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Progesterone Level Affects the Onset of Early Menopause in Women

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ABSTRACT

Early menopause occurs when woman stops bleeding at the age of 40 to 45 years. Menopausal symptoms include missed periods, hot flushes, night flushes and reduced sex drive. The ratio of early menopause is increasing day by day. Almost 10% women experiencing early menopause. This ratio increasing day by day due to shift in the reproductive cycles. Many factors affecting the early menopause, progesterone level is one of the main factors. The females above age of 40 have different level of progesterone. The low level of progesterone affects the menstrual cycle of women. The purpose of this study was to find out the effect of progesterone level on early menopause. For this study sample size was 40. The samples were divided into two groups, control and experimental group. The age of all participants were 40 to 45 years. For this research work 40 blood samples were collected. The collected blood samples were processed on Abbott by using chemiluminescence technique. The device used in this study were Abbott. The questionnaire was used to find out the symptoms of early menopause. The results were statistically analyzed by using t-test independent on SPSS. It has been found that the low level of progesterone affects the early menopause in women. This study will help to give awareness that if a woman has menopausal symptoms, she should take it seriously and visit doctor.

INTRODUCTION

The menopause is the complete stop of the menstrual cycle due to end of ovarian follicular activity, this loss ends the female fertility life. The menopause often starts after 40s, it is divided into early and late period [1]. When natural menopause occurs in female it stops the ability of women to bear a child, it is permanent end of her menstrual cycle [2]. Fertility also decreasing with age, ageing also increasing decline of other physiological functions. Cessation of reproductive function is mostly observed at the age of 50, if this decline does not occur then it may be observed at the age of 70. Moreover, this decline occurs more rapidly in women as compared to men and other physiological changes [3].

Early menopause also defined by the World Health Organization, as early menopause is the perpetual end of menstrual cycle, due to end of female reproductive activity [4]. The menopause occurs at the age of 40 t0 45 years is referred as early menopause, while natural menopause age is 51 years. Early menopause can be natural or induced, by chemotherapy, surgery or due to some medicinal uses. The hormonal deficiency in women also causes early menopause [5].

These symptoms highly effecting menopausal women. Women rate these symptoms from normal to sever. Many factors like social relation, social characteristics, daily life style and psychological behavior are factors, these factors affect severity of menopause [6]. All over from history women facing many menopausal symptoms, hot flushes, heart diseases, osteoporosis, weight gain and vaginal dryness. The menopausal symptoms are affecting the quality of life of women, for example hot flushes disturbs the daily routine of women life. The goal of study was to develop permanent treatment of hot flushes. If women do not bleed for 12 months then it is announced the complete end of menstrual cycle in women and start of menopause. The menopause in women terminates the menses in women. The menopausal symptoms include hot flushes and night sweats, these symptoms affecting almost 50-85% women. These symptoms affect women older than 45 years.

These treatments are used for menopausal symptoms. Beside these therapies medicines also referred. The hormonal replacement therapy creates some complications, depends on patient symptoms. many programmed suggests the hormonal replacement therapy for estrogen and progesterone hormones. These therapies considered harmless for seven years. The hot flushes can be treated by using medicines and hormonal therapies. Hot flushes are most common and ignored symptom of

menopause. Information on these symptoms backgrounds and pathophysiology are emphasized for the treatment [7].

Ovary is the main location for the release of progesterone in mammals. The primary follicles perform double function by releasing progesterone and estrogen hormone. During normal cycle in female's primary follicles also responsible for the secretion of ovum. Before ovulation, in follicle granulose cells biologically synthesized. Ovum releases after the rupture of follicle, the cells of granulosa mature and alter on forms corpus luteum. The corpus luteum responsible for the release of progesterone hormone in the end of the cycle. In humans during 1 to 2 days fertilization don not takes place then corpus luteum resumes to enlarge for 10 - 12 days. This process observes by the glands and it stops the discharge of progesterone hormone. And if fertilization takes place, then corpus luteum will remain grow and plays a prime role in first trimester of pregnancy. Later on, placenta forms and carry on the function of hormone synthesis and continue the pregnancy. In this phase corpus luteum eventually deteriorate [8].

The progesterone hormone plays the vital role in the female reproduction and fertility. Progesterone has important function during ovulation and 1st, second and third trimester of pregnancy. In menstrual cycle progesterone plays role in the uterine function. Progesterone controls the changes during menstrual cycle. Progesterone helps in the change of decidual tissues. The high level of progesterone required for fertilization, after fertilization if pregnancy takes place first, second and third trimester requires more level of progesterone. It also holds up the pregnancy and help in uterus growth. Progesterone also promotes the muscular contraction [9].

