ISSN: 2536-7153 SJMLS

Sokoto Journal of Medical Laboratory Science 2023; 8(4): 113 - 123

SJMLS - 8(4) - 014

Immunomodulatory Effects of *MANGIFERA INDICA* on Haematological Parameters and Kidney Architecture of Breast Cancer-Induced Albino Rats.

Alhassan Hussaini Mohammed*[1], Usman Malami Aliyu[2], Isiyaku Adamu[1], Samuel Bamaiyi Dadzie[1], Abdulrahman Abdullatif[1], Abdullahi S. Mainasara[3], Yakubu Abdulrahman[4], and Hamisu Abdullahi[1]

Department of Immunology, Faculty of Medical Laboratory Sciences, Usmanu Danfodiyo University, Sokoto-Nigeria[1], Department of Radiotherapy and Oncology, Usmanu Danfodiyo University Teaching Hospital, Sokoto-Nigeria[2], Department of Chemical pathology and Immunology, Faculty of Basic Medical Sciences, College of Health Sciences, Usmanu Danfodiyo University, Sokoto-Nigeria[3], Department of Haematology, Faculty of Medical Laboratory Sciences, Usmanu Danfodiyo University, Sokoto-Nigeria[4].

Author for Corresponding*: halhassanmohd@gmail.com/ hussaini.alhasan@udusok.edu.ng/ +234-803-970-5336

https://dx.doi.org/10.4314/sokjmls.v8i4.14

Abstract

Breast cancer is one of the most common cancers found in women worldwide, accounting for more than half a million deaths in 2015. Over 1.5 million women are diagnosed with breast cancer every year throughout the world. Breast cancer is a metastatic cancer which is commonly transferred to distant organs such as the bone, liver, lung and brain. Biological therapies have been developed in recent years but despite that, a lot needs to be done in addressing the treatment for these strata of patients. The present study investigated the effects of *Mangifera indica* in the modulation of haematological indices in breast cancer-induced albino rats. Twenty-four female albino rats were used for the study, and they were grouped into six groups of four albino rats each. Group 1 (normal control) was fed with normal growers feed. Group 2-6 were orally administered 65 mg/kg body weight of DMBA to induce tumour. Group 2 was untreated; Group 3 was treated with a daily intraperitoneal injection of ascorbic acid [12 mg/kg/day] for 2 weeks. Groups 4- 6 were treated with 500,1000 and 1500 mg/kg body weight of *M. indica* stem bark extract. The animals were sacrificed 24hrs after the last treatment. The present study reveals that the plant extract was able to reduce anaemia and inflammation posed as a result of breast cancer. WBC, Lymphocyte, RBC and PCV were significantly upregulated in the group treated across most of the group treated with the various doses of *M. indica*. *M. indica* may possess immunosuppressive and immunomodulatory potentials and may be of benefit in the management of breast cancer.

Keywords: Breast cancer, Mangifera indica, DMBA, Ascorbic acid

Introduction

Breast cancer is one of the most common cancers in women worldwide, accounting for approximately 570,000 deaths in 2015. Over 1.5 million women (25% of all women with cancer) are diagnosed with breast cancer globally (WHO, 2014). In Africa, it is estimated that 30% of all new cancer cases among women are breast cancer in 2017 (Siegel *et al.*, 2016). Early diagnosis of the disease can lead to a good prognosis and a high survival rate. In North America, the 5-year relative survival rate of breast cancer patients is above 80% due to the timely detection of this disease (WHO, 2014). There are numerous risk factors such as gender, aging, estrogen, family history, gene mutations and unhealthy lifestyle, which can increase the possibility of developing breast cancer (Majeed *et al.,* 2014). Most breast cancer occurs in women and the number of cases is 100 times higher in women than that in men (Siegel *et al.*, 2016).

Chemically induced rodent models of mammary cancer have been extensively used over the years to emulate human breast carcinogenesis. All models in mice and rats have specific advantages and limitations. Mammary tumors can be induced in susceptible rat strains after single doses of carcinogens such as DMBA or nitrosomethylurea (NMU). Rat tumors are not extremely invasive beyond the mammary fat pad, have short latency, seldom metastasize and are highly hormone-dependent and for that reason were widely used as models of estrogen

dependent breast cancers (Abba *et al*., 2014). On the other hand, most mouse strains are far more resistant than rats to chemical induced mammary gland carcinogenesis, typically requiring multiple doses of carcinogens such as DMBA and developing only after a long latency. In 1996 it was observed that MPA was not carcinogenic per se but in combination with DMBA mouse mammary tumors developed with a much shorter latency and requiring a lower number of DMBA doses (Alhassan *et al*., 2021).

Plants produce a wide range of chemical compounds that apparently have no direct role in the plants growth. These compounds are called secondary metabolites. Alkaloids, terpenoids, flavonoids, pigments, and tannins are important constituents of these compounds. Secondary metabolites have biologic effects such as anticancer, contraceptives, and different effects on haematopoeitic, immunological cells (Mansouri *et al*, 2015) and cardiovascular system (Kooti *et al.*, 2016). About 80% of the rural African population almost exclusively uses traditional medicine for its primary health care needs. For cultural and economic reasons, medicinal plants constitute a major part of traditional medicine. Globally, more than 3,000 plants have been reported to exert cytotoxicity toward cancer cells In the recent years, numerous African medicinal plants have been screened for their cytotoxic potential (Alhassan *et al*., 2021). Approximately 60% of drugs currently in use for mammary cancer treatment have been isolated from natural products and the plant kingdom constitute the the most significant (Gordaliza, 2017).

Mangifera indica popularly known as mango tree in English, is a plant that is locally abundant in Nigeria, particularly in towns and villages in the Savanna zones. Traditionally, *M. indica* stem bark has been employed in the treatment of several ailments like skin irritation, abnormal lumps or mast tissue (tumour), neck and breast. It is also used in the treatment of pyrexia of unknown origin. Extract of the bark, leaves and unripe fruit have demonstrated antibiotics, anti-inflammatory and in treatment for ulcers and anti-snake venom (Shah *et al.,*2011).

Differential leukocyte count is one of the most common laboratory tests performed today. It provides information about the immune system through the evaluation of the white blood cell (WBC) count with differential. These tests are helpful in diagnosing; anaemia, certain cancers, infection, acute haemorrhagic states, allergies, and immunodeficiency as well as monitoring the side effect of certain drugs that causes blood dyscrasias. (Alhassan *et al*., 2021).

