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# PREDICTION OF OOCYTE OUTPUT: How can we maximize the oocyte retrieving from follicles in Controlled Ovarian Hyperstimulation cycles?

# OOSİT ÇIKIŞININ ÖNGÖRÜLMESİ : Kontrollü Ovaryan Hiperstimulasyon sikluslarında foliküllerden çıkan oosit sayısını nasıl maksimize edebiliriz?

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## ÖZ

**Amaç:** Üremeye Yardımcı Teknik uygulamalarında oosit çıktısını öngörerek, daha çok oosit toplamak ve in vitro fertilizasyon (IVF) başarısını arttırmak için hangi değişkenleri kullanmamız gerektiğini incelemek amacıyla çalışmamızı tasarladık.

**Gereç ve Yöntemler:** 2016-2018 arasında bir üniversite infertilite merkezine başvuran 412 infertil hasta retrospektif olarak değerlendirildi. Kontrollü ovaryan hiperstimülasyon (KOH) sonucu elde edilen foliküllerden elde edilen oosit sayıları yüzdelik dilimlere ayrılarak gruplandırıldı. Gruplarda oosit çıktısına etkili değişkenler analiz edilerek, en çok sayıda oosit eldesini öngörecek değişkenleri tespit etmeye çalıştık.

**Bulgular:** Hastaları oosit/≥14 mm folikül oranlarına göre incelediğimizde, %10,2'sinin ≤%30, %4,1'inin %31-40, %6,6'sının %41-50, %2,9'unun %51-60, %5,6'sının %61-70, %8'inin %71-80, %4,9'unun %81-90 ve %57,8'inin %91-100 grubunda olduğu tespit edilmiştir. Çalışmamıza göre, oosit/≥14mm folikül oranına etkili değişkenler kadının yaşı, bazal FSH (eşik değer 9.5 mIU/mL için duyarlılık %70.6 ve özgüllük %62.1, P=0.001) ,bazal LH (eşik değer 5.5 mIU/mL için duyarlılık %64 ve özgüllük %69, P<0.001), AMH (eşik değer 0.280 ng/mL için duyarlılık %74.8 ve özgüllük %68.4, P<0.001), hCG günü E2 (eşik değer 388.50 pg/mL için duyarlılık %77.4 ve özgüllük %77.4, P<0.001) ve hCG günü ≥17 mm folikül sayısıdır. AMH için AUC 0.734 ve hCG günü E2 için AUC 0.768, bazal LH için AUC 0.721 ile iyi derecede; bazal E2 için AUC 0.623, bazal FSH için AUC 0.685 ile düşük derecede belirleyicidir.

**Sonuç:** Oosit çıktısına etkili değişkenler bazal FSH-LH, AMH, hCG günü E2, ve hCG günü ≥17mm folikül sayısıdır. AMH, bazal LH ve hCG günü E2 oosit çıktısı için daha iyi belirteçlerdir.

**Anahtar Kelimeler:** Oosit çıktısı, Yardımcı Üreme Teknikleri, oosit toplama

## ABSTRACT

**Aim:** We designed our study with the purpose of determining which variables should be used to predict oocyte output, to increase the number of collected oocytes and the success rate of IVF.

**Materials and Method:** A total of 412 infertile patients admitted to the IVF center of a tertiary university hospital between the years 2016 and 2018 were evaluated retrospectively. The number of oocytes obtained by OPU from the follicles as the result of COH was grouped in percentiles. The variables effective on oocyte output were analyzed in the groups, and tests to predict acquisition of the maximal number of oocytes were tried to be determined.

