eXpert

EXcellence in PCOS & Expertise in Reproductive Technology

MODULE 4:
ROLE OF ADJUVANTS IN INFERTILITY MANAGEMENT: FROM PHYSIOLOGY TO THERAPEUTICS

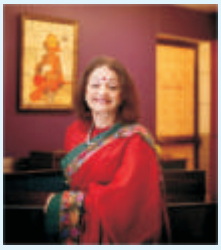
Brought to you by

THE PCOS SOCIETY

An Initiative of PCOS Society (India)

eXpert
EXcellence in PCOS & Expertise in Reproductive Technology

Foreword



Duru Shah
Founder President,
The PCOS Society of India,
Course Director, **EXPERT**

Madhuri Patil
Scientific Co-ordinator,
The PCOS Society of India,
Course Director, **EXPERT**

PCOS is quite often associated with infertility, especially in women with irregular periods and hyperandrogenemia. It is one of the most treatable forms of infertility, if ovulation induction is optimal. Unfortunately, many PCOS women, both lean and obese, behave erratically during ovulation induction sometimes leading to ovarian hyperstimulation syndrome which can become a serious iatrogenic complication. Keeping in mind that many PCOS women approach us for management for their infertility issues, it is important for us to understand the principles of management. Which when applied correctly, they can give us great success in making PCOS women pregnant. Of course experience counts, and as we continue treating women, we learn how to tweak our stimulation protocols to avoid complications and give us the best results!

After initiating the Basic Course on Infertility in 2018, we are delighted to introduce to you the Advanced Course called **"EXPERT"- (Excellence in PCOS and Expertise in Reproductive Technology")** a Certificate Course brought to you by the PCOS Society of India, through an unrestricted educational grant by Sun Pharma, Inca Life Sciences.

"EXPERT" will be presented to you in a set of 6 Modules which will update you on various aspects of the management. Infertility in PCOS, from minimal intervention to Assisted Reproduction.

Once you complete the 6 Modules, you could participate in an Online Exam, Assessment and on clearing it, you will be eligible to receive a beautiful certificate from the PCOS Society of India, which you will be extremely proud to display! To own this Certificate, you need to be a member of the PCOS Society, India!

To become a member, please log on to http://www.pcosindia.org/ to download your form and become a Life Member or Patron Member of the PCOS Society of India

If you have any queries, please write to us at thepcossociety@gmail.com

Both Madhuri and myself have worked hard on creating this program and we thank the team at Sun Pharma for their support in making this program a reality !

Enjoy reading.

With warm regards,

Duru Shah
Founder President, The PCOS Society of India.
Course Director, EXPERT

Madhuri Patil
Scientific Co-ordinator, The PCOS Society of India
Course Director, EXPERT

eXpert
EXcellence in PCOS & Expertise in Reproductive Technology

Contents

ROLE OF ADJUVANTS IN INFERTILITY MANAGEMENT: FROM PHYSIOLOGY TO THERAPEUTICS

eXpert
EXcellence in PCOS & Expertise in Reproductive Technology

ROLE OF ADJUVANTS IN INFERTILITY MANAGEMENT: FROM PHYSIOLOGY TO THERAPEUTICS

Adjuvant Therapies in Infertility Management: An Overview

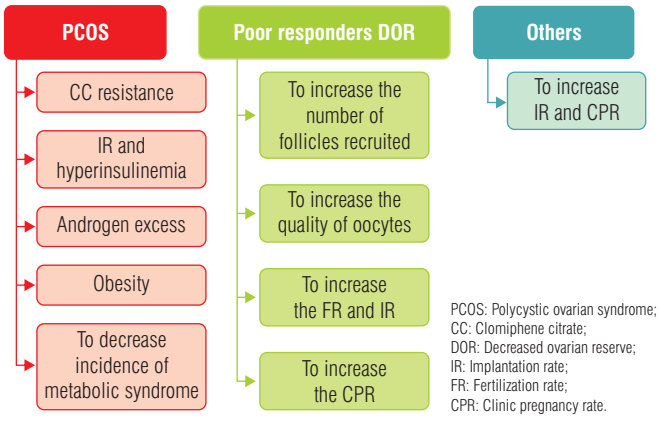
In vitro fertilization (IVF) has become a widely accepted procedure to help improve the chances of conception in infertile or subfertile conditions. However, despite improved technologies and better results in recent years, most patients do not have a successful pregnancy following a single IVF treatment. Repeated treatments and failures have negative repercussions on the quality of life, and each failed cycle incurs substantial financial costs. It is estimated that 10%–15% of couples seek professional help for difficulty in conceiving at sometime during childbearing years. The expense, time, stress, and frustration felt by couples and physicians have led to a search for new drugs and technologies that will increase success rates.[1]

The word adjuvant is derived from the Latin word "Adjuvare," which means to hold. Hence, adjuvant is a drug or substance that enhances the efficacy of a primary drug. Although mechanisms of actions have been proposed, justification for the use of adjuvant therapies is usually empirical and is based on physicians' personal views. The use of adjuvant therapy may improve the IVF outcome and may be particularly beneficial for women with a history of repeated IVF failure. Common adjuvants used to improve ovarian response include glucocorticoids and growth hormone (GH), and common adjuvants used to improve endometrial receptivity include induction decidualization of a local injury to the endometrium through the Pipelle procedure.[1]

Adjuvant Therapies Used During Ovarian Stimulation

Adjuvant therapies are helpful in various infertile situations, primarily in polycystic ovarian syndrome (PCOS) and in poor responders. A detailed need of adjuvant therapies has been listed in Figure 1.[1–3]

Figure 1: Need of adjuvant therapy in ovulation induction.[1–3]



PCOS: Polycystic ovarian syndrome; CC: Clomiphene citrate; DOR: Decreased ovarian reserve; IR: Implantation rate; FR: Fertilization rate; CPR: Clinic pregnancy rate.

Approved and Off-label Drugs Used

A few drugs are approved as adjuvants in ovulation induction, while others are off-label drugs. Figure 2 provides the list of approved and off-label drugs. Off-label drugs have been evaluated in the phase I or phase II trials of clinical research, but they have not been fully assessed in phase III or phase IV trials.[4,5]

Figure 2: Approved and off-label drugs used as adjuvant therapy in ovulation induction.[4,5]

Approved drugs	Off-label drugs
• CC	• Aromatase inhibitors
• Gonadotropins (FSH; LH + FSH)	• Tamoxifen
• hCG	• Dexamethasone
• GnRH antagonists and agonist	• Prednisone
• Progesterone	• Dopamine agonist
	• Metformin
	• Myoinositol
	• N-acetyl cysteine
	• Vitamin D
	• Growth hormone
	• DHEA/testosterone
	• L-arginine
	• Antioxidants and CoQ10
	• Aspirin
	• Sildenafil

LH: Luteinizing hormone; FSH: Follicle-stimulating hormone; hCG: Human chorionic gonadotropin; CC: Clomiphene citrate; GnRH: Gonadotropin-releasing hormone proved drugs; DHEA: Dehydroepiandrosterone; CoQ10: Coenzyme Q10.

Adjuvants in Ovulation Induction

Adjuvants in ovulation induction

Adjuvants in treatment of PCOS	Adjuvants in treatment of poor responders	Other adjuvants
Glucocorticoids prednisone, methylprednisolone and dexamethasone	Growth hormone/ GH-releasing factor (GHRF)	Antioxidants
Metformin	Pyridostigmine	Micronutrients
Myoinositol	DHEA/testosterone	Dopamine agonist
N-acetyl cysteine	L-arginine	Aspirin
Melatonin	Aromatase inhibitors	Sildenafil
Vitamin D	Estrogen pretreatment	
Chromium polynicotinate	Antioxidants	
	GCSF	

PCOS: Polycystic ovarian syndrome; GH: Growth hormone; DHEAS: Dehydroepiandrosterone; GCSF: Granulocyte colony-stimulating factor.

Glucocorticoids

Glucocorticoids have been proposed as a useful adjuvant to both clomiphene citrate (CC) and gonadotropin ovulation induction in women with PCOS with a therapeutic rationale based on reducing ovarian androgen levels, improving ovulatory function, and reducing resistance to ovulation induction agents. Although the source of high androgen secretion in anovulatory women with PCOS is primarily ovarian, 50%–70% women also demonstrate excessive adrenal androgen levels.[6,7]

Glucocorticoids reduce adrenal androgen production by negative feedback inhibition of adrenocorticotrophic hormone production. Hence, the administration of low doses of glucocorticoids may benefit women with hyperandrogenic anovulation. The mechanism of action presumably involves a reduction in adrenal androgen secretion, which in turn may reduce total circulating androgen levels by as much as 40%, improving folliculogenesis. While major complications from the adjuvant use of low-dose glucocorticoids are rare, weight gain is a common problem. Other reported side effects include glucose intolerance and osteoporosis. Given possible side effects, their use should remain as a second-line therapy.[6,7]

A study evaluated the effects of short-course administration of dexamethasone (DEX) combined with CC in CC-resistant patients with PCOS and normal dehydroepiandrosterones (DHEAS) levels and suggested that mean follicular diameters were 18.4124±2.4314 mm and 13.8585±2.0722 mm for the treatment and control groups, respectively. Eighty-eight percent of the treatment group and 20% of the control group had evidence of ovulation. Forty-six (40.5%) pregnancies resulted after the combined use of CC and DEX, whereas only five (4.2%) women in the control group conceived.[8]

