Serum Anti-Mullerian Hormone Levels in Early Follicular Phase Can Predict Ovarian Reserve and Pregnancy Outcome in In-Vitro Fertilization Cycles

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Abstract

Objective: The aim of this study is to investigate whether anti-Mullerian hormone (AMH) levels and antral follicle count (AF) can be useful in predicting the ovarian reserve and pregnancy outcome in IVF cycles.

Study Design: A prospective observational clinical study carried out at Kuwait Maternity Hospital, between March 2007 and December 2008.

Patients and Methods: A total of 60 patients undergoing their first ICSI treatment cycle using a long protocol with GnRH agonist were included. Patients with an oocyte count ≥4 were considered good responders (group I) and those with <4 were considered as poor responders (group II). On day three of a spontaneous menstrual cycle, blood sample was taken from each patient for measurement of serum levels of AMH, FSH, LH, E2, and inhibin B. Thereafter, ovarian ultrasound scanning was performed to assess the number and size of antral follicles.

Results: Parameters such as LH and E2 levels were not statistically different between the two groups. Whereas, the difference between serum AMH, FSH and inhibin B levels; AFC and retrieved oocyte counts were statistically significant in the two groups. The mean ± SD serum anti-Mullerian hormone level was 33.19 ± 12.84 and 11.51 ± 8.41 pmol/L in groups I, II, respectively (p < 0.001), the number of basal antral follicles was 9.73 ± 4.10 versus 2.60 ± 0.81 in normal and poor responders groups, respectively (p < 0.001). Serum AMH levels were highly correlated with the number of antral follicles (r = 0.77; p < 0.01) and the number of oocytes retrieved (r = 0.57; p < 0.01). A negative association was found between AMH levels and poor ovarian response (fewer than 4 oocytes or cycle cancellation; OR 0.80, 95% CI 0.73-0.89 p < 0.001). Inclusion of inhibin B and FSH concentrations to AMH in a multivariate model improved the prediction of ovarian response. The number of chemical pregnancies was 8 (7%) versus 3 (30%), p = 0.013 and the number of clinical pregnancies was 8 (89%) versus 1 (11%), p = 0.011 in groups I and II, respectively.

Conclusion: It appears that there is an association between the serum level of anti-Mullerian hormone in early follicular phase and ovarian reserve. Poor response in IVF treatment cycles, indicative of a diminished ovarian reserve, is associated with reduced baseline serum AMH levels. Furthermore, a higher serum level of AMH on day three is associated with chemical pregnancy success. It appears that AMH can be used as a marker for ovarian ageing.

Key Words: Anti-Mullerian hormone – Antral follicle count – Ovarian reserve – IVF – Poor response.

Introduction

ANTI-MULLERIAN hormone (AMH), a member of the transforming growth factor-β (TGF-β) family, was identified as a factor which is being synthesized by testicular Sertoli cells, induces regression of the Mullerian ducts during male foetal development [1,2]. Since AMH is secreted mainly in preantral and early small antral follicles [3,4], the circulating AMH level decreases through maturing follicle in normal menstrual cycle and further decrease in FSH-treated cycle [5,6]. In ovarian hyperstimulation and IVF cycles, however, continued recruitment of additional antral follicles during the stimulatory phase results in higher AMH levels, thus it may be correlated with the number of mature follicles and number of oocyte retrieved [7,8]. Since AMH is solely produced by the growing ovarian follicles, serum levels may be used as a marker for ovarian reserve, presenting the quantity and quality of the ovarian follicle pool [9]. Recent preliminary reports indeed indicate that AMH levels decline with increasing female age [10] and that initial AMH is associated with ovarian response in IVF patients with normal FSH levels [11,12].

