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在线全文

不同年龄段人群三种促排卵方案胚胎发育及临床结局比较*

范咏琪， 张文香， 章志国△
安徽医科大学第一附属医院 妇产科生殖中心（合肥 230032）

【摘要】 **目的** 比较不同年龄段人群使用促性腺激素释放激素（gonadotropin releasing hormone, GnRH）拮抗剂方案、GnRH激动剂长方案与早卵泡期长方案进行体外受精（IVF）过程中的胚胎发育及临床结局情况。**方法** 回顾性纳入2021年1月–2023年2月期间收治的患者。① 在总人群中，对三种促排方案〔拮抗剂方案组（4 173例），激动剂长方案组（2 410例）与早卵泡长方案组（341例）〕患者的基本情况、胚胎发育情况及临床结局进行比较；② 将总人群划分为三个年龄段〔<30岁（2 576例），30～35岁（3 249例），>35岁（1 099例）〕，并在此基础上，进一步对三种促排方案进行比较。分别比较<30岁、30～35岁、>35岁人群中使用三种促排方案的胚胎发育情况及临床结局。**结果** 在总人群中，激动剂长方案组的获卵数高于拮抗剂方案组〔（13.85±7.162）个vs.（13.36±7.862）个，P=0.022 4〕与早卵泡长方案组〔（13.85±7.162）个vs.（11.86±6.802）个，P<0.000 1〕；拮抗剂方案组的促性腺激素（gonadotropin, Gn）启动量、Gn使用天数均低于其余两组（P<0.05）；拮抗剂方案组的囊胚形成率高于激动剂长方案组（64.91% vs. 62.35%，P<0.000 1），同时也高于早卵泡长方案组（64.91% vs. 61.18%，P=0.000 1），而三种促排方案临床妊娠率、活产率差异无统计学意义（P>0.05）；②在<30岁人群中，拮抗剂方案组的囊胚形成率高于激动剂长方案组（66.12% vs. 63.33%，P<0.000 1）与早卵泡长方案组（66.12% vs. 62.13%，P=0.009 4）；在30～35岁人群中，拮抗剂方案组的囊胚形成率高于激动剂长方案组（64.88% vs. 62.93%，P=0.000 9）与早卵泡长方案组（64.88% vs. 60.39%，P=0.001 1）；在>35岁人群中，拮抗剂方案组的囊胚形成率高于激动剂长方案组（59.83% vs. 56.51%，P=0.009 3），而与早卵泡长方案组差异无统计学意义（P>0.05）。在三个年龄段人群中，三种促排方案的临床妊娠率、活产率、胎儿体质量及评分差异均无统计学意义（P>0.05）。**结论** 拮抗剂方案能减少促排时间、促排卵Gn剂量，增强患者就医依从性。高龄患者使用拮抗剂方案能提高囊胚形成率，但三种方案的活产率并没有明显差异。

【关键词】 促性腺激素释放激素拮抗剂 促性腺激素释放激素激动剂 胚胎发育

Comparative Study of the Embryo Development and Clinical Outcomes of 3 Ovarian Stimulation Protocols in Different Age Groups

FAN Yongqi, ZHANG Wenxiang, ZHANG Zhiguo△. *Reproductive Medicine Center, Department of Obstetrics and Gynecology, The First Affiliated Hospital of Anhui Medical University*, Hefei 230032, China
△ Corresponding author, E-mail: Zzg_100@163.com

【**Abstract**】 **Objective** The main purpose of this study is to compare the embryo development and clinical outcomes of women in different age groups undergoing *in vitro* fertilization (IVF) processes using gonadotrophin-releasing hormone (GnRH) antagonist protocol, GnRH agonist long protocol, and early follicular phase protocol. We aim to provide reliable reference for future clinical treatments. **Methods** We conducted a detailed analysis of patients who underwent treatment between January 2021 and February 2023. 1) In the overall patient population, we comprehensively compared the basic characteristics, the embryo development, and the clinical outcomes of patients treated with three different ovarian stimulation protocols, including the GnRH antagonist protocol group (n=4 173), the agonist long protocol group (n=2 410), and the early follicular phase long protocol group (n=341). 2) We divided the overall population into three age groups, one group for patients under 30 years old (n=2 576), one for patients aged 30-35 (n=3 249), and one for patients older than 35 years old (n=1 099). Then, we compared the three stimulation protocols based on the group division. We separately compared the embryo development and clinical outcomes of patients using the three stimulation protocols in the under 30 years old, the 30-35 years old, and the over 35 years old age groups. With this analysis, we aimed to explore the response of different age groups to different stimulation protocols and their impact on the success rate of IVF. **Results** 1) In the overall population, we found that the average number of oocytes retrieved in the GnRH agonist long protocol group was significantly higher than that in the GnRH antagonist protocol group ([13.85±7.162] vs. [13.36±7.862], P=0.022 4), as well as the early follicular phase long protocol group ([13.85±7.162] vs. [11.86±6.802], P<0.000 1). Patients in the GnRH antagonist protocol group not only had a significantly lower starting dose of gonadotrophin (Gn) compared to the other two groups (P<0.05) but also had a significantly lower number of days of Gn use (P<0.05). The blastocyst formation rate in the GnRH antagonist protocol group was the highest among the three