**Results:** When we investigated according to the oocyte/≥ 14 mm follicle ratios, we determined that 10.2% of the patients were in the ≤ 30% group, whereas 4.1% of them were in the group of 31-40 %. 6.6% of the patients were in the group of 41-50 %, 2.9% in the group of 51-60%, 5.6% in the group of 61-70 %, 8% in the group of 71-80%, 4.9% in the group of 81-90 %, and 57.8% in the group of 91-100 %. According to the results of our study, the variables effective on the oocyte/≥14 mm follicle ratio were the patient's age, basal FSH value (cutoff 9.5mIU/mL, sensitivity 0.706, specificity 0.621, p=0.001), basal LH (cutoff 5.5mIU/mL, sensitivity 0.640 specificity 0.690, p<0.001), AMH (cutoff 0.280ng/mL, sensitivity 0.748 specificity 0.684, p<0.001), trigger day E2 (cutoff 388.50pg/mL, sensitivity 0.774, specificity 0.774, p<0.001) and the number of ≥17 mm follicles on the trigger day. The area under the curve (AUC) was 0.734for AMH, 0.768 for trigger day E2, and 0.721 for basal LH, having a good predictivity. The AUC was 0.623 for basal E2, and 0.685 for basal FSH value, having poor predictivity.

**Conclusion:** The variables effective on oocyte output were the basal (day 2-4) FSH, LH, and AMH values, together with trigger day E2 value and the number of ≥17 mm follicles on the trigger day. AMH, basal LH, and trigger day E2 have good predictivity regarding oocyte output.

**Keywords:** Oocyte output, Assisted Reproductive Technologies, Oocyte pick-up

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## INTRODUCTION

One of the most important targets is oocyte retrieval in assisted reproductive technologies (ART). It is well known that the number of collected oocytes is strongly associated with live birth rate (LBR) (1). The primary outcome of in vitro fertilization (IVF) treatment deals with LBR and can be increased by good-quality embryo transfer. The greater number of retrieved oocytes, the greater number of embryos are formed. Therefore, the success rate of fresh and frozen-thawed embryo transfers is elevated by the number of oocytes retrieved during controlled ovarian hyperstimulation (COH). The retrieval of oocytes by the oocyte pick-up procedure (OPU) from ovarian follicles obtained via COH is a significant step of the IVF practice. Unfortunately, oocytes cannot be retrieved from all follicles grown within the hyper-stimulated ovary. Even though the incidence of the genuine empty follicle syndrome has been reported to reach up to 7% in the medical literature (2), in an IVF cohort study with 12000 cases, the prevalence of actual cases was reported to be as low as 0.16% (3). According to this study, the number of oocytes retrieved from the follicles grown to a certain size by the administration of exogenous gonadotropins should be approximately equal to the number of follicles or oocyte retrieval should be accomplished in more than 90% of follicles. In IVF treatment, it is not always possible to retrieve a maximum number of oocytes from the follicles. Theoretically, numerous reasons that influence the oocyte output would be present. The size of the follicle (4), the type and dosing of the gonadotropins used for hyperstimulation (5), the type and dosing of the drug used as trigger (6), the OPU setup and experience of the performer (7), whether follicular flushing is performed or not (8,9), and the patient age (10) are among the variables effective in oocyte output rate. Nevertheless, the etiology of variability in oocyte retrieval is still unidentified, and it continues to be a source of anxiety for both the people dealing with IVF treatment and the patient. In this study, we aimed to determine the variables predictive for retrieving a maximal number of oocytes by analyzing various tests and applications routinely used in our clinic.

## MATERIALS AND METHODS

For this oocyte output study, the infertile patients who had been admitted to the IVF Center of Ankara University Medical Faculty and had received ART treatment (n=412) between 2016 and 2018 were retrospectively evaluated following approval of the Ethics Committee. (App. date/no: 17-1122-18/October 22, 2018). All ethics protocols followed as per Declaration of Helsinki.

The number of follicles observed by ultrasound on the trigger day was recorded. OPU was performed an average of 36 hours following the administration of trigger for final oocyte maturation. In our clinic, we have routinely been using the flushing method for follicle aspiration. The aspiration fluid was evaluated by a group of experienced embryologists. The oocytes retrieved from the follicles were divided into percentage groups as < % 30, %31-40, %41-50, %51-60, %61-70, %71-80, %81-90, and %91-100. The patient's age, the marital duration, the total induction period, the basal (D2-4) E2-LH-FSH-TSH-PRL values, the history of previous ART treatment, the type of infertility, the type of gonadotropin used, and the type of trigger were recorded. The presence of statistically significant differences among the groups regarding the investigated parameters was sought for.