Ovarian reserve comprises two elements: the size of the stock of primordial follicles and the quality of the oocytes. Ovarian reserve may be assessed by static tests such as: measure of early follicular phase basal FSH, inhibin B, morphometric ultrasonographic measures such as antral follicle count (AFC) or ovarian volume [13].
**Aim of the work:**

The aim of the present study is to prospectively assess the significance of AMH as a marker for ovarian reserve in IVF cycles. The predictive performance of serum AMH levels towards poor response in relation to other ovarian reserve tests was investigated. Also to test whether serum concentration of AMH in early follicular phase is associated with ovarian response and pregnancy outcome in patients undergoing IVF programs.

**Patients and Methods**

This study was a prospective observational clinical trial which was approved by the ethical committee of Kuwait Maternity Hospital and Institutional Research Committee between March 2007 and December 2008. 60 infertile patients who were going to have their first ICSI treatment cycle in IVF-center were recruited.

**Subjects:**

All patients aged <40 years and were scheduled for ICSI program. The inclusion criteria were:

1. Regular menstrual cycle (cycle length, 25-35 days; duration of menstruation, 3-8 days).
2. Presence of both ovaries and lack of morphologic abnormalities.
3. No evidence of endocrine disorders (normal prolactin, thyroid stimulating hormones (TSH), free thyroxine (FT$_4$), testosterone and androstenedione).
4. A body mass index (BMI) ranging from 18-27kg/m$^2$.
5. Not on hormone therapy for three months.
6. Adequate visualization of ovarian at transvaginal ultrasound scanning.
7. Written informed consent.

**The exclusion criteria:**

1. Present or past history of ovarian endometriosis [14,15].
2. Ovulatory factor such as polycystic ovary syndrome (PCOS) was entirely excluded since women with PCOS may have unusually higher serum AMH levels during ovarian hyperstimulation [16].
3. Patient age >40 years.

**Hormonal and follicular measurements:**

On day 3 of spontaneous cycle within the 3 months preceding ICSI treatment, patients underwent a transvaginal ultrasound examination to assess the number of antral follicles, measuring 2-5mm as described previously by Bancsi et al. [17]. On the same day, 5ml of venous blood sample was obtained for the measurement of AMH, FSH, LH estradiol (E$_2$) and inhibin B. Serum and plasma samples were centrifuged at the 3500rpm for 10 minutes and all sera were stored at -20ºC until assayed. Serum AMH levels were determined using a “second generation” enzyme-linked immunosorbent assay (ELISA) (reference A16507; Immunotech Beckman Coulter laboratories). Intra- and inter-assay coefficients of variation (CV) were ≤12.2%, and ≤14.1% respectively.

The immunoassay was specific for AMH and the analytical sensitivity was <0.7 pmol/L. Inhibin B concentration were measured using a sensitive (ELISA); (DSL-10-84100, Diagnostic system laboratories, Webster, Texas, USA). The sensitivity was <0.5ng/L. The intra- and inter-assay coefficients of variation were ≤14.3% and ≤13.9%, respectively. Serum FSH, LH and E$_2$, were measured by the same laboratory technician for all patients by ELISA technical.

**Ovarian stimulation protocol:**

The patients were all treated with a long protocol for ovarian hyperstimulation. Pituitary down regulation was achieved by administering buserelin acetate (Superfact, Hoechst AG, Germany) (0.5mg SC) starting from day 21 of menstrual cycle. After menstruation, the ovarian hyper-stimulation was commenced using human menopausal gonadotropin (HMG) (Menogon, Ferring, Germany) from the second day of their menstrual cycle with a dose of 225-300 IU/day. Monitoring was carried out by transvaginal ultrasound (Ultramark 9 sonographic machine developed by Advanced Technology Laboratories) on day seven of HMG stimulation and E$_2$ was measured. After more than three leading follicles of ≥18mm in diameter were observed, 10,000 IU of human chorionic gonadotropin (HCG) (Pregnyl 5000, Organon) was administered intra-muscularly. Thirty-six hours later, oocyte retrieval was performed. Mature oocytes were placed in G-fert (version 3; Vitrolife, Goteborg, Sweden) and after intracytoplasmic sperm injection (ICSI), fertilization was observed; 2PN zygote was transferred to G-1 media (G-1 TM version 3; Vitrolife, Goteborg, Sweden). All embryo-transfers were performed two days after oocyte retrieval using labotec catheters (Labor-Technik, Germany). Before the transfer, embryos were evaluated microscopically and the best-quality embryos were selected for transfer. A maximum of 3-embryos were transferred.
Luteal phase was supported with cyclogest (Alpharma, Barnstaple, UK) with a dose of 1200mg vaginally per day and starting after oocyte retrieval.

