

MODERN APPROACH IN PREMATURE OVARIAN FAILURE

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Abstract

Premature ovarian failure (POF) is a condition affecting 1-2% of women younger than 40 years of age, characterized by amenorrhea, hypoestrogenism and elevated gonadotropin levels. In the last years it became a problem of social health interest as the frequency increased due to environmental factors and new, efficient methods for cancer treatment in young women. Few genes have been identified to explain cases of POF but there are also autoimmune associated conditions and an increasing number of iatrogenic cases (chemotherapy, surgery, radiotherapy). Modern approach in POF means not only a precise etiological diagnosis, but also a correct counseling for these patients who often want to become parents, and a chance for a healthy life without the long term consequences of estrogen deprivation from an early age. In vitro fertilization (IVF) techniques can be useful for certain cases but research is needed on strategies to improve fertility for women who have follicles remaining in the ovaries.

key words: *premature ovarian failure, estrogen deprivation, autoimmune etiology, infertility, hormonal substitution*

Introduction

Premature ovarian failure (POF), also known as premature menopause or premature ovarian insufficiency, is a common condition, affecting 1% of women under 40 years old. There are three criteria for the diagnosis: amenorrhea for minimum 4 months, hypoestrogenism and elevated gonadotropin levels - two serum follicle stimulating hormone (FSH) levels obtained at least 1 month apart [1]. Many reviews have discussed the appropriate name for this condition and it is obvious that no term is perfect as each

patient is different, not only regarding their symptoms, but also conception desire, associated comorbidities, or treatment approach [2]. The use of premature menopause seems to describe a permanent condition for these women who can induce shyness and social anxiety, impaired self-esteem and a perceived low level of social support among these women. Many women report experiencing severe emotional distress and want guidance on how to cope with the emotional implications.

Menopause is defined as a permanent cessation of menses, generally occurring about

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the age of 50 [1]. POF is not exactly a premature menopause since around 50% of affected women still experience unpredictable and intermittent ovarian function for many years [2,3]. When treating POF, hormone therapy is essential in replacing hormones the body would normally make on its own at this age. The term of premature ovarian failure can have problematic connotations of “failure” for a woman who has just received a traumatic diagnosis of future infertility [4]. This is why some clinicians prefer to use the term of premature ovarian dysfunction (POD) in an attempt to reflect the potential reversible nature of this condition and avoid the idea of failure [5].

The most recent terminology is premature ovarian insufficiency (POI) as it is thought to be more accurate and informative for patients. This terminology is widely supported by patients, as some women have a complete cessation of menstruation which does not perfectly match the concept of “insufficient follicles” since there are some conditions and treatment options which can restore ovulation temporarily and even permit to conceive [6]. Moreover, the disorder is not permanent in all women and remission of the disease is possible. In fact, up to 5% of women with POF may conceive without any specific fertility treatment and for these cases the preferred term is now POI [7].

Given that no term is perfect, probably the ongoing use of POF until an international consensus will be reached is the best option.

Pathophysiological aspects

The ovary is unique in the endocrine system in that an entirely new secretory structure is developed each month: the Graafian follicle which arises from a microscopic primordial follicle [1]. Ovarian follicular maturation is a highly organized and complex process that occurs continuously. Human females begin life

with a fixed number of primordial follicles but only a few hundred develop and release oocyte for ovulation [7]. The granulosa and thecal cells secrete different hormones and growth factors as inhibin, FOXL2 (Forkhead box L2), steroid hormones, BMP15 (bone morphogenic protein 15), GDF9 (growth differentiation factor 9) and, in turn, they are regulated by gonadotropins [4,8,9]. Menopause at older age results from the depletion of primordial follicles but POF cannot be explained by this mechanism [10].

There are two forms of ovarian insufficiency. Primary ovarian insufficiency is defined by failure of the ovary to respond to hormone signals sent from other parts of the body, like hypothalamus and pituitary gland. This is the form that is commonly referred to as premature ovarian failure [11].

Secondary ovarian insufficiency is when the problem lies directly in the hypothalamus or pituitary gland that fails to stimulate the ovaries and subsequent ovarian function. Currently, this is not considered to be POF [1,4].

Etiology

In 90% of POF cases the cause remains unknown [12,13]. Malnutrition and cigarette smoking are perhaps the only consistent environmental factors associated with earlier menopause, but for POF there does not seem to be any consistent environmental factors involved such as early menarche, use of exogenous hormones or body weight [14]. Even when we have a clear explanation and an identifiable cause for POF, we do not always understand the molecular series of events that lead to the development of this disorder. Two major mechanisms seem to be involved: follicle dysfunction and follicle depletion (no more follicles in the ovaries, similar to natural menopause) [15,16]. Follicle dysfunction indicates that there are follicles in the ovaries but

a pathological process prevents their normal function [17]. Follicle depletion may be due to a reduced initial pool of a primordial follicles, an accelerated expenditure of follicles, autoimmune or toxic destruction of follicles [16,18].

