Cellular and Molecular Biology

Journal homepage: www.cellmolbiol.org

Relationship between the Characteristics of Vaginal Secretion, Reproductive Immune Antibody and Infertility

Huabin Cao*, Lu Wang, Huanhuan Wu

Department of Reproductive Health, Jiangxi Maternal and Child Health Hospital, Nanchang, 330006, China

ARTICLE INFO

Original paper

Article history:
Received: September 18, 2021
Accepted: December 16, 2021
Published: February 28, 2022

Keywords:
reproductive immune antibody, infertility, anti-sperm antibody, vaginal secretion

ABSTRACT

Infertility is still a hot topic in the field of reproductive medicine. In this paper, the detection of vaginal secretions and the positive rate of serum reproductive immune antibody in infertile women and normal pregnant women were used to explore the correlation between infertility and the two, so as to provide an effective scientific basis for the diagnosis and treatment of infertility. In this paper, 80 infertile patients (experimental group) and normal pregnant women (control group) were selected for the experiment. Real-time PCR was used to detect vaginal secretion and enzyme-linked immunosorbent assay. The results showed that the positive rates of UU and CT were 42.5% and 40%, respectively. The positive rate of the control group was 8.8% (Uu) and 7.5% (CT). In addition, the positive rates of ANA and AsAb in the experimental group were 40% and 44%, respectively, and those in the control group were 7.5% and 8.8%, respectively, with a significant difference ($P<0.01$). The correlation coefficients between the two antibodies and the occurrence of infertility were 0.501 (ANA) and 0.663 (AsAb), and there was a correlation ($P<0.05$).

DOI: http://dx.doi.org/10.14715/cmb/2022.68.2.18

Introduction

Infertility refers to having a normal couple's life and not being pregnant for one year without contraception. The incidence rate of infertility varies in different places. The incidence rate of infertility in China is 8%~11%, of which l2%~18% of patients do not find out the cause. Infertility also affects family happiness, women's health and social harmony. There are many induced factors of infertility. The pathogenesis and related factors of infertility are scattered in China. In addition, the epidemiological survey of infertility is also rare, so the prevalence rate has not been accurate statistical data. If we treat the patients without knowing the cause of the disease, there is no way to achieve a considerable effect. Therefore, it is of far-reaching significance to study the correlation between vaginal secretions and reproductive immune antibodies and infertility, so as to provide a strong basis for the examination and treatment of infertility.

In recent years, more and more people pay close attention to the direct relationship between immune factors and infertility. Among them, 11% - 16% of infertility patients are caused by immune factors. In order to explore the correlation between reproductive immune antibodies and infertility, Lao selected 15 infertile patients as the experiment, and 15 normal childbirth women as the control group (1-2). During the experiment, the reproductive immune antibody in the serum of the experimenter was collected for detection and analysis of its influence (3-4). The results of this study showed that the positive rate of reproductive immune antibodies in the experimental group was significantly higher than that in the control group ($P<0.05$) (5-6). From this study, we can know that the reproductive immune antibody has a very important impact on the detection of infertility patients, and has a certain reference value for the development of related clinical research (7-8). Such a research method is novel and feasible, but due to the limitations of the samples, the research results have a certain contingency.

Many factors cause infertility. For patients with infertility, clinicians need to have a rigorous attitude, carefully monitor the change and development of their condition, try to find out the cause of the disease, and take the right treatment. Many scholars have researched this. For example, Zhang explored the

*Corresponding author. E-mail: shenqiaojiu61135@163.com

correlation between vaginal infection and infertility (9-10). Zhang randomly selected 50 infertile patients (experimental group) and normal pregnant women (control group) and set up a control group to test, analyze and compare the detection results of UU and CT (11-12). The results of this study showed that the CT infection rate of the experimental group was 33.63%, the UU infection rate was 41.15%, the combined infection rate was 11.53%, the control group was 4.22%, 6.32% and 2.21%, the difference was significant (P<0.05) (13). From this study, we can know that vaginal infection is a pathogenic factor of infertility (14). Generally speaking, Zhang's research has a certain reference value, but its reliability is low, and it still has a long way to go in the research of infertility.

Infertility is common in women's diseases at present. With the improvement in people's living quality, the incidence rate of infertility is increasing. There are many pathogenic mechanisms of infertility, and many scholars have done related research, scholars generally think that the immune factor is an important cause of infertility, in order to study the correlation between vaginal secretion and reproductive immune antibodies and infertility, this paper set up a control experiment to study. The results showed that the positive rates of UU and CT in the experimental group were 42.5% and 40% respectively, while those in the control group were 8.8% and 7.5% respectively. It can be seen that the positive rates in the experimental group were higher than those in the control group, and the differences were statistically significant. Moreover, the correlation coefficients between UU and CT and infertility were 0.608 and 0.513, respectively (P<0.05). In addition, the positive rates of ANA and AsAb in the experimental group were 40% and 45%, respectively, and those in the control group were 7.5% and 8.8%, respectively, with significant differences (P<0.01). The content of the two antibodies in the experimental group was higher than that in the control group, and the difference was significant. The correlation coefficients were 0.501 and 0.663, respectively. Therefore, there was a correlation between the reproductive immune antibody and the occurrence of infertility (P<0.05).

