Evaluation of Ovarian Reserve Based on Hormonal Parameters, Ovarian Volume and Antral Follicle Count in Infertile Women

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ABSTRACT:

Back Ground: It is important to regard the value of ovarian reserve in the evaluation of infertile women, because diminish in the ovarian reserve is a decrease in the quantity and quality of oocyte leading to impaired fertility**.**

Aim: To investigate the risk factors associated with diminished ovarian reserve.

Patients & Methods: This Case -control study conducted on 147seven women attending the infertility clinic in Al-Batool Teaching Hospital from 1st march to 1st November 2010 are included in this study. All women are in good health and not known to have any medication in the previous month which might affect her hormonal profile. Ovarian cyst was excluded. All women evaluated by history and examination, review of previous investigation. Clomiphene citrate challenge test and basal FSH hormone was performed to detect women with abnormal ovarian reserve. 86 infertile women proved to have diminished ovarian reserve were considered as cases. The control 61 infertile women with normal ovarian reserve .Different variable were assessed in women with diminished ovarian reserve including female age, BMI, causes of infertility, menstrual irregularity, type of infertility, history of pelvic operation.

Results: The results of examining main risk factors for developing diminished ovarian reserve appeared as following. unovulatory cycle ,unexplained infertility and hyperprolactineamia ,they were found significantly to be associated with diminished ovarian reserve. Women more 35 years old with history of irregular menstrual cycle and duration of infertility more than 10 years of primary type, history of operation on the ovaries, BMI<30 were all found to be highly significant for diminished ovarian reserve. Other variable like size of ovaries between (6.2-6.6), size of antral follicles between (0.63000 to 0.89477) were significant as risk factor for diminished ovarian reserve

Conclusion: Women more than 35years old, their BMI<30 with irregular unovulatry menstrual cycle and unexplained type infertility, more than 10 years duration of infertility, have positive family history of premature menopause and history of elecrothermal operation on the ovaries. Size of the ovaries between (6.2-6.6), size of antral follicles between (0.63000 & 0.89477); all regarded risk factors for diminished ovarian reserve.

Recommendations: The assessment of ovarian reserve help provide valuable information about status of ovarian function .This may help a couple make a more informed decision concerning treatment option and the risk factors affecting their ability to conceive and may to advice all women for screen for diminished ovarian reserve.

Key wards: Risk, Factors, Diminished Ovarian Reserve, Hyperprolactineamia, Polycystic ovaries.

Introduction:

Ovarian reserve testing is a critical part of an infertility evaluation in which we test the reproductive potential of the eggs. [1] Several methods have been used for its determination these include:

- Biochemical parameters such as estradiol, FSH/LH levels and ratio, inhibine-B and anti-mullerian hormone levels [2,3].
- Sonographic measurement of ovarian volume, antral follicle count & follicular volume.
- Dynamic tests of ovarian function such as clomiphene citrate challenge test exogenous FSH ovarian reserve test (EFORT) [4] and GnRH agonist stimulation (GAST) [5]
- Ovarian biopsy to determine follicular density [6].

With ovarian ageing, levels of oestradiol (E2) and inhibine-B decline, while level of FSH rise. These markers constituting the classical hypothalamus–pituitary gonadal feedback loop are interdependent. [7] The Clomiphene Challenge Test (CCT), which may provide an earlier warning sign of diminished ovarian reserve than measuring baseline FSH alone .It can identify patients with "diminished ovarian reserve" that was not detected with baseline FSH measurements [8].

It is simple test involves determination of the day 3-5 FSH level, administration of clomiphene 100 mg per day on days 5-9 and re-testing of the FSH level on day 10 which is often elevated in women with diminished ovarian reserve.[9] AMH, inhibine-B level a recently added markers to assess ovarian reserve, both decline with age.

