Comparison of the effectiveness and safety of the full-down regulation of early ovarian follicle phase and mid-luteal phase for controlled hyperstimulation: a single-center, randomized, controlled trial

Project Number: SAHoWMU-CR2019-07-115
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Unit responsible for the experiment: Second Affiliated Hospital of Wenzhou Medical University, Yuying Children's Hospital

Test statistics unit: Second Affiliated Hospital of Wenzhou Medical University, Yuying Children's Hospital

Person in charge of clinical researcher: Ying Yingfen

Confidentiality statement

Project signature page

Sponsor's signature

I have read and confirmed this plan (project number: SAHoWMU-CR2019-07-115; version number: 01; version date: 2020-04-03). I agree to perform relevant duties in accordance with Chinese law, the Declaration of Helsinki, the Chinese GCP, and this research plan.

Applicants: the Second Affiliated Hospital of Wenzhou Medical University, Yuying children's Hospital

Yingfen Ying	Yingfen Ying	08/15/2020
Person in charge (print)	Person in charge (signature)	Date of signature (mm / DD / yyyy)

Word page

Researcher signature

We have read and confirmed this plan (project number: SAHoWMU-CR2019-07-115; version number: 01; version date: 2020-04-03) and agree to the scientific and ethical nature of this plan. We will perform relevant responsibilities in accordance with Chinese laws, the Helsinki Declaration, the Chinese GCP, and the provisions of this research plan, and only modify the plan after notifying the sponsor, and only after approval by the ethics committee can it be implemented, unless it is for protection Measures must be taken for the safety, rights and interests of the person.

We will keep this research plan confidential.

Participants: the Second Affiliated Hospital of Wenzhou Medical University, Yuying children's Hospital

Yingfen Ying	Yingfen Ying	08/15/2020
Person in charge (print)	Person in charge (signature)	Date of signature (mm / DD / yyyy)

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Acronyms and Abbreviations

AFC	Antral follicle count
BMI	Body mass index
COH	Controlled hyperstimulation
E_2	Estradiol
FSH	Follicle stimulating hormone
GCP	Code for quality management of drug clinical trials
GnRH-a	Gonadotropin releasing hormone agonist
HCG	Human chorionic gonadotropin
ICSI	Intracytoplasmic sperm injection of oocytes
IVF-ET	In vitro fertilization-embryo transfer
LH	Luteinizing hormone
OHSS	Ovarian hyperstimulation syndrome
P	progesterone
r-FSH	Recombinant human follicle stimulating hormone
r-LH	Recombinant luteinizing hormone

