

**EFFECT OF METHANOLIC EXTRACT OF *OCIMUM*  
*GRATISSIMUM* (SCENT LEAF) ON SOME  
HEMATOLOGICAL PARAMETERS IN MALE WISTAR  
RATS**

**BY**

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## ABBREVIATIONS

- O. gratissimum* – *Ocimum gratissimum*  
MCHC – Mean Corpuscular Hemoglobin Concentration  
MCV – Mean Cell Volume  
GPV1 – Collagen-receptor glycoprotein IV

## ABSTRACT

Medicinal plants and the bioactive substances they contain have drawn the interest of various researchers over the past ten years due to their ability to cure different illnesses. *Ocimum gratissimum* is a member of the *Lamiaceae* family. It is cultivated in several gardens surrounding village huts in Nigeria under the popular name "scent leaf" for both medicinal and culinary purposes. The aim of this study is to elucidate the effect of *ocimum gratissimum* on some hematological parameters in male wistar rats. The effect of methanolic extract of *Ocimum gratissimum* on some red blood indices of Wister rats was studied using fifteen healthy adult wistar rats with weights ranging between 140-160g. The rats were divided into three groups; control group, low dose group and high dose group. Increasing doses (100mg and 300mgkg<sup>-1</sup> body weight) of the extract were administered orally to the other two groups for a period of four weeks. Sample collection was done via cardiac puncture using 5ml syringes. The extract displayed a significant increase ( $p < 0.05$ ) difference in platelet levels when compared with the normal control and a non significant difference ( $p < 0.05$ ) in the other parameters were observed. In conclusion, the extract of *gratissimum* might be a panacea in the management of anaemic conditions due to its erythropoietic, and/or haematopoietic effects, and beneficial to the blood's

oxygen supporting ability and thrombopoietin, putting into consideration that there were no alteration in the morphology and fragility of the RBCs.

## **CHAPTER ONE: INTRODUCTION**

### **1.0 BACKGROUND OF THE STUDY**

Because of their value in the management and prevention of serious and chronic diseases like cancer, diabetes, stroke, and arthritis (Sofowora *et al.*, 2013; WHO, 2019), as an alternative therapy for the treatment of psychiatric disorders (Venuprasad *et al.*, 2014), and in helping to meet the health needs of the global population, medicinal plants and their bioactive compounds have attracted the attention of several researchers over the past ten years (WHO, 2019). These medicinal plants are currently used to treat a wide range of illnesses as well as provide new medications for use in conventional or traditional medicine. From medical herbs including *Cinchona officinalis*, *Digitalis purpurea*, *Saix alba*, and

*Papaver somniferum*: quinine, digoxin, aspirin, and morphine, respectively, were made (Mbanaso *et al.*, 2000). *Ocimum gratissimum* belongs to the family Lamiaceae. It is commonly called ‘scent leaf’ and is cultivated in many gardens around village huts in Nigeria for its medicinal and culinary uses (Aguiyi *et al.*, 2000).

It is said to have come from South East Asia and Central Africa (Simon *et al.*, 1995). Numerous active substances, including flavonoids, triterpenes, alkaloids, citral, saponins, eugenol, linalool, methyl cinnamate, camphor, and thymol, have been identified in this plant using phytochemical screening (Pessoa *et al.*, 2002). Eugenol, an isolated compound from *O. gratissimum*, was found to have antihelminthic, nematocidal, and insecticidal effects (Begum *et al.*, 1993).

*Ocimum* is a genus that has been discovered to produce a variety of oils, which are often referred to as basilica oils. The oils made from *O. gratissimum* are effective against a number of bacteria and fungi, such as *Trichophyton rubrum* and *T. mentagrophytes*, as well as *Staphylococcus aureus*, *Listeria monocytogenes*, and *Escherichia coli* (Akinyemi *et al.*, 2004). The oils are used in the treatment of many ailments, including upper respiratory tract infections, diarrhea, headache, fever, eye problems, skin diseases, and pneumonia (Onayobi, 1986). The oil is also a potent antidiabetic agent (Mohammed *et al.*, 2007). Mbata and Saikia have reported the use of *O*

*gratissimum* for flavoring foods and as an antimicrobial agent (Mbata and Saika, 2007).

Blood is a tissue made of fluid plasma in which various created constituents are suspended. The relatively steady amounts of blood cells point to the presence of feedback regulatory systems (Guyton and Hall, 1996). Blood cells suspended in blood plasma make up blood (Merriam-Webster, 2017). Proteins, glucose, mineral ions, hormones, carbon dioxide (plasma is the principal medium for excretory product movement), and blood cells themselves are all found in plasma, which makes up 55 percent of the blood fluid. Plasma is 92 percent water by volume. The primary protein in plasma, albumin, controls the blood's colloidal osmotic pressure (National Cancer Institute, 2011). Red blood cells, also known as erythrocytes or RBCs; white blood cells, also known as leukocytes or WBCs, and in mammals, platelets make up the majority of the blood cells (also called thrombocytes). Red blood cells are the most numerous cells in the blood of vertebrates (Aryal, 2017). These have hemoglobin, a protein that contains iron and promotes oxygen delivery by reversibly binding to this respiratory gas and boosting its solubility in blood (The Franklin Institute, 2009). Contrarily, the majority of the extracellular transport of carbon dioxide occurs as the bicarbonate ion in plasma. When the hemoglobin in vertebrate blood is oxygenated, it is bright red, and when it is deoxygenated, it is dark red (Seguin, 2022). Hemocyanin, as opposed to hemoglobin, is sometimes used by

animals to carry oxygen, such as crustaceans and mollusks. Hemolymph, an alternative to blood, is used by insects and some mollusks. Hemolymph, unlike blood, is not enclosed in a closed circulatory system. Since most insects have small enough bodies for their tracheal systems to be adequate for giving oxygen, their "blood" does not contain oxygen-carrying molecules like hemoglobin (Kato *et al.*, 2018). Blood is circulated around the body through blood vessels by the pumping action of the heart. In animals with lungs, arterial blood carries oxygen from inhaled air to the tissues of the body, and venous blood carries carbon dioxide, a waste product of metabolism produced by cells, from the tissues to the lungs to be exhaled.

## 1.1 RESEARCH QUESTIONS

1. Does methanolic extract of *Ocimum gratissimum* have any effect on the functioning activity of blood cells?
2. What is the effect of methanolic extract of *Ocimum gratissimum* on hemoglobin (Hb) level?
3. What is the effect of methanolic extract of *Ocimum gratissimum* on packed cell volume (PCV)?
4. What is the effect of methanolic extract of *Ocimum gratissimum* on red blood cells (RBC)?
5. What is the effect of methanolic extract of *Ocimum gratissimum* on platelets?

6. What is the effect of methanolic extract of *Ocimum gratissimum* on MCV and MCHC levels?

## 1.2 JUSTIFICATION FOR STUDY

These medicinal plants are currently used to treat a wide range of illnesses as well as provide new medications for use in conventional or traditional medicine. Various in vivo and in vitro studies have shown that *O. gratissimum* and its bioactive constituents possess pharmacological properties such as antioxidant, anti-inflammatory, anticancer, hepatoprotective, antidiabetic, antihypertensive, antidiarrhoeal, and antimicrobial properties. This review demonstrated that *O. gratissimum* has a strong preventive and therapeutic effect against several diseases. The effectiveness of *O. gratissimum* to ameliorate various diseases may be attributed to its antimicrobial and antioxidant properties as well as its capacity to improve the antioxidant systems. With this study, I hope to add to the knowledge regarding the effect of *O. gratissimum* on some haematological parameters, especially hemoglobin (Hb) level, packed cell volume (PCV), red blood cells (RBC), mean cell volume and platelets level.

### **1.3 AIM OF STUDY**

This study aims to elucidate the effect of *Ocimum gratissimum* on some hematological parameters in male albino wistar rats.

