The effect of age, type and duration of infertility on prolactin concentration in the serum of hyperprolactinemic infertile women

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Summary :

Background: *Physiological prolactin level is necessary for normal GnRH secretion and necessary for the maintenance of luteal function. High prolactin secretion may interfere with the ovulation through inhibition of gonadotropin secretion and with the function of corpus luteum as demonstrated by short luteal phase and decrease progesterone.*

Objectives: *The objectives of this study were to 1) determine the upper normal value of prolactin hormone in Iraqi women and its range; 2) study the effect of age, type and duration of infertility on prolactin concentration.*

Materials and Methods: *One hundred forty seven hyperprolactinemic infertile women were enrolled in this study. Those were compared with 125 control women. Serum prolactin hormone were estimated in cycle day 2, 12 and 21.*

Results: *The present study showed that the prolactin level in the serum of hyperprolactinemic infertile patients were significantly higher compared to control group in regard to the age of patients, duration, and type of infertility. There were no significant differences in the level of prolactin hormone between primary and secondary infertile patients.*

Conclusion: *The upper limit for normal prolactin value in small sample Iraqi women was 20ng/ml with a range between 5-9 and 20 ng/ml. Age has a significant effect on prolactin concentration in infertile hyperprolactinemic women. Prolactin increased with increasing the duration of infertility.*

Introduction

Among the possible causes of female infertility are the hormonal disturbances and part of these disturbances may be attributed to hyperprolactinemia (Ben-David and Schenker, 1982; Clayton et al., 1986).

In fact the principle mood of presentation of hyperprolactinemia is infertility. Ten to fifteen percent of patients with infertility have hyperprolactinemia.

Physiological prolactin level is necessary for normal GnRH secretion (Tennekoon and Lenton, 1993) and necessary also for the maintenance of luteal function (del-Pozo et al., 1994).

Prolactin is synthesized and secreted from the lactotrophs, the acidophilic cells in the adenohypophysis (anterior pituitary), which constitute 40-50% of the total pituitary cell population (Yen and Jaffe, 1999).

Prolactin has been considered to have an inhibitory effect on ovarian function through the suppression of centrally derived gonadotropin secretion. It has a direct role in modifying granulose cell function, so it has a role in follicular development (Picazo et al., 2000).

Prolactin release is governed primarily by an inhibitory influence from the hypothalamus (Blackwell et al., 1998). It is released in pulses of varying amplitude superimposed on continuous basal secretion (Sievertsen et al., 1980).

Dopamine is the most important inhibiting factor from the hypothalamus, and there are high affinity dopamine receptors on the lactotrophs on which dopamine acts (Gibbs and Neill, 1978).

Materials and Methods:

One hundred forty seven hyperprolactinemic infertile women were entrolled in this study, attending IVF Institute for Embryo Research and Infertility Treatment, during the period of October 2001 to May 2002. Those were compared with 125 control group women, seventy five of them were normoprolactinemic infertile, and fifty were fertile (conceived within less than 1 year). The age range for control group was between 21 and 43 years (mean 31.5) and for hyperprolactinemic group was

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between 22 and 45 years (mean 33.52) and with duration of infertility from 2-20 years (mean 9.18).

Careful history was obtained from patients including the age, duration and type of infertility, the present infertility problem.

The patients were send for basal hormonal investigation and ultrasound. Hormonal assay include serum prolactin estimation in cycle day 2, 12 and 21.

Samples of blood for hormonal estimation were collected from patients between 9.00 AM and 11.00 AM. The patients were put into relatively calm quiet room, stressful venipuncture was avoided as much as possible, some were in a fasting state others were at least 3-4 hours away from the last meal (breakfast). Prolactin measurement was estimated for at least 2 occasions before recording it.

The patients were divided into 3 groups according to age (20-30, 31-40 and above 40 years group). Also they had been divided into 4 groups duration of infertility (2-5, 6-10, 11-15 and above 15 years groups), and according to the type of infertility, they were divided into primary and secondary infertility groups.

