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Current evidences and future perspectives on patient-oriented treatments for polycystic ovary syndrome: an overview

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ABSTRACT

Polycystic ovary syndrome (PCOS) is a common metabolic disorder that typically affects women of childbearing age. Several factors are involved in the genesis of this disease but a common agreement on its etiology is still far to be reached. According to Rotterdam criteria, PCOS is characterized by oligo and/ or anovulation, biochemical hyperandrogenism and polycystic ovaries on ultrasound; following main clinical features and symptoms, women with PCOS are distinguished in four different phenotypes. The combination of different medical and, in specific cases, surgical treatments is necessary to properly reduce symptoms and restore fertility. This review points out the last evidences on medical and surgical treatments for PCOS, especially it focuses on the most common assisted reproductive techniques currently performed in case of infertility due to this syndrome. We put a special attention on the possible approaches useful for sterile women who do not respond to first line medical and / or surgical treatments.

Key words: Assisted Reproductive Techniques, Infertility, Medical Therapy, PCOS

SOMMARIO

La sindrome dell'ovaio policistico (PCOS) è una sindrome metabolica di comune riscontro nella popolazione femminile fertile. Diversi fattori vengono chiamati in causa nel tentativo di spiegare la poliedrica fisiopatologia di questa malattia, tuttavia un'unica eziologia, condivisa da tutta la comunità scientifica, non è stata ancora trovata. Secondo i criteri di Rotterdam, la PCOS è caratterizzata da oligo e/o anovulazione, iperandrogenismo e ovaia ad aspetto policistico all'ecografia; inoltre, sulla base delle principali caratteristiche e sintomi clinici, le donne con PCOS possono essere suddivise in quattro distinti fenotipi. La combinazione di diverse terapie, sia mediche che chirurgiche, è necessaria per trattare adeguatamente i sintomi e ripristinare la fertilità della donna. Lo scopo di questa review è di sottolineare le più recenti evidenze scientifiche riguardanti il trattamento della PCOS, concentrandosi sulle più comuni tecniche di riproduzione assistita utilizzate in caso di infertilità. In secondo luogo verranno anche analizzate le tecniche messe in atto in caso reiterata sterilità nelle donne che non rispondono alle terapie mediche e / o chirurgiche di prima linea.

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INTRODUCTION

Polycystic ovary syndrome (PCOS) is the most common endocrine disorder among women of childbearing age with a prevalence between 5-10% [1-3]. Its specific etiology remains largely unknown. Ovarian hyperthecosis, abnormal serum androgen levels, insulin resistance, genetic and environmental factors are the pathological base of this heterogeneous disease [4]. This disorder is associated with an increased risk of hyperinsulinemia, due to tissues' insulin resistance, type 2 diabetes mellitus, dyslipidemia, and cardiovascular diseases [5]. According to Rotterdam criteria, PCOS is defined by the presence of at least two out of the three following features: oligo-anovulation (ANOV), hyperandrogenism (HA) and polycystic ovaries by ultrasound (≥12 follicles measuring 2–9 mm in diameter, or ovarian volume >10 ml in at least one ovary) (PCOm). Four distinct phenotypes are described using a combination of Rotterdam criteria with clinical features and medical response to the tailored treatment: phenotype 1 (ANOV + HA + PCOm), phenotype 2 (ANOV + HA), phenotype 3 (HA + PCOm), and phenotype 4 (ANOV + PCOm) [5-7]. Menstrual dysfunction is a typical feature of PCOS and reflects chronic anovulation: it is characterized by irregular and infrequent bleeding, in absence of premenstrual symptoms. Clomiphene citrate (CC) remains the first-line treatment for PCOS-related anovulatory infertility [8-11]. Unfortunately, CC-resistant PCOS women are about the 5–40% of cases. CC-resistance happens when a woman do not ovulate although she has received a maximum dosage of 150 mg per day after 6 cycles, since the third day of menstrual cycle [8, 12-14]. In fact, the increased extraglandular conversion of androgens into estrogens and the consequent high estrogen production cause irregular menses and anovulation [15, 16]. Hyperandrogenism is responsible for several symptoms that include acne, hirsutism and alopecia [17]. Insulin resistance is a prominent feature of PCOS, especially among those women with hyperandrogenism and chronic anovulation [17]. PCOS is associated with four time the risk for developing prediabetes (higher serum fasting glucose and insulin levels) in children born to women with hyperandrogenism compared to those born to normal women.