The ideal trigger time in our clinic is the presence of a follicle cohort consisting of two or more follicles sized ≥ 17 mm, and most of the follicles sized ≥ 14 mm. On the trigger day, two separate groups were formed according to the numbers of follicles sized ≥ 14 mm and ≥ 17 mm. The correlations of the factors effective on the oocyte/≥14 mm follicle ratio obtained via OPU were investigated.

Statistical Analysis

Data analyses were performed using SPSS Version 21.0 (IBM Corporation, Armonk, NYC, USA). Samples were tested with Shapiro-Wilk to determine the normality of distributions. According to the results, non-parametric tests were preferred. Continuous variables were compared with Kruskal-Wallis and Mann-Whitney U test. Categorical variables were compared with Chi-square test. The correlations among the variables were investigated by the Spearman rank correlation tests. A P value of <0.05 was considered statistically significant.

## RESULTS

Since the quantitative variables were not in conformity with normal distribution, the descriptive statistical results of our cohort were presented as median (min-max) and the results regarding the qualitative variables were presented as frequency and percentage in Table 1.

**Table 1.** Descriptive statistical results

| Quantative variables | Median | Min-Max |
|---|---|---|
| Age (years) | 34.0 | 19.0-48.0 |
| Marital duration (years) | 5.0 | 0.2-27.0 |
| Duration of infertility | 5.0 | 0.2-21.0 |
| OPU count | 4.0 | 0.0-19.0 |
| Trigger day ≥14mm follicle | 4.0 | 0.0-17.0 |
| Oocyte/≥14mm follicle ratio | 100.0 | 0.0-100.0 |
| Total dose of gonadotropin | 2400.0 | 300.0-7025.0 |
| Total duration of cycles | 12.0 | 5.0-31.0 |
| Total induction period | 10.0 | 0.0-30.0 |
| BasalE2 (pg/mL) | 47.0 | 3.0-594.0 |
| Basal FSH (mIU/mL) | 8.0 | 0.6-54.0 |
| Basal LH (mIU/mL) | 4.0 | 0.2-34.0 |
| Basal TSH (mIU/mL) | 2.0 | 0.1-51.0 |
| Basal PRL (ng/ml) | 13.0 | 0.0-50.0 |
| Trigger day E2 | 878.0 | 12.0-10958.0 |
| Trigger day ≥17mm follicle count | 2.0 | 0.0-17.0 |
| AMH (ng/ml) | 0.6 | 0.0-7.0 |
| **Qualitative variables** | **Frequency** | **Percentage** |
| The presence of oocytes at OPU | 375 | 91 |
| Previous IVF İCSİ | 203 | 49.9 |
| Previous IUI | 145 | 35.5 |
| The type of infertility | | |
| Male | 129 | 31.3 |
| Female | 166 | 40.3 |
| Unexplained | 94 | 22.8 |
| Female + Male | 14 | 3.4 |
| IVF for HLA compatible sibling | 9 | 2.2 |
| Gonadotropin | | |
| Only r-FSH | 82 | 19.9 |
| Hp-hMG | 118 | 28.6 |
| Regular-hMG | 24 | 5.8 |
| Hp-hMG and r-FSH | 138 | 33.5 |
| Regular-hMG and r-FSH | 50 | 12.1 |
| Oocyte/≥14 mm follicle ratio (%) | | |
| ≤30 | 42 | 10.2 |
| 31-40 | 17 | 4.1 |
| 41-50 | 27 | 6.6 |
| 51-60 | 12 | 2.9 |
| 61-70 | 23 | 5.6 |
| 71-80 | 33 | 8.0 |
| 81-90 | 20 | 4.9 |
| 91-100 | 238 | 57.8 |
| Trigger | | |
| HCG | 271 | 66.9 |
| aGnRH | 77 | 19.0 |
| HCG + aGnRH | 57 | 14.1 |

