induced diabetic wistar albino rats

	GRP 1	GRP 2	GRP 3	GRP 4	GRP 5	GRP 6	GRP 7	GRP 8
Week 1	62.37± 1.10[bdegh]	38.67± 6.51[acefgh]	71.37± 1.39[bde]	47.63± 0.64[acfgh]	51.65± 7.65[abcfgh]	72.17± 0.75[bde]	76.00± 2.00[abde]	78.00± 2.00[abde]
Week 2	65.37± 5.82[def]	35.00± 8.89	56.70± 15.11[e]	31.60± 1.44[a]	22.00± 21.00[ac]	23.47± 10.46[a]	40.73± 2.53	47.60± 14.41
Week 3	62.70± 1.47[bf]	36.00± 8.54[adh]	46.73± 6.47[bf]	60.67± 2.52[f]	53.67± 8.02[f]	23.25± 10.45[acdegh]	45.40± 5.79[f]	62.17± 3.35[bf]
Week 4	56.30± 3.00	30.75± 29.65	49.57± 20.87	43.27± 11.47	27.80± 2.60	55.30± 1.56	57.10± 0.58	49.10± 6.58

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Superscript "d" shows significant difference, (p<0.05) when Group 4 is compared with other groups.
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Table 7; Effect of costus afer stem extract on monocyte count (109/L) of alloxan-induced diabetic wistar albino rats

	GRP 1	GRP 2	GRP 3	GRP 4	GRP 5	GRP 6	GRP 7	GRP 8
Week 1	1.60± 0.17[h]	1.20± 0.35[h]	9.07± 3.17	12.47± 8.38	15.23± 12.82	14.67± 10.97	19.33± 4.16	21.67± 2.08[ab]
Week 2	1.70± 0.17	1.20± 0.35	5.20± 3.18	3.70± 3.15	2.13± 0.95	1.60± 0.50	1.15± 0.13	2.30± 1.15
Week 3	1.60± 0.17[c]	1.27± 0.42[c]	3.73± 1.07[abdefgh]	1.23± 0.85[c]	1.07± 0.70[c]	1.85± 0.25[c]	1.41± 0.35[c]	1.67± 0.81[c]
Week 4	2.80± 0.60	1.55± 1.05	3.93± 4.24	3.70± 3.10	1.27± 0.12	2.15± 0.05	2.32± 0.25	2.57± 1.36

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Table 8: Effect of costus afer stem extract on eosinophil count (109/L) of alloxan-induced diabetic wistar albino rats

	GRP 1	GRP 2	GRP 3	GRP 4	GRP 5	GRP 6	GRP 7	GRP 8
Week 1	0.47± 0.12[h]	0.33± 0.15[h]	2.70± 0.66	5.40± 1.31	5.10± 4.90	3.57± 5.57	4.80± 2.00	8.70± 2.26[ab]
Week 2	0.60± 0.20	0.33± 0.15[c]	1.70± 0.69[b]	1.09± 1.02	0.43± 0.15	0.50± 0.10	0.50± 0.10	0.43± 0.06
Week 3	0.67± 0.31	0.37± 0.12	1.30± 0.69	3.47± 3.88	0.80± 0.50	0.47± 0.06	0.57± 0.06	0.80± 0.53
Week 4	3.00± 2.36[g]	0.55± 0.18	1.90± 0.72[fg]	5.30± 1.32	8.30± 4.75	8.90± 2.00[c]	9.97± 1.01[ac]	3.70± 0.20

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Table 9: Effect of costus afer stem extract on basophil count (109/L) of alloxan-induced diabetic wistar albino rats

	GRP 1	GRP 2	GRP 3	GRP 4	GRP 5	GRP 6	GRP 7	GRP 8
Week 1	0.47± 0.06	0.50± 0.10	0.50± 0.00	0.43± 0.06	0.67± 0.15	0.60± 0.10	0.53± 0.06	0.57± 0.06
Week 2	0.47± 0.06	0.40± 0.10	0.43± 0.06	0.47± 0.06	0.43± 0.06	0.53± 0.06	0.43± 0.06	0.50± 0.10
Week 3	0.50± 0.00	0.43± 0.12	0.37± 0.06	0.20± 0.17[f]	0.23± 0.15	0.53± 0.06[d]	0.47± 0.12	0.33± 0.12
Week 4	0.10± 0.10	0.57± 0.60	0.37± 0.32	0.20± 0.00	0.57± 0.50	0.23± 0.06	0.27± 0.06	0.25± 0.15

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Superscript "f" shows significant difference, (p<0.05) when Group 6 is compared with other groups.