Another study was conducted to determine the safety and effectiveness of systemic glucocorticoids during ovarian stimulation for IVF and intracytoplasmic sperm injection (ICSI) cycles. Four randomized controlled trials (RCTs) were included in the review (416 women) and the trials compared glucocorticoid supplementation during IVF stimulation vs. placebo. The results suggested that if the chance of live birth with placebo is assumed to be 15%, the chance following supplementation would be between 7% and 31%. There was no conclusive evidence of a difference in the clinical pregnancy rate.[9]

The evidence suggests that if the chance of clinical pregnancy with placebo is assumed to be 24%, the chance following treatment with glucocorticoid supplementation would be between 23% and 47%. There was also insufficient evidence to determine whether there was any difference between the groups in multiple-pregnancy rate or miscarriage rate. Neither of the studies reported Ovarian hyperstimulation syndrome (OHSS) or side effects. Further, the safety and effectiveness of glucocorticoid administration in women undergoing controlled ovarian hyperstimulation for IVF/ICSI cycles are unclear due to the small number of studies and low event rates. While glucocorticoids possibly increase the clinical pregnancy rate, there may be little or no impact on live birth rate.[9]

Metformin

Metformin belongs to the class of biguanides and is the most common drug used as first-line oral therapy for the treatment of type 2 diabetes. Anovulatory women with PCOS and who are resistant to CC may be prescribed metformin as a second-line treatment as adjunct therapy to CC and gonadotropin ovulation induction. It may also be used as an adjunct therapy to reduce the risk of developing ovarian hyperstimulation syndrome in PCOS women undergoing Gonadotropin-releasing hormone (GnRH) agonist long protocol IVF/ICSI.[10]

Myoinositol

Myoinositol is the most commonly found form of inositol. Several studies have reported that one of the mechanisms of insulin deficiency may be initiated by inositol phosphoglycan mediator and that the insulin resistance is due to deficiency of inositol in the inositol phosphoglycans . Myoinositol plays an important role for the signal pathways of cells, particularly in individuals with PCOS. An improved insulin sensitivity may be observed due to the action of myoinositol in PCOS pathway. Several studies indicate that myoinositol is an effective alternative in the treatment of PCOS with no side effects with a standard dosage.[11]

N-Acetyl Cysteine

N-acetyl cysteine (NAC) is a mucolytic agent and has a role in the treatment of infertility. A limited number of studies conducted in the recent years has reported possible benefits of improving insulin sensitivity and better outcomes in ovulation induction in patients with PCOS. It is also recommended as an adjuvant to CC for induction of ovulation and improving pregnancy rates in PCOS patients.[12]

Melatonin

Melatonin is an indoleamine produced by pineal gland. Melatonin is linked to several reproductive functions in humans. It has a wide range of functions as circadian pacemakers, hypothalamic/pituitary axes to vasomotor effects, immunomodulatory, antioxidative actions and anti-apoptotic effects (direct and indirect). Owing to its safety and efficacy, it has been indicated as an adjuvant therapy for PCOS.[13]

Vitamin D

Vitamin D is a steroid hormone synthesized in the skin by ultraviolet light. Several studies have indicated that most of the women with PCOS are vitamin D deficient. Vitamin D deficiency may be associated with obesity, insulin resistance and metabolic syndrome, all of which are commonly observed in PCOS and ovulatory dysfunction. In women with PCOS, vitamin D can improve menstrual irregularity, follicular development, and pregnancy rate.[14]

Chromium Polynicotinate

Chromium is a metal that exists in a variety of oxidative states. Its deficiency may result in insulin resistance and evidence from clinical studies suggests that low levels of chromium may disrupt glucose and insulin regulation. It is used as an adjuvant therapy for the treatment of anovulation in infertile patients with PCOS.[15]

Summary

Adjuvant in PCOS	Application
Dexamethazone	• Beneficial in elevated DHEAS androgen levels • Highly effective adjunct to CC in PCOS women • Should be avoided in women with diabetes
Metformin	• Decreased the risk of OHSS when gonadotropins were used for ovulation induction • Increases the live birth rate (LBR) among women undergoing ovulation induction with gonadotrophins (GTs)
Myoinositol	• An insulin sensitizer which has beneficial effects on ovarian function and response to assisted reproductive technology (ART) in women with PCOS • No data on its effects on pregnancy rates (PR) and LBRs
Vitamin D	• Influences ovarian endocrine function and likelyhood of pregnancy • Inverse associations 25-hydroxy vitamin D levels and insulin resistance, features of hyperandrogenism and circulating androgens in women with PCOS
N-acetyl cysteine	• Improves insulin sensitivity and decreases androgen level • Can improve the ovulation and pregnancy rates • May also have some beneficial impacts on endometrial thickness
Melatonin	• Regulates a variety of important central and peripheral actions related to circadian rhythms and reproduction • Powerful free radical scavenger and has a broad-spectrum antioxidant property • Melatonin deficiency seems to be involved in pathophysiology of PCOS
Chromium polynicotinate	• Active component of glucose tolerance factor, which is responsible for binding insulin to the cell membrane receptor sites • Improves insulin sensitivity • Stimulates the metabolism of sugar, fat, and cholesterol

Adjuvants in the Treatment of Poor Ovarian Responders

Adjuvants used in the treatment of poor responders include GH, GH releasing factor, pyridostigmine, DHEA or testosterone, L-arginine, aromatase inhibitors, estrogen pre-treatment, antioxidants, and GCSF. Other adjuvants include antioxidants, micronutrients, dopamine agonist, aspirin, and sildenafil.[1-3]

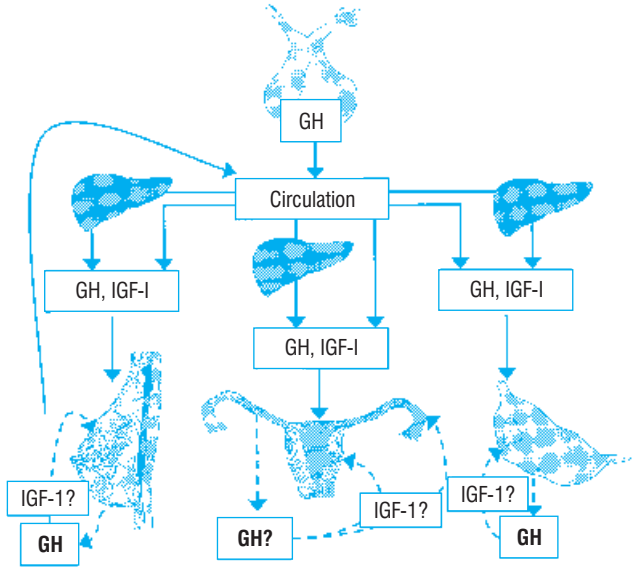
Growth Hormone

Physiology of GH in Infertility

Growth hormone is known to be involved in sexual differentiation and pubertal maturation, and it participates in gonadal steroidogenesis, gametogenesis, ovulation, pregnancy, and lactation. Moreover, pituitary GH and hepatic insulin-like growth factor-I (IGF-I) may be involved in the strategic maintenance of ovarian function, whereas ovarian GH may be involved in emergency modulation of ovarian function (Figure 3).[6]

Growth hormone is involved in the regulation of male and female infertility. Several clinical studies reveal that adjuvant GH treatment has a possible role in IVF, especially in poor ovarian responders (PORs).[16,17] Growth hormone enhances the effects on granulosa cells and stimulates IGF-1, which in turn stimulates follicular development, estrogen production, and oocyte maturation. Moreover, growth hormone–releasing hormone (GHRH) enhances gliotoxin-induced steroidogenesis cyclic adenosine monophosphate (cAMP) formation.[4,18,19] Although using GH in poor responders can significantly improve live birth rates, further research is necessary to fully define its role before recommending GH adjuvant in IVF treatment.[16,17]

Figure 3: Effects of GH in the female reproductive system.[6]



GH: Growth hormone; IGF: Insulin-like growth factor.

Adjuvant Therapy With GH: Beneficial Effects on the Probability of Clinical Pregnancy and Live Birth

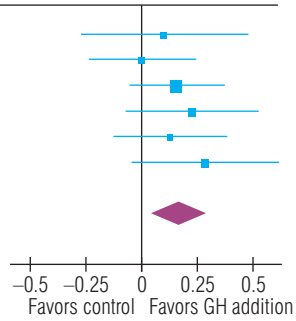
A systematic review and meta-analysis was conducted to evaluate the addition of GH to gonadotropins in ovarian stimulation of poor responders treated by IVF. The results suggested that GH addition significantly increased clinical pregnancy and live birth rates (Figure 4). Furthermore, GH addition was associated with a significantly higher proportion of

Figure 4: Risk difference for clinical pregnancy rate.[20]

Study or Subgroup	GH Events	GH Total	Control Events	Control Total	Weight	Risk difference M-H, fixed, 95% CI
Bergh *et al*., 1994	3	10	2	10	12.2%	0.10 (–0.28, 0.48)
Dor *et al*., 1995	0	7	0	7	8.5%	0.00 (–0.24, 0.24)
Kucuk *et al*., 2007	10	31	5	30	37.2%	0.16 (–0.06, 0.37)
Owen *et al*., 1991	4	13	1	12	15.2%	0.22 (–0.07, 0.52)
Suikkari *et al*., 1996	2	16	0	6	10.6%	0.13 (–0.13, 0.38)
Zhuang *et al*., 1994	5	12	2	15	16.3%	0.28 (–0.04, 0.61)
Total (95% CI)		**89**		**80**	**100.0%**	**0.16 (0.04, 0.28)**
Total events	24		10			

Heterogeneity: $Chi^2=2.71$, df=5 (p=0.75); $I^2=0\%$
Test for overall effect: Z=2.65 (p=0.008)

GH: Growth hormone; CI: Confidence interval.



patients reaching embryo transfer. The study, therefore, suggested that the GH addition increases the probability of clinical pregnancy and live birth in poor responders undergoing ovarian stimulation with GnRH analogs and gonadotropins for IVF.[20]

