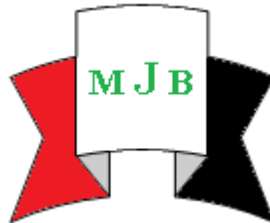


Risk Factors for Pre Mature Ovarian Failure in Women from Babylon/ Iraq

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Abstract

Background: Premature ovarian failure (POF) is a common cause of infertility in women characterized by amenorrhea, hypoenestrogenism and elevated gonadotrophin level in women under the age of 40. The ethnic prevalence variations pose possible etiologic differences also due to environmental, genetic, and nutritional factors. Conduction of etiologic studies in different settings or populations can help in improving the knowledge regarding etiology and predictors of POF.

Objective: To determine possible risk factors of POF in subset of women from Babylon/Iraq.

Patients and Methods: This study includes Fifty- three Selected women from Al- Amal clinic for infertility management and treatment in Babylon, patients were categorized in three groups, **A:** those women are the premature ovarian failure group, (comprise 26 patients, $n=26$, age <40 years (mean 30.23 years) follicular stimulating hormone (FSH) >14 mIU/ml). Group **B:** twelve women who reached natural menopause ($n=12$, age ≥ 40 years (mean 42.75 years) FSH >20 mIU/ml, Group **C:** Fifteen known normally cycling fertile women mean age 27.06 years, FSH 7.05 mIU/ml ($n=15$). All studied groups were asked about their maternal age of natural menopause, body mass index (BMI) were calculated and asked about menstrual cycle pattern and if previous fertility is present or not. Blood group type was stated for all women. All inhabitants registered with an address in Babylon/Iraq. **Results:** There was no significant difference in maternal age of menopause between groups, neither in BMI. Blood group type O ranked higher significant percentage in group A women than other two groups, while other blood types show no significant difference. Sixty-five percent of group A women presented with infertility and irregular menstruation was reported in 80.8% of participants in the same group, which is significantly different from group B and C.

Conclusion: Maternal age of menopause constitute no risk factor for POF in our study population this may be due to the fact that they have underwent extensive psychological and environmental pollution in the last 20 years that have detrimental effect on ovarian reserve of the female, or may be due to small sample size. Blood group O seems to be a risk factor as well as the presence of previous infertility and menstrual irregularities.

Key words: POF, premature menopause, risk factors, blood group type.

عوامل الخطر في حدوث فشل المبيض المبكر لدى نساء من بابل ١ العراق

الخلاصة

الخلفية: من الاسباب الشائعة للعقم لدى النساء هو فشل المبيض المبكر الذي يتميز بانقطاع الطمث و انخفاض مستوى هرمون الاستروجين في الدم مع ارتفاع الهرمونات القنديه تحت سن الاربعين . من الاسباب المحتمله هو الاختلافات العرقية و العوامل الوراثية و البيئية و الغذائية . ان اعداد دراسات مختلفه ممكن ان تساعد في معرفه الاسباب و التنبؤ بحدوث فشل المبيض المبكر .

الهدف : تحديد العوامل المحتمله كمسببات لحدوث فشل المبيض المبكر في مجموعه فرعية من نساء محافظة بابل – العراق .

المرضى و طرق العمل : تم اختيار ٥٣ مريضه و صنفوا الى ثلاث فرق , الفريق الاول (أ) هو الفريق السابق لاولانه في حدوث فشل المبيض يضم ٢٦ مريضه, العمر دون الـ ٤٠ (معدل العمر ٣٠,٣٠ سنة) مستوى الهرمون المحرض للجريب اكثر من ١٤ مللي وحدة دولية لكل مليلتر, الفريق الثاني (ب) يضم ١٢ مريضه في سن اليأس الطبيعي, العمر ≤ 40 (معدل ٤٢,٧٥ سنة) مستوى الهرمون المحرض للجريب اكثر من ٢٠, الفريق الثالث (ج) يضم ١٥ مريضه معروفات الخصوبه معدل العمر (٢٧,٠٦ سنة) مستوى الهرمون المحرض للجريب ٧,٠٥. تم سؤال جميع النساء المدروسات عن سن امهاتهن عند انقطاع الطمث بصورة طبيعيه و تم حساب مؤشر كتله الجسم و تم السؤال ايضا عن نمط دوره الطمث و اذا كان هناك خصوبه سابقه ام لا. و تم ايضا تحديد نوع فصيله الدم لجميع النساء و كل النساء المسجلات يسكن محافظه بابل - العراق.

