



Preliminary Phytochemical Analysis of Aqueous Extract of Leaf *Psidium guajava* and Its Effect on Some Haematological Parameters

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Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

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ABSTRACT

Various parts of *psidium guajava* linn are used in ethnomedicine in controls and treatments of different disorders such as liver, heart, and blood related disorders [11]. This research work investigate qualitative phytochemical composition of ethanol leaf extract of *psidium guajava* linn and its effect on some hematological parameters in albino rats. Qualitative phytochemical analysis of the extract was performed by standard procedures. A total of 20 adult male albino rats were used, they were randomly placed into 5 groups (A, B, C, D and E) each group containing 4 rats groups A, B, C, and D were administered, via oral intubation, the extract at doses of 200, 400, 600, and 800 mg/kg body weight respectively of the extract for 7 consecutive days. Phytochemical screening shows that the extract was rich in tannins anthraquinones, and flavonoid while alkaloids, saponins, terpenes and glycoside were slightly present. There was a decrease in the average body weight, physical activities, and feed and water intake of the rats in the test groups relative to the control hemoglobin concentrate, total white blood cell and packed cell volume recorded in the groups administered the extract were significantly ($p < 0.05$) higher than in the control this effect on the haematological parameters was found to be dose dependent. The findings of this study

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suggest that the aqueous extract of the fruit plants may be useful in control and treatment of blood related disorders. These useful applications may be as a result of the phytochemical present in the extract.

Keywords: Phytochemical analysis; heamatological parameters; guava leaf.

1. INTRODUCTION

The guava fruit plant is one of the numerous medicinal plant used all over the world especially Nigeria. Fruit plants are utilized for various medicinal purposes such as control, treatment, prevention of numerous disorder. The use started with the early traditional medicine practitioner, according to World Health Organization (WHO) 80% of people depends on the medicinal plant. For instance the popular dogonyaro and karikapapya serve as an antimalarial. Guava leaf are used for controls and treatment of several disorders such as diarrhea, constipation, GIT disorder, head ache and blood pressure. In this practice, when somebody is suspected to have lost large quantity of blood either by accident or during child delivery, they are given a concoction made of guava leaf to drink and boost their blood level [1].

The area of drug discovery is of utmost important to the whole world, the orthodox medicine of using synthetic drug has been shown to cause several side effect that are not desirable, also the drugs are expensive not really affordable by the poor man. So that is a big challenge the world today now look for an alternative in medicinal plant research. To identify the phytochemical constituents of the ethanol leaf extract of *Psidium guajava* linn and to investigates its effect on some hematological parameters. Preparation of ethanol leaf extract, Qualitative phytochemical screening of the extract, Determination of effect of the extract on hemoglobin, packed cell volume and total white blood cell count. Statistical Analysis and biochemical interpretation of the generated data [2].

The chemical analysis of guava fruit plant extract have revealed the presence of anti microbial compounds [3], tannins, phenol, triterpenes, flavonoids, guajivolic acid, guajavanoicacid, linolenic acid, guavacoumaricb acid, galaturonic acid, asphaltic acid, benzaldehyde, essential oils, saponins, carofenoid, cectin, fibre, fatty acids, and a high content of vitamins C and A in its fruit. The leaves are rich in flavonoid (quercetin) attributing for most of its therapeutic activity [4]. Hemoglobin(Hb) is the major protein of the red blood cell it is responsible for carrying oxygen

from lungs to tissues and then carry carbon dioxide from the tissues back to the lungs for exhalation. The Packed cell volume (PCV) is a measurement of the proportion of blood that is made of cells. White blood cell (WBC) plays an important role in protection of the living system against foreign invaders, against pathogens they are different types of white blood cell they include; neutrophils, lymphocytes, eosinophils, monocytes, basophils and granulocyte. An increase or decrease in white blood cell is an indicator of the over all immune system of an individual. WBC ranges from 4000-11000/mL [5].

The leaf of the guava fruit plant is widely used for the control and treatment of several disorders by the traditional medicine practitioners but most of this their claims that guava can do this or that have not been fully investigated that is why the present research on its effect on the haematological parameters was investigated.