Program summary

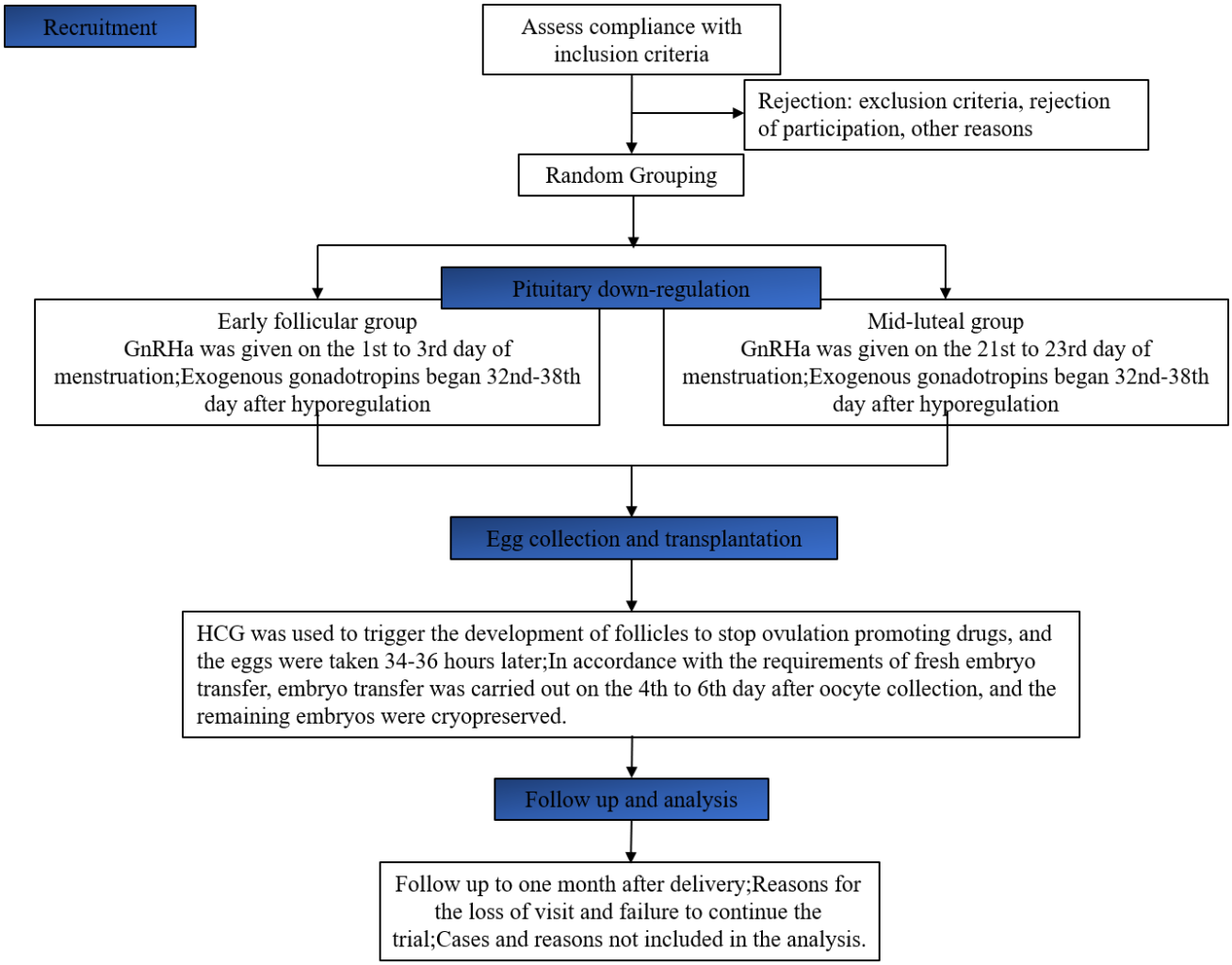
Research topics	**Comparison of the effectiveness and safety of the full-down regulation of early ovarian follicle phase and mid-luteal phase for controlled hyperstimulation: a single-center, randomized, controlled trial**
Study purpose	To clarify whether the whole dose long-acting GnRH-a prolonged protocol under different menstrual cycles really brings the best clinical benefits to infertile patients.
overall design	A prospective randomized controlled trial design was used in this study.
Screening criteria	**Inclusion criteria:** All of the following criteria must be met before they can be included in the test (1) Factors of fallopian tube: such as adhesion around fallopian tube, obstruction of fallopian tube, etc. if the patient has hydrosalpinx, they can be put into the group after hydrosalpinx operation; (2) Ovulation disorders, such as polycystic ovary syndrome, polycystic ovarian changes, etc; (3) Mild to moderate endometriosis; (4) Male factors: oligozoospermia, asthenospermia, dysspermia, etc; (5) Unexplained infertility: the patient who has a history of non contraception and non pregnancy for more than one year, who has no definite cause of infertility, such as ovulation, fallopian tube, endometrium and male factors, or who is still not pregnant after the above factors return to normal after treatment; (6) AFC≥5 in bilateral ovaries; **Exclusion criteria:** All of the following criteria must not be included in the test before screening (1) Those with a history of adverse pregnancy, including: repeated abortion: three or more times of spontaneous abortion, missed abortion, biochemical pregnancy; history of fetal malformation and chromosomal abnormality / intrauterine history of fetal death; (2) unilateral oophorectomy; (3) Patients with the following uterine abnormalities: uterine malformation (single angle uterus, double angle uterus, double uterus, mediastinum uterus); submucous

	myoma; intrauterine adhesions; (4) Chromosomal abnormality of one of the couples; (5) Patients with contraindications of assisted reproductive technology or pregnancy: such as uncontrolled diabetes mellitus, undiagnosed liver and kidney dysfunction, history of deep vein thrombosis, history of pulmonary embolism, history of cerebrovascular accident, uncontrolled hypertension, heart disease, suspected cervical cancer, endometrial cancer, breast cancer or previous history, undetermined vaginal bleeding; (6) Unable to follow up regularly or complete the study in all aspects; (7) Participants in other clinical trials;
Experimental design and method	In the no intervention group, a 3.75mg GnRH-a injection was injected on the 1st-3rd day of menstruation, the level of E_2, P, LH in peripheral blood was monitored on the 32nd-38th day after pituitary hyporegulation, and the number of follicles in bilateral ovarian sinuses was monitored by ultrasound. If pituitary desensitization was achieved, the start-up of Gn began. When the diameter of at least two follicles ≥ 18mm or more than three follicles ≥ 17mm, the trigger of hCG was given and oocyte were retrieved, and the selective fresh single blastocyst transplantation was carried out on the 4th-6th day after oocyte extraction, and 12 days after transplantation bloodβ-hCG test to see whether pregnancy or not, if had pregnancy，patients will continue to be followed-up until 42 days postpartum; In the no intervention group, a 3.75mg GnRH-a injection was injected on the 21st-23rd day of menstruation, the level of E_2, P, LH in peripheral blood was monitored on the 32nd-38th day after pituitary hyporegulation, and the number of follicles in bilateral ovarian sinuses was monitored by ultrasound. If pituitary desensitization was achieved, the start-up of Gn began. When the diameter of at least two follicles ≥ 18mm or more than three follicles ≥ 17mm, the trigger of hCG was given and oocyte were retrieved, and the selective fresh single blastocyst transplantation was carried out on the 4th-6th day after oocyte extraction, and 12

