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Allogeneic ovarian transplantation promotes the recovery of ovarian function in a rat model of cisplatin-induced premature ovarian failure

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In this study, ovarian transplantation at different sites was performed on the rat model of premature ovarian failure, to explore the effect on the recovery of ovarian function, and to evaluate the application value in the treatment of ovarian function decline caused by the application of chemotherapy drugs. Thirty 2-month-old female SD rats of SPF grade were randomly divided into five groups, and both ovaries in the model group were treated with cisplatin (11 mg/kg) for modelling. After successful modelling, another 18 rats were taken as donor rats: group A was the control group, group B was the model group and group CDE was the experimental group. Ovary transplantation was performed using muscle, subcutaneous and renal peritoneum, respectively, and the rats were monitored for their general condition, estrous cycle and serum hormone levels after surgery. At the end of the observation period, the rats were executed and follicular development was observed.We confirm that all methods are carried out in accordance with the relevant guidelines and regulations. After 14 days of xenotransplantation, the general condition of the rats in the experimental group was better than that of the model group. Serum E2 and AMH levels were significantly higher than those in the model group ($P = 0.000$), and FSH levels were significantly lower than those in the control group ($P = 0.000$). Cisplatin can be used to successfully establish the rat POF model; the three transplantation sites (muscle, subcutaneous and renal peritoneum) selected in this experiment can make the transplanted ovarian tissue survive and successfully restore the endocrine function of the body, and the effect of which showed no significant difference.

Keywords Rat POF model, Allogeneic ovary transplantation, Estrous cycle, Serum oestrogen, AMH

The ovary is responsible for ovulation and the secretion of female hormones, and its dysfunction or loss causes a number of symptoms of endocrine disorders[1]. In recent years, as the incidence of malignant tumours has increased and tended to be younger, more and more patients with tumours have had their survival prolonged by modern medical technology. At the same time, however, the problem of premature ovarian failure (POF) has emerged. More and more menstruating women are experiencing endocrine dysfunction, infertility and other problems that put them under great physical and psychological strain. In addition, estrogen deficiency leads to irreversible skeletal dysplasia and metabolic abnormalities during puberty, resulting in retarded bone growth and abnormal fat distribution[2]. Therefore, the question of how to restore or preserve female endocrine and reproductive functions has gradually become a concern.

Against the background of the successful development of organ transplantation technology and the successful development of various highly effective immunosuppressive agents, ovarian transplantation technology has made great progress and has been gradually applied in the clinic. There are no studies on whether this technique can be used to restore ovarian function in chemotherapy patients. In the technique of ovarian transplantation, one issue that deserves attention is how to enable the graft to rapidly form a vascular network on the contact surface of the recipient tissue to provide blood for the development of the ovary after transplantation. The formation of the vascular network is related to the site of transplantation of the ovary, which should not only

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facilitate manipulation, but also enable the graft to rapidly establish a blood supply on the contact surface of the recipient tissue. A large number of studies have confirmed the survival, growth and development of ovarian grafts in subperitoneal, subcutaneous and intramuscular kidney grafts[3]. However, whether there is a difference in endocrine function after ovarian transplantation at different sites remains to be investigated.

Cisplatin is one of the most commonly used chemotherapeutic drugs in gynaecology, and its toxic effect on the ovaries is moderate. In the present study, a rat POF model was established using the chemotherapeutic drug cisplatin, and after successful modelling, different parts of allogeneic ovaries were transplanted to observe the follicular development of the transplanted ovaries and the recovery of endocrine function. In this study, we investigated the feasibility of allogeneic ovarian transplantation (AOT) for the recovery of ovarian function after chemotherapy and compared the different transplantation sites in order to provide an experimental basis for the clinical application of this technique.

Materials and methods

Experimental animal

A total of 48 2-month-old female SD rats(Sprague-Dawley rats), SPF grade, weighing approximately 200 g, were purchased from Beijing Specific Bio-Technology Co. Ltd, Animal Licence No.: SCXK (Beijing) 2019-0010, 110,324,220,105,330,814.All rats were housed in an SPF grade animal house with free access to food and water, room temperature of 20–23 °C, humidity of 50–65%, and 12 h/12 h regular light day and night. This study was approved by the Research Ethics Committee of Hebei University Hospital **(Code: IACUC-2022XS032). All methods were carried out according to relevant guidelines and regulations and are reported under the ARRIVE guidelines.**

Main instruments and reagents

Cisplatin for injection (lyophilised) (purchased from Qilu Pharmaceutical Co., Ltd.); penicillin for injection (purchased from Zhongnuo Pharmaceutical Co., Ltd.); rat E2, FSH, AMH enzyme-linked immunosorbent assay (ELISA) kits (purchased from RAYTO Raytheon, Inc., USA); YP520N electronic scales, fully automated dewatering machine (Leica, Inc., USA), KD-BMIV, BLIV tissue embedding machine The following are the results of the study: YP520N electronic scale, fully automatic dehydrator (Leica, USA), KD-BMIV, BLIV tissue embedding machine, ambient tabletop high-speed centrifuge (Eppendorf), ci-1 biomicroscope (Nikon, Japan), enzyme labelling instrument (Raytor, USA), and CKX-41 microscope (Olympus, Japan).