The relationships between the oocyte/≥14 mm follicle ratio and the quantitative variables were shown in Table 2. A weak, statistically significant negative correlation was found to be present between the ratio and the total cycle duration (r=-0.121). There was a statistically weakly significant correlation between the ratio and the total induction period (r=-0.138). Weak-level statistically significant correlations were determined between the ratio, FSH and LH (r=-0.122 and r=-0.137, respectively). A weak positive correlation was present between the ratio and the HCG day E2 level (r=0.185). No significant relationships were found to be present between the ratio and the remaining parameters. The correlations determined between the oocyte/≥14mm follicle ratio and the qualitative variables were also shown in Table 2.

**Table 2.** The distributions of the relationships between the oocyte/≥14 mm follicle ratio and the variables

| Quantitative variables | r |
|---|---|
| Age | -0.056 |
| Marital duration | 0.059 |
| Total gonadotropin dose | -0.070 |
| Total cycle duration | **-0.121** |
| Total induction period | **-0.138** |
| Basal E2 | -0.091 |
| Basal FSH | **-0.122** |
| Basal LH | **-0.137** |
| Basal TSH | -0.094 |
| Basal PRL | -0.055 |
| AMH | 0.148 |
| Trigger day E2 | **0.185** |
| Trigger day ≥17 mm follicle count | 0.045 |
| **Qualitative variables** | **Median (min-max)** |
| Previous IVF-ICSI | |
| No (n=204) | 100 (0-100) |
| Yes(n=203) | 100 (0-100) |
| Previous IUI | |
| No (n=260) | 100 (0-100) |
| Yes(n=145) | 100 (0-100) |
| The type of infertility | |

| | | |
|---|---|---|
| Male (n=129) | 100 (0-100) ab | **0.002** |
| Female (n=167) | 85.7 (0-100) a | |
| Unexplained (n=94) | 100 (0-100) b | |
| Female + Male (n=14) | 91,7 (0-100) ab | |
| IVF for HLA compatible sibling (n=9) | 100 (50-100) ab | |
| Gonadotropin | | |
| rFSH (n=82) | 89.2 (0-100) | 0.716 |
| hp-hMG (n=129) | 100 (0-100) | |
| regular-hMG (n=24) | 100 (0-100) | |
| hp-hMG and r FSH (n=144) | 100 (0-100) | |
| regular-hMG and r FSH (n=34) | 100 (0-100) | |
| Trigger | | |
| HCG | 100 (0-100) | 0.318 |
| aGnRH | 100 (0-100) | |

a-b: No significant difference was present between the groups having similar letters

r: Spearman rank correlation coefficient, Basal: day 2-4, FSH: Follicle stimulating hormone, LH: Luteinizing hormone, TSH: Thyroid stimulating hormone, PRL: Prolactin, AMH: Anti-Mullerian hormone, rFSH: Recombinant FSH, hp-hMG: High-purified human menopausal gonadotropin, aGnRH: Gonadotropin-releasing hormone agonist, HCG: Human chorionic gonadotropin, HLA: Human leucocyte antigen (the group in which IVF was performed for bone marrow transplantation from an HLA compatible sibling)

The oocyte/≥14 mm follicle percentages were divided into eight groups in total. The degree of influence of the factors effective on oocyte output in these groups constituted according to the oocyte/≥14 mm follicle percentages were shown in Table 3. The age and the basal FSH level showed differences according to ratios (p<0.001). The marital duration, the total gonadotropin dose, the total cycle duration, and the basal E2 level did not reveal any differences according to the ratios (p values 0.211,0.309, 0.050, 0.335, respectively). Regarding the basal FSH value, the median value of the <30 group was significantly higher compared to the medians of the 51-60, 61-70, 71-80, 81-90, 91-100 groups. The basal LH value was found to differ according to the ratios (p=0.006). The median value of the <30 group was determined to be higher than the 91-100 group. The difference originated from these two groups. The basal TSH and PRL values were not found to differ according to the ratios (p values 0.528 and 0.775, respectively). The AMH level was determined to be different according to the ratios (p=0.001). The median AMH value of the 81-90 group was found to be higher compared to the 91-100 and 41-50 groups. The difference originated from these two groups. The HCG day E2 value showed differences according to the ratios (p<0.001). The median E2 level of the <30 group was determined to be lower compared to the 71-80 and 91-100 groups. The HCG day ≥17mm follicle count was found to differ according to the ratios (p<0.001); the median value of the ≥17mm follicle count was determined to be lower in the <30 group when compared to the 51-60, 61-70, 71-80, 81-90, 91-100 groups. The median value of the ≥17mm follicle count was significantly higher in the 81-90 group than the 31-40 and 91-100 groups.