2. MATERIALS AND METHODS

2.1 Collection of Plant Leaf (*psidium guajava* linn)

The fresh leaves of *psidium guajava* linn were collected from Mgbowo in Awgu Local Government Area, Enugu State Nigeria and was identified by a botanist.

2.2 Preparation of Plant Extract

Five hundred and twenty grams of fresh leaves of *psidium guajava* linn were washed with clean water, air dried under room temperature and afterwards ground into powdered using a mortar and a pestle, after which 300ml of distilled water were added and allowed to soak for an hour. It was then filtered and squeezed out using a cloth to get a greenish aqueous extract. The extract was stored in a refrigerator for use.

2.3 Collection and Acclimatization of Animals

Healthy 20 adult male albino rats weighing 70-130g were used for this study. Before starting the experiment, the animals were acclimatized to the laboratory conditions for 1 week at ambient temperature (24±25°C) and relative humidity (40-

60%). The animals were fed with standard laboratory diet and water. After administration the animal were starved overnight (24 hrs.) and subsequently sacrificed under mild Anastasia using chloroforms as an anesthetic agent, blood sample was collected from the albinos rat by cardiac puncture and the blood sample were put into a container with anti-coagulant (EDTA container). Qualitative analysis and heamatological parameters were measured.

2.4 Experimental Design

The twenty albino rats were allowed for several days to get adapted, they were placed in five cages, four rats in each cages group. The cages were marked (A, B, C, D). The extracts were administered to the animals at different doses after adaptation as follows; Group A - 200mg/kg body weight.

Group B - 400mg/kg body weight

Group C, 600mg/kg body weight

Group D 800mg/kg body weight

Group E Control Group

The administration was by oral intubation and lasted for fourteen (14) consecutive days.

2.5 Recording of Body Weight

Body weight was measured before and after the drug administration every day during the treatment in all groups.

2.6 Blood Sample Collection

After administration the animal were starved overnight (24 hrs.) and subsequently sacrificed under mild Anastasia using chloroforms as an anesthetic agent, blood sample was collected from the albinos rat by cardiac puncture and the blood sample were put into a container with anti-coagulant (EDTA container) and used for the analysis of the parameters Packed cell volume (PCV), Hemoglobin (Hb) and White blood cell (WBC).

2.7 Estimation of Heamatological Parameters

Hemoglobin concentration was measured by the method of [6], Packed cell volume was determine by the method of [7] and the White blood cell concentration was measured by the method of [5].

2.8 Statistical Analysis

The results gotten were subjected to statistical analysis using ANOVA (analysis of variance) to test for the significance and effectiveness of leaf extract solution to the experimental animals.

3. RESULTS

The result shown in Table 1 explains that the presence of Alkaloids is said to affect the central nervous system causing the decrease in the physical activities, feed and water intake. The presence of Flavonoid serves as an antioxidant that protects the white blood cell. The presence of Saponin in the constituent causes hemolysis. Tannin helps in cell growths in plants. The presence of Anthroquinone shows that the leaf is good for constipation because it is used for preparation of laxative. The presence of glycosidic are used in preparation of drugs that are used in treatment of heartbeat or heart failure [8].

The result shown from Table 2 explains that the increase values in the total white blood cell is due to the presence of flavonoid which serves as an antioxidant to protects the white blood cell. While the low value of hemoglobin and pack cell volume is due to the presence of saponin causes hemolysis.

4. DISCUSSION

The decrease in physical activities, feed and water intake in the groups administered the extract may be as a result of the chemical constituent of the extract. This chemical constituent may have affected the over all metabolic process taken place in the animals, same observation was reported by [9].

The decrease in the body weight of the test group may also be as result of the reported decrease in the feed and water intake [10].

Decrease in packed cell volume and hemoglobin, and increase in total white blood cell is observed in the groups A,B,C and D administered the extract may be as a result of the phytochemical constituent of the extract and suggest that the extract may be useful in the control and treatment of disorders of hematological system. The presence of flavonoid explains that guava leaf acts as blood tonic.

Table 1. Phytochemical composition of the extract

Phytochemical	Observation
Alkaloids	+
Flavonoid	++
Terpenes	+
Tannins	+++
Saponins	+
Glycosides	+
Anthraquinones	+++

Key

+ = slightly present

++ = moderately present

+++ = highly present

The qualitative phytochemical screening done and the result obtained shown in Table 1 above, from the table the aqueous extract of the leaves contains, Anthraquinone and Tannins in abundance, Flavonoid moderately and Alkaloids, Glycosides, Saponins and Terpenes were slightly present.