Researches into breast cancer in humans in various populations have shown an increase in morbidity and mortality of the breast cancer patients. Despite the considerable efforts, mammary cancer still remains an aggressive killer globally, due to the fact that, novel synthetic chemotherapeutic agents have not succeeded in fulfilling expectations despite the considerable cost of their development (Adawin *et al.,* 2017). There is therefore the need to develop new, effective, and affordable mammary anticancer drugs (Coseri *et al.,* 2018). The aim of this research was to determine the effects of *M. indica* in the modulation of immunological and haematological indices in breast cancer-induced albino rats.

Materials and Methods

Study Area

This research was carried out in the Department of Immunology, School of Medical Laboratory Science, UDUS and Faculty of Pharmaceutical Science, Usman Danfodiyo University Sokoto, Nigeria.

Plant Collection and Identification

Fresh bark of *Mangifera indica* was collected with an axe, from Sokoto North Local Government, Sokoto State. The plant collected was authenticated in the Department of Botany, Usman Danfodiyo University Sokoto, Nigeria.

Plant Extraction

Mangifera indica stem bark was shade-dried at room temperature, pounded into powder and extracted with methanol (50%□w/v) by gentle stirring in a covered beaker for 24hours at room temperature. The supernatant of the extracts was filtered, and methanol evaporated in a chemical hood for four days. The evaporated extract was lyophilized till dryness and kept at 4°C till use.

Stock solutions of the plant extract (200mg/mL) was prepared by weighing the powder and dissolving it in 10% dimethylsulphuroxide (DMSO). The solution was divided to aliquots and kept at - 20°C, until used (Alhassan *et al*., 2021).

Experimental animals

Female albino rats (10 weeks old) were purchased from the Veterinary Department of Usman Danfodio University Sokoto and were allowed to acclimatize for one week. The experimental house was cleaned, disinfected and fumigated before the commencement of the experiment. Cleaning of the animal cages was carried out daily, and on a regular basis. All the experimental protocols followed the Animal Ethics Committee Guidelines of Usmanu Danfodiyo University, Sokoto.

Study Design

Twenty-four female albino rats were used for the study, and they were grouped into six (6) groups of four (4) albino rats each

Table 1: Study Design

Groups	Induction of Tumour	Intervention
Grp 1	The albino rats in this were not induced with DMBA	The rats were fed with normal growers and normal saline throughout the period of the research.
Grp 2	The rats were induced with 65mg/kg body weight of DMBA once, and observed for 4weeks.	The rats in this group were not treated with either conventional drug nor extract. There were only fed on water and normal feeds.
Grp 3	The rats were induced with 65mg/kg body weight of DMBA and observed for 2weeks.	The rats were treated with a daily intraperitoneal injection of 12mg/kg body weight of Ascorbic acid for 2weeks.
Grp 4	The rats were induced with 65mg/kg body weight of DMBA and observed for 2weeks.	The rats were treated with a daily dose of 500mg/kg body weight of *M. indica* stem bark extract, daily for 2weeks
Grp 5	The rats were induced with 65mg/kg body weight of DMBA and observed for 2weeks.	The rats were treated with a daily dose of 1000mg/kg. body weight of *M. indica* stem bark extract, daily for 2weeks
Grp 6	The rats was induced with 65mg/kg body weight of DMBA and observed for 2weeks.	The rats were treated with a daily dose of 1500mg/kg body weight of *M. indica* stem bark extract, daily for 2weeks

Blood Sample Collection and Processing

The animals were sacrificed 24hrs after the last treatment. Blood samples were collected in sterile well-labeled EDTA bottles. The samples were mixed gently and taken to the laboratory for analysis immediately after collection. The kidney was collected into formalin for H/E staining.

Laboratory Analysis.

Full Blood Count

The immunological and hematological parameters were estimated using Sysmex KX-21N which is 3-part differential haematology analyzer. This analyzer has the capacity to differentiates, the white blood cells into three populations, based on their sizes. These are neutrophils, lymphocytes, and mixed population which consists of eosinophils, monocytes, and basophils. Other parameters include red blood cell count, white blood cell count, haemoglobin and haematocrit value, mean cell volume, mean cell haemoglobin and mean cell haemoglobin concentration.

Tissue Processing

Tissue sample of the kidney was manually processed at Histology Laboratory of Faculty of Medicine, Department of Anatomy Ahmadu Bello University, Zaria (ABU) following the

method of Bancroft and Steven 2008 using Automatic tissue processor.

Haematoxylin and Eosin (H & E) Staining Technique

Assay Principle

The principle behind H&E stain is the chemical attraction between tissue and dye. Hematoxylin, a basic dye imparts blue-purple contrast on basophilic structures, primarily those containing nucleic acid moieties such as chromatin, ribosomes and cytoplasmic regions rich in RNA. Acidic eosin counterstains the basic elements such as RBCs, cytoplasm, muscle and collagen in varying intensities of pink, orange and red (Giri, 2018).

Assay Procedure

Tissue sections were dewaxed in xylene two changes per minute each after then Sections were dehydrated through descending grades of alcohol (100 %, 90 % and 70%) respectively. Sections were stained in Harris haematoxylin stain for 8 minutes thereafter; sections were rinsed in running tap water for 30 seconds.

They were briefly differentiated in 1% acid alcohol, and sections were blued in Scott's tap water, they were rinsed in running tap water for another 30 seconds. After that, sections were counterstained in 1% Eosin for 2 minutes. were dehydrated through ascending grades of alcohol (70%, 90% and 100%) and were cleared in xylene briefly after then, sections were mounted in DPX and air-dried. Sections were examined using Olympus binocular light microscope (Ochei and Kolhatkar, 2008).

Statistical analysis

The results obtained were entered into SPSS version 21 for analysis. Continuous variables were expressed as mean ± standard deviation (SD) as well as percentage. Test for normality was performed to ascertain normal distribution of the variables. The p value ≤ 0.05 was used to determine the level of statistical significance.

Result

The Data were stated as mean ± SEM and analyzed using One-way Analysis of Variance (ANOVA) using SPSS. A level of $p \leq 0.05$ was considered statistically significant.

