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Original article:

EFFECT OF PROGESTERONE ON SERUM VEGF AND RECURRENT ABORTION IN INDIAN WOMEN

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ABSTRACT

Background: There is a mutual relationship between progesterone and Vascular Endothelial Growth Factor (VEGF) which are important for adaptation to pregnancy and in cases of Recurrent Abortion.

Materials and methods: This is a descriptive comparative study. The correlation of Progesterone and VEGF was evaluated in cases of recurrent abortion(RA) in North Indian pregnant women for 60 patients with a history of RA (Study group) and 60 normal pregnant women of early pregnancy (Control group) through this descriptive comparative study. The blood samples of the patients were collected to assess the serum level of Progesterone and VEGF by Enzyme-Linked Immunoassay (ELISA) technique Human VEGF-A BIOLISA kit. The concentration of circulating Progesterone and VEGF-A was evaluated by using Student-t test.

Results : There was a positive correlation between progesterone therapy and serum progesterone & VEGF levels in cases of RA.

Conclusion: The study concluded that increased levels of progesterone lead to increasing levels of VEGF and progesterone could help in the management of Recurrent Abortion patients.

Keywords: Progesterone, Vascular Endothelial Growth Factor (VEGF), Recurrent Abortion (RA), Early Pregnancy.

BACKGROUND

Recurrent pregnancy loss is a frustrating and heart-wrenching experience for both physician and patients. It is, unfortunately, the most common complication of human gestation in at least 75% of all women trying to conceive.

Recurrent abortion is defined as the 3 or more consecutive pregnancy losses. Most of these losses are unrecognised and occur before or within the next menstrual cycle. It affects about 1% of childbearing population and presents couples with a formidable challenge in successfully having a family. Approximately 5% of couples trying to conceive have 2 consecutive miscarriages and about 1% have 3 or more consecutive losses. (1,2)

Unexplained recurrent losses are associated with substantial adverse clinical and psychological consequences for the women and their families. There have been consistent efforts to evaluate therapeutic strategies to increase the rate of live births among these women, but no effective treatment has been identified. (3).

Physiological changes in the uterine circulation during early pregnancy are regulated by various hormones including progesterone and human chorionic gonadotropin (HCG)secreted by the syncytiotrophoblast cells (4,5). They play an

important role in the implantation processes during pregnancy, like cell growth and differentiation besides maintenance of the corpus luteum. (2,6,7) .

Progesterone is the hormone most clearly associated with maternal adaptation to pregnancy. It is more effective in the early stages of gestation (8).Progesterone seems to be necessary for supporting an early pregnancy and it has been used for the said purpose for several years (9).

During pregnancy, VEGF expression rises and is prominent in syncytiotrophoblasts and deciduas and exhibits positive functional regulation by estrogen and progesterone in a steroid receptor-dependent manner. Thus formation and maintenance of the vascularization and receptor endometrium is also dependent upon angiogenesis and is driven by expression of VEGF and other angiogenic factors.

In pregnant women, progesterone appears to be potent regulators of ovarian VEGF expression and promote the vascularization and stabilization of the functional steroid producing corpus luteum (4,5).

This study aims to find out the relationship between VEGF and progesterone and progesterone therapy in cases having a history of early recurrent losses.

MATERIALS AND METHODS

A descriptive comparative study was conducted in the Department of Anatomy and the subjects for the study were obtained from the Department of Obstetrics and Gynaecology of Lady Hardinge Medical College and Smt. Sucheta Kriplani Hospital, New Delhi. A total of 120 pregnant subjects were included in this study, which was divided into a study group (60 subjects) and control group (60 subjects). The inclusion criteria in the study group were women in the age of 20 - 35 yrs. with a history of 3 or more spontaneous abortions. Subjects having a single partner and with no anatomic, physiological, hormonal, chromosomal, infective or autoimmune causes for recurrent abortions were included. The subjects included in the control group were of a similar age group without any history of pregnancy loss and at least one live birth. Subjects with medical problems like thyroid disorders, autoimmune diseases, bleeding disorders, an endocrine disease like diabetes mellitus, uterine or any other congenital abnormality were excluded.