	days after transplantation blood β-hCG test to see whether pregnancy or not, if had pregnancy， patients will continue to be followed-up until 42 days postpartum.
Curative effect observation index	**Main outcome measures:** live birth rate per transplantation cycle (defined as delivery of live fetus ≥ 28 weeks) **Secondary outcome measures:** (1) Clinical pregnancy rate: 26-35 days after transplantation, fetal heart beat was confirmed by transvaginal ultrasound; (2) Good quality blastocyst rate: high quality blastocyst number / D3 embryo culture blastocyst number;
Safety observation index	(1) The incidence of severe ovarian hyperstimulation syndrome; (2) Pregnancy loss rate: the percentage of abortion and fetal death in utero; (3) Incidence of perinatal complications
Follow up time	Follow up 1: 12 ±2 days after transplantation: the subjects received routine pregnancy test (serum β - hCG) to confirm pregnancy, and those with positive (> 50iu / L) continued luteal support; Follow up 2: 28 ±2 days after transplantation: the subjects r eceived the first vaginal ultrasound examination to determine the continued pregnancy, and recorded the size and number of fetal sacs, the length of fetal buds and the presence or absence of fetal heart rate; Follow up 3: 42 ±2 days after transplantation: the subjects received the second ultrasound examination, and recorded the size and number of fetal sacs, the length of fetal buds and the presence or absence of fetal heart; Follow up 4: 11-13 weeks after transplantation: check the cases and telephone follow-up to find out whether there are complications in early pregnancy; Follow up 5: 24 weeks gestation: telephone follow-up to find out if there are complications in the second trimester; Follow up 6: 42 days postpartum: the pregnancy outcome, delivery time and neonatal parameters (gender, weight, abnormal results of physical examination and any deformity) were obtained by telephone follow-up, and the puerperal complications of the mother and the health status of the newborn were understood.

statistical analysis	(1) The sample size calculated by the effectiveness test: $\alpha = 0.05$, $1 - \beta = 0.80$, and the live rate of the traditional rectangular case can reach about 30%. According to the relevant literature reports and our previous retrospective research data, it is estimated that the success rate of the ultra long scheme is about 50%. The difference of clinical significance between the two groups was set at 10%, the ratio of sample size of the two groups was 1:1, and the rate of abscission was about 5%. (2) Statistical analysis method a) Comparability analysis Demographic characteristics and other baseline indicators were compared to weigh the comparability of the two groups. If the continuous variable accords with normal distribution, t test is used, otherwise rank sum test is used, or normal transformation of data is carried out. Chi square test or Fisher exact probability method are used to classify variables. b) Security analysis The adverse events and reactions of the two groups were described in the list, and the adverse reactions were statistically analyzed by chi square test.
sample size	In this study, 1150 subjects were enrolled, 575 in the experimental group and 575 in the control group.
Research cycle	May 2020 ~ May 2023

Technology Roadmap



Solution text

1. Research topics

Comparison of the effectiveness and safety of the full-down regulation of early ovarian follicle phase and mid-luteal phase for superovulation: a single-center, randomized, controlled trial

2. Research Background

Since the birth of the first test-tube baby in 1978, in vitro fertilization-embryo transfer (IVF-ET) has become an important means for infertility treatment in countries around the world. IVF-ET treatment requires different ovulation induction protocols. At present, the commonly used controlled hyper-ovulation schemes in clinic are divided into GnRH-a down-regulation schemes and non-GnRH-a down-regulation schemes according to whether gonadotropin releasing hormone agonist (GnRH-a) is used. It is a key problem to be solved in clinical work to develop an individualized controlled super ovulation program for patients in order to obtain high-quality eggs and high-quality embryos, improve clinical pregnancy rate and live birth rate, and reduce the occurrence of complications.

