Pro Index | Journal of Science in Medicine and Life
Volume: 3 Issue: 5 Year: 2025
ISSN: 2992-9202 | http://journals.proindex.uz

Biochemical Study of Oxidative Stress and its Role in Recurrent Pregnancy Loss

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Abstract: Background: Abortion is considered one of the important medical and social problems in the life of the couple in particular and society in general. In addition to the health risks it causes to the woman, it causes psychological, economic and family problems for the aborted women, and it is one of the factors affecting women of childbearing age. This study aimed to evaluate key biochemical and immunological markers in - Identify some of the causes of recurrent miscarriage in women The link between reactive stress and having multiple miscarriages. Ways: Ninety samples were put through a metabolic test. The amount of total reactive oxygen species in the blood was found using an ELISA reader from Germany and an immune method called Enzyme-Linked Immuno-Sorbent Assay. Before they are used, all materials and chemicals must be brought to room temperature. The chemistry tests were done in the Biology Department's labs at the University of Kufa's Faculty of Sciences. Results: The results show that abortion women are of different ages, as shown in Figure (1). There are 4 abortion women younger than 20 years, 32 abortion women younger than 30 years, and 24 abortion women younger than 40 years. There was a significant rise ($p<0.05$) in the level of reactive oxygen species in patients (3.285 ± 0.1225) compared to the control group (1.155±0.09705). There were no significant differences ($p<0.05$) in the levels of reactive oxygen species in abortion women based on their age. less than 20 years (3.328±0.3904), less than 30 years (3.168±0.1756), and less than 40 years (3.433±0.1895). Reactive oxygen species (ROS) levels were much higher in the women who had miscarriages than in the control group. This suggests that oxidative stress may play a part in the biology of miscarriage. However, ROS levels were not significantly different between women of different ages.

Keywords: ROS, women miscarried.

Introduction

About 1% to 2% of women of childbearing age have a tough reproductive problem called recurrent pregnancy loss (RPL). This is when they have two or more unplanned miscarriages in a row before 20 weeks of pregnancy. About half of the time, the cause of RPL is still unknown, even after a lot of study. This kind of RPL is often called unexplained. Genetic mistakes, uterine flaws, hormonal issues, thrombophilias, and inflammatory diseases have been thought to be the main reasons of unexplained RPL in the past. Now, though, there is more proof that oxidative stress is a major cause of this condition(3,4).

The body's antioxidant protection systems are out of balance when reactive oxygen species (ROS) are made too much. This is called oxidative stress. In a healthy body, ROS help cells talk to each other and protect the immune system(7,8). However, too many ROS can damage lipids, proteins, and DNA through oxidative damage, which can throw off the balance of cellular homeostasis(9, 10). Redox balance must be carefully controlled during pregnancy for fertilization, placental development, and baby growth to go as planned(11,12). Too much reactive stress can damage trophoblasts, cause cell death, and cause inflammatory reactions, which could end the pregnancy(13,14).

As a result of several studies, it has been found that women with RPL have lower levels of antioxidants such as glutathione peroxidase (GPx) and higher levels of reactive markers such as malondialdehyde (MDA) and 8-hydroxy-2′-deoxyguanosine (8-OHdG)(15, 16). It seems that oxidative stress may play a role in the development of RPL, especially when there is no clear cause (17,18).

This study compares the levels of oxidative stress and antioxidant defense systems in women who have lost multiple pregnancies to healthy pregnant and non-pregnant controls(19,20). This is done because an imbalance in oxidative stress may make it harder for a woman to get pregnant. Learning about the antioxidant profile of RPL patients could help doctors find new ways to diagnose the condition and find better ways to treat it, which would ultimately help women with this condition have healthier pregnancies(21,22).

Materials and methods

During the months of November 2024 and March 2025, this study was carried out in the labs of the University of Kufa's Department of Biology and the Al-Amin Center in Najaf Governorate, which is part of the Ministry of Health in Iraq. At Al-Zahraa Teaching Hospital, blood samples were taken from both the control group, which was made up of healthy pregnant women, and from women who had abortions. For women who had an abortion, the average age was (28.18 ± 0.77) years, and the range of ages was from 18 to 39 years.

Methods

Ninety samples were put through a metabolic test. The amount of total reactive oxygen species in the blood was found using an ELISA reader from Germany and an immune method called Enzyme-Linked Immuno-Sorbent Assay. Before they are used, all materials and chemicals must be brought to room temperature. The chemistry tests were done in the Biology Department's labs at the University of Kufa's Faculty of Sciences.