The consent form was provided, and informed consent was obtained from all the subjects. A detailed history, clinical examination, and investigations of the subjects were recorded. Ultrasound of the pelvis was done to rule out any malformations of the genital tract, which might have been responsible for abortion. The blood was collected from the subjects to check the Progesterone levels and VEGF levels through Enzyme-Linked Immunoassay (ELISA) technique - Human VEGF-A BIOLISA kit (Diaclone). The concentration of circulating Progesterone and VEGF-A was evaluated by using Student-t test.

RESULTS:

The serum VEGF levels in the recurrent pregnancy loss before administration of progesterone and control group are show below.

Serum VEGF (pg/ml)	Study Group -1 (Before Progesterone) n=60, (%)	Control Group n=60,(%)
0-100	30(50%)	0
100-200	29(48.3%)	0
200-300	-	0
300-400	1(1.6%)	0
400-500	0	0
500-600	0	2(3.3%)
600-700	0	0
700-800	0	0
800-900	0	4(6.6%)
900-1000	0	3(5%)
1000-2000	0	39(65%)
2000-3000	0	12(20%)

Table 1: Distribution of patients according to S.VEGF LEVELS IN STUDY GP- 1 and CONTROL GROUP

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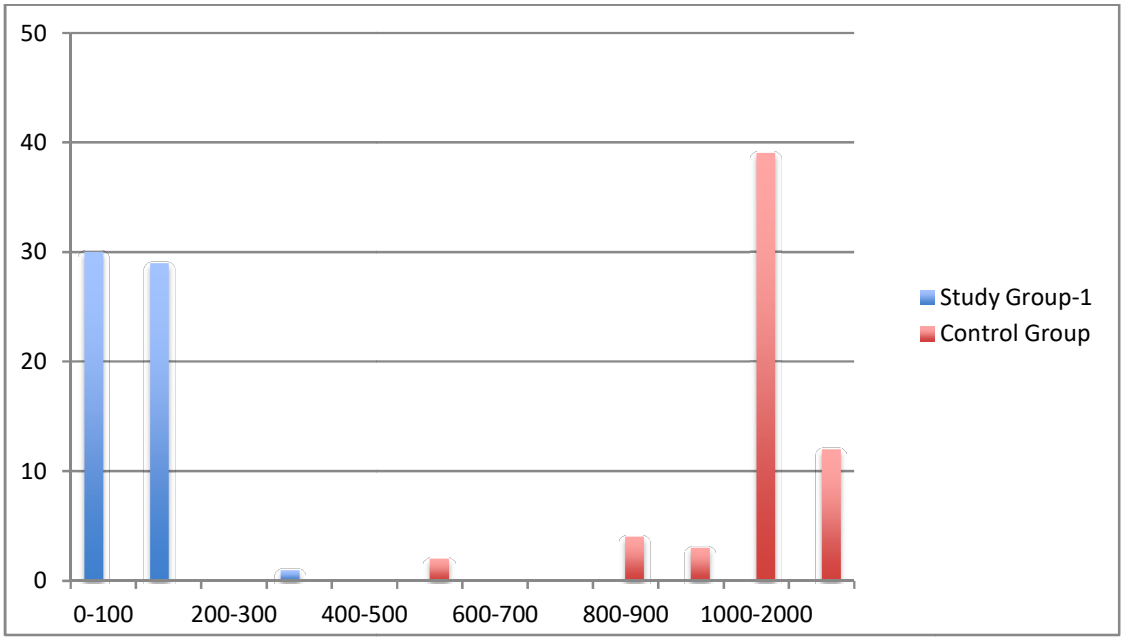

Fig 1: Distribution of patients according to S.VEGF LEVELS IN STUDY GP- 1 and CONTROL GROUP

Using student – t test, Mean Value of Serum VEGF in Study Group – 1= 104.58 +/- 465.85 pg/ml

Mean Value of Serum VEGF in Control Group = 1524.0+/- 465.85 pg/ml

P<0.001, was very highly significant.

