

Evaluation of Some Spontaneous Abortion-Causative Factors for Women in Baghdad Province A Serological and Histological Study

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ABSTRACT:

BACKGROUND:

The spontaneous abortion is considered as one of the important clinical problems that may affects pregnant women during the 1st 20-22 weeks of pregnancy causing dangers to the mother and her fetus. The present study was designed to focus on some spontaneous abortion causes and intrauterine fetal death.

OBJECTIVE:

Study certain risk factors that may play a role in abortion such as serum level of , progesterone hormone , estradiol hormone , interlukine-6 and perform a histological study on placenta to identify any specific histop-

METHODS:

Seventy (70) Women were selected to represent the samples of spontaneous aborted women. The women were classified into two groups , the first represented aborted women during the first trimester of pregnancy (52) and the second were females in the second trimester of pregnancy (18) , in comparison with result of (20) normally delivered pregnancies. The samples (sera and placenta) were collected during the period from November 2008 to April 2009 in Baghdad.

RESULTS:

Significant decrease ($p < 0.01$) in the level of progesterone and estradiol hormone was found in all aborted women at different stages of pregnancy compared with control. There was no significant difference ($p > 0.05$) in level of IL-6 between aborted women and control. There was a correlation (positive) between progesterone with IL-6 in first and second trimester ($p < 0.05$, $p < 0.01$), respectively. And progesterone with estradiol ($p < 0.05$) in second trimester. The histological study shows hydropic degeneration fibroids with hyalinization of fibrinosis changes.

CONCLUSION:

These results suggest an important role of progesterone, estradiol and IL-6 in spontaneous abortion women during first and second trimester. And apparition of the histological changes corresponding to the aborted women.

KEY WORDS: miscarriage, Progesterone, estradiole, intelukine-6.

INTRODUCTION:

Spontaneous abortion is the most common complication of early pregnancy, also known as miscarriage, refers to a pregnancy that ends spontaneously before the fetus has reached a viable gestational age ⁽¹⁾ . The world Health Organization defined it as expulsion or extraction of an embryo or fetus weighing 500 g or less from it's mother. This typically corresponds to gestational age of 20 to 22 weeks⁽²⁾. Other terms

includes miscarriage. Miscarriage is very common, with nearly one in four women experiencing an early pregnancy loss in her lifetime. Approximately 30 percent of all pregnancies end in miscarriage, although the majority of these are miscarried before they are recognized. The rate of pregnancy loss among clinically diagnosed pregnancies is 8 to 15 percent. As many as 80 percent of miscarriages occur before 12 weeks' gestation, with miscarriage rates decreasing sharply after the first trimester. ⁽³⁾

There are many reasons associated with higher rate of pregnancy loss includes: Chromosomal

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abnormalities which are found in more than half of embryos miscarried in the first 13 weeks⁽⁴⁾. Another cause may be endocrine abnormalities such as progesterone deficiency, uncontrolled diabetes and sever cases of hypothyroidism⁽⁵⁾ up to 15% of pregnancy losses in the second trimester may be due to uterine malformation, growths in the uterus fibroids or cervical problems⁽⁶⁾.

In fact the etiology remains unexplained in a high proportion of cases. Of the causes that have been investigated is the possible immunologic rejection of paternal antigen by the maternal immune system, resulting in abnormal immune cells and cytokine production⁽⁷⁾

PATENTS AND METHODS:

This study was conducted on 70 spontaneous aborted women 52 of them aborted and rest during first trimester and second trimester. Control group consisted of 20 normal delivery women with an age range from (15 – 40) year, and the patients samples were collected in Baghdad city from Ibn – AL- Balady hospital and Fatima – AL- Zahraa hospital during the period from November 2008 to April 2009.

Collection of blood samples

Six to eight mL of peripheral venous blood samples had been collected from each patient before curettage and before normal delivery according to control in sterile plane tube and left to clot for (15 -30) minutes at room temperature, then centrifuged for 5 minutes at 3000 rpm. Sera were separated and divided into apendroff tubes and stored in deep freez at -20°C till examination.

Histopathological study

From each aborted women one piece of placentas was collected, and fixed in 10% formalin overnight for preparation of tissue sections, which were stained by H&E according to Bancroft & Steven(1980)⁽⁸⁾.

Determination of hormone concentration

Progesterone & Estradiol concentrations in blood serum were determined through solid phase competitive ELISA by using commercially available kit (DRG, Germany).

Determination of IL-6 concentration

IL-6 concentration in blood serum was determined through solid phase competitive ELISA by using commercially available kit (Immunoteck, France).

Statistical analysis

The data were statistically analyzed by using analysis of variance test (ANOVA), least

significant difference (LSD) and person correlation coefficient (r) depending on SPSS program statistical package of social science.vergion⁽¹⁹⁾.