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Table 10: Effect of costus afer stem extract on red blood cell (RBC) count (10-6L) of alloxan-induced diabetic wistar albino rats

	GRP 1	GRP 2	GRP 3	GRP 4	GRP 5	GRP 6	GRP 7	GRP 8
Week 1	4.75± 3.06[b]	0.37± 0.14[acdefgh]	5.35± 0.09[b]	6.59± 0.56[b]	6.19± 0.64[b]	7.20± 0.80[b]	6.06± 0.71[b]	6.02± 0.33[b]
Week 2	6.74± 0.45[b]	0.70± 0.72[acdefgh]	5.35± 0.09[be]	6.33± 0.11[b]	7.01± 0.03[bc]	6.71± 0.53[b]	6.00± 0.63[b]	6.54± 0.83[b]
Week 3	7.01± 0.25[b]	0.88± 0.65[acdefgh]	5.30± 0.17[b]	6.81± 0.81[b]	6.38± 0.54[b]	6.96± 0.54[b]	6.24± 1.00[b]	5.38± 0.64[b]
Week 4	6.47± 1.29	0.96± 0.82	5.67± 0.56	5.90± 1.29	4.72± 3.72	6.42± 0.68	4.25± 3.36	3.53± 2.94

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Table 11: Effect of costus afer stem extract on haemoglobin (Hb) concentration (g/dL) of alloxan-induced diabetic wistar albino rats

	GRP 1	GRP 2	GRP 3	GRP 4	GRP 5	GRP 6	GRP 7	GRP 8
Week 1	9.50± 5.38	3.80± 5.54[f]	10.85± 0.05	11.43± 0.71	11.63± 1.07	11.87± 1.10[b]	9.80± 0.60	11.27± 1.01
Week 2	9.87± 4.75	4.27± 5.20[de]	10.80± 0.10	12.05± 0.05[b]	13.25± 0.55[b]	11.05± 0.37	10.33± 1.95	11.30± 0.90
Week 3	11.73± 1.55[b]	4.63± 5.01[adefh]	10.13± 1.07	11.80± 0.20[b]	11.90± 0.40[b]	10.68± 0.50[b]	9.67± 1.10	11.05± 0.95[b]
Week 4	13.10± 0.50	6.45± 6.15	10.85± 0.05	12.50± 1.47	9.38± 3.53	12.20± 6.26	8.53± 7.93	10.10± 7.93

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Superscript "h" shows significant difference, (p<0.05) when Group 8 is compared with other groups.

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Table 12: Effect of costus afer stem extract on haematocrit (HCT) (%) of alloxan-induced diabetic wistar albino rats

	GRP 1	GRP 2	GRP 3	GRP 4	GRP 5	GRP 6	GRP 7	GRP 8
Week 1	31.10± 19.16	15.27± 11.71	33.40± 2.90	35.83± 1.52	37.77± 4.96	36.80± 2.17	27.53± 2.25	26.47± 3.11
Week 2	38.10± 7.21	18.60± 15.74[g]	33.57± 2.91	37.23± 0.38	42.55± 1.85	39.00± 16.37	46.10± 2.74[b]	41.90± 4.46
Week 3	40.07± 4.05	26.60± 22.30	34.73± 1.36	37.40± 1.20	35.90± 1.20	44.73± 7.55	43.53± 4.42	33.60± 2.60
Week 4	40.53± 4.32[c]	26.83± 6.75[c]	33.40± 2.90[abdefgh]	32.87± 1.71[c]	25.70± 2.02[c]	38.83± 8.15[c]	26.90± 2.09[c]	45.80± 5.13[c]

