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ORIGINAL ARTICLE *Infertility*

Inactivated COVID-19 vaccination does not affect *in vitro* fertilization outcomes in women

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STUDY QUESTION: Do inactivated coronavirus disease-2019 (COVID-19) vaccines affect IVF outcomes among the vaccine recipients?

SUMMARY ANSWER: The receipt of inactivated COVID-19 vaccines before ovarian stimulation has little effect on the outcomes of IVF, including ovarian stimulation outcomes, embryo development and pregnancy rates.

WHAT IS KNOWN ALREADY: Limited studies have reported that COVID-19 vaccines do not affect ovarian function, embryo development or pregnancy outcomes.

STUDY DESIGN, SIZE, DURATION: This was a retrospective cohort study performed at the Third Affiliated Hospital of Guangzhou Medical University on 240 women vaccinated with either CoronaVac or Sinopharm COVID-19 before ovarian stimulation in the exposed group and 1343 unvaccinated women before ovarian stimulation in the unexposed group. All participants received fresh embryo transfers between 1 March 2021 and 15 September 2021. The included women were followed up until 12 weeks of gestation.

PARTICIPANTS/MATERIALS, SETTING, METHODS: Vaccination information of all subjects was followed up by a nurse, and the IVF data were obtained from the IVF data system. The following aspects were compared between the vaccinated and the unvaccinated groups: parameters of ovarian stimulation, embryo development and pregnancy rates. Regression analyses were performed to control for confounders of embryo development and pregnancy rates. Propensity score matching (PSM) was performed to balance the baseline parameters of the two groups. The primary outcome was the ongoing pregnancy rate.

MAIN RESULTS AND THE ROLE OF CHANCE: Liner regression analysis revealed that the number of oocytes retrieved (regression coefficient (B) = −0.299, P = 0.264), embryos suitable for transfer (B = −0.203, P = 0.127) and blastocysts (B = −0.250, P = 0.105) were not associated with the status of vaccination before ovarian stimulation, after adjusting for the confounders. The ongoing pregnancy rate in the women of the vaccinated group was not significantly lower than that in the unvaccinated group (36.3% vs 40.7%, P = 0.199) (adjust odd ratio = 0.91, 95% CI = 0.68–1.22, P = 0.52). After PSM, the rates of ongoing pregnancy (36.0% vs 39.9%, P = 0.272), implantation (35.4% vs 38.3%, P = 0.325), biochemical pregnancy (47.3% vs 51.6%, P = 0.232), clinical pregnancy (44.4% vs 47.4%, P = 0.398) and early miscarriage (15.0% vs 12.1%, P = 0.399) were not significantly different between the vaccinated and the unvaccinated groups.

LIMITATIONS, REASONS FOR CAUTION: This is a retrospective study of women with infertility. The results from the present study warrant confirmation by prospective studies with a larger cohort.

WIDER IMPLICATIONS OF THE FINDINGS: This is the first study with a large sample size on the effect of inactivated COVID-19 vaccines on ongoing pregnancy rates of women undergoing IVF. The present results showed that vaccination has no detrimental effect on IVF outcomes. Therefore, women are recommended to receive COVID-19 vaccines before undergoing their IVF treatment.

[†]The first three authors contributed equally to this work.

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Introduction

Coronavirus disease-2019 (COVID-19), which emerged at the end of 2019, is caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). A total of 543 352 927 cases of COVID-19 have been confirmed so far, leading to at least 6 331 059 deaths worldwide (updated on 29 June 2022). The COVID-19 pandemic has caused extreme burdens on public health and the economy worldwide. The COVID-19 surveillance system of the Centers for Disease Control and Prevention (CDC) reports that pregnant women are at a higher risk of admission to an intensive care unit (ICU), requirement for mechanical ventilation and death following COVID-19 infection (Zambrano *et al.*, 2020). Therefore, pregnant or preconception women must exercise particular caution against getting infected with SARS-CoV-2.

Vaccination is the best way to prevent the spread of COVID-19. The completely vaccinated population globally is 46.5% by 8 December 2021 (Pettersson *et al.*, 2021). The completely vaccinated population in China has reached 77.3% (Pettersson *et al.*, 2021). Based on the present retrospective data, the vaccination rate in men receiving ART was ~80%, which is comparable to the national vaccination rate in China. However, misinformation about the vaccines has raised hesitancy regarding vaccination among some people, especially among women who are trying to conceive. Whether inactivated COVID-19 vaccination can lead to any adverse effect on conception in women is unclear. Therefore, women undergoing ART have showed a much lower vaccination rate of 33.2% (our present data).