Progesterone supports the pregnancy by changing mucosa membrane. the progesterone hormone brings changes in mucosa membrane from growing stage to secretory stage, these changes help and supports pregnancy. The name of progesterone came from Latin word and its name define it well as pro gestational. Other than reproduction progesterone plays an important role in other body parts for example it plays vital role in the breast feeding, heart and vessels, brain, spinal cord and in bones. In 1993 scientist Allen gave the molar formula of progesterone [10].

Early menopause is serious fertility issue in young women and its ratio increasing from past 10 years. It causes serious health issues in women like osteoporosis, cardiovascular diseases, hypertension, obesity and infertility. Progesterone level is corelated with the onset of early menopause. The low level of progesterone may be the cause of early menopause.

Research Objective

The objectives of this research work are as following:

- To determine effect of progesterone level on early menopause.
- To find out the symptoms of early menopause.

This research will support to acknowledge the effect of hormone progesterone level onset of the early menopause in women. Women above age 40 to 45 years have different level of progesterone and this will cause many health issues and infertility in women. This research will create awareness among women who are afraid of talk about it and can be used to early diagnosis.

MATERIALS AND METHODS

For collection of data, approval letter from Human Research Ethics Committee, University of Sialkot was received. After getting approval letter a copy of letter submitted to the Aslam Medical Complex and Azeem clinic for allocation of sample collection. For data collection a criterion was made. Two groups participated in this research, one was control group and other was experimental group. For participation age group of females was 40-45 years. And woman had menopausal symptoms for experimental group. And participants of control group also from same age group with no menopausal symptoms.

Sample size were 40, 20 participants from control group and 20 from experimental group. For sample collection Dr. Ambreen from Azeem hospital and Dr. Nighat from Aslam Medical Complex helped, they allowed for sampling and data collection. Blood collection was done by venipuncture. Blood collected in gel test tube having clot activator. From each woman 2 specimens were collected and placed in dry test tube. For sampling dry glass test tube used which have separating gel [11].

In this research questionnaire were used. Questionnaire were given to 40 women, 20 women from control group and 20 were from experimental group. 7K77 ARCHITECT Progesterone Reagent Kit was used (Strauss & Hsueh, 2001). Satisfactory specimens contain serum or plasma (EDTA or lithium heparin). Yet heat inactivated sera is not developed so, sera were not heated. The sample were stored at 2-8oC for up to 48 hours. Samples were freeze for longer storage at - 25 ± 6°C for up to two months [12,13].

The Abbott progesterone procedure was performed on an automated instrument and enzyme linked florescent immunoassay (ELFA) technique was used [14]. All steps in this procedure and temperature controlled by instrument. For Solid Phase Receptacle (SPR), a disposable device pipette tip-like act as solid phase for assay and pipetting device [15]. SPR was coated with mouse monoclonal anti-progesterone antibodies [16].

The layout of VIDAS progesterone prevents common reaction with SPR. Reagents was loaded on sealed reagent strips for assy. Sample was loaded into well which have alkaline phosphatase derivative with progesterone derivative. The progesterone conjugate moved in and out and cycled and competes with progesterone already present in the sample in order to bind with antibiotic [17]. Later on, in the process of washing unbound conjugate remove. 4-methylumbelliferyl phosphate, a fluorescent substrate circulates, the enzyme which already present in the wall accelerate the reaction to convert reactants into products [18]. A product 4methylumbelliferone was formed wavelength of 450nm and for measurement of intensity optical scanner used. This intensity was inverse relation to progesterone of samples. After completion of cycle results were analyzed automatically and report printed for each sample [19].

The interior of the SPR is coated with anti-human u chain antibodies. Each SPR was marked with specific
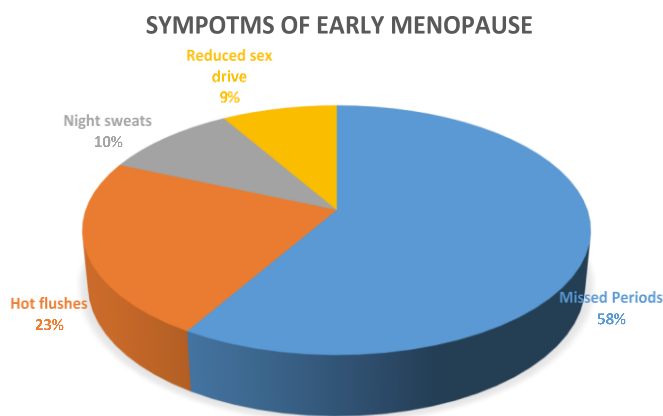
progesterone code, remove required number and reseal it carefully [20]. The strip consists of 10 wells which have specific reagents required for the assay. These wells are labeled and sealed foil [21].