Statistical Analysis

The statistical analyses were done by using Spss (V. 20) to find the mean and standard error (± SE). Completely randomized design (CRD) and least significant differences (L.S.D) tests were used to test the results at a level ($P < 0.05$) to show the significance of the results.

Results

Women with CMV (Cytomegalovirus), Toxoplasma gondii, and Rubella IgM were excluded and reactive oxygen species were measured.

The result show different age in abortion women according to the age as showed in the figure(1) ,which show (4) abortion women less than 20 year ,(32) abortion women less than 30 year and (24) abortion women less than 40 year, as shown in figure (1) .

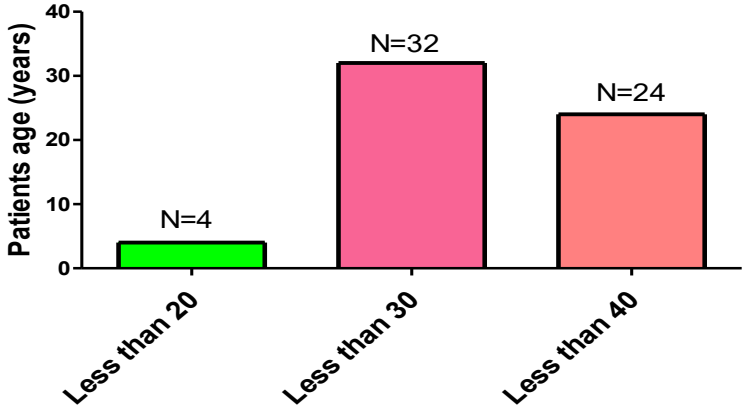
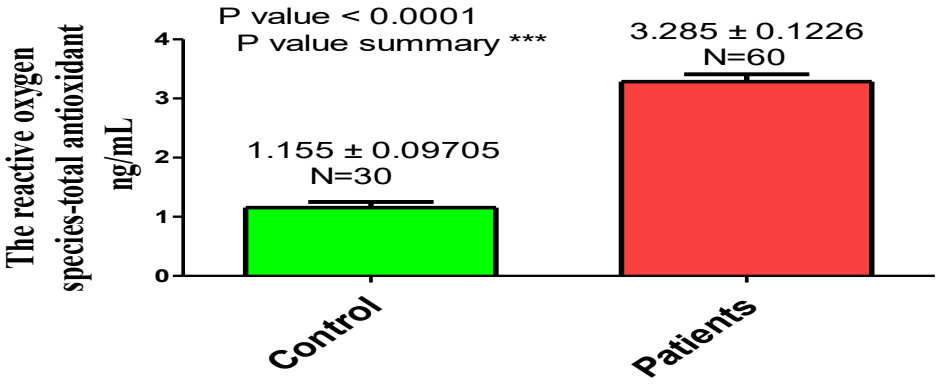


Figure (1) show number of patient according to age

The results showed a significant increase ($p<0.05$) the reactive oxygen species level in patients (3.285 ± 0.1226) compared with control group (1.155±0.09705), as shown in figure (2).



Figure(2)show The effect of the reactive oxygen species-total anti-oxidant on abortion women compared with control group

The results showed non-significant different ($p<0.05$) of the reactive oxygen species to abortion women according to their age . less than 20 year(3.328± 0.3904) ,less than 30 year (3.168±0.1756) and less than 40 year(3.433±0.1895), as shown in figure (3).

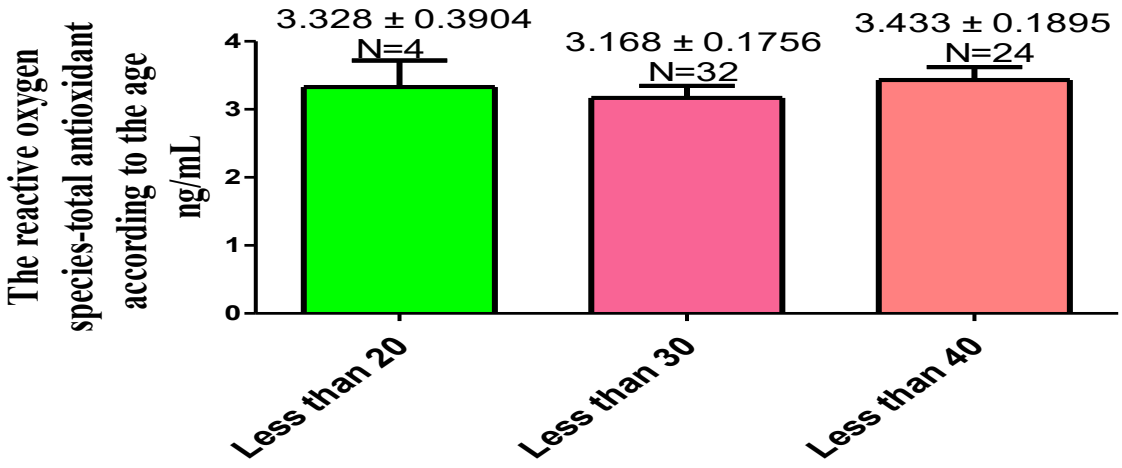


Figure (3) shown The effect of the reactive oxygen species-total anti oxidant on abortion women according to the age

