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Estrogen, Progesterone and Serum Prolactin Levels In Infertility Cases 0f Women

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Abstract: *The present study was undertaken to evaluate the endocrinal causes of infertility in women .Infertility affects 10-15% 0f couples 0f reproductive age groups and female factor is responsible for 40 – 50 %of cases , most common causes of female infertility are ovulatory dysfunction and thyroid hormone ,and estrogen ,progesterone ,and serum Prolactin dysfunction . Estimation of estrogens,progesterone and serum prolactin hormones useful in diagnosis the type of ovulatory dysfunction and useful in deciding the mode of treatment. The present study has been undertaken to evaluate hormone levels (estrogens,progesterone and serum prolactin) in ovulatory dysfunction cases of female infertility after elimination of PCOD. The fertility of female reproductive system is maintained by prevaling hormonal mainly which is delicately balanced by Hypothalamic ,Pituitary ,Thyroid, adrenal and gonadal axis .The problem of infertility are faced from the time of evaluation of man will probably remain with as forever .little is added every year to the present understanding and more remains unknown Infertility affects 10-15% of couples of reproductive age groups .Male factor in 25-40 % female factor in 40- 55% .male and female in 10%, unexplained in 10% cases are responsible for infertility .ovulatory dysfunction is responsible for upto 40% of female cases and for 20-25% of total infertility cases*

Keywords: Female infertility, estrogens,progesterone and serum prolactin, Hypo& Hyper Gonadotrophic anovulation

I. Introduction

Human beings can not help thinking alone ,may longing for what they don't have ,particularly when every one round them has it in abundance .procreation is a very basic biological urge. It is hence logical that the infertile couple be offered every scientific aid available to them to attain freedom from their sense of void, so that they may have the pleasure and fulfillment of bearing their own pregnancy. Endocrinological abnormalities are frequently encuontered and most effectivly treated causes of infertility. [1] We must in particular recognize the right of women to bear her own child ,if she so desires .Infertility is defined as one year of unprotected coitus without conception this condition may be further classified as primary infertility in which a prior pregnancy has occurred .Infertility affects approximately 10-15% reproductive age couples in U.S.A -17% ,26% IN U.K ,21% in canada ,13% IN DENMARK ,14% IN SCOTLAND ,5.4% IN ISRAELI ,In INDIA incidence varies from 5-15%in south INDIA 8-10% Infertility is a common accompaniment of disorders of various gonadal hormones are associated with variety of changes in reproductive function including of delayed onset of puberty, an ovulation and abnormally high fetal wastage .subtle changes in thyroid function may have permissive role in the production of absolute &relative infertility in some individuals.[2] Prolactin can also stimulate the enzyme 3-beta –ol-dehydrogenase in the gonads,resulting in steroidogenisis, particularly in the synthesis of progesterone. prolactine levels also may be elevated in women who have subtle alterations of fertility such as anovulations and amenorrhea .Both primary and secondary types of infertility generally share common causes. The incidence of infertility has been increasing in the last decade. Awareness of infertility has increased as couples have become more willing to seek medical advice .Infertility problems often arise as a result of hormonal dysfunction of hypothalamo-pituitary gonadal axis.[3] Measurement of peptide and steroid hormones in the serum are therefore essential aspects of the evaluation of infertility.