The decrease occurring earlier than changes in FSH; the lower the AMH level the lower the fertility potential. (10)

The sonographic variables used for assessing ovarian function include ovarian volume, antral follicle count (AFC) antral follicle count (Tomás *et al*., 1997; Chang *et al*., 1998; Frattarelli *et al*., 2000; Ng *et al*., 2000b; Hsieh *et al*., 2001; Nahum *et al*., 2001; Bancsi *et al*., 2002). (11,10)

Finally; an ovarian biopsy conducted to determine follicular density directly. It is invasive procedure obtained by laparoscopy carries the risk of later adhesion formation. [12] In this study we choice to apply basal measurement of FSH and CCCT in combination with transvaginal ultrasound for assessing women with diminished ovarian reserve because these tests are simple, inexpensive and routinely available. [13]

Researchers have explored links between diminished ovarian reserve and variables like severe endometriosis or trauma (such as might be experienced during surgical procedures to remove ovarian cysts or endometriomas), environmental exposure (cigarette smoking) or pelvic radiation, certain chemotherapies [14].

In spite of dramatic advance in infertility treatment, female age related infertility remain big challenge, age is one of the most important factors affecting female fertility. Absence of ovulation is important predictor of reduced fertility and reduces ovarian reserve as it may indicate disturbances of the hypothalamic pituitary ovarian axis. [15]

Patients and Method:

Over a period of 6 months, 148 women attending the infertility clinic are included in this study .All women are in good health and not known to have any medication in the previous month which might affect her hormonal profile. Ovarian cyst was excluded. All women evaluated by history and examination review of previous investigation. Using transvaginal ultrasound, the volume of each ovary can be calculated by measuring the length, width and depth. Normal dimensions are generally considered to be 1.5cm x 2 cm x 3.5 cm. and the number of antral follicle. AFC is a sum of follicles measuring 2-10 mm in diameter in both ovaries; the total follicular volume is a sum of the volumes of all the follicles up to 10 mm in diameter in both ovaries.

Body mass index was measured as weight in kilogram per square of height in meter (Kg/m2). The women were categorized as follows: Severely thin<16.9, under weight 17–18.4, desirable weight 18.5–24.9, over weight 25–29.9 and obese when BMI was>30 Kg/m2.

Interventions:

1. Trasvaginal ultrasound day 3 to measure the size of the ovaries, number of antral follicles size of antral follicles.
2. Measurement of basal FSH and E2 level day 3 of cycle.
3. Clomephen challenge test was carried out by giving clomiphene citrate at dose of 100g/day from day 5-9.
4. FSH with E2 level reassessed at day 10.

MINI VIDAS is used for measurement FSH and E2 level, serum measured by automated quantitative test Using the EIFA technique (Enzyme limited Fluorescent Assay). FSH level >10miu/ml and E2 level, >80pg.ml suggest the diagnosis of diminished ovarian reserve in 81 infertile women.

The control group 61 women with normal ovarian reserve. Statistical analysis of the results was assessed by multivariate analysis of variance to study p value, 95% CI.

Different variable assessed in women to examine the main risk factors with DOR including different causes of infertility , women age, BMI . Menstrual cycle abnormality, history of electro thermal surgery on ovaries, ovaries size and antral follicles number and size by base line vaginal ultrasound.

Results:

The results of examining main risk factors for developing DOR appeared as following.

Table (1) shows distributions of cases and controls according to causes of infertility and reveals ovulatory dysfunction is significantly associated with DOR (OR= 2.36, P- value0.043 and CI 1.015-5.523).

Endometriosis is found to be highly and significantly associated with DOR (OR =3.76, P-value 0.034 and 95% CI 1.031-13.711).

Unexplained causes of infertility is found significantly associated with DOR (OR=3.18, P-value 0.0025 and 95% CI 0.131-9.19).

Table 1: Distribution of risk factors according to causes of infertility.

