The Impact of *Citrus aurantium* Fruit Juice on Bodyweight and Haematological Parameters of Wistar Rats

ABSTRACT

Background: *Citrus aurantium* L. (Rutaceae), commonly known as bitter orange is widely consumed owing to its numerous therapeutic potentials.

Purpose: This study evaluated the impact of *Citrus aurantium* fruit (bitter orange) juice on bodyweight and haematological parameters of wistar rats.

Method: The animals were administered *Citrus aurantium* juice orally at three divided doses (2 ml/kg, 4ml/kg and 8ml/kg) for a period of three months and their bodyweight were monitored and taken weekly with the aid of electronic weighing balance while the haematological parameters were analyzed at the end of every month using standard methods.

Results: Result showed that administration of *C. aurantium* juice caused a significant ($p<0.05$) weight reduction in the experimental animals from the first week through the twelfth week. Result further showed significant increase ($p<0.05$) in haemoglobin, platelet, red blood cells (RBC) and white blood cell (WBC) counts of the animals with no significant difference in the mean corpuscular volume (MCV), the mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) levels.

Conclusion: The findings in this study have shown that oral administration of *C. aurantium* juice is capable of causing weight loss without any detrimental effect on the haematological parameters.

Keywords: *C. aurantium*, Bodyweight, Haematological parameters, Health.

1.0 INTRODUCTION

The use of fruits as a major constituent of diet for the populace is gaining popularity worldwide [1]. This is partly due to its nutritional and health benefits. Fruits are vital for their content in phytochemicals, antioxidants, vitamins, minerals and dietary fibre [2]. These are responsible for reducing the risk of development of health problems such as certain types of cancer, heart-related diseases, type 2 diabetes, obesity and constipation [3,1]. *Citrus aurantium* (bitter orange) is one of the promising fruits that has been known and consumed widely for its health promoting qualities [4].

Citrus aurantium L. (Rutaceae), commonly known as bitter orange is a polyembryonic species with greenish yellow smooth surfaced, thin-skinned fruits, and solid core at maturity with high acidic juice [5]. *C. aurantium* is believed to be native of Southeast Asia [6], and was carried by Arab traders to the Middle East and eventually came to Europe during the crusades, and are today cultivated in many countries of the world. *C. aurantium* is majorly used for food, refreshing drinks, tasty desserts and for seasoning meats, vegetables, salads and sauces [7]. The fruits of *C. aurantium* are sources of flavonoid-type compounds with various biological effects [8]. Due to the abundance of health-giving secondary metabolites, *C. aurantium* is also used for the treatment of several ailments such as anxiety [9], lung and prostate cancers [10].

The extract of the fruit and peel of *C. aurantium* has been widely used in weight loss dietary supplements and in sports performance products [11]. Specifically, after the ban of the sale of all ephedra-containing supplements by the Food and Drug Administration (FDA) in 2004, *C.*

aurantium has further gained popularity as a safe alternative to Ephedra in herbal weight loss products [12]. Some Medicinal plants such as *Telfairia occidentalis* leaves and *Mucuna pruriens* are capable of improving the haematological parameters of experimental animals [13]. *C. aurantium* has been used as an ingredient of the dietary supplements marketed for weight loss aid due to its claimed effects on metabolism, increasing the basal metabolic rate and lipolysis, and as appetite suppressant [14,15]. Despite the quantum of research on the weight-loss potential of *C. aurantium*, there is a dearth of literature reports on its effects on haematological parameters. It is therefore necessary that research focuses on the effects of medicinal fruits especially *C. aurantium* on haematological parameters. This study was therefore aimed at exploring the impact of *C. aurantium* fruit on bodyweight and haematological parameters of Wistar rats.

2.0 MATERIALS AND METHODS

2.1 Sample Collection and Identification

The *Citrus aurantium* fruits *were* collected from Mgbakwu, Awka North Local Government Area, Anambra State, Nigeria. The sample was identified by a taxonomist in the Department of Botany, Nnamdi Azikiwe University, Awka. The voucher number as deposited in the herbarium of Nnamdi Azikiwe University, Awka is NAUH 197[A].