A higher expression of insulin-like growth factor 2 (IGF2) has been showed in the oocytes belonging to PCOS hyperandrogenic women [2, 18, 19]. Hyperandrogenemia is associated, in PCOS women, with a more severe steatosis and with increased ALT levels, independent from obesity and insulin resistance [20-23]. Insulin resistance and the consequent hyperinsulinemia are considered of primary importance in women affected by PCOS [24, 25]. Hyperinsulinemia occurs in about 80% of obese women, while is observed only in 30–40% of cases among PCOS-lean women [26]. In order to contrast altered insulin signalling and reduce hyperinsulinemia, several insulin sensitizing drugs, have been proposed as safe and effective long-term treatments [19, 26-31]. Despite metformin is the most common used drug in the treatment for PCOS' related hyperinsulinemia, it is not well tolerated, because of its side effects, such as gastrointestinal discomforts (bloating, nausea, and diarrhea) [32, 33]. Some studies have highlighted that the combination of hyperandrogenism and anovulatory cycles may influence the risk for sex hormone related cancers in patients affected by PCOS. Insulin is an important growth factor and the insulin receptors are overexpressed in cancer cells [34, 35]. Chronic anovulation and deficient progesterone activity increase the risk for endometrial cancer, the fourth most common cancer in women in Europe [36, 37]. Although obesity is not a main criterion for PCOS, the majority of these patients is overweight; it is well known the existence of a relationship between obesity and cancers, including not only premenopausal endometrial cancer, but also biliary tract system cancer and pancreatic cancer, multiple myeloma and renal cancer [36, 38, 39]. This is why the link between PCOS and other cancer types is not obvious. PCOS patients with cycle irregularity have a reduced risk of high-grade serous ovarian tumours, whereas serous borderline ovarian tumours are more common in overweight women and in those who have never been treated with oral contraceptives [40, 41]. Moreover, the high testosterone and estrogen levels could be associated with an increased risk of breast cancer [42, 43], but a recent meta-analysis do not show an increased incidence in patients with PCOS [44]. Anyway, reproductive counselling and psychological support are necessary when cancer affects women in childbearing age because treatments could affect reproductive capacity [41, 45-49]. In this article, we reviewed the current evidences and future perspectives on PCOS with the specific aim of providing a large summary of its current treatments, knowing that an early and correct diagnosis of PCOS leads to appropriate intervention.

TREATMENTS FOR PCOS

Polycystic ovary syndrome (PCOS) is one of the most common female endocrine disorders. A close relationship exists between obesity, insulin resistance (IR) and PCOS. Hyperandrogenism (HA) and menstrual irregularities until complete amenorrhoea are the major clinical features of young PCOS women, related to abnormal circulating androgen levels. Current evidence support a close relationship between the severity of cycle irregularities and the grade of endocrine and metabolic disorders among PCOS women [14, 50]. The management of women with PCOS should include treatments for hirsutism, alopecia, acne, insulin resistance, hyperinsulinemia and ovulatory dysfunction. Moreover, treatment of ovulatory dysfunction may also reduce the prevalence of infertility, endometrial hyperplasia and, consequently endometrial cancer. Hirsutism develops gradually and it is more serious with weight gain. Girls with severe acne or acne resistance to oral and topical agents may have a 40% increased risk of developing PCOS' related physical and psychological sequels, with the necessity of proper treatments [51]. Hair loss patterns are variable, related to severity of hyperandrogenism: the areas typically involved are the vertex, the crown and, in severe cases, there should be bitemporal hair loss and loss of the frontal hairline [17]. PCOS is also related to infertility. Infertility is defined as the couple's inability or failure to conceive after regular unprotected sexual intercourses for 6 months (women aged >35 years) or 12 months (women aged <35 years) [52]. Infertility may be linked to organic causes ("organic" infertility), such as PCOS, or it can be due to non-organic causes ("functional" infertility) [47]. In both organic and functional infertility, the psychological well-being of the couple is affected [53]. How to treat infertile women with PCOS is widely discussed. The ovulatory defects and unexplained causes of infertility account for >50% of infertility etiologies and PCOS represent the primary cause of anovulatory infertility [50, 54]. Before starting any medical interventions, preconceptional counseling should be provided emphasizing the importance of life style [47]. It is well-known that the recommended first-line treatments for ovulation induction remain the anti-estrogen clomiphene citrate (CC) [55, 56]. Second-line interventions are exogenous gonadotrophins (GNs) or laparoscopic ovarian surgery (LOS). Third-line treatments are nowadays in vitro fertilization (IVF) [57]. The use of insulin sensitizing agents in ovulation induction is not recommended, but their use is surely recommended in women with glucose intolerance [11, 14, 19, 58].

The treatment of PCOS should interest all clinical disorders.

