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Research Paper

Determination of Anemia control of Moringa oleifera leaf Extract and Oral Iron Supplements in Wistar Rats

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ABSTRACT:
Anemia remains one of the most difficult public health problems to manage, of which nutritional anaemia is the leading cause of anaemia worldwide. Iron deficiency anaemia (IDA), being the most serious nutritional problem globally. This study aims at evaluating the hematopoietic effect of Moringa oleifera leaf extract (MOLE) in comparison to oral iron supplements in rats. Twenty five adult male rats were randomly divided into 5 groups of (n=5). The control was fed with 10 mls/kg/BW of distilled water and iron free animal feed. Second group received 250 mg/kg/BW of MOLE and normal animal feed third and 4th received 500 mg/kg BW and 1000 mg/kg BW of MOLE plus animal feed respectively. Last group received animal feeds containing 100mg/KG/BW of (FeSo4) iron supplement alone. The result showed a dose dependent increase in the mean cell volume, mean cell hematocrit, mean cell hematocrit concentration, hemoglobin and pack cell volume. When compared to the standard, there was a slight increase in hematologic parameters. There was also slight increase in weight of rats fed with (MOLE), with slightly higher increase in the group that received oral iron supplement. The findings thus indicate that MOLE, are as potent as iron supplements in management of anaemia.

***KEY WORDS**: Moringa oleifera leaf extract, Anemia, Oral iron Supplements, Hematopoiesis*

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I. INTRODUCTION:

Anemia is a condition in which the number of red blood cells, and consequently their oxygen-carrying capacity is insufficient to meet the body's physiologic needs (Dacie, 2020).The word "Anemia" explain itself an" mean without and "Anemia "mean red blood cells. If you have anemia then you feel fatigue and weakness all the day.

Anemia is a public health problem globally, and its prevalence has being continuously on the rise. It has being known to pose different consequences both economically and on general health care (WHO, 2024). World health Organization (WHO) estimates that more than 2 billion people, or one quarter of the world's population suffer from anemia (WHO, 2025).

Between the year 2023 and 2024, the prevalence rose to 47.3% in women of reproductive age and 71% in children under five years of age. A study by (Alo and Adetuji, 2025), found 52.7% 0f adult respondents females and a higher prevalence of anaemia (39%) compared to males (17.5%). Pregnant women exceeded 50%.

Factors that cause anemia include helminthic infections, malaria, nutritional deficiencies and chronic disease conditions. Also the poor socio economic state in developing nations was seen to play a role as lower educational levels and rising unemployment rates were strong predictors.

Iron deficiency is said to be the most common cause of anemia globally, constituting 50% of global cause (Stevens and Finucane, 2013).

Other nutritional deficiencies include folate, vitamin B12 and vitamin A and C production (Dacie, 2020). Sickle cell disease and thalassemia are also indicated. In women of child bearing age, reproductive health is affected, leading to maternal and infant mortality, low birth weight babies and complications at delivery (Sun and Wu, 2021).

Diverse measures have being undertaken to curb the growing prevalence of nutritional anemia, with (Fe) tablets supplements given to women of reproductive age and pregnant women at ante-natal clinic visits (Fauziandari ,2019). Supplements have also being added to correct deficiency of essential vitamins, micro and macro minerals.

Healthy iron rich diet however, remains vital, consisting of vital nutrients that aid in adequate nutrition. Here, an adequate caloric intake of carbohydrate, proteins, iron, folic acid, vitamin A, Zinc, Iodine is important (Aminah, 2015).

Moringa oleifera is known to have many benefits and has been recognized globally and recommended for pregnant women, infants and children for correcting nutritional anemia and malnutrition (Aminah, 2015). It contains 28.2mg of iron, also rich in protein, Vitamin A, Vitamin C, potassium and calcium (Aminah, 2015). The aim of this article is to evaluate and compare the hematopoietic effect of Moringa oleifera leaves extract and oral iron supplements.

II. Materials and Methods:

2.1. Plant Materials:

Fresh leaves of M. oleifera were collected from a natural habitat in Umuahia, South East Nigeria, and the botanical identity of the plant was confirmed by Dr. Jimoh Abiola At the herbarium unit of the department of plant science technology, Michael Okpara University of Agriculture, Umudike, south east Nigeria, where a voucher specimen was deposited for future reference with voucher number MOUAU/VPP/10124.