Several cohorts or case-controlled studies have focused on the safety of COVID-19 mRNA vaccines in pregnant women (Bleicher *et al.*, 2021; Kharbanda *et al.*, 2021; Magnus *et al.*, 2021; Shimabukuro *et al.*, 2021; Trostle *et al.*, 2021; Wainstock *et al.*, 2021; Zauche *et al.*, 2021; Blakeway *et al.*, 2022; Rottenstreich *et al.*, 2022; Trostle *et al.*, 2022). Zauche *et al.* (2021) conducted a retrospective cohort study to determine the effect of COVID-19 mRNA vaccines on the risk of spontaneous pregnancy loss after 6 weeks of gestation. They concluded that women who received mRNA vaccines before conception or during pregnancy did not show any increased incidence of spontaneous pregnancy loss when compared with that in the previous year (Zauche *et al.*, 2021). Shimabukuro *et al.* (2021) analyzed the data obtained from the v-safe pregnancy registry, which included information on women who received COVID-19 mRNA vaccines during pregnancy or in the preconception period. Among the 3958 women enrolled in the study, 827 women completed the pregnancy. The rate of pregnancy loss was 13.9% (115/827) and the live birth rate was 86.1% (712/827). The incidence rate of adverse pregnancy was similar to those before the onset of the COVID-19 pandemic (Shimabukuro *et al.*, 2021). Two case-control studies showed that the rate of COVID-19 mRNA vaccinations was not significantly different in the groups of miscarriage when compared with that of the ongoing pregnancy group (Kharbanda *et al.*, 2021; Magnus *et al.*, 2021).

Inactivated vaccines have been widely used in China and have been proven safe in people aged >18 years (Jara *et al.*, 2021). However, the effectiveness and safety of inactivated COVID-19 vaccines have not been tested in pregnant women in almost all clinical trials. Moreover, studies on the effect of any COVID-19 vaccines on ART outcomes are extremely rare. To date, only four studies have investigated the effect of mRNA vaccines among women undergoing ART. Bentov *et al.* (2021) showed that vaccination by Pfizer BioNtech vaccines led to no detrimental effect on follicular growth. Orvieto *et al.* (2021) showed that the parameters of ovarian stimulation and embryo development in women were comparable before and after the administration of mRNA COVID-19 vaccination. However, both studies involved a limited number of patients and did not report pregnancy outcomes. A retrospective study by Aharon *et al.* (2022) reported that women who received COVID-19 mRNA vaccines did not exhibit any negative effect on the outcomes of ovarian stimulation and early pregnancy. Furthermore, in their retrospective study, Huang *et al.* (2022) found that the COVID-19 inactivated vaccines had no detrimental effect on either laboratory outcomes or pregnancy rates after IVF.

These studies have mainly focused on mRNA vaccines, and the only study on inactivated vaccines conducted so far did not report ongoing pregnancy rates. Therefore, our study aimed to investigate the effect of COVID-19 inactivated vaccines on IVF outcomes, especially ongoing pregnancies, with a larger sample size.

Materials and methods

Study design and patients

This retrospective cohort study was conducted at the Third Affiliated Hospital of Guangzhou Medical University and included patients who underwent fresh embryo transfer between 1 March 2021 and 15 September 2021. The exclusion criteria included a history of COVID infection and cycles with donor sperm or oocytes. The exposed group included women who were previously vaccinated before ovarian stimulation, while the control group included women who were not vaccinated before ovarian stimulation. The present research was approved by the Ethics Committee at the Third Affiliated Hospital of Guangzhou Medical University (Ethic no. (2021) 116). All personal information of the subjects was kept confidential during the study period.

Data extraction and processing

All data of patients were well recorded in the IVF data system, except for the vaccination information. In China, the vaccination information, including the vaccination date, name of vaccines and manufacturer, is recorded in an Application (APP, Suikang) developed by the Provincial CDC (Supplementary Fig. S1). All vaccinated individuals could obtain the vaccination information through the APP. The vaccination data were validated by nurses via telephone. During the follow-up, the nurse asked the patients to check the vaccination information through the APP. Patients who were still receiving IVF treatments were provided with questionnaires on vaccination when they visited the hospital. The assigned nurse ensured that the patient was referred to the APP and completed the questionnaire according to the data in the APP.

COVID-19 vaccination

The COVID-19 vaccines used in China are inactivated virus vaccines (CoronaVac, Sinovac Life Science Co, Ltd, Beijing, China and Sinopharm COVID-19, Beijing Institute of Biological Products Co., Ltd., Beijing, China). The entire course of vaccination includes three vaccine dosages. The interval between the first and second vaccine is at least 14 days, while the interval between the second and third is at least 6 months.