Effects of MISBE On Haematological Indices

Table 2: Haematological and Immunological Parameters

Groups	WBC	NEU	LYM	EOSIN	MONO	BASO
Group 1	12.52 (1.31)	29.33 (3.65)	67.33 (2.07)	1.44 (0.26)	1.20 (0.46)	0.07 (0.05)
Group 2	17.28 (2.53)^{a}	31.33 (6.87)^{b}	62.33 (9.34)^{b}	3.22 (0.63)^{a}	3.12 (0.13)^{a}	0.04 (0.03)^{a}
Group 3	20.05 (1.29)^{a}	29.33 (5.50)^{b}	71.00 (5.55)^{a}	2.66 (0.32)^{b}	2.20 (0.42)^{b}	0.03 (0.02)^{b}
Group 4	16.13 (0.65)^{a,d,f}	21.33 (1.14)^{b,d,f}	65.00 (1.05)^{b,d,e}	2.55 (0.44)^{b,d,f}	2.41 (0.35)^{b,d,f}	0.05 (0.03)^{b,d,f}
Group 5	12.98 (1.04)^{b,d,f}	24.33 (1.04)^{a,c,e}	73.23 (4.47)^{a,d,e}	2.23 (0.25)^{b,c,f}	2.01 (.60)^{b,c,f}	0.045(0.01)^{b,c,f}
Group 6	19.00 (1.29)^{a,d,f}	32.00 (3.72)^{a,d,f}	66.33 (1.20)^{b,d,e}	1.66 (0.22)^{b,c,f}	1.29 (0.55)^{b,c,f}	0..06 (0.02)^{b,c,f}

Table 1 shows Immunological cell indices (white blood cell, Neutrophil, lymphocyte, Eosinophil, Monocyte, and Basophils). There was statistically significance ($p \leq 0.05$) in the White blood cells, Neutrophil between groups. and Lymphocyte when compare with those treated with 65 mg/kg b.w. dimethylbenz (α) anthracene and a daily intraperitoneal injection of ascorbate (12 mg/kg/day) for 18 days; Group 3.

Effects of MISBE on Erythropoietic Indices

Table 3: Erythropoietic Indices

Groups	RBC	HGB	PCV	MCV
Group 1	6.10 (0.66)	10.90 (0.30)	32.80 (18.64)	69.38 (4.13)
Group 2	6.36 (0.27) [b]	8.85 (4.93) [b]	45.18 (4.02) [a]	71.30 (6.08) [b]
Group 3	5.89 (0.05) [b]	10.2 (0.87) [b]	38.85 (1.38) [a]	69.68 (2.15) [b]
Group 4	6.89 (0.24)[b,d,f]	12.10 (0.49)[b,d,f]	43.65 (1.37)[a,d,f]	66.08 (2.12)[b,d,f]
Group 5	6.47 (0.33)[b,d,f]	11.65 (0.60)[b,d,f]	43.33 (3.69)[a,d,f]	74.05 (3.62)[b,d,f]
Group 6	6.61 (0.38)[b,d,f]	12.10 (0.35)[b,d,f]	41.40 (1.54)[a,d,f]	62.35 (0.87)[a,c,e]

Table 2 shows the erythopoietic indices of red blood cell, haemoglobin, Packed cell volume, and mean cell volume. There was no statistically significant difference ($p \leq 0.05$) in the red blood cell and the haemoglobin but there was statistical increase ($p \leq 0.05$) in the haematocrit values when compare with the control. *Values are stated as Mean (Standard Deviation), p-value = 0.05 using Chi-Square.*

Effects of MISBE on Red Cell and Platelet Concentrations

Table 4: Red Cell and Platelet Concentrations

Groups	MCH	MCHC	PLT
Group 1	18.15 (2.01)	25.95 (1.59)	823.00 (81.33)
Group 2	17.88 (0.73) [b]	25.08 (1.27)[b]	624.00 (67.27) [a]
Group 3	18.50 (1.18) [b]	27.33 (1.27)[b]	669.00 (23.07) [a]
Group 4	18.30 (0.41)[b,d,f]	27.70 (0.44)[b,d,f]	701.00 (19.84) [a,d,f]
Group 5	18.93 (1.25)[b,d,f]	27.38 (0.73)[b,d,f]	889.75 (14.18) [a,c,e]
Group 6	18.33 (0.78)[b,d,f]	29.40 (1.12)[a,c,f]	740.50 (39.18) [a,c,f]

Table 3 indicates the statistical description of, mean cell haemoglobin and mean cell haemoglobin concentration and Platelet of various groups. There was no statistical distinction ($p \leq 0.05$) between various group with exception of Group 6; treated with 65 mg/kg b.w. dimethylbenz (α) anthracene and 1500 mg/kg b.w. methanol extract of *Mangifera indica* stem bark. There was statistically significant ($p \leq 0.05$) increase in mean cell volume and decrease in mean cell haemoglobin concentration. *Values are stated as Mean (Standard Deviation), p-value = 0.05 using Chi-Square.*

KEY

WBC = White blood cells, RBC = Red blood cells, HGB = Hemoglobin, HCV = Hematocrit, MCV = Mean Cell Volume, MCH = Mean Cell Hemoglobin, MCHC = Mean Cell Hemoglobin Concentration, PLT = Platelet, LYM = Lymphocyte, MXD = Monocyte, NEU = Neutrophil, are stated as Mean (Standard Deviation), p-value = 0.05 using Chi-Square.

3.4 Effect of MISBE on Kidney (H&E 250)

Fig. 1 is the photomicrograph of normal renal section from a healthy control of experimental Albino Rat, showing normal histological architecture. The renal cortex shows normal Malpighian renal corpuscle containing glomerulus and Bowman's space. The proximal convoluted tubules have narrow lumina and are lined with normal cuboidal cells with rounded vesicular basal nuclei and deeply acidophilic cytoplasm. The distal convoluted tubules have normal lumina lined with cubical cells with rounded vesicular central nuclei and paler acidophilic cytoplasm.

Fig. 2 is the photomicrograph of renal section from a Positive Control of experimental Albino Rat (65 mg DMBA) reveals cortex with complete destruction of the renal corpuscles with loss of glomerular tufts, the tubular lining cells show vacuolated cytoplasm with pyknotic nuclei and desquamation of the lining epithelium. Most of the tubular lumina contain cellular debris and few red blood cells with infiltrated inflammatory cells showing that the tumour has already been inducted.