**Study outcome:**

The main outcome measures of the study were the number of oocytes retrieved and poor ovarian response. As previously described by Bancsi et al. [17], poor ovarian response was defined as fewer than 4 oocytes at follicle puncture or an cancellation due to impaired (fewer than 3 follicles) or absent follicular growth in response to ovarian hyperstimulation. Patients were considered high responders in case of collection of more than 20 oocytes at ovum retrieval or when the cycle was cancelled due to exaggerated response (more than 30 follicles in both ovaries and/or peak E2 > 15000pmol/L). High response was considered a secondary outcome measure, and in the analysis of high response both the poor and normal responders are considered as one group.

Secondary outcome measures were chemical pregnancy that was defined as serum β-HCG >50mIU/mL, two weeks after ICSI; clinical or ongoing pregnancy that was determined by detection of foetal heart beat by abdominal ultrasound eight weeks after the initiation of ART cycles; and miscarriage that was defined as pregnancy loss before 20 weeks based upon the date of the first day of the last normal menses [18].

The study population was divided into two subgroups according to the number of oocytes retrieved. Patients with an oocyte count of ≥4 were considered normal responders (n=42); patients with <4 oocyte were considered poor responders (n=18).

**Statistical analysis:**

Data were analyzed with the statistical program for social science (SPSS Inc., Chicago, IL, USA), version 13. To compare normal with poor responders, the student's t-test, Chi-square and fisher exact test were used whenever appropriate. The correlation between different variables is expressed as Spearman’s correlation coefficient. Univariate and multivariate logistic regression with the main outcome measure poor ovarian response and secondary outcome measures of high response and ongoing pregnancy were performed. For each single variable used in the univariate analysis and for the models, the ability to discriminate between poor and normal responders was assessed by calculating the area under the receiver operating characteristics curves (ROCAUC) as described by Harrell et al. [19]. The ROCAUC may vary between 0.5 (no discriminative power) to 1.0 (perfect discrimination). p<0.05 was considered statistically significant.

**Results**

Patients clinical characteristics of the complete group and of normal and poor responders separately are presented in Table (1). There was no significant difference between normal and poor responders regarding age, body mass index (BMI), menstrual cycle length, infertility period and diagnosis of primary infertility (p>0.05). As expected, patients with a poor response were more often treated for unexplained infertility than in normal responders [13 (72%) vs 5 (12%), respectively] (p<0.01).

Considering the ovarian reserve test parameters, it was found that serum anti-Mullerian hormone (AMH) and inhibit B levels on day 3 of a spontaneous cycle were significantly higher in the normal than in the poor responders groups (33.19±12.84 vs 11.51±8.41pmol/L) and (117.17±12.35 vs 70.40±12.30ng/L, respectively) (p<0.001). Also, the difference between the basal number of antral follicles (AFC) (11.61±3.06 vs 4.34±3.60), basal serum FSH levels (6.13±3.21 vs 10.46±3.45 mIU/mL) and the number of retrieved oocytes (7.73±4.10 vs 2.60±0.81) were statistically significant in the normal and poor responders groups, respectively (p<0.001).

Table (1): Clinical characteristics in the total group of ICSI patients and in good and poor responders separately.