Serum VEGF (pg/ml)	Control Group n=60,(%)	Study Group -2 (After Progesterone) n=60, (%)
0-100	0	0
100-200	0	0
200-300	0	3(5%)
300-400	0	6(10%)
400-500	0	15(25%)
500-600	2(3.3%)	18(30%)
600-700	0	11(18.3%)
700-800	0	6(10%)
800-900	4(6.6%)	0
900-1000	3(5%)	0

1000-2000	39(65%)	1(1.6%)
2000-3000	12(20%)	0

The serum VEGF levels in the recurrent pregnancy loss patients after administration of progesterone and control group are show below

Table 2: Distribution of patients according to S.VEGF LEVELS IN CONTROL GROUP and STUDY GP- 2

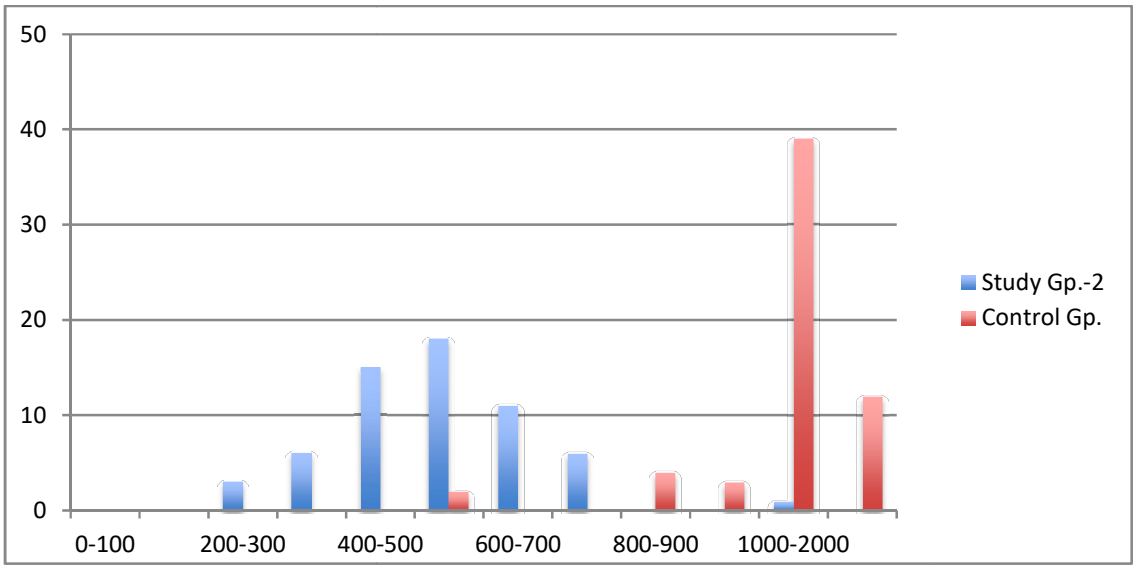

Fig 2: Distribution of patients according to S.VEGF LEVELS IN CONTROL GROUP and STUDY GP- 2

Using Student –t tests , Mean Value of Serum VEGF in Control Group = 1524.0+/- 465.85 pg/ml

Mean Value of Serum VEGF in Study Group – 2= 524.0+/- 182.31 pg/ml

P<0.001, was very highly significant.

The serum VEGF levels in the recurrent pregnancy loss patients before and after administration of progesterone and control group are show below.

Serum VEGF (pg/ml)	Study Group -1 (Before Progesterone) n=60, (%)	Study Group -2 (After Progesterone) n=60, (%)
0-100	30(50%)	0
100-200	29(48.3%)	0
200-300	-	3(5%)
300-400	1(1.6%)	6(10%)
400-500	0	15(25%)
500-600	0	18(30%)
600-700	0	11(18.3%)
700-800	0	6(10%)
800-900	0	0
900-1000	0	0
1000-2000	0	1(1.6%)
2000-3000	0	0

Table 3: Distribution of patients according to S.VEGF LEVELS IN STUDY GROUP-1 and STUDY GP- 2