Materials and methods

Infertility

Recently, with the rapid development of the economy, people's pace of life is faster and faster. The factors of living environment pollution, diet type and structure change, the globalization of sexual diseases, and the change of outlook on life and childbearing of young people and so on have caused a rapid increase in the incidence rate of infertility. (15-16). In clinical medicine, infertility is different from other diseases. Both physiological and psychological factors coexist, affecting the harmony of patients' families and social stability (17). According to the statistics and analysis of the World Health Organization, infertility has become the third-largest disease, after cardiovascular and cerebrovascular diseases and tumor diseases. Currently, infertility has affected the physical and mental health and family harmony of infertility patients (18-19). The most common causes of infertility are vaginal infection and reproductive immune antibodies.

Vaginal Infection

At present, 16% - 22% of infertility occur every year in the world, of which vagina infection accounts for 18% - 62% of total infertility. Studies have shown that UU and CT are the main pathogens of infertility, among which UU and CT lead to an increased incidence of uterovaginal infection (20-21).

UU belongs to one of the ureaplasma, which needs urea to grow. UU is mainly transmitted through sexual life, so young people are more common. In addition, people who have unclean sexual intercourse are also more common. When the vagina is inflamed, its surface mucous membrane is damaged, and UU enters while it is empty, which leads to vagina infection. After UU infection, the symptoms of patients are not obvious, it is difficult to be found, and at the same time, it will make doctors misjudge and miss the diagnosis. It can be seen that the important reason for infertility caused by UU infection is the pathological changes in female reproductive organs (22).

CT is the main pathogen of nongonococcal urethritis. Once the infection occurs, abnormal vaginal secretions will appear, and severe cases will also have bleeding after sexual intercourse or non-menstrual period. Frequent urination, dysuria and urgency are common symptoms of urethritis. If the urethra

infection of CT is not treated, there will be lower abdominal pain and sexual contact pain and other symptoms, which can cause pelvic inflammation in serious cases. Long-term continuous infection can lead to infertility, abortion and chronic abdominal pain. CT infection in the reproductive tract of pregnant women also increases the risk of premature delivery, low birth weight and premature rupture of membranes. If the treatment is not timely and effective, it may also infect the newborn baby, resulting in pneumonia and ophthalmia of the newborn baby.

Reproductive Immune Antibody

Reproductive immunity is a unique immune response system. It prevents the body's immune system from rejecting sperm, eggs, fertilized eggs and embryos to ensure the success of the entire pregnancy. Any abnormality may lead to infertility (23). Studies have shown that with the detection of serum lecithin in male and female infertility patients, immune infertility has gradually attracted people's attention, and has become a hot topic in infertility research. Reproductive immune antibodies can affect many aspects of normal fertility through various mechanisms, leading to infertility.

Anti-sperm antibodies can be produced under certain inducements, the most common one is vaginal infection or damage to reproductive tract mucosa. In this condition, sperm, as an antigen, stimulates the body to produce T cell immunity, thus inducing the production of anti-sperm antibodies. It mainly affects the immune mechanism of sperm.

The production of sperm antibodies must be based on specific motivation, the most common of which is a vaginal infection or genital mucosal damage. In this case, spermatozoa act as antigens, stimulating the body to produce T cell immunity, thus producing antibodies against spermatozoa (24). This is a major immune mechanism that affects sperm. Sperm antibodies can be divided into three subtypes: IgM, IgA and IgG. IgM is the main factor to reduce the fertility rate, and IgA is the second. The results showed that IgM blocked the position of endosperm and blocked the release of the enzyme system to the acrosome, then stopped the acrosome reaction. In addition, IgM may also affect sperm viability. It hinders the ability of sperm and leads to infertility, mainly due to the role of dissolving cells and promoting sperm aggregation. IgG can promote the connection between sperm and macrophage or neutrophil receptor (FC) by combining with sperm, and then play a role in the pathogenesis of pregnancy.

As a common female reproductive immune antibody, Ana affects the cell division of the fertilized egg by combining with the nuclear components, resulting in pregnancy failure (25). Previous reports have shown that the antigen level on the surface of the membrane of early fertilized eggs is very low. Among the multiple divisions of fertilized eggs, Ana, whose main target is the nuclear component, is the most likely immune factor leading to the failure of embryo transfer. Because of this important reason, in vivo, ANA has become a hot research topic.