Co-treatment With GH has Benefit on Poor Responders

A Cochrane review was conducted to assess the effectiveness of adjuvant GH in IVF protocols. The review suggested a statistically significant difference with the use of GH adjuvant in IVF protocols when compared with IVF protocols in women with a poor prognosis (Figure 5).[21]

Results of a meta-analysis showed that GH addition significantly increased serum E2 level on the day of human chorionic gonadotropin (HCG) (odds ratio [OR]=0.55; 95% confidence interval [CI]=0.127–0.973) and Metaphase II (MII) oocyte number (OR=0.827; 95% CI=0.470–1.184). Furthermore, GH addition significantly improved the number of male and female pronuclei (2PN) (OR=0.934; 95% CI=0.206–1.661) and obtained embryos (OR=0.934; 95% CI=0.206–1.661). However, no significant difference was found for the overall implantation rate at 8.8% (95% CI=–0.062 to 0.237) and clinical pregnancy rate at 5.1% (95% CI=–0.033 to 0.134).[17]

Another randomized, double-blind, placebo-controlled trial was conducted to evaluate the efficacy and tolerability of GH-releasing factor (GRF) in 196 patients. Following downregulation with a gonadotropin-releasing hormone agonist (GnRHa), patients were randomized to receive GRF (500 μg twice daily; n=96) or placebo (n=100) in addition to follicle stimulating hormone (FSH); treatment was continued until HCG was given, or for a maximum of 14 days. GRF had no significant effect on the mean number of follicles with a diameter of ≥16 mm, the number of FSH ampoules required to achieve ovarian stimulation, or on secondary measures of ovarian response and treatment outcome. There were, however, significant increases in circulating GH and IGF-1 concentration (Figure 6). It is concluded that despite producing significant increases in GH and IGF-1, concomitant treatment with GRF does not improve the ovarian response to FSH in poorly responsive women undergoing IVF.[22]

Several investigators have reported that GH cotreatment may enhance the ovarian response to gonadotropins and reduce the total dose of gonadotropins required. However, this reduction in gonadotropin requirements is not economically beneficial. Moreover, there is still controversy concerning the ability of GH or GH-releasing hormone co-treatment to improve the ovarian response and increase the pregnancy rate. These inconclusive findings and the high cost of GH and GH-releasing hormone make clinicians reluctant to initiate a trial of GH or GH-releasing hormone co-treatment for low responders.[17–22]

L-Arginine

The importance of nitric oxide (NO) as an intra- and intercellular modulator has been recognized in many biological processes, including ovarian

Figure 5: Forest plot of comparison: Growth hormone vs. placebo (pregnancy rate per woman).[21]

Study or subgroup	GH Events	GH Total	Placebo Events	Placebo Total	Weight	Odd ratio M-H, fixed, 95% CI
Bergh 1994	3	9	2	9	11.8%	1.75 (0.22, 14.22)
Hazout 2003 4 IU	7	12	2	6	98%	2.80 (0.36, 21.73)
Hazout 2003 8 IU	3	12	1	6	8.8%	1.67 (0.13, 20.58)
Kueuk 2008	10	30	5	30	29.4%	2.50 (0.74, 8.50)
Owen 1991	4	13	1	12	6.4%	4.89 (0.46, 51.87)
Suikkari 1996 4 IU	2	10	0	3	5.0%	2.06 [0.08, 54.80)
Tesarik 2005	13	50	3	50	19.6%	5.50 (1.46, 20.76)
Zhuang 1994	5	12	2	15	92%	4.64 (0.71, 30.42)
Total (95% CI)		**148**		**131**	**100.0%**	**3.28 (1.74, 6.20)**
Total events	47		16			

Heterogeneity: $Chi^2=1.74$, df=7 (p=0.97), $I^2=0\%$
Test for overall effect Z=3.66 (p=0.0002)

GH: Growth hormone; CI: Confidence interval.

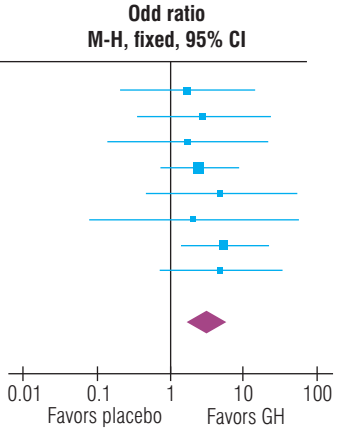
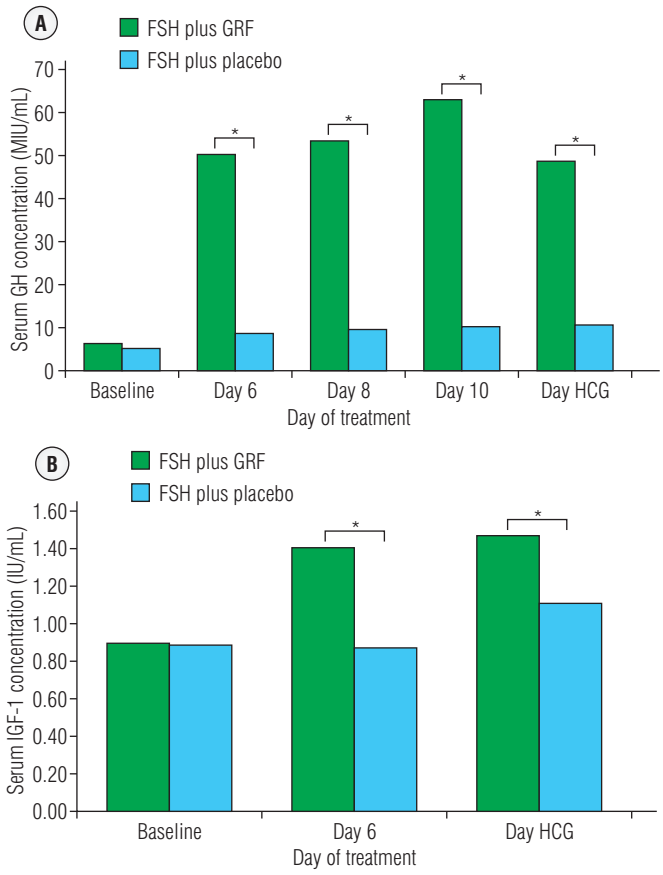


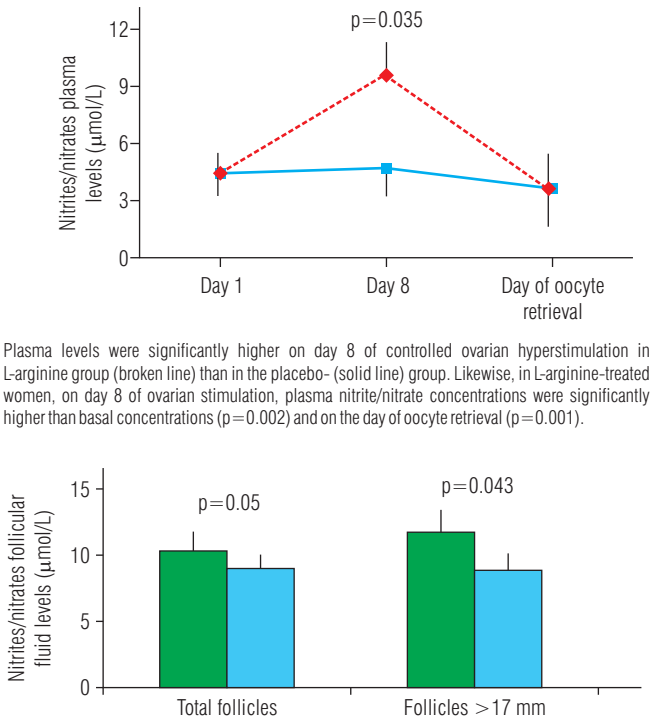
Figure 6: Efficacy and tolerability of GRF.[22]



*p<0.0001, growthhormone-releasing factor vs. placebo.

GH: Growth hormone; FSH: Follicle stimulating hormone; GRF: Growth hormone-releasing factor

Figure 7: Plasma and follicular fluid nitrite/nitrate concentrations.[19]



Plasma levels were significantly higher on day 8 of controlled ovarian hyperstimulation in L-arginine group (broken line) than in the placebo- (solid line) group. Likewise, in L-arginine-treated women, on day 8 of ovarian stimulation, plasma nitrite/nitrate concentrations were significantly higher than basal concentrations (p=0.002) and on the day of oocyte retrieval (p=0.001).