Hormonal Estimation:

1 - Sample Preparation:

Blood sample was taken from the patients and prepared by centrifugation (3000-RPM for 10 minutes). The serum was collected and frozen at -20 degree centigrade in freezer until examination. Hormones were examined by instrument called mini VIDAS (made in France by Bio Mericux company in 1992, model VIDAS 12). The technique of enzyme immunoassay was used for the determination of human prolactin in human serum using ELFA (Enzyme linked fluorescent assay) technique.

The principle of this machine is automated assay, which enables human prolactin in serum to be quantitively measured.

2 - Assay Kit:

A – The single reagent strip has ten wells the first one is an empty well in which to place the sample, the next eight wells contain reagent or washed, the last well is optical cuvette where the substrate reaction is measured, from its strip fluorescent reading.

B - SPR (solid phase Recplacte):

The SPR is a specially designed plastic pipette – shaped device.

During manufacturing, the interior of the SPR is coated with antibody, antigen, or other treatment that capture a target analyzes.

Each SPR has a corresponding mini VIDAS reagent strip is included in the test kit, both are coded with matching color dots and assay code.

Results:

The effect of the age of the normoprolactinemic infertile women on prolactin concentration was studied. Although the concentration of prolactin (15.34 ± 2.14 ng/ml) in the more than 40 years old women was higher than the other 2 groups (14.54 ± 0.74 ng/ml in the 31-40 years old and 12.88 ± 0.86 ng/ml in 20-30 years old) but it was not significant (Table 1). The concentration of prolactin in the serum of fertile women with a mean age of 32.5 years, and with a range of 20-45 years old women is shown in table number 2. The mean concentration of prolactin was 11.72 ± 0.52 in cycle day 12 of the menstrual cycle.

The concentration of prolactin in the hyperprolactinemic infertile groups was compared to its value in the control fertile (Normoprolactinemic) group according to the age of the patients. In cycle day 2, 12 and 21 (Figure 1, 2 and 3 respectively), the concentration of prolactin in the 3 aged groups were significantly higher ($P<0.0001$) than the control group. The prolactin concentration in the above 40 years age group was significantly higher ($P<0.05$) than that of 20-30 years age group.

Figure 4 shows the concentration of prolactin in the serum of infertile hyperprolactinemic patients according to the duration of infertility and compared to its value in the control (Normoprolactinemic) group in cycle day 2, and it was found that the prolactin concentrations in the 4 different ranges in the duration of infertility were significantly different ($P<0.0001$) than that in the control group. Moreover its concentration in the above 15 years duration of infertility was significantly higher ($P<0.05$) than that in the 2-5 years and 6-10 years duration of infertility.

The same previous comparison of prolactin concentration between hyperprolactinemic and control groups is shown in figures 5 and 6 with a significant difference ($P<0.0001$) between them in cycle days 12 and 21 respectively, and a significant difference also ($P<0.05$) between the above 15 years and 2-5 years duration of infertility.

The hyperprolactinemic patients were divided into primary and secondary infertility groups. The concentration of prolactin hormone in the primary infertility groups was 41.54 ± 3.48 ng/ml, and that in secondary infertility group was 37.33 ± 2.53 ng/ml. The difference between the 2 groups was of no significant difference, but their concentrations in these groups were significantly higher ($P<0.001$) than the control group (Figure 7).

Prolactin Conc. (ng/ml) Patient Number	Age of normoprolactinemic infertile women		
	20-30 years	31-40 years	>40 years
1	13.3	12.3	18.2
2	17.7	16.2	19.9
3	8.3	12.8	12.49
4	7.2	16.9	7.5
5	19.6	14.3	6.3
6	18.7	20	7.9
7	17.3	17.3	10.3
8	9.3	7.8	12.4
9	8.9	10.8	15.3
10	13.3	8.4	18.9
11	9.8	19.2	12.9
12	8.5	18.8	13.5
13	17.2	17.5	10.3
14	19.5	19.6	9.2
15	12.3	12.5	6.4
16	11.4	14.9	7.8
17	10.3	20.1	12.9
18	20.1	16.5	20.1
19	16.9	15.9	13.1
20	14.5	11.2	10.1
21	10.9	13.8	16.3
22	11.8	12.1	15.9
23	9.2	14.9	14.2
24	7.5	10.3	18.1
25	8.4	9.5	16.7
Mean + SEM	12.88 + 0.86*	14.54 + 0.74	15.34 + 2.14

* P>0.05 No significant difference from other age groups.