Table 2. Haemoglobin concentration, total white blood cell and packed cell volume of the rats

Groups	Haemoglobin (g/dl)	Total white blood cell (x108/L)	Packed cell (%)
Group A	14.61 ± 0.45	144.44 ± 7.48	63.13 ± 2.71
Group B	15.40 ± 0.39	120.87 ± 10.6	75.30 ± 2.41
Group C	17.36 ± 0.27	126.24 ± 9.07	87.01 ± 4.58
Group D	10.58 ± 1.12	87.96 ± 2.04	47.44 ± 3.39
Group E	11.18 ± 0.56	85.68 ± 3.03	51.91 ± 0.86

Values are mean ± SD; n = 4

Flavonoid are known to be antioxidant. Antioxidant scavenge free radicals thereby preventing oxidative stress and enhancing the normal functioning of the body cell [11]. This means that total white blood cell having the highest number explains that guava leaf serves as an antioxidant. The function of the white blood cell is to protect the body from foreign bodies, thereby in function to protect the body, the white blood cell tends to be strong thereby increasing in value but the intake of guava leaf increases the value of the white blood cell making it stronger due to the presence of flavonoid that are antioxidant.

5. CONCLUSION

The findings from this research suggest that leaves of guava may be useful in management and treatment of disorders of the hematological system. Since the administration increase the hemoglobin, packed cell volume and total white blood cell count.

ETHICAL APPROVAL

Animal Ethic committee approval has been taken to carry out this study.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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APPENDIX

Groups weight (gram)

Group 1

Green	back	180
Red	head	180
Plain		190

Group 2

Read	Head	320
Green	body	320
Green	back	320
Black	head	320

Group 3

Black	head	200
Green	body	200
Green	tail	200
Plain		200

Group 4

Red	Tail	165
Blue	body	190
Green	body	180
Plain		165

Group 5

Red	body	210
Red	head	400
Green	body	210
Plain		360

Volume of dose given in groups

Group 1

$$\frac{190 \times 0.1}{200} = 0.095$$

$$\frac{180 \times 0.1}{200} = 0.09$$

$$\frac{190 \times 0.1}{200} = 0.95$$

$$\frac{180 \times 0.1}{200} = 0.09$$

Group 2

$$\frac{320 \times 0.1}{400} = 0.08$$

$$\frac{320 \times 0.1}{400} = 0.08$$

$$\frac{320 \times 0.1}{400} = 0.08$$

$$\frac{320 \times 0.1}{400} = 0.08$$

Group 3

$$\frac{200 \times 0.1}{600} = 0.033$$

$$\frac{200 \times 0.1}{600} = 0.033$$

$$\frac{200 \times 0.1}{600} = 0.033$$

$$\frac{200 \times 0.1}{600} = 0.033$$

Group 4

$$\frac{165 \times 0.1}{800} = 0.020$$

$$\frac{190 \times 0.1}{800} = 0.023$$

$$\frac{180 \times 0.1}{800} = 0.022$$

$$\frac{165 \times 0.1}{800} = 0.020$$

Calculation of the concentration haemoglobin, white blood cell and pack cell volume

General formula for Standard Deviation $\sigma = \sqrt{\frac{\sum(x-x)^2}{N}}$

Group A			
	$(x-x)^2$	$x - \bar{x}$	
13.84 - 14.61	$-(0.77)^2$	0.5929	$\sqrt{0.200875}$
14.75 - 14.61	$(0.15)^2$	0.0225	
14.88 - 14.61	$(0.27)^2$	0.0729	
14.95 - 14.61	$(0.34)^2$	0.1156	
		0.8039 ÷ 4	0.448

Group B			
	$(x-x)^2$	$x - \bar{x}$	
15.60-15.402	0.04	0.04	$\sqrt{0.1498}$
15.94-15.042	0.54^2	0.2916	
15.01-15.402	-0.30^2	0.1521	
15.66-15.402	-0.342	0.1156	
		0.5993 ÷ 4	=0.387