Experiment Preparation

Subjects: 80 infertile women (without other diseases), aged between 23-34 years old, in addition to 80 normal pregnant women, of the same age. There was no significant difference between the two groups ($P > 0.05$). Main instruments and reagents: centrifuge, enzyme labeling instrument, pipette, burette, 86-hole enzyme labeling plate, standard, lysate, chloroform, anhydrous ethanol, etc.

Experimental Group and Control Experimental Setup

In this paper, 80 infertile women and normal pregnant women were selected respectively, and then a control group was set up for the experiment.

Real-Time PCR Test

Pretreatment of the experiment: put the vaginal secretions collected from the experimenter into a sterile test tube, then add 1 ml of sterile physiological saline, shake the test tube fully to mix the two components, and immediately transfer all the mixed solution to a sterile centrifuge tube of 1.5 ml, place it in a centrifuge for centrifugation, remove the supernatant and retain the solid precipitation, add 1 ml of sterile normal saline to the sediment with a rubber head dropper, repeat the above experiment, add 30 μl of DNA extract to the sediment, shake it well, stand at 100 ℃ (± 2 ℃) for 10 minutes, and then centrifuge it for 6 minutes, and keep it for standby. For the quality control standard and quantitative positive standard,

the positive judgment value DNA > 50 copies/ml is positive.

Detection of UU and CT: add the experimental quality control materials to the samples, put the samples that have been added to the experimental quality control materials into the 86-hole sample tank, use the instrument correctly in strict accordance with the operating instructions of the instrument, and ensure the accuracy of the data. Establish a standard curve (linearity requirement is more than 0.995) and test it. The instrument will automatically analyze data and record results.

Enzyme-Linked Immunosorbent Assay

First, 5ml of the blood of the experimenter was collected, then the serum was thawed and naturally cooled to room temperature, and the sample was diluted according to a certain proportion and the washing solution was prepared, which was kept for future use. Establish a new standard curve: set 10 standard holes, add 120 μl sample diluent to each hole, and then add 120 μl standard to the first hole, mix it well, and then use the pipette gun to suck out 120 μl standard and put it into the second hole. Repeat the above steps to the seventh hole, and finally use the pipette gun to suck out 120 μl in the seventh hole and discard it, so as to ensure that the volume of each hole remains the same, The eighth hole is set as blank control; take 120 μl of sample to be tested with pipette gun and add it to the eight reaction holes, mix and seal the holes, incubate in 37 ℃ (± 2) incubator for 1.5h, wash the plates for 6 times and dry them, then add 60 μl of first antibody working solution to each hole, mix and seal the holes, incubate in 37 ℃ (± 2) incubator for 1h; repeat the above steps twice, Finally, according to the operation instruction of the instrument, set up the procedure of enzyme labeling instrument, measure the absorbance value of each sample at the wavelength of 492nm, and calculate the content of antibody in the sample according to the standard curve.

Statistical Treatment

In order to better analyze the experimental data, SPSS 21.0 was used to analyze the data, P<0.05 was statistically significant. Pearson was used to analyze the correlation between the data. Pearson value > 0 indicates positive correlation, Pearson value < 0 indicates negative correlation, P<0.05 indicates significant correlation.

Results and discussion

Real-Time PCR Results

The standard curve of UU and CT positive standards was established, and then the quantitative analysis of each sample was carried out. The results are shown in Table 1 and Figure 1.

Table 1. Analysis of quantitative results of UU and CT

Group	Number	UU Number	UU Positive rate	CT Number	CT Positive rate
Infertility group	80	34	42.5%	32	40%
Control group	80	7	8.8%	6	7.5%
X2	-	13.29		12.92	
P	-	P<0.05		P<0.05	

Figure 1. Analysis of quantitative results of UU and CT

From Table 1 and Figure 1, we can see that the positive number of UU in the experimental group is 34, the positive rate is 42.5%, the positive number of CT in the experimenter is 32, the positive rate is 40%, the positive number of UU in the control group is 7, the positive rate is 8.8%, the positive number of CT in the control group is 6, the positive rate is 7.5%. The positive rates of UU and CT in the two groups were statistically significant (P<0.005).

The correlation analysis of each group of experimental results is shown in Figure 2.