Table (2): Prolactin concentration in the serum of normoprolactinemic fertile women in cycle day 12.

	Fertile normoprolactinemic women 20-45 years			
	Patients number	Prolactin concentration	Patients number	Prolactin concentration
Prolactin concentration (ng/mL)	1	8.9	26	10.3
	2	9.2	27	8.9
	3	7.3	28	14.5
	4	10.2	29	13.8
	5	11.4	30	16.1
	6	12.5	31	17.3
	7	6.3	32	15.2
	8	8.7	33	10.9
	9	10.5	34	12.3
	10	13.4	35	8.2
	11	15.9	36	7.6
	12	12.9	37	6.9
	13	11.8	38	13.1
	14	10.6	39	11.7
	15	7.9	40	18.1
	16	5.9	41	19.1
	17	12.4	42	8.4
	18	17.3	43	20.0
	19	15.2	44	9.2
	20	14.8	45	7.8
	21	13.2	46	15.1
	22	9.5	47	11.4
	23	10.9	48	12.1
	24	19.1	49	6.5
	25	8.3	50	7.9
Mean + SEM	11.72 + 0.52			

Discussion:

In order to have an idea about the normal prolactin (PRL) concentration in Iraqi females, this study showed that its mean concentration in the inferLile women was 14.25 ng/ml with slight increase in relation to increasing age. The range of PRL concentration was between 6.3 and 20.1 ng/ml in infertile women. The mean concentration in normal fertile was 11.72 ng/ml with a range of 5.6 to 20 ng/ml. The difference in PRL concentration between fertile and infertile women were not significant (P 0.05). the upper limit for normal range is considered to be 15 ng/ml in other women (Breckwoldit et al 1994). Gunarantne in 1993 recorded that the normal range is between 3 and 21.6 ng/ml. The results of this study goes in agreement with the results of Batrinos et al. (1994) who reported that the normal PRL values can be considered up lo 20 ng/ml and values between 20 and 30 ng/ml considered to be hyperprolactincmic.

These differences in the normal range between different reports may be due Lo the variation between laboratories and the pulsatile nature of PRL release (Gunarantne, 1993). Also it may be due to the style of life including stress and food quality.

The concentration of PRL in the serum of infertile hyperprolactinemic patients was significantly different from that in fertile control group. The older age group (>40 years) has PRL concentration significantly higher than that in the younger age group (20-30 years). This is due to increasing stress with increasing age, which is associated with alteration in hypothalamic and pituitary functions (Greenspan et al., 1990). In addition to the life style, psychological status of the patient and embargo of Iraqi infertile women. With the increase in patients age there is age related changes in PRL cell population as their volume density is higher with older age as well as PRL secretion (Console ct al., 1997). In addition to that PRL concentration in patients with pituitary adenoma (Prolactinoma) was significantly lower in those less than 30 years than those >40 years (Yonezawa et aL, 1997). Animal work showed that the level of PR1 receptor mRNA in choroids plexus, periventricular area of the preoptic and arcuate nuclei increased significantly by the time the animals were old (Chiu and Wise. 1996).

The differences between older and younger age groups were noted in cycle days 2, 12 and 21. This may be due to the presence of stress all over the cycle days in older age group. Other investigators found no significant effect of age on PRL concentration (Greenspan et al., 1990; Okada et al., 1996).

Patients with duration of infertility of more than 15 years had PRL level significantly higher than those with 2-5 years. This may be also due to the stress of increasing duration of infertility and the increase of age, which accompanies it. The difference in duration of infertility in regard to PRL concentration was significantly different from control group. This is in good agreement with the results reported by Eggert-Kruseetal.. (1991).

Concerning the type of infertility, PRL concentration in primary type was slightly higher but not significant (P>0.05) than that of the secondary one. This agrees with the study done by Parra et al. (1997) who found that parious females have higher dopamine tone (i.e. lower PRL concentration than nalliparocs females).

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