<table>
<thead>
<tr>
<th>Variables</th>
<th>Total (n=60)</th>
<th>Normal responders (n=42)</th>
<th>Poor responders (n=18)</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (yr)</td>
<td>29.16±3.02</td>
<td>27.51±3.02</td>
<td>30.06±3.21</td>
<td>NS</td>
</tr>
<tr>
<td>Body mass index (kg/m²)</td>
<td>25.32±2.6</td>
<td>24.43±3.2</td>
<td>26.36±4.12</td>
<td>NS</td>
</tr>
<tr>
<td>Menstrual cycle length (days)</td>
<td>28.6±2.7</td>
<td>27.3±1.2</td>
<td>29.5±3.1</td>
<td>NS</td>
</tr>
<tr>
<td>Duration of infertility (yr)</td>
<td>6.64±3.1</td>
<td>6.46±3.2</td>
<td>7.23±3.1</td>
<td>NS</td>
</tr>
<tr>
<td>Primary infertility n (%)</td>
<td>58 (97)</td>
<td>41 (97.6)</td>
<td>17 (94)</td>
<td>NS</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Causes of infertility: Male factor (%)</th>
<th>29 (48.3)</th>
<th>26 (62)</th>
<th>3 (17)</th>
<th>NS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tubal factor (%)</td>
<td>13 (21.6)</td>
<td>11 (26)</td>
<td>2 (11)</td>
<td>NS</td>
</tr>
<tr>
<td>Unexplained (%)</td>
<td>18 (30)</td>
<td>5 (12)</td>
<td>13 (72)</td>
<td>&lt;0.01</td>
</tr>
</tbody>
</table>

Values are presented as mean ±SD or as n (%). NS: Not significant (p>0.05).
No oocyte retrieval took place in two patients with poor response because of insufficient follicular growth. Meanwhile, parameters such as basal serum LH and E-2 were not statistically different ($p>0.05$) (Table 2).

Table (2): Ovarian reserve test characteristics in the studied groups.

<table>
<thead>
<tr>
<th>Variables</th>
<th>Total (n=60)</th>
<th>Normal responders (n=42)</th>
<th>Poor responders (n=18)</th>
<th>$p$ value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Day-3 AMH (pmol/L)</td>
<td>23.72±11.10</td>
<td>33.19±12.84</td>
<td>11.51±8.41</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Day-3 FSH (mIU/mL)</td>
<td>6.71±4.22</td>
<td>6.13±3.21</td>
<td>10.46±3.45</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Day-3 LH (mIU/mL)</td>
<td>5.64±2.51</td>
<td>5.44±2.61</td>
<td>6.78±3.21</td>
<td>NS</td>
</tr>
<tr>
<td>Day-3 E2 (pmol/L)</td>
<td>158.42±11.20</td>
<td>160.35±12.64</td>
<td>158.46±13.60</td>
<td>NS</td>
</tr>
<tr>
<td>Day-3 inhibin B (ng/L)</td>
<td>102.21±10.20</td>
<td>117.17±12.35</td>
<td>70.40±12.30</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>AFC (n)</td>
<td>8.37±3.20</td>
<td>11.61±3.06</td>
<td>4.34±3.60</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Number of retrieved oocytes</td>
<td>6.51±4.04</td>
<td>9.73±4.10</td>
<td>2.60±0.1</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

Values are presented as mean ±SD or as n (%). NS: Not significant ($p>0.05$).

The correlation coefficients for the association between AMH levels on one hand and several ovarian reserve test variables and the total number of oocytes retrieved on the other hand are depicted in (Table 3). AMH was highly correlated with the number of antral follicles (AFC) ($r=0.77$, $p<0.01$) and the number of oocytes retrieved after ovarian hyperstimulation ($r=0.57$, $p<0.01$). In addition, inhibin B was positively correlated with the number of antral follicles (AFC) ($r=0.45$, $p<0.01$) and the number of retrieved oocytes ($r=0.70$, $p<0.01$). Besides AMH, only AFC was significantly correlated with chronological age ($r=0.39$, $p<0.05$). A good correlation was found between AFC and the number of oocytes retrieved ($r=0.58$, $p<0.05$), comparable with that of AMH and number of retrieved oocytes.