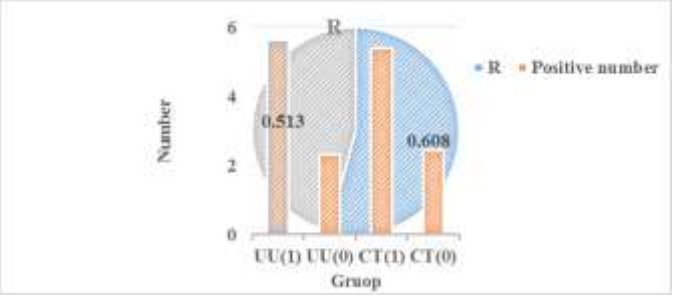

Figure 2. Analysis of the correlation between UU and CT and infertility

According to Figure 2, the correlation coefficients between UU, CT and infertility are 0.608 and 0.513 respectively, with statistical significance (P<0.005). The positive quantitative results of UU in the experimental group were 5.62±1.02, 2.31±1.09 in the control group, 5.39±0.33 in the experimental group and 2.45±0.63 in the control group. The difference between the two groups was statistically significant (P<0.05). The quantitative data of UU and CT in the experimental group were higher than that in the control group.

Detection Results of Antinuclear Antibody and Antisperm Antibody in Serum

The results of ANA and AsAb in serum are shown in Figure 3.

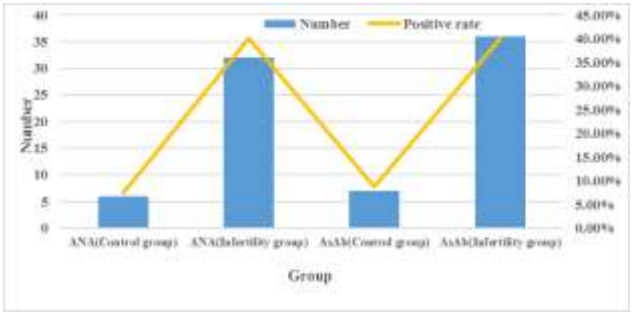

Figure 3. Analysis of ANA and AsAb in serum

According to Figure 3, the positive rates of ANA and AsAb in the experimental group were 40% and 45% respectively, and those in the control group were 7.5% and 8.8% respectively. There was a significant difference between the control group and the infertile patients (P<0.01). The antibody content in the blood samples of infertile patients was higher than that of the normal control group, and the difference was significant.

The correlation analysis between the content of antibodies in the patient's serum and the occurrence of infertility is shown in Figure 4.

According to Figure 4, the correlation coefficient (R-value) between anti-nuclear antibody and anti-sperm antibody and infertility is 0.501 and 0.663, respectively. There was a correlation (P<0.05). The content of ANA and AsAb in the blood of the experimental group was 2.37 ± 0.29 and 2.49 ± 0.44, respectively. The content of ANA and AsAb in the serum of the control group was 1.02 ± 0.32 and 1.24±0.29, respectively. The difference between the two groups was significant and statistically significant (P<0.05). The quantitative data of UU and CT in the experimental group were higher than that in the control group.

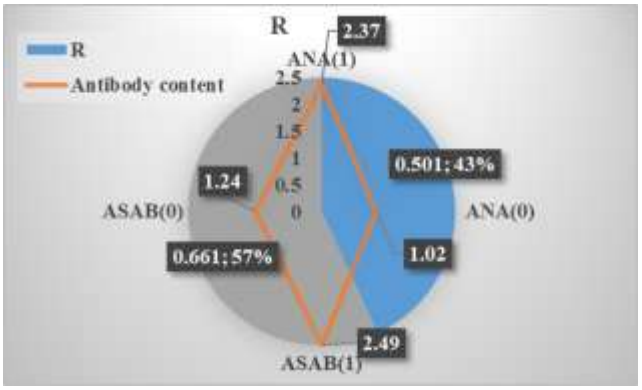

Figure 4. Correlation analysis of ANA and AsAb in serum

As we all know, infertility has been affecting the family happiness and social stability of patients in recent years, and the number of infertile women is also increasing year by year. As society pays more and more attention to the cause of infertility and the research goes deeper and deeper, the research results on infertility have achieved good results. Of course, vaginal infection and reproductive immune antibodies are still hot spots in medical research. Therefore, this paper studies the relationship between vaginal secretions, reproductive immune antibodies and infertility.

Relevant medical research shows that there is a close relationship between reproductive immune antibodies and vaginal infection and the occurrence of infertility. Therefore, we need to understand the pathogenesis of infertility in time, so that clinicians can better understand the symptoms of patients, improve the treatment effect of infertility patients as much as possible, and finally achieve the purpose of reducing the incidence of infertility.

In this paper, by setting up a control experiment, the relationship between vaginal secretions and reproductive immune antibodies and infertility was studied. The results show that Urea-plasma urealyticum (Uu) and Chlamydia trachomatis (CT) in vaginal infection may be the causes of infertility; the factors of female reproductive immune antibodies, such as an anti-nuclear antibody (ANA) and anti-sperm antibody (AsAb), may also lead to infertility;

the screening of vaginal infection and reproductive immune antibody is carried out at the same time, It can more accurately judge the occurrence of infertility, and provide an effective scientific basis for the diagnosis and treatment of infertility.

Acknowledgments

None.

Conflict interest

The authors declare no conflict of interest.

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