Table 13: Effect of costus afer stem extract on mean corpuscular volume (MCV) (10-15L) of alloxan-induced diabetic wistar albino rats

	GRP 1	GRP 2	GRP 3	GRP 4	GRP 5	GRP 6	GRP 7	GRP 8
Week 1	67.80± 5.62	55.50± 16.24	62.40± 4.40	55.37± 3.49	60.83± 1.82	57.13± 1.22	56.17± 1.78	53.20± 2.44
Week 2	75.30± 12.19	55.90± 17.24	61.35± 2.91	59.30± 0.92	60.70± 2.40	59.00± 1.25	59.23± 0.91	62.80± 5.51
Week 3	73.23± 12.97	58.73± 12.54	64.07± 2.39	55.50± 4.90	56.40± 1.90	55.90± 6.45	61.93± 4.65	61.15± 1.95
Week 4	63.63± 7.35	54.70± 2.30	62.40± 4.90	60.73± 7.84	58.17± 3.61	58.93± 3.86	61.60± 3.21	55.67± 0.68

Table 14: Effect of costus afer stem extract on mean corpuscular haemoglobin (MCH) (10-12L) of alloxan-induced diabetic wistar albino rats

	GRP 1	GRP 2	GRP 3	GRP 4	GRP 5	GRP 6	GRP 7	GRP 8
Week 1	21.83± 4.30	16.60± 3.75	20.30± 0.40	17.70± 0.85	18.80± 0.80	18.53± 0.31	17.90± 0.44	19.40± 0.82
Week 2	20.73± 5.52	15.47± 4.69	20.23± 0.42	19.05± 0.25	18.90± 0.70	18.53± 0.95	19.20± 0.30	20.00± 1.75
Week 3	16.57± 2.60	17.47± 3.63	19.53± 0.81	17.50± 1.80	18.70± 0.60	18.07± 1.60	18.87± 0.35	20.05± 0.45
Week 4	20.90± 4.82	18.00± 0.60	20.30± 0.40	20.90± 4.29	35.67± 9.82	18.13± 0.31	22.87± 2.91	36.25± 7.45

Superscript "a" shows significant difference, (p<0.05) when Group 1 is compared with other groups.
Superscript "b" shows significant difference, (p<0.05) when Group 2 is compared with other groups.
Superscript "c" shows significant difference, (p<0.05) when Group 3 is compared with other groups.
Superscript "d" shows significant difference, (p<0.05) when Group 4 is compared with other groups.
Superscript "e" shows significant difference, (p<0.0 5) when Group 5 is compared with other groups.
Superscript "f" shows significant difference, (p<0.05) when Group 6 is compared with other groups.
Superscript "g" shows significant difference, (p<0.05) when Group 7 is compared with other groups.
Superscript "h" shows significant difference, (p<0.05) when Group 8 is compared with other groups.

KEY: GRP 1= Normal control; GRP 2= Diabetic control; GRP 3= Normal (non-diabetic) rats + 1ml Extract; GRP 4= Diabetic rats treated with Metformin; GRP 5= Diabetic rats treated with 0.25ml Extract; GRP 6= Diabetic rats treated with 0.50ml Extract; GRP 7= Diabetic rats treated with 1.0ml Extract; GRP 8= Diabetic rats treated with 2.0ml Extract.

Table 15: Effect of costus afer stem extract on mean corpuscular haemoglobin concentration (MCHC) (g/dL) of alloxan-induced diabetic wistar albino rats

	GRP 1	GRP 2	GRP 3	GRP 4	GRP 5	GRP 6	GRP 7	GRP 8
Week 1	32.03± 3.64^b	26.37± 1.98^{acdh}	32.70± 3.00^b	33.13± 0.76^b	30.90± 1.73	31.83± 0.15	31.70± 0.30	32.60± 1.22^b
Week 2	30.97± 1.82^b	29.57± 2.65^{acdefgh}	30.73± 2.30^b	32.10± 0.10^b	31.15± 0.05^b	31.48± 1.02^b	31.87± 1.16^b	31.97± 0.35^b
Week 3	29.50± 0.90	25.70± 2.44^{cdefgh}	33.47± 4.20^b	31.55± 0.45^b	33.27± 0.21^b	32.01± 1.92^b	32.30± 0.72^b	32.87± 0.35^b
Week 4	32.53± 3.66	32.90± 0.20	35.70± 3.40	36.57± 10.82	83.43± 88.83	32.17± 0.76	38.23± 10.39	65.70± 32.10