RESULTS:

Progesterone hormone concentration was significantly lower $p=0.0001$ in the aborted women in 1st trimester (10.28 ± 3.99) ng/mL and 2nd trimester (26.90 ± 9.68) ng/mL compared with control (219.79 ± 5.11) ng/mL as shown in table (1). There was high significant ($p=0.0001$) between aborted group and control but there was no significant difference between aborted groups themselves.

Estradiol concentration was significantly lower $p=0.0001$ in the aborted women in 1st (110.21 ± 19.22) pg/mL and in 2nd (166.55 ± 39.86) pg/mL than in control (3910.74 ± 271.35) pg/mL. There is a high significant difference $p=0.0001$ between aborted women groups and control but there was no significant difference between first trimester and second trimester of aborted women group (table 2).

Interleukine-6

The mean levels of IL-6 in serum were not different between aborted women groups and control group ($p=0.602$). The mean value \pm SEM for IL-6 was (53.81 ± 2.33) pg/ml in first trimester, (56.40 ± 2.33) pg/ml in second trimester of abortion and (57.26 ± 2.30) pg/ml in control (table 3).

Correlations

Table (4) shows a significant positive correlation between progesterone with IL-6 ($r=0.285$ $p=0.041$, $r=0.739$, $p=0.000$) in first trimester and second trimester, respectively.

Also progesterone showed positive significant correlation with estradiol ($r=0.552$, $p=0.018$) in second trimester.

Histological findings

The figure (1) and (2) show sections of placenta of aborted women with low level of progesterone and estradiol in serum. Degenerated changes with slight fibrosis chorionic villi are seen. However patent blood vessel inside the stroma are still seen

DISCUSSION:

The lower progesterone levels that had been observed in this study are similar to those reported by Nelson *et al*⁽⁹⁾ Muttukrishna *et al*⁽¹⁰⁾ and AL-Najjar⁽¹¹⁾.

The decrease of progesterone hormone levels lower than 10 ng/ml causes abortion⁽¹²⁾.

According to the data discussed, three endocrine disorders related to spontaneous and recurrent

SPONTANEOUS ABORTION-CAUSATIVE FACTORS

abortions can be differentiated: Corpus luteum insufficiency, defective luteo-placental progesterone and placental delay for progesterone synthesis and secretion⁽¹³⁾. Most of the PRO hormone in early pregnancy is produced by the corpus luteum and placental PRO comes later on⁽¹⁴⁾. The importance of corpus luteum that after its formation, PRO acts on the endometrium and stimulate blastocyst growth and elongation to a filamentous conceptus (embryo/ fetus)⁽¹⁵⁾.

Significantly lower estradiol concentrations were recorded in aborted women compared with control in this study. This result is correlated with many previous studies Nelson *et al*⁽⁸⁾; Muttukrishana *et al*⁽¹⁰⁾ Who found that estradiol concentration level was significantly lower ($p=0.0001$) in women with miscarriage than those with alive birth. In contrast AL-Barwary,⁽¹⁶⁾ that estradiol concentration is significantly higher in aborted women than those with a live birth.

It was reported that estradiol is crucial for normal pregnancy development⁽¹⁷⁾. Women at high risk for subsequent miscarriage had oligomenorrhea and an isolated deficiency of estradiol in the luteal phase of menstrual cycle⁽¹⁸⁾. The source of estradiol in the first trimester of pregnancy is corpus luteum and placenta⁽¹⁴⁾. Therefore any deficiency of corpus luteum or delayed estradiol biosynthesis by the placenta may cause abortion. The result of IL-6 was similar to the results observed by Hattori *et al*⁽¹⁹⁾. Who showed no significant difference of IL-6 level in serum between serum miscarriage and live birth group. On other hand other study conducted by Makhseed *et al*⁽²⁰⁾ showed that IL-6 level was significantly higher in live birth women than in women with spontaneous abortion.

Hill *et al*⁽²¹⁾ indicated that peripheral blood lymphocytes of women with RSA secrete higher concentration of certain Th1 cytokines and lower concentration of Th2 cytokines upon stimulation with antigen as compared with women with successful pregnancy, we did find low concentration of IL-6 level in aborted women

without significant difference compared with control group; the cause may be the sample size.

In this study, a positive significant correlation was recorded between IL-6 and progesterone in women with abortion, may be because progesterone hormone is essential for the maintenance of pregnancy. It is also known to modulate immune function and elicit an immunological critical response for normal gestation. It has been shown to favor the development of human T.lymphocytes production Th₂ cytokines, which inhibit Th₁ cytokine, thus allowing the survival of fetal allograft⁽²²⁾ and block Th₁ immunity to trophoblast by PBMC of women with recurrent pregnancy loss in vitro⁽²³⁾. Therefore, they propose that progesterone may be responsible, at least in part, for a Th₁ to Th₂ switch at the maternal – fetal interface⁽²⁴⁾.

Also progesterone showed positive significant correlation with estradiol in second trimester of abortion.