2.2. Preparation of Extract:

The leaves were rinsed with distilled water to remove dust particles. They were dried to a constant weight and pulverized with a blender. A portion of the powdered leaves was cold macerated with ethanol for 24 hours and filtered with (whatman size number 1) filter paper, to obtain MOLE. The extract was concentrated over a water bath at 40^0 Celsius and stored in a refrigerator.

2.3. Animal Source:

25 healthy adult male wistar rats weighing (180-200g) were sourced from the animal house department of vetinary medicine, Michael Okpara University of Agriculture Umudike, Abia Sate Nigeria, and were used for this study. They were maintained in stainless steel cages and given clean water and pelleted rat feed. All experiments were performed according to the principles of laboratory animal care (NIH and NIPRD). Ethical clearance was also gotten.

2.4: DRUGS and Chemicals:

Oral iron tablets 200mg, produced by Chemiron International Limited, Lagos Nigeria, were purchased from Zigar Pharmacy Aba, Nigeria. Absolute ethanol (99.5%) was purchased from the local market in Aba, Nigeria.

2.5. Acute Toxicity Study:

The oral median lethal dose (LD50) of the extract was determined in the rats according to method described by Lorke (1983); the study was carried out in two phases. In the first phase, nine rats were randomized into three groups 10, 100 and 100 mg/kg body weight. The rats were kept under the same condition and observed for signs of toxicity, change in behavior or deaths and recorded.

Based on the result of initial phase, three other rats were administered with 1600, 2900 and 5000 mg/kg/BW respectively. The rats were monitored closely for 24 hours for sings of toxicity or mortality. The result obtained in the 2^{nd} phase was used to calculate the LD50.

2.6. Experiment Protocol:

5 groups of 5 male adult wistar rats were involved in this study and were weighed at start.

Group 1: control group, received 10mls /kg /BW of distilled water orally and fed with normal animal feed.

Group 2: Received 250 mg/kg/BW of MOLE via orally, and also fed with normal animal feed.

Group 3: Received 500 mg/kg/BW of MOLE orally, and fed with normal animal feed.

Group 4: received 1000 mg/kg/BW of MOLE orally, and fed with normal animal feed.

Group 5 (STANDARD GROUP): received 100mg/kg/BW of FeS04 tablets in animal feed alone.

At the end of the 28 day treatment, animals were anaesthetized and killed; blood samples were collected via ocular puncture, put into anti-coagulated tubes to access haematological profiles Hb, MCV, Hematocrit, MCHC and MCH.
Animals were also weighed.

2.7. SERUM BIOCHEMISTRY:
2.7.1. Hematological Profile:
Hemoglobin, mean cell volume, mean cell hemoglobin, hematocrit and mean cell hemoglobin concentration were measured with an automated hematology analyzer (Mind ray Bc-6800).
2.8. Statistical Analysis:
Data generated were analyzed using statistical package for social science (SPSS) version 23. Student T- test and analysis of variance (ANOVA) was used for comparing values of measured parameters between control and experimental groups, values $P < 0.05$ was considered statistically significant. Data collected was expressed as mean ± (SEM) standard error of mean.

2.9 RESULTS:
2.9.1. Acute Toxicity Studies:
MOLE did not produce any sign of toxicity or mortality at all to the doses administered orally. The oral median lethal dose (LD50) MOLE was therefore estimated to be greater than 500mg/kg weight in the rats.
2.9.2. Effect of MOLE administration and supplementary iron tablets on Haematological profile:
From table 3.1, it was indicated that there was a dose dependent significant increase in all the mean hematological parameters, (Hb, MCV, MCHC, HCT and MCH) in the rats fed with ethanol MOLE, when compared with the control group at P value < 0.05. Similarity was also seen in comparison with the standard group with P value < 0.05.
2.9.3. Effect of MOLE administration on weight of study rats after 4 weeks:
From table 3.2, it showed a significant increase in the mean weight of the rats fed with MOLE, with rats in the standard group weighing slightly higher, with P value < 0.05.