النتائج: لم يمكن هناك فرق معنوي في عمر الامهات عند وصولهن سن اليأس الطبيعي بين الفرق الثلاث ولا في مؤشر كتله الجسم. استحصل نوع فصيله الدم ٠ اعلى نسبه مئويه عند النساء في الفريق الاول بينما انواع الدم الاخرى لم تظهر فرقا معنويا بين الفرق الثلاثه . ٦٥% من النساء في الفريق الاول كانوا مصابات بالعقم بينما كان عدم انتظام نمط الطمث موجود في ٨٠,٨% من نفس المجموعه و هذا يشكل فرقا معنويا عن الفريقين الاخرين. **الاستنتاج:** لا يشكل عمر الامهات عند وصولهن سن اليأس عامل خطوره في حدوث سن اليأس عند بناتهن المدروسات في الفريق الاول و هذا ممكن ان يعزى الى الظروف النفسيه والتلوثات البيئيه الشديده في السنوات الـ ٢٠ الاخيره التي لها تاثير ضار على احتياط المبيض في النساء المدروسات أو الى صغر حجم العينة المدروسة. ان فصيله الدم ٠ كذلك وجود العقم السابق و عدم انتظام نمط الطمث ممكن ان تشكل عامل خطوره في حدوث فشل المبيض المبكر.

المفاتيح : فشل المبيض المبكر , سن اليأس المبكر , عوامل الخطوره , فصيله الدم.

Introduction

Premature ovarian failure (POF) is a common cause of infertility in women, and is characterized by amenorrhea, hypo-oestrogenism and elevated gonadotrophin levels in women under the age of 40. The condition affects 1–2% of women younger than 40 years of age and 0.1% of women younger than 30 years of age [1]. Known causes include iatrogenic agents that cause permanent damage to the ovaries, such as chemotherapy, radiation therapy and surgery; autoimmune conditions, X-chromosome abnormalities and autosomal genetic condition are also known causes [2]. However, few genes have been identified that can explain a substantial proportion of cases of POF. Turner syndrome and gonadal dysgenesis are the best known causes of early POF. Nevertheless the etiology is most often unknown [3].

Most women with POF are deeply upset by the diagnosis, partly due to the unexpected menopausal symptoms, but also due to infertility [4], which puts it on

high priority of reproductive and health research work [5]. Therefore, early detection would provide better opportunity for early intervention, and furthermore, will help to direct potential targets for future treatment [6].

There are ethnic differences in prevalence of POF in different populations [7], this ethnic prevalence variations pose possible etiologic differences also due to environmental, genetic, and nutritional factors. Conduction of etiologic studies in different settings or populations can help in improving the knowledge regarding etiology and predictors of POF. The aim of this study was to determine possible risk factors of POF in subset of women from Babylon/Iraq.

Patients and Methods

All cases were selected from Al-Amal clinic for infertility diagnosis and management between November 2012 – August 2013; they attend the clinic for infertility, menstrual irregularity or secondary amenorrhea. Fifty- three

Selected patients were categorized in three groups, **A**: those women are the POF group, (comprise 26 patients, n= 26). All cases were interviewed in detail for medical & surgical history, a family history of POF, autoimmune diseases, or cognitive abnormalities (suggestive of fragile X syndrome), and any treatment (particularly exogenous estrogen & cytotoxic chemotherapy or radiation) examined clinically (general & systemic) to exclude any gross underlying disease and they were investigated extensively (including ultrasonography, reproductive hormones ((FSH) ,Lutinizing Hormone (LH) and Esrradiol (E2)) , hematology, biochemistry, Tyroid Function Test (TFT), Prolactin, Anti-Nuclear Antibodies (ANA) , and pregnancy test. They also underwent blood pressure measurement for orthostatic hypotension, before inclusion in the study. All related information was obtained in detail. The following criteria must be present before inclusion:

1. Age (< 40years) (1).
2. Menstrual cycle patterns (Menstrual irregularities or secondary amenorrhea for 4-6 months).
3. Elevated basal serum FSH level above 14mIU/ml in two occasions. We reviewed the literature and we find wide range of inclusion criteria regarding basal FSH levels, we selected this level since our laboratory analysis consider any level above 12 mIU/ml as starting menopause and since our patients are exclusively nonsmokers and noncontraceptive users, since these can cause early elevation in FSH level (8) so this number in our opinion is quite sufficient for inclusion criteria. **Group B**: twelve women who reached natural

menopause (n= 12, age ≥ 40 years FSH > 20 mIU/ml, no history of hormone replacement therapy and 12 consecutive months of amenorrhea. These women were selected while they attend the clinic for vaginal dryness or hot flashes and included for comparison. **Group C**: Fifteen known normally cycling fertile women (n=15) no contraceptive pill intake, proven fertility, normal regular menstrual cycle pattern, not pregnant or using hormones or drugs that interfere with the menstrual cycle, no history of hysterectomy, miomectomy, oophorectomy, or any other surgery on their ovaries. Blood sampling from group C women and women from group A who have irregular cycle was collected on day 2 or 3 of menstrual cycle. Those women from group B blood samples were collected at any day. Serum was isolated after centrifugation and stored at -20C before assay. BMI were calculated after height and weight measurement for all women included in this study. All women were asked about their blood group type (A, B, AB or O) and their mother age at menopause if they have reached natural menopause. All inhabitants registered with an address in Babylon.

Statistical analysis:

Continuous data are presented as mean $\pm$ SD and categorical data are presented as percentage (%). Comparison between groups was done using Wilcoxon signed Rank test. Relationships between categorical variables were assessed using chi-square test. P- Values < 0.05 were considered to be significant. Statistical analysis was performed using SPSS version 17.

Results

Table 1 Demographic data of different studied groups (including blood group types).

Parameter	GroupA(n=26)	GroupB(n=12)	GroupC(n=15)	P value	
				A/B	A/C
Age (years)	30.23 ± 6.65	42.75 ± 1.91	27.06 ± 3.95	<0.05	>0.05
FSH (mIU/ml)	25.17 ± 9.29	21.74 ± 7.83	7.05 ± 3.77	>0.05	<0.05
Mothers age (years)	48.15 ± 2.7	49.33 ± 3.31	50.33 ± 2.25	>0.05	>0.05
BMI (Kg/m ²)	26.18 ± 3.15	24.73 ± 3.57	25.06 ± 2.84	>0.05	>0.05
Blood group					
O	53.8 %	25 %	33.3 %	<0.05	<0.05
A	30.8 %	33.3 %	33.3 %	>0.05	>0.05
B	11.5 %	33.3 %	20 %	>0.05	>0.05
AB	3.8 %	8.3 %	13.3 %	>0.05	>0.05

Table 1 demonstrate that Mean± standard deviation(SD) of the age for group A was 30.23 ± 6.65 years and 42.75 ± 1.91 years for group B respectively, and that for group C was 27.06 ± 3.95 years. Significant difference in age (A\B) (p< 0.05) and insignificant difference in age (A\C) (P>0.05). Mean Basal hormonal level for

FSH in group A is 25.17 ± 9.29mIU/ml which is significantly different from the C group and insignificantly different from B group. Maternal menopausal age, if reached, was not significantly different between these three groups (P>0.05). The same applied to BMI in these groups (A : 26.18 ± 3.15, B: 24.73 ± 3.57, C: 25.06 ± 2.84 Kg/m²)(p>0.05).