Superscript "a" shows significant difference, (p<0.05) when Group 1 is compared with other groups.
Superscript "b" shows significant difference, (p<0.05) when Group 2 is compared with other groups.
Superscript "c" shows significant difference, (p<0.05) when Group 3 is compared with other groups.
Superscript "d" shows significant difference, (p<0.05) when Group 4 is compared with other groups.
Superscript "e" shows significant difference, (p<0.0 5) when Group 5 is compared with other groups.
Superscript "f" shows significant difference, (p<0.05) when Group 6 is compared with other groups.
Superscript "g" shows significant difference, (p<0.05) when Group 7 is compared with other groups.
Superscript "h" shows significant difference, (p<0.05) when Group 8 is compared with other groups.

KEY: GRP 1= Normal control; GRP 2= Diabetic control; GRP 3= Normal (non-diabetic) rats + 1ml Extract; GRP 4= Diabetic rats treated with Metformin; GRP 5= Diabetic rats treated with 0.25ml Extract; GRP 6= Diabetic rats treated with 0.50ml Extract; GRP 7= Diabetic rats treated with 1.0ml Extract; GRP 8= Diabetic rats treated with 2.0ml Extract.

Table 16: Effect of costus afer stem extract on red blood cell distribution width (RDW.SD) (%) of alloxan-induced diabetic wistar albino rats

	GRP 1	GRP 2	GRP 3	GRP 4	GRP 5	GRP 6	GRP 7	GRP 8
Week 1	43.50± 5.44^b	56.87± 1.03^{acdefgh}	44.33± 5.51^b	36.20± 1.71^b	42.60± 2.35^b	40.41± 0.34^b	39.53± 1.07^b	42.80± 3.42^b
Week 2	40.93± 1.29^b	53.87± 2.65^{acdefgh}	41.00± 7.00^b	41.70± 1.90^b	41.35± 1.85^b	43.37± 2.50^b	45.80± 2.46^b	45.83± 3.10^b
Week 3	41.53± 0.40	51.78± 7.68^{de}	42.33± 9.07	38.25± 3.35^b	35.85± 1.35^b	43.60± 2.45	46.57± 2.92	39.55± 1.65
Week 4	43.43± 11.60	36.70± 3.70	40.73± 10.93	41.83± 9.27	37.83± 4.13	35.60± 2.10	38.67± 7.33	35.25± 0.85

Superscript "a" shows significant difference, (p<0.05) when Group 1 is compared with other groups.
Superscript "b" shows significant difference, (p<0.05) when Group 2 is compared with other groups.
Superscript "c" shows significant difference, (p<0.05) when Group 3 is compared with other groups.
Superscript "d" shows significant difference, (p<0.05) when Group 4 is compared with other groups.
Superscript "e" shows significant difference, (p<0.0 5) when Group 5 is compared with other groups.
Superscript "f" shows significant difference, (p<0.05) when Group 6 is compared with other groups.
Superscript "g" shows significant difference, (p<0.05) when Group 7 is compared with other groups.
Superscript "h" shows significant difference, (p<0.05) when Group 8 is compared with other groups.

KEY: GRP 1= Normal control; GRP 2= Diabetic control; GRP 3= Normal (non-diabetic) rats + 1ml Extract; GRP 4= Diabetic rats treated with Metformin; GRP 5= Diabetic rats treated with 0.25ml Extract; GRP 6= Diabetic rats treated with 0.50ml Extract; GRP 7= Diabetic rats treated with 1.0ml Extract; GRP 8= Diabetic rats treated with 2.0ml Extract.