According to these mechanisms we can summarize the causes of spontaneous primary ovarian insufficiency as described in Figure 1.

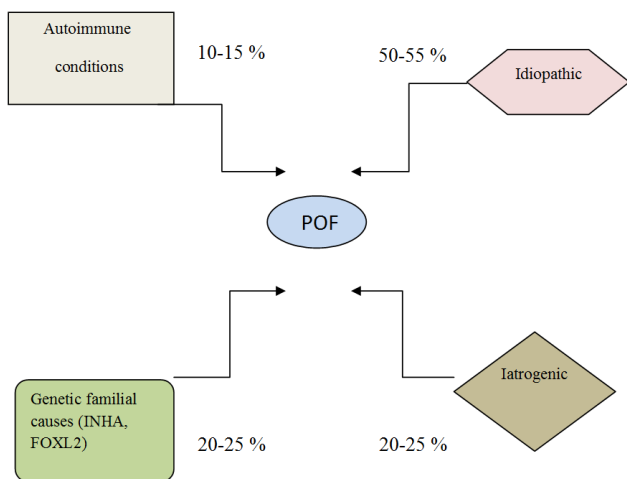


Figure 1. Causes of premature ovarian failure.

Causes of ovarian follicle dysfunction

1. *FSH or LH (luteinizing hormone) receptor mutations* - women have ovarian follicles on ultrasound evaluation, confirmed by biopsy, but without follicle development, hormonal secretion and ovulation [19]. This is a rare condition, most cases being reported in Finland;

2. *G-protein mutations* – rare mutations associated with hypoparathyroidism. Patients respond to high doses of gonadotropins [1,20];

3. *Enzyme deficiency (isolated 17,20-lyase deficiency, aromatase deficiency)* – ovarian follicles on biopsy with moderate enlargement of ovaries and functional inability of ovarian cells for estrogen synthesis [21];

4. *Autoimmune lymphocytic oophoritis* (10-20% of cases) - antral follicles with lymphocytic infiltration into theca, multifollicular ovaries. It is frequently associated with other autoimmune

conditions but not always with ovarian antibodies. Around 20% of women with POF develop autoimmune thyroiditis [22];

5. *Luteinized Graafian follicles* – antral follicles on ultrasound evaluation but histological findings show that over 60% of antral follicles are luteinized [23].

Causes of ovarian follicle depletion

1. *Insufficient initial follicle number* – mutation in FOX2; rare disorder associated in some families with blepharophimosis, ptosis, epicanthus, inversus syndrome [24].

2. *Spontaneous accelerated follicle loss* – Turner syndrome with follicle loss through apoptosis, primordial follicles being depleted before puberty; autosomal translocations occurring sporadically by chance; fragile X syndrome [25,26].

3. *Autosomal genes mutations* – inhibin α (INHA) gene has been studied as a potential candidate gene based on strong biological evidence associated with its function in the regulation of follicle loss. A decline in circulating inhibin levels associated with a decline in follicular reserve has been shown to result in raised FSH levels, increased follicular recruitment and rate of follicular depletion [26]. About 5% of women with POF have this specific mutation (gene INHA G769A).

4. *Environmental-toxin-induced follicle loss* – exposure to some chemical agents (industrial exposure to 2-bromopropane) [1].

5. *Damage to the ovaries by iatrogenic agents* such as chemotherapy or radiotherapy – the extent of the follicle loss is related to the level of exposure [27]. On the other hand, there are some studies indicating an association of Breast Cancer 1 (BRCA1) gene with occult primary ovarian insufficiency and this finding may, at least in part, explain the link between infertility and breast/ovarian cancer risk [28].

Some evidence shows that pelvic surgery can be associated with ovarian failure probably due to damage to the ovarian blood vessels as a result of surgical procedures. Young women about to begin cancer treatment are encouraged to attempt a cycle of in vitro fertilization (IVF) if it is possible to store embryos for later use [29]. Very young women may store ovarian tissue for an eventual reimplantation in the future, or for in vitro growth and maturation of immature follicles that might restore fertility [30].