The results of the logistic regression analysis for the prediction of poor response are shown in (Table 4). AFC presented the highest $\text{ROC}_{\text{AUC}}$ of 0.85, indicating a good discriminating potential for prediction of poor ovarian response. The $\text{ROC}_{\text{AUC}}$ for AMH (0.84) was almost identical, followed by the AUC values for FSH (0.82) and inhibin B (0.75). Age was not significantly related to poor response. With a multivariate analysis AFC, inhibin B and FSH were selected. Because of the high correlation between AMH and AFC, AMH was not selected in the model. Leaving AFC out of the model led to selection of AMH together with inhibin B and FSH. Both models showed a comparable discriminative potential towards poor ovarian response prediction. When a similar analysis was performed with high response as the outcome measure, AFC and AMH were again the best performing variables. The AUC values were 0.88 and 0.87 for AFC and AMH respectively, in the univariate analysis. AFC and inhibin B were selected in the multivariate analysis. AFC could be exchanged for AMH and giving a similar performing model for the prediction of high response.

Pregnancy outcome of the studied groups are presented in (Table 5). In total, 11 of the 60 couples had a positive β-HCG test. The number of chemical pregnancies was 8 (70%) in normal responders versus 3 (30%) in poor responders ($p=0.01$).

Whereas, the number of clinical pregnancies was 9 (89%) versus 1 (11%) in normal and poor responders, respectively ($p=0.011$). Two pregnancies were aborted before 20 weeks of gestation (one miscarriage occurred in each group), and the remaining of the pregnancies continued as singletons. There was no significant difference in miscarriage rate between the two groups ($p>0.05$).

No oocyte retrieval took place in two patients with poor response because of insufficient follicle growth. One patient with high response had cancelled her cycle (more than 30 follicles in both ovaries).

Table (3): Correlation analysis between serum anti Mullerian hormone (AMH) and several ovarian reserve test variables relevant to number of oocytes retrieved.

<table>
<thead>
<tr>
<th>Variables</th>
<th>AMH (pmol/L)</th>
<th>Inhibin B (ng/L)</th>
<th>FSH (mIU/mL)</th>
<th>No. of antral follicles</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of retrieved oocytes</td>
<td>$r=0.57$</td>
<td>$r=0.70$</td>
<td>$r=0.47$</td>
<td>$r=0.58$</td>
</tr>
<tr>
<td>$p&lt;0.01$</td>
<td>$p&lt;0.01$</td>
<td>$p&lt;0.01$</td>
<td>$p&lt;0.01$</td>
<td></td>
</tr>
<tr>
<td>No. of antral follicles</td>
<td>$r=0.77$</td>
<td>$r=0.45$</td>
<td>$r=0.46$</td>
<td>–</td>
</tr>
<tr>
<td>$p&lt;0.01$</td>
<td>$p&lt;0.01$</td>
<td>$p&lt;0.01$</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>Inhibin B (ng/L)</td>
<td>$r=0.52$</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>$p&lt;0.01$</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>FSH (mIU/mL)</td>
<td>$r=0.53$</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>$p&lt;0.01$</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>E2 (pmol/L)</td>
<td>$r=0.36$</td>
<td>$r=0.35$</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>$p&lt;0.05$</td>
<td>$p&lt;0.05$</td>
<td>–</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>Age (yr)</td>
<td>$r=0.30$</td>
<td>–</td>
<td>–</td>
<td>$r=0.39$</td>
</tr>
<tr>
<td>$p&lt;0.05$</td>
<td>–</td>
<td>–</td>
<td>$p&lt;0.05$</td>
<td></td>
</tr>
</tbody>
</table>

$r$ is spearman's correlation coefficient.
Table (4): Logistic regression for prediction of poor response following ovarian hyperstimulation in ICSI cycles.