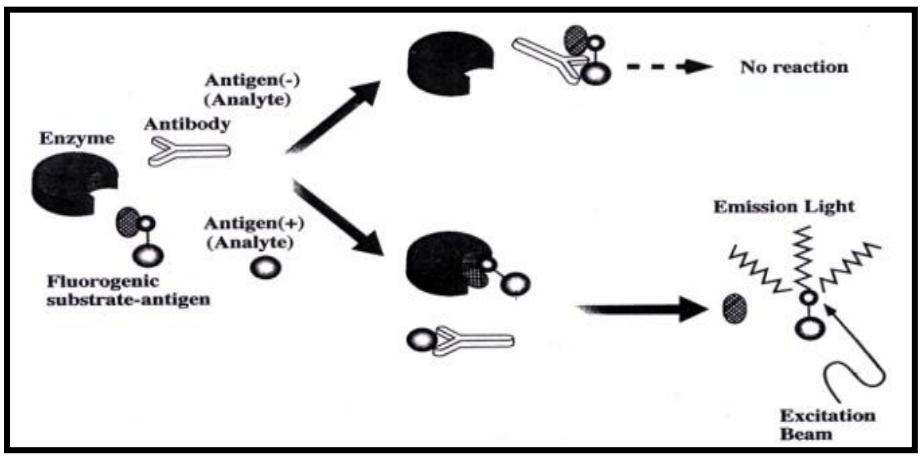
II. Materials And Methods

The study included a total no of 63 cases of infertility cases of women and the present study was carried out in the Dept of obstetrics and Gynaecology of maternity hospital of SVMC and collaboration with Dept of Endocrinology, SVIMS Tirupati .The couples with either primary or secondary infertility were selected successively and evaluated for male factor, non- endocrinological causes of infertility and then subjected to endocrinological evaluation

Controls : Controls were selected on the basis of normal menstrual history, no systemic illness ,no contraceptive history and are fertile ,and in good health ,63 an- ovulatory patients were studied against 30 controls of 20 to 30 age group.

Collection of samples:Whole blood was collected on day 7 of menstrual cycle from controls.3 samples of whole blood was collected from patients at an interval of 20 minutes and pooled. Clear serum without hemolysis is separated into clean bottles and labeled and measured for hormonal assay. Depending upon the values obtained the patients are divided into 3 categories [10]

Group-1 – Hormonal levels below normal limits
Group-2 –Hormonal levels within normal limits
Group-3 –Hormonal levels above normal limits



Estimation:
Estimation of estrogens,progesterone and serum prolactin levels by

Fluoro-Immuno assay method (FIA) This methods is most sensitive than the radio immuno assay and ELISA. We can detect even less than 1ng/ml of analyte and we can also avoid risk of radiation by this method.

Enzyme linked fluorescent Assay
Principle
Enzyme linked fluorescent Assay are identical to other EIAS except that they use fluorescent Substrates. In the ElFA a florophore is generated by an enzyme reaction.

The substrate β-galactosyl umbelliferone is conjugated with the antigen (analyte) and forms non fluorescent substrate. The substrate can be cleaved by an enzyme β-galactosidase to form a fluorescent product. However when the substrate – antigen conjugate is allowed to react with specified antibody to the antigen, there is no cleavage of the substrate complex with the β-galactosidase enzyme. In this assay the concentration of the antigen (analyte) is directly proportional to the fluorescent intensity measured. Flurophor used–4-methylumbelliferone (4-MU)Instrument calibration is provided using a standard calibration solution in a VIDAS strip prepared by bioMerieux. These calibration strips are checked by QC/QA and values are assigned to the strip [approximately a 3,200 RFU reading with an 8,000nM solution of solution of 4-methylumbelliferone (4-MU) in a buffer].

Flurophor used–4-methylumbelliferone (4-MU): Instrument calibration is provided using a standard calibration solution in a VIDAS strip prepared by bioMerieux. These calibration strips are checked by QC/QA and values are assigned to the strip [approximately a 3,200 RFU reading with an 8,000nM solution of solution of 4-methylumbelliferone (4-MU) in a buffer].

Methods
The FIA uses several different methods to calculate results. There are three basic categories of analysis methods:

Patient to standard: A comparison of the Relative Fluorescence Value (RFV) of the test sample to that of a standard. This method is used in single reagent strip qualitative and most semi-quantitative assays.

Patient to reference: A comparison of the RFV of the test sample to that of a reference blank This method IS used In dual reagent strip qualitative assays

Curve fitting equations: The RFV of a test sample is mathematically placed on a calibration curve. This method is used for all quantitative and some semi-quantitative assays.