Anti-Androgen Treatments

Oral Contraceptives (OCs)

The first-line therapy for patients with PCOS with hyperandrogenic phenotype is OCs therapy. It has been demonstrated OCs therapy's safety and usefulness, especially for the most annoying clinical expressions of the disease, such as hirsutism and acne [59]. OCs, passing through the liver, increase the sex hormone binding globulin (SHBG) synthesis with the consequent decrease in free testosterone (T) levels. Thanks to this specific pharmacodynamics effect, they are more effective in the treatment of hirsutism and acne than transdermal or vaginal ring preparations [17, 30, 60] without effects on glucose metabolism. Long term OC pill use may have some limited benefits in IR, but data are still limited. Some studies have demonstrated some improvements in IR after a 6 months course of OC treatment in obese PCOS adolescents [11, 61]. Although OCs are the most common treatments for PCOS, there are doubts concerning their impact on the cardiovascular system and glucose metabolism [62-66]. OCs contain estrogen (almost exclusively ethinyl estradiol) and a progestin. A daily dose of 20 to 35 μg of ethinyl-estradiol effectively suppresses pituitary-ovarian stimulation decreasing ovarian androgen production without effects on sexual behaviour as happened for ultra-low estrogen concentration (15 micrograms) [67]. The ideal progesterone for PCOS should have the lowest androgenic profile, such as chlormadinone and drospirenone [17]. Drospirenone, a relatively new progestin, shows anti-androgenic and anti-mineralocorticoid features, even though it is weaker than the other antiandrogens in the treatment of hirsutism. Drospirenone has been widely used in combination with estrogens in women with PCOS. Recent studies have reported that a 6-month treatment with OC containing drospirenone is effective in improving hirsutism in women with PCOS [68].

Spironolactone (SPA), cyproterone acetate, finasteride, minoxidil

OCs used alone are not completely effective in arresting mild to moderate alopecia. So, when symptoms persist, it is reasonable to use a combined therapy with an anti-androgen agent to achieve

a better result. SPA, an aldosterone antagonist, competes with 5α dihydrotestosterone (DHT) to bind the androgen receptor. SPA also has several further effects, including a moderate local block of 5-alpha reductase (5αR) activity, a competition with androgens for binding to SHBG, an arrest of the conversion of Testosterone to DHT in the cells of dermal papilla and an interruption of the DHT's androgenic effect on the hair follicles [60, 69]. Its activity as progesterone, may also reduce levels of gonadotrophin-releasing hormone and attenuate the Luteinizing Hormone (LH) effects on androgen steroidogenesis. The dosage of SPA is 100 to 200 mg daily, given in two split doses [17]. Other antiandrogenic drugs such as: cyproterone acetate (at a daily dosage of 50 or 100 mg for 10 days/cycle) or flutamide (at daily dosage of 500 mg) in association with OCs are particularly effective in the hyperandrogenic phenotypes [70]. The 5α reductase inhibitors (finasteride at a daily dosage of 5mg) are also effective in these women, however they have the additional side effects, not useful in this patients, of increasing the body mass [71]. Minoxidil, used in 2% solution twice a day for at least 6 months, has shown a good effect on alopecia. Minoxidil, after its local conversion into its active form (minoxidil sulfate), opening the cells ATP-dependent potassium channels, increases the production of vascular endothelial growth factor (VEGF) in dermal papillae causing vasodilatation and improvement in hair growth. The same effect is obtained through the increase in hepatocyte growth factor (HGF) production and the consequent synthesis of prostaglandins [72, 73].

Isotretinoin

Among the new therapeutic options for PCOS patients, isotretinoin – a popular drug used to treat acne – must be mentioned. In a study by Cakir et al., women with acne (46 without PCOS and 50 with PCOS) received intramuscular injections of 0.5–1 mg/kg/dL isotretinoin, and the effects of the treatment were observed after one - two years of treatment. In both groups, the therapy was highly effective (91.6% achieved complete remission of acne), should lead us to recommend its use as a first-line treatment for PCOS patients with acne, secondary only to oral contraceptive therapy. Despite that, its high costs, the several adverse effects and its problematic effectiveness, isotretinoin treatment is not yet widely recommended in PCOS, although the findings of its effectiveness are promising [74].

Insulin sensitizing agents

Treatment of insulin resistance and hyperinsulinemia

IR affects approximately 10–15% of normal weight PCOS women and 20–40% of obese PCOS women. Additionally, women with PCOS have an increased risk of developing diabetes type II (DM2) [75]. IR is a reduced glucose response to a given amount of insulin and usually results from faults within the insulin receptor and post-receptor signalling. Therefore, circulating insulin levels rise. In the ovary, high levels of circulating insulin are thought to contribute to excess androgen production and to anovulation. Not all women with PCOS have increased IR [76]. Insulin sensitizing agents are currently being utilized to treat diabetes, and there is considerable interest for their use in the treatment of women with PCOS. Insulin sensitizers available include metformin and the thiazolidinediones (pioglitazone and rosiglitazone).

Metformin

Metformin is a synthetically derived biguanide and nowadays it is the most used and cost-effective first-line oral therapy for the treatment of DM2 [77]. The liver is the primary site of its action, in fact it reduces hepatic glucose production, it also stimulates insulin-mediated glucose uptake by hepatocytes and myocytes and it reduces substrates availability for gluconeogenesis by lowering serum lipid levels. Gastrointestinal side effects are managed by starting at a low dose of 500 mg once daily with a gradual increase to 850–1000 mg twice daily [78]. Metformin is commonly used in adolescents with PCOS as first-line monotherapy or in combination with OCs and anti-androgen medications. Metformin is currently used to target hyperandrogenemia and symptoms of androgen excess, to restore normal menses, to aid in weight reduction, and to intervene in metabolic parameters of IR [11, 16, 17, 79].