Steroids are fundamental in pregnancy maintenance and parturition. The progesterone hormone is a female sex hormone which, in conjunction with estrogens regulates the accessory organs the implantation of the blastocyst and in maintaining pregnancy⁽²⁵⁾.

Because the high abortion rate in cases with over – stimulation syndrome, where progesterone as well as estradiol fall from a very high level support this concept. Furthermore, abnormal endocrine condition could be generated by the delayed and lower progesterone and estradiol biosynthesis by placenta. This delay is very extensive and could perhaps be the reason of late abortion⁽¹⁴⁾.

CONCLUSION:

These results suggest an important role of progesterone, estradiol and IL-6 in spontaneous abortion women during first and second trimester. And apparition of the histological changes corresponding to the aborted women.

Table 1: Concentration of progesterone hormone in the studied group.

Study group	Number	Progesterone concentration (ng /mL)	ANOVA
		Mean ± SEM	P.Value
First trimester	52	10.28 ± 3.99 ^a	0.0001
Second trimester	18	26.90 ± 9.68 ^a	
Control	20	219.79 ± 5.11 ^b	

Value with different superscripts in the same columns (small letters) differ significantly ($p=0.0001$).

SPONTANEOUS ABORTION-CAUSATIVE FACTORS

Table 2: Concentration of estradiol hormone in the studied group.

Study group	Number	estradiol concentration (pg / mL)	ANOVA
		Mean \pm SEM	P.Value
First trimester	52	101.21 \pm 19.22 ^a	0.0001
Second trimester	18	166.55 \pm 39.86 ^a	
Control	20	3910.74 \pm 271.35 ^b	

Value with different superscripts in the same columns (small letters) differ significantly ($p=0.0001$).

Table 3: Concentration of IL-6 in the studied group.

Study group	Number	IL-6 concentration (pg / mL)	ANOVA
		Mean \pm SEM	P.Value
First trimester	52	53.81 \pm 2.33	0.0001
Second trimester	18	56.40 \pm 2.33	
Control	20	57.26 \pm 2.30	

Table 4: Person correlation between selected variables in first and second trimester of abortion

	First trimester				Second trimester			
	Interleukine-6		Estradiol		Interleukine-6		Estradiol	
	r	p	r	p	r	p	r	p
Progesterone	0.285*	0.041	0.264	0.059	0.739**	0.000	0.552*	0.018

Correlation (r value), p: Probability*.

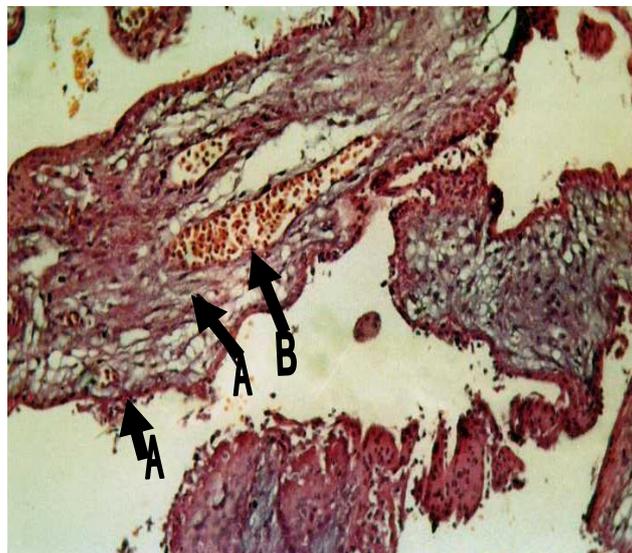


Figure 1: Section of Placenta from aborted women with low level of progesterone and estradiol showing certain degenerated chorionic villi with slight fibrosis (A) but still patent blood vessel inside stroma (B) (H&E, x 200).

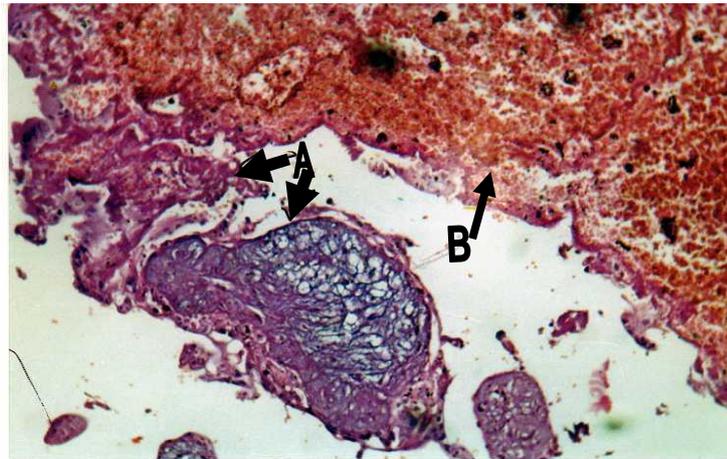


Figure 2: Section of placenta from aborted women with low level progesterone and estradiol showing (A) degenerative changes and necrosis (B) ((high power view x400).

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