The early follicular phase ultra-long program due to the controlled ovarian hyperstimulation (COH) before the full amount of GnRH-a injection, so that the test tube baby patient's ovulation promotion cycle can obtain a more ideal number of eggs, increase the endometrium for embryo transfer Tolerability, suppression of endogenous LH peaks, and reduction of cycle cancellation rate showed obvious advantages. The mid-luteal corpus luteum long-term plan also applies the full amount of GnRH-a in front of COH, and its down-regulated pharmacological mechanism of action is similar. Our previous retrospective studies have shown that the two programs have comparable clinical effects. However, at present, whether the clinical and perinatal outcomes of the early follicular phase ultra-long regimen have the same benefits as the mid-luteal phase ultra-long regimen have not been prospectively reported at home and abroad.

3. Test purpose and test design basis

3.1 Test purpose

The research team hopes to further build on our retrospective research, and further proceed from single-center, randomized, prospective, and controlled studies, with IVF as the primary end point, and relevant laboratory and clinical indicators as secondary The end point of the study to further clarify whether the full-length long-acting GnRH-a ovulation induction program under different menstrual cycles really brings the best clinical benefit to IVF patients. If prospective studies also show that the two groups have comparable clinical and perinatal outcomes, it will provide IVF patients with a better choice of treatment options, rather than just full down regulation in the early follicular phase It can flexibly administer the full amount of long-acting dose of down-regulating drugs according to the time of the patient's visit.

3.2 Research overall design

This study used a prospective randomized controlled trial design.

3.3 Research sample size

3.3.1 Calculate the sample size using the superiority test: $\alpha = 0.05$, $1-\beta = 0.80$, the live birth rate of the traditional long plan can generally reach about 30%, according to relevant literature reports and our previous retrospective research data, it is expected The success rate of the ultra-long program is about 50%. The clinically significant difference between the two groups is set at 10%, and the ratio of the sample size of the two groups is 1: 1, then the sample size of each group is 547. Considering the shedding rate of about 5%, the sample size required for this experiment was 1150 cases, with 575 cases in each group.

3.3.2 Statistical analysis methods

3.3.2.1 Comparability analysis

Compare demographic characteristics and other baseline indicators to weigh the comparability of the two groups. If the continuous variable conforms to the normal distribution, the t test is used, otherwise the rank sum test is used, or the data is transformed normally. The categorical variables are chi-square test or Fisher exact probability method.

3.3.2.1 Security analysis

The list describes the adverse events and adverse reactions of the two groups, and the chi-square test was used to statistically analyze the adverse reactions.

3.4 Study the test population

Women aged 20-42 years of infertility (with a history of more than 1 year of infertility), BMI 18-30kg / m2, non-smokers, subjects receiving IVF / ICSI treatment.

4.1 standard constrain

4.1.1 Fallopian tube factors: such as adhesions around the fallopian tube, obstruction of the fallopian tube, etc., if the patient is combined with hydrops of the fallopian tube, he can be enrolled after hydrops operation;

4.1.2 Diseases related to ovulation disorders, such as polycystic ovary syndrome, polycystic ovarian changes, etc .;

4.1.3 Patients with mild to moderate endometriosis;

4.1.4. Male factors: such as oligozoospermia, asthenozoospermia, and deformity spermia;

4.1.5 Unexplained infertility: a history of infertility without contraception for more than 1 year, no clear cause of infertility such as ovulation, fallopian tube, endometrium and male factors, or the above factors returned to normal after treatment and still not pregnant By.

4.1.6 Bilateral ovarian AFC≥5;

4.1.7 Informed consent

4.2 Exclusion criteria (exclude if one of them is met)

4.2.1 Those with a history of poor pregnancy, including: repeated abortions: three or more spontaneous abortions, missed abortions, biochemical pregnancy, fetal malformation and history

of chromosomal abnormalities / intrauterine death

4.2.2 After one ovariectomy;

4.2.3 Patients with the following uterine abnormalities: uterine malformation (single horn uterus, double horn uterus, double uterus, mediastinal uterus); submucosal uterine fibroids; intrauterine adhesions;

4.2.4 One of the husband and wife has abnormal chromosomes;

4.2.5 Patients with contraindications to assisted reproductive technology or pregnancy contraindications: such as uncontrolled diabetes, undiagnosed liver and kidney dysfunction, history of deep vein thrombosis, history of pulmonary embolism, history of cerebrovascular accidents, uncontrolled hypertension , Heart disease, suspicious cervical cancer, endometrial cancer, breast cancer or previous medical history, vaginal bleeding without a clear diagnosis;

4.2.6 Unable to follow up regularly or complete the study from all aspects;

4.2.7 Those who are participating in other clinical trials.

4.3 Exit criteria

Subjects who experienced drug allergies, poor compliance, did not acquire eggs due to poor ovarian function, could not transfer fresh embryos, and could not complete the follow-up task should withdraw from the trial.