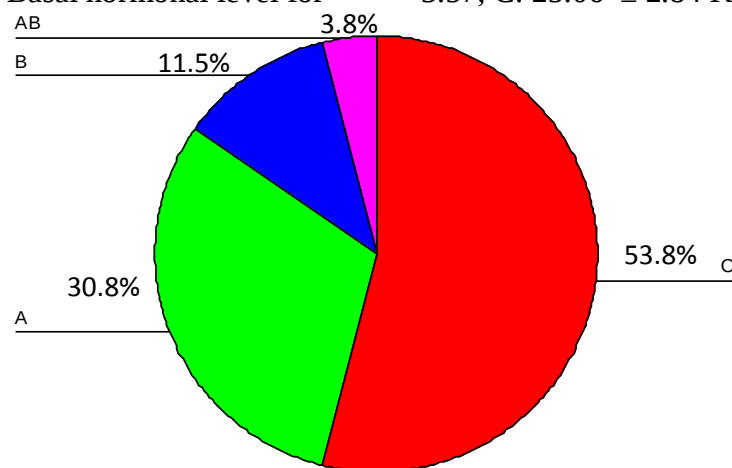


Figure 1 Prevalence of blood types according to the order of frequency in group A women.

Figure 1 It shows that blood type O rank 53.8 % of all blood types in group A women, A (30.8%), B (11.5%) and AB

(3.8%), Blood group O is significantly higher in group A women than other two groups (B, C) P<0.05(25% and 33.3%

respectively). Blood type A is distributed with no significant difference between these three groups (30.8 %, 33.3% and 33.3 %) ($p>0.05$). The same for blood type B & AB. (Table 1)

Sixty-five percent of group A women presented with infertility, while 66.7% & 86.7% are fertile in group B and C which is of significant difference ($P<0.05$).

A history of lifelong irregular menstruation was reported in 80.8% of participants in group A, while no one reported an irregular menstruation history in group C and of course its absent in B group. None of the patients in all groups had a history of radiotherapy or chemotherapy or unusual exposure to environmental toxins in the workplace. Renal disease, diabetes, thyroid disease, lupus erythematosus, and rheumatoid arthritis didn't exist in any of the participants of the study groups. None of the participants were smokers. No one had history of autoimmune disease or surgical removal of gonads.

Discussion

An understanding of why certain factors contribute to a more rapid decline in ovarian function may, for some women, help to prevent premature loss of fecundity and the subsequent impact of health problems secondary to long-term estrogen deficiency such as osteoporosis, cardiovascular disease, and possibly Alzheimer's disease. Many recent reviews have discussed the appropriate name for the condition [4, 9, 10] and it is clear that no term is perfect to name the disorder. We prefer the term premature ovarian failure (POF), until an international consensus can be gained. The condition is characterized by cessation of menstruation before the age of 40 years. A definitive diagnosis of

POF is difficult to make, and the criteria for defining POF are not always standard [9]. The diagnosis of POF can take more than 5 years [11] and clinician education' is required to raise the level of suspicion that the diagnosis might be POF in young women whose periods have become irregular and/or stopped. This concern is underlined by the fact that the diagnosis includes a simple and relatively inexpensive blood test to measure FSH levels.

In this paper a summary of the evidence regarding some factors that have been proposed to contribute to an early onset of natural menopause in subpopulation of Iraqi women from Babylon is presented. These factors include mothers age at natural menopause, BMI, blood group type, presence of menstrual irregularities and if previous fertility is present or not.

Table 1 shows that group A women have significantly lower age than women in group B and comparable to group C, a same conclusion reached by an Italian group, 2003 , and by Halder and his team in 2007 [8, 12].