Fig. 3 is the photomicrograph of renal section of 65 mg DMBA+100mg Ascorbic acid groups, which shows apparently normal renal corpuscle with glomerular tuft infiltrated with high inflammatory cells with few tubular cells that shows cytoplasmic vacuoles, and nearly all tubular cells have vesicular nuclei.

Fig. 4 is the photomicrograph of renal section of 65 mg DMBA+500mg MISBE shows renal cortex that reveals apparently normal renal corpuscle with glomerular tuft, with renal corpuscles, infiltrated with few inflammatory cells. Most of the tubular cells show cytoplasmic degeneration and nearly all tubular cells have vesicular nuclei.

Fig. 5 is the photomicrograph of renal section of experimental Albino Rat (65mg DMBA+1000 mg MISBE) shows the renal cortex with apparently normal renal corpuscle that reveals glomerular tuft within the bowman's capsule, The proximal convoluted tubules have normal narrow lumina and are lined with cuboidal cells with rounded vesicular basal nuclei. The distal convoluted tubules have wider lumina and are lined with cubical cells with rounded vesicular central nuclei. No inflammatory infiltration of cells was observed which shows that MISBE has an active anticancer effect.

Fig. 6 is 65mg DMBA+1500mg MISBE, photomicrograph of renal section of experimental Albino Rat shows renal cortex with Malpighian renal corpuscle containing glomerulus and Bowman's space that appear normal. The proximal convoluted tubules show narrow lumina that are lined with cuboidal cells with rounded vesicular basal nuclei and deeply acidophilic cytoplasm. The distal convoluted tubules have wider lumina and are lined with cubical cells with rounded vesicular central nuclei. There was no inflammatory infiltration of cells observed which remain a pointer that MISBE has bioactive properties that can ameliorate breast cancer.

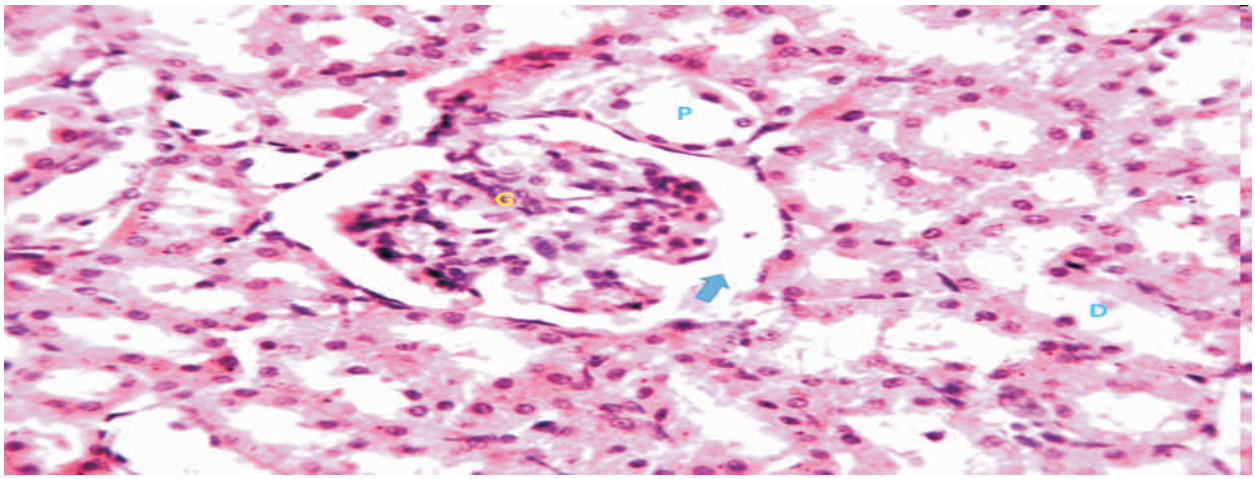


Fig. 1: Photomicrograph of normal renal section of experimental albino rats (Non DMBA) showing normal histological architecture. The renal cortex shows normal Malpighian renal corpuscle containing glomerulus (G) and Bowman's space (arrowhead). The proximal convoluted tubules (P) have narrow lumina and are lined with normal cuboidal cells with rounded vesicular basal nuclei and deeply acidophilic cytoplasm. The distal convoluted tubules (D) have normal lumina lined with cubical cells, rounded vesicular central nuclei and paler acidophilic cytoplasm. H&E x250.