2.2 Grouping of Experimental Animals

A total of 20 rats of both sex weighing between 150g and 170g were purchased from Chris Experimental Animal Farm and Research Laboratory, Awka, Anambra State and used for the experiment. They were maintained and housed in cages under standard environmental conditions (27°C±3°C, 12-hour light/dark cycle) in the Department of Applied Biochemistry Laboratory, Nnamdi Azikiwe University, Awka. The experimental animals were acclimatized with the environment for one week and fed with Vital grower's mash pellets and water *ad libitum*. At the end of the one-week acclimatization period, the animals were weighed, grouped and administered the freshly squeezed juice of *C. aurantium* for a period of three (3) months as shown in table 1 below.

Comment [j1]: Don't think that an extract would have given better comparable results as different climate and soils may give different results??

Table 1: Grouping of Experimental Animals

Group	Dosage
A	Normal Control (Administered distilled water)
B	2 ml/kg bodyweight of *Citrus aurantium* fruit juice
C	4 ml/kg bodyweight of *Citrus aurantium* fruit juice
D	8 ml/kg bodyweight of *Citrus aurantium* fruit juice

2.3 Determination of Bodyweight

The weight of the experimental animals was checked using an electronic weighing scale. The weights of the rats were monitored before, during, and after the experiment to know whether the continuous administration of lime juice caused a noticeable increase or decrease in bodyweight. Percentage bodyweight was also calculated using the formular below:

Comment [j2]: Lime juice is introduced for the first time.

$$\text{Percentage weight} = \frac{Weekly\ weight - Initial\ weight}{Initial\ weight} \times \frac{100}{1}$$

2.4 Haematological Analysis

Haematological parameters were determined using automated haematology analyzer (Mindray-BC-5300). The haematological parameters that were analysed include Haemoglobin (HGB), Packed Cell Volume (PCV), Red Blood Cells (RBC), Platelets (PLT), Mean Corpuscular Volume (MCV), Mean Corpuscular Haemoglobin (MCH), Mean Corpuscular Haemoglobin Concentration (MCHC), White Blood Cells (WBC), Neutrophils (NEUT), Lymphocytes (LYMPH), Monocytes (MON), Eosinophils (EOS), Basophils (BAS).

2.5 Statistical Analysis

Data obtained from the experiments were analyzed using the Statistical Package for Social Sciences software for windows version 23 (SPSS Inc., Chicago, Illinois, USA). All the data collected were expressed as Mean ± SEM. Statistical analysis of the results obtained were performed by using ANOVA Tests to determine if significant difference exists between the mean of the test and control groups. The limit of significance was set at $p<0.05$.

Comment [j3]: There was only one control group. Is thare any other group that was not included in Table 1?

3.0 RESULTS

Table 2.0 represents the effect of oral administration of *C. aurantium* fruit to the experimental animals. Results showed that administration of *C. aurantium* fruit causes a significant decrease in the body weight of animals administered varying doses (2 ml/kg, 4 ml/kg and 8 ml/kg) from week 1 to week 12 when compared with the control group.

Table 2.0: Weekly Bodyweight of rats administered different doses of *C. aurantium* fruit juice.

Groups	Weight (g) Week 0	Weight (g) Week 1	Weight (g) Week 2	Weight (g) Week 3	Weight (g) Week 4	Weight (g) Week 5	Weight (g) Week 6	Weight (g) Week 7	Weight (g) Week 8	Weight (g) Week 9	Weight (g) Week 10	Weight (g) Week 11	Weight (g) Week 12
Normal Control	156.23 ±1.82	165.20 ±1.57	172.17 ±1.42a	180.65 ±1.93a	189.39 ±0.82a	196.57 ±1.31a	203.24 ±2.61a	213.72 ±1.30a	220.11 ±1.80a	228.26 ±2.05a	237.32 ±1.53a	248.01 ±1.70a	256.33 ±1.15a
2ml/kg bw. of *Citrus aurantium* fruit juice	153.50 ±1.09	158.35 ±1.23	165.91 ±1.16	170.64 ±2.34	176.21 ±1.25a	181.57 ±2.73a	187.29 ±1.19a	190.32 ±2.33a	194.10 ±2.86a	195.73 ±1.20a	195.12 ±2.03a	192.34 ±2.76a	183.52 ±2.49a
4ml/kg bw. of *Citrus aurantium* fruit juice	157.31 ±1.96	161.52 ±1.82	168.46 ±1.73	173.32 ±0.99	177.56 ±1.56	179.83 ±2.30a	186.58 ±1.07a	188.23 ±2.00a	190.03 ±1.32a	185.65 ±2.19a	172.82 ±1.72	170.63 ±2.03	163.45 ±1.36
8ml/kg bw. of *Citrus aurantium* fruit juice	152.83 ±1.36	159.75 ±1.54	165.71 ±1.01	171.13 ±1.40	176.63 ±1.11	179.21 ±2.08a	182.95 ±1.35a	183.18 ±2.57a	180.32 ±1.45a	176.90 ±1.27	171.23 ±2.12	163.83 1.60	157.02 ±2.53