Follicular fluid nitrite/nitrate concentrations were significantly higher in the L-arginine group (green box) than in the placebo group (blue box).

physiology. Enhanced vascularization appears to be important for follicular selection and maturation in both spontaneous and stimulated IVF cycles. Although the precise role of NO has not been elucidated, it has been suggested that it is involved in follicular maturation and ovulation. Oral L-arginine supplementation during controlled ovarian hyperstimulation in poor responders decreases blood flow resistance in both perifollicular and uterine arteries. Hence, it was speculated that L-arginine, by modulating the permeability of follicular epithelium to plasma proteins and increasing uterine perfusion, might improve ovarian response, endometrial receptivity, and pregnancy rate.[23]

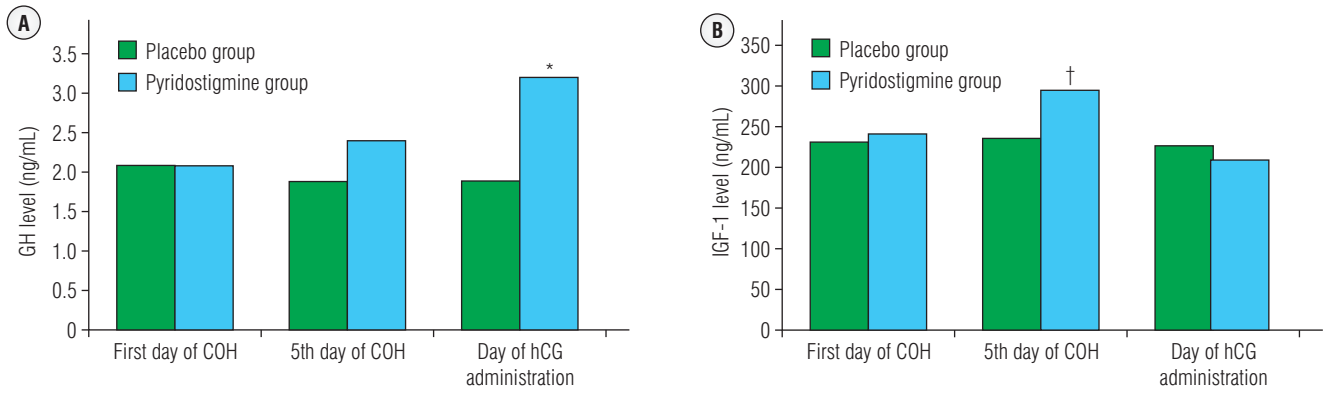
To evaluate the role of L-arginine supplementation in controlled ovarian hyperstimulation, 37 IVF patients were divided into two groups according to ovarian stimulation protocols: group I, GnRHa plus pure (p) FSH plus oral L-arginine (n=18); and group II, GnRHa plus pFSH plus placebo (n=19). In group I (n=16), plasma L-arginine concentrations increased from 87±12 µmol at baseline to 279±31 µmol (p=0.002) on the day of β-HCG administration. In this group, pFSH treatment was shorter (p=0.039) than in group II (n=16). The "good quality" embryos were fewer in number (p=0.034) and pregnancy rate, both per patient (p=0.024) and per embryo transfer (p=0.019), was lower in group I. In the L-arginine group, an increased follicular fluid concentration of nitrite/nitrate was observed. On day 8 of the cycle, elevated plasma estradiol levels were associated with decreased blood flow resistances of perifollicular arteries. Follicular fluid concentrations of nitrite/nitrate were inversely correlated with embryo quality (r=–0.613; p=0.005) and perifollicular artery pulsatility index (r=–0.609; p= 0.021) (Figure 7).[19]

Pyridostigmine

Pyridostigmine is an acetylcholinesterase inhibitor and, thus, may activate the cholinergic pathway, leading to inhibition of somatostatin release in the brain and increasing GH secretion. A randomized, double-blind, placebo-controlled study was conducted to investigate the effect of pyridostigmine, an acetylcholinesterase inhibitor, as co-treatment for controlled ovarian hyperstimulation (COH) in low responders. Sixty milligrams of pyridostigmine or placebo was administered to 20 infertile women with a history of low ovarian response to COH using a GnRHa orally twice daily from the first day of COH until the day of HCG injection in patients undergoing IVF-embryo transfer (ET) cycles.[20]

Pyridostigmine co-treatment was associated with significant decreases in the amount of gonadotropins and the duration of stimulation required. The clinical pregnancy rate was higher in the pyridostigmine group, but this difference was not statistically significant (25.7% vs. 11.4%). The serum GH level on the day of HCG injection was significantly higher in the pyridostigmine group than in the placebo group. Follicular fluid concentrations of GH and IGF-1 were significantly higher in the pyridostigmine group. The study suggested that pyridostigmine co-treatment for COH could affect the serum and intrafollicular GH and IGF-1 concentrations and, hence, improves the ovarian response to COH and the results of IVF in low responders undergoing IVF-ET (Figure 8).[20]

Figure 8: Comparison of serum concentrations of GH (A) and insulin-like growth factor-1 (IGF-1) (B) between the placebo and pyridostigmine groups.[20]



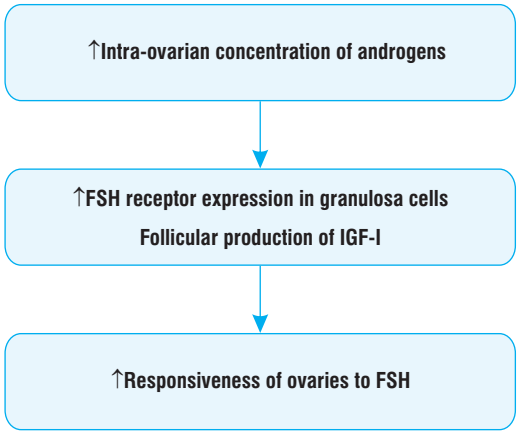
*p<0.05 (vs. the placebo group); †p<0.01 (vs. the placebo group).

GH: Growth hormone; COH: Controlled ovarian hyperstimulation protocol; hCG: Human chorionic gonadotropin.

Androgens or Androgen-Modulating Agents

The rationale and application of androgens or androgen-modulating agents such as testosterone or dehydroepiandrosterone (DHEA) have been explained in Figure 9.[21]

Figure 9: Rationale to the use of androgens or androgen-modulating agents for poor responders.[21]



It has been proposed that pre-IVF DHEA adjuvant therapy may improve ovarian response and pregnancy rates in women with diminished ovarian reserve. A meta-analysis was conducted to investigate the efficacy of DHEA as an adjuvant to improve ovarian response and IVF outcome in women with diminished ovarian reserve. The study suggested that there was no significant difference in the clinical pregnancy rate and miscarriage rates between women pre-treated with DHEA compared to those who were not pre-treated with DHEA. It is proposed that DHEA changes the follicular microenvironment by reducing hypoxic inducible factor-1, thus improving the quality of oocytes. Pooled data from two RCTs showed no significant difference in Clinic pregnancy rate (CPR) with DHEA supplementation. The number of oocytes retrieved was significantly lower in the DHEA group (Figure 10). In conclusion, based on the limited available evidence from a total of approximately 200 IVF cycles, there are insufficient data to support a beneficial role of DHEA as an adjuvant to COS in IVF cycle.[21]

Multiple studies have been conducted to evaluate the potential of androgens in ovulation induction; however, the extreme heterogeneity of treatment group and results makes it difficult to confirm its therapeutic efficacy. A Cochrane study suggests that:[22]

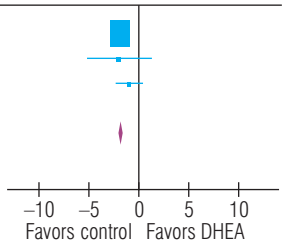
- Pretreatment with DHEA or testosterone may be associated with improved live birth rates
- The overall quality of evidence is moderate

Figure 10: Meta-analysis of numbers of oocytes.[21]

Study or subgroup	DHEA Mean	DHEA SD	DHEA Total	Control Mean	Control SD	Control Total	Weight	Mean difference IV, random, 95% CI	Year
Barad *et al*., 2007	3.9	0.4	89	5.8	0.5	101	96.8%	–1.90(–2.03, –1.77)	2007
Gleicher *et al*., 2010	9.6	6.2	22	11.7	6.3	44	0.4%	–2.10 (–5.29, 1.09)	2010
Wiser *et al*., 2010	2.8	1.7	17	3.8	1.9	16	2.8%	–1.00 (–2.23, 0.23)	2010
Total (95% CI)			**128**			**161**	**100.0%**	**–1.88 (–2.08, –1.67)**	

Heterogeneity: Tau^2=0.01; Chi^2=2.04, df: 2 (p=0.36); I^2=2%

Test for overall effect: Z=17.71 (p<0.00001)



SD: Standard Deviation; CI: Confidence interval; DHEA: Dehydroepiandrosterone; IV: *In vitro*.

- There is insufficient evidence to draw any conclusions about the safety of either androgen
- Definitive conclusions regarding the clinical role of either androgen await evidence from further well-designed studies
- Androgen supplementation prior to ovarian stimulation is not supported by the best available evidence, although it may be associated with slightly improved LBR.

Letrozole

Letrozole is the third-generation selective aromatase inhibitor most widely used in assisted reproduction. Aromatase inhibitors induce ovulation by inhibiting estrogen production; the consequent hypoestrogenic state increases GnRH release and pituitary FSH synthesis. In *in vitro* studies, letrozole showed the lowest IC50 and the greatest relative potency, which indicates a higher *in vitro* inhibitory effect on the enzyme aromatase. Aromatase is a microsomal member of the cytochrome P450 hemoprotein-containing enzyme complex superfamily (P450 arom, the product of the CYP19 gene) that synthesizes estrogens by catalyzing three consecutive hydroxylation reactions converting C19 androgens to aromatic C18 estrogenic steroids.[17] Aromatase converts androstenedione to estrone and testosterone to estradiol. Its activity can be demonstrated in several tissues, including the ovaries, brain, placenta, adipose tissue, muscle, liver, breast, and estrogen-dependent breast cancer. Aromatase is expressed in a tissue-specific manner. This enzyme is mainly expressed in the ovaries of premenopausal women. A very high level of aromatase is expressed in the placenta of pregnant women. In postmenopausal women, adipose tissue is the main source of estrogens.[23]

Side effects from letrozole are uncommon and related to suppression of the production of estrogens as a result of aromatase inhibition induced by the drug. Side effects include hot flashes (11%), nausea (7%), fatigue (5%), alopecia, and vaginal bleeding, which occur more frequently in breast cancer patients than in women treated for ovulation induction due to differences in the duration of treatment. Finally, administration of cimetidine had no effect on the pharmacokinetics of letrozole, and letrozole had no effect on the pharmacokinetics of warfarin. However, co-administration with tamoxifen leads to a significant decrease in letrozole plasma levels.[23]