4. Test design

5.1 Test procedure

The study method used a prospective randomized controlled trial design:

5.1.1 After signing the informed consent form, conduct a complete medical history collection and detailed physical examination, and patients who meet the inclusion criteria but do not meet the exclusion criteria will be randomly divided into the study.

5.1.2 The subjects were treated according to random grouping after entering the group. The experimental group had a total corpus luteum down-regulation scheme, and the control group: an early follicular phase down-regulation scheme.

5.2 Screening period

The screening period of the outpatient visit is to complete the collection of relevant medical history of IVF, physical examination, complete inspection of IVF and sign the informed consent.

Remarks: 1. If the subject has completed the laboratory examination, electrocardiogram, etc. of the same item in the same or second-level medical institution or more within 365 days before

randomization, in order to avoid repeated examination, the completed examination report can be used as a screening Period results.

5.3 Research period

5.3.1 GnRH-a down regulation in early follicular stage or mid-luteal phase;

5.3.2 Before the start of ovarian volume, AFC, hormones LH, E2, P check FSH to start super ovulation;

5.3.3 hCG trigger;

5.3.4 Egg retrieval;

5.3.5 Embryo transfer;

5.3.6 Serum β-hCG detection 14 ±2 days after transplantation;

5.3.7 If the serum β-hCG test is positive (≥50IU / mL), the test tube baby will be tested first;

5.3.8 Test tube baby first ultrasound examination is good (at least see intrauterine pregnancy sac), test tube baby second ultrasound examination;

5.4 End of study

42 days postpartum: telephone follow-up to obtain pregnancy outcomes, delivery time and neonatal parameters (gender, weight, physical examination abnormal results and any deformities), to understand the mother's puerperium complications and the health status of the newborn.

5. Efficacy evaluation index

6.1 Main curative effect indexes

Live birth rate per transplantation period (defined as those who gave birth to live births ≧ 28 weeks).

6.2 Secondary efficacy indicators

(1) Clinical pregnancy rate: 28-35 days after transplantation, transvaginal ultrasound examination confirmed fetal heartbeat;

(2) Embryo implantation rate: number of embryos implanted / number of embryos transferred;

(3) The rate of high-quality embryos: the number of D3 high-quality embryos / the total number of embryos in D3, of which the number of D3 high-quality embryos is AB embryos with 7-9 cells;

(4) Blastocyst formation rate: the number of blastocysts formed by D4, D5 or D6 / the number of blastocysts cultured by D3 embryos;

(5) High-quality blastocyst rate: the number of high-quality blastocysts / the number of blastocysts in D3 embryo culture;

(6) Serum LH, P and E2 levels on HCG day.

6. Safety evaluation

(1) The incidence of moderate to severe ovarian hyperstimulation syndrome (OHSS);

(2) Pregnancy loss rate: abortion and fetal death account for the percentage of all pregnancy;

(3) Pregnancy complications and comorbidities, such as pregnancy-induced hypertension, gestational diabetes, etc .;

(4) Fetal malformation, intrauterine fetal death, stillbirth and other adverse outcomes.

7.1 Definition of adverse events

From the time the subject signed the informed consent to the completion of all visits, any adverse medical events reported by the subject or observed by the research physician, regardless of whether there is a causal relationship with the trial drug, are considered adverse events. Adverse events include general adverse events, important adverse events and serious adverse events.

7.2 Ways to obtain information on adverse events

Research physicians report all adverse events directly observed in concise medical terms.

7.3 Record of adverse events

The adverse event report form should be filled out truthfully during the test. Record the time, severity, duration, measures and outcomes of adverse events. Adverse events should be recorded in the designated CRF adverse event report form.

When an adverse event is discovered, the investigator should immediately deal with it and report it to the person in charge of the trial, decide the necessary diagnosis and treatment measures according to the condition, and decide whether to discontinue clinical trial observation. All adverse events should be tracked and investigated, and the process and results should be recorded in detail until it is properly resolved or the condition is stable, and the correlation between it and the test drug is determined. If it is related to the test drug, it should be tracked to return to the pre-dose level. Follow-up and follow-up methods can choose hospitalization, outpatient clinic, home visit, telephone, communication and other methods according to the severity of adverse reaction events.

7.4 Criteria for the severity of adverse events

When filling out the CRF adverse event form, the researchers used the CTCAE 4.0 standard to describe the intensity of adverse events using mild, moderate, severe, life-threatening, and death. In order to unify the standards, the criteria for judging light, moderate and severe events are as follows:

Mild: does not affect the normal function of the subject;

Moderate; to some extent affects the normal function of the subject;

Severe: Obviously affect the normal function of the subject.