[a]Significant increase with respect to week 0; [b]Significant decrease with respect to week 0.

The results of the percentage change in body weight are presented in table 2.1. Results showed that administration of *C. aurantium* fruit causes a significant decrease in the bodyweight of animals administered varying doses (2 ml/kg, 4 ml/kg and 8 ml/kg) from week 1 to week 12 when compared with the control group.

Table 2.1: Percentage (%) increase/decrease in bodyweight of rats administered different doses of *C. aurantium* fruit juice.

Groups	Week 0	Week 1	Week 2	Week 3	Week 4	Week 5	Week 6	Week 7	Week 8	Week 9	Week 10	Week 11	Week 12
Normal Control	-	5.76	10.18	15.62	21.19	25.80	30.09	36.81	40.91	46.09	52.05	58.77	64.08
2ml/kg bw. of *Citrus aurantium* fruit juice	-	3.13	8.08	11.14	14.79	18.24	33.72	36.80	40.65	42.21	41.60#	25.28#	19.54#
4ml/kg bw. of *Citrus aurantium* fruit juice	-	2.67	7.06	10.17	12.84	14.30	18.56	19.64	20.79	17.99#	9.85#	8.46#	3.88#
8ml/kg bw. of *Citrus aurantium* fruit juice	-	4.52	8.44	11.98	15.58	17.28	19.70	19.83	18.00#	15.77#	12.04#	7.20#	2.75#

shows percentage decrease compared with the bodyweight before it.

Table 2.2 presents the effect of oral administration of *C. aurantium* on the haemoglobin concentration of the experimental animals. Results showed a significant increase (p<0.05) in the haemoglobin (HGB) level of the test groups when compared with the control group.

Table 2.2: Haemoglobin concentration of rats administered different doses of *C. aurantium* fruit juice.

	HGB (g/dl)			
	Initial	**1st Month**	**2nd Month**	**3rd Month**
Normal Control	13.53±0.02	12.72±0.03	13.91±0.01	13.63±0.05
2ml/kg bw. of *Citrus aurantium* fruit juice	13.81±0.08	15.82±0.05	13.96±0.31	22.94±0.03
4ml/kg bw. of *Citrus aurantium* fruit juice	12.92±0.03	13.80±0.02	13.73±0.07	15.35±0.03
8ml/kg bw. of *Citrus aurantium* fruit juice	14.25±0.01	14.13±0.03	13.34±0.03	12.41±0.06

Table 2.3 presents the effect of oral administration of *C. aurantium* on the haematocrit level of the experimental animals. Results showed a significant increase (p<0.05) in the haematocrit level of the group administered 2 ml/kg at the third month of administration while a significant decrease of haematocrit level was observed in group administered 8 ml/kg at the third month of oral administration.

Table 2.3: Haematocrit of rats administered different doses of *C. aurantium* fruit juice.

	HCT (%)			
	Initial	**1st Month**	**2nd Month**	**3rd Month**
Normal Control	46.33±1.78	44.20±1.21	46.93±0.33	45.27±1.48
2ml/kg bw. of *Citrus aurantium* fruit juice	46.72±3.12	48.12±0.90	46.32±2.51	66.73±0.72m
4ml/kg bw. of *Citrus*	43.53±0.67	43.91±0.88	45.17±0.68	47.15±2.10

***aurantium* fruit juice**				
8ml/kg bw. of *Citrus* *aurantium* fruit juice	47.16±1.23	45.32±0.62	42.60±1.25	37.73±0.36n

[i]Significant increase with respect to 1st Month; [j]Significant decrease with respect to 1st Month; [k]Significant increase with respect to 2nd Month; [l]Significant decrease with respect to 2nd Month; [m]Significant increase with respect to 3rd Month; [n]Significant decrease with respect to 3rd Month.