### **1.4 OBJECTIVES**

- To evaluate the effect of *Ocimum gratissimum* on hemoglobin level.
- To evaluate the effect of *Ocimum gratissimum* on packed cell volume.
- To evaluate the effect of *Ocimum gratissimum* on red blood cells.
- To evaluate the effect of *Ocimum gratissimum* on platelets, MCV and MCHC.

## CHAPTER TWO: LITERATURE REVIEW

### 2.0 *OCIMUM GRATISSIMUM* (OG)

One of the recently found medicinal plants with the potential to be used as a novel medication source or as an alternative therapy for the treatment of a number of illnesses is *Ocimum gratissimum* L., also known as scent leaf. It is a common perennial herbaceous plant with a potent aroma that is also commercially viable. It can be found in Africa, Asia, and South America and is a member of the Lamiaceae family (Tanko *et al.*, 2008; Akara *et al.*, 2021). In the cooking of fish, meat, soup, and stew, it serves as a natural flavoring, seasoning, or vegetable. It is also used in conventional medicine to treat a variety of conditions, including bacterial and fungal infections, cough, pneumonia, fever, inflammation, anemia, diarrhoea, and aches according to scientific studies (Ironi *et al.*, 2016 and Venuprasad *et al.*, 2014), *O. gratissimum* contains a variety of bioactive substances, including flavonoids and

polyphenols, as well as essential oils that have a number of advantageous effects (Benitez *et al.*, 2009, Melo *et al.*, 2019).

Additionally, numerous studies have demonstrated that this plant has a wide range of pharmacological properties, including anti-hyperglycaemic (Aguiyi *et al.*, 2000; Casanova *et al.*, 2014), hypoglycaemic (Shittu *et al.*, 2019), anti-inflammatory (Ajayi *et al.*, 2019), anti-diarrheal (Offiah and Chikwendu, 1999). It is commonly called African basil or shrubby basil. It is Efinrin in Yoruba, Ebavbokho in Bini, Aai doya ta gida in Hausa and Nchonwu in Igbo (Owolade, 2004).

## **2.1 BOTANICAL DESCRIPTION**

The herbaceous plant *O. gratissimum* is also known as alfavaca, basil, and basil-clove. It is a member of the Lamiaceae family (Ocimum genus, gratissimum species) (Nweze and Eze, 2009). It stands 1-3 cm tall, has an upright stem that branches out to form a spherical, quadrangular base, and its leaves are opposite, slender, and marginalized. Most members of the Lamiaceae family are categorized as herbs, spices, and other aromatic varieties. There are 7200 species of trees, shrubs, and vines in the Lamiaceae family, which has 236 genera. About 60 species make up the genus *Ocimum*, with majority of them being found in Africa (Tanko *et al.*, 2008). *O. gratissimum* can be found in a variety of shapes and is frequently divided into various species and subspecies.



Fig 1.1 Pictorial sample of *O. gratissimum* with flowering buds (Eze, 2009)

## **2.2 GEOGRAPHICAL LOCATION**

*O. gratissimum* is a perennial and odoriferous shrub found in tropical regions such as Brazil, India, Vietnam, Rwanda, Nigeria (Lahlou *et al.*, 2004; Nweze and Eze, 2009), Cameroon, Togo, Cote d'Ivoire, Kenya, Benin Republic (Kpoviessi *et al.*, 2014), and South Africa (Venuprasad *et al.*, 2014).

### **2.2.1 Taxonomy**

Most members of the Lamiaceae family are categorized as herbs, spices, and other aromatic varieties. There are 7200 species of trees, shrubs, and vines in the Lamiaceae family, which has 236 genera. About 60 species make up the genus *Ocimum*, with majority of them being found in Africa (Tanko *et al.*, 2008). *Ocimum gratissimum* exists in a variety of forms and is frequently divided into various species and subspecies.

## **2.3 PHYTOCHEMISTRY**

### **2.3.1 Polyphenols and flavonoids found in *Ocimum gratissimum***

The phenolic compounds found in *Ocimum gratissimum* include rosmarinic acid, sinapic acid, salvigenin, gallic acid, catechins, methyl eugenol, caffeic acid, L-caftaric, ellagic acid, trans-ferulic acid, L-chicoric acid, and flavonoids such as xanthomicrol, cirsimaritin, rutin, apigenin, kaempferol, vicenin-2, luteolin 5-

O-glucoside, luteolin 7-O-glucoside, 7,4,0-dimethyl ether, vitexin, isovitexin, nepetoidin A, quercetin 3-Oglucoside, nevadensin, cirsimaritin, hymenoxin, myricetin, basilimoside, morin, isothymusin (Grayer *et al.*, 2000; Costa *et al.*, 2012; Ouyang *et al.*, 2013; Casanova *et al.*, 2014; Venuprasad *et al.*, 2014; Ajayi *et al.*, 2019), epicatechin, quercitrin, quercetin (Ironi *et al.*, 2016), and triterpenes (oleanolic, pomolic acid, ursolic acids, and tormentic acid) (Dzoyem *et al.*, 2021).

### **2.3.2 Chemical constituents of essential oil present in *O. gratissimum***

Compounds present in the essential oil of *O. gratissimum* include hydrocarbonated monoterpenes such as camphene,  $\alpha$ -thujene,  $\alpha$ -pinene, sabinene,  $\beta$ -pinene,  $\beta$ -myrcene,  $\alpha$  and  $\beta$ -phellandrene,  $\delta$ -3-carene, limonene,  $\alpha$ -terpinene, p-cymene, trans- $\beta$ -ocimene,  $\gamma$ -terpinene, terpinolene, p-cymenene, and p-menthane-1,3,8-triene; oxygenated monoterpenes such as 1,8-cineole, cis-sabinene hydrate, linalool, trans-sabinene hydrate, trans-thujone, citronellal, umbellulone, borneol, terpinen-4-ol, p-cymen-8-ol,  $\alpha$ -terpineol, thymol methyl ether, estragol, p-cymen-7-ol, thymol, and carvacrol; hydrocarbonated sesquiterpenes such as  $\alpha$ -copaene,  $\beta$ -elemene,  $\gamma$ -elemene,  $\beta$ -caryophyllene,  $\alpha$ -trans-bergamotene,  $\alpha$ -humulene,  $\beta$ -bourbunene,  $\alpha$ -guaiene,  $\delta$ -cadinene, germacrene D,  $\gamma$ -selinene,  $\beta$ -selinene,  $\alpha$ -selinene, (Z,E)- $\alpha$ -farnesene, and 7-epi- $\alpha$ -selinene; and oxygenated sesquiterpenes such as caryophyllene oxide, 1,2epoxydehumulene, and 3,7-(11)-eudesmadiene, (Pessoa *et al.*, 2002; Lahlou

*et al.*, 2004; Tchoumboungang *et al.*, 2005; Lemos *et al.*, 2005; Benitez *et al.*, 2009; Kpoviessi *et al.* 2012, 2014; Nguemtchouin *et al.*, 2013; Aguiar *et al.*, 2015; Mohr *et al.*, 2017; Chimnoi *et al.*, 2018; Melo *et al.*, 2019; Onyebuchi and Kavaz, 2020; Essoung *et al.*, 2020).

## **2.4 PHARMACOLOGICAL ACTIVITIES**

### **2.4.1 Anti-oxidant Activity**

The medicinal advantages of OG have been attributed to its anti-inflammatory and antioxidant characteristics (Olamilosoye *et al.*, 2019; Oyem *et al.*, 2021). Alpha-tocopherol and ascorbic acid, two antioxidant vitamins, have been found to be present in its leaf extracts (Olamilosoye *et al.*, 2019). Previous studies have demonstrated that phenols and flavonoids guard against the cellular damage brought on by oxidative stress. By scavenging or quenching free radicals, chelating metal ions, or inhibiting enzyme systems that produce free radicals, flavonoids and phenols have the ability to reduce inflammation and prevent oxidative damage (Olamilosoye *et al.*, 2016). The extract of OG contains saponins, terpenoids, glycosides, and alkaloids that may help explain why it has anti-inflammatory and antioxidant properties (Olamilosoye *et al.*, 2019; Oyem *et al.*, 2021).