Experimental methods

Animal groupings

Experiment I. Replication of the animal model. Thirty female rats were randomly divided into five groups of 6 rats each: group A was the control group, and rats in group BCDE received cisplatin (11 mg/kg) in both ovaries, and the route of administration was direct injection into the open ovary.

Experiment 2: Allogeneic ovarian transplantation Eighteen rats were taken as donor rats, kept under the same conditions and underwent unilateral ovariectomy on the day of transplantation. Group A rats were used as control group, group B rats as model group and group CDE rats as experimental group and were transplanted with homografted ovarian tissue into the muscle, subcutaneously and under the renal peritoneum, respectively.

Methods of anaesthesia

Ethyl carbamate anaesthesia by intraperitoneal injection. Specific procedure: After intraperitoneal injection of ethyl carbamate in rats, anaesthesia can be achieved in approximately 5 ~ 10 min. The specific manifestation of successful anaesthesia is the disappearance of the upright position of the forepaw and the absence of somatic response to tail clamping[4].

Surgical methods

Cisplatin Ovarian Injection: Rats were anaesthetised by intraperitoneal injection of ethyl carbamate solution at a concentration of 20% at 4 ml/kg body weight. After approximately 2 min, when the body and limbs were limp and in the anaesthetic state, the rats were placed supine on the operating table, the lower abdominal wall was shaved and disinfected with iodophor. Ophthalmic forceps were used in the left hand to clip the skin on the middle and lower 1/3 of the abdominal wall, and ophthalmic scissors were used in the right hand to make a longitudinal incision of approximately 0.5 cm. The abdominal wall muscle was then clamped with forceps and the muscle and peritoneum were cut in the same direction to expose the bilateral ovaries[5]. An insulin needle was inserted approximately 1.5 mm and cisplatin solution was injected directly into the cortical part of each ovary at 11 mg/kg body weight. The muscle and skin were then sutured sequentially.

Ovariectomy in donor rats: Anaesthesia, positioning, disinfection and opening of the surgical access were performed as described above. The ovary and oviduct were exposed, the root of the ovary was clamped with haemostatic forceps, ligated with a 4-gauge wire, and the ovary was cut along the root with ophthalmic scissors and placed in saline. The muscle and skin were then sutured sequentially.

Allogeneic ovarian transplantation: Anaesthesia, positioning and disinfection methods are the same as before. Submuscular transplantation: the skin was cut along the original incision, the muscle was separated into two layers with small curved forceps, and a complete donor ovary tissue was cut into small 2mm3 pieces[6], placed between the two muscle layers and fixed with absorbable protein suture. The skin was sutured and the wound sterilised with iodine povidone. ② Subcutaneous transplantation: the skin was incised along the original incision and a complete donor ovarian tissue was cut into small 2mm3 pieces and placed under the skin. The skin was sutured and the wound disinfected with iodophor. (iii) Subperitoneal transplantation: slightly above the original incision, the skin and muscle (with the peritoneum) are cut sequentially, the length of the incision is

approximately 1 cm, and the kidney is exposed. The kidney is carefully lifted out of the incision and fixed, a small opening is made in the renal peritoneum using forceps, a complete donor ovary is cut into small 2mm3 pieces and placed under the renal peritoneum to reposition the kidney. The muscle and skin were sutured in layers and the wound sterilised with iodine povidone[6] (see Fig. 1 for details).

Observation indicators

General status of rats

Observe the fur, paw colour, body shape and locomotor status of the rats in each group.

Changes in body weight after modelling and transplantation in rats

Motivational cycle detection

Vaginal swabs were taken to monitor the fluctuation of the motility cycle in the rats from day 8. Every day at 8:00 am, a sterilised cotton swab dipped in saline was inserted about 0.5 cm into the vagina of the rats and gently rotated for 2 turns. The secretions were collected and smeared onto microscope slides, stained with HE and the results observed.

ELISA for E2, FSH and AMH levels

The ELISA kit was equilibrated at room temperature for 30 min. serum specimen was dissolved on ice for 30 min. standards were diluted, three wells were set up for blank, standard and samples to be examined, samples were added, incubated at 37℃ for 30 min, washed, 50 µl of enzyme reagent was added, incubated, and colour development solution A and B were added sequentially, and the colour was developed at 37℃ for 10 min. termination solution was added dropwise, and the concentration of each sample was calculated according to the OD value and concentration of standards in the standard curve at 450 nm. The optical density (OD) value of each well was detected at 450 nm of the enzyme marker. Plot the standard curve according to the OD value and concentration of the standard, and calculate the concentration of each sample.