Table 2.4 presents the effect of oral administration of *C. aurantium* on the Mean Corpuscular Volume (MCV) of the experimental animals. Results showed no significant difference in the MCV of the experimental animals**.**

Table 2.4: Mean Corpuscular Volume of rats administered different doses of *C. aurantium* fruit juice.

	MCV (fl)			
	Initial	**1st Month**	**2nd Month**	**3rd Month**
Normal Control	58.54±1.58	56.70±0.25	56.93±0.52	58.32±2.35
2ml/kg bw. of *Citrus* *aurantium* fruit juice	55.72±0.76	55.87±0.83	55.22±2.31	55.12±0.89
4ml/kg bw. of *Citrus* *aurantium* fruit juice	59.34±2.63	56.02±0.16	58.84±2.35	59.74±1.35
8ml/kg bw. of *Citrus* *aurantium* fruit juice	57.28±0.56	57.62±1.10	56.12±1.21	57.93±2.67

Table 2.5 presents the effect of oral administration of *C. aurantium* on the Mean Corpuscular Haemoglobin (MCH) of the experimental animals. Results showed no significant difference in the MCH of the experimental animals**.**

Table 2.5: Mean Corpuscular Haemoglobin of rats administered different doses of lemon juice.

	MCH (pg)			
	Initial	**1st Month**	**2nd Month**	**3rd Month**
Normal Control	18.74±0.01	18.50±0.02	19.51±0.01	19.20±0.07
2ml/kg bw. of *Citrus* *aurantium* fruit juice	18.21±0.02	18.15±0.01	18.30±0.01	18.92±0.03
4ml/kg bw. of *Citrus* *aurantium* fruit juice	19.53±0.04	18.62±0.01	19.32±0.04	19.47±0.01
8ml/kg bw. of *Citrus* *aurantium* fruit juice	19.13±0.01	19.19±0.01	19.46±0.01	19.04±0.02

Table 2.6 presents the effect of oral administration of *C. aurantium* on the Mean Corpuscular Haemoglobin Concentration (MCHC) of the experimental animals. Results showed no significant difference in the MCHC of the experimental animals**.**

Table 2.6: Mean Corpuscular Haemoglobin Concentration f rats administered different doses of *C. aurantium* fruit juice.

	MCHC (g/dl)			
	Initial	**1st Month**	**2nd Month**	**3rd Month**
Normal Control	31.04±0.21	32.52±0.03	33.61±0.05	34.80±0.01
2ml/kg bw. of *Citrus aurantium* fruit juice	31.54±0.02	31.71±0.05	33.27±0.02	34.45±0.02
4ml/kg bw. of *Citrus aurantium* fruit juice	33.62±0.02	31.85±0.08	32.63±0.01	32.47±0.06
8ml/kg bw. of *Citrus aurantium* fruit juice	31.81±0.41	32.07±0.04	31.75±0.32	32.83±0.03

Table 2.7 presents the effect of oral administration of *C. aurantium* on the Red Blood Cell count (RBC) of the experimental animals. Results showed no significant difference in the RBC of the experimental animals**.**

Table 2.7: Red Blood Cell count of rats administered different doses of *C. aurantium* fruit juice.

	RBC (x $10^{12/L}$)			
	Initial	**1st Month**	**2nd Month**	**3rd Month**
Normal Control	7.62±0.73	7.58±0.02	6.91±0.09	7.53±0.05
2ml/kg bw. of *Citrus aurantium* fruit juice	7.56±0.34	7.25±0.04	7.62±0.01	12.10±0.01
4ml/kg bw. of *Citrus aurantium* fruit juice	7.01±0.12	7.14±0.01	7.18±0.04	7.89±0.05
8ml/kg bw. of *Citrus aurantium* fruit juice	6.95±0.25	7.71±0.09	6.63±0.13	6.52±0.06

Table 2.8 presents the effect of oral administration of *C. aurantium* on the Platelet count of the experimental animals. Results showed a significant decrease in the Platelet count of the experimental animals administered 2 ml/kg and 8 ml/kg while group administered 4 ml/kg showed otherwise at the third month of oral administration.