Chemotherapy reduces ovarian reserve and ovarian failure is more common in women older than 35 or 40 years [28,31]. Because drug combinations are continually evolving, it is difficult to predict the degree of ovarian damage for a particular chemotherapy regimen. It is estimated that each month of chemotherapy translates into 1.5 years of lost reproductive age [32]. Most studies of fertility after cancer treatment have simply used menstruation as an outcome measure, but many cancer survivors with regular menses have reduced ovarian reserve and impaired fertility [28]. Women who stop menstruating after chemotherapy and then resume cycling have an elevated risk of premature ovarian failure compared with women who continue to menstruate during treatment [33].

Symptoms and evaluation

In most cases, the condition develops after a normal puberty and established regular menses. Only 10% of cases present as primary amenorrhea. Occasionally menses stop abruptly. In some cases menses fail to resume after a pregnancy or after cessation of oral pills. In most cases there is a period of oligomenorrhea, polymenorrhea or dysfunctional menstrual bleedings which precedes amenorrhea. Estrogen deprivation is responsible for vasomotor symptoms (hot flushes, night sweats), sleep

disturbances and dyspareunia due to vaginal dryness. Usually, at the beginning of POF, the symptoms of hypoestrogenism are mild [34].

Although most cases of POF occur sporadically, there is a positive family history with an affected first-degree relative in 10-15% of cases [6]. Patients should be queried about other autoimmune conditions such as myasthenia gravis, autoimmune thyroiditis, idiopathic thrombocytopenic purpura, type 1 diabetes (T1DM), rheumatoid arthritis, hypoparathyroidism, adrenal insufficiency, vitiligo, systemic lupus erythematosus, etc. [10]. The association with T1DM is common. Young women with type 1 diabetes have a delayed age at menarche and are at higher risk for having menstrual irregularities than nondiabetic women of similar age. Of the women with T1DM, 30% report problems such as amenorrhea, polymenorrhea or POF throughout their reproductive years. This is approximately double the prevalence of menstrual disorders observed among women without the disease, with differences most pronounced when diabetes occurs before puberty. T1DM women are also more likely to have adverse pregnancy outcomes than nondiabetic women. Spontaneous abortions, stillbirths, and congenital anomalies characterize the reproductive histories of T1DM women with poor glycemic control. With advances in intensive insulin therapy, women with T1DM are now able to maintain better glycemic control and have successful pregnancies and healthy children [6].

Peripheral hyperinsulinemia and insulin resistance occurs among approximately one-half of individuals with T1DM [3]. Hyperinsulinemia is usually associated with the polycystic ovarian syndrome (PCOS) and is characterized by hyperandrogenemia and amenorrhea. Because insulin and androgen levels are highly correlated in women with

PCOS, one may speculate that the young age at menopause in women with T1DM may be mediated, in part, through peripheral hyperinsulinemia and/or hyperandrogenemia [1].

The diagnosis of POF is based on the criteria described previously. After pregnancy is ruled out, it is important to assess serum levels of FSH, prolactin, estradiol and thyrotropin. If FSH is high, another evaluation should be made after one month, along with serum estradiol [35].

Being able to determine the ovarian reserve is important since it not only predicts fertility, but also the timing of permanent ovarian failure. The most relevant marker is anti-Mullerian hormone, which is secreted by the granulosa cells until the early antral phase. It is more precise than inhibin B in predicting the ovarian reserve [36,37]. Pelvic ultrasonography is very useful in determining the number of antral follicles and the volume of ovaries, two parameters that are also strong predictors of ovarian reserve. Women who have diminished ovarian reserve but normal menstruation and normal levels of estradiol can have impaired fertility, but are unlikely to suffer menopausal symptoms. Many studies showed that a number of follicles lower than 1000 on both ovaries indicate a permanent menopause [32].

If we want to establish the precise etiology of POF that is not associated with a syndrome, we can request a karyotype analysis and testing for a FMR1 (fragile X gene) permutation (14% of these women have this permutation and they have a 4% risk to have a child with fragile X syndrome) [38]. Testing for adrenal antibodies is required but ovarian antibodies tests are not useful as they do not have specificity [22]. Even ovarian biopsy (almost always performed during a surgical procedure with another reason) is not a very precise diagnosis method as pregnancy occurred even in cases when biopsy specimen showed that follicles were absent [39]. About

4% of women with POF have adrenal antibodies (steroidogenic cell autoimmunity and lymphocytic autoimmune oophoritis as the mechanism of deficiency) [40].

Management

The diagnosis of primary ovarian insufficiency affects a woman's physical and emotional well-being and the management should address both issues [12]. It is also very important to take into account the patient's desire for conceiving and family planning. A special case, with great anxiety and depression, is the iatrogenic POF after cancer treatment, the management of which can be very difficult in some cases.