Table 2.1: Effect of MOLE and Supplementary oral Fe tablets on Haematological profile after 4 weeks

Haem profile	IU	groups					
		K	D250	D500	D 1000	100mg/kg/ bw FeSo4	P value
Hb	g/dl	11.30 ± 0.22	13.33± 0.31	13.93 ± 0.16	14.46 ± 0.80	15.3 ± 0.04	0.20
PCV	%	35.3 ± 1.53	39.67 ± 1.52	39.67 ± 1.52	4 2.67± 1.53	45.67± 1.53	0.002
MCV	FL	48.18 ± 2.34	55.65 ± 1.2	55.65 ± 1.20	51.71 ± 0.83	56.25± 1.15	0.002
MCH	g/dl	13.71 ± 0.53	55.65 ± 1.2	15.10 ± 0.32	14.68 ±0.26	16.85± 0.15	0.30
MCHC	g/dl	20.07 ± 0.38	20.07 ± 0.38	26.91± 0.83	27.71 ± 0.68	31.85 ± 0.55	0.001

Data represented as mean ± S.E.M. Data was analyzed by one way ANOVA. n = (5). $P < 0.05$, thus statistically significant as compared to control.

Table 2.2: Effect of MOLE on Percentage weight gain of Study rats at Day 0 and 28

Group	treatment	Initial weight (g)	Final weight (g)	%weight gain	P value
K	10mls/kg/bw distilled water	134.6 ± 6.20		32.5%	0.001
250mg/kg/BW	250 mg/kg/bw mole	136.1 ± 4.05		8.64%	0.002
500mg/kg/bw	500 mg/kg/bw mole	132.1 ± 4.0		5.63%	0.01
1000mg//kg bw	1000 mg/kg/bw mole	135.2 ± 5.4	142.7 ± 4.1	7.62%	0.002
100 mg/kg/bw FeSo4	100 mg/kg/bw Fe tablets	135.2 ± 5.4	144.1 ± 6.5	6.62%	0.002

Data is presented in mean± S.E.M. (P value< 0.05).