Table 2.8: Platelet count of rats administered different doses of *C. aurantium* fruit juice.

	PLT (x $10^{9/L}$)			
	Initial	**1st Month**	**2nd Month**	**3rd Month**
Normal Control	821.24±4.64	753.55±6.10j	802.32±3.88	782.93±2.92n
2ml/kg bw. of *Citrus*	760.52±3.33	698.71±2.95j	686.56±3.75l	609.31±3.11n

***aurantium* fruit juice**				
4ml/kg bw. of *Citrus aurantium* fruit juice	815.28±6.02	775.32±4.63j	713.20±4.12l	890.54±2.36m
8ml/kg bw. of *Citrus aurantium* fruit juice	917.43±2.11	830.20±6.00j	826.52±4.31l	751.21±3.23n

[i]Significant increase with respect to 1st Month; [j]Significant decrease with respect to 1st Month; [k]Significant increase with respect to 2nd Month; [l]Significant decrease with respect to 2nd Month; [m]Significant increase with respect to 3rd Month; [n]Significant decrease with respect to 3rd Month.

Table 2.9 presents the effect of oral administration of *C. aurantium* on the White Blood Cell (WBC) count of the experimental animals. Results showed a significant decrease in the WBC of the experimental animals administered 2 ml/kg while groups administered 4 ml/kg and 8 ml/kg showed no significant difference at the third month of oral administration.

Table 2.9: White Blood Cell count of rats administered different doses of *C. aurantium* fruit juice.

	WBC (x $10^{9/L}$)			
	Initial	**1st Month**	**2nd Month**	**3rd Month**
Normal Control	9.53±0.85	10.26±0.92	9.81±0.45	10.73±0.69
2ml/kg bw. of *Citrus aurantium* fruit juice	10.82±0.61	10.56±0.03	7.11±0.12l	6.82±0.08n
4ml/kg bw. of *Citrus aurantium* fruit juice	7.92±0.99	10.13±1.02	10.64±0.51k	6.69±0.42
8ml/kg bw. of *Citrus aurantium* fruit juice	9.51±0.68	10.35±0.93	8.92±0.17	11.36±0.38

[i]Significant increase with respect to 1st Month; [j]Significant decrease with respect to 1st Month; [k]Significant increase with respect to 2nd Month; [l]Significant decrease with respect to 2nd Month; [m]Significant increase with respect to 3rd Month; [n]Significant decrease with respect to 3rd Month.

Table 2.10 presents the effect of oral administration of *C. aurantium* on the Neutrophil count of the experimental animals. Results showed a significant increase in the neutrophil count of the experimental animals administered 2 ml/kg and 4 ml/kg while group administered 8 ml/kg showed no significant difference at the third month of oral administration.

Table 2.10: Neutrophils of rats administered different doses of *C. aurantium* fruit juice.

	NEU (%)			
	Initial	**1st Month**	**2nd Month**	**3rd Month**
Normal Control	4.20±0.11	4.83±0.41	5.03±0.65	5.65±0.12
2ml/kg bw. of *Citrus aurantium* fruit juice	4.83±0.54	4.77±0.32	4.91±0.06	11.55±1.12m
4ml/kg bw. of *Citrus aurantium* fruit juice	5.25±0.15	4.65±0.36	6.38±0.78	7.17±0.28m
8ml/kg bw. of *Citrus aurantium* fruit juice	4.51±0.02	4.11±0.15	4.25±0.23	3.41±0.04

[i]Significant increase with respect to 1st Month; [j]Significant decrease with respect to 1st Month; [k]Significant increase with respect to 2nd Month; [l]Significant decrease with respect to 2nd Month; [m]Significant increase with respect to 3rd Month; [n]Significant decrease with respect to 3rd Month.

Table 2.11 presents the effect of oral administration of *C. aurantium* on the Lymphocytes count of the experimental animals. Results showed no significant difference in the lymphocytes count of the experimental animals at the third month of oral administration.