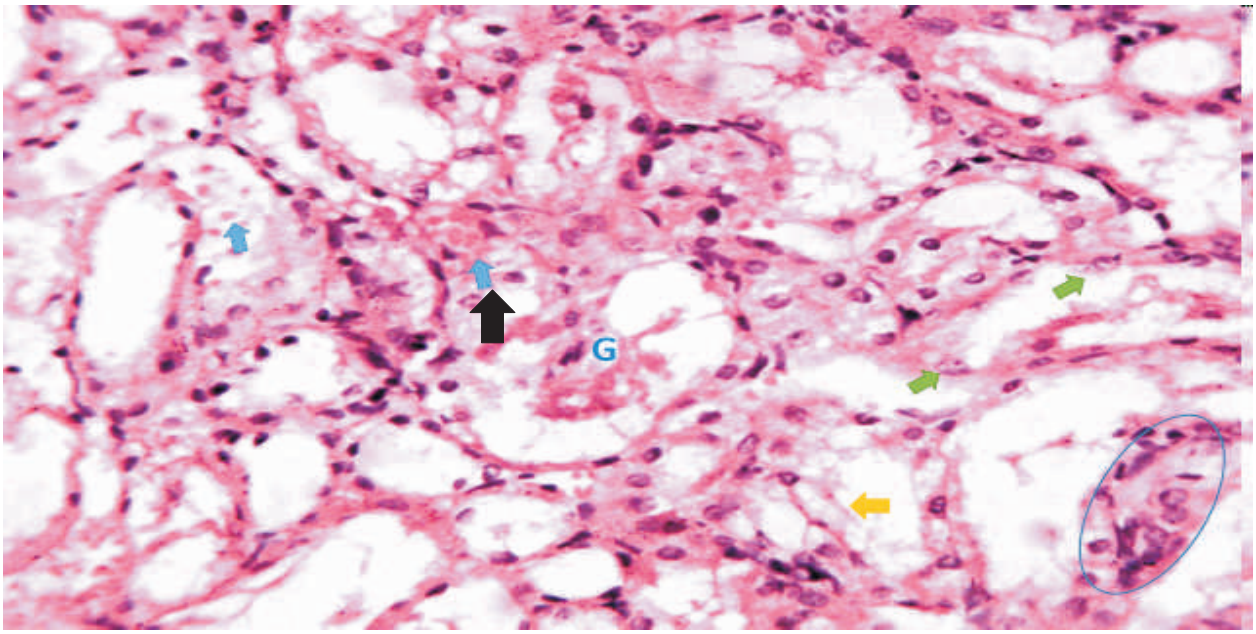
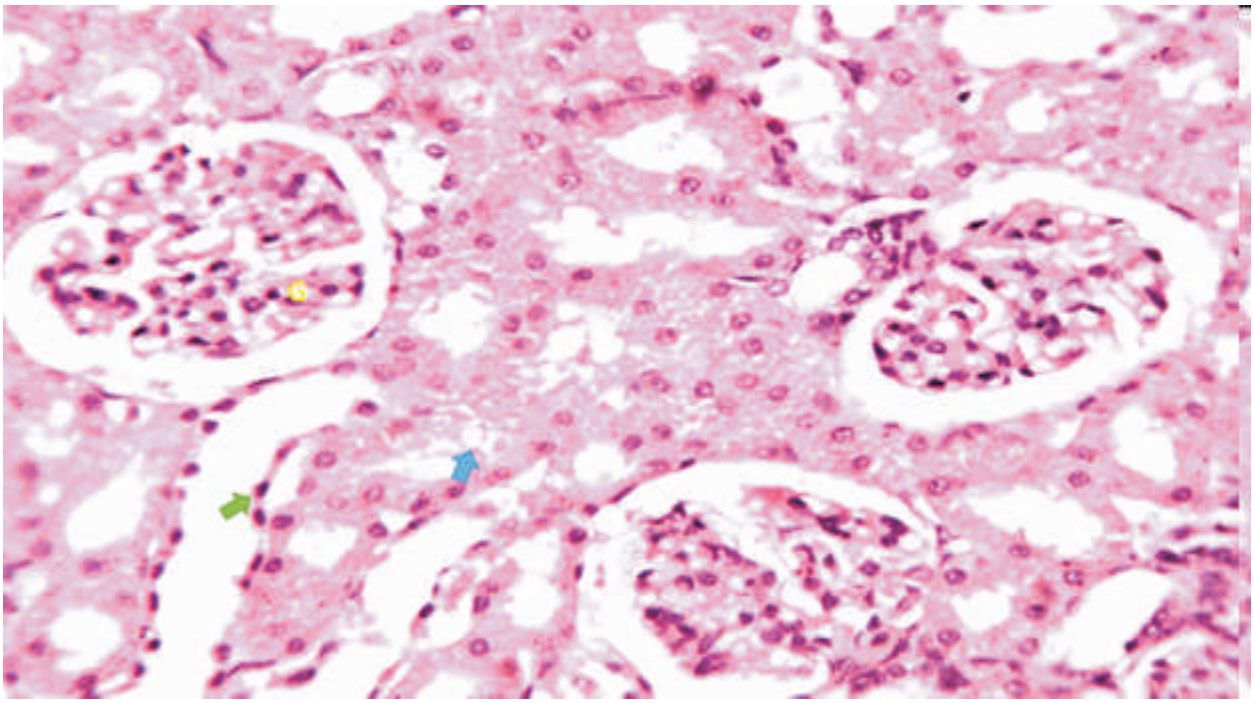
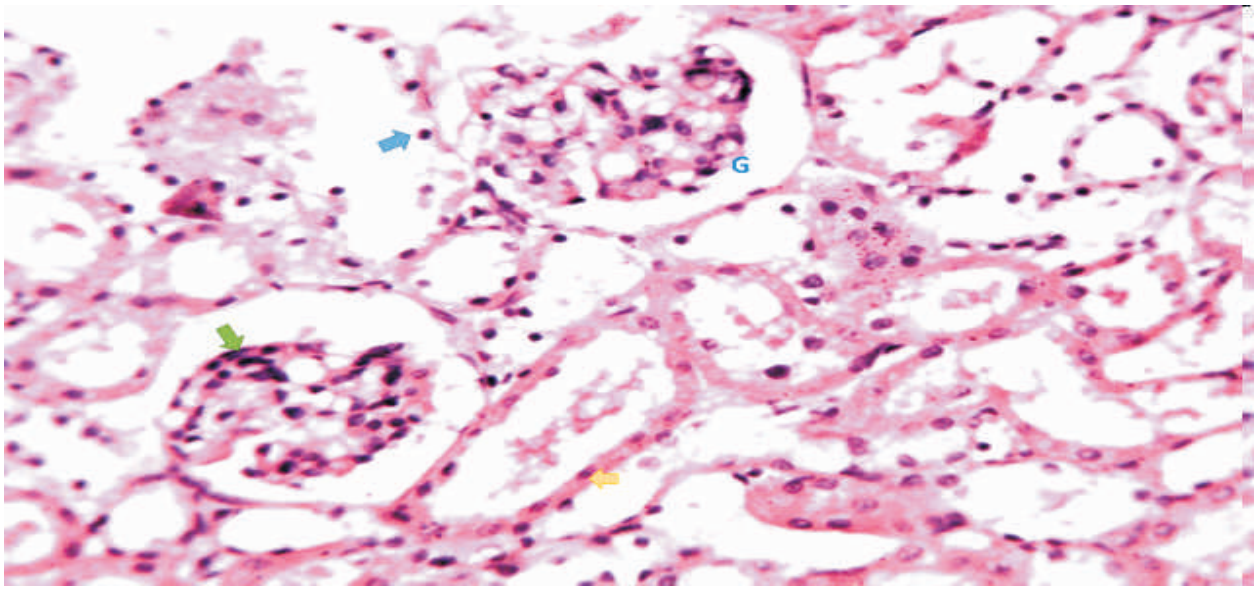