Ovarian stimulation and embryo transfer

In total, three protocols were employed for ovarian stimulation: antagonist protocol, agonist protocol and mild stimulation. For the antagonist protocol, the 150–300 IU recombinant FSH (Gonal-F, Merck Serono, S.p.A.) was administered on Days 2–3 of the cycle. The antagonist was started on Day 5 of ovarian stimulation and continued until the day of ovulation triggering. For the agonist protocol, the long-acting agonist (triptorelin acetate; Upson Biotech, Inc., France) was administered in the luteal phase of the previous cycle. Recombinant FSH (150–300 IU) was initiated 14 days after downregulation. For mild stimulation, 5 mg letrozole and 150 IU recombinant FSH were started on Days 2–3 of the cycle. The use of antagonists was the same as that for the antagonist protocol. In the case of at least two follicles sized $\geq$18 mm or three follicles sized $\geq$17 mm, 250 μg of recombinant hCG (Ovitrelle, Merck Serono) was administered for triggering, and oocyte retrieval was performed after 36 h. One or two cleavage-stage embryos or one blastocyst were transferred on Day 3 or 5 after oocyte retrieval. For the luteal support, 90-mg vaginal progesterone (Crinone, Merck Serono) was administered once daily on the day of oocyte retrieval and continued up to the 10th week of gestation if pregnancy was achieved.

Outcomes

The primary outcome was the ongoing pregnancy rate. The secondary outcomes included the number of oocytes retrieved, blastocysts per cycle, embryos suitable for transfer and rates of implantation, biochemical pregnancy, clinical pregnancy, biochemical pregnancy loss, ectopic pregnancy and early miscarriage.

Definitions

Ongoing pregnancy was defined as a pregnancy that continued beyond 12 weeks with a live fetus. An intrauterine gestational sac was identified by an ultrasound, and together with positive serum hCG was considered to indicate clinical pregnancy. Early miscarriage was defined as intrauterine pregnancy loss before 12 weeks of gestation, while biochemical pregnancy loss was defined as pregnancy loss before the gestational sac could be identified by ultrasonography. Embryos eligible for transfer or frozen were considered as embryos suitable for transfer.

Statistical analysis

The following aspects of the vaccinated versus unvaccinated group were compared: the baseline characteristics, ovarian stimulation parameters, embryo development and pregnancy rates. Statistical analysis was performed by using the SPSS version 22.0 software (IBM, Armonk, NY, USA). Quantitative variables with normal distribution and homogenous variance were expressed as mean ± SD, with the means compared by the Students' *t*-test. Quantitative variables with abnormal distribution or heterogeneous variance were expressed as median (1st and 3rd quartile), with the medians compared by Mann–Whitney *U*-test. Differences in the rates were compared by the Chi-squared test. When the expected count was $<$5 or the total sample size was $<$40, Fisher's exact test was performed to compare the differences in the rates. $P < 0.05$ was considered to indicate a statistical difference.

Multivariable logistic regression was performed to control for confounders of pregnancy rates (female age, male age, infertility duration, anti-Mullerian hormone (AMH), antral follicle count (AFC), type of infertility, the number and the day (D3 versus D5) of embryos transferred, BMI and the number of past oocyte retrieval). Multiple linear regression was applied to analyze the influence of COVID-19 vaccination on the number of oocytes retrieved, blastocysts developed and embryos suitable for transfer. Variables that may have an impact on the primary or secondary outcomes were included in the multivariable models.

Propensity score matching (PSM) was applied to screen a group of patients, such that the baseline parameters of the vaccinated group were quite similar to those of the unvaccinated group. The propensity score was calculated by using a multiple logistic regression model, with vaccinated versus unvaccinated serving as the dependent variable, and female age, AMH levels, total AFC, BMI, duration and types of infertility, stimulation protocol, starting dose of gonadotrophins, number of previous oocyte retrieval cycles, number of embryos transferred and the day of embryo transferred serving as independent variables. The PSM was conducted with a caliper width of 0.2 of the SD of the logit of the propensity score. The ratio of matching was 1:4 by closest neighbor matching. The SD for independent variables before and after PSM was calculated. An absolute value of SD $<$10% was considered to indicate a balance (Wu *et al.*, 2021).

Results

Vaccination status of couples with infertility

A total of 1781 fresh embryo transfer cycles between 1 March 2021 and 20 September 2021 were enrolled, among which 1588 cycles had vaccination information. A total of 527 (33.2%) women were vaccinated, whereas 1061 women were unvaccinated. Among the vaccinated

women, 240 were vaccinated before ovarian stimulation and 282 were vaccinated after IVF. A total of five women did not have the detail of the vaccination time. Therefore, the 240 women who were vaccinated before ovarian stimulation were included in the vaccinated group, whereas the remaining 1343 patients were included in the unvaccinated group (Fig. 1, Table I). Of the 240 women who were vaccinated before ovarian stimulation, 220 (91.7%) had received the second dose, 16 (6.7%) women had received the first dose and only 4 (1.7%) women had completed the full course of vaccinations before follow-up. A total of 80.7% (1281/1588) of the male partners were vaccinated, although the details of the vaccination time were unavailable (Table I).