Causes	Cases (n=86)		Controls (n=61)		OR	χ^2	p-value	95 %Confidence interval	
	NO.	%	NO.	%				Lower limit	Upper limit
An ovulation	25	29.07	9	14.75	2.36	4.114	0.043	1.015	5.523
Endometrioses	14	16.28	3	4.92	3.76	4.504	0.034	1.031	13.711
Unexplained infertility	19	22.09	5	8.20	3.18	5.045	0.025	1.131	9.191
Hyper prolactinemia	9	10.47	8	13.11	0.77	0.117	0.732	0.281	2.136
Tubal factor	10	11.63	13	21.32	0.49	2.536	0.111	0.197	1.195
Male factor	5	5.81	9	14.75	0.36	3.310	0.069	0.113	1.123

Table (2) reveal that women age>30 years (OR=2.69, P-value 0.004, and 95% CI 1.355-3417), is highly significant for developing DOR. Obese women with BM I > 30 (OR =2.29, P-value 0.005, and 95% CI 1.339-5.794) is also found to be associated with reduction in the ovarian reserve. Women with duration of infertility more than 10 years (OR=3.06, P-value 0.001, 95% CI 1.544-6.063), with abnormality in the menstrual cycle in form of irregular period (OR=2.19, P-value 0.022 and 95% CI 1.113-4.311) were all found to be associated with the development of DOR.

Women with history of electro thermal surgery on the ovaries is highly significant for developing

DOR. (OR=2.12,P-value 0.037, and 95% CI 1.041-4.331) .Presence of positive history of premature menopause is another risk factor found to be associated with reduction in the ovarian reserve.

Table 2: Distribution of DOR according to different women variable.

Age	Cases (n=86)		Controls (n=61)		OR	χ^2	p-value	95 % C.I	
	NO.	%	NO.	%				Lower limit	Upper limit
>35	61	70.93	29	47.54	2.69	8.224	0.004	1.357	5.341
<35	25	29.07	32	52.45					
BMI									
<18	9	10.47	17	27.87	0.30	7.425	0.006	0.124	0.736
18-25	22	25.58	19	31.15	0.076	0.550	0.458	0.367	1.572
25-30	16	18.60	11	18.03	1.04	0.008	0.930	0.444	2.429
>30	39	45.35	14	22.95	2.79	7.765	0.005	1.339	5.794
Menstrual cycle									
Regular cycle	24	27.91	23	37.70	0.64	1.575	0.209	0.318	1.288
Irregular	46	53.49	21	34.43	2.19	5.228	0.022	1.113	4.311
Oligomenorrha	16	18.60	17	27.87	0.59	1.124	0.289	0.271	1.29
Ovarian surgery									
No surgery	29	33.72	28	45.90	0.60	2.230	0.135	0.306	1.176
Oopherictomy	9	10.47	8	13.11	0.77	0.245	0.621	0.281	2.136
Cystectomy	11	12.79	9	14.76	0.847	0.117	0.732	0.328	2.19
Electrothermy	37	43.02	16	26.23	2.12	4.366	0.037	1.041	4.331

Table (3): other variables like size of ovaries between (6.2-6.6). and size of antral follicles between (0.63000 to 0.89477) are highly significant risk factor for DOR.

Table 3: Distribution of DOR according to ovarian size and size of antral follicles.

Size of ovaries	Cases (n=86)	Controls (n=61)	p-value
	Mean ± S.d	Mean ± S.d	
R.ovary	6.9245 ± 3.0348	7.4828 ± 2.7372	0.25
L.ovary	6.6826 ± 2.3985	7.2623 ± 2.9413	0.21
Size of follicles			
R.ovary	0.89477 ± 0.59778	0.52377 ± 0.20971	0.0000
L.ovary	0.63000 ± 0.33392	0.43115± 0.19696	0.0000

Discussion:

Recent study have been reveled that there are potential causes for infertility that are not diagnosed by routine evaluation, one of these is ovarian reserve which is defined as reduction in the women remaining oocyte to point that the probability of producing viable gestation is low.[16]

Amongst the main causes of female infertility found to be associated with diminishes in their ovarian reserve in this study are chronic unovualtion (P-value 0.043, OR 2.36) women with menstrual abnormality in form of irregular cycle (p-value 0.022, OR 2.19) women with unexplained infertility (P-0.034 ,OR 3.76) women with endometriosis (P-value 0.025, OR 3.18).