### **2.4.2 Anti-Microbial Activity**

Studies have verified the antibacterial properties of OG (Ilori *et al.*, 1996; Nweze and Eze, 2009; Prakash *et al.*, 2011; Melo *et al.*, 2019). According to Prakash *et al.* (2011), OG essential oil is a plant-based preservative that can be used to protect spices against microbial and aflatoxin contamination as well as to extend the shelf life of foods due to its antioxidant activity. According to Chimnoi *et al.* (2018), *O. gratissimum* leaf essential oil extract (0.015-8.00 mg/ml) quickly inhibited *S. typhimurium* and *E. coli*. Additionally, prior studies demonstrated that the aqueous extract of OG significantly inhibited *Staphylococcus aureus* and strongly inhibited *Pseudomonas aeruginosa*. However, the aqueous ethanolic leaf powder extract showed a wider variety of antimicrobial activities with notable inhibitory properties against *E. coli*, *Bacillus cereus*, *P. aeruginosa*, and *S. aureus* (Talabi and Makanjuola, 2017). Joshi (2013) tested the antibacterial effects of OG essential oils using the tube-dilution method. Its main component, eugenol, showed potent antibacterial effects against *Klebsiella pneumoniae*, *Serratia marcescens*, and *E. coli*.

### **2.4.3 Anti-Inflammatory Properties**

According to reports, the herb possesses anti-inflammatory properties (Ajayi *et al.*, 2017; Alabi *et al.*, 2018). According to earlier research, carrageenan-induced inflammation in rats might be reduced when OG extract was administered at doses of 50, 100, and 200 mg/kg body weight. By reducing carrageenan-induced paw oedema in rats, the extract at 100 mg/kg body weight

may have therapeutic value in the management of inflammations (Ajayi *et al.*, 2017). In the study by Alabi *et al.* (2018), it was discovered that OG, at dosages of 100–800 mg/kg, had anti-inflammatory effects on dextran sodium sulphate (DSS)–induced colitis in rats, where evidence of healing were visible. The extract was found to be useful in the treatment of eosinophilic airway inflammation in male AJ mice induced by *Blomia tropicalis*. In a murine model, doses of 25, 50, and 100 mg/kg of methanolic extract of the plant were found to be effective in alleviating respiratory allergy (Costa *et al.*, 2012).

#### **2.4.4 Anti-Hypertensive Activity**

OG at 100 and 200 mg/kg improved blood pressure and toxic processes in cobalt chloride-induced cardio-renal dysfunction in rats (Akinrinde *et al.*, 2016). Investigations on the inhibitory effect of OG (8 weeks at 100 or 500 mg/kg) were carried out on angiotensin-converting enzyme (ACE) in hypertensive rats (Shaw *et al.*, 2017).

#### **2.4.5 Immunomodulatory Activity**

Mahapatra *et al.* (2011) examined the immunological actions and reactions in nicotine-induced macrophages (10 mM), as well as the immunomodulatory

efficacy of OG extract. After the injection of a 10 g/mL aqueous extract of the plant, nicotine-induced NO generation and iNOS II expression were significantly decreased. By inhibiting Th1 cytokines in nicotine-treated macrophages and inducing Th2 responses, the plant's aqueous extract showed protective effects on mice peritoneal macrophages (Mahapatra *et al.*, 2011).

#### **2.4.6 Wound-Healing Properties**

Chang *et al.* (2021) reported that 100 µg/mL *O. gratissimum* restored cell activity and protected against ultraviolet C-induced inhibition of cell proliferation and migration of skin cells, and therefore can serve as a potent natural wound care agent. According to a study done by Orafidiya *et al.*, *S. aureus* was resistant to the formulation's antibacterial effects when it contained 2 percent OG and honey as a surfactant. It claimed that ocimum oil's antibacterial action is influenced by the net electrical charge on the surfactant used in its production. Together with honey's known ability to promote wound healing, the outstanding antibacterial properties of the 2 percent ocimum oil in honey formulation raise the possibility that it could be used as a topical antiseptic for wounds (Orafidiya *et al.*, 2006).

#### **2.4.7 Cytotoxic activity**

Using the 3-(4,5-dimethylthiazol-2-yl)-2,5 diphenyltetrazolium bromide (MTT) method, the cytotoxic effects of methanolic extract *O. gratissimum* (ME-Og) were examined in murine peritoneal macrophages. The results showed that nicotine-induced free radical generation, lipid-protein damage, and antioxidant status in peritoneal macrophages are significantly modulated by *O. gratissimum* plant extracts. It was discovered that ME-Og had a protective effect against nicotine toxicity after dangerous 10 mM nicotine had been exposed to mouse peritoneal macrophages for 12 hours in culture media. The impact of the deadly chemical and the negative effects were greatly diminished in rats given the extract treatment. As a result, the findings support *O. gratissimum*'s ability to modulate dangerous substances like nicotine (Mahapatra *et al.*, 2009). In rats used as a model for chronic hepatic damage, Chiu *et al.*, (2012) discovered that *O. gratissimum* aqueous extract (OGAE) protected the liver against CCl<sub>4</sub>-induced damage. The CCl<sub>4</sub>-administered animals treated with the aqueous plant extract had an increase in blood catalase activity that was significantly higher than that of the control rats treated with saline solution. In a study comparing rats given OGAE to those given saline or CCl<sub>4</sub>, it was found that the rats given the plant extract while receiving CCl<sub>4</sub> had significantly lower levels of stress proteins such heat shock protein (HSP70) and inducible nitric oxide synthase in their livers (iNOS). A significant reduction in the ratio of MMP-9/MMP-2, phosphorylated ERK (p-ERK), and NF-kB (p-P65) was observed, suggesting

the possible protective effect of OGAE against CCl4-induced toxicity in rats (Chiu *et al.*, 2012).

When administered at doses ranging from 200 to 400 mg/kg/day for 20 days straight, the aqueous extract of *O. gratissimum* showed ameliorative properties that made it possible for it to provide protective advantages against acetic acid-induced colitis in male rats (Olamilosoye *et al.*, 2019). In a similar way, it improved the haematological parameters in colitis animals and greatly decreased the activities of MPO, SOD, and NO while raising GSH levels (Olamilosoye *et al.*, 2019). At concentrations between 150 and 200 g/mL, *O. gratissimum* modulated the apoptotic pathway, reducing the cytotoxic effects of H<sub>2</sub>O<sub>2</sub>-induced apoptosis (Chao *et al.*, 2017).

#### **2.4.8 Anti-infertility effect**

When administered, *Ocimum gratissimum* was observed to have an effect on penile and testicular tissues present in rats in the treatment of erectile dysfunction (Ojo *et al.*, 2019). Joseph *et al.* (2019) reported that methanolic and oil extracts of *O. gratissimum* leaves, when administered at two dosages of 250 and 500 mg for 14 and 28 days, had no negative effect on the reproductive capabilities of the male rats.

#### **2.4.9. Hepatoprotective activity**

According to research by Ajayi *et al.*, (2011), *O. gratissimum* administered to mice at various dosages may shield the liver by reducing the rise in catabolic enzyme levels brought on by CCl<sub>4</sub> damage to liver cells. According to Chiu *et al.* (2014), *O. gratissimum* extracts containing 0–40 mg/kg decreased liver steatosis, fibrosis, and damage while increasing catalase and anti-oxidative enzymes. According to Huang *et al.* (2020), after treatment with *O. gratissimum* at concentrations more than 20 mg/mL, an elevation in p/ERK1/2 levels was seen in hepatocarcinomal cells (HCC), suggesting that it affects the survival signaling of the liver cancer cells. The flavonoid-rich ethyl acetate fraction of *O. gratissimum* was discovered to exhibit hepatoprotective properties, indicating that it may be useful in slowing down the LPS-mediated sickness process in mice with LPS-induced sickness behavior (Ajayi *et al.*, 2019).