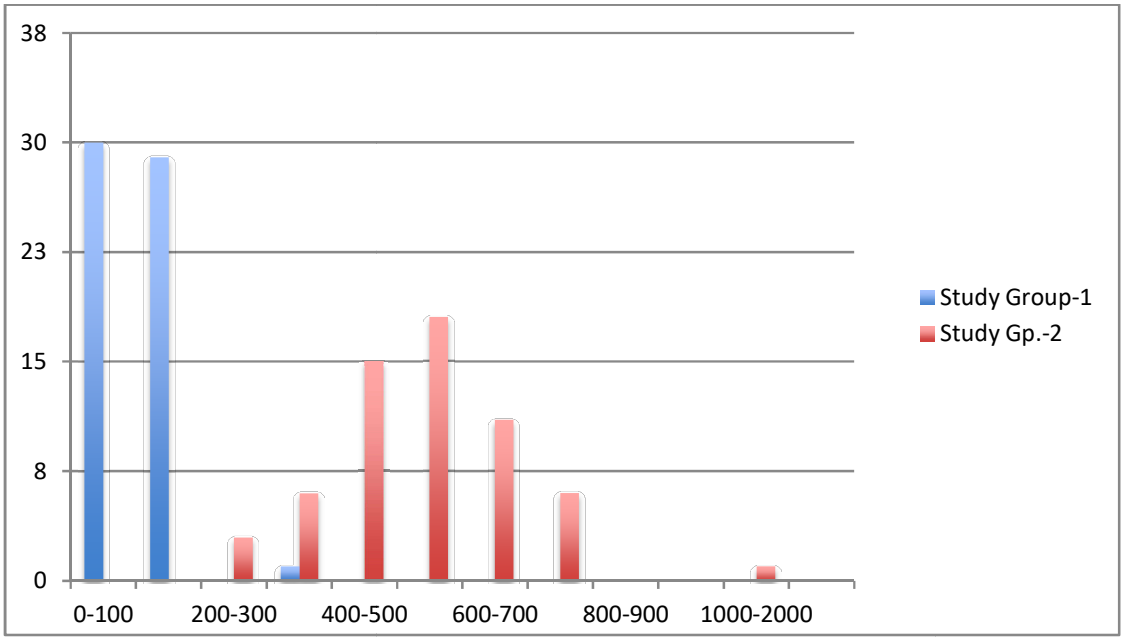

Fig. 3: Distribution of patients according to S.VEGF LEVELS IN STUDY GROUP-1 and STUDY GP- 2

Using Student –t tests , the Mean Value of Serum VEGF in Control Group = 104.58.0+/- 57.97pg/ml

Mean Value of Serum VEGF in Study Group – 2= 524.0+/- 182.31 pg/ml

P<0.001, was very highly significant.

DISCUSSION

Progesterone secreted by the corpus luteum and then by the placenta is a key hormone during pregnancy and is required for implantation and maintenance of pregnancy. Additionally, it also has some immunomodulatory effects that could further influence endometrial receptivity . This meta-analysis supports the use of synthetic progesterone in women having a history of unexplained recurrent pregnancy loss.(10)

Hormonal control of gene expression is of fundamental importance in many tissues. The uterine endometrium is a prime example of such tissue and highly responsive to the ovarian sex steroids. These responses include rapid tissue growth and remodeling, blood vessels development and various protein secretions. Each these process necessitates complex cell-cell interactions (11).VEGF is the growth factor which mediates this cell-cell interaction.

VEGF is a potent angiogenic growth factor and produces in both glands and stromal cells of the endometrium and also myometrial smooth muscle cells.(12,13,14)

Recurrent abortion is labeled on the basis of total number of abortions (3 or more than 3 consecutive abortions). The mean serum VEGF decreased as the number of recurrent abortions increased in the patients. Thus mean serum VEGF was 165.8,113.7,100.5,when there 3,4,5 recurrent abortions respectively. According to the present study number of abortions was found to have a highly significant negative correlation (r=-0.495) with serum VEGF concentration (p<0.001). These findings imply that patients with a higher number of abortions in the early trimester had lower serum VEGF concentrations. No other study could be found for comparison.