Follicular development in transplanted ovarian tissue

Observe the growth and development of the transplanted ovarian tissues and the surrounding blood supply, and exfoliate the transplanted ovaries completely for histological evaluation. Tissues were dehydrated, hyaline, fixed, dehydrated, xylene hyaline, dehydrated step by step, paraffin embedded, and then serially sectioned at a thickness of 3–4 µm.The fifth section of each ovarian tissue was taken for HE staining[7] and follicular development was assessed by light microscopy.

Sample size calculation

The pre-experiment showed that the treatment effect was significant (mean difference = Δ, SD = σ), the minimum demand $n = 5$ was calculated by G*power (independent t-test, $\alpha = 0.05$, power = 0.8), and the actual sample size was slightly higher than the theoretical value. Statistical validation: post-hoc efficacy analysis and sensitivity analysis confirmed that the results wererobust; Ethical compliance: Following the 3Rs principle, the sample size is approved by the Ethics Committee.

Statistical methods

Data analysis was performed by applying SPSS22.0 statistical software, and the measurement information was expressed as mean ± standard deviation ($x \pm s$); intergroup and intragroup comparisons were performed by one-way ANOVA or ANOVA with repeated-measures design; and further two-by-two comparisons were performed by LSD-t test. $p < 0.05$ was taken as the difference was statistically significant.

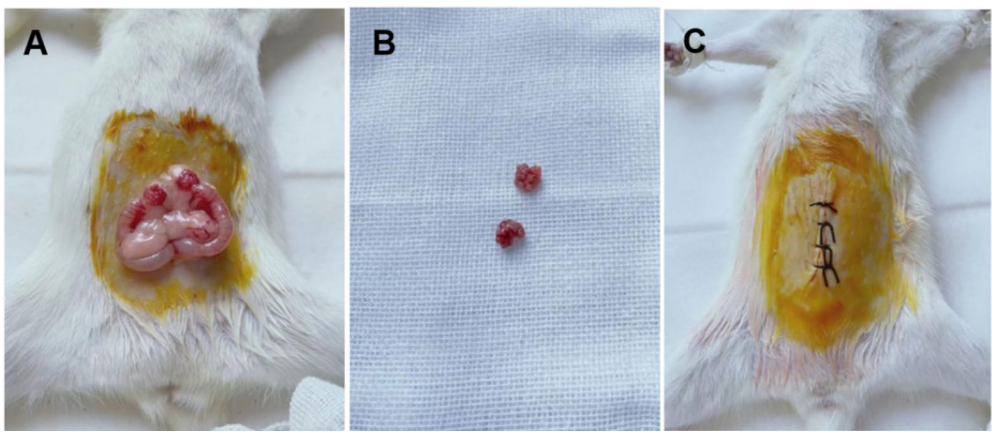

Fig. 1. Specific Surgical Procedures (**A**):Incision of the abdominal wall exposes the uterus and bilateral ovaries; (**B**):a complete donor ovary tissue was cut into small 2mm3 pieces; (**C**):The abdominal wall is sutured after surgery.

Results

General conditions of rats

After modelling, the rats in the normal control group (group A) had a glossy coat, sensitive and active movements, frequent feeding and good responsiveness to external stimuli. The rats in each model group (groups B, C, D and E) had dull and lustrous fur, were slow and sluggish, atrophied, ate little and responded poorly to external stimuli. After transplantation, the rats in the experimental groups (groups C, D and E) showed increased coat sheen, agile activity, increased food intake and increased responsiveness and flexibility to external stimuli compared to the pre-transplantation period.

Changes in body weight of rats

After modelling: Comparison of the body weights of rats in each model group from day 10 to day 14 with those of rats in the control group: ① There was a difference in the body weights of rats at different time points ($F = 1405.134$, $P = 0.000$); ② The difference in the body weights of rats in the model group was not statistically significant ($P > 0.05$). Compared with the body weight of rats in the normal control group, the difference was statistically significant ($F = 1119.841$, $P = 0.000$); ③ The difference in the trend of body weight of rats in the model group was not statistically significant ($P > 0.05$). Compared with the control group, the body weight of rats in the control group showed a steady increasing trend; the body weight of rats in each model group showed a decreasing trend ($F = 469.189$, $P = 0.000$). See Fig. 2 A.

Post-transplantation: Comparison of body weights of rats in each experimental group with those in the control and model groups from day 10 to day 14: ① The difference in the body weights of rats at different time points was statistically significant ($F = 230.158$, $P = 0.000$); ② There was no statistically significant difference in the body weights of rats among the experimental groups ($P > 0.05$), and there was a statistically significant difference in the body weights of rats comparing the experimental groups with the control and model groups ($F = 1831.\ 06$, $P = 0.000$); and ($F = 1831.06$, $P = 0.000$); (iii) the difference in body weight of rats between the experimental groups was not statistically significant ($P > 0.05$), and the body weight of the model group showed a decreasing tendency compared with that of the control group; and the body weight of the model group showed a slow increasing tendency compared with that of the experimental group ($F = 171.11$, $P = 0.000$). See Fig. 2 B.