Comparisons of baseline characteristics between the vaccinated and unvaccinated groups

The average age of the participants was 33.5 years (range: 21–46 years), and the BMI was 22.4 kg/m^2 (range: 14.4–38.6 kg/m^2).

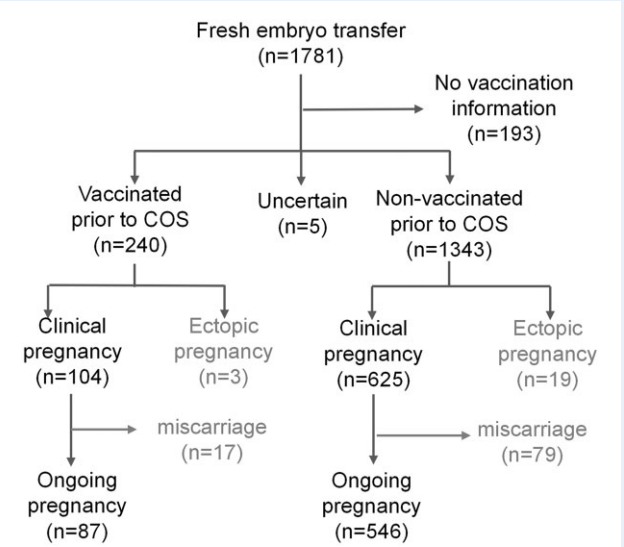

Figure 1. Flow chart. COS, controlled ovarian stimulation.

Table I Vaccination status of infertile couples.

	Female	Male
Vaccination % (n)		
Yes	33.2 (527/1588)	80.7 (1281/1588)
No	66.8 (1061/1588)	18.4 (292/1588)
Missing	NA	0.9 (15/1588)
Time of vaccination % (n)		
Before ovarian stimulation	45.5 (240/527)	NA
After IVF	53.5 (282/527)	
Uncertain	1.0 (5/527)	

The last day of follow-up was 31 October 2021.
NA, not available/applicable.

The baseline characteristics were comparable between the two groups, when considering their age, AMH, BMI, number of previous oocyte retrieval cycles, causes of infertility and types and duration of infertility. The median number of total AFC in the vaccinated group was significantly lower than that in the unvaccinated group (14.5 vs 16, $P = 0.009$). For the women vaccinated before ovarian stimulation, 27.5% had the last vaccination within 30 days, 38.4% within 31–60 days and 34.1% after 61 days (Table II).

Comparison of cycle characteristics of ovarian stimulation between the vaccinated and unvaccinated groups

The ovarian stimulation protocols and other ovarian stimulation parameters (i.e. starting dose, total dose, duration of gonadotrophin treatment and incidence of ovarian hyperstimulation syndrome) were similar between the vaccinated and the unvaccinated groups. The serum levels of estradiol and progesterone on the trigger day were also comparable between the two groups. However, the serum levels of LH on the trigger day were statistically higher in the vaccinated group when compared with that in the unvaccinated group (mean value: 1.41 IU/l vs 1.21 IU/l, $P = 0.024$), although this was probably clinically insignificant. The endometrial thickness (10.7 mm vs 10.6 mm, $P = 0.009$) on the trigger day was also statistically different between the two groups, which is also likely to be clinically insignificant (Table III).

Comparisons of oocytes retrieved and embryo development between the vaccinated and unvaccinated groups

As shown in Table III, the vaccinated group had notably lower numbers of dominant follicles (diameter $\geq$ 14 mm) on the trigger day when compared with the unvaccinated group (mean rank: 556 vs 622, $P = 0.017$). The vaccinated women had fewer oocytes retrieved (median: 8 vs 9, $P = 0.039$), blastocysts (mean rank: 737 vs 802, $P = 0.037$) and embryos suitable for transfer (mean rank: 737 vs 803, $P = 0.023$) when compared with those in the unvaccinated group (Table IV). Apart from the above-mentioned differences, the rates of fertilization and blastocyst development and types of fertilization were similar between the two groups. The proportion of single embryo transfer (57.1% vs 53.2%, $P = 0.241$) and blastocyst transfer (20.8% vs 26.7%, $P = 0.057$) were also comparable between the two groups (Table IV). The results of linear regression analysis revealed that the number of oocytes retrieved ($B = -0.299$, $P = 0.264$), embryos suitable for transfer ($B = -0.203$, $P = 0.127$) and blastocysts ($B = -0.250$, $P = 0.105$) was not associated with the status of vaccination before ovarian stimulation, after adjusting for the confounders (Supplementary Table SI).