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△ 通信作者，E-mail: Zzg_100@163.com
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groups, significantly higher compared to the GnRH agonist long protocol group (64.91% vs. 62.35%, *P*<0.000 1) and the early follicular phase long protocol group (64.91% vs. 61.18%, *P*=0.000 1). However, there were no significant differences in the clinical pregnancy rates or the live birth rates among the three groups treated with different ovarian stimulation protocols (*P*>0.05). 2) In the <30 age group, the blastocyst formation rate in the GnRH antagonist protocol group was the highest among the three groups, significantly higher compared to the GnRH agonist long protocol group (66.12% vs. 63.33%, *P*<0.000 1) and the early follicular phase long protocol group (66.12% vs. 62.13%, *P*=0.009 4). In the 30-35 age group, the blastocyst formation rate in the GnRH antagonist protocol group was the highest among the three groups, significantly higher compared to the GnRH agonist long protocol group (64.88% vs. 62.93%, *P*=0.000 9) and the early follicular phase long protocol group (64.88% vs. 60.39%, *P*=0.001 1). In the >35 age group, the blastocyst formation rate in the GnRH antagonist protocol group was significantly higher than that in the GnRH agonist long protocol group (59.83% vs. 56.51%, *P*=0.009 3), while there was no significant difference compared to that of the early follicular phase long protocol group (*P*>0.05). In the three age groups, we found that there were no significant differences in clinical pregnancy rate, live birth rate, and neonatal outcome indicators (fetal weight and Apgar score) among the three stimulation protocols (antagonist protocol, GnRH agonist long protocol, and early follicular phase long protocol) (*P*>0.05). The findings showed no significant differences between clinical and neonatal outcomes in patients of all ages, regardless of the ovarian stimulation protocol, suggesting that the three ovarian stimulation protocols have similar therapeutic effects in patients of different ages. The results of this study have important implications for the selection of an appropriate ovarian stimulation protocol and the prediction of treatment outcomes. **Conclusion**　In the younger than 30 and 30-35 age groups, the GnRH antagonist protocol showed a more significant advantage over the GnRH agonist long protocol and the early follicular phase long protocol. This suggests that for younger and middle-aged patients, the antagonist protocol may lead to better outcomes during ovarian stimulation. In the older than 35 age group, while the antagonist protocol still outperformed the GnRH agonist long protocol, there was no significant difference compared to the early follicular phase long protocol. This may imply that with increasing age, the early follicular phase long protocol may have effects similar to the antagonist protocol to some extent. The advantages of the antagonist protocol lie in its ability to reduce stimulation duration and the dosage of GnRH, while enhancing patient compliance with treatment. This means that patients may find it easier to accept and adhere to this treatment protocol, thereby improving treatment success rates. Particularly for older patients, the use of the antagonist protocol may significantly increase the blastocyst formation rate, which is crucial for improving the success rates. Although there were no significant differences in the clinical outcomes of patients treated with the three protocols in each age group, further research is still needed to validate these findings. Future multicenter studies and increased sample sizes may help comprehensively assess the efficacy of different stimulation protocols. Additionally, prospective studies are needed to further validate these findings and determine the optimal treatment strategies.

【Key words】　Gonadotropin releasing hormone antagonist　Gonadotropin releasing hormone agonist　Development of embryos

辅助生殖技术(assisted reproductive technology, ART)是治疗不孕症的重要手段[1]。控制性超排卵技术(controlled ovarian hyperstimulation, COH)是辅助生殖技术中关键步骤之一。促性腺激素释放激素拮抗剂(gonadotrophin releasing hormone antagonist, GnRH-ant)通过竞争性拮抗GnRH受体,短期内可抑制内源性促性腺激素(gonadotrophin, Gn)的释放,具有促排时间短,促排卵用药少,且降低卵巢过度刺激综合征风险的优点,目前临床上使用越来越多[2]。促性腺激素释放激素激动剂(gonadotrophin releasing hormone agonist, GnRH-a)作为最早的促排方案,起源于1984年[3]。GnRH-a 的大量持续作用,会耗竭垂体的GnRH受体,从而抑制早发黄体生成素(luteinizing hormone, LH) 峰的发生。早卵泡期长方案通常在卵泡发育早期给予GnRH-a降调节,28 ~ 30 d后检测性激素和B超,达到降调节标准后,使用促性腺激素促排卵。本研究通过将总人群划分三个年龄段,探究不同年龄段人群使用三种促排方案的胚胎发育与临床结局,旨在为临床上决定各年龄段人群促排方案作出一定参考。

1　资料与方法

1.1　对象资料

本研究为回顾性研究,可豁免知情同意。研究方案由安徽医科大学伦理委员会批准(批准号 20200114)。选取2021年1月–2023年2月于我院生殖中心行(*in vitro* fertilization, IVF)助孕的患者临床资料,共计6 924例,其中 GnRH拮抗剂 4 173例 , GnRH激动剂长方案 2 410例,

早卵泡期长方案341例。纳入标准:①夫妻双方有体外受精IVF指征;②女方年龄20～43岁;③不孕因素为排卵障碍、盆腔或输卵管因素、子宫内膜异位症、多囊卵巢综合征、卵巢功能减退。排除标准:①促排卵方案为除GnRH拮抗剂方案、GnRH激动剂长方案或早卵泡期长方案以外的其他方案;②患者某项信息不全;③患者体质量指数(body mass index, BMI)> 30 kg/m^2;④严重输卵管积液、子宫腺肌病、胚胎植入前遗传学检测(preimplantation genetic testing, PGT)周期患者。纳入30岁以下患者共2 576例,其中拮抗剂组1 610 例,长方案组850例,早卵泡期长方案组116 例;纳入30～35岁患者共3 249例,其中拮抗剂组1 881例,长方案组1 193例,早卵泡期长方案组175例。纳入35岁以上患者共1 099例,其中拮抗剂组682例,长方案组367例,早卵泡期长方案组50例。见图1 。