**Table 3.** The comparison of the qualitative variables effective on the oocyte/≥14 mm follicle percentages

| | <30 | 31-40 | 41-50 | 51-60 | 61-70 | 71-80 | 81-90 | 91-100 | p |
|---|---|---|---|---|---|---|---|---|---|
| Age | 35.21 ± 6.36b | 34.88 ± 6.03b | 35.11 ± 5.92b | 28.42 ± 5.99a | 34.52 ± 6.44b | 31.91 ± 6.14ab | 28.65 ± 4.61a | 33.32 ± 5.56b | **<0.001** |
| Marital duration | 7 (0.17 - 22) | 3.5 (0.92 - 15) | 4 (1 - 26) | 4.75 (1.5 - 8) | 3.5 (0.5 - 22) | 4 (1 - 21) | 4 (1 - 9) | 5 (0.5 - 37) | 0.211 |
| Total gonadotropin dose | 2287.5 (300 - 7025) | 3000 (1025 - 4825) | 2700 (900 - 3975) | 2700 (1050 - 3975) | 2250 (675 - 4350) | 2700 (1050 - 4875) | 2562.5 (1200 - 3900) | 2313 (300 - 6225) | 0.309 |
| Total cycle duration | 12 (6 - 31) | 13 (6 - 26) | 11 (7 - 17) | 12 (8 - 19) | 12 (9 - 19) | 13 (9 - 29) | 12.5 (10 - 28) | 12 (5 - 31) | 0.050 |
| Total induction period | 11 (4 - 29) | 11 (4 - 24) | 9 (6 - 14) | 10 (7 - 16) | 10 (7 - 17) | 11 (7 - 26) | 11 (8 - 26) | 10 (0 - 30) | 0.050 |
| BasalE2 | 54 (12 - 234) | 50 (14 - 75) | 46 (13 - 130) | 44 (18 - 70) | 53 (23 - 428) | 52 (20 - 146) | 47 (20 - 141) | 45 (3 - 594) | 0.335 |
| BasalFSH | 12 (3 - 40)a | 8 (3 - 34) ab | 11 (6 - 30) ac | 6 (3 - 13)b | 7 (0.6 - 19bc | 7 (2 - 17)b | 7 (2 - 11)b | 7.5 (1 - 54)bc | **<0.001** |
| BasalLH | 6 (2 - 34)a | 4 (0.9 - 15)ab | 4.5 (1 - 24)ab | 4 (1 - 10)ab | 4 (0.2 - 13)ab | 4 (2 - 20) ab | 5 (2 - 12)ab | 4 (0.9 - 28)b | **0.006** |
| BasalTSH | 2 (0.4 - 18) | 2 (1 - 18) | 2 (0.2 - 18) | 2 (0.6 - 5) | 1.52 (0.5 - 3) | 1 (0.3 - 5) | 2 (0.9 - 4) | 2 (0.1 - 51) | 0.528 |
| BasalPRL | 14 (5 - 50) | 10 (6 - 28) | 14 (6 - 24) | 12 (3 - 31) | 14 (7 - 50) | 11.5 (6 - 41) | 14 (4 - 46) | 13 (0 - 50) | 0.775 |
| AMH | 0.21 (0.02 - 5)b | 1.8 (0.6 - 2)ab | 0.26 (0.06 - 3.01)b | 1.15 (1 - 1.3)ab | 0.6 (0.1 - 1)ab | 0.9 (0 - 2.2)ab | 2.14 (0.9 - 7)a | 0.67 (0 - 5)b | **0.001** |
| Trigger day E2 | 208 (12 - 4895)a | 869 (73 - 1579)ab | 502 (72 - 4800)ab | 1365 (486 - 2046)ab | 944 (122 - 4856)ab | 1231 (130 - 7746)b | 1152 (202 - 6685)ab | 1045 (50 - 10958)b | **<0.001** |
| Trigger day ≥17mm fol. | 1 (0 - 6)a | 2 (1 - 4)ba | 2 (1 - 5)ba | 4 (2 - 9)bc | 3 (1 - 5)bc | 3 (1 - 12) bc | 4 (1 - 11)c | 2 (0 - 17)b | **<0.001** |