A systematic search of the literature was performed for both prospective and retrospective studies. Meta-analyses of RCTs were performed for three comparisons: letrozole vs. clomiphene citrate (CC), letrozole 1 FSH vs. FSH in intrauterine insemination (IUI), and letrozole 1 FSH vs. FSH in IVF (Figure 11). The meta-analysis suggested the letrozole is as effective as other methods of ovulation induction. Further RCTs are warranted to define more clearly the efficacy and safety of letrozole in human reproduction.[23]

Conclusive data regarding the optimal doses of aromatase inhibitors (AIs) in reproductive medicine are lacking. In most studies, letrozole has been administered at once-daily doses of 2.5–5 mg for 5 days. Higher doses are associated with a persistent inhibition of aromatase and a very low estrogen level to ensure an adequate endometrial growth at the time of ovulation.[23]

Another study was conducted to compare the efficacy and cost-effectiveness of extended high-dose letrozole regimen/HPuFSH-gonadotropin releasing hormone antagonist (GnRHant) protocol with those of short low-dose letrozole regimen/HPuFSH-GnRHant protocol in 136 poor responders undergoing IVF-ET. There were no significant differences between both groups with regard to number of oocytes retrieved and clinical pregnancy rate (5.39+2.08 vs. 5.20+1.88 and 22.06% vs. 16.18%, respectively). The total gonadotropins dose and medications cost per cycle were significantly lower in the extended letrozole group (44.87+9.16 vs. 59.97+14.91 ampoules and USD 616.52+94.97 vs. USD 746.84+149.21, respectively). The cost-effectiveness ratio was USD 2794 in the extended letrozole group and USD 4616 in the short letrozole group. Thus, extended letrozole regimen/HPuFSH-GnRHant protocol was more cost-effective than short letrozole regimen/HPuFSH-GnRHant protocol in poor responders undergoing IVF-ET.[24]

Aspirin

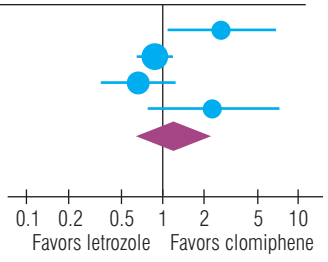
Aspirin, also called acetylsalicylic acid, has many clinical applications, including as an antipyretic, analgesic, antithrombotic, and micro-circulation improvizer. Its proposed mechanism in ovulation induction has been elaborated in Figure 12.[7,8]

A systematic review and meta-analysis was conducted to evaluate the different outcomes of low-dose aspirin on patients undergoing in IVF/ICSI, including clinical pregnancy rate, implantation rate, live birth rate, miscarriage rate, fertilization rate, and number of oocytes retrieved. Thirteen RCTs that included 3104 participants were selected. There were no significant differences in implantation rate, live birth rate, miscarriage rate, fertilization rate, and endometrial thickness. However, research showed that aspirin treatment may improve clinical pregnancy rates compared with placebo or no treatment and reduce the number of oocytes

Figure 11: Effect of letrozole on ovulation rate per cycle in PCOS.[23]

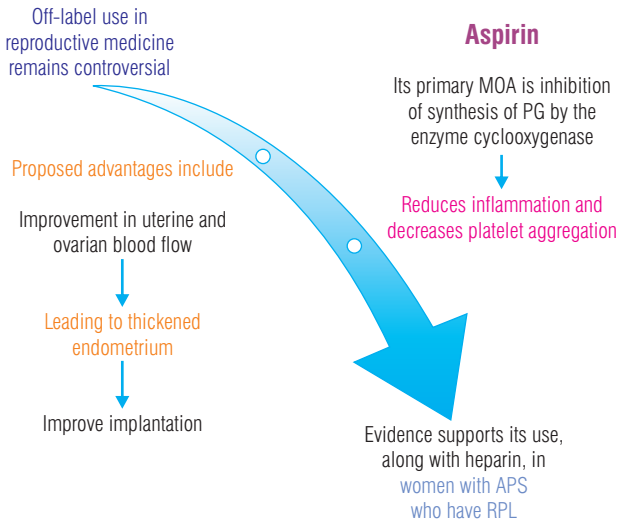
Study name	Odds ratio	Lower limit	Upper limit	Events/Total: Letrozole	Events/Total: Clomiphene
Atay, 2006	2.67	1.08	6.60	42/51	35/55
Badawy, 2007	0.85	0.66	1.11	365/540	371/523
Bayar, 2006	0.65	0.35	1.20	65/99	71/95
Sohrabvand, 2006	2.31	0.77	6.96	48/53	54/67
	1.17	0.66	2.09	520/743	531/740

Test for heterogeneity: $\chi^2=9.54$, df=3 (p=0.02), $I^2=68.6\%$. Test for overall effect: Z=0.53 (p=0.59).



PCOS: Polycystic ovarian syndrome; CI: Confidence interval.

Figure 12: Mechanism of action of aspirin in ovulation induction.[7,8]



APS: Antiphospholipid antibody syndrome; RPL: Recurrent pregnancy loss; MOA: Mechanism of action.

retrieved. They concluded that low-dose aspirin may improve the pregnancy rates in IVF/ICSI, with the recommended clinical use dose of 100 mg/day.[25]

Summary

- The use of adjuvant therapy may improve the outcome of IVF and may be particularly beneficial for women with a history of repeated IVF failure.
- A few drugs are approved for ovulation induction, while others are off-label drugs used as adjuvants.
- Off-label drugs have been evaluated in the phase I or phase II trials of clinical research, but they have not been fully assessed in phase III or phase IV trials.
- Growth hormone enhances the effects on granulosa cells and stimulates IGF-1, which in turn stimulates follicular development, estrogen production, and oocyte maturation. Moreover, GHRF enhances gliotoxin-induced steroidogenesis cyclic adenosine monophosphate (cAMP) formation.
- Inconclusive findings and the high cost of GH and GH-releasing hormone make clinicians reluctant to initiate a trial of GH or GH-releasing hormone co-treatment for low responders.
- Cochrane review 2009 supports the use of dexamethasone plus CC in CC-resistant women.
- L-arginine, by modulating the permeability of follicular epithelium to plasma proteins and increasing uterine perfusion, might improve ovarian response, endometrial receptivity, and pregnancy rate.
- Pyridostigmine is an acetylcholinesterase inhibitor and, thus, may activate the cholinergic pathway, leading to inhibition of somatostatin release in the brain and increasing GH secretion.
- It has been proposed that pre-IVF DHEA adjuvant therapy may improve ovarian response and pregnancy rates in women with diminished ovarian reserve.
- Letrozole is the third-generation selective aromatase inhibitor most widely used in assisted reproduction.
- Few studies support the use of DHEA to improve LBR in decreased ovarian reserve (DOR) undergoing IVF/ICSI treatment.
- Aromatase inhibitors induce ovulation by inhibiting estrogen production; the consequent hypoestrogenic state increases GnRH release and pituitary FSH synthesis.
- Glucocorticoids have been proposed as a useful adjuvant to both CC and gonadotropin ovulation induction in women with PCOS with a therapeutic rationale based on reducing ovarian androgen levels, improving ovulatory function, and reducing resistance to ovulation-induction agents.
- Androgen supplementation prior to ovarian stimulation is not supported by the best available evidence, although it may be associated with slightly improved LBR.

Adjuvant Therapy for Improving Endometrial Receptivity

Improving endometrial receptivity can be divided into two broad categories:

- Treatment of thin endometrium
- Endometrial stimulation
- Treatment of endometriosis to improve endometrial receptivity

Aspirin

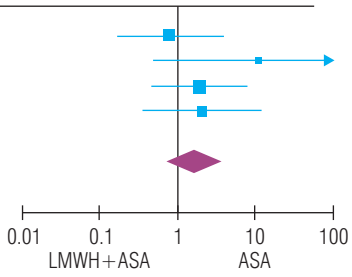
Aspirin has been commonly used in an attempt to increase the chance of live birth in women undergoing assisted reproductive technology (ART). However, there is contradictory evidence on the effectiveness of this treatment and on the appropriate time to commence treatment and its duration. Although physiologically, aspirin exerts a beneficial effect on some aspects required for a successful pregnancy, aspirin intake has also been associated with miscarriage and vaginal bleeding. It was, therefore, important to evaluate current evidence on the effectiveness of this treatment. While some studies could not demonstrate any benefit in IVF outcome, others reported an increase in pregnancy rate, sometimes even statistically significant. No less than four meta-analyses have been published on the subject thus far. Thus, despite these conventional meta-analyses conducted on the subject, there is still no conclusive answer to the question of whether aspirin is truly effective in IVF. In addition, a Cochrane review suggested that there is no evidence in favor of routine use of aspirin to improve pregnancy rates for a general IVF population.[26,27]

A study was conducted to assess whether the combination of low-molecular–weight heparin (LMWH) and aspirin (ASA) is better than ASA alone in women with hereditary thrombophilia. Four trials were

Figure 13: Odds ratio of live births.[28]

Study or subgroup	ASA Events	ASA Total	LMWH+ASA Events	LMWH+ASA Total	Weight	Odds ratio M-H, random, 95% CI
Kaandorp	11	17	9	13	30.8%	0.81 (0.17, 3.81)
Laskin	10	10	6	9	7.5%	11 .31 [0.50, 256.20)
Visser	13	19	8	15	37.3%	1.90 (0.47, 7.70)
Vries	67	69	66	70	24.4%	2.03 (0.36, 11 .46)
Total (95% CI)		**115**		**107**	**100.0%**	**1.70 (0.72, 4.00)**
Total events	101		89			