If the test value is:	**the result is:**
> or = high threshold	positive
< high threshold and	equivocal
≥ low threshold	equivocal
< low threshold	negative

Expected Values in FIA method

1. Estrogen = Follicular phase 8-147 pg/ml
   Preovulatory phase 93-475 pg/ml
   Luteal phase 43-214 pg/ml
   Meno phase < 58 pg/ml
2 . Progesterone= Ovulatory phase 9-60 ng/ml
3. Serum Prolactin= Normal menstruating 5-25 ng/ml

Results: The dysfunctioning of **estrogens,progesterone and serum prolactin** hormone levels are interfering the functions of normal female fertility. The inspiration for this study to know the present infertility cases (female) by analyzing the following biochemical parameters by estimating the levels of these parameters we can assess the infertility rate in women.

The following biochemical parameters were done. 1.Estrogen 2.Progesterone 3.Serum prolactin

III. Results

Female reproductive hormones measurement are of Diagnostic and therapeutic value in ascertaining the homeostasis of fertility regulation via the hypothalamic-pituitary-gonadal axis.[9]

Hypogonadotropic anovulation is caused hypothalamic or pituitary failure. After excluding space occupying lesions in hypothalamic - pituitary regions ovulation can be induced by pulsatile or recombinant therapy in majority of cases [8] . In our present study cases categorized under normogonadotropic anovulation have significant low levels of gonadotropins compared to controls. Though the values are within normal limits they are not sufficient to initiate normal ovulatory cycle or due to disturbances in hypothalmic – pituitary–ovarian axis. 33 cases of ovulatory dysfunction were chosen for the present study of evaluation of estrogen,progesterone serum prolactin hormones levels after elimination of systemic illness, pregnancy,PCOD. In our present study hypogonadotropic anovulation (Group I) was observed in 23 cases (about 10%) with and the estrogen level with mean value 35.78 and with SD of ±9.67 and progesterone with mean value 5.70 and with SD of ±1.88 and serum prolactin level with mean value 2.00 and with SD value of ±0.64.

Normogonadotropic anovulation groups was observed in 30 cases (about 75%) with estrogen level with mean value 85.5 and with SD of ±11.87 and progesterone with mean value 15.93 and with SD of ±2.28 and serum prolactin level with mean value 15.69 and with SD value of ±5.69.

Hypergonadotropic anovulation groups observed in 10 cases with estrogen level with mean value 217.2 and with SD of ±10.62 and progesterone with mean value 77.75 and with SD of ±2.95 and serum prolactin level with mean value 25.36 and with SD value of ±1.65.

Observations were depicted in table 1,2,3. Results were statistically evaluated using student ‘t’ test

Table-1

Different Bio-chemical parameters of cases (total cases : 33)

S.No.	Estrogen (ng/ml)	Pro-g (ng/ml)	Prolactin (ng/ml)
1.	25	0.6	3.1
2.	31	8.4	2.8
3.	43	7.3	2.1
4.	27	7.8	2.5
5.	21	6.9	2.8
6.	23	5.5	2.6
7.	32	4.8	1.1
8.	31	3.6	1.2
9.	27	7.7	1.8
10.	34	6.8	1.6
11.	36	5.7	2.1
12.	51	4.3	1.8

13.	47	6.1	1.2
14.	45	8.0	1.0
15.	32	7.3	1.8
16.	38	7.1	2.3
17.	37	6.1	2.2
18.	28	5.6	25.1
19.	25	6.0	24.8
20.	53	4.8	23.6
21.	42	4.1	25.2
22.	52	3.7	28.1
23.	43	3.1	
24.	225	75.1	
25.	231	76.8	
26.	203	78.9	
27.	221	80.1	
28.	228	78.1	
29.	225	77.3	
30.	202	78.2	
31.	219	71.3	
32.	208	79.8	
33.	210	81.9	
Mean	**90.71**	**27.53**	**75.3**
SD	**±85.23**	**±33.69**	**±10.06**

Table-2
Different Bio-chemical parameters in controls (total controls n=30)