## **2.5 BLOOD, CELLS AND COMPOSITION**

### **2.5.1 Blood**

Blood cells suspended in blood plasma make up blood (Merriam-Webster, 2017). Proteins, glucose, mineral ions, hormones, carbon dioxide (plasma is the principal medium for excretory product movement), and blood cells themselves are all found in plasma, which makes up 55 percent of the blood fluid. Plasma is 92 percent water by volume. The primary protein in plasma, albumin, controls the blood's colloidal osmotic pressure (National Cancer Institute, 2011). Red

blood cells, also known as erythrocytes or RBCs, white blood cells, also known as leukocytes or WBCs, and in mammals, platelets make up the majority of the blood cells (also called thrombocytes). Red blood cells are the most numerous cells in the blood of vertebrates (Aryal, 2017). These have hemoglobin, a protein that contains iron and promotes oxygen delivery by reversibly binding to this respiratory gas and boosting its solubility in blood (The Franklin Institute, 2009). Contrarily, the majority of the extracellular transport of carbon dioxide occurs as the bicarbonate ion in plasma. When the hemoglobin in vertebrate blood is oxygenated, it is bright red, and when it is deoxygenated, it is dark red (Seguin, 2022). Blood-related medical phrases frequently begin with the Greek word (haima), which means "blood," or with hemo-, hemato-, haemo-, or haemato-. Given its genesis in the bones and the possibility of molecular fibers in the form of fibrinogen, blood is regarded as a specialized type of connective tissue in terms of anatomy and histology (Krause, 2009). Numerous vital bodily processes are carried out by blood, including the delivery of oxygen to tissues (bound to hemoglobin, which is carried by red blood cells), the supply of nutrients (such as glucose, amino acids, and fatty acids) to tissues, the removal of waste products (such as carbon dioxide, urea, and lactic acid), the circulation of white blood cells in the immune system, and the detection of foreign objects by antibodies, coagulase, and other proteins. Hydraulic processes and core body temperature regulation.

## 2.5.2 Blood Components

- Composition

According to Alberts (2012), blood makes up 7% of a person's body weight and has an average density of 1060 kg/m<sup>3</sup>, which is quite similar to the density of pure water at 1000 kg/m<sup>3</sup> (Elert, 2012). The blood volume of a typical adult is around 5 litres (11 US points), or 1.3 gallons, and is made up of plasma and formed components. Red blood cells (erythrocytes) and white blood cells (leukocytes), as well as platelets—cell fragments involved in clotting—are the two types of blood cells or corpuscles that are created (Shmurkler, 2004). Red blood cells make up around 45% of whole blood by volume, plasma about 54.3 %, and white blood cells about 0.7 %. The main factor affecting the color of blood in vertebrates is hemoglobin. The four heme groups on each molecule interact with other molecules to change the precise color. As oxygen strongly reddens the heme group, arterial blood and capillary blood in vertebrates and other hemoglobin-using organisms are brilliant red. Deoxygenated blood is present in veins and is visible during blood donations and when venous blood samples are taken. It is a darker shade of red. This is because the spectrum of light that hemoglobin absorbs varies depending on whether it is oxygenated or not (Prahl, 2002)

- Cells

One microliter of blood contains about:

- a. 4.7 to 6.1 million red blood cells (for males) and 4.2 to 5.7 million red blood cells (for females).
- b. 4,000–11,000 leukocytes (or white blood cells).
- c. 200,000–500,000 thrombocytes (or platelets).

- Plasma

The blood's liquid medium, blood plasma, which is by itself straw-yellow in hue, makes up around 55% of the blood. An average individual has a blood plasma volume of 2.7–3.0 liters (2.8–3.2 quarts). Essentially, it is an aqueous solution made up of 92% water, 8% blood plasma proteins, and traces of other substances. Plasma transports nutrients that are dissolved in the blood, such as glucose, amino acids, and fatty acids, as well as waste products like carbon dioxide, urea, and lactic acid. .

- pH Values

The narrow range of 7.35 to 7.45 is where blood pH is controlled, making it somewhat basic (compensation) (Waugh and Grant, 2007). Blood with an extracellular fluid pH below 7.35 is too acidic, and blood with an extracellular fluid pH over 7.45 is too basic (Donna *et al.*,2018). Typically, a pH of 6.9 or higher is fatal.

### **2.5.3 Red Blood Cells (or Erythrocytes)**

#### **Structure**

Red blood cells (RBCs), also known as erythrocytes, are the functional part of blood that transport gases and nutrients throughout the body. These specialized cells can perform their critical jobs due to their particular structure and makeup. The adult erythrocyte is anucleated and has a biconcave discoid form (Smith, 1987). This architecture provides the mobility required to move through the circulatory system and an expanded surface area that facilitates enough gas exchange and enables the cell to function. This particular cell's structure is framed by a phospholipid bilayer membrane that is kept in place by the cytoskeleton, a network of proteins. This cytoskeleton, which is made up of spectrin, actin, band 3, protein 4.1, and ankyrin, enables both the structural integrity and malleability of cells. These molecules interact to support a structure that is both rigid and flexible (Kuhn *et al.*, 2017).

#### **Function**

The lifespan of a red blood cell is just about 120 days. In that little period, it must transport oxygen from the lungs to peripheral tissues to help with metabolic processes like ATP generation, and it must also gather carbon dioxide produced in the peripheral tissues and return it to the lungs for removal from the body. The hemoglobin with ferrous heme (Fe) that is present in the deoxygenated blood that reaches the lungs has an affinity for oxygen. The heme

loses its affinity for the oxygen as it reaches the deoxygenated tissues due to the low pH and decreased partial pressure of oxygen, which allows the oxygen to reach the tissue. The cell then absorbs carbon dioxide, which is subsequently mixed with water to produce bicarbonate and hydrogen via carbonic anhydrase. Most of the carbon dioxide will travel back to the lungs in the form of bicarbonate and be exhaled (Kuhn *et al.*, 2017).

### **Pathophysiology**

Erythrocytes are very responsive to their environment, altering shape and responding to it. The erythrocyte exists as a biconcave disc in its optimal state. The cell transforms in reaction to exposure to specific chemicals or substances. It has been suggested that this occurs when either the erythrocyte's phospholipid bilayer is altered by the environment or it is exposed to environmental oxidants (Ford, 2013). Red blood cells, for instance, take on the appearance of an echinocyte when their energy supply, ATP, is exhausted or when intracellular calcium levels are elevated. A erythrocyte also develops into a stomatocyte when it swells with water. These modifications take place as a result of how these circumstances affect the lipid bilayer membrane (Pretorius *et al.*, 2016). Additionally, hydrogen peroxide can be effectively converted by the red blood cell into water to stop its ability to oxidize lipids and degrade proteins. The red blood cell experiences oxidative stress because it lacks the enzymes necessary to perform this activity in some hereditary diseases.

#### **2.5.4 White blood cells (or lymphocytes)**

##### **Structure**

Granulocytes, lymphocytes, and monocytes are subtypes of WBC. Granulocytes have distinctive cytoplasmic granulation, which gives them their name. Neutrophils (or polymorphonuclear granulocytes), eosinophils, and basophils are the three types that are recognized (Cline and Hunt, 1983). Colony forming unit (CFU)-S or CFU-GEMM, two types of common pluripotent stem cells, are the source of myeloid cells. Both the myeloid precursor and lymphoid cells are derived from a more basic stem cell. The stem cells develop into the adult blood components through a series of intermediary processes under the control of poietins and microenvironmental factors. Burst forming unit (BFU)-E, CFU-E, and the erythroid series are all descended from CFU-S, along with CFU-MEGA, which produces megakaryocytes, and CFU-GM, which produces monocytes and granulocytes. Morphologically, stem cells resemble lymphocytes. Their in vitro growth characteristics in semisolid media containing various growth agents can be used to identify them (Quesenberry and Levitt, 1979).

