COMPLETE BLOOD COUNT AND HORMONAL PARAMETERS IN SOME MENOPAUSAL WOMEN IN ILORIN, KWARA STATE. NIGERIA.

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Abstract

Hormonal levels vary in women at different stages of life from puberty with remarkable observable changes from menopause. The purpose of this project was to carry out complete blood count on menopausal women with premenopausal women who served as control and to assess selected hormonal parameters in them by measuring their Follicle Stimulating Hormone and progesterone levels respectively. This was a case-control study involving menopausal and premenopausal women conducted within Ilorin metropolis, Kwara state, Nigeria. The inclusion criteria were adult female subjects within age range of 18-65 years old. A standard venipuncture technique was used to collect blood sample in which 4ml was drawn aseptically from the vein using vacutainer tubes. The hormonal parameters were carried out using ab108678 follicle stimulating hormone and ab108670 progesterone human ELISA kit following the standard operating procedure (SOP). The complete blood count CBC was carried out using the Sysmex1000i analyzer following the manufacturer's instruction. The study found that at menopause, the blood indices such as packed cells volume and lymphocyte were increased, while platelet count and neutrophil levels were higher in premenopausal women. Haemoglobin concentration, total white blood cells count, and the mixed leucocyte parameters (Basophils, Monocytes, Eosinophils) were unaffected. The mean value of postmenopausal progesterone of the menopausal women increased in comparison with that of the premenopausal women. There was also a significant difference between the mean value of the postmenopausal FSH (follicle stimulating hormone) in menopausal level as compared with that of the premenopausal women. The luteal progesterone in premenopausal women was significantly higher than in menopausal women. These findings indicate that some parameters in the complete blood count and selected hormonal parameters are affected by menopause.

Keywords: hormone, menopausal, premenopausal, women, progesterone

INTRODUCTION

Background of study

The relationship between sex and health perception is close. Females tend to have a worse experience in their own health than males at virtually all ages, mostly when they are more than 50 years old (Aneke et al., 2016). This is probably connected to the onset of menopause, which at the same time, leads to an increase in heart issues, blood vessel issues and other health issues. However, it is a well-known fact that women generally have a higher life expectancy than

their male counterparts (Aneke et al., 2016). Some women may not be aware that they have reached menopause, which happens to be the stage at which their ovaries do not produce estrogen and progesterone any longer. They only become aware about a year after they get to this period. Biologically, a woman can still produce new life until she gets to that point. The diagnosis of menopause is based on the menstruation records and general medical history; it also relies on Medical Laboratory tests that evaluate certain hormones and blood components that can be used for verification and confirmation of menopause (Bain, 1985). As a woman advances in age, important changes occur, and these affect the output of her female hormones. These hormones, which are vital in controlling the menstrual cycle and having a successful gestation, are synthesized by the ovaries, which are the two oval-shaped organs, located on both sides of the uterus (Complete Blood Count (CBC), 2021).

In the year just before menopause, also known as perimenopause, the ovaries begin to shrink in size and form. Estrogen and progesterone levels will be unsteady as the ovaries keep working to maintain the production of the hormones. Females at this stage can experience variable menstrual cycles, alongside fluctuating episodes of heavy bleeding of menstruation. Perimenopause typically lasts for many years before the menstruation finally stops by Menopause. Menopause signals the era of the last menstrual period. It is not regarded as the last until the woman has been shown to be period-free for a period of one year without illness, pregnancy, breast-feeding, or using certain medication. These and many more factors can also cause menstruation to stop flowing (Complete Blood Count (CBC), 2021). Menopause is the last menstruation in which the ovaries have halted production of the hormones that leads to menstrual flow (Medicinenet: Complete Blood Count (CBC): Types, Preparation & Procedure, 2021).

The Complete blood count is a group of Haematological parameters, which is usually requested, in order to screen for blood cell-based disorders that can affect health. Any increase or decrease in the blood cell counts of either white blood cells, red blood cells or platelets in the blood could represent a deviation from normal health. The normal range for each parameter is dependent on the age and gender of the patient. These tests are vital in the diagnosis of different health conditions, ranging from anaemia and other pathological states, to cancers, as well as menopause (Healthline: What Are the Symptoms of Low Estrogen in Women and How Are They Treated?, 2021). Complete blood count is the computation (calculation) of formed elements of blood. There is a special machine known as an autoanalyzer, which helps in the calculation of these parameters. The tests can also be performed manually (del Sueldo et al., 2018). Results will determine whether the number of red blood cells, white blood cells, and platelets in the blood is sufficient or not (Hall, 2015).