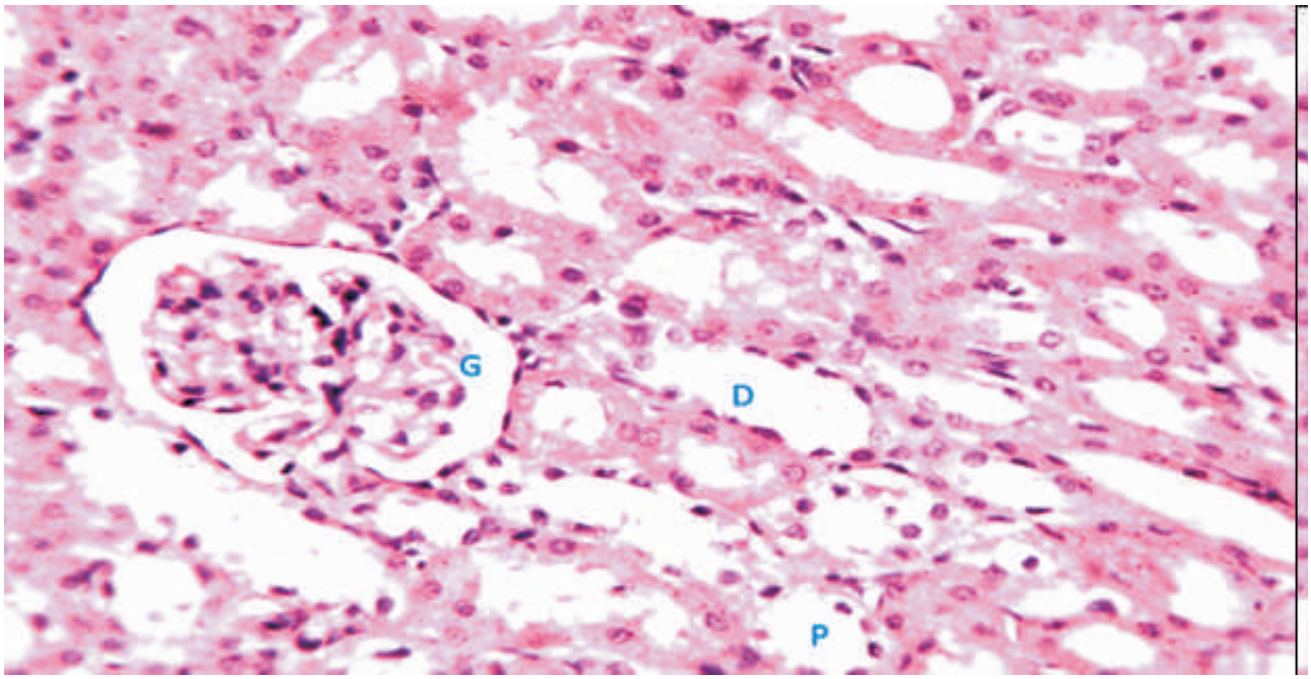
Fig. 2: Photomicrograph of renal section from a positive control of experimental Albino Rats (65 mg DMBA/single dose). The cortex reveals complete destruction of the renal corpuscles with loss of glomerular tufts (G), the tubular lining cells show vacuolated cytoplasm (blue arrow head).The pyknotic nuclei (green arrows) and desquamation of the lining epithelium (yellow arrow). Most of the tubular lumina contain cellular debris, few red blood cells (blue arrow) and infiltrated inflammatory cells (black arrow) showing that the tumour has already being inducted. H&E x250



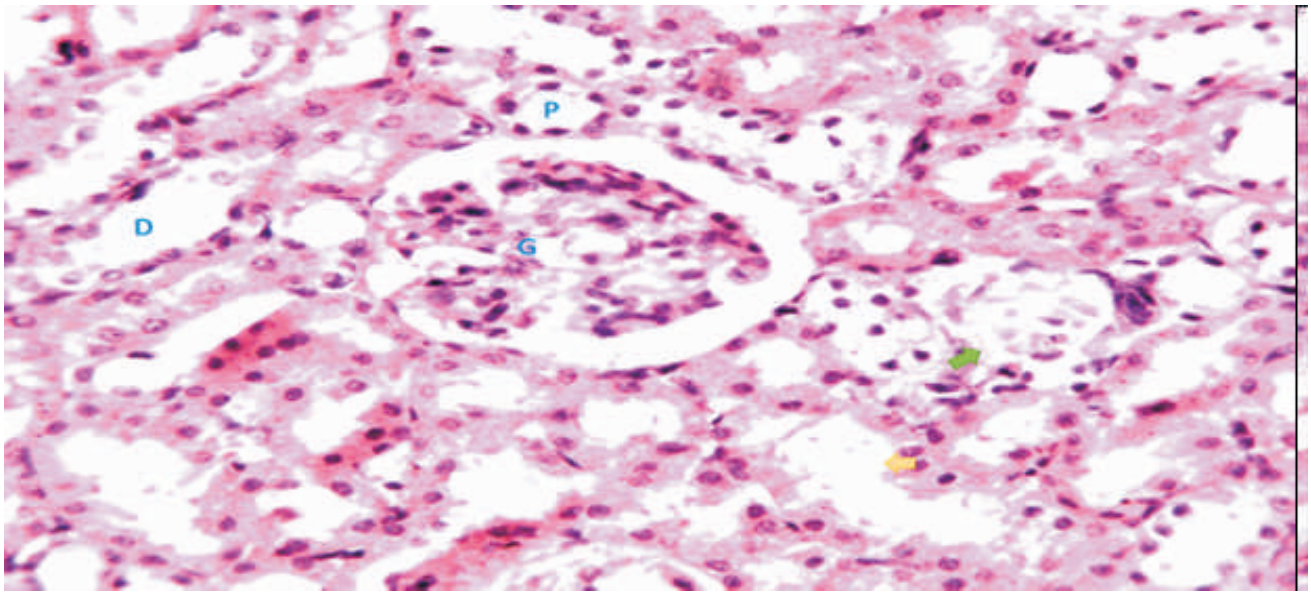
Ffig. 3: Photomicrograph of renal section of experimental Albino Rat (65 mg DMBA + 100 mg AA). The renal cortex shows apparently normal renal corpuscle with glomerular tuft infiltrated with high inflammatory cells (G), few tubular cells show cytoplasmic vacuoles (blue arrow), and nearly all tubular cells have vesicular nuclei (green arrows).H&E x250



Ffig. 4: Photomicrograph of renal section of experimental Albino Rat (65 mg DMBA + 500 mg MISBE). The renal cortex shows apparently normal renal corpuscle with glomerular tuft (G), with renal corpuscles (green arrow). Infiltrated with few inflammatory cells. Most of the tubular cells show cytoplasmic degeneration (blue arrow), and nearly all tubular cells have vesicular nuclei (yellow arrow). H&E x250.