The myeloblast is the first cell that can be distinguished morphologically in the genesis of the neutrophilic granulocyte. The myeloblast develops into a promyelocyte and then a myelocyte as it matures. These developmental stages constitute predominantly a proliferative compartment, in which the cell number increases geometrically. The next form, the metamyelocyte, is unable to

undergo further mitosis but transforms into a band (Foon *et al.*, 1982). This cell is either released into circulation (3 to 5% of WBC) where it completes its maturation or enters a storage compartment in the marrow where it becomes a neutrophil and is released later into the circulation. About half of the intravascular polymorphonuclear cells are circulating, maintaining a dynamic equilibrium with the other half, which are marginated against the vascular endothelium. Only the circulating neutrophils are accounted for in the WBC count. The half-life of mature neutrophils in circulation is about 7 hours. They irreversibly traverse the vascular endothelium into the tissues, where they die after 1 or 2 days. The phagocytosis of germs is the primary function of neutrophilic granulocytes. A lysosome is fused with through a complex multistage process that involves engulfing the organism, incorporating it into the cytoplasm, and releasing enzymes that will kill the bacterium while releasing energy. The development of eosinophils and basophils is comparable. Eosinophils quickly leave the intravascular compartment (where they make up to 5% of WBC) after being released from the bone marrow and go into the tissues. They can't get back into the blood. The GI tract, lungs, and skin all contain high levels of eosinophils. It is unclear exactly what these complicated cells do. They may aid in reducing inflammation and providing protection against multicellular parasites (Cline and Hunt, 1983). About 1% to 2% of the leukocytes in circulation are basophils. Their precise physiologic function is also unknown. They include histamine and heparin in their granules. They have

IgE bonded to their surface. Mononuclear leukocytes are a collective term for lymphocytes and macrophages. Both cellular and humoral immunity depend on both of them. These cells can leave and reenter circulation while still functioning. They could stay in the lymph nodes or the tissues for some time.

The CFU-GM in the bone marrow is the source of the cells that make up the monocyte-macrophage system. Instead of being kept, they are quickly discharged into the bloodstream, where they make up 5% of the WBC. They transform into macrophages in tissues (Wintrobe, 1987). Monocyte-macrophages process antigenic material and "communicate" with T lymphocytes through a cell-cell interaction process. They phagocytose bacteria and particulate matter, contribute to the inflammatory response, and are crucial in the immune system. Interleukin, a chemical that potentiates B and T lymphocytes, can be secreted by monocytes. They release plasminogen activators, which aid in fibrinolysis. Immune cells called lymphocytes are essential for both cellular and humoral immunity. They account for 20 to 45 percent of WBC in the blood. They are a part of the T (thymus) or B (bursa or bone marrow) systems. There is no morphological difference between the two cells. Antibody synthesis is carried out via the B system. When a B cell is suitably activated, it first multiplies before changing into a plasma cell, the immune system's effector limb. Each B cell can only produce one type of antibody (Wintrobe, 1981).

The cellular immune system is made up of the T system, which also controls the entire immune system. Monoclonal antibodies directed against various membrane antigens can be used to distinguish between different T cell subsets. For instance, suppressor T cells hinder B cell function while helper T cells promote it. Natural killer (NK) lymphocytes are in charge of the non-specific lysis of some cells, while some T cells are responsible for cell-mediated cytotoxicity.

Between 15 and 25 percent of the lymphocytes in the peripheral circulation are B cells, while between 40 and 75 percent are T cells (Ogawa *et al.*, 1983).

### **2.5.5 Platelets**

The purpose of platelets, also known as thrombocytes (from the Greek, "clot," and, "cell"), which are a blood component, is to react to bleeding from blood vessel injury by clumping and starting a blood clot. (Laki, 1972). The megakaryocytes (Machlus *et al.*, 2014; Lefrancais *et al.*, 2017) of the bone marrow or lung produce platelets, which are cytoplasmic pieces that then enter the bloodstream. Only mammals have platelets, whereas birds and amphibians have thrombocytes that circulate as whole mononuclear cells (Michelson, 2013). The platelets (P) are signaled by the ligands (L) to migrate to the wound (Site A). The production of ligands increases as more platelets congregate near the opening, amplifying the reaction. To form a cap to halt blood flow out of the

tissue, platelets assemble around the wound. One major function of platelets is to contribute to hemostasis: the process of stopping bleeding at the site of interrupted endothelium. They gather at the site and, unless the interruption is physically too large, they plug the hole. First, platelets attach to substances outside the interrupted endothelium: adhesion. Second, they change shape, turn on receptors and secrete chemical messengers: activation. Third, they connect to each other through receptor bridges: aggregation (Yip *et al*, 2005). The coagulation cascade is activated during the formation of this platelet clog (primary hemostasis), which results in fibrin deposition and linkage (secondary hemostasis). There may be overlap between these processes; the spectrum ranges from a mainly platelet plug, or "white clot," to a mostly fibrin plug, or "red clot," or the more common mixture. As the fourth and fifth steps in the process' completion, some would also include the subsequent retraction and platelet inhibition (Berridge, 2014), while others would add a sixth step—wound repair. Additionally, platelets take involved in both innate and adaptive intravascular immunological responses (Gaertner and Massberg, 2016; Hampton, 2018). Low platelet concentration is called thrombocytopenia, and is due to either decreased production or increased destruction. Elevated platelet concentration is called thrombocytosis, and is either congenital, reactive (to cytokines), or due to unregulated production: one of the myeloproliferative neoplasms or certain other myeloid neoplasms. A disorder of platelet function is called a thrombocytopathy or a platelet function disorder. Normal platelets may

react to an anomaly on the vessel wall rather than to bleeding, which can cause incorrect platelet adhesion/activation and thrombosis, or the creation of a clot inside an intact conduit. This kind of thrombosis develops by different causes than a typical clot, including the extension of the fibrin in venous thrombosis, the extension of an unstable or ruptured arterial plaque, which results in arterial thrombosis, and microcirculatory thrombosis. An arterial thrombus can either fully block blood flow, which results in downstream tissue death, or it can totally block it, resulting in ischemia upstream (Michelson, 2013). Platelets have a central role in innate immunity, initiating and participating in multiple inflammatory processes, directly binding pathogens and even destroying them. This is consistent with clinical evidence that many people with serious bacterial or viral infections have thrombocytopenia, which lessens their ability to cause inflammation. Typical platelet-leukocyte aggregates (PLAs) in sepsis or inflammatory bowel illness demonstrate the relationship between thrombocytes and immune cells (Jenne, 2013). The membrane of the platelet cell contains receptors for collagen. The platelets are exposed once the blood artery wall ruptures, and they stick to the collagen in the nearby connective tissue. During inflammation, Platelets are rapidly deployed to sites of injury or infection, and potentially modulate inflammatory processes by interacting with leukocytes and secreting cytokines, chemokines, and other inflammatory mediators. Platelets also secrete platelet-derived growth factor (PDGF). Platelets modulate neutrophils by forming platelet-leukocyte aggregates (PLAs). These formations

induce upregulated production of  $\alpha\text{M}\beta\text{2}$  (Mac-1) integrin in neutrophils. Interaction with PLAs also induce degranulation and increased phagocytosis in neutrophils. Platelets are also the largest source of soluble CD40L which induces production of reactive oxygen species (ROS) and upregulate expression of adhesion molecules, such as E-selectin, ICAM-1 and VCAM-1, in neutrophils, activates macrophages and activates cytotoxic response in T and B lymphocytes (Jenne, 2013).