Table 2.11: Lymphocytes of rats administered different doses of *C. aurantium* fruit juice.

	LYM (%)			
	Initial	**1st Month**	**2nd Month**	**3rd Month**
Normal Control	97.63±2.46	96.10±1.50	95.82±2.14	98.20±2.87
2ml/kg bw. of *Citrus aurantium* fruit juice	85.08±1.15	87.62±2.57	90.27±2.87	87.62±1.52
4ml/kg bw. of *Citrus aurantium* fruit juice	92.73±2.21	85.38±2.09	88.23±1.24	90.90±2.59
8ml/kg bw. of *Citrus aurantium* fruit juice	92.57±1.27	97.35±2.31	96.15±0.93	95.82±1.44

Table 2.12 presents the effect of oral administration of *C. aurantium* on the Monocyte count of the experimental animals. Results showed a significant increase in the monocyte count of the experimental animals administered 2 ml/kg and 4 ml/kg while group administered 8 ml/kg showed no significant difference at the third month of oral administration.

Table 2.12: Monocytes of rats administered different doses of *C. aurantium* fruit juice.

	MON (%)			
	Initial	**1st Month**	**2nd Month**	**3rd Month**
Normal Control	0.00±0.00	0.20±0.00i	0.13±0.00k	0.16±0.00m
2ml/kg bw. of *Citrus aurantium* fruit juice	0.00±0.00	0.12±0.00i	0.00±0.00	0.27±0.00m
4ml/kg bw. of *Citrus aurantium* fruit juice	0.27±0.00	0.43±0.00i	0.71±0.01k	0.93±0.02m
8ml/kg bw. of *Citrus aurantium* fruit juice	0.12±0.00	0.15±0.00	0.29±0.00k	0.14±0.00

[i]Significant increase with respect to 1st Month; [j]Significant decrease with respect to 1st Month; [k]Significant increase with respect to 2nd Month; [l]Significant decrease with respect to 2nd Month; [m]Significant increase with respect to 3rd Month; [n]Significant decrease with respect to 3rd Month.

Table 2.13 presents the effect of oral administration of *C. aurantium* on the Eosinophil of the experimental animals. Results showed a significant increase in the eosinophil of the experimental animals administered 2 ml/kg, 4 ml/kg and 8 ml/kg at the third month of oral administration.

Table 2.13: Eosinophils of rats administered different doses of *C. aurantium* fruit juice.

	EOS (%)			
	Initial	**1st Month**	**2nd Month**	**3rd Month**
Normal Control	0.31±0.00	0.42±0.00	0.20±0.00l	0.30±0.00
2ml/kg bw. of *Citrus aurantium* fruit juice	0.33±0.00	0.28±0.00	0.33±0.00	0.74±0.00m
4ml/kg bw. of *Citrus aurantium* fruit juice	0.21±0.00	0.62±0.00i	0.13±0.00	1.11±0.01m
8ml/kg bw. of *Citrus aurantium* fruit juice	0.57±0.00	0.32±0.00j	0.57±0.01	0.76±0.00m

[i]Significant increase with respect to 1st Month; [j]Significant decrease with respect to 1st Month; [k]Significant increase with respect to 2nd Month; [l]Significant decrease with respect to 2nd Month; [m]Significant increase with respect to 3rd Month; [n]Significant decrease with respect to 3rd Month.

Table 2.14 presents the effect of oral administration of *C. aurantium* on the Basophil of the experimental animals. Results showed a zero count for Basophil in all the experimental animals.

Table 2.14: Basophils of rats administered different doses of *C. aurantium* fruit juice.

	BAS (%)			
	Initial	**1st Month**	**2nd Month**	**3rd Month**
Normal Control	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
2ml/kg bw. of *Citrus aurantium* fruit juice	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
4ml/kg bw. of *Citrus aurantium* fruit juice	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
8ml/kg bw. of *Citrus aurantium* fruit juice	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00

4.0 DISCUSSION

Over the years, phytomedicine have shown an outstanding role in new drug discovery scaffolds [16]. Both in the crude form as well as pure chemical entities, a large population around the globe are getting therapeutic benefits from them [17]. Overweight and obesity are major risk factors for a number of chronic diseases, including cardiovascular diseases such as heart disease and stroke. Hence, controlling excess weight is crucial for a healthy living.