S.No.	Estro (pg/ml)	Pro-g (ng/ml)	Prolactin (ng/ml)
1.	61.8	12.1	4.1
2.	65.3	14.3	4.6
3.	71.8	14.1	5.6
4.	93.7	13.8	5.8
5.	78.9	11.8	7.2
6.	68.9	14.1	6.9
7.	73.2	13.8	10.2
8.	81.3	16.7	11.3
9.	101.2	15.3	18.6
10.	111.3	17.1	17.3
11.	78.7	14.8	18.7
12.	91.8	13.6	19.2
13.	79.8	14.1	20.1
14.	86.8	14.6	19.3
15.	79.8	13.5	18.7
16.	81.3	14.3	17.8
17.	90.8	16.1	19.3
18.	97.2	17.8	16.8
19.	98.3	18.3	17.2
20.	97.2	19.1	16.7
21.	92.3	17.3	16.3
22.	78.3	16.8	21.2
23.	81.4	14.5	21.3
24.	78.9	19.1	20.8
25.	71.8	17.2	21.3

26.	92.3	18.3	21.4
27.	97.4	19.6	20.8
28.	96.3	17.8	16.7
29.	97.4	18.2	17.3
30.	91.6	19.8	18.4
Mean	**85.56**	**15.93**	**15.69**
S.D.	**±11.87**	**±2.28**	**±5.69**

Table3Mean and SD VALUES of various biochemical parameters in controles and total cases(hypo +hyper)

S.No.	Parameter	Mean & SD		P value	Remarks
		Control (30)	Cases (33)		
1	Estrogen	85.56±11.87	90.75±85.23	<0.001	Significant
2	Progesterone	15.93±2.28	27.53±33.69	<0.001	Significant
3	Prolactin	15.69±5.69	75.3±10.06	<0.001	Significant

IV. Discussion

The dysfunctioning of the estrogen levels, progesterone levels and serum prolactin levels are interfering the functions of normal female fertility The inspiration for this study to know the present infertility cases (female) by analyzing the following biochemical parameters. By estimating the levels of these parameters we can assess the infertility rate in women. Hypergonadotropic anovulation indicates premature ovarian failure and early menopause. Fertility can not be restored in these cases. These cases should be followed up for autoimmune disorders.Measurement of these hormones differentiates between ovarian and hypothalamic - pituitary causes of hypoestrogenism.

Estrogen hormone levels

The mean Estrogen hormone level in the study group was 90.75±85.23. The difference between mean of the two groups statistically significant and we also compare the mean of Hypohormonal cases i.e. 35.78±9.67 and in hyperhormonal cases i.e. 217.2±10.62. It is also shows significant levels.

Progesterone hormone levels

The mean Progesterone hormone level in the study group was 27.52±33.69. The difference between mean of the two groups statistically significant and we also compare the mean of Hypohormonal cases i.e. 5.70±1.88 and in hyperhormonal cases i.e. 77.75±2.95. It is also shows significant levels.

Serum prolactin levels

The mean Serum prolactin level in the study group was 75.30±10.06. The difference between mean of the two groups statistically significant and we also compare the mean of Hypohormonal cases i.e. 2.00±0.64 and in hyperhormonal cases i.e. 25.36±1.65. It is also shows significant levels.

V. Summary & Conclusions

Evaluation of estrogen,progesterone and serum prolactin hormone levels, in cases of female infertility due to ovulatory dysfunction is extremely useful in determination of the diagnosis. estrogen,progesterone and serum prolactin hormone levels are useful in selection of treatment plan and can also assess the outcome of the treatment. In our present study variations of estrogen,progesterone and serum prolactin hormone levels in patients suffering from ovulatory dysfunction were significant to classify the patients into three district functional groups.

Further evaluation of normogonadotropic anovulation revealed that functional hypothalamic - pituitary dysfunction is a major cause of ovulatory dysfunction. We tried to assess by our study the some endocrinal hormonal levels in infertility cases of women. We have done the comparative study of all parameters with control subjects and cases of both hypo and hyper hormone levels. We have also compared the level of all parameters between the hypo and hyper gonadotrophic anovulation.

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