These result's similar to study by K. Lutchman 2005 [17] which confirmed that women with polycystic ovarian syndrome have an increased risk of long term of chronic unovualtion , irregular cycles, obesity ,hyperandrogenism, health problems including metabolic syndrome strong familial aggregation suggests genetic basis, all theses regarded as risk factors for diminished ovarian reserve. Scott, RT. et al 2006 [18] in his study found that 30% women with disorder in the ovulation or unexplained causes for their infertility associated with occult ovarian failure.

The reason behind this *Younger women with unexplained infertility should be screened because an abnormal test may approach 50% of these patients*. M.L.Haadsam, 2005[19], Glen E. 2002 [20] found that endometriosis is a major cause of infertility affecting 30-40% of infertile women, is highly significant factor for decrease in ovarian reserve.

In cases with endometriosis the eggs in the ovaries can be damaged resulting in decreased ovarian reserve and reduced egg quantity and quality

This study found that women >35 years of age at very high risk for having diminishes in ovarian reserve.

Our result similar to a study done by Monle D. et al., 2008 [21] concluded that diminished reserve did not affect the quality of oocytes and any reduction in quality in diminished reserve women was age related. Worldwide experience with in vitro fertilization (IVF) has shown a similar decline in fertility beginning in the mid 30s, with a more rapid decline in fertility after the age of 40s. [22]

Obesity is associated with many medical conditions, as well as social and psychological disorders, and links between obesity and fertility-related disorders have recently been recognized [23]. Women with BMI >30 in our study group are significantly at high risk of developing diminished ovarian reserve, this result proved by the research works of [Wenjie Sun. ,2009][24] who found that body mass index ≥25 Kg/m^2 and irregular cycle appear to be associated with diminished ovarian reserve and severe endocrine and metabolic abnormalities. Gleicher N. 2009 [25] found that Basal FSH concentration achieved the best predictive value for diminished ovarian in relation to the number of oocytes obtained, followed by BMI.

In our study women with history previous surgery on the ovaries inform of ovarian electro diathermy or oohperectomy or cystectomy are associated with significant risk for decrease in ovarian reserve .Same result found by study done in Haseki Education and Research Hospital, Istanbul, Turkey 2010; there were statistically significant differences between Day 3 FSH, inhibin B levels, ovarian volume and antral follicle count before and after LOD(laparoscope ovarian drilling) in women with polycystic ovaries (PCOS) .

Although the after LOD values were found to be lower than the before LOD values by means of ovarian reserve markers, the after values stayed higher than normal when compared with normal women without PCOS. [23,25] Daniel A. 2007 study shows the main risk factors for diminished ovarian reserve include age > 35, previous ovarian surgery, single ovary, unexplained infertility, significant pelvic surgery, pelvic infection. [Cramer et., 1995] [26]has been found that women who had lost one ovary at an early age –particulary before the age of 35years old had strong risk factor for early menopause.

The number of primordial follicles appears to be correlated with the number of growing follicles (Gougeon, 1984) [27].

The decline in primordial follicle reserve leads to a decrease in size of the antral follicle cohort [Schefer, 1999] [28].

This study found that there is significant correlation between size of the ovaries , number of the antral follicles with the development of diminished ovarian reserve. In an 'Ovarian Cancer Screening' program conducted at the University of Kentucky, involving 13,963 women who underwent transvaginal sonography annually with basal FSH , a statistically significant decrease in ovarian volume and reserve was shown with each decade of life from 30 to 70 years[29].

There is a relationship between follicle count, size of ovaries, ovarian reserve, age <40 years, serum day 3 FSH levels of < 10 mlU/mL. [Pache, 1990]. [30]. in women with small ovaries (<3cm^2) the cancellation rate of IVF is higher (like hood ratio 3.8-6.8). [31]

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