Comparisons of pregnancy outcomes between the vaccinated and unvaccinated women

The ongoing pregnancy rate in the vaccinated group was not significantly lower than that in the unvaccinated group (36.3% vs 40.7%, $P = 0.199$) (adjust odd ratio (aOR) = 0.91, 95% CI = 0.68–1.22,

Table II Baseline characteristics of the vaccinated versus the unvaccinated group.

	Vaccinated	Unvaccinated	P
n	240	1343	
Female age (year)	33.8 ± 4.7	33.4 ± 4.7	0.200
Male age (year)	35 (32, 40)	35 (31, 38)	0.119
Infertility duration (year)	4 (3, 6)	4 (2.5, 6)	0.967
AMH (ng/ml)	2.65 (1.28, 4.00)	2.52 (1.49, 4.12)	0.750
Total AFC	14.5 (10, 19)	16 (11, 21)	0.009
BMI (kg/m^2)	22.4 ± 3.0	22.4 ± 3.2	0.933
No. of previous OPU cycles	0 (0, 0)	0 (0, 1)	0.555
Causes of infertility % (n)			0.088
Male	20.0 (48)	18.8 (252)	
Tubal factors	48.8 (117)	49.1 (660)	
Ovulatory disorder	8.8 (21)	7.8 (105)	
Endometriosis	4.2 (10)	3.0 (40)	
Unexplained infertility	5.4 (13)	11.2 (151)	
Mixed factors	12.8 (31)	10.1 (135)	
Type of infertility % (n)			0.152
Primary	45.8 (110)	50.9 (683)	
Secondary	54.2 (130)	49.1 (660)	
Interval between last vaccination and ovarian stimulation (day)		NA	
≤30	27.5 (66)		
31–60	38.4 (92)		
≥61	34.1 (82)		

AMH, anti-Mullerian hormone; AFC, antral follicle count; OPU, oocyte retrieval; NA, not available/applicable.

$P = 0.52$). The rates of implantation (34.4% vs 37.9%, $P = 0.217$), biochemical pregnancy (47.5% vs 52.6%, $P = 0.148$) and clinical pregnancy (43.3% vs 46.5%, $P = 0.359$) (aOR = 0.95, 95% CI = 0.71–1.27, $P = 0.72$) were not significantly different between the two groups. The rate of early miscarriage was similar in the vaccinated group when compared with that in the unvaccinated group (14.9% vs 11.2%, $P = 0.251$) (aOR = 1.36, 95% CI = 0.76–2.43, $P = 0.30$) (Table V and Fig. 2).

A total of seven women underwent two IVF cycles in the vaccinated group. Among them, two women received two doses of vaccines before ovarian stimulation and five women received two doses of vaccines between the two oocyte retrievals. In the unvaccinated group, 52 women underwent 2 IVF cycles. Even after eliminating the repeated cycles, the pregnancy outcomes were still not significantly different between the vaccinated and the unvaccinated groups (Supplementary Table SII).

PSM analyses

We applied the PSM to balance the baseline characteristics, with 239 vaccinated versus 928 unvaccinated women before ovarian stimulation. The results revealed that the baseline characteristics were well balanced after PSM, with the SD values <10% (Supplementary Table SIII). The parameters of ovarian stimulation, including ovarian stimulation protocols, starting dose, the total dose of gonadotrophins and days of stimulation and hormonal levels were comparable between the two groups after PSM (Supplementary Table SIV). The results of the PSM analysis indicated the absence of difference in the number of oocytes retrieved and embryological development, including the rates of fertilization and blastocyst development and the number of blastocysts, and embryos suitable for transfer (Table IV). No significant differences were detected in the rates of ongoing pregnancy (36.0% vs 39.9%, $P = 0.272$) and clinical pregnancy (44.4% vs 47.4%, $P = 0.398$) after PSM. Other outcomes, such as the rates of implantation, biochemical pregnancy and early miscarriage, were also similar between the two groups after PSM (Table V).

Discussion

To the best of our knowledge, the present study is the first one to focus on the ongoing pregnancy rate of IVF patients after vaccination with an inactivated COVID-19 vaccine. Our research revealed that vaccination before ovarian stimulation did not have any effects on IVF outcomes, including rates of ongoing pregnancy, clinical pregnancy and early pregnancy loss. COVID-19 vaccination also did not affect ovarian stimulation, the number of oocytes retrieved or the development of the embryos, including fertilization rate, blastocyst development rate, number of blastocysts and embryos suitable for transfer. These results suggested that the patients can be vaccinated before IVF because the vaccination would not have negative effects on the IVF outcomes.

Since its emergence at the end of 2019, the COVID-19 pandemic has become a global issue. The inactivated virus vaccines of COVID-19 (CoronaVac), which is the dominant type of COVID-19 vaccine in China, were efficient in preventing COVID-19 spread, showcasing 65.9% efficiency in preventing COVID-19 infection, 87.5% efficiency in avoiding hospitalization, 90.3% efficiency in preventing ICU admission and 86.3% efficiency in preventing deaths caused by COVID-19 (Jara *et al.*, 2021).