a-c: No difference was present among the groups with similar letters

A logistic regression analysis was performed for determining the variables effective on the oocyte/≥14mm follicle ratio. The variables with statistical significance were shown in Table 4. According to the results of logistic regression analysis, prolongation of the total cycle period reduces the probability of oocyte retrieval (p=0.010). On the other hand, prolongation of the total induction period leads to 5.29-fold increase in the probability of oocyte retrieval. The increase in the basal LH value reduces the retrieval probability of oocytes (p=0.010). The trigger day E2 value has a statistically significant effect on the ratio, and as E2 value increases, the number of retrieved oocytes becomes different from zero.

**Table 4.** The results of the logistic regression analysis

| | OR (%95 CI) | p |
|---|---|---|
| Total cycle duration | 0.165 (0.041 – 0.656) | **0.010** |
| Total induction period | 5.29 (1.392 – 20.1) | **0.014** |
| Basal LH | 0.778 (0.643 – 0.941) | **0.010** |
| Trigger day E2 | 1.009 (1.003-1.015) | **0.030** |

The predictive values of several variables effective on the oocyte/≥14mm follicle ratio were evaluated using the ROC analysis method, and the cut-off values, together with the sensitivity and specificity rates of statistically significant variables for basal hormones were shown in Table 5.

**Table 5.** The results of the ROC analysis for the basal E2, FSH, LH, TSH, and PRL values

| | AUC (%95 CI) | p | Cut-off value | Sensitivity | Specificity |
|---|---|---|---|---|---|
| E2 | 0.623 (0.509-0.737) | **0.031** | 53.50 | 0.610 | 0.586 |
| FSH | 0.685 (0.572-0.797) | **0.001** | 9.5 | 0.706 | 0.621 |
| LH | 0.721 (0.632-0.810) | **<0.001** | 5.5 | 0.640 | 0.690 |
| TSH | 0.608 (0.486-0.730) | 0.059 | --- | --- | --- |
| Prolactin | 0.610 (0.499-0.721) | 0.054 | --- | --- | --- |

ROC curves were shown in Figure 1, and the ROC analysis for trigger day E2 and AMH were shown in Figures 2 and 3.