Ffig. 5: Photomicrograph of renal section of experimental Albino Rat (65 mg DMBA + 1000mg MISBE). The renal cortex shows apparently normal renal corpuscle with glomerular tuft (G) within the bowman's capsule, The proximal convoluted tubules (P) have normal narrow lumina and are lined with cuboidal cells with rounded vesicular basal nuclei. The distal convoluted tubules (D) have wider lumina and are lined with cubical cells with rounded vesicular central nuclei. There was **no** inflammatory infiltration of cells observed. H&E x250



Ffig. 6: Photomicrograph of renal section of experimental Albino Rat (65 mg DMBA + 1500 mg MISBE). The renal cortex shows Malpighian renal corpuscle containing glomerulus (G) and Bowman's space (arrowhead) appear normal. The proximal convoluted tubules (P) have narrow lumina and are lined with cuboidal cells with rounded vesicular basal nuclei and deeply acidophilic cytoplasm. The distal convoluted tubules (D) have wider lumina and are lined with cubical cells with rounded vesicular central nuclei. There was no inflammatory infiltration of cells observed. H&E x250

Discussion

White blood cells are vital components of the blood; their role is to fight infection. A high white blood cell count indicates that the immune system is working to destroy an infection. A white blood cell (WBC) below the normal, pose risk for infection (James, 2013), whereas a higher WBC count increases risk of developing invasive breast cancer (Park *et al*., 2019). Our studies shows a statistical significance ($p \leq 0.05$) in the white blood cells between groups 1 Normal Control rats fed orally with phosphate buffer saline (PBS) and group 2 treated with 65 mg/kg b.w. dimethylbenz (α) anthracene and fed orally with PBS when compared. The three groups (groups 4, 5 and 6) treated with methanol extract of *Mangifera indica* stem bark showed an elevation in white blood cell count when compared to the normal control. This commensurate with the findings of park *et al*. (2019) whose study demonstrated an increase in white blood cell count. The extracts of *Mangifera Indica* stem bark have some positive effect on the hemopoietic system of normal rats (Park *et al.,* 2019). *The methanol extract demonstrated* a significant decrease ($p \leq 0.05$) in neutrophil and platelets, in the treated groups when compared to the DMBA group. This is similar to the findings of Siniorakis *et al*. (2017) that there was a decrease in neutrophil, N/L, P/L and platelets level in the treated groups treated with other plant extracts when compared to the DMBA group (though not significant, $p \leq 0.05$).

All the groups treated with methanol extract of *Mangifera indica* stem bark demonstrated statistical significance when compared to members of group 3 treated with 65 mg/kg b.w. dimethylbenz (α) anthracene and a daily intraperitoneal injection of ascorbate (12 mg/kg/day) for 14 days. This indicates the functionality of the extract. Thus, members of group 3 treated with a daily intraperitoneal injection of ascorbate (12 mg/kg/day) for 18 days were significant when compared with the normal control. Lymphocytes fight cancer, low levels of lymphocytes in the blood encourages a relapse and decreased survival rates, while higher lymphocyte count increases the overall survival (Akinbami *et al*., 2013).

The mixed cells consist of eosinophil, basophil and monocyes. Members of group 5 and 6 were significant when compared to the disease control.Red blood cell, haemoglobin and packed cell volume are markers of anaemia, predicting the increased danger of cancer patients' death due to heart attack (Akinbami *et al*., 2013, Arika *et al*.,

2016). There was no statistical significant difference ($p \leq 0.05$) in the red blood cell and the haemoglobin but there was statistical increase ($p \leq 0.05$) in the haemotocrit values when compared with the normal control. Anaemia is observed commonly in cancer patients, which might result from bleeding, nutritional deficiencies, damage of the bone marrow, tumour infiltration and malignancy (Ali, 2014). The methanol extract of *Mangifera indica* stem bark have shown to have anti-anaemic properties. This commensurate with the findings of (Nwinuka *et al*., 2005) which demonstrated that the aqueous extract of *Mangifera indica* increased the heamatocrit value of normal albino rats.

There was statistically significant ($p \leq 0.05$) increase in mean cell volume in the red blood cells in the positive control making them macrocytic when compared with the the treatment group of the methanol extract of *Mangifera indica*.

Table 4.4 indicates the platelet concentration of various groups in the study, the statistical analysis indicates statistical difference ($p \leq 0.05$) in most of the groups when compared to the group treated with ascorbic acid. A high platelet count is associated with prognosis of gynecological cancers (Park et al., 2019), since it secretes various growth factors and cytokines that promote angiogenesis which is a critical step in breast cancer metastasis (Alhassan et al.,2021). Group 5; treated with 65 mg/kg b.w. dimethylbenz (α) anthracene and 1000 mg/kg b.w. methanol extract of *Mangifera indica* stem bark, shows obvious increase in platelet concentration. This is in contrast to Akuru *et al.* (2019), the difference may be as a result of the methodology used and the difference in the extract used.

Conclusion/Recommendation

Conclusion

The methanol extract of *Mangifera indica* stem bark have been able to increase red blood cell indices indicating its anti-anaemic properties, it also stabilized white blood count indicating its ability to suppress tumour. However, from this study the methanol extract of *Mangifera indica* stem bark has proved to possess effective healing properties in breast cancer management.

Recommendation

Considering the findings of the present study, it could be recommended that: Immunological and haematological parameters should be assessed periodically in breast cancer patients as this could be useful in the early diagnosis and management of breast cancer patients. Further studies are necessary to focus on using plants and plant extract to manage breast cancer.

Conflict of Interest:

There is no conflict of interest to declare.

References

Abba, M. Zhong, y., Lee, J., Kil, H., Lu, Y., Takata, Yoko & Simper, Melissa & Gaddis, Sally & Shen, Jianjun & Aldaz, C.Marcelo. (2014). DMBA induced mouse mammary tumors display high incidence of activating Pik3caH1047 and loss of function Pten mutations. Oncotarget; **7**: 10.18632/oncotarget.11733.