Pregnancy may enhance the severity of SARS-CoV-2 infection. As demonstrated by CDC in America, pregnant women with SARS-CoV-2 infection possessed a higher risk of a requirement for mechanical ventilation, ICU admission and death relative to that in infected non-pregnant women (Zambrano *et al.*, 2020). Moreover, 2.2% of infants of mothers infected with COVID-19 tested positive for the virus. The worsening symptoms of maternal infection may result in more adverse neonatal outcomes related to preterm birth (Angelidou *et al.*, 2021; Lv *et al.*, 2021). On the other hand, vaccination of the mother provided passive immunization to the fetus via the placenta (Shook *et al.*, 2021; Zdanowski and Wasniewski, 2021). Therefore, the preconception vaccination of women is particularly important.

However, a low vaccination rate was observed in preconception women in our reproductive medical center (33.2%) (15 October 2021). In contrast, the vaccination rate of their male partners was similar to that of the national data. Two main reasons may account for the low vaccination rate in women receiving ART. First, due to the limited evidence of the safety of COVID-19 vaccines on assisted reproductive outcomes, these women were hesitant regarding COVID-19 vaccination. Second, no such data were available suggesting the appropriate time interval between the COVID-19 vaccination and

Table III Ovarian stimulation of the vaccinated versus unvaccinated patients.

	Vaccinated	Unvaccinated	P
n	240	1343	
Ovarian stimulation protocols % (n)			0.203
Antagonist	61.3 (147)	61.6 (827)	
Agonist	27.5 (66)	30.5 (409)	
Mild stimulation	11.3 (27)	8.0 (107)	
Starting dose of Gn (IU)	150 (150, 225)	175 (150, 225)	0.834
Total dose of Gn (IU)	1950 (1425, 2700)	2025 (1500, 2700)	0.343
Days of stimulation	10 (9, 11)	10 (9, 12)	0.294
Serum level of LH on triggering day (IU/l)	1.41 (0.86, 2.58)	1.21 (0.79, 2.03)	0.024
Serum level of E_2 on triggering day(pmol/l)	7451 (4769, 10568)	7591 (4803, 10681)	0.772
Serum level of P on triggering day(nmol/l)	1.9 (1.1, 2.5)	1.9 (1.3, 2.7)	0.311
No. of follicles $\geq$14 mm on triggering day	7 (4, 9)	7 (5, 10)	0.017
Endometrial thickness (mm)	10.7 (9.4, 11.2)	10.6 (9.8, 11.6)	0.009
Endometrial type			0.692
A	44.6 (107)	44.9 (603)	
B	34.2 (82)	31.8 (427)	
Missing	21.3 (51)	23.3 (313)	
OHSS % (n)	1.7 (4)	2.4 (32)	0.493

Gn, gonadotropin; E_2, estradiol; P, progesterone, OHSS, ovarian hyperstimulation.

Table IV Embryo development of the vaccinated versus unvaccinated patients before and after propensity score matching (PSM).

	Before PSM			After PSM		
	Vaccinated	Unvaccinated	P	Vaccinated[c]	Unvaccinated[c]	P
n	240	1343		239	928	
Oocytes retrieved	8 (5, 12)	9 (6, 12)	0.039	8 (5, 12)	9 (5, 12)	0.244
Fertilization rate % (n)	79.87 (1647/2062)	80.18 (9927/12381)	0.748	80.0 (1642/2053)	79.7 (6623/8314)	0.747
Fertilization type			0.566			0.412
IVF	70.4 (169)	73.7 (990)		70.3 (168)	74.5 (691)	
ICSI	26.7 (64)	23.6 (317)		26.8 (64)	23.3 (216)	
IVF + ICSI	2.9 (7)	2.7 (36)		2.9 (7)	2.3 (21)	
Blastulation rate[a] % (n)	48.9 (466/953)	50.3 (2993/5953)	0.429	48.9 (465/951)	49.3 (1937/3927)	0.812
No. of blastocytes	1 (0, 3)	1 (0, 4)	0.037	1 (0, 3)	1 (0, 3)	0.229
No. of frozen embryos	1 (0, 3)	1 (0, 3)	0.051	1 (0, 3)	1 (0, 3)	0.255
No. of embryos suitable for transfer[b]	3 (2, 4)	3 (2, 5)	0.023	3 (2, 4)	3 (2, 4)	0.266
No. of embryos transferred			0.241			0.951
1	57.1 (137)	53.2 (7153)		56.9 (136)	56.7 (526)	
2	42.9 (103)	46.8 (628)		43.1 (103)	43.3 (402)	
Days of embryos transferred			0.057			0.439
3	79.2 (190)	73.3 (985)		79.1 (189)	76.7 (712)	
5	20.8 (50)	26.7 (358)		20.9 (50)	23.3 (216)	