The kit was opened and all necessary components. Unused components were repacked at 2 to 8 Celsius. The components were placed for a while to reach them at room temperature. It was made sure samples storage pouch was completely sealed. One PRG and one SPR strips used for each sample. Calibrator was used and was tested. Calibrator marked with S1 and identified by code. The PRG reagent strips was labelled with specific sample identification number. Sample and calibrator were mixed and it was made sure each one has 200 micro liter portion. SPRs and PRG reagent strips were inserted in specific position on instrument. The color labels were checked and identified with codes mentioned on strips.

The process was initiated while an instrument run automatically. The vials were closed and pipetted at required temperature. Procedure was run for 45 minutes. After procedure completed, Used SPRs and strips were removed and disposed in appropriate way. Two instrument readings took for fluorescence. First reading took before SPR introduced and second reading took when substrate exposed to enzyme conjugate. For final results of florescence value, the background readings were subtracted from final values. Procedure completed then results calculated automatically by using calibrator cure which is stored in the memory. Reports were printed and value of progesterone calculated in ng/ml.

RESULTS
Figure 1

SYMPOTMS OF EARLY MENOPAUSE

Reduced sex drive 9%
Night sweats 10%
Hot flushes 23%
Missed Periods 58%

For statistical analysis T-test applied. Test ran on SPSS. Results shows the mean of patients as well as mean of controlled group. Mean value of patients is 2.0 while mean value of controlled group is 14.9. And the mean difference between their mean values is 12.9. This mean difference shows this hypothesis is not null. Above mentioned tables showed the T-test applied on the data. 0 represents control group while 1 represents patients' group. Results shows mean of two groups and mean differences of two groups. It also shows standard deviation of two groups. For progesterone level in women age group from 40 to 45 years old an independent t-test was conducted. Results indicates that group 0 females that is control group (M=14.91, SD= 1.53) have more values of progesterone as
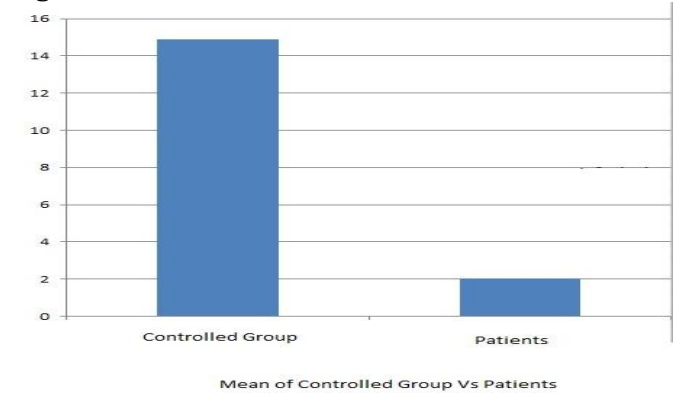
compared to patients (M=2.045, SD= 1.0200).

Table 1

*Progesterone Level ng/mL of Control and Experimental Group*

Group	Number of samples N	Mean	Standard deviation	Mean differences
Control group	20	14.90	1.53854	12.03014
Patients	20	2.0450	1.02005	12.03014

Figure 2


Mean of Controlled Group Vs Patients

This figure demonstrates that the control group's progesterone levels were higher than those of the control group.

Figure 3
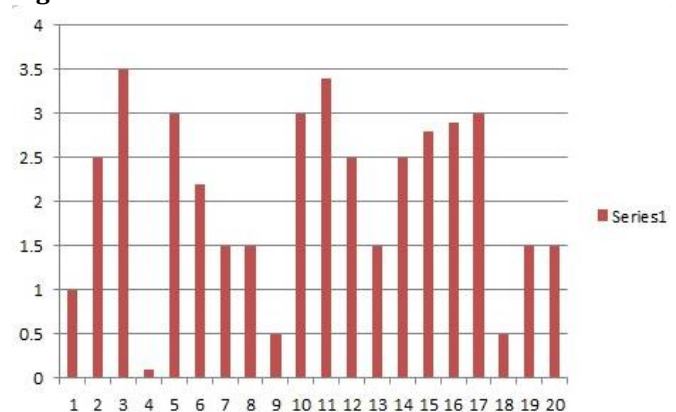
This figure shows the progesterone level in patients. Patient 3 has the highest progesterone level, followed by patient 11 in the middle, and patient 17 in the third-highest. Patient 4 has the lowest level of progesterone.