<table>
<thead>
<tr>
<th>Variables</th>
<th>Odds ratio (95% CL)</th>
<th>ROC AUC</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>AFC (per follicle)</td>
<td>0.72 (0.59-0.81)</td>
<td>0.85</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>AMH (per pmol/mL)</td>
<td>0.80 (0.73-0.89)</td>
<td>0.84</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>FSH (per mIU/mL)</td>
<td>1.38 (1.18-1.60)</td>
<td>0.82</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Inhibin B (per ng/L)</td>
<td>0.97 (0.96-0.99)</td>
<td>0.75</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Age (per year)</td>
<td>1.06 (0.97-1.13)</td>
<td>0</td>
<td>NS</td>
</tr>
<tr>
<td>E2 (per pmol/mL)</td>
<td>1.02 (1-1.01)</td>
<td>0.52</td>
<td>NS</td>
</tr>
</tbody>
</table>

Multivariate analysis

<table>
<thead>
<tr>
<th>Variables</th>
<th>OR (95% CL)</th>
<th>ROC AUC (Final model)</th>
</tr>
</thead>
<tbody>
<tr>
<td>All variables</td>
<td>0.79 (0.63-0.90)</td>
<td>0.001</td>
</tr>
<tr>
<td>AFC excluded from analysis</td>
<td>0.79 (0.63-0.90)</td>
<td>0.001</td>
</tr>
<tr>
<td>AMH (per pmol/mL)</td>
<td>0.88 (0.80-97)</td>
<td>0.16</td>
</tr>
<tr>
<td>Inhibin B (per ng/L)</td>
<td>0.97 (0.96-0.99)</td>
<td>0.004</td>
</tr>
<tr>
<td>FSH (per mIU/mL)</td>
<td>1.24 (1.03-1.47)</td>
<td>0.005</td>
</tr>
</tbody>
</table>

Table (5): Pregnancy outcome in the studied groups.

<table>
<thead>
<tr>
<th>Variables</th>
<th>Total (n=60)</th>
<th>Normal responders (n=42)</th>
<th>Poor responders (n=18)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of chemical pregnancies</td>
<td>11</td>
<td>8 (70%)</td>
<td>3 (30%)</td>
<td>0.013</td>
</tr>
<tr>
<td>Number of clinical pregnancies</td>
<td>9</td>
<td>8 (89%)</td>
<td>1 (11%)</td>
<td>0.011</td>
</tr>
<tr>
<td>Number of miscarriages</td>
<td>2</td>
<td>1 (50%)</td>
<td>1 (50%)</td>
<td>NS</td>
</tr>
</tbody>
</table>

Values are presented as n (%).

**Discussion**

This study was prospectively investigated whether serum AMH levels can predict ovarian response during first IVF treatment cycles. High correlation of serum AMH in early follicular phase and the ovarian response as expressed by the number of retrieved oocytes, was observed in the current study and this concurs with the results reported by Van Rooi J et al. [20] and Firozabae et al. [21].

Ovarian response during exposure to high levels of gonadotrophins can be considered to be a measure of the selectable cohort of antral follicles. As this number of antral follicles appears to be related to the size of the primordial follicle [22], ovarian response can be regarded a reflection of the ovarian reserve. Recent studies have shown that a low response to exogenous gonadotrophin stimulation is associated with an early menopause, supporting the idea that ovarian response indeed reflects the ovarian aging process [23,24]. The excellent correlation between initial AMH levels and subsequent ovarian response in IVF therefore implies that AMH is a promising marker for ovarian reserve.

It was found that serum AMH levels were highly correlated with the number of antral follicles and the number of retrieved oocyte than did E2, FSH or LH on day 3 of the cycle. In normal responders, with increasing AMH levels, the antral follicle, the growing follicle, and oocyte retrieval counts would also increase. These results were in agreement with those of Bancsi et al. [17] and Van Rooi J et al. [20].

Fanchin et al. found that serum AMH levels were more strongly correlated with antral follicle counts than did the serum levels of inhibin B, E2, LH and FSH.