1.2 治疗方案

1.2.1 GnRH 拮抗剂方案

在月经周期的第2或3天开始使用Gn启动,药物为果纳芬(默克雪兰诺,瑞士)、普丽康(默沙东,美国)、丽申宝(珠海丽珠医药贸易有限公司,中国)等。于用药后第6天或主导卵泡达到14 mm直径时,结合激素水平开始使用GnRH拮抗剂思则凯(默克雪兰诺,瑞士)等直至人绒毛膜促性腺激素(human chorionic gonadotrophin, HCG)注射日,促排期间根据B超及检测性激素结果,判断卵巢对药物的反应并依此调整促排卵药物的使用剂量。

1.2.2 GnRH激动剂长方案

在前次月经周期的第18～22天(黄体中期)使用长效GnRH激动剂达菲林(益普生,法国)等使垂体降调节;后使用促性腺激素(果纳芬、普丽康或丽申宝等)启动,促排期间根据B超及检测性激素结果,判断卵巢对药物的反应并依此调整促排卵药物的使用剂量。

1.2.3 早卵泡期长方案

自月经期第2～3天,应用长效GnRH激动剂达菲林等,第28～30天检测性激素和B超,达到降调节标准后,同GnRH-a长方案。

1.2.4 取卵、体外受精-胚胎移植

B超见2个或2个以上直径> 18 mm的卵泡时立即停止促排卵药物,并于当晚21: 00–22: 00患者接受10 000 IU HCG(珠海丽珠医药贸易有限公司,中国)肌内注射;HCG注射36 h后行经阴道超声监测下取卵。对于接受IVF的患者,在取卵后4 h进行人工授精。精卵共孵育维持约4 h,去除卵丘细胞,将卵母细胞转移到卵裂培养基中,在离体培养第3天检测卵裂期胚的发育情况。7～9个卵裂球且破碎率小于20%的胚胎为优质胚胎。将胚胎转移到囊胚培养基中继续孵育2～3 d,在第5天和第6天使用Gardner囊胚分级法对囊胚进行评估[4]。优质囊胚定义为第5天评分为3BB及以上,第6天评分为4BB及以上的囊胚[5]。行鲜胚移植术患者经超声引导,将囊胚植入子宫,生化妊娠定义为胚胎移植后2周血清β- HCG水平≥25 IU/L。临床妊娠定义为胚胎移植后35 d超声扫描发现妊娠囊。

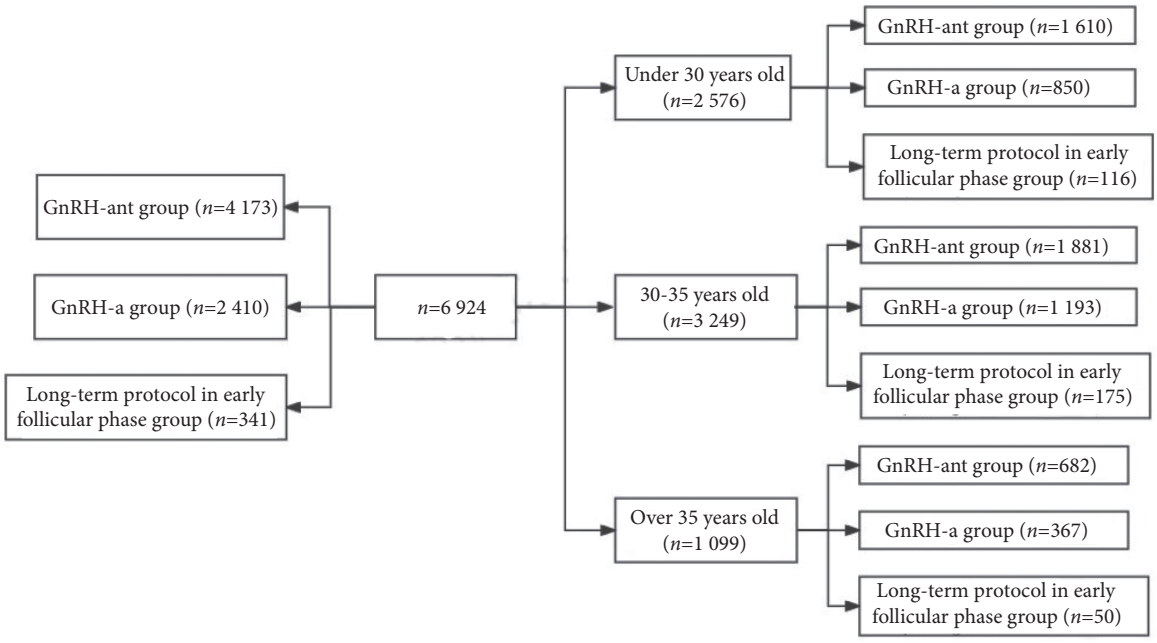

图 1 研究设计图

Fig 1 Study design

1.3 统计学方法

采用GraphPad Prism 8.0和SPSS 23.0软件进行统计分析。计量资料以$\bar{x}\pm s$表示，计数资料以百分数表示。计量资料采用单因素方差分析，计数资料采用χ^2检验。$P<0.05$为差异有统计学意义。