Heterogeneity: $Tau^2=0.00$; $Chi^2=2.38$, df=3 (p=0.50); $I^2=0\%$
Test for overall effect: Z=1.22 (p=0.22)



LMWH: Low-molecular–weight heparin; ASA: Aspirin; CI: Confidence interval.

included in the quantitative synthesis in a total of 222 randomized women. Effect of LMWH+ASA vs. ASA with regard to live births was evaluable in all four RCTs with a similar overall treatment effect for the therapies (OR: 1.7) and without heterogeneity. No significant differences or heterogeneity were observed between groups for secondary outcomes, namely first-trimester miscarriages (OR=0.69 [0.22–2.16]), prematurity (OR=0.99 [0.4–2.08]), pre-eclampsia (OR=1.49 [0.63–3.5]), and small-for-gestational–age babies (OR=2.08 [0.96–4.47]) (Figure 13).[28]

Heparin

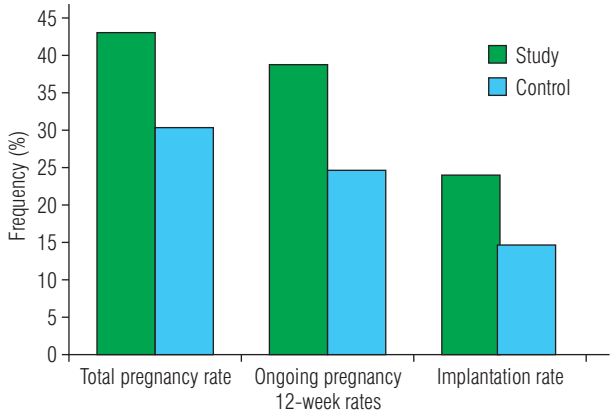
A study conducted to understand the role of LMWH in women with recurrent implantation failure after IVF treatment demonstrated an improvement in clinical pregnancy rates (34.7% [n=75] vs. 26.7% [n=75]), although the low number of patients precluded statistical significance in the results. The observed relative increase of approximately 30% in live-birth rate with LMWH was proposed by the investigators as a clinically significant trend, necessitating further research on the use of empirical LMWH in assisted reproduction. The study showed no statistically significant difference in the live-birth rate, clinical pregnancy rate, or implantation rate between the study and control groups.[29]

Corticosteroids

A prospective quasi-randomized, controlled trial evaluated the effect of combined oral prednisolone and LMW heparin in ICSI in women with previously unexplained, failed implantation. A total of 334 cycles (women with previously unexplained, failed one or two ICSI attempts) were assigned randomly to receive standard treatment or combined prednisolone (20 mg/day), starting on the first day of ovarian stimulation, and LMWH (1 mg/kg/day) starting 1 day after oocyte retrieval in addition to standard treatment. The results suggested that the mean age, number of previously failed IVF attempts, and basal FSH levels were comparable between the groups. The pregnancy and implantation rates were significantly different between the study and control groups (Figure 14). Thus, a combination of oral prednisolone and LMWH may have a significant effect on pregnancy and implantation rates in prior unexplained, failed implantation.[30]

Further, a meta-analysis was performed to evaluate the efficiency of prednisolone administration on unexplained recurrent miscarriage (RM) and the process of ART. The meta-analysis provides evidence that

Figure 14: Comparison of the clinical outcome between participants and controls.[30]



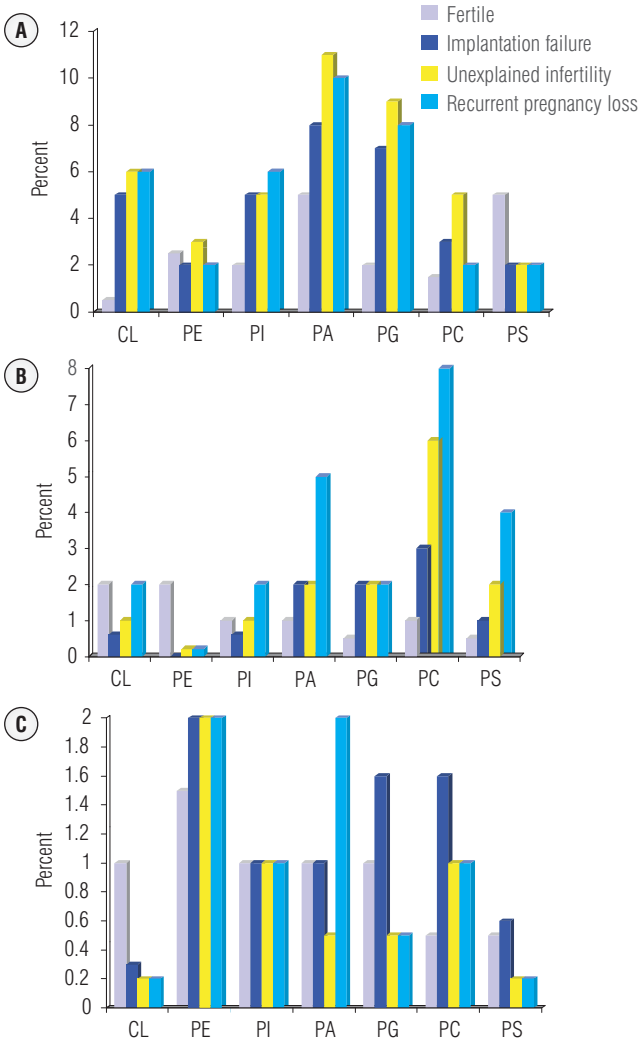
prednisolone therapy improves pregnancy outcomes in women with idiopathic RM (live birth rate: RR=1.58, 95% CI=1.23–2.02; successful pregnancy outcome: RR=7.63, 95% CI=3.71–15.69; miscarriage rate: RR=0.42, 95% CI=0.28–0.61). Moreover, the meta-analysis revealed a non-significant effect of prednisolone on pregnancy outcome during ICSI cycles (pregnancy rate: RR=1.02, 95% CI=0.84–1.24; clinical pregnancy rate: RR=1.01, 95% CI=0.82–1.24; implantation rate: RR=1.04, 95% CI=0.85–1.28). Thus, prednisolone administration may improve pregnancy outcomes in women with idiopathic RM; its efficacy in women undergoing ICSI is not significant.[31]

Immunotherapy

Considering the common occurrence of reproductive failure, the cause of reproductive failure is important to be determined. The most frequently studied risk factors to identify an immunologic cause of reproductive failure have included the presence of antiphospholipid antibodies (APA) and elevated natural killer (NK) cell killing. Elevated levels of NK cell cytotoxicity have been linked to increased rates of spontaneous abortions and IVF failure. Increased killing activity can be the result of elevated numbers of NK cells or increased cytotoxicity within each cell. Detection of elevation of circulating CD56+ (NK) cells and NK cell activity has been shown to be helpful in identifying individuals at risk for not implanting embryos and for losing karyotypically normal pregnancies.[32]

A study suggested that the prevalence of APA was the same among women with the diagnosis of unexplained infertility, recurrent implantation failure, and recurrent miscarriage. Heparin and aspirin are successful in the treatment of elevated APA among women with recurrent miscarriage but not with recurrent implantation failure. Intravenous immunoglobulin (IVIg) has been successful in the treatment of recurrent miscarriage and recurrent implantation failure among women with elevated APA and/or NK cell activity. When the pregnancy outcomes of women with a history of reproductive failure and elevated NK cell cytotoxicity treated with intralipid were compared with women treated with IVIg, no differences were seen. Immunotherapy for the treatment of reproductive failure enhances live birth but only in those women displaying abnormal immunologic risk factors (Figure 15).[32]

Figure 15: Percentage of abnormally elevated antiphospholipid antibody serum concentrations.[32]



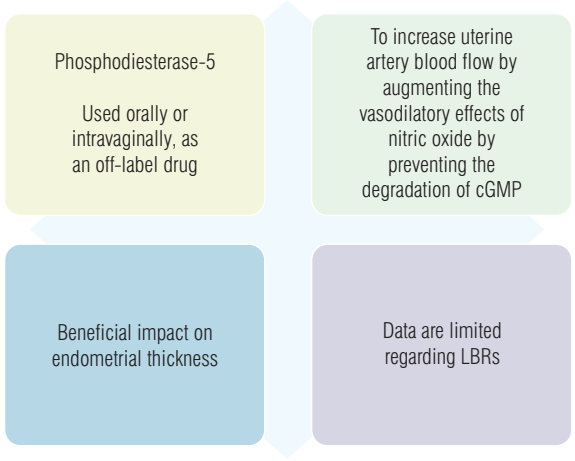
CL: Anticardiolipin antibodies; PE: Antiphospatidylethanolamine; PI: Antiphosphatidylinositol; PA: Antiphosphatidic acid; PG: Antiphosphatidylglycerol; PC: Antphopatidylcholine; PS: Antiphosphatidylserine

Further, another systematic review was conducted to evaluate the routine use of adjuvant therapies for women with elevated NK cells undergoing ARTs to improve live birth rate. The review suggested a beneficial effect of the interventions on clinical pregnancy rates with an RR of 1.63 for prednisolone and 3.41 for IVIg. Studies assessing the efficacy of IVIg have also reported live birth rate with an relative risk (RR) of 3.94 favoring the intervention. Data heterogeneity was substantial (I2=66%), suggesting a cautious interpretation of the results. The analysis suggested that some data show adjuvant therapies (mainly IVIg) to confer some benefit on ART outcome. However, overall, the review does not support the use of prednisolone, IVIg, or any other adjuvant treatment in women undergoing ART who are found to have elevated absolute numbers or activity of NK cells, purely due to the paucity of, or poor quality of, the evidence (Figure 16).[33]

Sildenafil

A few insights into the application of sildenafil have been described in Figure 17.[7,8]

Figure 17: Application of sildenafil.[7,8]

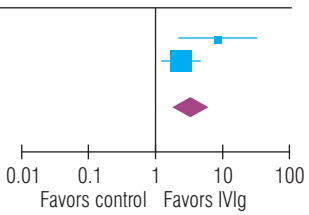


cGMP: Cyclic guanosine monophosphate; LBRs: Live birth rates.