Motivational cycle observation

The estrous cycle of normal rats is generally 4-6d, divided into four stages, namely, pre-estrus, estrus, post-estrus and inter-estrus. The changes of vaginal exfoliative cells in each period were as follows: (1) a large number of oval-shaped nucleated epithelial cells and a small number of keratinised cells in the pre-motility period, and basically no leukocytes; (2) a large number of flaky anucleated keratinised epithelial cells and a small number of nucleated epithelial cells, with very few leukocytes; (3) a large number of leukocytes in the late stage of the motility period, and a small number of nucleated epithelial cells and flaky anucleated keratinised epithelial cells; (4) the full field of view of leukocytes was visible in the inter-motility period. leucocytes are seen in the full field of view. See Fig. 3.

After modelling: The control group had a normal and regular estrous cycle, and the difference between the two model groups was not statistically significant when compared with the two model groups ($P > 0.05$). Compared with the control group, the ratio of inter-oestrous interval (the ratio of inter-oestrous interval to the whole oestrous cycle) in the model group was higher than that in the control group ($P < 0.05$). See Fig. 4.

After transplantation: The control group had a normal and regular estrous cycle.There was no statistically significant difference between the three experimental groups ($P > 0.05$);when further compared with the control group and the model group, the ratio of the inter-estrous interval in the model group was higher than that in the control group ($P < 0.05$);the ratio of the inter-estrous interval in the experimental group was lower than that in the model group ($P < 0.05$),and the difference was not statistically significant when compared with that in the control group ($P > 0.05$). See Fig. 5.

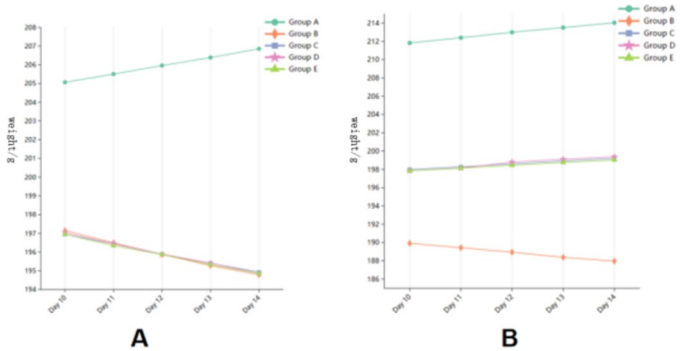

Fig. 2. Changes in body weight of rats (Figure (**A**): Trends of body weight changes in 5 groups of rats after modelling; Figure (**B**): Trends of body weight changes in 5 groups of rats after transplantation).

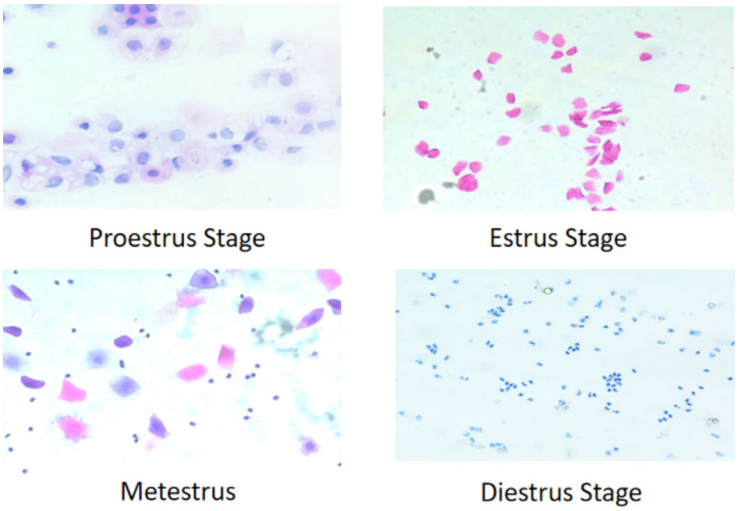

Fig. 3. Vaginograms of 4 periods of the estrous cycle in normal rats (HE staining × 200).