**Figure 1.** ROC analysis for the basal E2, FSH, LH, TSH, and PRL values

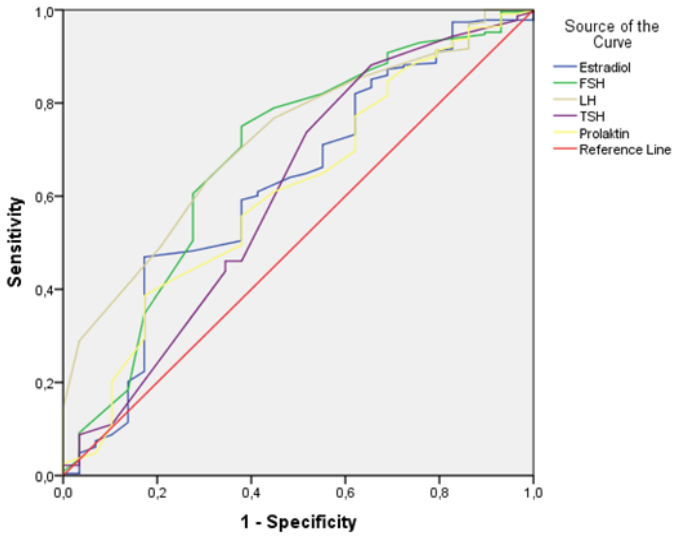



**Figure 2.** ROC analysis for the trigger day E2 value

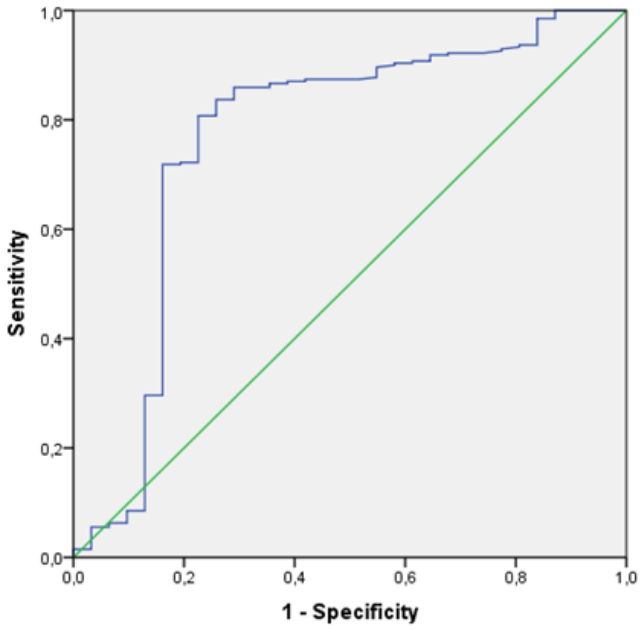
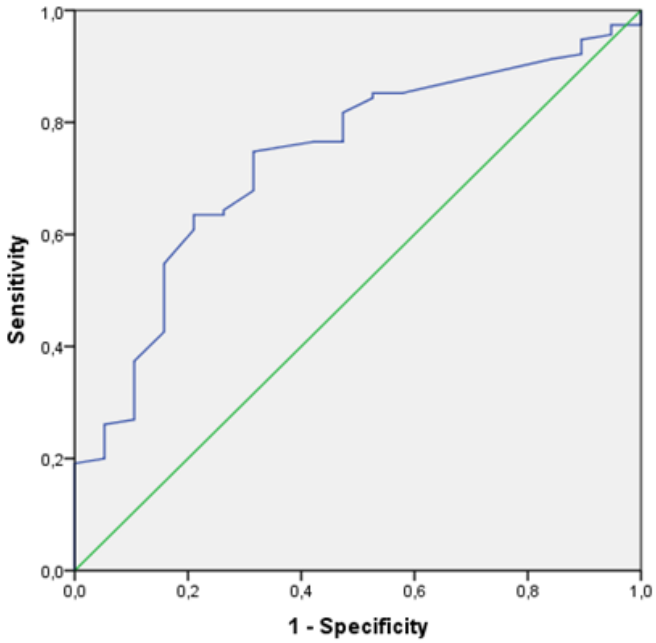


**Figure 3.** ROC analysis for the AMH value




The area under the curve (AUC) was calculated to be 0.768 (0.659-0.877) for the trigger day E2 value, and this value was statistically significant (p<0.001). When the cut-off value for HCG day E2 level is considered as 388.50, the sensitivity is calculated as 0.774 and the specificity value as 0.774.AUC was calculated as 0.734 (0.621-0.847) for the AMH value, and this result was statistically significant (p=0.001). When the cut-off value for AMH level is considered as 0.280, the sensitivity is calculated as 0.748 and the specificity value as 0.684. Inability to retrieve any oocyte from sufficiently sized five or more follicles was considered as the criterion for genuine EFS. Two cases (0.5%) complying this criterion were determined to be present in our study cohort.