Adawin, D.E. (2017): Potential role of TNF alpha and TNF gama in the pathogenesis of tumor. *Tumor Immunology;* **101:** 212–217.

Alhassan, H.M., Yeldu, M.H., Safiya, Y., Usman M., Isiyaku, A., Mustapha, U.K., Ahmad H.M., and Hamisu A. (2021). Anti-tumour activities of *Allium sativum* on Serum IL-33 TNF-α and Breast tissue in Cancer Induced Wistar albino rats. *Archives of Immunology and Allergy*; **4(1)**: 4-15.

Ali, R. and Wendt, M. K., (2017) The paradoxical functions of EGFR during breast cancer progression. Signal transduction and targeted therapy. *Proceedings of the National Academy of Sciences of USA*; **2**: 16042.

Akinbami A, Popoola A, Adediran A, Dosunmu A, Oshinaike O, Adebola P, Ajibola S. (2013). Full blood count pattern of pre-chemotherapy breast cancer patients in Lagos, Nigeria. *Caspian Journal of Internal Medicine*. Winter; **4(1)**:574-579.

Arika, W.M., Nyamai, D.W., Musila, M.N., Ngugi, M.P., Njagi, E.N.M. (2016). Hematological Markers of In Vivo Toxicity. *Journal of Haematology and Thrombotic Disease*; **4**: 236. doi:10.4172/2329-8790.1000236.

Coseri, S. (2018). "Natural products and their analogues as efficient anticancer drugs," *Mini-Reviews in Medicinal Chemistry*;**9(5)**: 560–571.

Eunice A. Akuru, Chika I. Chukwuma, Chika E. Oyeagu, Ochuko L. Erukainure, Boitumelo Mashile3, Reaotshepa Setlhodi, Samson S. Mashele, Tshepiso J. Makhafola, Jeremiah O. Unuofin, Taiwo O. Abifarin,Thando C. Mpendulo (2021). Nutritional and phytochemical profile of pomegranate peel and its effects on hepatic oxidative stress and metabolic alterations. *Journal of Food Biochemistry, ISSN:* 1745-4514, DOI: 10.1111/jfbc.

Gordaliza, M. (2017)."Natural products as leads to anticancer drugs" . *Clinical and TranslationalOncology*; **9(12)**: 767–776.

Giri, B., Dey, S., Das, T., Sarkar, M., Banerjee, J., Dash, S.K. (2018). Chronic hyperglycemia mediated physiological alteration and metabolic distortion leads to organ dysfunction, infection, cancer progression and other pathophysiological consequences: An update on glucose toxicity. *Biomedical Pharmacotherapy*; **107**: 306-328. doi: 10.1016/j.biopha.2018.07.157.

James, L., Bauer, J. A., Chen, X., Sanders, M. E., Chakravarthy, A. B., Shyr, Y., Pietenpol, J. A. (2017). Identification of human triple-negative breast cancer subtypes and preclinical models for selection of targeted therapies. Journal of Clinical Investigation; **121 (7):** 2750–2767.

Kooti W, Hasanzadeh-Noohi Z, Sharafi-Ahvazi N, Asadi-Samani M, Ashtary-Larky D. Phytochemistry, pharmacology, and therapeutic uses of black seed (Nigella sativa). *Chinese Journal of Natural Medicine*; **14**:732-745.

Majeedi, S.F., Roqaiya, M., Jahan, D., Khan, A. A. (2015). Immunomodulatory herbs of Unani medicine: A review. *International Journal of Herbal Medicine*; **3(4)**: 19-21.

Mansouri E, Kooti W, Bazvand M, *et al*. (2015). The effect of hydroalcoholic extract of Foeniculum vulgare Mill on leukocytes and hematological tests in male rats. Jundishapur *Journal of Natural Pharmacological Production*;**10**: e18396.

Nwinuka, N.M., , G.I., Ibeh, G.O. (2005). Proximate Composition and Levels of Some Toxicants in Four Commonly Consumed Spices. *Journal of Applied Sciences and Environmental Management; **9(1)**: 150-155*

Ochei, J. and Kolhatkar A., (2008), Medical Laboratory Science, Theory and Practices, Tata McGraw-Hill:311-347.

Park, B., Lee, H.S., Lee, J.W., Park, S. (2019). Association of white blood cell count with breast cancer burden varies according to menopausal status, body mass index, and hormone receptor status: a case-control study. *Science Reports*; **9(1)**:5762. doi: 10.1038/s41598-019-42234-6.

Shah, J. I.,Zhao, W., Zhang, M., Usary, J., Murrow, G., Edwards, D., Knezevic, J., Greene, S. B., Darr, D., Troester, M. A., Hilsenbeck, S. G., Medina, D., Perou, C. M., Rosen, J. M. (2011). Breast Cancer Special Feature: Proceedings of the National Academy of Sciences; **109 (8)**: 2778–2783.

Siegel, L., Zambetti, M., Clark K. (2018). Gene expression profiles in paraffin-embedded core biopsy tissue predict response to chemotherapy in women with locally advanced breast cancer. Journal of Clinical. Oncology; **23 (29):** 7265–7277.

Siniorakis, E.E., Tsarouhas, K., Tsitsimpikou, C., Batila, T.C., Panta, S., Zampelis, K.G., Rentoukas, E., Triposkiadis, F., Limberi, S.J. (2017). Neutrophil to Lymphocyte (N/L) and Platelet to Lymphocyte (P/L) Ratios in Differentiating Acute Heart Failure from Respiratory Infection. Current Vascular Pharmacology;**15(5)**:477-481. doi: 10.2174/1570161115666170126141809. PMID: 28128065.

World Cancer Report (2014). International Agency for Research on Cancer, World Health Organization. 2014. ISBN 978-92-832-0432-9.

Citation: Alhassan Hussaini Mohammed, Usman Malami Aliyu, Isiyaku Adamu, Samuel Bamaiyi Dadzie, Abdulrahman Abdullatif, Abdullahi S. Mainasara, Yakubu Abdulrahman and Hamisu Abdullahi. Immunomodulatory Effects of *MANGIFERA INDICA* on Haematological Parameters and Kidney Architecture of Breast Cancer-Induced Albino Rats. ***Sokoto Journal of Medical Laboratory Science*; 8(4): 113 – 123**. https://dx.doi.org/10.4314/sokjmls.v8i4.14

Copyright: This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.