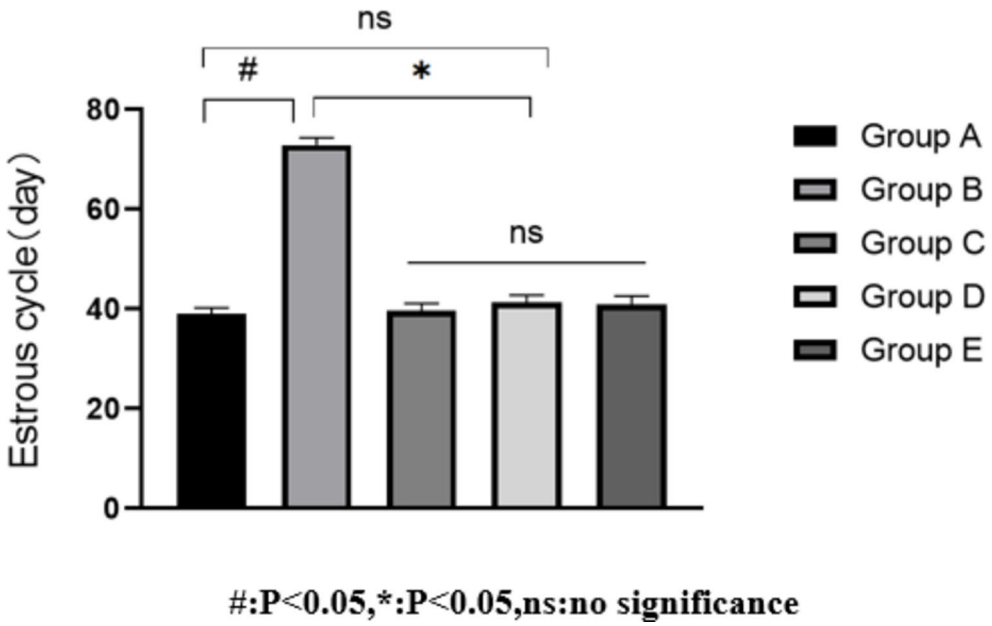

Fig. 4. Post-modelling motility cycles in groups of rats.

Comparison of serum E2, AMH and FSH levels in 5 groups of rats

After 14 days of modelling: The difference between the model groups was not statistically significant ($P > 0.05$). Compared with the control group, the serum E2 level of rats in the model group was reduced, and the difference was statistically significant ($F = 11.909$, $P = 0.000$); the AMH level was reduced, and the difference was statistically significant ($F = 26.190$, $P = 0.000$); and the FSH level was increased, and the difference was statistically significant ($F = 38.607$, $P = 0.000$). See Table 1; Fig. 6.

After 14 days of transplantation: The difference was not statistically significant between the experimental groups ($P > 0.05$), and the difference was not statistically significant when compared with the control group ($P > 0.05$). Compared with the model group, serum E2 level increased, and the difference was statistically significant ($F = 5.979$, $P = 0.000$); AMH level increased, and the difference was statistically significant ($F = 60.931$, $P = 0.000$); FSH level decreased, and the difference was statistically significant ($F = 9.261$, $P = 0.000$). See Table 2; Fig. 7.

Histomorphology of the ovary in 5 groups of rats

In the control group (Group A), follicles of different developmental stages could be seen, mature follicles could be seen at the edge of the ovarian cortex, and the number of granulosa cells was large and regularly arranged; in the model group (Group B), the interstitium of the ovary was atrophied, and more atretic follicles could be seen

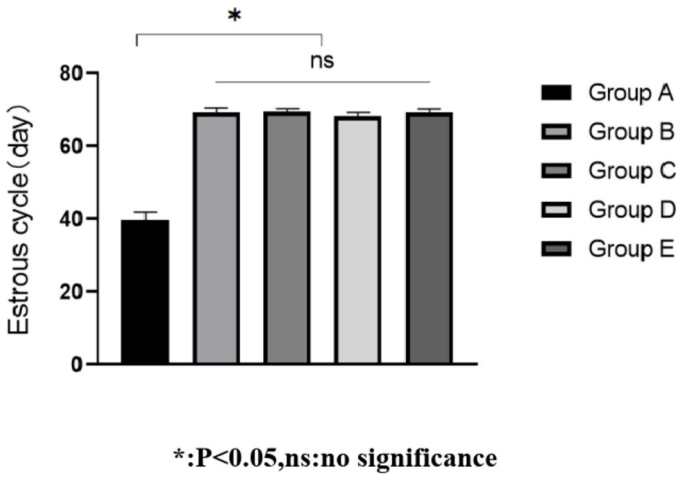

Fig. 5. Post-transplantation motility cycles in groups of rats.

Groups	E2(pg/ml)	FSH(IU/L)	AMH(pg/ml)
Group A	86.12 ± 11.48	8.06 ± 0.24	3088.88 ± 288.16
Group B	43.52 ± 19.24	10.99 ± 0.55	2424.58 ± 247.56
Group C	57.57 ± 10.03	10.71 ± 0.46	2544.79 ± 224.28
Group D	51.15 ± 9.79	10.35 ± 0.57	2629.74 ± 220.38
Group E	46.77 ± 6.16	10.65 ± 0.46	2228.89 ± 192.98

Table 1. Comparison of serum hormone levels in 5 groups of rats after 14 days of modelling ($n = 6$ x ± s).