[a]Blastulation rate = no. of II–VI blastocysts/no. of cleavage-stage embryos suitable for culture × 100%.
[b]No. of embryos suitable for transfer = no. of embryos transferred + no. of embryos frozen.
[c]The two groups were matched for female age, AMH levels, total AFC, BMI, duration and types of infertility, stimulation protocol, starting dose of Gn, no. of previous oocyte retrieval, no. of embryo transferred and the day of embryo transferred.
AMH, anti-Mullerian hormone; AFC, antral follicle count; Gn, gonadotropin.

Table V Pregnancy outcomes of the vaccinated versus unvaccinated patients before and after propensity score matching (PSM).

	Before PSM			After PSM		
Rates % (n)	**Vaccinated**	**Unvaccinated**	***P***	**Vaccinated**	**Unvaccinated**	***P***
n	240	1343		239	928	
Implantation	34.4 (118/343)	37.9 (747/1971)	0.217	35.4 (121/342)	38.3 (509/1330)	0.325
Biochemical pregnancy	47.5 (114/240)	52.6 (706/1343)	0.148	47.3 (113/239)	51.6 (479/928)	0.232
Clinical pregnancy	43.3 (104/240)	46.5 (625/1343)	0.359	44.4 (106/239)	47.4 (440/928)	0.398
Biochemical pregnancy loss	6.1 (7/114)	8.8 (62/706)	0.346	6.2 (7/113)	9.6 (39/479)	0.487
Early miscarriage	14.9 (17/114)	11.2 (79/706)	0.251	15.0 (17/113)	12.1 (58/479)	0.399
Ectopic pregnancy	2.6 (3/114)	2.7 (19/706)	0.971[a]	2.7 (3/113)	2.5 (12/479)	1.000[a]
Ongoing pregnancy	36.3 (87/240)	40.7 (546/1343)	0.199	36.0 (86/239)	39.9 (370/928)	0.272

[a]Fisher's exact test was used.

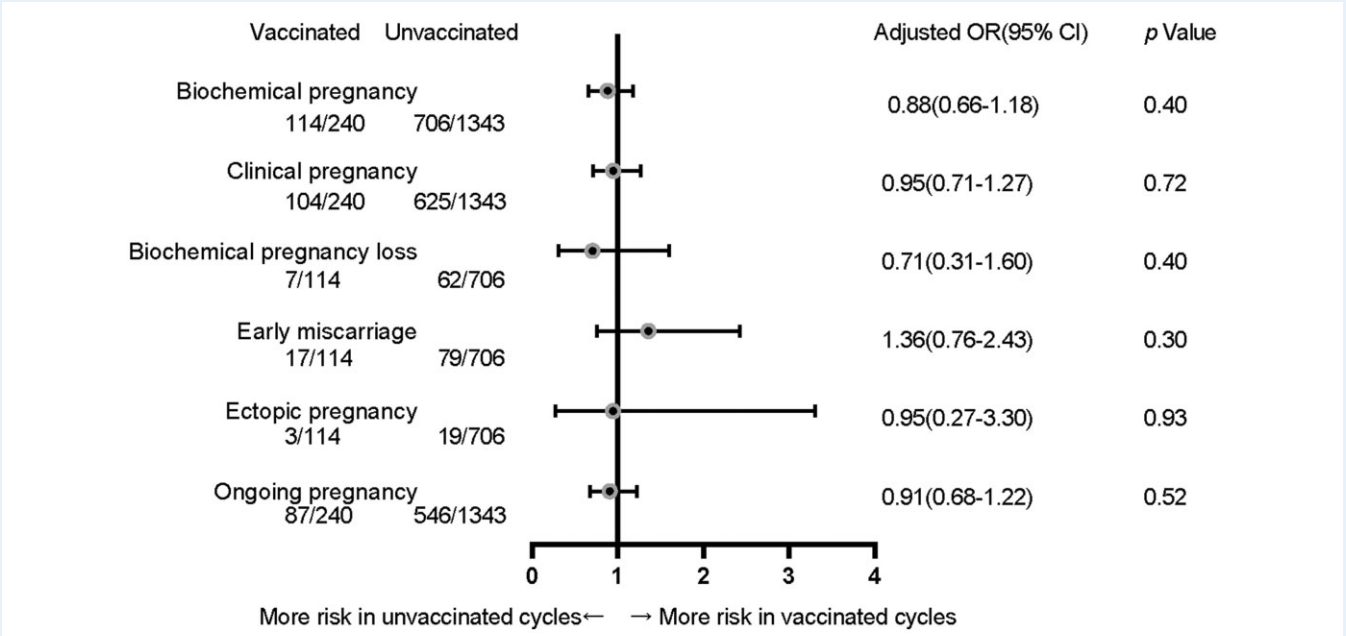

Figure 2. Logistic regression of the pregnancy outcomes in vaccinated women versus unvaccinated women. Adjusted for the age of female and male subjects, infertility duration, AMH, AFC, type of infertility, the number and the day (D3 versus D5) of embryos transferred, BMI and the number of previous oocyte retrievals. AMH, anti-Mullerian hormone; AFC, antral follicle count; OR, odds ratio.