Table 17: Effect of Costus afer stem extract on platelet (PLT) count (K/UL) of alloxan-induced diabetic wistar albino rats

	GRP 1	GRP 2	GRP 3	GRP 4	GRP 5	GRP 6	GRP 7	GRP 8
Week 1	464.00± 333.75	115.67± 62.13	447.00± 31.00	373.00± 212.11	276.67± 106.84	417.83± 49.54	465.00± 30.00	472.67± 35.57
Week 2	583.00± 137.57	167.00± 50.76^{de}	438.67± 34.20	613.00± 217.77^b	612.33± 145.50^b	496.67± 204.39	444.00± 28.83	584.33± 210.06
Week 3	557.00± 178.53	211.00± 100.50	493.33± 80.11	578.50± 109.50	516.00± 223.00	475.33± 215.07	508.33± 110.83	519.00± 197.34
Week 4	289.33± 78.62	180.50± 13.50	467.60± 37.20	412.67± 141.68	217.67± 186.35	411.00± 64.28	442.00± 112.97	374.33± 145.24

Superscript "a" shows significant difference, (p<0.05) when Group 1 is compared with other groups.
Superscript "b" shows significant difference, (p<0.05) when Group 2 is compared with other groups.
Superscript "c" shows significant difference, (p<0.05) when Group 3 is compared with other groups.
Superscript "d" shows significant difference, (p<0.05) when Group 4 is compared with other groups.
Superscript "e" shows significant difference, (p<0.0 5) when Group 5 is compared with other groups.
Superscript "f" shows significant difference, (p<0.05) when Group 6 is compared with other groups.
Superscript "g" shows significant difference, (p<0.05) when Group 7 is compared with other groups.
Superscript "h" shows significant difference, (p<0.05) when Group 8 is compared with other groups.

KEY: GRP 1= Normal control; GRP 2= Diabetic control; GRP 3= Normal (non-diabetic) rats + 1ml Extract; GRP 4= Diabetic rats treated with Metformin; GRP 5= Diabetic rats treated with 0.25ml Extract; GRP 6= Diabetic rats treated with 0.50ml Extract; GRP 7= Diabetic rats treated with 1.0ml Extract; GRP 8= Diabetic rats treated with 2.0ml Extract

Table 18: Effect of costus afer stem extract on mean platelet volume (MPV) (10-15L) of alloxan-induced diabetic wistar albino rats.

	GRP 1	GRP 2	GRP 3	GRP 4	GRP 5	GRP 6	GRP 7	GRP 8
Week 1	6.97± 0.25	6.23± 0.12[e]	6.75± 0.05	6.87± 0.25	7.23± 0.32[b]	6.80± 0.40	6.70± 0.20	6.60± 0.44
Week 2	6.77± 0.21	6.63± 0.32	6.70± 0.10	7.05± 0.45	6.50± 0.20	6.67± 0.45	6.57± 0.31	6.67± 0.74
Week	6.90± 0.26	6.20± 0.10[h]	6.57± 0.25[h]	6.60± 0.20[h]	6.60± 0.50[h]	6.60± 0.36[h]	6.63± 0.31	7.45± 0.15[bcdef]
Week 4	6.30± 0.98	6.20± 0.10	6.67± 0.32	7.17± 0.90	6.63± 2.21	5.50± 2.55	5.47± 1.12	6.50± 2.62

Superscript "a" shows significant difference, (p<0.05) when Group 1 is compared with other groups.
Superscript "b" shows significant difference, (p<0.05) when Group 2 is compared with other groups.
Superscript "c" shows significant difference, (p<0.05) when Group 3 is compared with other groups.
Superscript "d" shows significant difference, (p<0.05) when Group 4 is compared with other groups.
Superscript "e" shows significant difference, (p<0.0 5) when Group 5 is compared with other groups.
Superscript "f" shows significant difference, (p<0.05) when Group 6 is compared with other groups.
Superscript "g" shows significant difference, (p<0.05) when Group 7 is compared with other groups.
Superscript "h" shows significant difference, (p<0.05) when Group 8 is compared with other groups.