FIGURE 2.1: MEAN Hemoglobin concentration at day 28

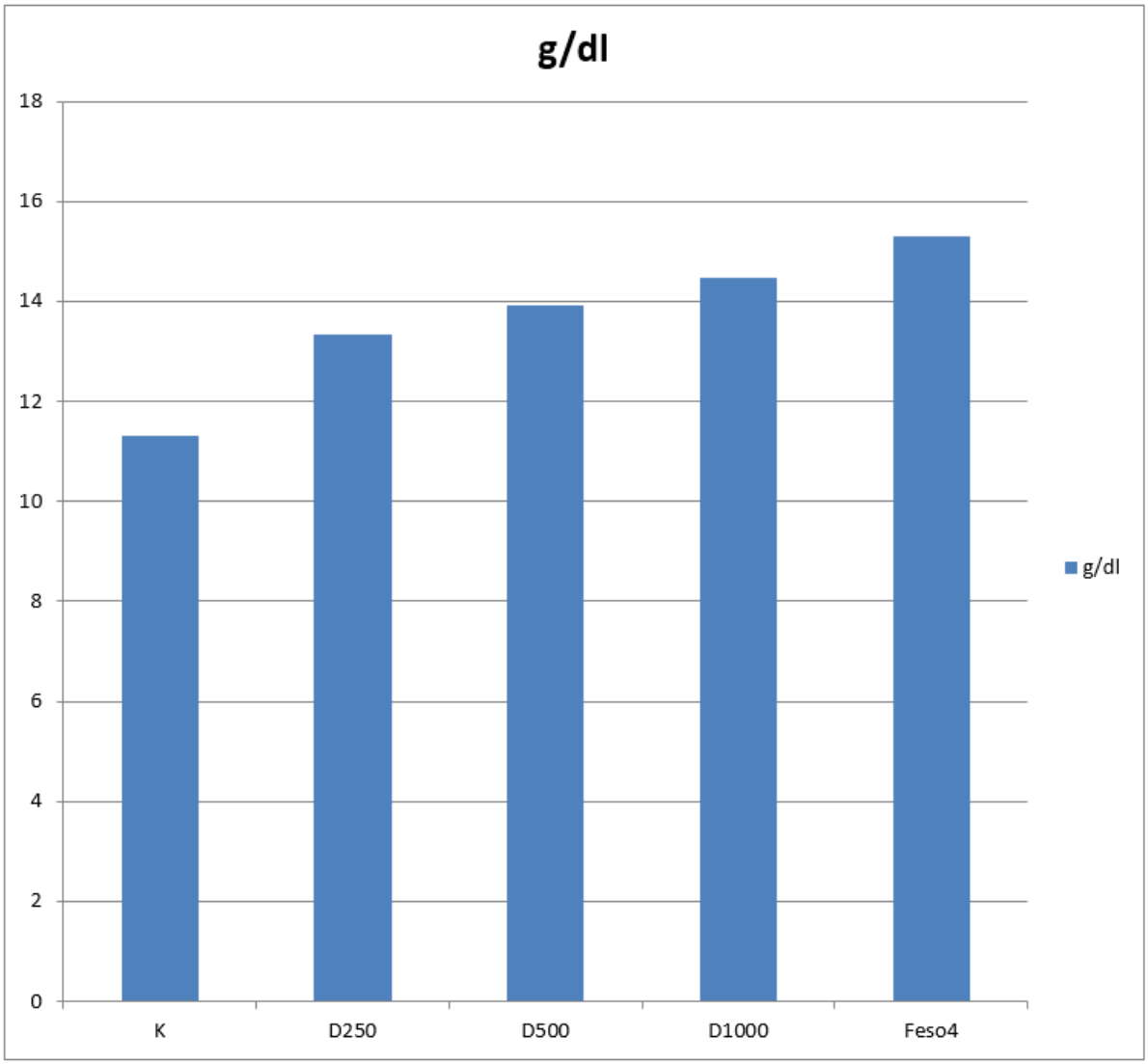