Recently, the theory that mammalian platelets without nuclei are incapable of autonomous locomotion was refuted (Gaertner *et al.*, 2017). In actuality, the platelets are active scavengers who scale blood vessel walls and reorganize the thrombus. As a result of their ability to fully enclose bacteria in their open canalicular system (OCP), which is merely an invagination of the outer plasma membrane, they are able to recognize and adhere to a wide variety of surfaces, including bacteria. As a result, the process is sometimes referred to as "covercytosis" rather than "phagocytosis." The neutrophils then use these platelet-bacteria bundles as a platform for interaction to NETose and phagocytose the bacteria, destroying them. . Platelets also participate in chronic inflammatory disease, such as synovitis or rheumatoid arthritis (Boilard *et al.*, 2010). Platelets are activated by collagen receptor glycoprotein IV (GPVI). Proinflammatory platelet microvesicles trigger constant cytokine secretion from neighboring fibroblast-like synoviocytes, most prominently Il-6 and Il-8.

Inflammatory damage to the surrounding extracellular matrix continuously reveals more collagen, maintaining the microvesicle production.

### **2.5.6 Mean Cell Volume**

The mean corpuscular volume (MCV), sometimes known as the mean cell volume (MCV), is a measurement of the typical size of a red blood cell (or red blood cell). The measurement is made by multiplying the volume of blood by the hematocrit, or the percentage of blood that is made up of red blood cells, and then dividing the result by the volume's erythrocyte content. In a typical full blood count, the mean corpuscular volume is measured.

The MCV measurement in anemic patients allows for classification as either having microcytic anemia (MCV below normal range), normocytic anemia (MCV within normal range), or macrocytic anemia (MCV within normal range) (MCV above normal range). Normocytic anemia is typically thought to exist when the bone marrow has not yet changed the volume of the cells in response. Hemolysis and blood loss are two acute diseases where it might occasionally happen.

The outcome, if the MCV was calculated using automated equipment, can be compared to the RBC morphology on a peripheral blood smear, where a normal RBC is roughly the size of a normal lymphocyte nucleus. Though there are some diseases that present with high MCV without megaloblastic cells, any

divergence would typically be suggestive of either malfunctioning apparatus or operator mistake. For further specification, it can be used to calculate red blood cell distribution width (RDW). The RDW is a statistical calculation made by automated analyzers that reflects the variability in size and shape of the RBCs. To calculate MCV, the hematocrit (Hct) is divided by the concentration of RBCs (Mondal, 2020)

$$\text{MCV} = \frac{\text{Ht}}{\text{RBC}}$$

Normally, MCV is expressed in femtoliters (fL, or  $10^{-15}$  L), and [RBC] in millions per microliter ( $10^6 / \mu\text{L}$ ). The normal range for MCV is 80–100 fL.

The MCV can be calculated by automatic analyzers in a variety of ways using assumed units. The red blood cells travel one at a time through a tiny aperture in volume-sensitive automated blood cell counters like the Coulter counter, where they produce a signal that is precisely proportionate to their volume. Other automated counters use methods to measure light that is refracted, diffracted, or dispersed to calculate the volume of red blood cells (Stanley, 2011). The common reference range is between 80 and 100 fL (Medical Encyclopedia, 2020).

### **2.5.7 Haemoglobin and Haematocrit**

Red blood cells include the protein hemoglobin (Hb), which is in charge of carrying oxygen to the tissues. Maintaining a suitable hemoglobin level is necessary to guarantee appropriate tissue oxygenation. G/dl is the unit used to measure the amount of hemoglobin in whole blood. The normal Hb range for men is 14 to 18 g/dl, whereas it is 12 to 16 g/dl for women. Anemia is present in the patient when the hemoglobin level is low. The outcome of having too many red blood cells is an erythrocytosis, which raises the hemoglobin level over normal (Adamson and others, 1975). Hematocrit quantifies the proportion of red blood cells to total blood volume (red blood cells and plasma). The typical hematocrit for men is 40 to 54 percent ; for women it is 36 to 48 percent . Microhematocrit centrifugation can be used to directly compute this value or to make an indirect calculation. Automated cell counters multiply the red cell count (in millions/mm<sup>3</sup>) by the mean cell volume to determine the hematocrit (MCV, in femtoliters). While so assessed, it is susceptible to the unforeseen circumstances that arise when trying to quantify the MCV accurately (Bunn and Beck, 1981). Since whole blood is the basis for both the hemoglobin and the hematocrit, plasma volume is necessary. Hemoglobin and hematocrit in a patient with severe dehydration will seem greater than they would if the patient were normovolemic; in a patient with fluid overload, they will appear lower than their real level (Scott, 1981) .

### **2.5.8 Mean Corpuscular Haemoglobin Concentration**

The average amount of hemoglobin in a given volume of packed red blood cells is called the mean corpuscular hemoglobin concentration (MCHC). By dividing hemoglobin by hematocrit, it is calculated. Reference ranges for blood tests are between 4.81 and 5.58 mmol/L, or 32 to 36 g/dL (320 to 360 g/L) (Medical Encyclopedia, 2011). A mass or molar concentration is what it is. However, MCHC is frequently expressed as a percentage (percent), rather than a mass fraction (mHb / mRBC) (BloodBook, 2009 and MedNet, 1999). However, when assuming an RBC density of 1 g/mL and negligible hemoglobin in plasma, the MCHC in g/dL and the mass percentage of hemoglobin in red blood cells are numerically equal. A low MCHC can be used to indicate a drop in production of haemoglobin. MCHC can be normal even when hemoglobin production is decreased (such as in iron deficiency) due to a calculation artifact. MCHC can be elevated ("hyperchromic") in hereditary spherocytosis, sickle cell disease and homozygous hemoglobin C disease, depending upon the hemocytometer (Hill *et al.*, 2017). MCHC can be elevated in some megaloblastic anemias. MCHC can be falsely elevated when there is agglutination of red cells (falsely lowering the measured RBC count) or when there is opacification of the plasma (falsely increasing the measured hemoglobin). Causes of plasma opacification that can falsely increase the MCHC include hyperbilirubinemia, hypertriglyceridemia, and free hemoglobin in the plasma (due to hemolysis).

## **CHAPTER THREE**

### **3.0 MATERIALS AND METHOD**

#### **3.1 Materials**

These are some of the materials used in the course of this experiment. They include:

- 16G calibrated gavage
- Disposable 5ml needle syringes
- EDTA blood sample vials
- Gloves and nose masks.
- Feeders and drinkers
- Plastic cages

- Calibrated syringes

### **3.2 Experimental Animals**

Fifteen Male Albino Wistar rats weighing between 160-180g were used for the study. The rats were purchased from the animal house of the Department of Anatomy, University of Benin. They were bred for 4 weeks in the animal house of the Department of Anatomy, University of Benin. The rats were fed with pellets grower mash obtained from Top Feeds Mill, Benin and with water *ad libitum* during the breeding period.

### **3.3 Preparation of Extracts and Dose concentration**

Fresh leaves of the *OG* were bought at Urelu Market, Benin. Botanical identification of the plants was done at the herbarium of the Botany Department of University of Benin. Extraction was done at the Department of Pharmacognosy, Faculty of Pharmacy of the University of Benin. The leaves of *O. gratissimum* were collected and dried at room temperature for a period of 21days until constant weight was obtained. The dried leaves were then pulverized to powder form by a machine blender and sieved. 50g of the powdered leaves was stirred into 450 ml of methanol containing 70% alcohol. The mixture was then kept aside for 48 hrs to allow it to infuse. It was then filtered using a filter paper. The filtrate was concentrated to 200ml (1ml of the

extract being equivalent to 0.25g of the starting material). The extract was kept in petri dishes with tight fitting covers in a refrigerator until it was time to use.

The extract was dissolved into a solution, with 2g of the extract into 20ml of distilled water. The dosage was calculated in this equation;

$$\frac{\text{Weight of experimental animals (in kg) x dosage}}{\text{Concentration of solution (in ml)}}$$

### **3.4 Administration of Extracts**

Administration of the aqueous extract was done orally by means of calibrated syringe with attached 16G gavage. The animals received their doses daily for 28 days.