Discussion

There were significantly more reactive oxygen species ($p<0.05$) in women who had abortions than in women who did not have abortions. It was found that women of all ages who had abortions had reactive oxygen species, but there was no strong difference ($p<0.05$) between the groups. Because it boosts reactive stress, abortion is an illness in and of itself, especially in the early stages of sexual life. This could be the reason. When there is something wrong with the baby, the placenta may not be fully made enough to provide air and nutrients. Lack of oxygen, or hypoxia, can happen because of this, and it can lead to a loss in the end(23,24).

Also, some abortions are caused by or happen at the same time as pain or inflammation. This makes defense cells work, and to protect themselves, they release ROS (25,26).

To compare the uterine tissue of women who have had multiple spontaneous abortions (RSA) to that of women who are not pregnant, the study set out to find molecular changes and specific pathways. Twenty-five people with RSA and twenty-five healthy pregnant women were picked for the study. We used HPLC-MS (high-performance liquid chromatography paired with mass spectrometry) to do a study of uterine tissue's raw metabolome. To find molecules that were different, we used PCA and OPLS-DA, which stands for orthogonal partial least squares

discriminant analysis. There is a library called KEGG that was used to find biological pathways that are related. This is called pathway enrichment analysis. To find out how much of these important proteins were present, a method known as quantitative polymerase chain reaction (qPCR) was used. Indoleamine 2,3-dioxygenase (IDO), long-chain acyl-CoA synthetase 4 (ACSL4), and glutathione peroxidase 4 (GPX4) are some of these proteins. The study found that a big increase in ROS might lead to more losses (27,28).

Endometrial biopsies were used to see if the following bacterial and viral groups could be to blame for miscarriages in women: Chlamydia trachomatis, Ureaplasma urealyticum/parvum, Mycoplasma hominis/genitalium, Gardnerella vaginalis, herpes simplex virus type 1/2, Epstein-Barr virus, cytomegalovirus, varicella zoster virus, human herpesvirus type 6, human herpesvirus type 7, and human herpesvirus type 8. In 65 women who had an early or late loss in the past, we looked at the anaerobic and aerobic germs that cause bacterial imbalance and uterine bacterial illness because of difference in growth. The amount of reactive oxygen species rose significantly ($p<0.05$) in women who had abortions, as shown by DNA extraction and quantitative polymerase chain reaction (qPCR) (29,30).

Over the course of a study, oxidative stress markers were measured in pregnant women who had an early loss and pregnant women who were healthy. As cases and controls, the study used 32 women who had an early loss and 32 healthy pregnant women in the first trimester. The women were all the same age, race, and gender. We measured bile, uric acid, malondialdehyde (MDA), and total antioxidant capacity (TAC) in the blood of both groups. Women and men in the two groups were the same age, the pregnancies were the same length, and the subjects' body mass index (BMI) was the same. Women who had an abortion on their own had significantly higher levels of MDA (4.35±1.47 µmol/L vs. 3.42±1.68 µmol/L; P=0.026), and women who had an abortion had significantly lower levels of total uric acid (TAC) (552.34±212.79 U/mL vs. 1003.23±1168.68 U/mL; P=0.040). Both groups had the same amount of bile and uric acid. The study found that having a higher overall survival rate has an effect on the number of unexpected abortions that happen early in the first trimester of pregnancy. Having a lot of MDA in your blood and not much total uric acid (TAC) can make you more likely to have an abortion by yourself while you are pregnant. It's very important for spontaneous abortion to happen that there is a lot of MDA in the body (31,32). Since the ROS level went up in all of the women who had miscarriages, which led to more miscarriages, the results of these three other studies may be similar to ours.

Conclusion:

Compared to the control group, the women who had miscarriages had much higher amounts of reactive oxygen species (ROS). This suggests that oxidative stress may play a part in how miscarriages happen. However, ROS levels were not significantly different between women of different ages.

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