2 结果

2.1 三种促排方案患者一般情况比较

三种不同促排方案组患者女方年龄、不孕年限，基础雌二醇（estradiol, E2）、抗苗勒氏管激素（anti-Müllerian hormone, AMH）差异均无统计学意义（$P>0.05$）；拮抗剂方案组的BMI、基础LH水平显著高于其余两组（$P<0.05$）；拮抗剂方案组的窦卵泡数（antral faollicle counting, AFC）高于激动剂长方案组〔（15.23±7.268）个vs.（14.45±5.754）个，$P=0.001\,1$〕，而激动剂长方案组的AFC高于早卵泡期长方案组〔（14.45±5.754）个 vs.（12.96±6.686）个，$P=0.012\,2$〕；拮抗剂方案组的促性腺激素（gonadotropin, Gn）启动量、Gn使用天数均低于其余两组（$P<0.05$）；早卵泡期长方案组的基础卵泡刺激素（follicle-stimulating hormone, FSH）水平低于拮抗剂方案〔（6.777±2.327）IU/L vs.（7.183±2.854）IU/L, $P=0.013\,0$〕与激动剂长方案组〔（6.777±2.327）IU/L vs.（7.199±2.460）IU/L, $P=0.013\,7$〕。见表1。

2.2 三种不同促排方案患者胚胎发育及临床结局

在总人群中，激动剂长方案组的获卵数高于拮抗剂组〔（13.85±7.162）个vs.（13.36±7.862）个，$P=0.022\,4$〕和早卵泡期长方案组〔（13.85±7.162）个vs.（11.86±6.802）个，$P<0.000\,1$〕；早卵泡期长方案组的受精率低于拮抗剂方案组（77.02% vs. 78.61%，$P=0.016\,8$）与激动剂长方案组（77.02% vs. 79.65%，$P<0.000\,1$）；拮抗剂方案组的优质胚胎率高于激动剂长方案组（52.31% vs. 49.75%, $P<0.000\,1$）与早卵泡期长方案组（52.31% vs. 47.68%, $P<0.000\,1$）；拮抗剂方案组的囊胚形成率高于激动剂长方案组（64.91% vs. 62.35%, $P<0.000\,1$），同时也高于早卵泡期长方案组（64.91% vs. 61.18%, $P=0.000\,1$）；拮抗剂方案组的优质囊胚率高于激动剂长方案组（53.58% vs. 50.81%, $P<0.000\,1$）与早卵泡期长方案组（53.58% vs. 48.57%, $P<0.000\,1$）；三种促排方案的临床妊娠率和活产率差异并无统计学意义（$P>0.05$）。见表2。

2.3 30岁以下人群中三种促排方案胚胎发育及临床结局

在30岁以下人群中，拮抗剂方案组的优质胚胎率高于激动剂长方案组（53.84% vs. 50.85%, $P<0.000\,1$）和早卵泡期长方案组（53.84% vs. 48.99%, $P=0.002\,6$）；拮抗剂方案组的囊胚形成率高于激动剂长方案组（66.12% vs. 63.33%, $P<0.000\,1$），同时也高于早卵泡期长方案组（66.12% vs. 62.13%, $P=0.009\,4$）；拮抗剂方案组的优质囊胚率高于激动剂长方案组（55.28% vs. 52.44%, $P<0.000\,1$）与早卵泡期长方案组（55.28% vs. 50.05%, $P=0.001$）；三种促排方案临床妊娠率和活产率差异无统计学意义（$P>0.05$）。见表3。

2.4 30～35岁人群三种不同促排方案胚胎发育及临床结局

在30～35岁人群中，激动剂长方案组的获卵数高于拮抗剂方案组〔（14.18±7.146）个 vs.（12.68±7.535）个，$P<0.000\,1$〕和早卵泡期长方案组〔（14.18±7.146）个vs.（11.96±6.821）

表 1　总人群中三种促排方案患者一般情况

Table 1　General characteristics of patients in the three groups of ovarian stimulation protocols in the total population

Index	GnRH-ant group (A)	GnRH-a group (B)	Long-term protocol in early follicular phase group (C)	*P* (A vs. B)	*P* (A vs. C)	*P* (B vs. C)
Total cycle/*n*	4 173	2 410	341	/	/	/
Female age/yr.	31.310±4.711	31.390±4.166	31.330±4.039	0.716 9	0.995 6	0.960 1
Duration of infertility/year	3.087±2.273	3.219±2.403	3.046±2.382	0.089 4	0.944 5	0.401 5
BMI/(kg/m^2)	22.730±2.965	22.190±2.850	22.040±2.778	<0.000 1	<0.000 1	0.705 6
Basic FSH/(IU/L)	7.183±2.854	7.199±2.460	6.777±2.327	0.969 1	0.013 0	0.013 7
Basic LH/(IU/L)	5.652±4.188	4.930±3.048	4.750±3.051	<0.000 1	<0.000 1	0.669 8
Basic E2/(pmol/L)	133.60±69.49	132.90±63.75	130.20±64.17	0.921 9	0.662 0	0.792 5
AMH/(ng/mL)	3.520±2.765	3.457±2.513	3.003±1.742	0.978 9	0.673 0	0.760 1
AFC/number	15.23±7.268	14.45±5.754	12.96±6.686	0.001 1	<0.000 1	0.012 2
Gn doses/IU	2 020±862	2 587±1 001	2 774±1 029	<0.000 1	<0.000 1	0.000 8
Gn times/d	9.426±1.870	11.780±1.736	11.560±2.229	<0.000 1	<0.000 1	0.092 0

BMI: body mass index; FSH: follicle-stimulating hormone; LH: luteinizing hormone; E2: estradiol; AMH: anti-Müllerian hormone; AFC: antral faollicle counting; Gn: gonadotropin.