Vasomotor symptoms can disturb sleep and contribute to chronic fatigue. These symptoms are more frequent in POF after chemotherapy or surgical procedures. All over, bothersome hot flashes were reported in 80% of women with ovarian suppression [41]. Although they are linked to estrogen deprivation, their frequency is also correlated with negative effect, particularly anxiety. The direction of this relationship is unclear, but psychotropic medications have beneficial effect both on mood and on hot flashes [43]. Placebo typically reduces hot flashes by 20-30% [42]. Although estrogen and progesterone agents reduce hot flashes by as much as 90%, the likelihood of increased risk for breast cancer rules out their use for some patients. Selective serotonin reuptake inhibitors are effective in about 50-60% of cases. However, for women with breast cancer receiving tamoxifen, these drugs (especially paroxetine) can interfere with an isoenzyme that converts tamoxifen in its active metabolite. For these cases the most common used drug is the antidepressant venlafaxine (increase central levels of serotonin and norepinephrine) and gabapentine [44]. Studies for complementary

and alternative medicines, like dietary supplements with isoflavones or phytoestrogens, have failed to show a significant reduction in vasomotor symptoms [45]. As hot flashes are strongly correlated with emotional distress it is important to use any treatment to interfere with this mechanism, including relaxation training and cognitive-behavioral therapy.

Hormone-replacement therapy is an important issue for patients with POF. Discussing about the long term effects, it is obvious that premature menopause has been associated with an increased incidence of bone fractures and increased mortality due to ischemic heart disease [36]. There aren't many studies about these long term complications for young women with POF and, in fact, usually data were extrapolated from studies which involved women over 60 years of age with physiologic menopause [37]. Data from most studies suggest that lack of progesterone and estrogen must be replaced and hormone therapy should be continued until women reach the age when menopause usually occurs. A daily dose of 100 µg of estradiol (orally or transdermal patches) achieves serum estradiol levels in the physiologic range and effectively treats the symptoms [36]. Transdermal estradiol has limited effect on hemostatic factors and it is safer to use from this point of view, having a lower risk for venous thromboembolism than oral estrogens. Association between estrogen supplementation with cyclic progestogens is safer for the endometrium (it induces a secretory endometrium and provides protection for endometrial cancer). The most used regimen includes 10 mg medroxyprogesterone acetate per day for 10-12 days a month (from day 14-16 of menstrual cycle) [46]. It is important to advise the patient that this is not a contraceptive therapy and during this treatment it is possible to

conceive. The therapy should be stopped if the period is late and pregnancy test is positive.

For women with POF who want to be sure they are not going to become pregnant during hormone replacement therapy, contraceptive pills are useful but they must know that they provide more steroid hormone than is needed (for hormone replacement.)

Regarding *bone health*, there are no studies for this age group patients, but it seems to be OK to follow the guidelines developed for perimenopausal and postmenopausal women: 1200mg of calcium per day and maintaining an adequate vitamin D status (serum 25-hydroxivitamin D level of 30 ng/ml or higher) [47]. Vitamin D deficiency is common and it has been recommended that adults with inadequate exposure to the sun to take at least 800-1000 UI vitamin D per day. Bisphosphonates are not advised if pregnancy is possible, since these agents have long skeletal half-lives and the effects on the fetus are uncertain.)

Sexual function can be impaired by increased vaginal dryness and dyspareunia. There are also psychological factors with an important role in these cases of young women who are in a difficult moment of their lives. General psychological distress and depression are also correlated with higher rates of sexual dysfunction [47]. Painful intercourse related to postmenopausal vaginal atrophy is the most frequent sexual dysfunction and appears to be a key factor in decreased sexual desire, the second most common sexual problem. Some women react by avoiding sex, but others try to hide the pain to satisfy their partners. Thus, all these patients should be discouraged from using testosterone therapy [48]. For sexual dysfunction, many patients would benefit from a brief counseling, including information about vaginal moisturizer, water or silicone-based vaginal lubricants, and training to relax pelvic

floor muscles [49]. There are some studies with a new melanocortin receptor agonist, bremelanotide, which appears to be the first centrally acting aphrodisiac [47]. Low dose topical estrogens in a form of a slow release ring or vaginal suppository is quite effective. Some estrogen can escape in general circulation in the first 2-4 weeks of treatment, but serum estrogen levels remain at postmenopausal range [49].