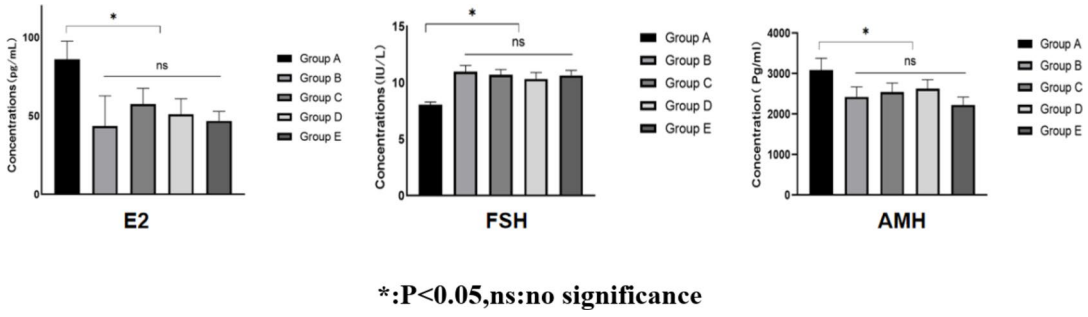

Fig. 6. Comparison of serum hormone levels in 5 groups of rats after 14 days of modelling ($n = 6$ x ± s).

Groups	E2(pg/ml)	FSH(IU/L)	AMH(pg/ml)
Group A	97.13 ± 16.57	8.77 ± 0.59	3573.38 ± 267.62
Group B	59.90 ± 11.93	12.10 ± 1.17	2148.37 ± 169.68
Group C	82.21 ± 12.47	8.97 ± 1.55	2812.11 ± 120.98
Group D	81.73 ± 6.37	9.06 ± 0.96	2875.76 ± 55.62
Group E	77.60 ± 16.68	8.69 ± 1.33	2836.20 ± 84.5

Table 2. Comparison of serum hormone levels in 5 groups of rats 14 days after transplantation ($n = 6$ x ± s).

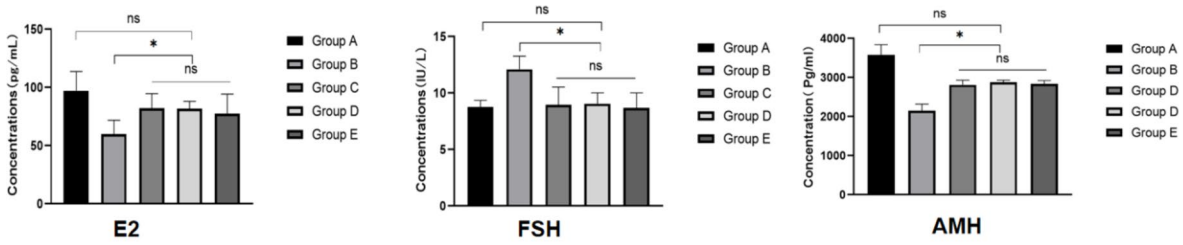

Fig. 7. Comparison of serum hormone levels in 5 groups of rats after 14 days of modelling ($n = 6 \text{ x} \pm \text{s}$).

at the cortical part, with reduced and disorganised granulosa cells; and in the experimental group (Groups C, D, and E), a very small number of atretic follicles could be seen, and mature follicles could be seen at the edge of the ovaries, and the number of granulosa cells was large and orderly arranged. See Fig. 8.

Discussion

In our study, we find that cisplatin can be used to successfully establish the rat POF model.The three transplantation sites (muscle, subcutaneous and renal peritoneum) selected in this experiment can make the transplanted ovarian tissue survive and successfully restore the endocrine function of the body, and the effects were similar between these three groups with no significant differences.

Premature ovarian failure in women has a major impact on patients' health and daily lives. Premature ovarian failure can occur for a variety of reasons, such as chromosomal abnormalities, autoimmune diseases, and a history of multiple surgeries[9-11].The most important of which are chemotherapy and radiotherapy. Chemotherapy or radiotherapy can cause ovarian damage as a side effect of treating cancer patients[10]. Cisplatin, a first-line drug widely used in various solid tumours, has a moderate degree of gonadotoxicity and affects normal cells while killing tumour cells. At the same time, female gonads are particularly sensitive to cisplatin, which has led to an increasing number of cases of ovarian failure[11]. In this experiment, the rat POF model was established by injecting cisplatin solution directly into the ovary. After modelling, the general condition, body weight changes, erotic cycle fluctuations and serum hormone levels of the rats in each group were compared to show that the ovarian function of the rats in the model group was impaired, which further confirmed that direct injection of cisplatin into the ovary could be used as a means to replicate the POF model.