conception. Women receiving ART would rather choose to defer vaccination than conception. The ASRM guideline declares that women who are receiving fertility treatment should be encouraged to receive the vaccination if found eligible (The ASRM Coronavirus/COVID-19 Task Force, 2020). However, the evidence of vaccination safety and its effect on IVF outcomes remains scarce. Orvieto *et al.* (2021) conducted a self-controlled study on 36 couples undergoing IVF treatment. They compared the characteristics of ovarian stimulation, the number of oocytes retrieved and sperm parameters in cycles before and after COVID-19 vaccination and found no differences between the aforementioned parameters (Orvieto *et al.*, 2021). Bentov *et al.* (2021) performed a cohort study in a COVID-19 infection group, COVID-19-vaccination group and control group. Ovarian function, as determined by the number of oocytes retrieved, serum and follicular fluid hormones were compared, and no compromise in the ovarian functions was detected after COVID-19 vaccination (Bentov *et al.*, 2021). However, the sample sizes of the aforementioned studies were small (36 and 9 for the vaccination group, respectively) and no reproductive outcomes were detected. In their retrospective analyses, Aharon *et al.* (2022) found that the

receipt of COVID-19 mRNA vaccines was not correlated with the fertilization rate ($\beta = 0.02 \pm 0.02$, $P = 20$), number of oocytes retrieved, blastulation rate or euploid embryo rate. For the 214 women in whom frozen embryos were transferred, the pregnancy outcomes, including ongoing pregnancy (aOR = 0.90, 95% CI = 0.61–1.31), biochemical pregnancy loss (aOR = 1.21, 95% CI = 0.69–2.14) and clinical pregnancy loss (aOR = 1.02, 95% CI = 0.51–2.06) were not significantly different from those of the unvaccinated women (Aharon *et al.*, 2022). Huang *et al.* (2022) also reported no significant differences in the outcomes of numbers of oocytes retrieved (9.9 ± 7.1 vs 9.9 ± 6.7; $P = 0.893$) and the rates of good-quality embryos (33.5 ± 29.8% vs 29.9 ± 28.6%; $P = 0.184$) and clinical pregnancy (59.1% vs 63.6%; $P = 0.507$) for patients with or without receipt of COVID-19 inactivated vaccines. Unfortunately, they did not report the outcomes of any ongoing pregnancy rates (Huang *et al.*, 2022).

Several strengths of the present study are worth mentioning. First, to the best of our knowledge, our study enrolled the largest sample size comparing the reproductive outcomes of vaccinated and unvaccinated women in the assisted reproduction medicine field. Furthermore, multivariable regression analysis and PSM were performed to control for confounders that may affect the outcomes. Moreover, although this is a retrospective study, with the help of electronic health databases, detailed and accurate vaccination information, including the vaccine manufacturer and administration date, and fertility treatment information can be obtained to ensure the reliability and authenticity of the retrospective results.

However, two limitations of the study should also be noted. First, due to the retrospective nature of the study, a couple of baseline clinical characteristics were not adequately balanced. Therefore, both multivariable regression analysis and PSM were employed to control for the effect of confounders. Second, due to the limited follow-up period, live birth outcomes and maternal and neonatal outcomes were not available in the present study, which will be further evaluated with an extended follow-up period.

Conclusions

The results of the present study indicated that the vaccination of women with inactivated COVID-19 vaccines before ovarian stimulation showed minimal effect on assisted reproductive outcomes. Women attempting ART should not postpone their COVID-19 vaccination because of their ovarian stimulation and embryo transfer schedules.

Supplementary data

Supplementary data are available at *Human Reproduction* online.

Data availability

The data underlying this article are available in the article and in its online supplementary material.

Authors' roles

Y.W. contributed to the conception and design of the study and the drafting of the article and was accountable for all aspects of the work. Y.L., C.A., L.L., Q.H. and Y.M. contributed to the follow-up of the patients. Z.X. and Z.L. contributed to the statistical analyses. M.C. and S.L. revised the manuscript. H.L. contributed to the conception and design of the study. J.L. was responsible for approval of the final version.

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Conflict of interest

None of the authors declare any conflict of interest.

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