KEY: GRP 1= Normal control; GRP 2= Diabetic control; GRP 3= Normal (non-diabetic) rats + 1ml Extract; GRP 4= Diabetic rats treated with Metformin; GRP 5= Diabetic rats treated with 0.25ml Extract; GRP 6= Diabetic rats treated with 0.50ml Extract; GRP 7= Diabetic rats treated with 1.0ml Extract; GRP 8= Diabetic rats treated with 2.0ml Extract.

Table 19: Effect of costus afer stem extract on platelet distribution width (PDW) (10-15L) of alloxan-induced diabetic wistar albino rats

	GRP 1	GRP 2	GRP 3	GRP 4	GRP 5	GRP 6	GRP 7	GRP 8
Week 1	15.80± 0.26	15.60± 0.46	15.50± 0.17	15.40± 0.26	15.63± 0.06	15.37± 0.21	15.41± 0.22	15.30± 0.17
Week 2	15.83± 0.28	15.63± 0.47	15.57± 0.57	15.33± 0.15	15.47± 0.06	15.47± 0.25	15.30± 0.10	15.53± 0.31
Week 3	15.80± 0.26[d]	15.67± 0.38	15.30± 0.17	14.83± 0.60[a]	15.25± 0.15	15.53± 0.15	15.13± 0.38	15.60± 0.10
Week 4	15.53± 0.35	15.67± 0.38	15.47± 0.12	15.47± 0.46	15.03± 0.21	15.50± 0.53	15.77± 0.67	15.50± 0.20

Superscript "a" shows significant difference, (p<0.05) when Group 1 is compared with other groups.
Superscript "b" shows significant difference, (p<0.05) when Group 2 is compared with other groups.
Superscript "c" shows significant difference, (p<0.05) when Group 3 is compared with other groups.
Superscript "d" shows significant difference, (p<0.05) when Group 4 is compared with other groups.
Superscript "e" shows significant difference, (p<0.0 5) when Group 5 is compared with other groups.
Superscript "f" shows significant difference, (p<0.05) when Group 6 is compared with other groups.
Superscript "g" shows significant difference, (p<0.05) when Group 7 is compared with other groups.
Superscript "h" shows significant difference, (p<0.05) when Group 8 is compared with other groups.

KEY: GRP 1= Normal control; GRP 2= Diabetic control; GRP 3= Normal (non-diabetic) rats + 1ml Extract; GRP 4= Diabetic rats treated with Metformin; GRP 5= Diabetic rats treated with 0.25ml

Extract; GRP 6= Diabetic rats treated with 0.50ml Extract; GRP 7= Diabetic rats treated with 1.0ml Extract; GRP 8= Diabetic rats treated with 2.0ml Extract.

Table 20: Effect of costus afer stem extract on procalcitonin (PCT) concentration (µg/L) of alloxan-induced diabetic wistar albino rats

	GRP 1	GRP 2	GRP 3	GRP 4	GRP 5	GRP 6	GRP 7	GRP 8
Week 1	0.33± 0.24[b]	0.04± 0.02[a]	0.17± 0.05	0.11± 0.02	0.20± 0.09	0.23± 0.04	0.21± 0.10	0.23± 0.04
Week 2	0.33± 0.24	0.06± 0.05[defg]	0.44± 0.12[b]	0.48± 0.09[b]	0.40± 0.08[b]	0.35± 0.03[b]	0.35± 0.02[b]	0.32± 0.01
Week 3	0.48± 0.06[b]	0.11± 0.11[acdfg]	0.40± 0.07[b]	0.40± 0.08[b]	0.33± 0.12	0.34± 0.02[b]	0.38± 0.05[b]	0.31± 0.07
Week 4	0.42± 0.18[b]	0.09± 0.13[a]	0.37± 0.08	0.34± 0.10	0.36± 0.06	0.34± 0.03	0.35± 0.03	0.27± 0.13

Superscript "a" shows significant difference, (p<0.05) when Group 1 is compared with other groups.
Superscript "b" shows significant difference, (p<0.05) when Group 2 is compared with other groups.
Superscript "c" shows significant difference, (p<0.05) when Group 3 is compared with other groups.
Superscript "d" shows significant difference, (p<0.05) when Group 4 is compared with other groups.
Superscript "e" shows significant difference, (p<0.0 5) when Group 5 is compared with other groups.
Superscript "f" shows significant difference, (p<0.05) when Group 6 is compared with other groups.
Superscript "g" shows significant difference, (p<0.05) when Group 7 is compared with other groups.
Superscript "h" shows significant difference, (p<0.05) when Group 8 is compared with other groups.