Figure 2.2: MEAN HEMATOCRIT on Day 28

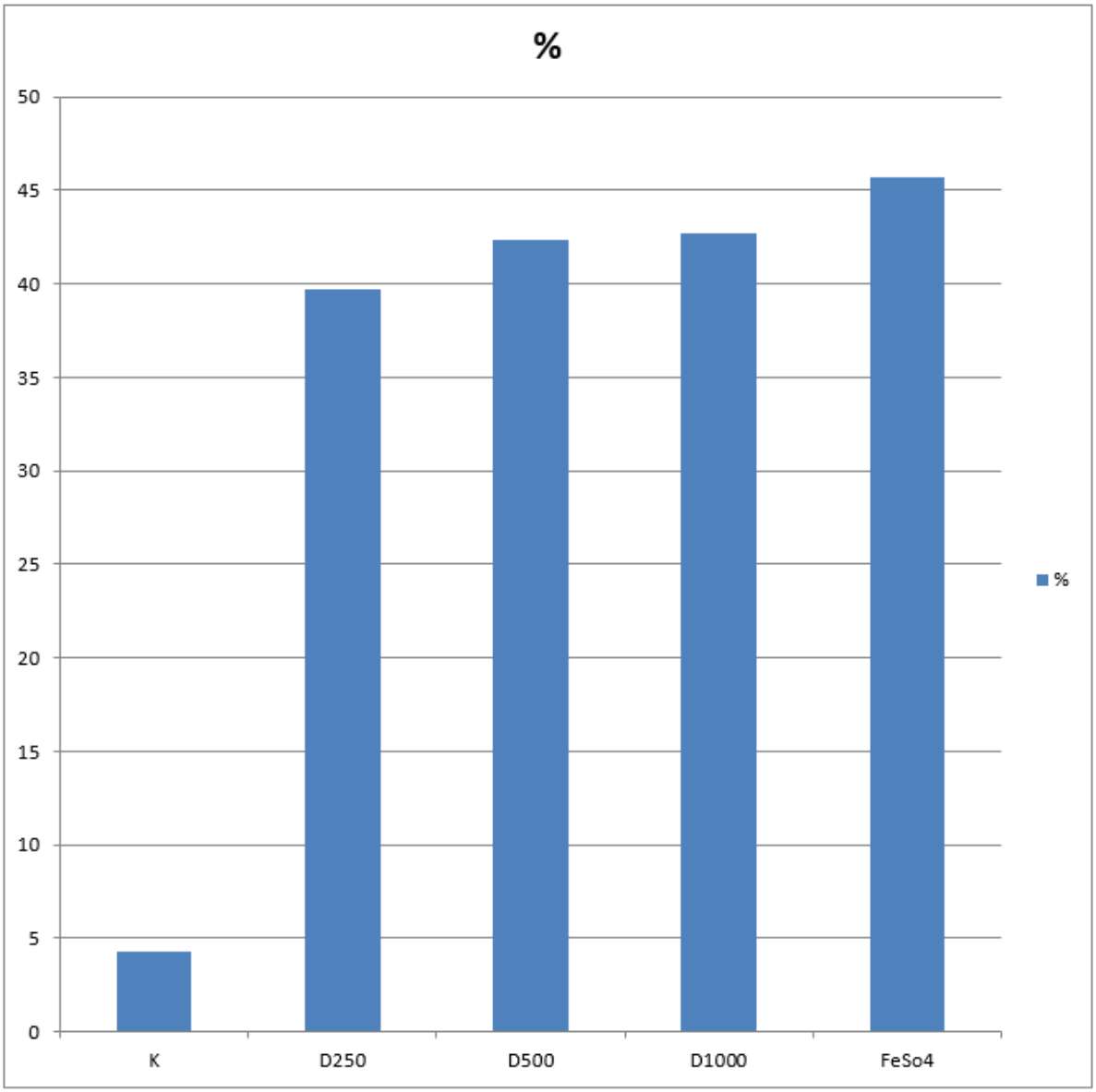
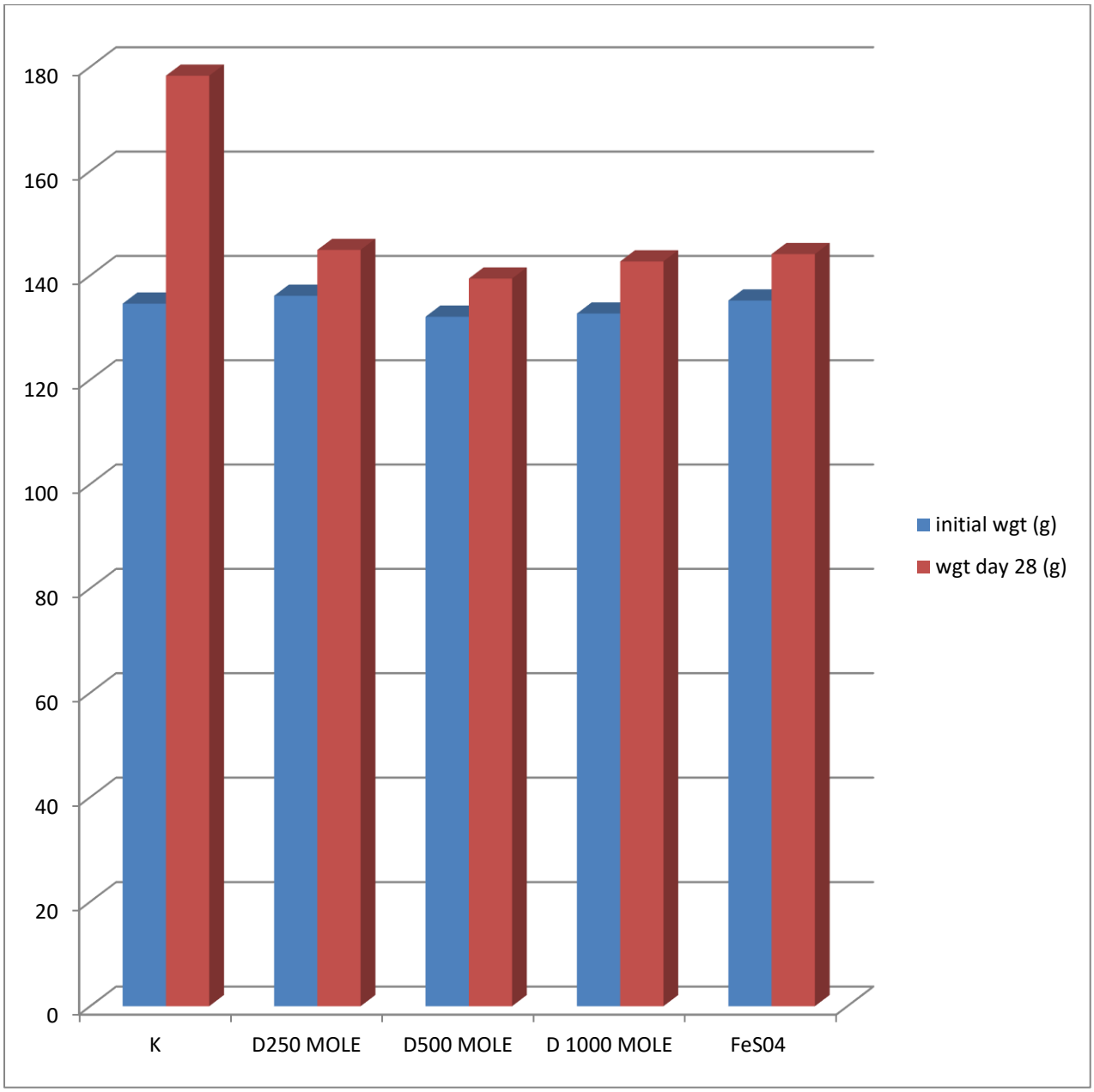