Pay attention to distinguish the severity and intensity of adverse events. Severe is used to describe intensity, not necessarily SAE. For example, a headache may be severe in intensity, but it cannot be included in SAE unless it meets SAE standards.

7.5 Judgment criteria for the relationship between adverse events and trial medication

Refer to the following 5-level classification criteria for evaluation.

Definitely related: the occurrence of adverse events conforms to a reasonable chronological sequence after medication, and the response conforms to the known type of reaction of the suspected drug: improvement after discontinuation of the drug, repeated administration of the drug and the occurrence of the reaction;

Probably related: the occurrence of adverse events conforms to a reasonable time sequence after medication, and the response conforms to the known type of response of the suspected drug; it is unlikely that there will be another explanation, such as co-administration or concomitant disease;

May be related: Adverse events occur in a reasonable time sequence after medication, and the response conforms to the known response type of the suspected drug; the patient's clinical status or other treatment methods may also produce the response;

May be irrelevant: the occurrence of adverse events does not conform to a reasonable time sequence after medication, and the response does not conform to the known type of reaction of the suspected drug; it is more likely to be related to other factors;

Certainly irrelevant: the reaction does not meet the reasonable chronological sequence after the drug is taken, the response has a type of response known to the non-experimental drug; the patient's clinical status or other treatment methods may produce the response, the disease state improves or the treatment is stopped by other treatment methods. , Repeated use of other treatment methods appears.

Adverse drug reactions include all adverse events that are definitely related, likely related, and likely related.

7.6 Serious adverse events

Serious Adverse Event (Serious Adverse Event, SAE) refers to events that require hospitalization, prolonged hospital stay, disability, work ability, life-threatening or death, congenital malformation, etc. during the clinical trial.

7.7 Treatment plan for serious adverse events

When a serious adverse event occurs, all efforts should be made to rescue and actively take various measures to avoid permanent damage. From the start of the test to any SAE that occurs during the observation period, the investigator must notify the person in charge of the immediate trial, the clinical trial management department of our hospital, and the ethics committee, fill out the "serious adverse event form", and report to the sponsor in 24 hours , Our hospital clinical trial management department, ethics committee and related drug regulatory management department. After the treatment of serious adverse events, the subject ’s situation should be submitted to a follow-up report.

Contacts for serious adverse events are as follows:

Contacts for serious adverse events

Company	contacts	contact information
The Second Affiliated Hospital of Wenzhou Medical University	**Yingfen Ying**	**+086-13732091230**
The Second Affiliated Hospital of Wenzhou Medical University	**Medical ethics committee**	**0577-88002560 feykjkcy@126.com**
The Second Affiliated Hospital of Wenzhou Medical University	**Clinical research center / Office of drug clinical trial institution**	**0577-88002664、88002738 ywlcsy409@163.com**

8. Statistical Analysis

7.1 Calculate the sample size using the superiority test: $\alpha = 0.05$, $1-\beta = 0.80$, the live birth rate of the traditional long plan can generally reach about 30%, according to relevant literature reports and our previous retrospective research data, it is expected to be super long The success rate of the program is about 50%. The clinically significant difference between the two groups was set at 10%, and the ratio of the sample size between the two groups was 1: 1, with a shedding rate of around 5%.

7.2 Statistical analysis methods

7.3 Comparability analysis

Compare demographic characteristics and other baseline indicators to weigh the comparability of the two groups. If the continuous variable conforms to the normal distribution, the t test is used, otherwise the rank sum test is used, or the data is transformed normally. The categorical variables are chi-square test or Fisher exact probability method.

a) Security analysis

The list describes the adverse events and adverse reactions of the two groups, and the chi-square test was used to statistically analyze the adverse reactions.

9. Data management and statistical analysis

9.1 Data selection for statistical analysis

Full Analysis Data Set (FAS): All cases that have been randomized into groups, have used study drugs and have post-medication efficacy evaluation form a full analysis set. For the efficacy data that failed to observe the entire treatment process, the last measured efficacy data was used as the endpoint data for analysis.

Compliant protocol data set (PPS): included in the FAS, meets the inclusion criteria, does not meet the exclusion criteria, no major protocol violations, complete the treatment period and has a curative effect evaluation, good compliance cases, constitute a compliant protocol set for this study.

Safety analysis data set (SS): All cases that were randomized into groups, took at least one study drug, and had post-medication safety evaluation data constitute the safety analysis data set of this study.