KEY: GRP 1= Normal control; GRP 2= Diabetic control; GRP 3= Normal (non-diabetic) rats + 1ml Extract; GRP 4= Diabetic rats treated with Metformin; GRP 5= Diabetic rats treated with 0.25ml Extract; GRP 6= Diabetic rats treated with 0.50ml Extract; GRP 7= Diabetic rats treated with 1.0ml Extract; GRP 8= Diabetic rats treated with 2.0ml Extract.

Discussions and conclusion

Plants belonging to the costaceae family are known to be effective in the management of diabetes14; results from this study for qualitative phytochemical screening of costus afer stem indicated the presence of steroids, cardenolide, tannin, flavonoids and alkaloid. Flavonoid prevents oxidative activities, inflammatory/allergies protection while15 alkaloid has antibacterial and antispasmodic potentials[16].

Carbohydrate serves as source of energy thus supporting the traditional claims of rehydrating its users; costus afer plant would be and is effective in the management of diabetes because of high composition of crude fiber. Obesity is a risk factor for diabetes17 and it is mainly caused as a result of increase in energy absorption to energy expenditure ratio18, a diet rich in fiber is more preferable as the energy absorbed in fiber diet is reduced probably due to decreased digestibility of fiber diets19. Crude fiber aids absorption of trace elements in the guts and also prevents diverticulosis20, it could be used as foliage for animals providing them with adequate nutrients as presence of protein, fats, moisture and even ash was found in the plant as it proximate constituent.

Steroids, cardenolide, tannin, flavonoids and alkaloid are constituted in costus afer plant, the presence of these phytochemicals are responsible for the traditional usage of the plant. Based on the proximate composition of the plant it serves as an adequate source of food for animals.

Effect of costus afer stem extract on haematological parameters of

alloxan-induced diabetic albino wistar rats As observed in the study in the presence of untreated diabetes the following parameters were reduced WBC count, neutrophil count, lymphocyte count, monocyte count, eosinophil count, basophil count, RBC count, HGB, HCT, MCV, MCH, MCHC, RDW.SD, PLT, MPV and PDW while treatment with both metformin and plant extracts at varying concentration elevated these parameters, increase of these parameters was only significant for RBC and RDW.SD in this study. The assessment of haematological parameters is a biomarker for evaluating the haematotoxic potential of the extract in area of pharmacognosy and also to explain blood relating functions of a plant extract or its products21. It can also be stated that these parameters can be used to determine the extent of deleterious effect of foreign compounds, including plant extracts, on the blood constituents of an animal22. This observation of increase in the levels of these parameters by this plant extract shows that the principal function of phagocytes, which is to defend against invading microorganisms by ingesting and destroying them, thus contributing to cellular inflammatory processes23 which may account for its antibacterial activity and antimicrobial activity24 . It has also being confirmed that the high antimicrobial activity shown by this extract on organisms may be either due to the presence of alkaloids, flavonoids, saponins, tannins, steroids and cardiac glycosides[24].

As a result of glycosylation of haemoglobin due to hyperglycemia associated with diabetes, the total haemoglobin of is reduced25, haemoglobin plays a major role in the transport of oxygen. The observed reduction in red blood cell of untreated experimental animals is as a result of reactive oxygen species generated, generation of reactive specie is the mechanism through which alloxan executes it's cytotoxic diabetogenic action26, costus afer plant are effective in the treatment of various oxidative related disease27. Reduced PCV, RBC and hemoglobin observed in this experiment is in accordance with other previous study[28, 29].

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