Women with menopausal signs, who may have premature ovarian failure, or who are being evaluated for risk of other health problems, such as osteoporosis, may have one or more or all of the following tests ordered:

Follicle stimulating hormone (FSH): the test is carried out to ascertain if a woman is approaching, evolving to, or has gone through menopause; in general, the FSH level mounts at transition. Follicle stimulating hormone (FSH) is a hormone for regulating the reproductive processes of the body. Follicle stimulating hormone is produced by the pituitary gland in man. It is responsible for controlling growth and development, and also pubertal changes in both males and

females. The hormone assists in the reproductive progression in females who are in their reproductive period. For females older than 45 years, increased FSH levels may signal an impending menopause (Langford et al., 2003).

Estrogen: measures the estrogen produced by the ovaries and evaluates whether the menstrual cycle is normal (Lindberg et al., 2021). Estrogen is a hormone that influences the sexual and reproductive development of females. It has 3 types: estradiol, estrone and estriol. The ovaries make most of the body's estrogen. The adrenal glands and fat cells also produce small amounts (National Institutes of Health, 2015). Estradiol is a form of estrogen that is responsible for regulating the menstrual cycle and supporting the female reproductive tract (National Institutes of Health, 2015). The decline in estrogen levels during the perimenopausal period is responsible for some of the symptoms in menopausal women (Menopause and FSH Levels, 2009).

Progesterone is a pregnancy hormone that supports fertility and maintains pregnancy. It is synthesized by the ovaries after ovulation. This is important for women in the reproductive stage because it prepares the uterus to receive, implant and nurture the fertilized egg (Lindberg et al., 2021). It is a steroid hormone belonging to the progestogens class and is secreted by the corpus luteum (Lindberg et al., 2021). Low progesterone levels can lead to high estrogen levels, which can lead to decreased libido, gallbladder problems, or weight gain in menopausal women (Lindberg et al., 2021).

Anti-Müllerian hormone (AMH): this measures blood anti-mullerian hormone levels, which can provide data about fertility. It is also useful in the diagnosis of menstrual disorders or predict the onset of menopause (MedlinePlus Medical Encyclopedia, 2021).

A woman who suspects that she may be going through menopause should get herself tested, because it can help her to decide in time, how best to manage her health and some of the symptoms that she will encounter in the future (Hall, 2015). Hormonal therapy can reduce some of the symptoms of menopause, either by using estrogen alone or in combination with progesterone or progestin in its synthetic form. Estradiol promotes cell growth and replication, whereas progesterone counteracts proliferation and causes cell maturation (Cleveland Clinic, 2021). These two hormones normally help to regulate a woman's menstrual cycle. There are differences in the manifestations and seriousness of menopausal symptoms from woman to woman. Women who are not on hormonal therapy tend to show more menopausal symptoms with higher severity compared to those on hormonal treatment. The use of hormonal therapy should be considered legal, as it appreciably reduces the undesirable symptoms of menopause and can promote women's daily health (Lab tests online, 2021).

Menopausal change is constant in the life of every women, during which hormonal imbalance occurs, and is accompanied by menopausal syndrome which is expected to have effect on reproductive hormones. Unfortunately, there is paucity of data on hematological parameters in Menopausal women. It is also important that every women should be adequately educated in order to prepare them for this stage of life and also emphasize on the importance of going for regular hormonal evaluation in order to establish the onset or presence of menopause.

Materials and methods

This study was conducted on some women within Ilorin metropolis, Kwara state, Nigeria. It was a prospective case-control study involving postmenopausal women and women in their reproductive stage resident within Ilorin. A total of 73 women comprising 50 menopausal women and 23

women who were in their reproductive stages and used as controls, participated in the study. Ethical approval was obtained from the Ethics and Research Committee of the Kwara State Ministry of Health, Ilorin, Kwara State, Nigeria, while informed consent of the participants was duly obtained prior to commencement of the study. Subjects who were aged between 18 and 65 years were included in the study, while subjects at the extremes of age, pregnant women, lactating mothers and male subjects were excluded from the study.

A standard venipuncture technique was used to collect blood sample. Four millilitres (4ml) of blood was drawn aseptically from the vein using vacutainer tubes as follows; 2ml of blood into Ethylene diamine tetra-acetic acid (EDTA) vacutainer tube for the complete blood count, and
2ml of the blood drawn into a plain vacutainer tube, where the serum was obtained after the blood sample was centrifuged at 10,000rpm for 5minutes for assessment of the hormonal parameters.