### **3.5 Study Area**

The study was carried out at the Animal House of the Department of Anatomy, University of Benin. The study was done in plastic cages measuring breadth as 30cm, length as 42cm and height as 24cm, having a volume of 30,240cubiccm. The rats were selected at random and grouped into three (n=5).

### **3.6 Ethical Consideration/Precautions**

Throughout the experimental period, animals were handled and maintained in accordance with the guidelines of the Institutional Animal Care and Use Committee (IACUC). Animals were handled with caution using protective

latex gloves and care was taken to prevent holding the animal too stiffly by the neck or abdomen.

### **3.7 Study Design**

The rats were selected at random and grouped into three (n=5). A control group, while groups 1 and 2 were the experimental groups. Each of the rats were marked at different parts of their bodies for easy identification and put into different segments of the cage, according to their group.

Control group received a quantity of distilled water equivalent to the volume in group 2 and 3. Groups 2 and 3 received the methanolic extract of *Ocimum gratissimum* at doses of 100 and 300mgkg<sup>-1</sup> respectively.

### **3.8 Measurement of Body Weight**

Body weight was measured on a weekly basis using a lithium-cell battery operated weight scale. Weight ranges for the first week span between 128g to 161g. Subsequently, increase in weight was observed in the following weeks.

### **3.9 Sample Collection**

At the end of the experimental period, the final weights of the rats were taken using *compact electric weighing scale calibrated in grams*. Cotton wool was soaked with chloroform of about 50ml in an enclosed container and the rat was placed in the enclosed container containing the chloroform for about 30-50 sec

to anaesthetize it. After anaesthetization, the rat was placed in a supine position on the dissection table.

Abdomino-thoracic incision was made on the rat to expose the abdominal viscera. Thereafter, blood samples were collected through the inferior vena cava and through the heart by the process of venous and cardiac puncture respectively. Using 5ml syringes, the blood samples were transferred into an EDTA bottle for haematological analysis.

### **3.10 Haematological Analysis**

Full blood count was performed using a DH-36 Haematology analyzer (Shenzhen Dymind, China), a three-part auto analyzer able to test 21 parameters per sample including Hb concentration, PCV, RBC concentration, MCH, MCV, MCHC, WBC count, and PLT count. Standardization, calibration of the instrument, and processing of the samples were done according to the manufacturer's instructions.

Each blood sample was mixed well and then approximately 9  $\mu$ L was aspirated by allowing the analyzer's sampling probe into the blood serum sample and depressing the start button. Results of the analysis were displayed after about 30 seconds, after which the analyzer generated hard copy of the results on thermal printing paper.

### **3.11 Statistical Analysis**

Data was analysed using pad statistical software version 8.1 and results analysed were presented in mean +/- SEM. The P-values <0.05 were considered statistically significant.

## CHAPTER FOUR

### RESULTS AND ANALYSIS

#### 4.0 RESULTS

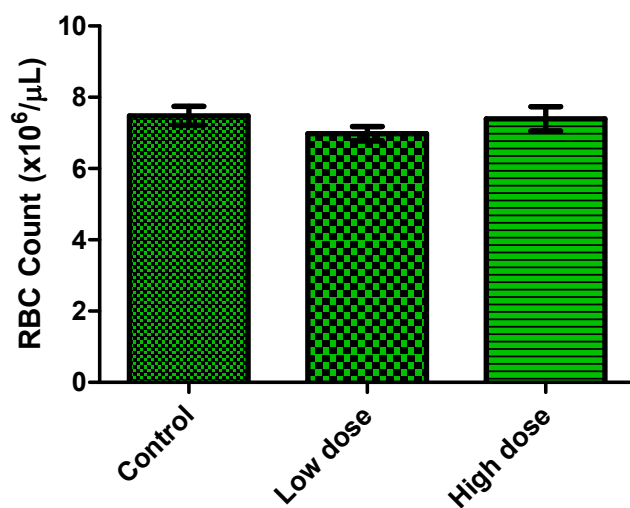
Table 1: Comparing the mean values of red blood cell indices in male Albino rats administered with methanolic extract of *Ocimum gratissimum* leaf.

Parameters	Control	Low dose	High dose
RBC count (x10 <sup>3</sup> /μl)	7.480 ± 0.265	6.976 ± 0.201	7.396 ± 0.339
Haemoglobin (g/dl)	15.40 ± 0.535	14.54 ± 0.568	15.70 ± 0.846
Haematocrit level (%)	40.20 ± 1.485	39.32 ± 1.265	44.68 ± 0.893 <sup>xa</sup>

MCV (fl)	53.84 ± 0.950	56.52 ± 1.803	61.14 ± 3.608
MCH (pg)	20.52 ± 0.074	20.78 ± 0.412	21.16 ± 0.361
MCHC (mg/dl)	38.28 ± 0.605	36.94 ± 0.879	35.16 ± 1.996
Platelet count	602.2 ± 55.48	660.4 ± 110.5	759.2 ± 105.8*

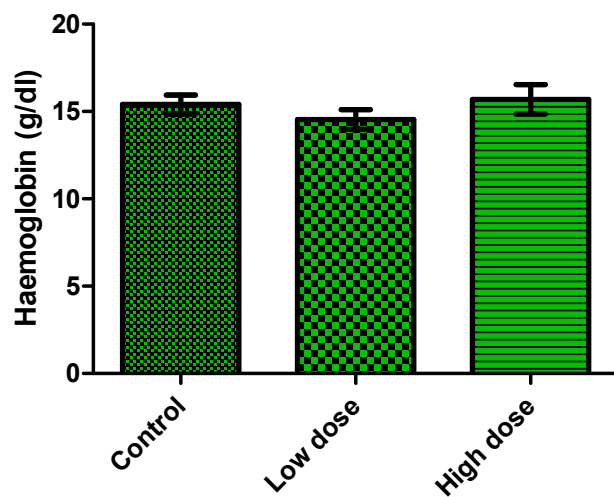
\*P < 0.05 indicates significant difference with control.

**Chart I: Effect of methanolic extract of *Occimum gratissimum* on red blood cells count of male Abino rats**



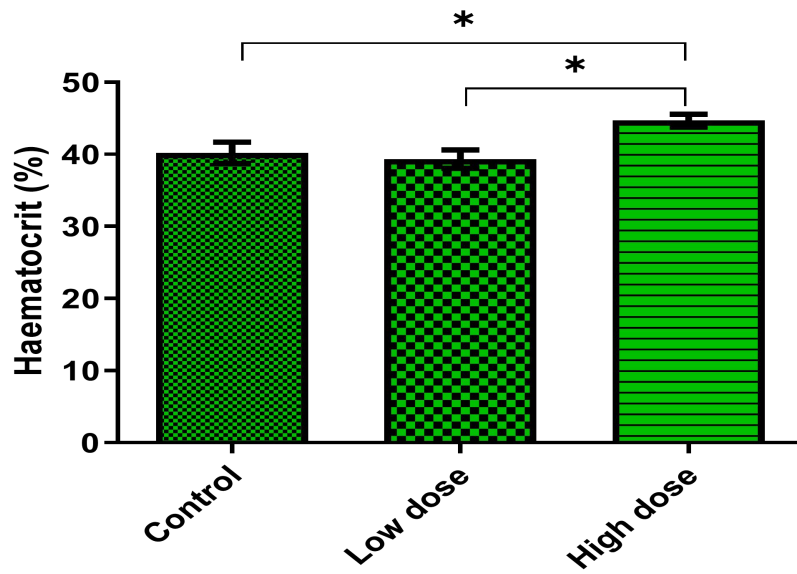
There were no significant differences in low and high doses compared with control