表 2 总人群中三种不同促排方案患者胚胎发育及临床结局

Table 2 Embryo development and clinical outcomes of patients with different ovarian stimulation protocols in the total population

Index	GnRH-ant group (A)	GnRH-a group (B)	Long-term protocol in early follicular phase group (C)	*P* (A vs. B)	*P* (A vs. C)	*P* (B vs. C)
Total cycle/*n*	4 173	2 410	341	/	/	/
Retrieved oocytes/number	13.360±7.862	13.850±7.162	11.860±6.802	0.022 4	0.000 1	<0.000 1
Fertilization rate	78.61% (42 564/54 148)	79.65% (26 287/3 303)	77.02% (3 197/4 151)	0.000 2	0.016 8	<0.000 1
Cleavage rate	97.74% (41 602/42 564)	97.92% (25 739/26 287)	97.84% (3 128/3 197)	0.133 7	0.757 4	0.793 4
High-quality embryo rate	52.31% (18 465/35 296)	49.75% (10 972/22 053)	47.68% (1 282/2 689)	<0.000 1	<0.000 1	0.043 1
Blastocyst formation rate	64.91% (22 910/35 296)	62.35% (13 749/22 053)	61.18% (1 645/2 689)	<0.000 1	0.000 1	0.237 4
High-quality blastocyst rate	53.58% (18 911/35 296)	50.81% (11 205/22 053)	48.57% (1 306/2 689)	<0.000 1	<0.000 1	0.028 8
Fresh transferred cycles	259	132	28	/	/	/
Clinical pregnancy rate	60.09% (417/694)	60.40% (273/452)	64.89% (61/94)	0.249 7	0.150 6	0.485 4
Live birth rate	51.44 % (357/694)	51.99% (235/452)	57.45% (54/94)	0.856 3	0.322 2	0.364 6
Abortion rate	13.67% (57/417)	13.55% (37/273)	11.48 % (7/61)	0.497 4	0.443 6	0.834 5
Neonatal body mass/g	3 183±615.7	3 205±477.9	3 149±690.7	0.871 1	0.894 9	0.754 1
Apgar score	9.991±0.100 0	9.995±0.070 0	9.963±0.272 2	0.905 2	0.287 8	0.176 0

表 3 <30岁人群中三种促排方案胚胎发育及临床结局

Table 3 Embryo development and clinical outcomes of three ovarian stimulation protocols in women under 30 years old

Index	GnRH-ant group (A)	GnRH-a group (B)	Long-term protocol in early follicular phase group (C)	*P* (A vs. B)	*P* (A vs. C)	*P* (B vs. C)
Total cycle/*n*	1 610	850	116	/	/	/
Female age/yr.	26.720±2.036	26.880±1.951	26.950±2.004	0.162 1	0.478 7	0.943 0
BMI/(kg/m^2)	22.280±2.952	21.980±2.983	21.820±2.823	0.053 6	0.235 6	0.842 9
Basic FSH/(IU/L)	6.839±2.196	7.053±2.024	6.646±2.263	0.090 3	0.666 0	0.185 8
Basic E2/(pmol/L)	143.90±73.10	137.40±65.23	143.00±63.78	0.166 5	0.993 2	0.778 9
Retrieved oocytes/number	15.260±8.370	15.430±7.275	13.840±7.006	0.880 9	0.156 0	0.114 8
Fertilization rate	78.37% (19 247/24 559)	79.18% (9 668/12 210)	77.20% (1 229/1 592)	0.074 6	0.272 3	0.072 2
Cleavage rate	97.63% (18 850/19 247)	97.76% (9 476/9 668)	98.10% (1 209/1 229)	0.691 4	0.347 9	0.443 5
High-quality embryo rate	53.84% (8 584/15 944)	50.85% (4 051/7 966)	48.99% (511/1 043)	<0.000 1	0.002 6	0.263 0
Blastocyst formation rate	66.12% (10 542/15 944)	63.33% (5 045/7 966)	62.13% (648/1 043)	<0.000 1	0.009 4	0.452 7
High-quality blastocyst rate	55.28% (8 814/15 944)	52.44% (4 177/7 966)	50.05% (522/1 043)	<0.000 1	0.001 0	0.147 2
Fresh transferred cycles	259	132	28	/	/	/
Clinical pregnancy rate	67.57% (175/259)	65.91% (87/132)	71.43% (20/28)	0.734 7	0.831 9	0.662 2
Live birth rate	58.69% (152/259)	59.85% (79/132)	53.57% (15/28)	0.913 4	0.687 8	0.673 1
Abortion rate	8.57% (15/175)	8.05% (7/87)	10.00% (2/20)	>0.999 9	0.687 9	0.674 1
Neonatal body mass/g	3 273±613.1	3 198±466.2	3 199±560.3	0.632 0	0.861 5	>0.999 9
Apgar score	9.910±0.287 6	9.927±0.260 9	9.792±0.414 9	0.610 6	0.980 1	0.204 4

BMI: body mass index; FSH: follicle-stimulating hormone; E2: estradiol.