The diagnosis of POF is based on finding of amenorrhoea before age 40 associated with FSH levels in the menopausal range [13]. A serum FSH level >10 mIU/ml is a commonly utilized threshold to identify women at risk for suboptimal quantitative response to ovarian stimulation and poor reproductive success, an entity alluded to as diminished ovarian reserve [14]. Mean serum FSH level in group A was >14 mIU/ml in two occasions at interval of 1 or more months .FSH levels was high in group A&B women in our study, which is obviously significantly different from group C. This finding is comparable to that of Halder et al., 2007 [12]. On comparing the three groups regarding

their maternal age at natural menopause we find no significant difference between them ($p>0.05$). This points that POF group has mothers reached the menopause at age comparable to other two groups. This finding is in contrary to other epidemiological studies who established association between menopausal age in mothers and daughters [15-17]. In the general population, genetic factors are considered to account for ~31–87% of the individual variability in age at menopause [18, 19]. A possible limitation is that our study consisted of a population that have underwent extensive psychological impact (in term of bombing and terrorist militancy) and the exposure to environmental pollution and radioactive material in term of depleted uranium in the last 20 years that have detrimental effect on ovarian reserve of the female. Social and physical stress have been linked to a greater frequency of anovulatory cycles [20, 16], this explanation is the most acceptable one, small sample size could be claimed for this result as well.

Our study concluded that there were insignificant difference between groups regarding BMI ($p>0.05$) [21,22]; but we observed that group A& C falls in overweight line according to WHO classification of obesity, while group B falls in normal weight category [23], but still they are all non-obese. BMI seems to be another determining factor for age at menopause; however, the results found in some studies are inconsistent. Parazzini, 2007 find that Body mass index was associated with the age at inception of the perimenopause only among smokers and ex-smokers, with underweight women having the earliest perimenopause(24), this could not be

applied to our study group since they were all nonsmokers.

We explored the relationship between ABO blood type and the occurrence of POF, figure 1 shows that the prevalence of blood types according to order of frequency in group A women which was: O (53%), A (30%), B (11.5%) and AB (3.8%). These results are very similar to that of Nejat and coworkers, 2011 who studied the association of blood group types and ovarian reserve according to early follicular FSH, they concluded that A blood group antigen appears to be protective for ovarian reserve, whereas blood type O appears to be associated with diminished ovarian reserve, in a relationship that is independent of advancing age [25]. This study concluded that prevalence of blood type O in group A women is significantly different from the other two groups (B&C) and further studies are needed to establish causality and identify the underlying mechanisms for the association. Other blood types were not significantly different between groups. We are very interested in these findings, in particular because this study is one of the first to successfully find a constitutional risk factor for POF.

Historically, researchers have not found an association between ABO blood group and fertility, although decreased reproductive success in women exhibiting ABO incompatibility with their male partners has been reported [26- 28].

Sixty-five percent of women from A group presented with infertility, while 66.7% & 86.7% are fertile in group B and C respectively, the difference is significant ($P< 0.05$). Our results are in line with several studies which reported an association between higher parity and later age at menopause [29, 30] or

perimenopause , the trend suggests that nulliparous women have an earlier onset of menopause than do parous women [31].

In the present study, a history of irregular menstruation was found to be strongly associated with POF. A history of lifelong irregular menstruation was reported in 80.8% of participants in the POF group, while no one reported an irregular menstruation history in the control group and of course its absent in B group

This was in line with a large Italian and Iranian studies[8 ,32] reporting that the risk of POF and of early menopause was higher among women reporting lifelong irregular menstrual cycles ; however, due to lack of clear-cut temporality assessment, in their studies and ours, caution needs to be taken in interpreting the results. Considering this point and also lack of a reasonable biological plausibility to explain a direct causality association, it cannot be clarified with certainty whether irregular menstrual cycles can be considered as a risk factor, a predisposing factor, or even an early symptom of POF [33]. Nevertheless, some studies have also supported lack of an association between irregular menstrual cycles and ovarian failure [34].

Conclusions

Maternal age of menopause constitute no risk factor for POF in our study population since they have underwent extensive psychological and environmental pollution in the last 20 years that have detrimental effect on ovarian reserve of the female. Blood group O seems to be a risk factor as well as the presence of previous infertility and menstrual irregularities. Immune,

genetic and other idiopathic causes of POF await further clarification. It is clear that this is an area of great research potential. These results must be regarded as preliminary and unconfirmed due to small sample size.

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