**Chart II: Effect of methanolic extract of *Occimum gratissimum* on haemoglobin concentration of male Abino rats**



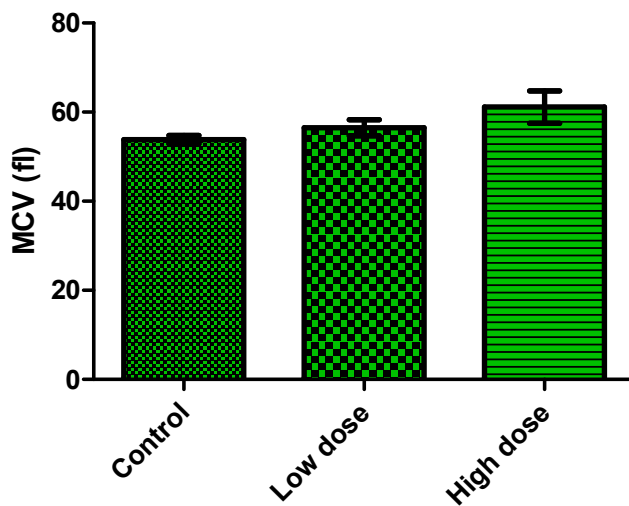
There were no significant differences in low and high doses compared with control

**Chart III: Effect of methanolic extract of *Occimum gratissimum* on haematocrit level of male Abino rats**



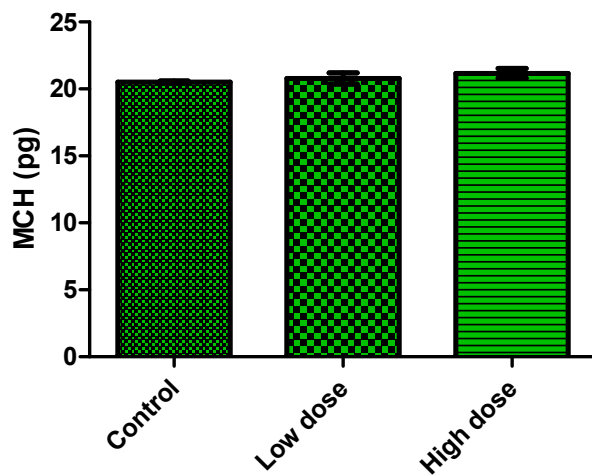
There was a significant increase in high dose compared with control and low dose respectively.

**Chart IV: Effect of methanolic extract of *Occimum gratissimum* on mean cell volume of male Abino rats**



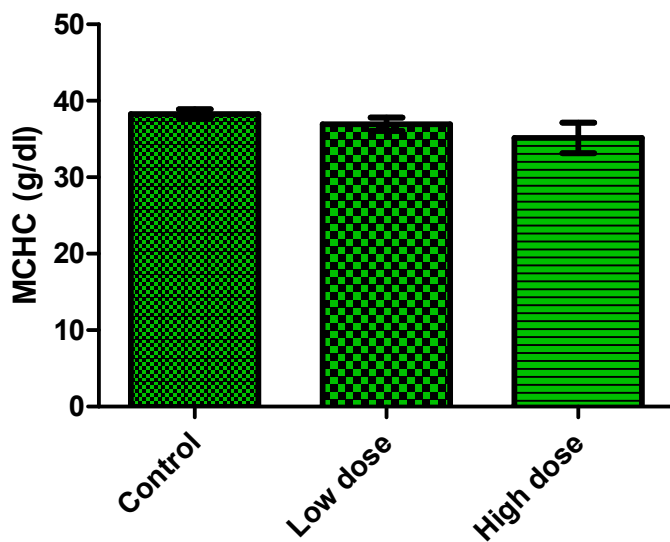
There were no significant differences in low and high doses compared with control

**Chart V: Effect of methanolic extract of *Occimum gratissimum* on mean corpuscular haemoglobin volume of male Abino rats**



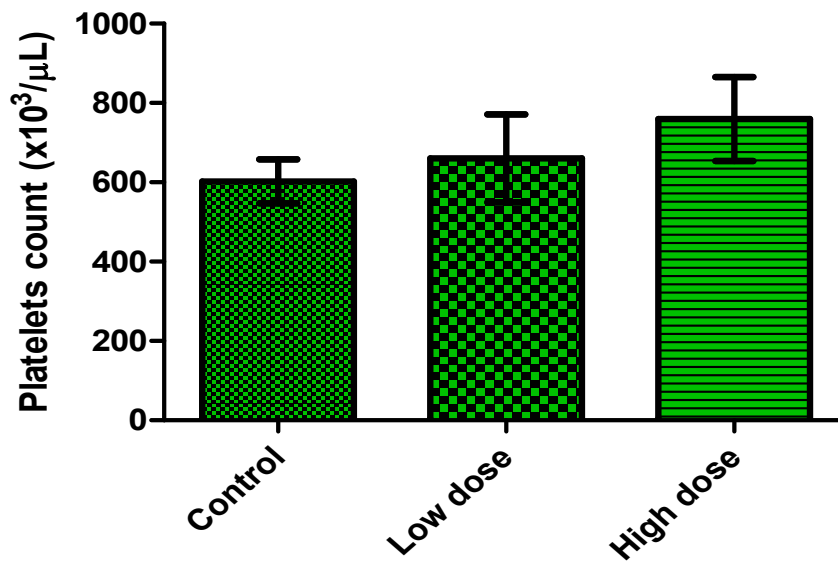
There were no significant differences in low and high doses compared with control

**Chart VI: Effect of methanolic extract of *Occimum gratissimum* on mean corpuscular haemoglobin concentration of male Abino rats**



There were no significant differences in low and high doses compared with control

**Chart VII: Effect of methanolic extract of *Occimum gratissimum* on platelet count of male Abino rats \***



There was a significant difference in low and high doses compared with control

## CHAPTER FIVE

### DISCUSSION AND CONCLUSION

#### 5.0 DISCUSSION

Haematological measures are helpful indicators for determining the negative effects of plant extracts or medications on the components of the blood (Ashafa et al., 2010). Determined hematological parameters are used to evaluate an animal's level of wellbeing (Ajayi and Raji, 2012). Therefore, haematological measures are accurate predictors of an animal's physiological and metabolic state (Khan and Zafar, 2005). The proportion (percent) of red blood cells in

blood is measured by packed cell volume (PCV), also known as haematocrit (Ht or Hct) or erythrocyte volume fraction (EVF) (Purves et al., 2003). It gauges the volume of red blood cells as a percentage of total blood volume; anemia is linked to a lack of synthesis of red blood cells (Guenter & Lawrence, 2005). Following administration of *O. gratissimum* methanol extract at 100 and 300 mg/kg body weight, the non-significant differences in MCV, haemoglobin, MCH, RBC, HCT and MCHC levels suggest that the extract may not contain any bioactive ingredients that could cause iron deficiency anemia, according to the findings of the current study. As a result, the fact that MCH, MCHC, MCV, and Hb are unaffected by the administration of methanolic extract of *ocimum gratissimum* at doses of 100 and 300 mg/kgbw raises the possibility that there were no alteration in the morphology and fragility of RBCs.

In this study, treatment with the plant extract led to a significant increase in PLTs in rats administered with 100 and 300 mg/kgbw of the methanol extract. According to Dasofunjo *et al.*, (2020), increase in platelets in experimental rats indicates good action on the blood's oxygen transporting ability as well as thrombopoietin. The observed increase in the platelets in this study indicates that the extract may improve the blood oxygen transporting ability. Likewise, the oxygen-transporting ability of the blood and the oxygen supplied to the tissues as well as hematosynthesis may be improved following the administration of the extract.

## 5.1 CONCLUSION

From the data obtained, it is safe to conclude that the extract of *Ocimum gratissimum* might be a panacea in the management of anaemic conditions due to its erythropoietic, and/or haematopoietic effects and also indicates good action on the blood's oxygen transporting ability as well as thrombopoietin, putting into consideration that there was no alteration in the morphology and fragility of the RBCs.

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