个, *P*= 0.000 6〕; 早卵泡期长方案组的受精率低于拮抗剂方案组(77.08% vs. 78.97% , *P*= 0.044 1)和激动剂长方案组(77.08% vs. 79.98%, *P*= 0.002 2); 拮抗剂方案组的优质胚胎率高于激动剂长方案组(52.55% vs. 50.66%, *P*= 0.002 1)与早卵泡期长方案组 (52.55% vs. 47.36%, *P*= 0.000 3); 拮抗剂方案组的囊胚形成率高于激动剂长方案组(64.88% vs. 62.93%, *P*= 0.000 9)与早卵泡期长方案组(64.88% vs. 60.39%, *P*= 0.001 1); 拮抗剂方案组的优质囊胚率高于激动剂长方案组(53.70% vs. 51.43%, *P*= 0.000 2)与早卵泡期长方案组(53.70% vs. 47.88%, *P*< 0.000 1) ; 三种促排方案临床妊娠率和活产率差异无统计学意义 (*P*> 0.05)。见表4。

2.5 35岁以上人群三种不同促排方案胚胎发育及临床结局

见表5。在35岁以上人群中, 激动剂长方案组的获卵数高于拮抗剂方案组〔(10.63±6.015)个vs. (8.433±6.335)个, *P*< 0.000 1〕; 拮抗剂方案组的优质胚胎率高于激动剂

表 4　30 ~ 35岁人群三种不同促排方案胚胎发育及临床结局

Table 4　Embryo development and clinical outcomes in three groups of patients aged 30-35 years with different ovarian stimulation protocols

Index	GnRH-ant group (A)	GnRH-a group (B)	Long-term protocol in early follicular phase group (C)	*P* (A vs. B)	*P* (A vs. C)	*P* (B vs. C)
Total cycle/*n*	1 881	1 193	175	/	/	/
Female age/yr.	32.060±1.619	32.150±1.593	32.140±1.677	0.225 3	0.809 6	0.994 5
BMI/(kg/m^2)	22.070±2.407	22.080±2.730	21.720±2.530	0.998 7	0.171 2	0.190 2
Basic FSH/(IU/L)	7.250±2.715	7.197±2.142	6.826±2.168	0.782 7	0.116 1	0.199 0
Basic E2/(pmol/L)	137.70±70.99	136.10±66.76	134.40±70.71	0.771 5	0.806 7	0.946 7
Retrieved oocytes/number	12.68±7.535	14.18±7.146	11.96±6.821	<0.000 1	0.428 3	0.000 6
Fertilization rate	78.97% (18 830/23 846)	79.98 (13 519/16 904)	77.08% (1 604/2 081)	0.013 4	0.044 1	0.002 2
Cleavage rate	97.61% (18 380/18 830)	97.87% (13 231/13 519)	97.44% (1 563/1 604)	0.131 0	0.670 8	0.276 7
High-quality embryo rate	52.55% (8 225/15 653)	50.66% (5 868/11 583)	47.36% (636/1 343)	0.002 1	0.000 3	0.022 8
Blastocyst formation rate	64.88% (10 155/15 653)	62.93% (7 289/11 583)	60.39% (811/1 343)	0.000 9	0.001 1	0.069 2
High -quality blastocyst rate	53.70% (8 406/15 653)	51.43% (5 957/11 583)	47.88% (643/1 343)	0.000 2	<0.000 1	0.014 2
Fresh transferred cycles	316	241	47	/	/	/
Clinical pregnancy rate	58.86% (186/316)	61.83% (149/241)	65.96% (31/47)	0.486 2	0.426 1	0.625 4
Live birth rate	47.78% (151/316)	49.38% (119/241)	57.45% (27/47)	0.732 6	0.273 6	0.341 4
Abortion rate	15.59% (29/186)	14.09% (21/149)	12.90% (4/31)	0.758 9	>0.999 9	>0.999 9
Neonatal body mass/g	3 110±607.4	3 247±543.5	3 241±543.5	0.139 0	0.510 3	0.998 7
Apgar score	9.866±0.341 7	9.875±0.331 5	9.906±0.296 1	0.955 3	0.798 0	0.874 3

BMI: body mass index; FSH: follicle-stimulating hormone; E2: estradiol.

表 5　>35岁人群三种不同促排方案胚胎发育及临床结局

Table 5　Embryo development and clinical outcomes in three groups of patients over 35 years old with different ovarian stimulation protocols