Thiazolidinediones (TZDs)

Pioglitazone and rosiglitazone, the two currently available TZDs, are effective in improving some metabolic (IR and impaired glucose tolerance IGT), hormonal (hyperandrogenaemia) as well as reproductive parameters (ovulation rate and menstrual cycle) of PCOS. Therefore, there are not sufficient evidences to support that TZDs are superior then metformin in metabolic and reproductive aspects of PCOS [80].

Glucagon-like peptide 1 (GLP1) analogues

GLP1 analogues (exenatide and liraglutide)

are the last therapeutic options for DM2 and they represent an attractive option for the treatment of obese women with PCOS who display impaired first- and second-phase insulin secretion [81]. Elkind-Hirsch et al. [82] evaluated exenatide and metformin alone and in combination with menstrual cyclicity, hormonal parameters, metabolic profiles, and inflammatory markers in overweight, insulin-resistant women with PCOS. Combination therapy was superior to exenatide or metformin monotherapy in improving menstrual cyclicity, ovulation rate, free androgen index, insulin sensitivity measures and reducing weight and abdominal fat. Both exenatide arms were more effective in promoting weight loss than metformin (P = 0.003).

Inositol

Among the insulin-sensitizing compounds, there is myo-inositol (MYO). Several studies have demonstrated that MYO is capable of restoring spontaneous ovarian activity, and consequently fertility, in most patients with PCOS [83-88]. Inositol belongs to the vitamin B complex. Epimerization of the six-hydroxyl groups of inositol leads to the formation of up to nine stereoisomers, including myo-inositol (MYO) and D-chiro- inositol (DCI); both stereoisomers were used, as insulin sensitizer drugs, in the treatment of PCOS. Despite their common origin, MYO and DCI play different roles in PCOS etiology and therapy. Indeed, in tissue such as the liver both molecules are involved in the insulin signaling, i.e. MYO promotes glucose uptake and DCI glycogen synthesis [89-91].

Insulin sensitizing agents and infertility

The Endocrine Society Clinical Practice Guidelines suggest the use of metformin in PCOS women as an adjuvant therapy for infertility (92). Metformin can improve menses and ovulatory dysfunction and, although it should not be considered a pro-ovulatory drug, it may sensitize the ovaries to the effect of CC [93-95]. It should be considered that individual responses may be variable, depending on: duration of the treatment, degree of the insulin resistance state, presence of excess body weight, particularly abdominal fat and, finally, genetic background [96]. Tang et al. [93] performed a randomized, placebo-controlled, double-blind study in 143 obese (body mass index >30 kg/m2), oligo-/amenorrhoeic women with PCOS. Metformin (850 mg) twice daily (69 patients) was compared with placebo (74 patients) over 6 months. All patients received the same advice about diet. Both groups showed significant improvements in menses; however, there were no significant differences between the two groups in insulin sensitivity or lipid profiles. Morin-Papunen et al. [94] performed a multicenter, randomized (1:1), double-blind, placebo-controlled study. Three hundred twenty women with PCOS and anovulatory infertility were randomized to metformin (n = 160 obese women, 1000 mg two times daily; non-obese subjects, 500 mg + 1000 mg daily) or identical doses of placebo (n = 160). After 3 months' treatment, when was valued necessary, another appropriate infertility treatment was combined. If pregnancy occurred, metformin/ placebo was continued up to the 12th week. Miscarriage rates were low and similar in the two groups (metformin 15.2% vs. placebo 17.9%, P = 0.8). The analysis showed that metformin significantly improved pregnancy rate (PR) and live birth rate (LBR) vs. placebo in the whole study population (PR: 53.6 vs. 40.4%, P = 0.006; LBR: 41.9 vs. 28.8%, P = 0.014) and PR in obese women (49.0 vs. 31.4%, P = 0.04), and there was a similar trend in non-obese (PR: 58.6 vs. 47.6%, P = 0.09; LBR: 46.7 vs. 34.5%, P = 0.09) and in obese women with regard to LBR (35.7 vs. 21.9%, P = 0.07). Cox regression analysis showed that metformin with standard infertility treatment increased the chance of pregnancy 1.6 times. They concluded that obese women seem to have benefited from 3 months' pre-treatment with metformin combined with routine ovulation induction in anovulatory infertility. In women with PCOS, metformin treatment before or during assisted reproductive technology cycles increases pregnancy rates and decreases the risk of ovarian hyperstimulation syndrome. However, there is no conclusive evidence of a benefit in live birth rates [95]. Inositol has an important role in infertility. In reproductive tissues such as the ovary, MYO regulates glucose uptake and FSH signaling, whereas DCI is devoted to the insulin-mediated androgen production. The new hypothesis on "DCI paradox" in the ovary has provided the key for a better and clear understanding. Unlike other tissues, the ovary is not insulin resistant. In fact, the consistent presence of epimerase enzyme, which converts MYO to DCI, makes the ovary insulin dependent. In the ovary of PCOS women, an increased epimerase activity leads to a DCI overproduction and MYO depletion. This imbalance could be the cause of the poor oocyte quality and the impairment in the FSH signaling [97]. Because of this condition, the focal point is the administration of both MYO and DCI in a proper ratio for treating PCOS [86, 98, 99]. DCI alone, mostly