Figure 2.3: weight gain of study rats at day 28



III. Discussion:

Anemia can be grouped by causes, into blood loss, reduced blood cell production, and increased blood cell destruction (Sun and Wu, 2021). Iron deficiency is a habitual nutritional disorder worldwide, accounting for 50% of anemic cases. Its diagnosis is confirmed by finding low iron stores and a hemoglobin level, two standard deviations below normal (Jason and Mathew, 2013).

The study evaluated the hematopoietic effect of Moringa oleifera leaf extract and oral iron supplements. From the results obtained, there was a dose dependent significant increase in the Hemoglobin, hematocrit, mean cell volume and mean cell hemoglobin concentration. The mean hemoglobin concentration showed dose dependent increase with a P-value $0.20 > 0.05$. The mean hematocrit on the other hand had similar result with a P- value $0.002 < 0.05$, as compared to the control. The standard group however had the highest rate of increase in values. Moringa oleifera leaf extract was thus seen to potent as potent as the iron tablets.

Literature studies carried out by (Fauziandari, 2019), showed an increase in hemoglobin levels on administration of MOLE, thus being a potent alternative to iron and folic acid tablets (Fauziandari,2019). Similar studies done by (Nurhidayat , 2019), showed the average Hb levels after consuming Moringa oleifera leaf extract to be higher than it was before consumption, with P- value $0.009 < 0.05$ (10). The leaves were also seen to be strong alternatives to iron tablets, as was noticed in index study.

Studies done using Moringa leaf capsules, where seen to have as much potency as they were seen to contain iron, vitamin C and vitamins needed in absorption of iron (Nurhidayat, 2019). (Iskander et.al, 2015), in their study compared the control group where mothers in control group showed no change in in hemoglobin. They also discovered that MOLE could maintain serum ferritin range to 50%. There was also no case of low birth weight in mothers that consumed Moringa leaf extract. There was also increase in the weight of the study rats at day 28 in index study, at the end of the experiment, with non-showing muscle atrophy or shrinking of nose with a P value <0.05.

(Tinna, 2018), in the study, showed from analysis results that administration of iron together with the provision of Moringa flour in the intervention group increased erythrocyte levels with (P value 0.033 <0.05), compared to the central group that received only iron tablets.

IV. Conclusion:

The nutrient content in Moringa leaf extract plays an important role in anemia control, with its strong hematopoietic effect. Other factors that can increase the risk of anemia include micro nutrient deficiency, infectious diseases, malaria, HIV/AIDS and tuberculosis. Other causes would include sickle cell disease and thalassemia (Naveed and Hameed, 2023). Thus with the diverse ways in which Moringa oleifera can be used, its incorporation should be encouraged to reduce the prevalence of anemia globally.

V. Recommendation:

From findings of study, and the diverse properties of this plant, it would be necessary to incorporate Moringa oleifera extracts into our diets, and in health care in the control of anemia.
Due to its anti- oxidative, hematopoietic and other diverse functions, it is recommended, in women of reproductive age and adolescents and infants.

Disclaimer:
Authors declare that no generative AI technologies were used in writing this manuscript.

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Conflict of Interest:
The authors declare that they have no conflict of interest.

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