Index	GnRH-ant group (A)	GnRH-a group (B)	Long-term protocol in early follicular phase group (C)	*P* (A vs. B)	*P* (A vs. C)	*P* (B vs. C)
Total cycle/*n*	682	367	50	/	/	/
Female age/yr.	38.140±1.799	37.940±2.058	38.330±2.331	0.174 9	0.767 4	0.367 6
BMI/(kg/m^2)	22.940±2.410	22.810±2.723	22.820±2.566	0.529 9	0.921 0	0.998 9
Basic FSH/(IU/L)	7.590±2.167	7.383±2.097	7.178±3.050	0.165 8	0.309 8	0.763 0
Basic E2/(pmol/L)	145.90±71.44	152.30±71.62	151.50±73.92	0.265 1	0.849 4	0.997 0
Retrieved oocytes/number	8.433±6.335	10.630±6.015	9.775±6.694	<0.000 1	0.325 4	0.630 2
Fertilization rate	78.12% (4 487/5 743)	79.71% (3 100/3 889)	76.15% (364/478)	0.063 8	0.328 7	0.072 7
Cleavage rate	97.44% (4 372/4 487)	97.81% (3 032/3 100)	97.80% (356/364)	0.322 9	0.861 9	>0.999 9
High-quality embryo rate	44.77% (1 656/3 699)	42.05% (1 053/2 504)	44.55% (135/303)	0.034 6	0.952 2	0.423 7
Blastocyst formation rate	59.83% (2 213/3 699)	56.51% (1 415/2 504)	61.39% (186/303)	0.009 3	0.625 9	0.110 3
High-quality blastocyst rate	45.72% (1 691/3 699)	42.77% (1 071/2 504)	46.53% (141/303)	0.023 5	0.810 5	0.219 7
Fresh transferred cycles	119	79	19	/	/	/
Clinical pregnancy rate	47.06% (56/119)	46.84% (37/79)	52.63% (10/19)	>0.999 9	0.805 4	0.799 0
Live birth rate	36.13% (43/119)	34.18% (27/79)	47.37% (9/19)	0.879 5	0.445 3	0.300 9
Abortion rate	23.21% (13/56)	24.32% (9/37)	10.0% (1/10)	>0.999 9	0.675 4	0.664 5
Neonatal body mass/g	3 110.0±607.4	3 247.0±543.5	3 241.0±713.2	0.139 0	0.510 3	0.998 7
Apgar score	9.855±0.404 5	9.929±0.262 3	9.727±0.467 1	0.674 4	0.563 4	0.293 9

BMI: body mass index; FSH: follicle-stimulating hormone; E2: estradiol.

长方案组(44.77% vs. 42.05%, P= 0.034 6); 拮抗剂方案组的囊胚形成率高于激动剂长方案组(59.83% vs. 56.51%, P= 0.009 3), 而与早卵泡期长方案组差异无统计学意义(P> 0.05); 拮抗剂方案组的优质囊胚率高于激动剂长方

案组(45.72% vs. 42.77%, P= 0.023 5);三种促排方案临床妊娠率和活产率差异无统计学意义(P> 0.05)。

3 讨论

近年来,辅助生殖技术成为具有生育需求而无法自然受孕的患者的首要选择。GnRH长方案通过大量GnRH-a作用于垂体后,在短期内释放大量Gn,产生"Flare up"效应,耗竭GnRH受体,有效抑制可能过早的LH峰值的出现,同时具有良好的卵泡发育同步性,是经典的促排方案之一。拮抗剂方案的主要优点在耗时短,数小时就能结合受体,产生Gn的抑制作用,大大降低治疗时间[6]。

DIETRICH等[7]发现,接受GnRH拮抗剂的患者卵泡液E2浓度显著低于接受GnRH-a的患者,而卵泡液中雌激素与雄激素的比例可能反映了颗粒细胞将雄激素转化为雌激素的能力。此外,雄激素增强卵巢颗粒细胞凋亡,而雌二醇发挥抑制作用。使用GnRH拮抗剂时,LH 循环浓度被抑制,导致雄激素产生受阻,从而降低滤泡内雄激素浓度[8]。雌、雄激素比率与卵泡健康和成熟度之间存在相关性,提示拮抗剂方案可能更有利于卵泡的发育[9]。本研究也发现拮抗剂方案组优质胚胎率、囊胚形成率、优质囊胚率显著高于其余两组。

CORREA-De-ARAUJO等[10]发现在使用辅助生殖技术(ART)时,随着女性年龄增加临床妊娠率逐渐下降。而本研究中也发现,无论采取哪种促排方案,随着年龄的增加,患者的获卵数、临床妊娠率都在逐渐下降。本研究中,每个年龄段人群,使用三种方案的临床妊娠率与活产率并无统计学意义。XU等[11]的研究表明,GnRH-ant治疗患者的子宫内膜中子宫自然杀伤细胞数量和穿孔素表达水平均增加,表明GnRH-ant可能增加着床期促炎因子的释放。由于孕体被认为是母体免疫系统的半同种异体移植物,从免疫学的角度来看,母体免疫耐受对于胚胎植入至关重要。RAGA等[12]发现,GnRH-ant可以改变子宫内膜免疫调节细胞因子的表达,这可能会影响胚胎植入。有研究表明[13],Gn 剂量的增加,会延长垂体功能恢复时间。而长时间低性腺激素水平状态下,有利于卵巢残余窦卵泡的清除、抑制甚至清除炎性内膜的增生,使 Gn 刺激日卵泡的募集更加同步,从而使子宫动脉血流参数(RI、PI、S/D)减小,使子宫内膜灌注良好,子宫内膜容积增大,进一步使内膜容受性更佳,从而为成功妊娠提供有利条件。ALPAÑÉS等[14]发现早卵泡期长方案可显著改良女性不孕患者子宫内膜容受性,卵泡期单剂量GnRH-a注射可诱导子宫内膜表达整合素,改善子宫内膜容受性,有利于胚胎着床。有研究者[15]发现,早卵泡期长方案临床妊娠率显著高于拮抗剂方案(58.57% vs. 37.14%, P< 0.05),提示早卵泡期长方案对不孕症患者子宫内膜容受性及临床妊娠具有更优的影响,在本研究中,早卵泡长方案组的临床妊娠率有上升趋势,但可能由于纳入早卵泡期长方案的周期过少,导致差异不明显。

就胚胎的发育方面而言,拮抗剂方案在< 30岁、30 ~ 35岁人群中较长方案与早卵泡期长方案优势更加显著;而> 35岁人群,拮抗剂方案优于长方案,与早卵泡期长方案无显著差异。在每个年龄段人群中使用三种方案的临床结局并无显著差异。拮抗剂方案能减少促排时间、促排卵Gn剂量,增强患者就医依从性,具有一定优势。高龄患者使用拮抗剂方案能提高囊胚形成率。

由于本研究为回顾性研究,存在一定的局限性,每个临床医生在进行促排方案与促排药物选择时,具有主观性。本研究中并未对患者的卵巢储备功能进行分层处理,未来还需要更大样本、多中心的实验性研究来验证。

* * *

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参 考 文 献

[1] VANDER BORGHT M, WYNS C. Fertility and infertility: definition and epidemiology. Clin Biochem, 2018, 62: 2–10. doi: 10.1016/j.clinbiochem.2018.03.012.

