Studying the Effect of Cholesterol and Activin a Hormone on Inducing Abortion in Pregnant Women

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ABSTRACT

The current study aimed to show the relationship between the levels of (activin-A hormone) and the concentration of lipids (cholesterol) and the occurrence of some cases of abortion and threatened pregnancy. The study included 150 blood samples taken from aborted women and women with a threatened pregnancy, which is diagnosed by ultrasound scan and other methods such as clinical examinations and the history of the pregnant woman. Their ages ranged between (16-45) who visited Tikrit Teaching Hospital and private outpatient clinics. It was divided into three groups: The first included 50 blood samples from aborted women, 20 blood samples from women with a threatened pregnancy, and 20 blood samples from women with normal pregnancies as a control group. Blood samples were taken during the first trimester of pregnancy. The results of this study showed a significant increase in the concentration of (activin-A hormone) at a significant level (P ≤ 0.05) in the aborted women group and women threatened with abortion compared with the control group, as well as a significant increase in the level of lipids (cholesterol) at a significant level (P ≤ 0.05) in the abortion group and the group of women threatened with abortion compared with the control group.

Keywords- abortion, cholesterol, activin hormone.

I. INTRODUCTION

Abortion is defined as the early outflow of all or part of the contents of the uterus, before the 24th week of pregnancy as before the fetus is able to survive outside the uterus. Abortion is a natural attempt to preserve the future generations to be normal and to try to get rid of deformed fetuses at an early date (Magawan, 2001). Abortion most often occurs during the early stages of pregnancy during the first trimester and may occur during the second trimester of pregnancy (Chamberlain, 1996). The causes of abortion remain somewhat unknown, especially the type of spontaneous abortion, at a rate of (24-60%) of all abortions, despite the involvement of many factors in its occurrence, such as anatomical, genetic and secretory factors (Wouters et al., 1993; Hill, 1998) Activin hormone: It is a complex glycoprotein hormone with a molecular structure of (25,000) thousand daltons and its role is to stimulate the secretion of pituitary FSH and this hormone belongs to the family (TGF B), (Vale et al., 1986. Loveland et al., 1996) Produced by the placenta and fetal amniotic fluid. Recently, it was suggested that this hormone may have a role in human parturition. It is a glycoprotein belonging to the TGF-B superfamily, a structurally similar group, but in practice the diverse growth rates are involved in cellular proliferation, differentiation, and cell fate design. During pregnancy, the placenta is the main source of this hormone (Petraglia et al., 1987).

Measurement of the level of ACTIVES A at the beginning of pregnancy is useful in diagnosing trophoblastic dysfunction and also helps in controlling early pregnancy problems to predict the outcome of the first trimester of pregnancy in women who experience...
vaginal bleeding during pregnancy leading to a threatened abortion. [Florio et al., 2002]

II. MATERIALS AND METHODS

2.1 study sample:

This study was conducted in Tikrit Teaching Hospital and one of the women's outpatient clinics in the city of Tikrit for the period from July (2010) to January (2011). The study included (150) A woman who has signs or symptoms of abortion of all kinds was diagnosed by performing an ultrasound scan to determine the nature of the abortion, and their ages ranged between (16-45) years. The studied samples were divided into three groups:

1. The first group: included women with normal pregnancies (3-4 months) (control group).
2. The second group: included women threatened with abortion (3-4 months)
3. The third group: included women who had aborted (3-4 months)

2. Measurement of the concentration of activin A hormone in the blood serum:

2.1.1 Test principle:

The basis of the hormone's action is an enzymatic reaction that includes the presence of a microtiter plate equipped in this kit covered with an antibody specific to ACV-A. Then the samples, standard or control are added to the microplate well with an associated enzyme (HRP) that reacts with the hormone-specific antibody And incubated at 37C. Then the solution of the substrates A and B is added to all the wells, after which the excess of the bond, which did not react, is removed. Only these wells that contain the hormone and HRP, which will have a change in color, the enzyme-base reaction ends with the addition of the stop solution and the color change is measured by a spectrophotometer at a wavelength (450nm).

2.1.2 Prepare the solution:

1. Put all solutions at room temperature for at least 30 minutes before use.
2. Washing solution: 15ml of the solution is dissolved with 300ml of distilled water.
3. Standard solution: 0.5ml of dd.H2O distilled water was added to the standard sample and it remained stable without stirring for 15 minutes and then stirred with a slight gesture before use.