An attempt was made to determine the role of progesterone in case of recurrent abortions with decreased S.VEGF levels, women with a history of recurrent abortions were put on progesterone therapy. Mean serum VEGF concentration in study GP-1(Before progesterone) was 104.58+/-465.85pg/ml. Levels of VEGF were repeated after four weeks of progesterone therapy and mean VEGF concentration rose significantly to 524.88+/- 182.31pg/ml,the levels were increased almost 5 times. We found a positive correlation (r=+0.351) between serum VEGF concentration and serum progesterone levels. Mean serum concentration of progesterone in the study group prior to progesterone therapy was 5.57=/-2.31ng/ml and means serum concentration of VEGF was 104.58+/-465.85pg/ml. After four weeks of progesterone therapy, however, mean serum conc. of progesterone increased to 14.81+/-2.83ng/ml and mean serum VEGF conc. rose to 5 folds, approximately 524.88+/-182.31pg/ml which was showing a positive and highly significant correlation (r=+0.351, p<0.005). Although serum VEGF values increased by 5 folds in the study group in spite of progesterone therapy it never reached the mean value of 1524+/-465.85pg/ml seen in the control group of present study. Mean serum progesterone in the study group continued to be lower being 14.81ng/ml as compared to the mean value of serum progesterone 24.36+/-3.4pg/ml in the control group. This difference again was highly significant. When the range of serum progesterone was compared in two groups of present study 45 out of 60 patients in the study group had serum progesterone between 8-16ng/ml whereas none of the patients the control group had serum progesterone (less than)put sign 16ng/ml. 14 patients in the study group, however, attained serum progesterone values between 16-32ng/ml. The results of the present study show that the rise in VEGF levels following progesterone therapy perhaps reflects improvement in placental blood flow. These levels of VEGF can perhaps be modulated by drug therapy. Future research to address this issue may help in better management of patients with recurrent abortions.

It is known that physiological changes in the uterine circulation during pregnancy are hormonally regulated. Hormones like progesterone and HCG are mainly responsible for numerous pregnancy-related and pregnancy maintaining changes such as implantation and preserving intrauterine pregnancy. It is possible that progesterone has direct angiogenic action and also stimulate angiogenesis by inducing VEGF production as was seen by rising of serum levels of VEGF in study group after progesterone therapy.

Cellinan Bone & Hyder et al observed up-regulation of VEGF m RNA in mature rat uterus in response to ovarian sex steroids.(15,16)

Cochrane et al reviewed four trails showed a significantly lower risk of miscarriages among women who had given progesterone than among women who were on placebo.(17).

Dr. Stephenson found that progesterone is a very beneficial and safe treatment for women with a history of recurrent pregnancy loss. The author found the higher birth rate in women administered natural progesterone per vaginally.(18)

This has been authenticated by a number of studies have reported that the progesterone has direct angiogenic action and also acts as our study as an indirect angiogenic factor by inducing VEGF production in macrophages. (19,12). Thus according to our study suboptimal amount of progesterone can result in defective endometrial development due to decreased VEGF production, which ultimately leads to abortions.

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Martin et al found a similar effect of progesterone in endometrial angiogenesis in pregnant and ovariectomized mice(20).

Greb et al, reported progesterone induced endothelial cell proliferation mediated by multiple angiogenic mechanism and regulatory factors.(21)

Ma et al, reported analogous findings and found that progesterone was proliferative in vivo in ovariectomized mice and suggested the mechanism of action to be via induction of VEGF and its receptors VEGF-2 (FLK-1) in conjunction with neuropilin-1 (22). Walter et al demonstrated that progesterone stimulates endometrial and endothelial cell proliferation mediated by VEGF.(20)

Xu et al in, observed that 100pg/ml progesterone increased VEGF –A and B in cultured leiomyoma and myometrial cells. (23)

CONCLUSION

The present study suggest that administration of progesterone can help as therapeutic drug for maintenance of pregnancy in recurrent abortion .

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