Group C

17.44 – 1736	0.082	0.0064	
16.89 -17.36	-0.472	0.2209	$\sqrt{0.07495}$
17.50 – 17.36	0.142	0.0196	
17.59 – 17.36	0.232	0.0529	= 0.2737
			≅ 0.274
		0.2998 ÷ 4	

Group D

12.55 – 10.58	1.972	3.8809	
10.72 – 10.58	0.14 ²	0.0196	$\sqrt{1.247125}$
9.54 – 10.58	-0.04 ²	1.0816	= 1.1167
10.50 – 10.58	0.08 ²	0.0064	≅ 1.117
		4.9885 ÷ 4	

Group E

10.89 – 11.18	-0.29 ²	0.084	$\sqrt{0.31395}$
10.78 – 11.18	-0.4 ²	0.16	= 0.560
11.46 – 11.18	0.08 ²	0.0784	
11.06 – 11.18	0.42 ²	0.1764	
		1.2558 ÷ 4	

White Blood Cell

GROUP A

110.30 – 114.44	-4.142	17.1396	
104.22 - 114.44	-10.22 ²	104.4484	$\sqrt{56.1055}$
121.52 - 114.44	7.08 ²	50.1264	= 7.490
121.70 - 114.44	7.26 ²	52.7076	
		227.422 ÷ 4	

GROUP B

104.72 – 120-97	-16.25 ²	264.063	
118.50 – 120-97	-2.47 ²	6.1009	$\sqrt{111.348}$
130.26 – 120-97	9.29 ²	86.3041	10.55//
130.40 – 120-97	9.43 ²	99.9249	
		445.3929 ÷ 4	

GROUP C

133.40 – 126-24	7.16 ²	51.2656	
112.53– 126-24	-13.71 ²	187.9641	
129.01– 126-24	2.77 ²	7.6729	$\sqrt{65.1924}$
130.00– 126-24	117.76 ²	13.867	= 8.074
		260.7696 ÷ 4	

GROUP D

84.60-87.86	-3.36 ²	11.2896	
89.11-87.86	2.15 ²	4.6225	
88.52-87.86	0.56 ²	0.3136	$\sqrt{4.158825}$
88.60.87.96	0.64 ²	0.4096	= 2.039
		16.6353 ÷ 4	

GROUP E

92.77-85.68	7.08 ²	50.2681	
84.60-85.68	8.92 ²	71.5664	$\sqrt{64.50645}$
77.55-85.68	-813 ²	66.0969	8.0315//
77.80-85.68	-7.88 ²	62.0844	$\cong 8.032$
		258.026 ÷ 4	

Packed Cell Volume %

GROUP A

61.60 – 63.13	-1.53 ²	2.3409	
59.50 – 63.13	-3.63 ²	13.1769	$\sqrt{7.3277}$
65.22 – 63.13	2.08 ²	4.3681	= 2.7069
66.20– 63.13	3.07 ²	9.4249	$\cong 2.707$
		29.3108 ÷ 4	

GROUP B

74.01 – 75.30	-1.29 ²	1.6641	
72.18 – 75.30	-3.12 ²	9.7344	$\sqrt{5.801725}$
78.52 – 75.30	3.22 ²	10.3684	2.4086
76.50 – 75.30	1.2 ²	1.44	$\cong 2.409$
		23.2068 ÷ 4	

GROUP C

87.20 – 87.01	0.19 ²	0.0361	
79.53 – 87.01	7.48 ²	55.9504	$\sqrt{21.01005}$
90.25 – 87.01	3.24 ²	10.4876	4.5836
81.20 – 87.01	4.19 ²	17.5561	$\cong 4.584$
		84.0402 ÷ 4	

GROUP D

48.33-47.44	0.89 ²	0.7921	
52.61-47.44	5.17 ²	26.7289	$\sqrt{11.459175}$
44.33-47.44	-3.11 ²	9.6721	3.385//
44.50-47.44	-2.94 ²	8.6436	
		45.8367 ÷ 4	

GROUP E

50.60-51.91	-1.31 ²	1.7161	
52.18-51.91	0.27 ²	0.0729	$\sqrt{0.7455}$
51.84-51.91	-0.07 ²	0.0049	0.863
53.00-51.91	1.09 ²	1.1881	
		2.982 ÷ 4	

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