As depicted in table 2.0, administration of *C. aurantium* to the experimental animals at varying doses (2ml/kg, 4ml/kg and 8ml/kg) caused a significant decrease in their bodyweight from the first week through the twelfth week (table 2.1). Administration of *C. aurantium* to the animals could plausibly have caused a delay in the basal metabolic rate of the animals and subsequently suppress their appetite which in turn impact on their bodyweight. Stohs *et al*. [14] asserted that administration of *C. aurantium* impacted the lipolysis as well as basal metabolic rate of the experimental animals. Alisa *et al*. [18] reported that oral administration of aqueous extract of Kuding leaves causes a significant decrease in bodyweight of albino rats.

The assessments of haematological parameters are useful guide to ascertaining the effect of foreign substances including plant extracts in a biological system [19]. They are used to determine possible alterations in the levels of biomolecules such as enzymes, metabolic products,

haematology, normal functioning and histopathology of the organs [20]. Tables 2.2 to 2.14 showed the effect of oral administration of *C. aurantium* juice on Haematological parameters in albino rats. As depicted in table 2.2, there was significant increase ($p< 0.05$) in the haemoglobin (HGB) level of the test groups when compared with the control group. Haemoglobin is the iron-containing oxygen-transport metalloprotein in the red blood cells of almost all vertebrates as well as the tissues of some invertebrates. They function primarily in the circulation of oxygen from the lungs to the rest of the body. Administration of *C. aurantium* juice to the experimental animals could therefore be said to have improved oxygen transport into the red blood cells of these animals and consequently impact on their health. However, there was no significant difference ($p<0.05$) in the mean corpuscular volume (MCV), the mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC).

Comment [j4]: This statement is difficult to appreciate especially when tere was no change in the MCH and MCHC

Red blood cells (RBCs), also referred to as red cells are the most common type of blood cell. As depicted in table 2.7, there was a significant increase ($p<0.05$) in the test group at the third month compared with the initial. The result of this study is similar to the report of Okon *et al*. [21] and Nwankpa *et al*. [22] in their separate studies. RBCs are known to functions primarily in picking up inhaled oxygen from the lungs and transport it to the body's tissues. Oladejo and Osukoya [19] reported that RBCs also pick up about 24% carbon dioxide waste at the tissues and transport it to the lungs for exhalation. Hence, administration of *C. aurantium* juice to the experimental animals could therefore improve oxygen circulation in a biological system.

Platelet is another haematological parameter that is of note. As shown in table 2.8, there was significant increase ($p<0.05$) in the group administered 4ml/kg bodyweight of *C. aurantium* juice at the third month compared with the initial stage. Platelets are made in the bone marrow, the sponge-like tissue inside the bones that contains the stem cells. They are known to help the body form clots to stop bleeding during injury.

Comment [j5]: So????

Tables 2.9 to 2.14 showed the results for white blood cells and its differentials (Neutrophils, Lymphocytes, Monocytes, Eosinophils and Basophils) respectively. There was a significant increase ($p<0.05$) in the white blood cell count, neutrophils, monocytes and eosinophils in the test groups at the third month compared with the control group.

WBC and its differentials are measurable indices of the blood, which can be used to evaluate hematopoietic function [23]. They are essential for the protection of the animal against foreign invaders and help to stimulate cytokine erythropoietin which subsequently stimulates blood cell synthesis [19]. Oral administration of *C. aurantium* to the experimental animals may plausibly have stimulated cytokine erythropoietin to stimulate blood cell synthesis.

Comment [j6]: Which blood cells?

5.0 CONCLUSION

The result from this study demonstrated that the oral administration of *C. aurantium* juice is capable of causing weight loss without any detrimental effect on the haematological parameters. Hence, the fruit could be useful in the preparation of weight loss formulas and consequently improve general wellbeing.

Comment [j7]: This could not be concluded from this study.

CONSENT

This is not applicable here.

ETHICAL APPROVAL

All experiments were inspected and approved by the Nnamdi Azikiwe University Animal Research Ethics Committee (NAU-AREC).

COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

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