Fertility and family planning is probably the most important concern for these women. Patients should understand that spontaneous remission resulting in pregnancy occurs in 5-10% of cases; the remission can be temporary but in rare case can last for years [1]. Currently there are no known markers associated with an increased rate of remission and there are no general therapies for restoring ovarian function and fertility. Patients who wish to avoid pregnancy should use a barrier method, oral contraceptives or intrauterine device [23]. For couples who want to become parents the options are: adoption, egg donation, embryo donation and, in special cases, different therapeutic protocols for restoring ovarian function temporary for IVF procedures. There is no medical emergency to proceed to egg donation because rates of pregnancy with egg donation appear to be similar among older and younger patients [49]. Women with POF who become pregnant as a result of oocyte donation may have an increased risk of delivering babies who are small for gestational age and having pregnancy complications (pregnancy-induced hypertension and postpartum hemorrhage) [50].

Cryopreservation of ovarian tissue or oocytes for later in vitro growth and maturation can be possible; however, given that women who are presenting with symptoms of POF will probably have follicles of lower quality, this would require that only women who are aware of future impending ovarian failure (breast cancer

before treatment) would be able to use this technology [51]. At present, in vitro maturation of immature follicles is possible but in vitro growth and maturation from stored ovarian tissue isn't.

For patients with a proven autoimmune cause of POF there are some therapeutic strategies for restoring spontaneous ovulation [38]. Corticotherapy can restore ovarian function for these patients. The diagnosis should be made through ovarian biopsy characterized by the presence of inflammatory histological features, other coexisting autoimmune disorders and circulating organ-specific antibodies. Identifying these patients presents the opportunity to temporarily restore ovarian function by treating them with the proper immune modulation therapy: prednisone 25mg 4 times a day or dexametasone for 4-16 weeks [40]. Prednisone regimen is safer than dexametasone. On the other hand, potent immune modulation therapy can have severe adverse effects and major complications. Glucocorticoid administration is the most frequent cause of osteonecrosis of the hip or knee. This is why there is no role for empirical use of corticosteroids for POF [2]. Since POF is not a life threatening condition aggressive immune-suppression with glucocorticoids is not indicated. Thus, the use of glucocorticoids for these cases must be limited to special cases with proven autoimmune oophoritis and for a short period of time. There is no need to wait for spontaneous pregnancy during treatment and the recommendation is to try drug stimulation strategies for IVF. The results are still controversial [23].

Associated disorders can be demonstrated in POF patients. There is 50% risk of adrenal insufficiency development in women with adrenal autoimmunity. Theoretically, one would expect adrenal-cell antibodies to be present when ovarian insufficiency develops if the mechanism

of POF is steroidogenic cell autoimmunity [40]. All patients with POF should be educated regarding the symptoms of adrenal insufficiency and should undergo evaluation of adrenal function if symptoms appear. Hashimoto thyroiditis is present in 15-30% of women with POF. This is why all POF women should be tested for thyrotropin levels and presence of thyroid peroxidase antibodies [25].

Conclusions

POF diagnosis is often unexpected (and is always undesired for most women). Confirmation of diagnosis can only be made using the elevated level of FSH and low level of estradiol in a woman under 40 years old, with secondary amenorrhea (minimum 4 consecutive months). The etiology of the disorder is multifactorial, genetic factors being quite important. There are many gene mutations responsible for POF and genetic testing can be done especially in families with a history of the disease in order to advise young women without any symptoms to have children earlier.

The most common association suggesting an autoimmune etiology is with autoimmune adrenal insufficiency and autoimmune thyroiditis.

Infertility treatment in POF cases can be very difficult. Ovarian reserve (the most important marker being anti-Mullerian hormone) must be evaluated in all these cases. It is important for patients to understand that

remission may occur and that pregnancy, though unlikely, can occur in 5-8% of cases. It is important to perform IVF techniques during the intervals of spontaneous remission. For POF cases with autoimmune etiology, better results can be obtained using a short term immunosuppressive treatment (glucocorticoids) with temporary remission and medicamentous (chemical) ovarian stimulation with gonadotropins for IVF. For women diagnosed with cancer, fertility preservation can be made by freezing embryos or unfertilized mature oocytes before starting chemotherapy.

Hormonal replacement therapy has to be initiated not only because of undesirable vasomotor symptoms but also for preventing all the long term detrimental effects of an early menopause. These women should be encouraged to maintain a lifestyle that optimizes bone and cardiovascular health: regular weight-bearing exercises, maintaining adequate intake of calcium and vitamin D, avoiding obesity and screening for cardiovascular risk factors.

There is no evidence that POF incidence increases but it is becoming a more important problem since women more often delay having children until later in their lives. In addition, there is an increasing number of young women undergoing surgical, radiation or chemotherapy for cancer.

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