when it is administered at high dosage, negatively affects oocyte quality, whereas the association MYO/DCI, in a combination reproducing the plasma physiological ratio (40:1), represents a promising alternative in achieving better clinical results, by facing PCOS on both systemic and ovarian level [91].

VITAMIN D

Vitamin D acts through the "vitamin D receptor" (VDR), that is the final common pathway through which it works on target tissues, in particular on the reproductive system. Several studies have demonstrated a strong correlation between vitamin D deficiency (concentration of 25-hydroxycalciferol < 20 ng/mL) and PCOS. Vitamin D deficiency in PCOS women is associated with more severe metabolic disorders, such as obesity and insulin resistance. Therefore, vitamin D supplementation is recommended in the case of considerable deficiency, in particularly in obese and insulin-resistant PCOS women because it might improve their metabolic parameters [100].

LIFESTYLE MODIFICATION AND BARIATRIC SURGERY

Obesity is common in women with PCOS. Weight loss is recommended in obese women with PCOS looking for pregnancy. This recommendation is based on the benefits of weight loss seen in multiple other conditions, such as diabetes and cardiovascular disease, but it is also related to recognition of a poor reproductive outcome in obese patients. Multiple studies have shown that weight loss is associated with improved spontaneous ovulation rates in women with PCOS [101]. The treatment of obesity involves several strategies: behavioral counseling, lifestyle therapy (diet and exercise), pharmacological treatment and bariatric surgery [102].

Several studies have examined as therapy the combination of diet and physical exercise. Regular physical activity plays an important role in weight loss programmes because it is associated with better long-term weight loss maintenance. In overweight and obese women with PCOS and infertility, lifestyle modification alone should be the baseline therapy for 3-6 months, in fact it is able to determine a 5-7% of weight reduction and to improve ovulation [103]. In some cases, the available literature supports the adjuvant use of bariatric surgery and pharmacological weight loss for the treatment of obesity in PCOS, although large clinical trials are needed [14]. In women with Body Max Index (BMI) >35 Kg/m2 after failed attempts at weight loss for more than 1 year and/ or in presence of obesity-related co-morbidities should be considered bariatric surgery. This procedure is recommended to monitor foetal growth during pregnancy [103, 104].

OVULATION INDUCTION TREATMENT

Clomiphene citrate, Tamoxifene, Aromatase inhibitors (AIs)

Clomiphene citrate (CC) is the first choice treatment for induction of ovulation in anovulatory women with PCOS. Cost of medication is low, the oral administration is easy and there are relatively few adverse effects. CC is thought to be involved in the interruption of the negative feedback mechanism that results in increased secretion of follicle-stimulating hormone (FSH) [103]. Selection of patients for treatment should take account of body weight/ BMI, age (poorer outcome in older patients may justify consideration of alternative treatments such as exogenous GNs or IVF) and other infertility factors [14]. The starting dose of CC generally should be 50 mg/day (for five days, starting on day 2 – 5 following a spontaneous or progestin-induced bleeding). The recommended maximum dose is 150 mg/day, as there is no clear evidence of efficacy at higher doses. Monitoring by ultrasound (US) is not mandatory to ensure good outcome but it is suggested to allow adjustment of the dose in following cycles, according to the observed response [105]. In the absence of complete cycle monitoring, a pre-treatment US evaluation is often performed to scan ovarian and endometrial morphology, which may be followed by serum progesterone measurements (typically one or two samples in the estimated luteal phase). There is no evidence that administration of human chorionic gonadotrophin (hCG) in mid-cycle improves the chances of conception [106]. Approximately 75 – 80% of patients with PCOS will ovulate after CC. Most reliable studies indicates a conception rate of up to 22% per cycle in those ovulating on CC. Treatment generally should be limited to six (ovulatory) cycles. Further cycles (maximum 12 in total) may be considered on an individual basis after discussion with the patient. Cumulative live birth rate is between 50–60% for up to six cycles [8]. Side effects are: hot flushes, headaches and