9.2 Statistical analysis plan

9.2.1 Calculate the sample size using the superiority test: $\alpha = 0.05$, $1-\beta = 0.80$, the live birth rate of the traditional long plan can generally reach about 30%, according to relevant literature reports and our previous retrospective research data, it is expected The success rate of the ultra-long program is about 50%. The clinically significant difference between the two groups was set at 10%, and the ratio of the sample size between the two groups was 1: 1, with a shedding rate of around 5%.

9.2.2 Statistical analysis methods

9.2.2.1 Comparability analysis

Compare demographic characteristics and other baseline indicators to weigh the comparability of the two groups. If the continuous variable conforms to the normal distribution, the t test is used, otherwise the rank sum test is used, or the data is transformed normally. The categorical variables are chi-square test or Fisher exact probability method.

9.2.2.2 Security analysis

The list describes the adverse events and adverse reactions of the two groups, and the chi-square test was used to statistically analyze the adverse reactions.

10. Modification of the plan

Any necessary changes to the protocol must be carried out in the form of a protocol revision, which needs to be signed and approved by the principal investigator and submitted to the ethics committee for approval or filing.

10.1 Filling out and handing over the case report form

The case report form (CRF) is filled out by the investigator. The data in the CRF comes from the original documents such as the original medical record and the physical and chemical examination report and should be consistent with the original documents. Any observations and inspection results in the test should be filled in the CRF in a timely, correct, complete, clear, standardized, and true manner, and cannot be changed at will. All items in the CRF must be filled in, and no blank or missing items are allowed. If necessary, CRF needs to fill in the reason for data modification when making data correction.

10.2 Data entry and modification

The data entry is the responsibility of the researcher, and the data management is the responsibility of the Clinical Research Center of our hospital. The PS data entry system is used to establish a dedicated database for data entry and management. In order to ensure the accuracy of the data, two data administrators independently perform double entry and proofreading. Any data modification system is recorded during the data entry process.

For the questions in the case report form, the data administrator will generate a question answering form (DRQ), and issue an inquiry to the investigator through the clinical monitor. The investigator should answer and return as soon as possible. Data modification, confirmation and entry, if necessary, can issue DRQ again.

10.3 Data lock

A blind review before statistical analysis is jointly conducted by the research leader, sponsor, statistician, and data administrator. Its important content is to determine the analysis data set (including FAS, PPS, and SS) that each case belongs to. Judgment of missing values and treatment of outliers. Decisions made under blind review cannot be modified after the blindness is removed, and any decisions must be documented.

After blindly reviewing and confirming that the established database is correct, the main researchers, sponsors, and statistical analysts lock the data. The locked data file will not be changed. The data after the database is locked must be kept for future reference, and the blind should be uncovered. At the same time, the blind base and the database should be submitted to statistical experts for statistical analysis.

10.4 Unmasking

The first stage: After all the research data is entered and locked, the staff who keeps the blind background will do a blinding, and submit it to the statistical analyst for statistical analysis according to the statistical plan. The statistical analyst will write a statistical analysis report based on statistical analysis The report results determine the optimal dose for the first stage.

The second stage: after all the research data is entered and locked, the staff who keeps the blind background will do the first blinding, and submit it to the statistical analyst for statistical analysis according to the statistical plan. The statistical analyst will write a statistical analysis report. Submit the main researcher to write a clinical trial summary report and do the second blinding.

11. GCP Principle-Subject's Rights and Safety

1. This plan must be approved by the ethics committee of the lead organization.

2. All subjects must understand the purpose, method, drug effect and response of the trial, participate in the trial voluntarily, and sign a written informed consent.

3. Subjects have the rights specified in the informed consent form, and can withdraw from the trial according to their own wishes without affecting their normal medical treatment.

4. If the subject has an adverse event related to the trial or drug during the trial, the sponsor is responsible for providing the treatment cost or corresponding compensation for the adverse event related to the trial or drug.

5. The test unit should ensure that the medical monitoring system is working properly, and the subject can contact the research doctor at any required time to get timely medical diagnosis and treatment, and can be hospitalized if necessary.

6. Emphasize the responsibilities of medical staff at all levels and observe closely to ensure the safety of the subjects.

7. If the subject has an adverse event, it should be dealt with in time. Follow up the subject until the adverse event is cured and the outcome is stable. The investigator believes that it is no longer clinically meaningful or the subject is lost to follow-up.

8. If serious adverse events occur, necessary measures should be taken to ensure the safety and rights of the subjects, and they should be reported to the ethics committee and the sponsor within 24 hours, and to the State Food and Drug Administration.

9. The informed consent is in compliance with the latest revised version of the Declaration of Helsinki, as well as any applicable regulations and policies. Each participant or legal representative participating in the screening must fully understand the trial and sign the name and date on the informed consent form to express their consent.