The first successful autologous ovarian transplantation in animals was performed by Knauer in 1896, followed by successful ovarian transplants in a variety of animals and humans, which laid the foundation for the development of ovarian transplantation techniques[12]. Ovarian tissue transplantation includes autologous transplantation, allogeneic transplantation, xenotransplantation, ovarian cortical transplantation, intact ovarian transplantation, in situ transplantation and ectopic transplantation. Research into ovarian transplantation techniques dates back to the 1860s[13], and today, with the increasing sophistication of organ transplantation techniques, ovarian transplantation techniques have also progressed considerably. Autologous ovarian transplantation may carry the risk of spilling cancer cells from the ovary back into the body, causing recurrence of the primary tumour. Both perfusion and cryopreservation can have a detrimental effect on the ovaries and result in the loss of large numbers of follicles. Allogeneic ovarian transplantation (AOT) is a powerful technique that can effectively improve the patient's symptoms while avoiding the high risk of medical therapy and the risk of tumour recurrence and dissemination caused by damage to the ovarian tissue during cryopreservation and thawing of the autologous ovarian graft. There is experimental evidence for the existence of effective immunomodulatory compounds that prevent organ rejection and preserve ovarian function well[14]. In recent years, ovarian allotransplantation has shown promising applications in the treatment of ovarian diseases requiring surgical removal of the ovaries due to ovarian pathology, premature ovarian failure or loss of ovarian function due to radiotherapy, as well as in menopausal women due to deterioration of ovarian function. At present, the feasibility and efficacy of allogeneic ovarian transplantation (AOT) to restore female endocrine function and fertility require further experimental data. There are no studies on whether this technique can be used to restore ovarian function in chemotherapy patients. In this experiment, after successfully reproducing the POF model, we implanted ovarian tissue allografts into the experimental group of rats, and after transplantation, we observed that the hair glossiness, activity, feeding and responsiveness to external stimuli of the rats improved compared with those before transplantation, and their body weight slowly increased, and they could gradually restore the normal estrous cycle and the levels of serum E2, FSH and AMH, and the primordial follicles of the transplanted ovarian tissue developed and matured under microscopic observation. The above results showed that the transplanted ovarian tissue could survive and maintain the physiological function of sex hormone secretion.

The most important factor affecting the survival of ovarian transplantation is the choice of transplantation site, which plays a key role in the haemorrhage of the transplanted ovary, and ischaemia is the main cause of follicular depletion[15]. Follicular depletion due to local ischaemic injury is a key factor limiting the use of this