visual complaints; this drug is generally well tolerated. The multiple pregnancy rate is 10%, while hyperstimulation syndrome is rare. Anti-estrogenic effects on endometrium and cervical mucus may occur but appear to represent an idiosyncratic response [107]. Tamoxifen appears to be as effective as CC for induction of ovulation, but its use for that purpose is off-label. It may be considered only in women who suffer intolerable CC' side effects such as hot flushes [107]. AIs are widely discussed in ovulation induction in PCOS patients. Initial preliminary studies suggest that letrozole appears to be as effective as CC for induction of ovulation, but the drug is currently not approved for treatment of infertility. It may be considered as an 'off-label' option for some patients after appropriate counselling with the patient on its risks and benefits [14]. AIs prevent estrogen biosynthesis from androgens and through the hypothalamic/pituitary cross-talk, increase FSH secretion. AIs theoretically avoid adverse effects of CC, as they do not affect estrogen receptors centrally or within the endometrium [108]. Legro et al. [109] performed a double-blind, multicenter trial. They random assigned 750 women, in a 1:1 ratio, to receive letrozole or CC for up to five treatment cycles. Women who had received letrozole had higher live birth rates than those who received CC (27.5% vs. 19.1%, p=0.007). There were no significant differences in overall congenital anomalies, though there were four major congenital anomalies in the letrozole group versus one in the CC group (p=0.65). The cumulative ovulation rate was higher with letrozole than with CC (61.7% vs. 48.3%, p<0.001). There were no significant differences in pregnancy loss (31.8% in letrozole group vs. 29.1% in CC group) or twin pregnancy (3.4% and 7.4%, respectively). CC was associated with a higher incidence of hot flushes, and letrozole was associated with higher incidences of fatigue and dizziness. Rates of other adverse events were similar in the two treatment groups. So they concluded that letrozole was associated with higher live-birth and ovulation rates among infertile women with PCOS. In a Cochrane review Franik et al. [110] included 26 RCTs (5,560 women). Twelve RCTs studied live birth rates showing that in letrozole group compared with CC group, the birth rate was higher, while letrozole group compared with those treated with laparoscopic ovarian drilling, showed no statistically significant differences. Sixteen RCTs demonstrated that there was no difference in the rate of ovarian hyperstimulation syndrome (OHSS) comparing letrozole, alone or in combination, with any of the following: placebo, CC, CC followed by intrauterine insemination (IUI), laparoscopic ovarian drilling. Twenty-five RCTs underlined higher pregnancy rates in the letrozole group in comparison with CC (alone or in combination in one or both arms) followed by timed intercourse. The pregnancy rate was higher in the letrozole group comparing it with CC, alone or in combination, followed by IUI, while there was no difference between the groups when letrozole, with or without metformin, was compared with laparoscopic ovarian drilling. Therefore letrozole can be used as second-line pharmacological therapy in women with PCOS who have CC resistance and/or failure, and are affected by pure anovulatory infertility [103].

Gonadotrophins

The ovulation induction using gonadotrophin therapy is based on the physiological concept that initiation and maintenance of follicle growth may be achieved by a transient increase in FSH, above a threshold dose for sufficient duration, in order to generate a limited number of developing follicles. Low-dose protocols (37.5 – 75 IU/day) have been developed to reduce the frequency of ovarian hyperstimulation. The increased LH secretion in PCOS may interfere with fertility. The mechanisms include premature oocyte maturation and elevated LH levels, associated with an increased pregnancy loss. Overall, low-dose regimens result in a monofollicular ovulation rate of 70%, a pregnancy rate of 20% and a multiple live birth rate of 5.7% [111].

Laparoscopic surgery

Another therapeutic option for PCOS women suffering from infertility is the laparoscopic surgery, especially in those who had shown a resistance to pharmacological therapies. Laparoscopic techniques that can successfully trigger ovulation include ovarian biopsy and electrocautery, laparoscopic ovarian drilling, transvaginal hydrolaparoscopy, ultrasound-guided transvaginal ovarian needle drilling or laparoscopic ovarian multi-needle intervention [9, 10, 12].

Since its introduction in 1984, laparoscopic ovarian drilling has evolved into a safe and effective surgical treatment for anovulatory, infertile PCOS women resistant to CC. It is as effective as GNs in terms of pregnancy and lives birth rates, but without the risks of ovarian hyperstimulation syndrome and multiple pregnancies. It improves ovarian responsiveness

to following use of ovulation induction agents. Its use in unselected cases of PCOS or for non-fertility indications is not prudent leading to the risks of iatrogenic adhesions and ovarian insufficiency [112]. Badawy et al. [113] performed a study to evaluate the outcome of ovarian needle drilling using transvaginal ultrasound guidance as an alternative to the traditional laparoscopic electrosurgical drilling for patients with PCOS. The study involved 163 PCOS patients with clomiphene-resistance. Patients were randomly allocated to either treatment with ultrasound-guided transvaginal needle ovarian drilling (UTND; n = 82) or laparoscopic electrosurgery ovarian drilling (n = 81). There were no significant differences between the two groups with regard to body mass index, hormonal profiles, clinical manifestations, and ultrasound findings of PCOS. There were no significant differences between the two groups about normal menstruation, hirsutism, acne, ovulation, and pregnancy. UTND resulted in significant improvement in the ovulation, pregnancy, hirsutism, and acne. There were significant decreases in the serum LH and Testosterone levels but not in the FSH or LH/FSH levels after UTND as well. It is important to underline that in 50% of laparoscopic surgery-treated women, adjuvant therapy often are required [14].