Laboratory procedures

The Follicle Stimulating Hormone (FSH) and Progesterone hormonal assays were separately carried out by ELISA (Milman & Pedersen, 2008), according to manufacturer's instructions. The mean absorbance values of all duplicates were calculated where applicable.
For the complete blood count (Nordin et al., 2004), the blood sample collected in the EDTA bottle was mixed properly. Then the stopper of the container was opened and the tube was set to the sample probe. The start switch was turned on. The tube was held continually to the sample probe until the buzzer sounded twice – "beep-beep"; and when the LCD screen displayed "**Analyzing**" the tube was removed. After that, the unit executed automatic analysis and displayed the result on the LCD screen. Then the unit turned to "ready" status, becoming ready for analysis of the next sample. When analyses of all samples were finished, shutdown was executed before turning the power off.
Data collected were analyzed using the IBM SPSS statistic software (version 25). The students' t-test for independent samples was used to test for the significance of differences between the two groups. $P < 0.05$ was considered statistically significant.

Results

Table 1 presents some socio-demographic characteristics of the Menopausal and Pre-Menopausal women**.** There were a number of 73 subjects, comprising 50 Menopausal women and 23 Pre-Menopausal women who served as the control subjects.
All the subjects were females within the following age bracket: For the menopausal group, subjects who were between 44-48 years were 12 in number (24.00%), those aged between 49-54 years were 18 (36.00%), while those aged 55 years and above were 20 (40.00%). For the premenopausal women, 6 subjects were aged between 18-22 years (26.09%), 12 were aged between 23-27 years (52.17%), while 5 subjects were aged between 28-32 years (21.74%). There were no subjects aged 33 years and above (0.00%).

Table 1: Some socio-demographic characteristics of the subjects

	Menopausal women	Pre-menopausal women
Age bracket (Yrs)	44-48(24.00%) n=12	18-22 (26.09%) n=6
	49-54(36.00%) n=18	23-27 (52.17%) n=12
	≥55(40.00%) n=20	28-32 (21.74%) n=5
Total	(100%) N= 50	(100%) N = 23

Table 2 depicts the mean and standard deviation of selected parameters of the Full blood Count in menopausal and premenopausal women.

The mean of the Packed Cells Volume (41.78 ± 6.74) (p=0.006) and the lymphocyte % (46.48±12.14) (p= 0.001) in the menopausal women were significantly higher than the values in the premenopausal women which were 36.57 ± 5.00 for the packed cells volume and 28.52 ±12.62 for the lymphocyte % respectively. On the other hand, the Neutrophil % (68.39 ±12.72) (p=0.001) and the platelet count (273.65 ±84.90) (p=0.003) of the premenopausal women were significantly higher compared to that of the menopausal women which were 49.50±12.00 and 185.64 ±76.75 respectively. However, there were no significant differences between the means of the Haemoglobin concentration (Hb), Mix Basophil, Monocytes and Eosinophil percentage (BME %) and Total WBC count in both groups

Table 3 shows the mean and standard deviation of the Follicle Stimulating Hormone (FSH) and progesterone hormonal levels in menopausal and premenopausal women.

The mean value of FSH at the follicular phase of the premenopausal women (4.51± 1.51) showed no significant difference when compared with that of the menopausal women (4.10 ±1.59) (p=0.159). On the other hand, there was observable high significant statistical differences between the mean values of the luteal phase Progesterone levels in premenopausal women (9.40±6.18) as compared to that of the menopausal women(4.73±3.17) (p=0.001). The mean value of postmenopausal progesterone of the menopausal women (2.16±0.14) was significantly higher when compared with that of the premenopausal women (0.60±0.12) (p=0.042).

Table 2: The mean and standard deviation of selected parameters of the complete blood count in menopausal and premenopausal women

Parameters	Menopausal Women (n=50)	Premenopausal women (n=23)	*P* value
Hb	12.30±1.55	11.60±1.51	0.144
PCV	41.78±6.74	36.57±5.00	0.006*
Lymph%	46.48±12.14	28.52±12.62	0.001*
Mix BME%	4.04±3.27	3.09±2.84	0.117
Neut%	49.50±12.00	68.39±12.72	0.001*
TWBC	6.32±2.41	8.34±3.06	0.112
Platelet	185.64±76.75	273.65±84.90	0.003*

Key: Hb=Haemoglobin concentration (Normal range: Women=12-16 g/dl);
TWBC=total white blood cell count (Normal range: 4.5-11.0 $X10^9$/l);
PCV=packed cells volume (Normal range: Women=35-47%);
Platelet=platelet count (Normal range: 150-400 $X10^9$/l);
Lymph=Lymphocyes (Normal range: %);
Neut=Neutrophil (Normal range: %);
BME=Basophil, Monocytes and Eosinophil (Normal range: <10%);
*p-value is significant at 0.05. N = 73

There was also a significant difference between the mean value of the postmenopausal FSH in menopausal women (52.18±12.07) as compared to that of the premenopausal women (0.03±0.06) (p<0.001). However, the mean level of the Follicular phase progesterone, Luteal FSH and Midcycle FSH were not statistically significant in both menopausal and premenopausal groups, (p=0.083), (p=0.257) and (p=0.268) respectively.