2.1.3 The procedure:

1. 50µl of standard solution, control, and samples taken from patients were added to each small expression pit.
2. 50 µl of HRP-Conjugate was added to each small expression pit.

3. We mix for 10 seconds and incubate at c37 for one hour.
4. All the solution was poured out and washed with wash buffer 3 times for 10 seconds.
5. Add 50µl of substrate A to each of the small expression pits and immediately after that we add substrate B to all the pits as well and we mix for 30 seconds.
6. Then put it in the incubator at C 37 for 15 minutes completely in dense darkness.
7. It was noticed that the water color changed to blue color and it varies according to the concentration.
8. 50µl of the stop solution was added to stop the reaction to all the pits sequentially and the color change to yellow was immediately observed and then placed in the LISA reader at a wavelength of 450nm to get results within 10 minutes.

2.1.4 Calculations:

The samples were read by drawing a standard curve on a graph paper and the obtained absorbance values for each sample were dropped on the concentration graph for each sample. The absorbance values were on the vertical Y axis and the concentration was on the horizontal X axis. Concentrations obtained from the graph were measured in ng/ml.

![Standard curve for the estimation of the hormone activin A](image)

2.2 Measurement of cholesterol concentration in blood serum:

2.2.1 Test principle:

Cholesterol concentration was estimated using the enzymatic method, which depends on the enzymatic oxidation of free cholesterol and cholesterol ester (Tietz, 1999). The principle of operation of the detector is illustrated by the following equations:

\[
\text{Cholesterol esters} \xrightarrow{CE} \text{Cholesterol + free fatty acids}
\]

\[
\text{Cholesterol + O2} \xrightarrow{CO} \text{cholesten4 one 3 + H2o2}
\]

\[
\text{H2o2 + phenol + PAP} \xrightarrow{POD} \text{Quinoneimine (pink) + 4H2o}
\]
2.2.2 Preparation of working reagents:

The working solution was prepared by mixing well the contents of the second vial containing the enzyme with the contents of the first vial containing the Buffer buffer solution. The working solution was stored in the first bottle, and the stability of the solution continued for (5-7) days when stored at a temperature of (2-8) °C, but at a temperature of (20) °C, the stability continued for 3 months.

2.2.3 The procedure:

Three test tubes (the sample, the standard solution, the blank) were used, each containing the solutions fixed below:

<table>
<thead>
<tr>
<th></th>
<th>Blank</th>
<th>Standard</th>
<th>Assay</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reagent R1</td>
<td>1.02 ml</td>
<td>1 ml</td>
<td>1 ml</td>
</tr>
<tr>
<td>Reagent R2</td>
<td>1 ml</td>
<td>1 ml</td>
<td>1 ml</td>
</tr>
<tr>
<td>Standard</td>
<td>-</td>
<td>20 ml</td>
<td>-</td>
</tr>
<tr>
<td>Specimen</td>
<td>-</td>
<td>-</td>
<td>20 ml</td>
</tr>
</tbody>
</table>

The tubes were mixed well and left for (5) minutes at a temperature of 37°C or for 10 minutes at a temperature of (20-25)°C, after which the optical absorbance was measured by means of a spectrophotometer at the wavelength of 500 nm.

2.2.4 Calculations:

The cholesterol concentration was calculated according to the following law:

\[
\text{Cholesterol conc.} = \frac{\text{Abs (Assay)}}{\text{Abs (standard)}} \times \text{standard concentrate (5.17) mno/l}
\]

2.3 Statistical Analysis

The results were analyzed according to one-way analysis of variance using the MINITYPE system and the arithmetic means of the coefficients were tested according to the Duncun Multiple Rang test with a significant level \( P \leq 0.05 \) (Kirkwood, 1988)

III. RESULTS AND DISCUSSION

3.1 The concentration of activin-A hormone in the blood serum

Figure (2) shows that there were significant differences in the concentration of the hormone in groups of women (abortive women and those threatened with abortion) compared with the control group at a significant level \( P \leq 0.05 \). where the highest concentration of the hormone was in the aborted women, and the concentration was \( (275.0 \pm 38.9) \) pg/ml compared to With the control group, which reached a level of \( (34.67 \pm 7.75) \) pg/ml, while in women threatened with abortion, the hormone was significantly high, which reached a level of \( (168.6 \pm 37.7) \) pg/ml, compared with the control group, while the hormone was significantly lower in the group of women threatened with abortion, compared to the control group With a group of aborted women.