ASSISTED REPRODUCTION TECHNIQUES (ART): INTRAUTERINE INSEMINATION (IUI) AND IN VITRO FERTILIZATION (IVF)

Induction of ovulation followed by IUI is indicated in PCOS women when there is associated male factor. Semen preparation is necessary before IUI. The clinical pregnancy rates in each cycle were between 11 to 20% and the multiple pregnancy rates from 11 to 36% [14]. IVF is considered another optional treatment, usually chosen when there are tubal factors and severe male factors. Although infertile women with PCOS may typically present with increasing oocytes retrieved in IVF cycles, most oocytes have poor quality [114]. Several stimulation protocols have been published for the treatment of patients with PCOS undergoing IVF, including CC associated with human menopausal gonadotrophins (hMG), hMG alone, recombinant FSH (recFSH) alone, GnRH agonist associated with hMG or recFSH and GnRH antagonist associated with hMG or recFSH. Currently, the most standard protocol is a long desensitization protocol associated with FSH. The clinical pregnancy rate for each started cycle is similar between PCOS and non-PCOS patients (about 35%). The most important complication of ovarian stimulation is the occurrence of OHSS [115]. Several studies try to determine the best method to control ovarian hyperstimulation (COH) in PCOS patients. Onofriescu et al. [116] compare the GnRH agonist long protocol with the flexible GnRH antagonist protocol in infertile PCOS women undergoing COH in terms of clinical pregnancy rate (CPR), with special reference to the incidence of OHSS. No differences were observed in clinical pregnancy rate (CPR) in the agonist and antagonist protocols, respectively. Incidence of OHSS was lower in the antagonist compared with agonist group (4% versus 28%). Duration of stimulation (13,80 + 1,4 vs 11,85 + 2,4 $p < 0,001$) and total gonadotrophin doses required (2435,5 + 884,5 versus 2005, 5 + 545,5 IU $p < 0.003$) were also lower in the antagonist treated group compared with those treated with agonist protocol. Krishna et al. [62] showed that gonadotropin-releasing hormone agonist (GnRHa) trigger is a better alternative to human chorionic gonadotrophin (hCG) in PCOS undergoing IVF cycles with GnRH antagonist protocol for the prevention of ovarian hyperstimulation syndrome (OHSS). A total of 227 patients diagnosed with PCOS, undergoing IVF in an antagonist protocol were recruited and randomly assigned to two groups: Group A (study group): GnRHa trigger 0.2 mg (n = 92) and Group B (control group): 250 µg of recombinant hCG as trigger (n = 101) 35 h before oocyte retrieval. The incidence of moderate to severe OHSS in the hCG group was 37.6% and 0% in the GnRHa group with $P < 0.001$. The GnRHa group had significantly more mature oocytes retrieved (19.1 ± 11.7 vs. 14.1 ± 4.3), more fertilized oocytes (15.6 ± 5.6 vs. 11.7 ± 3.6), and a higher number of top quality cleavage embryos on day 3 (12.9 ± 4.7 vs. 7.5 ± 4.3) than the hCG group.

DISCUSSION

PCOS represents a complex syndrome that includes clinical, endocrine and metabolic disorders that commonly lead to infertility (14). The clinical and endocrine involvements, such as HA and menstrual irregularities, are the major complaints in young PCOS women. OCs seem to represent an effective and safe treatment in every PCOS phenotype. Some authors reported