Figure 4
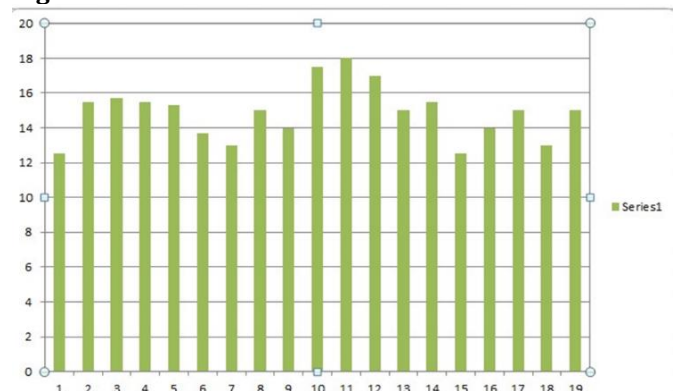
This figure shows the progesterone level in control group. Patient 11 has the highest progesterone level, followed by

patient 10 in the middle, and patient 12 in the third-highest. Patient 12 and15 has the lowest level of progesterone.

Table 2
Responses of n=40

Question	Responses			Total
	1	2	3	
Do You have bleeding issues?	50%	50%	0%	40
Do You have hot flushes?	23%	50%	27%	40
Do you often miss your periods?	48%	50%	2%	40
Do you feel night sweats?	10%	50%	40%	40
Do you feel reduced sex drive?	9%	50%	41%	40

Menopausal symptoms based on questionnaire answers (n = 40). Yes, on scale 1, nay on scale 2, and occasionally on scale 3.

DISCUSSION

The reproductive time period in female is normally much shortened than the total life duration. The primary reason for this is the constant failure or degeneration of expected ova, and the deficiency of any condition for their substitution. When no more ova stay, reproductive quality evidently comes to a final stage. On the other side, the endocrine action of the ovary does not change in this sudden way, and so there is a stage of decrease function [22]. Progesterone features a necessary role in control women fertility rate, with salient actions end-to-end the female reproductive mechanism. The endocrinology activity of progesterone has been viewed as captious for the relation of the women reproductive cycle. This basic generalization has been improved by in vivo experimental inflection, using hormone replacement as well as medicine and genetic interruption of the progesterone receptor (PGR). Phenotype description of PGR null rats modify roles for progesterone in the dominance of female fertility, but not function for progesterone as a necessary determinative of women reproductive regularity, hard an elemental role of women reproductive life [10].

This study demonstrated that low level of progesterone causing early menopause in 40 to 45 years old women. Women with low level of progesterone have bleeding issues and early menopausal symptoms, some of them are facing early menopause. Other than hot flushes and night sweats women also experience anxiety disorders and psychological symptoms, low sex life and some memory issues.

On the other side women with normal level of progesterone living normal regular life with no symptoms of early menopause and no bleeding issues.

The menopause either onset or obliged. Women experiences premature or early menopause. The premature menopause starts before 40s age of women, while early menopause starts after 40s age of women. Premature and early menopause brings increases the health risk factors of women by bringing cardiovascular diseases, neurological issues, osteoporosis and many other health problems. The risk factors of women health increases with the menopausal age. These risks factors and symptoms control by giving estrogen therapy, although estrogen alone not sufficient and will not give long lasting results. Beside all these women who experiences hormonal menopause due to estrogen and progesterone are at high risk of immortality. For these women estrogen and progesterone hormonal treatment advised but it may give harmful results [9].

This study showed that low progesterone level affecting menopause. And low level of progesterone causes menopausal symptoms and early menopause in women as Loren et al described the progesterone effect on early menopause [23].

CONCLUSION

Progesterone plays important role in female fertility and reproduction. The progesterone level regulates female reproductive system. Each month after ovulation progesterone thickens the lining of uterus for fertilization. During pregnancy progesterone level helps to keep up pregnancy. The goal of this research was to find out relation between level of progesterone and early menopause in age group of 40 to 45 years old women. The women experienced early menopausal symptoms and early menopause had clear decline in the level of progesterone. This research found that there is direct relation between the low level of progesterone and early menopause in woman. The findings of this study will create awareness about early menopause and low level of progesterone.

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