[2] HUIRNE J A, HOMBURG R, LAMBALK C B. Are GnRH antagonists comparable to agonists for use in IVF? Hum Reprod, 2007, 22: 2805–2813. doi: 10.1093/humrep/dem270.

[3] CHEN C, ZENG X, ZHANG H, *et al*. Comparison between the modified long gonadotropin-releasing hormone agonist protocol and the non-downregulation protocol in POSEIDON groups: a propensity score matching retrospective cohort study. Front Endocrinol (Lausanne), 2023,

14: 1189357. doi: 10.3389/fendo.2023.1189357.

[4] ZHAO H, LIU H, LI M, *et al*. Clinical outcomes following frozen-thawed blastocyst transfers with blastocysts derived from different cell numbers on day 3: a retrospective cohort study. J Assist Reprod Genet, 2020, 37: 641–648. doi: 10.1007/s10815-019-01664-x.

[5] ZHANG G L, SUN T Y, LI S, *et al*. The pregnancy outcomes of day-5 poor-quality and day-6 high-quality blastocysts in single blastocyst transfer cycles. Clin Exp Reprod Med, 2023, 50: 63–68. doi: 10.5653/cerm.2022.05540.

[6] HOSSEINI M A, ALEYASIN A, SAEEDI H, *et al*. Comparison of gonadotropin-releasing hormone agonists and antagonists in assisted reproduction cycles of polycystic ovarian syndrome patients. J Obstet Gynaecol Res, 2010, 36: 605–610. doi: 10.1111/j.1447-0756.2010.01247.x.

[7] DIETRICH J E, FREIS A, BEEDGEN F, *et al*. Intraindividual embryo morphokinetics are not affected by a switch of the ovarian stimulation protocol between GnRH agonist vs. antagonist regimens in consecutive cycles. Front Endocrinol (Lausanne), 2020, 11: 246. doi: 10.3389/fendo.2020.00246.

[8] LAWRENZ B, DEPRET BIXIO L, COUGHLAN C, *et al*. Inhibin A-A promising predictive parameter for determination of final oocyte maturation in ovarian stimulation for IVF/ICSI. Front Endocrinol (Lausanne), 2020, 11: 307. doi: 10.3389/fendo.2020.00307.

[9] KAR T K, SIL S, GHOSH A, *et al*. Mitigation of letrozole induced polycystic ovarian syndrome associated inflammatory response and endocrinal dysfunction by Vitex negundo seeds. J Ovarian Res, 2024, 17(1): 76. doi:10.1186/s13048-024-01378-4.

[10] CORREA-De-ARAUJO R, YOON S S S. Clinical outcomes in high-risk pregnancies due to advanced maternal age. J Womens Health (Larchmt), 2021, 30: 160–167. doi: 10.1089/jwh.2020.8860.

[11] XU B, WANG J, XIA L, *et al*. Increased uterine NK cell numbers and perforin expression during the implantation phase in IVF cycles with GnRH antagonist protocol. Sci Rep, 2017, 7: 39912. doi: 10.1038/srep39912.

[12] RAGA F, CASAN E M, BONILLA-MUSOLES F. Gonadotropin-releasing hormone (GnRH)-I regulation of interleukin (IL)-1b and IL-1 receptor antagonist 1279AIF-1 and TNF-α in GnRH antagonist protocol expression in cultured human endometrial stromal cells. J Obstet Gynaecol Res, 2008, 34: 464–472. doi: 10.1111/j.1447-0756.2008.00737.x.

[13] 张帆. 卵泡期长效长方案与黄体期短效长方案在卵巢储备良好患者IVF-ET 中的应用比较. 中国优生与遗传杂志, 2019, 27(2): 245–246.
ZHANG F. Comparison of follicular phase long-acting protocol and luteal phase short-acting long-acting protocol in patients with good ovarian reserve in IVF-ET. Chin J Euge Gene, 2019, 27(2): 245–246.

[14] ALPAÑÉS M, ALVAREZ-BLASCO F, FERNANDEZ-DURAN E, *et al*. Combined oral contraceptives plus spironolactone compared with metformin in women with polycystic ovary syndrome: a one-year randomized clinical trial. Eur J Endocrinol, 2017, 177(5): 399–408. doi: 10.1530/EJE-17-0516.

[15] 夏婷婷, 曾克非. 早卵泡期长方案与拮抗剂方案对不孕症患者子宫内膜容受性及临床妊娠的影响. 医学信息, 2023, 36(12): 140–143. doi: 10.3969/j.issn.1006-1959.2023.12.028.
XIA T T, ZENG K F. Effects of long-term regimen and antagonist regimen in early follicular phase on endometrial receptivity and clinical pregnancy in infertile patients. Med Inform, 2023, 36(12): 140–143. doi: 10.3969/j.issn.1006-1959.2023.12.028.

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