that in HA/PCOS patients, OCs can be used for symptom relief. The real problem is to find a good OC in patients with metabolic risk, overweight or moderate IR: surely a vaginal contraceptive ring should be preferred to oral OC. In these patients, the combination of OCs and myo-inositol may be more effective in controlling endocrine and metabolic profiles [59]. Several studies suggested the use of antiandrogen drugs alone or combined with another antiandrogen, with an insulin sensitizer or with an oral contraceptive pill to control androgenic symptoms. The combined treatments are generally more effective and preferred than single drug based ones. In particular, a combination of an antiandrogen OCs with metformin may lead to more beneficial metabolic effects than monotherapy with an antiandrogen or an OC alone. The anti-androgen should be used at the lowest effective to avoid dose-related side effects and reduce costs [68]. SPA and finasteride are often added to OCs in adolescents with PCOS in order to treat the androgen excess expressions, such as acne, hirsutism, and alopecia. Unfortunately the major risk of both of these medications is teratogenesis [17]. Metabolic disorders should be treated with insulin sensitizer agents alone or in combination with other drugs. Evidence supports the use of metformin in obese/overweight adult women and adolescent girls to help reduction in androgen excess and improvement of ovarian function. Many adolescents who do not want to use OCs may benefit from metformin monotherapy [17]. There are no properly designed studies pointing the best therapies for infertility in women with PCOS. Generally, a combination of medical and behavioral therapies can lead to the greatest weight loss, though long-term bariatric surgery is associated with the best weight maintenance after weight loss [104]. The lifestyle modification is the baseline therapy for PCOS women looking for a pregnancy [101, 102]. For women who do not find benefits from the lifestyle modification alone or bariatric surgery in selected cases, it is necessary to move to ovulation induction procedures. CC is the first line treatment for its easy administration, its effectiveness and its few collateral effects [8, 106, 107, 109]. Also AIs are proposed as ovulation inductors, but prospective studies on their efficacy and safety should be done, before recommending the use of aromatase inhibitors [14]. Roque et al. performed a systematic review and meta-analysis in order to identify the results of randomized controlled trials (RCTs) comparing the use of letrozole to CC for ovulation induction in patients with PCOS. They showed a statistically significant increase in live birth and pregnancy rates in the letrozole group when compared to the CC group and they showed no differences in multiple pregnancy, miscarriage and ovulation rates between the two groups. Regardless of these opinions, up to now AIs are not recommended for routine ovulation induction [117]. In patients resistant to CC, it could be suggested the use of Gonadotrophins, that seem to represent the second-line treatment for PCOS patients seeking pregnancy [8]. The use of exogenous gonadotrophins is associated with increased chances for multiple pregnancies and, therefore, intense monitoring of ovarian response is required. Another ovulation induction method is represented by Laparoscopic surgery [118]. Usually alone is effective in <50% of women and additional ovulation induction medication is required [14]. UTND can be adopted as an outpatient office procedure. The easy scheduling, the low costs and rapid recovery suggest it as the first alternative treatment for PCOS women resistant to CC [112]. Overall, ovulation induction is reported to be highly effective with a cumulative singleton live birth rate of 72%. More patient-tailored approaches should be developed for ovulation induction based on initial screening characteristics of women with PCOS [14]. Several studies have demonstrated that metformin is an adjuvant therapy for infertility [92]. However, its use in PCOS patients is widely discussed and we are still far from a common agreement about its routine use. Tang et al. showed that metformin does not improve weight loss or menstrual frequency in obese patients with PCOS. They demonstrated that weight loss alone through lifestyle changes improves menstrual frequency [93]. Metformin is preferred to insulin-sensitizing agents for PCOS, even if there are many other available drugs. For example, TZDs may be considered as an alternative treatment in insulin-resistant or obese PCOS women who do not tolerate or do not respond to metformin therapy [93]. ART should be considered the right choice in patients resistant to other treatments or with other infertility factors. Low dose gonadotrophins protocols are preferred (37.5-75 IU/day) in PCOS patients undergoing IUI. In case of IVF, the goal remains to find the right protocol. Recent studies suggested that the flexible GnRH antagonist protocol is associated with a similar ongoing pregnancy rate, lower incidence of OHSS grade II, lower gonadotrophin doses requirement and shorter duration of stimulation, compared with GnRH agonist. Therefore, the GnRH antagonist might be the treatment of choice

for patients with PCOS undergoing IVF [113]. Jiang et al. [119] performed a retrospective study (174 obese PCOS) to explore the effect of CC on the cycle characteristics and outcomes of obese women with PCOS undergoing ovarian stimulation for IVF. In the study group (n = 90), CC and human menopausal gonadotrophin (HMG) were administered simultaneously starting since the third cycle day, while in control group (n=84) HMG was used only. MPA was used to treat both of the two groups for preventing premature LH peak. Ovulation was co-triggered by a GnRH agonist and hCG when dominant follicles matured. The study group received obviously lower total HMG dose but similar HMG duration. While the antral follicle count (AFC) was higher in study group, the number of oocytes retrieved and top-quality embryos were remarkably less. The mature oocyte rate was higher in study group (p = .036). No significant differences were detected in fertilization rate and cleavage rate between the two groups. CC has a positive influence on cycle characteristics, but might be correlated with the impaired IVF outcomes (top quality embryos and lower oocyte retrieval rate) in obese PCOS patients undergoing IVF, when HMG and MPA are used simultaneously. An important aspect of IVF in PCOS patients is the risk of OHSS. In PCOS patients, undergoing ovarian stimulation through antagonist IVF cycles, the most effective strategy, significantly reducing the occurrence of OHSS, is the use of GnRHa trigger, that is also able to improve the number of mature oocytes and the quality of embryos compared with hCG trigger [115]. Beside all these different therapies, nowadays further studies are necessary to find the best approach to such a complex syndrome, in order to target the right treatment according to the clinical characteristics for each single PCOS patient.

DECLARATION OF INTEREST

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper. No specific funding was obtained.

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