Figure 16: Forest plots of clinical pregnancy rates where IVIg was administered.[33]

Study or subgroup	IVIg Events	IVIg Total	Control Events	Control Total	Weight	Risk ratio M-H, fixed, 95% CI
Moraru *et al*., 2012	17	20	2	20	17.0%	8.50 (2.25, 32.06)
Winger *et al*., 2011b	38	62	7	27	83.0%	2.36 (1.21, 4.61)
Total (95% CI)		**82**		**47**	**100.0%**	**3.41 (1.90, 6.11)**
Total events	55		9			

Heterogeneity: Chi^2=2.97, df=1 (p=0.08); I^2=66%
Test for overall effect: Z=4.12 (p<0.0001)



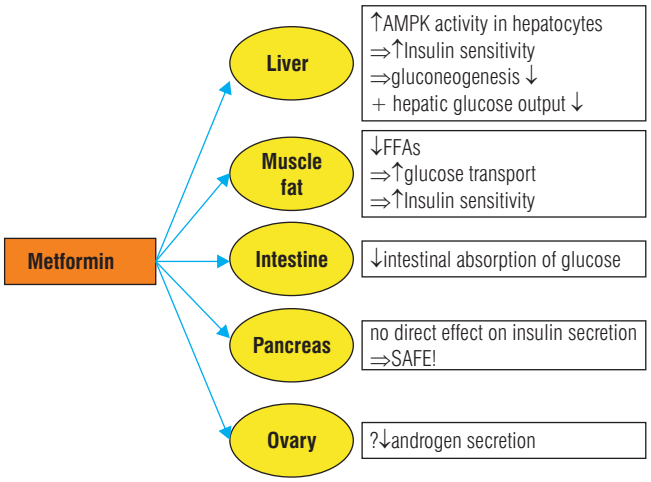
IVIg: Intravenous immunoglobulin; CI: Confidence interval.

Metformin as Adjuvant Medical Therapy in the Treatment of Anovular Infertility in PCOS Patients

The application of metformin has been elaborated in Figure 18. Metformin has been used alone to try and promote ovulation and conception in women with PCOS, although the effects appear limited. However, the drug has also been examined as a co-treatment in IVF in PCOS women.[34,35]

The potential advantages and disadvantages of metformin have been given in Figure 19.

Figure 18: Mechanism of action of metformin.[34,35]



AMPK: AMP-activated protein kinase; FFAs: Plasma free fatty acids.

Figure 19: Potential advantages and disadvantages of metformin.

Potential advantages	Potential disadvantages
• ↑ glucose tolerance • ↑ insulin sensitivity • ↓ blood lipid levels • ↑ weight loss or stabilization • Improved fat distribution • ↓ blood pressure • ↓ androgen levels • Restoration of regular menses • Stimulates folliculogenesis • Postponement of diabetes	• Gastrointestinal disturbance in one-third of patients • Generalized feeling of unwellness • Decreased absorption of vitamin B12 • Lactic acid buildup

Benefits of Metformin Supplementation in Infertility Treatment: Is the Evidence Conflicting?

A double-blind, multicenter, randomized, placebo-controlled study was conducted to investigate whether pretreatment with metformin before and during IVF increases the LBR compared with placebo in women with sonographic evidence of polycystic ovary syndrome (PCO) but without any clinical manifestations of PCOS. The study population included 134 women undergoing IVF treatment at three tertiary referral IVF units. In total, 134 women were randomized to receive either metformin or placebo: 69 to metformin and 65 to placebo. There were no statistically significant differences between the two groups in baseline characteristics. With regard to IVF outcome, no significant improvements were found in the metformin group when compared with the placebo group (Figure 20). In particular, there was no difference between the groups in rates of live birth, clinical pregnancy, or severe OHSS. The study suggested no benefit in metformin co-treatment before and during IVF in women with PCO without any other features of PCOS.[36]

Figure 20: *In vitro* fertilization cycle outcomes in the two groups.[36]

Outcome	Metformin (n=69)	Placebo (n=65)	p-value
LBR/cycle started n (%)	27 (39.1%)	30 (46.2%)	0.411[a]
CPR/cycle started n (%)	29 (42.0%)	33 (50.8%)	0.311[a]
PR/cycle started n (%)	36 (52.2%)	37 (56.9%)	0.581[a]
No. of patients—severe OHSS n (%)	6 (8.7%)	5 (7.7%)	0.833[a]
Cancellation/FAE n (%)	2 (2.9%)	3 (4.6%)	0.485[a]
No. of patients coasted n (%)	6 (8.7%)	11 (16.9%)	0.161[a]
Severe OHSS or avoidance technique n (%)	13 (18.8%)	12 (18.5%)	1.00[a]
Total dose of FSH (IU/L)	1650 (684)[b]	1650 (625)[b]	0.232[c]
No. of days of stimulation	10.9 (2.5)	11.0 (2.8)	0.930
Peak E_2 level (pmol/L)	4353 (4164)[b]	3367 (4292)[b]	0.798[c]
No. of oocytes	15.7 (7.5)	14.6 (9.3)	0.56
No. of oocytes fertilized	9.6 (5.2)	7.6 (4.8)	0.024
No. of embryos transferred	2.0 (0)[b]	2.0 (0)[b]	0.06[c]
No. of embryos frozen	4.0 (7.0)[b]	2.0 (5.0)[b]	0.102[c]
Implantation rate (%)	30.6%	38.9%	0.188[a]
No. of patients withdrawn	6 (8.7%)	3 (4.6%)	0.494[a]

Results are means (SD) unless indicated.

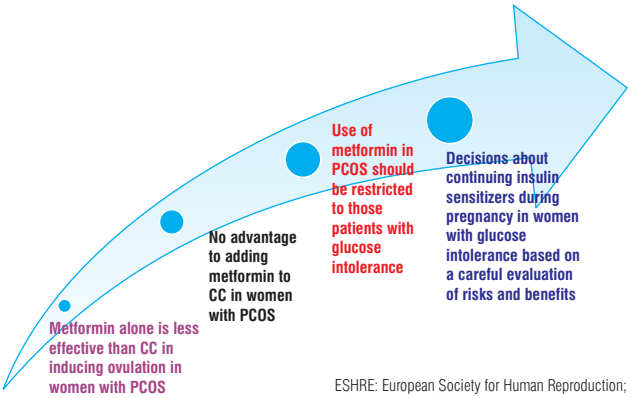
SD: Standard deviation; LBR: Live birth rate; CPR: Clinic pregnancy rate; PR: Pregnancy rates; OHSS: Ovarian hyperstimulation syndrome; FSH: Follicle stimulating hormone.

[a]Chi-square test/Fisher's exact test. [b]Median (interquartile range). [c]Mann–Whitney U-test.

Another randomized, placebo-controlled, double-blind study was conducted to explore the effect of metformin in women with PCOS undergoing IVF. Patients with PCOS undergoing IVF/ICSI treatment using a long GnRHa protocol were randomized to receive metformin, 850 mg or placebo tablets twice daily from the start of the downregulation process until the day of oocyte collection. There was no difference in the total dose of rFSH required per cycle. The median number of oocytes retrieved per cycle and the overall fertilization rates did not differ. However, both the clinical pregnancy rates beyond 12 weeks gestation per cycle and per embryo transfer were significantly higher in those treated with metformin (Figure 21). Thus, short-term co-treatment with metformin for patients with PCOS undergoing IVF/ICSI cycles does not improve the response to stimulation but significantly improves the pregnancy outcome and reduces the risk of OHSS.[37]

Figure 21: Metformin proved to be ineffective in large-scale RCT—second ESHRE/ASRM Consensus Meeting at Greece, March 2007[38]

	Clomiphene	Metformin	Combination
Number	209	208	209
Ovulation rate	49%	29%	60%
Conception rate	30%	12%	38%
Live birth rate	23%	7%	27%



ESHRE: European Society for Human Reproduction; ASRM: American Society of Reproductive Medicine.

Recommendations of Adjuvant Therapy for Infertility Treatment

British Fertility Society Policy and Practice Committee

The vital need to optimize uterine conditions for a successful ovarian response requires guidelines to provide the role and application of adjuvants in patients. The evidence base for the routine use of different adjuvants, alone or in combination, for women undergoing their first IVF treatment cycle and for those with poor prognosis is inadequate. However, the British Fertility Society Policy and Practice Committee has provided fertility professionals with evidence-based guidance and recommendations regarding the use of immunotherapy, vasodilators, uterine relaxants, aspirin, heparin, growth hormone, dehydroepiandrosterone, estrogen, and metformin as adjuvants in IVF. The summary of recommendations for good clinical practice is listed below.