Table 3: The mean and standard deviation of FSH and progesterone hormonal levels in menopausal and premenopausal women.

Parameters	Menopausal Women (n=50)	Premenopausal women (n=23)	*p*-value
Follicular FSH	4.10±1.59	4.51±1.51	0.159
Follicular Progesterone	0.58±0.48	0.71±0.52	0.083
Luteal FSH	5.61±2.22	5.20±1.82	0.257
Luteal Progesterone	4.73±3.17	9.40±6.18	0.001*
Midcycle FSH	9.33±3.54	11.58±4.20	0.268
Postmenopausal FSH	52.18±12.07	0.03±0.06	<0.001*
Postmenopausal Progesterone	2.16±0.14	0.60±0.12	0.042*

Key: Follicular FSH=Follicular phase (Normal range: 2.9-7.0 mIU/ml);
Luteal FSH= Luteal phase (Normal range: 12-16 mIU/ml);
Midcycle FSH= Midcycle or ovulation phase (Normal range: 5.2-17.0 mIU/ml);
Postmenopausal FSH= (Normal range: 35 – 151 mIU/ml);
Follicular Progesterone= (Normal range: 0.1-1.4ng/ml);
Luteal Progesterone= (Normal range: 4.0–25.0 ng/ml);
Postmenopausal Progesterone= (Normal range: <1.0 ng/ml);
*p-value is significant at 0.05. N = 73

Discussion

The study investigated some hormonal and haematological parameters among menopausal and premenopausal women in Ilorin, Kwara State, Nigeria.

Findings revealed that at menopause, blood indices such as packed cells volume and lymphocyte percentage increased, while platelet and neutrophil levels were higher in premenopausal women compared to menopausal women. This is similar to the findings of (Langford et al., 2003).

There was a non-significant increase in Haemoglobin concentration among menopausal women in comparison to premenopausal women. This is consistent with the findings of (Prior, 2018) and (Putyński & Janicka, 2011).

The white blood cells and mixed Basophil, Monocytes and Eosinophil percentage (BME %) were unaffected. The mean value of postmenopausal progesterone level of the menopausal women were increased in comparison to that of the premenopausal women (p=0.042). The postmenopausal FSH in menopausal women was also significantly higher than the value in premenopausal women (p<0.001). Even though this study recorded high level of hemoglobin in menopausal women compared with the premenopausal women, these differences were not statistically important (Table 2). Prior (2018), indicated that hemoglobin concentration may be higher in menopausal women, most probably due to the cessation of menstruation.

The platelet count was significantly higher in premenopausal women compared with menopausal women (p=0.001, Table 2). These observations have been emphasized in an earlier study (Langford et al., 2003) which suggested that high platelet counts found in premenopausal women was a compensatory response to endothelial vascular rupture in order to facilitate effective hemostasis in the endometrium. This explains the cause of high bleeding tendencies in the menopausal age group. However, the decreased platelet count in menopausal women may be due to decreased marrow activity with age or an effect of decreasing hormones with age (Reed & Carr, 2000).

There was a non-significant reduction in the follicular FSH and follicular progesterone in menopausal women and a significant increase in postmenopausal progesterone. The luteal progesterone in menopausal women was however, significantly reduced in comparison to the premenopausal women. These findings agree with the results of (Welt et al., 2003) and could be due to declining hormonal levels. At the time of transition from premenopausal to menopausal stage, the production of the ovaries and estrogen are

decreased. This eventually extends to reduce level of circulating 17β-estradiol, with high levels of follicle stimulating hormone (FSH) as seen in table 3 and agrees with the finding of (Healthline, 2021). Due to this, women develop estrogen deficiency once they get to this period of life. This in turn causes physiological impairments in menopausal women. The alteration in hormones leads to some of the symptoms experienced by menopausal women like chills and hot flushes (Healthline, 2021). The variation could be due to changes in the body physiology with aging among females probably due to stoppage of mensuration and the beginning of menopause.

There was an observed increase in the postmenopausal FSH among menopausal women. This could be due to ovarian aging and declining ovarian function, and also due to a possible reduction in the levels of inhibin B, a hormone that helps in the gonadal feedback regulation of FSH. This agrees with findings from (Everday health, 2021).

Findings from this study suggest that the alteration in some haematological and hormonal parameters due to menopause, could predispose women to pathological states. Therefore, regular medical checkups are strongly advocated.

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Conflicts of interest

The authors declare that they have no conflicts of interest.

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