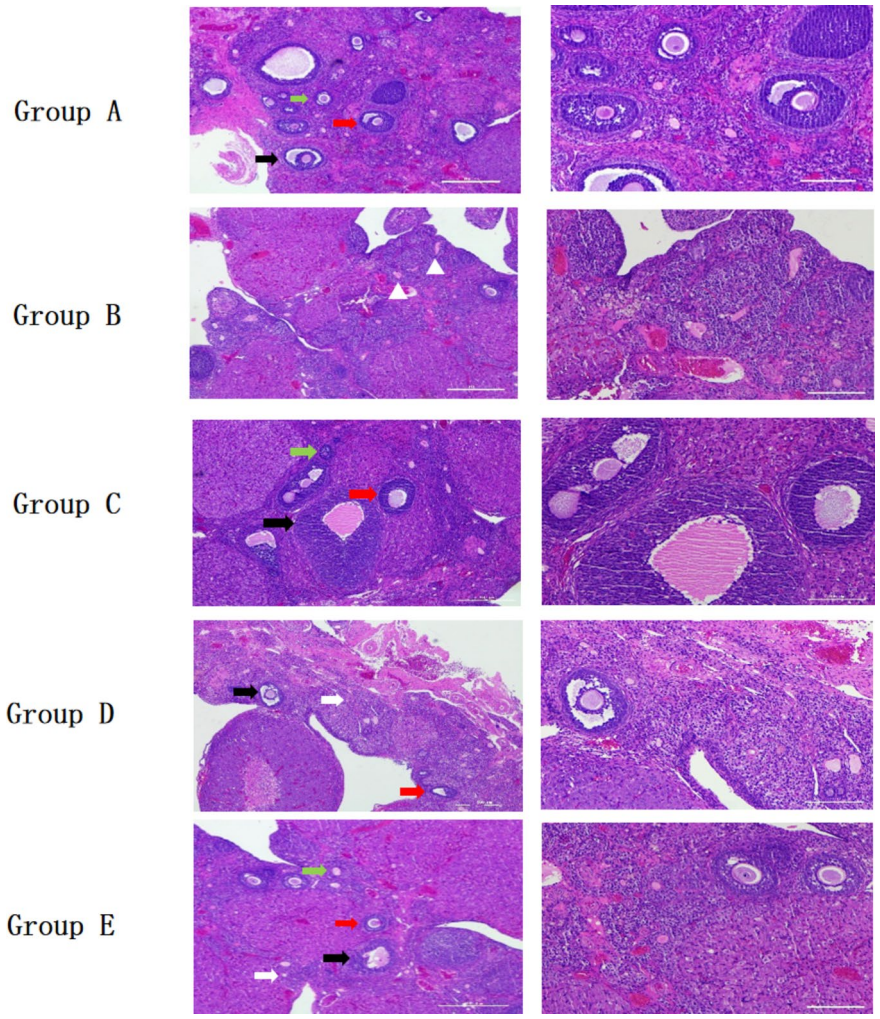

Fig. 8. Histomorphology of the ovary in each group of rats(White arrow: primordial follicle; green arrow: primary follicle; red arrow: secondary follicle; black arrow: sinus follicle; white triangle: atretic follicle.)The picture on the right is enlarged on the left.

technique, as it affects the hormonal environment and recovery of reproductive function after transplantation. Reducing the ischaemic time between ovarian reimplantation and spontaneous microvascular regeneration of the graft is essential to maintain follicular viability[16]. A number of strategies have been developed to minimise hypoxic tissue damage and restore blood supply more rapidly to improve follicular outcomes in terms of increased follicularity. In ovarian transplantation techniques, a topic of interest is how to enable the graft to rapidly generate a vascular network on the recipient tissue contact surface to provide blood for ovarian development after transplantation. The generation of the vascular network is related to the transplantation site of the ovary, which should not only be convenient for manipulation, but also enable the graft to rapidly establish a blood supply on the contact surface of the recipient tissue. According to the literature, the available sites for ovarian transplantation include intracapsular, subperitoneal, intraperitoneal, muscle and subcutaneous[17], but whether there is a difference in endocrine function after ovarian transplantation at different sites remains to be investigated. The three transplantation sites of muscle, subcutaneous and subperitoneal of the kidney selected in this study did not require vascular anastomosis, and at the same time, the transplantation sites were superficial and less invasive, which facilitated the clinical operation. When the results of the experimental group were compared, no significant differences were found in the general condition, body weight, estrous cycle, serum hormone levels and recovery of follicular development in the ovarian tissue of the rats. Compared with the model group, all indices were improved and the results showed that all three sites could make the transplanted ovaries survive and maintain a more normal ovarian function, providing more experimental data for the selection of allogeneic ovarian transplantation sites.

In conclusion, cisplatin can induce ovarian dysfunction in rats, which can be used to establish a POF model in experimental animals; the method of direct injection into the ovary reduces the systemic side effects of the drug and improves the survival rate of experimental animals; rat ovarian allografts are able to survive and possess normal endocrine functions; the three transplantation sites of muscle, subcutaneous and subperitoneal of the kidney in the selection area of the present experiment were able to survive and successfully restore the

endocrine function of the organism. In this experiment, all three transplantation sites, i.e. muscle, subcutaneous and subperitoneal, were able to restore the endocrine function of the organism.

Ovarian tissue allografts can maintain the supply of oestrogen and nutrients in the body, which is undoubtedly a major contribution to improving the quality of life of these groups of women, and this is obviously a worthwhile and far-reaching issue. The technique is not yet fully developed and the results of transplantation are influenced by many factors. In addition, the technology of ovarian transplantation faces many problems that need to be solved quickly, such as low success rates and the ethics associated with transplantation. More basic and clinical research is needed in the future.

The specific mechanism of ovarian tissue allotransplantation can be further verified by molecular and cellular studies, so that more efforts can be made to improve the success rate and efficacy of clinical application of this technique. It is believed that in the near future, ovarian transplantation will be as widely used in clinical practice as other organ transplantation techniques, contributing to the improvement of physical and mental health and quality of life of patients.

Shortcomings and prospects

The possibility of maintaining the body's supply of oestrogen and nutrient factors through ovarian tissue transplantation would undoubtedly make a major contribution to improving the quality of life of these groups of women, and this is clearly a topic that deserves to be explored with far-reaching implications. The technique is not yet fully developed and the results of transplantation are influenced by many factors. In addition, the technology of ovarian transplantation faces many problems that need to be solved quickly, such as low success rates and the ethics associated with transplantation. More basic and clinical research is needed in the future.

The specific mechanism of ovarian tissue allotransplantation can be further verified by molecular and cellular studies, so that more efforts can be made to improve the success rate and efficacy of clinical application of this technique. It is believed that in the near future, ovarian transplantation will be as widely used in clinical practice as other organ transplantation techniques, contributing to the improvement of physical and mental health and quality of life of patients.

The datasets used and/or analysed during the current study available from the corresponding author on reasonable request.

Data availability

The datasets used and/or analysed during the current study available from the corresponding author on reasonable request.

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Author contributions

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Declarations

Competing interests

The authors declare no competing interests.

Statement

We claim that the study was reported in accordance with the ARRIVE guidelines. The number of main body:3954 words

Additional information

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