12. Researcher's responsibilities

The principal investigator and the participating investigator should perform this trial according to the clinical research protocol, and abide by the Declaration of Helsinki, relevant Chinese regulations, and current GCP principles.

13. Test schedule

2020.05-2020.05 RCT system registration, related process sorting, researcher task assignment

2020.05-2020.10 included trial patients, data collation, follow-up

2020.11-2021.04 Patients included in the trial, data collation, follow-up

2021.05-2021.10 Patients included in the trial, data collation, follow-up

2021.11-2022.04 patients included in the trial, data collation, follow-up

2022.05-2022.10 Patients included in the trial, data collation, follow-up

2022.11-2023.05 Data summary, statistical analysis, research results published

14. references

1. Londra L, Moreau C, Strobino D, Bhasin A, Zhao Y. 2016. Is the type of gonadotropin-releasing hormone suppression protocol for ovarian hyperstimulation associated with ectopic pregnancy in fresh autologous cycles for in vitro fertilization? Fertility and Sterility 106 (3) : 666–672 DOI 10.1016 / j.fertnstert.2016.05.019.

2.Siristatidis CS, Gibreel A, Basios G, Maheshwari A, Bhattacharya S. 2015. Gonadotrophin releasing hormone agonist protocols for pituitary suppression in assisted reproduction. Cochrane Database of Systematic Reviews 9 (11): CD006919

DOI 10.1002 / 14651858.CD006919.pub4.

3.Xiao JS, Su CM, Zeng XT. 2014. Comparisons of GnRH antagonist versus GnRH agonist protocol in supposed normal ovarian responders undergoing IVF: a systematic review and meta-analysis. PLOS ONE 9 (9): e106854 DOI 10.1371 / journal. pone.0106854.

4.Cai J, Liu L, Zheng J, Zhang L, Jiang X, Li P, Sha A, Ren J. 2018. Differential response of AMH to GnRH agonist among individuals: the effect on ovarian stimulation outcomes. Journal of Assisted Reproduction and Genetics 35 (3): 467–473

DOI 10.1007 / s10815-017-1095-z.

5.Liao C, Huang R, Scherer RW, Liang XY. 2015. Prognostic factors associated with clinical pregnancy in in vitro fertilization using pituitary down-regulation with depot and daily low-dose luteal phase gonadotropin releasing hormone agonists: a single center's experience. Journal of Human Reproductive Sciences 8 (1): 30–36 DOI 10.4103 / 0974-1208.153124.

6.Martínez F, Clua E, Devesa M, Rodríguez I, Arroyo G, González C, SolérM, Tur R, Coroleu B, Barri PN. 2014. Comparison of st arting ovarian stimulation on day 2 versus day 15 of the menstrual cycle in the same oocyte donor and pregnancy rates among the corresponding recipients of vitrified oocytes. Fertility and Sterility 102 (5): 1307–1311 DOI 10.1016 / j.fertnstert.2014.07.741.

7.Cheon KW, Song SJ, Choi BC, Lee SC, Lee HB, Yu SY, Yoo KJ. 2008. Comparison of clinical efficacy between a single administration of long-acting gonadotrophin-releasing hormone agonist (GnRHa) and daily administrations of short -acting GnRHa in in vitrofertilizationembryo transfer cycles. Journal of Korean Medical Science 23 (4): 662–666 DOI 10.3346 / jkms.2008.23.4.662.

8.Gao J, Xu YW, Miao BY, Zhou CQ. 2014. Self-control study on reduced-dose depot versus daily administration of gonadotrophin-releasing hormone agonists for pituitary desensitization in in vitro fertilization cycles. Journal of Obstetrics and Gynaecology Research 40 (7): 1901–1906 DOI 10.1111 / jog.12417.

9.Londra L, Moreau C, Strobino D, Bhasin A, Zhao Y. 2016. Is the type of gonadotropin-releasing hormone suppression protocol for ovarian hyperstimulation associated with ectopic pregnancy in fresh autologous cycles for in vitro fertilization? Fertility and Sterility 106 (3): 666–672 DOI 10.1016 / j.fertnstert. 2016.05.019.

10. Broekmans FJ, Bernardus RE, Berkhout G, Schoemaker J. 1992. Pituitary and ovarian suppression after early follicular and mid-luteal administration of a LHRH agonist in a depot formulation: decapeptyl Cr. Gynecological Endocrinology 6 (3): 153–161 DOI 10.3109 / 09513599209015549.

11.Ying Y, Yang T, Zhang H, Liu C, Zhao J. 2019. Prolonged pituitary down-

regulation with full-dose of gonadotropin-releasing hormone agonist in different menstrual cycles: a retrospective cohort study. PeerJ 7: e6837 DOI 10.7717 / peerj.6837