Figure 1: shows the concentration of Activin-A hormone in the women under study.

Where the study agreed with what was stated by the researcher Muttukrishna and his group (2000), who indicated that the concentration of the hormone rises in the case of complete Abortion and removal of the placenta, as the placenta is the main source of the hormone activin A in the maternal circulatory system during pregnancy. As maternal hormone concentrations after removal of the fetal placenta gradually begin to decrease within the first hours, this supports the concept that the human placenta is the main source of activin A [Muttukrishna., 1997]

Also, the current study agreed with the study of researcher Florio and his group (2004) (2003), as the concentrations of the hormone in the blood circulation of pregnant women are higher than in non-pregnant women, and this increase in the hormone continues during pregnancy until birth.

Measurement of the level of Activin A at the beginning of pregnancy is useful in diagnosing trophoblastic dysfunction and also helps in controlling early pregnancy problems to predict the outcome of the first trimester of pregnancy in women who develop vaginal bleeding during pregnancy that leads to a fetus threatened with abortion.

The researcher Blackburn and his group (2003) stated that the placenta trophoblasts are the main source of the hormone in the maternal circulatory system during pregnancy, as the hormone concentrations after removing the fetal human placenta gradually begin to decrease within the first hours, and this supports the concept that the placenta is the main source of activin A.
The researcher Eramaa and his group (1993) stated that the hormone changes during abortion this is because, during the first trimester of pregnancy, the ovary is responsible for secreting activin-A. When a pregnancy disorder occurs, the secretion of the hormone increases by the granulocytes in the walls of the uterus.

3.2 The concentration of Cholesterol in the blood serum

Figure (3) shows significant differences in the concentration of lipids in groups of women (abortive women and those threatened with abortion) compared with the control group at a significant level (P ≤ 0.05), where we note that there is a significant increase in cholesterol concentration in the aborted women group, which reached the level of mg/dl (281.6±156.0) compared with the control group, which reached a level of (166.5 ± 33.5) mg/dl. In women threatened with abortion, we also notice a significant increase in cholesterol concentration, which reached a level of (305.6 ± 230.0) mg/dl compared with the control group, while Cholesterol concentration was significantly lower in the aborted women group compared with the women threatened with abortion.

Where we believe that this rise in lipid can be a cause of abortion and death of the fetus, especially if the pregnant woman is taking a drug for high body lipid. We found in this current study there is a high percentage of fat in women who have aborted and threatened abortion, unlike natural women, but we did not We find research that shows there is a relationship between cholesterol concentration and the occurrence of abortion.

Changes in the liver and lipid metabolism cause a change in the concentrations of triglycerides, cholesterol, fatty acids and phosphorylated fats. After the initial decrease in the first eight weeks of pregnancy, there is a steady increase in triglycerides, cholesterol, fatty acids and phosphorylated fats. Lipoproteins (Butte, 2000) and Tietz (1999) reported that the normal levels of cholesterol in the blood are (3.9-6.5mmol/L). The rise in total cholesterol is due to the pattern of nutrition, as nutrition is one of the factors that cause high concentrations of fat in the plasma, as eating food containing a high percentage of saturated fat causes an increase in cholesterol levels and the percentage is higher than the normal rate when suffering from some diseases such as diabetes. Atherosclerosis and high blood pressure, as well as an increase in cholesterol from the normal rate when jaundice is present (Vander et al., 1998).

IV. CONCLUSIONS

1. Some instances of pregnancy loss among women are attributable to elevated concentrations of Activin-A hormone. Statistical analysis revealed significant differences in hormone concentrations when compared to control samples. These alterations can induce deviations in normal gestation, leading to threatened miscarriage and subsequent miscarriage.

2. Elevated Cholesterol Levels Among Miscarrying and Threatened Miscarriage Women Compared to the Control Group.

3. The Prevalence of Miscarriages Among Women in Rural Areas Surpasses That Among Urban Women.

REFERENCES