Ovarian reserve comprises two elements: the size of stock of primordial follicles and the quality of the oocytes [26]. From the primordial follicle pool, primary follicles will start a maturation process and develop through secondary (preantral) follicles into the pool of antral follicles from which the monthly follicle to be ovulated is selected [27]. AMH protein expression beings in the third trimester of gestation, actually long before it can be detected in serum. It is also seen in granular cells of follicles from primary stage up to the larger antral stage when follicles have gained FSH dependence [28]. As a result it thought to play an important role in early follicular development. Once ovarian cycles begin, serum AMH levels vary slightly from baseline due to the stimulation of a small cohort of follicles and loss of AMH production from corpus luteum [29,30]. The present study showed an association between AMH level, antral follicles and retrieved oocyte counts. Therefore, serum AMH levels may reflect the size of the antral follicle pool and hence, may provide a marker associated with the anticipated number of oocytes.
to retrieved after controlled ovarian stimulation for IVF/ICSI.

Since the size of the primordial follicle stock is difficult to measure directly, a marker that reflects all numbers of follicles that have made the transition from the primordial follicle pool to the growing pool may be a good indirect measurement. AMH might be such a marker, as it is involved in the regulation of primordial follicle recruitment [31,32], an important mechanism for the depletion of the primordial follicle pool [33] and it is produced by all follicle stages until FSH-dependency recently. Ficicioglu and associates [34] have found that there was an association between AMH levels, antral follicle counts and the number of retrieved oocyte. Also, Seifer et al. [35] demonstrated that a higher day three serum AMH concentrations were associated with greater number of retrieved oocytes. In their study, the mean serum AMH levels was more than two and a half-fold in the group with ≥11 oocytes retrieved compared to the group with <6 oocytes. In the present study, it was observed that patients with <4 retrieved oocytes had lower day three AMH concentrations and fewer antral follicles. Thus, the basal antral follicle count and basal AMH levels are good tools to use in counseling patients [25,35,36].

The combination of three ovarian reserve tests, AFC, inhibin B and FSH, in a multivariate logistic model appeared to improve the response prediction. In the present study, AMH is found to have a predictive performance comparable with that of AFC. The multivariate analysis on the prediction of poor ovarian response to hyperstimulation in the study revealed that AMH will contribute independently to this prediction but only if AFC is removed from the analysis. This agrees with that reported by Fanchin et al. [37]; Louafi et al. [38] and Visser et al. [39]. There are advantages of the use of AMH over AFC in the multivariate model for the prediction of ovarian response, since all predictive information is obtained with blood sampling and no extra ultrasound is needed. This concurs with that concluded by Van Rooij et al. [20]; Harrell et al. [40] and Scheffer et al. [41].

Application of AMH in the current study, AFC Bancsi et al. [17], inhibin B Seifer et al. [42]; Hall et al. [43] and FSH Sharif et al. [44] and Bancsi et al. [45] as a predictor of ongoing pregnancy appears to be limited in view of the fact that they only present the quantitative aspect of ovarian reserve, whereas pregnancy is also dependent on the oocyte quality. Furthermore, it is also possible for patients with normal ovarian reserve not to become pregnant for instance as a result of fertilization failure. Nevertheless, the ability to predict poor response may be a valuable tool for patient counseling.

In the present study, it was found that a higher serum AMH level has a relation with chemical pregnancy outcome and that it does not have any correlation with clinical pregnancy and miscarriage rates. This agrees with results reported by Firouzabadi et al. [21] and Deffieux et al. [46].

Another study by Muttukrishna et al. [47] showed that AFC has a significant association with the number of retrieved oocytes and is a predictive clinical pregnancy.

Lately, Ficicioglu et al. [48] have found that levels of AMH predict the number of oocytes with a positive predictive value of 96%, although it has little value for prediction of pregnancy.

Conclusion:

It appears that AMH serum levels are associated with ovarian response in stimulated IVF cycles and can be served as a novel marker for ovarian reserve. The predictive value of AMH for poor ovarian response is comparable with that of AFC and therefore AFC could be replaced by AMH in the prediction of ovarian response to controlled ovarian hyper-stimulation in IVF. With respect to significant difference in chemical pregnancy outcome, serum levels of AMH may be used as a marker for predicting the chemical pregnancy rate. However, further studies are needed to determine whether AMH can accurately predict the IVF outcome or not.

References

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