1. There is no convincing evidence for the use and safety of intravenous immunoglobulin as adjuvants in women with recurrent implantation failure embarking on IVF.
2. There is a lack of evidence to indicate the effectiveness and safety of using anti-TNF-α agents as adjuvants in IVF cycles.
3. There is a lack of evidence to recommend intralipid infusion therapy as an adjuvant in IVF cycles.
4. There is a lack of robust evidence to support the routine use of corticosteroids as adjuvants in IVF cycles. There is limited evidence that corticosteroids may improve pregnancy rates in women undergoing conventional IVF and in the subgroup of women with autoimmunity or unexplained implantation failure.
5. There is a lack of evidence that NTG and sildenafil have significant beneficial effects on IVF outcome, and their routine use as adjuvants in IVF cycles is not recommended.
6. There is a lack of robust evidence to support the use of uterine relaxants (NTG, selective β_2-adrenergic blockers, and progesterone) as adjuvants in IVF cycles.
7. There is a lack of proven efficacy for routine use of aspirin as an adjuvant in IVF cycles.
8. There is a lack of robust evidence to warrant routine use of LMWH in the wide population of women undergoing IVF treatment, but its administration should be carefully considered in women with thrombophilia.
9. The available evidence does not recommend routine use of GH as an adjuvant in IVF cycles.
10. There is a lack of evidence to support the use of DHEA as an adjuvant in IVF cycles.
11. There is a lack of evidence to recommend estrogen as an adjuvant for follicular priming and/or for endometrial development in the luteal phase of fresh IVF cycles.
12. The available evidence suggests that metformin may have some beneficial effects in women with PCOS undergoing IVF by reducing the risk of developing OHSS and increasing clinical pregnancy rates.

Summary

- Aspirin has been commonly used in an attempt to increase the chance of live birth in women undergoing ART. However, there is contradictory evidence on the effectiveness of this treatment and on the appropriate time to commence treatment and its duration.
- A combination of oral prednisolone and low-molecular-weight heparin may have a significant effect on pregnancy and implantation rates in prior unexplained, failed implantation.
- Elevated levels of NK cell cytotoxicity have been linked to increased rates of spontaneous abortions and to IVF failure. Increased killing activity can be the result of elevated numbers of NK cells or increased cytotoxicity within each cell.
- Metformin has been used alone to try and promote ovulation and conception in women with PCOS, though the effects appear limited.
- Metformin proved to be ineffective in a large-scale RCT—second ESHRE/ASRM consensus meeting at Greece on March 2007.

References

1. Segev Y, Carp H, Auslender R, *et al*. Is there aplace for adjuvant therapy in IVF? *Obstet Gynecol Surv*. 2010;65(4):260–272.
2. Holzer H, Casper R, Tulandi T. A new era in ovulation induction. *Fertil Steril*. 2006;85(2):277–284.
3. Jeve Y, Bhandari H. Effective treatment protocol for poor ovarian response: A systematic review and meta-analysis. *J Hum Reprod Sci*. 2016;9(2):70.
4. Usadi R, Merriam K. On-label and off-label drug use in the treatment of female infertility. *Fertil Steril*. 2015;103(3):583–594.
5. Vitek W, Alur S, Hoeger K. Off-label drug use in the treatment of polycystic ovary syndrome. *Fertil Steril*. 2015;103(3):605–611.
6. Rashida Begum M, Ehsan M, Shahina Begum M, *et al*. Beneficial effects of addition of glucocorticoid during induction of ovulation by letrozole in polycystic ovarian syndrome. *J SAFOG DVD*. 2012;4:85–89.
7. Macklon N, Fauser B. Medical approaches to ovarian stimulation for infertility. *Yen Jaffe Reprod Endocrinol*. 2009:689–724.
8. Parsanezhad M, Alborzi S, Motazedian S, *et al*. Use of dexamethasone and clomiphene citrate in the treatment of clomiphene citrate-resistant patients with polycystic ovary syndrome and normal dehydroepiandrosterone sulfate levels: A prospective, double-blind, placebo-controlled trial. *Fertil Steril*. 2002;78(5):1001–1004.
9. Kalampokas T, Pandian Z, Keay S, *et al*. Glucocorticoid supplementation during ovarian stimulation for IVF or ICSI. *Cochr Database Syst Rev*. 2017.
10. Costello MF. The current role of metformin in the treatment of infertility in PCOS: A mini-review. *Ann Clin Exp Metabol*. 2017;2(2):1018.
11. Regidor PA, Schindler AE. Myoinositol as a safe and alternative approach in the treatment of infertile PCOS women: A German observational study. *Int J Endocrinol*. 2016;Article ID 9537632.
12. Habib H, Naseer S. Efficacy of N acetylcysteine as an adjunct to clomipene citrate in induction of ovulation in anovulatory infertility. *IJSRP*. 2016;6(11):2250–3153.
13. Basheer M, Rai S. Melatonin vs. phytomelatonin: Therapeutic uses with special reference to polycystic ovarian syndrome (PCOS). *Cogent Biology*. 2016;2:1136257.
14. Irani M, Merhi Z. Role of vitamin D in ovarian physiology and its implication in reproduction: a systemic review. *Fertility and Sterility*. 2014;102(2):460–468.e3.
15. Deshpande H. Practical management of ovulation induction. *JP Medical Ltd*. 2016.
16. Hull K. Growth hormone: Roles in female reproduction. *J Endocrinol*. 2001;168(1):1–23.
17. Yu X, Ruan J, He L. Efficacy of growth hormone supplementation with gonadotrophins *in vitro* fertilization for poor ovarian responders: An updated meta-analysis. *Int J Clin Exp Med*. 2015;8(4):4954–4967.
18. Lanzone A, Di Simone N, Castellani R, *et al*. Human growth hormone enhances progesterone production by human luteal cells *in vitro*: Evidence of a synergistic effect with human chorionic gonadotropin. *Fertil Steril*. 1992;57(1):92–96.
19. Doldi N, Bassan M, Bonzi V, *et al*. Effects of growth hormone and growth hormone-releasing hormone on steroid synthesis in cultured human luteinizing granulosa cells. *Gynecol Endocrinol*. 1996;10(2):101–108.
20. Kolibianakis E, Venetis C, Diedrich K, *et al*. Addition of growth hormone to gonadotrophins in ovarian stimulation of poor responders treated by *in-vitro* fertilization: A systematic review and meta-analysis. *Hum Reprod Update*. 2009;15(6):613–622.
21. Duffy J, Ahmad G, Mohiyiddeen L, *et al*. Growth hormone for *in vitro* fertilization. *Cochr Database Sys Rev*. 2010.
22. Howles C, Loumaye E, Germond M, *et al*. Does growth hormone-releasing factor assist follicular development in poor responder patients undergoing ovarian stimulation for in-vitro fertilization?. *Human Reproduction*. 1999;14(8):1939–1943.
23. Battaglia C, Regnani G, Marsella T, *et al*. Adjuvant L-arginine treatment in controlled ovarian hyperstimulation: A double-blind, randomized study. *Human Reproduction*. 2002;17(3):659–665.
24. Kim C, Chae H, Chang Y. Pyridostigmine cotreatment for controlled ovarian hyperstimulation in low responders undergoing in vitro fertilization–embryo transfer. *Fertil Steril*. 1999;71(4):652–657.
25. Wang L, Huang X, Li X, *et al*. Efficacy evaluation of low-dose aspirin in IVF/ICSI patients evidence from 13 RCTs. *Medicine (Baltimore)*. 2017;96(37):e7720.
26. Siristatidis C, Basios G, Pergialiotis V, *et al*. Aspirin for *in vitro* fertilisation. *Coch Database Syst Rev*. 2016.
27. Groeneveld E, Broeze K, Lambers M, *et al*. Is aspirin effective in women undergoing *in vitro* fertilization (IVF)? Results from an individual patient data meta-analysis (IPD MA). *Hum Reprod Update*. 2011;17(4):501–509.
28. Areia A, Fonseca E, Areia M, *et al*. Low-molecular-weight heparin plus aspirin versus aspirin alone in pregnant women with hereditary thrombophilia to improve live birth rate: Meta-analysis of randomized controlled trials. *Arch Gynecol Obstet*. 2015;293(1):81–86.
29. Seshadri S, Sunkara S. Low-molecular-weight-heparin in recurrent implantation failure. *Fertil Steril*. 2011;95(7):e29.
30. Fawzy M, El-Refaeey A. Does combined prednisolone and low molecular weight heparin have a role in unexplained implantation failure? *Arch Gynecol Obstet*. 2013;289(3):677–680.
31. Dan S, Wei W, Yichao S, *et al*. Effect of prednisolone administration on patients with unexplained recurrent miscarriage and in routine intracytoplasmic sperm injection: A meta-analysis. *Am J Reprod Immunol*. 2015;74(1):89–97.
32. Coulam C, Acacio B. Does immunotherapy for treatment of reproductive failure enhance live births? *Am J Reprod Immunol*. 2012;67(4):296–304.
33. Polanski L, Barbosa M, Martins W, *et al*. Interventions to improve reproductive outcomes in women with elevated natural killer cells undergoing assisted reproduction techniques: A systematic review of literature. *Human Reproduction*. 2013;29(1):65–75.
34. Zhou G, Myers R, Li Y, *et al*. Role of AMP-activated protein kinase in mechanism of metformin action. *J Clin Investigation*. 2001;108(8):1167–1174.
35. Natali A, Ferrannini E. Effects of metformin and thiazolidinediones on suppression of hepatic glucose production and stimulation of glucose uptake in type 2 diabetes: A systematic review. *Diabetologia*. 2006;49(3):434–441.
36. Swanton A, Lighten A, Granne I, *et al*. Do women with ovaries of polycystic morphology without any other features of PCOS benefit from short-term metformin co-treatment during IVF? A double-blind, placebo-controlled, randomized trial. *Human Reproduction*. 2011;26(8):2178–2184.
37. Tang T, Glanville J, Orsi N, *et al*. The use of metformin for women with PCOS undergoing IVF treatment. *Human Reproduction*. 2006;21(6):1416–1425.
38. Consensus on infertility treatment related to polycystic ovary syndrome. *Fertil Steril*. 2008;89(3):505–522.

Notes

8344909 L21804E25