## DISCUSSION

The present study was conducted to investigate the variables which are predictive for maximum oocyte output in patients who undergo in vitro fertilization treatment. According to the results obtained from our study, the most important variables to predict oocyte yield are serum AMH level, basal LH level, and trigger day E2 level. In addition, serum basal FSH level and the number of ≥17 mm follicles on hCG day are also associated with oocyte yield.

Substantial evidence is present regarding that each retrieved oocyte increases the chance of pregnancy, and that an optimal number of oocytes should be present for a successful outcome in ART (1,11,12) .

The primary outcome of ART is the live birth rate (LBR). LBR increases in direct proportion until 15 oocytes are obtained, it makes a plateau between 15 and 20 and then decreases after 20 (1). Inability to retrieve any oocyte post-OPU from the appropriately sized follicles formed by COH in ART is a quite troublesome issue for the clinician. Various causes have been suggested as the result of studies conducted for investigating the reasons for the inability to retrieve a maximal number of oocytes. The type and dose of the trigger administered for the final maturation, the period between the trigger and OPU, the OPU technique, the experience of the clinician performing OPU, whether flushing was performed or not, EFS, and the follicle size at the time of trigger are some of these causes.

The follicle size is considered as essential for oocyte output. Too small or too large follicles are not appropriate for oocyte retrieval (13). In a study, it was reported that the maximal number of oocytes had been retrieved from the follicles sized between 12 mm and 19 mm on the trigger day (4). In our clinic, the numbers of ≥14 mm and ≥17 mm follicles are recorded on the trigger day. With the increasing number of follicles sized ≥17 mm on the trigger day, the oocyte retrieval rate also increased in our study (p<0.001). The median value for oocyte retrieval was found to be lower in the <30% group when compared to the other percentage groups. Interestingly, the median value for oocyte retrieval was higher in the 81-90% group than the 91-100% group.

Rosen et al. reported that oocyte output was higher in 13-15 mm follicles compared to follicles sized >18 mm (14). Our series involved the number of oocytes retrieved from follicles sized ≥14mm. As practiced in most of the IVF clinics, in our center, when 2-3 leading follicles sized ≥17mm were observed, various triggers were used for final maturation. More oocytes were retrieved in patients who had ≥17mm follicle on the trigger day. The group in which oocytes were retrieved in more than 50% of the follicles consisted of cases with increased number of ≥17mm follicles. Similarly, Dubey et al. reported that 85% of the oocytes were retrieved from 14-24 mm follicles (15).

One of the significant applications affecting oocyte output from the follicle is the trigger. Currently, various drugs are administered for final oocyte maturation. HCG and aGnRH are administered singly or in combination, with Kisspeptin-54 (16) and rLH (6) being used less frequently. While detachment of the immature oocyte from the follicular wall is difficult, the mature oocyte can be easily separated. Thus, more oocytes can be retrieved during OPU. Data obtained from the studies on which trigger is better are contradictory (17,18).

The triggers used in our cases were grouped as HCG, aGnRH, and HCG+aGnRH, and which group was superior to the others regarding oocyte output was investigated. No significant differences were found to be present among the groups regarding the effect on the number of the oocytes retrieved from the ≥14 mm follicles (p=0.318) (Table 2). However, more follicles were observed in the HCG + aGnRH group, and the difference was significant (p<0.01). Inability to retrieve more oocytes despite the presence of more follicles in this group indicates that other factors were effective.

According to our results, oocyte retrieval was not possible in 37 of 412 cases (9%). The oocyte/≥14 mm follicle ratio was below 40% in more than 40% of the patients. Why oocyte is not retrievable from almost all appropriately sized follicles despite the quite low incidence of the genuine empty follicle syndrome in the literature is the starting point of this study. In our series, we considered null oocyte retrieval from five appropriately sized follicles as the criterion for calculating the rate pf the genuine EFS. Based on this criterion, the genuine EFS was 2/412 (0.05%) in our patients, and it was consistent with the literature.

According to our study, the variables effective on the oocyte/≥14 mm follicle ratio were the female patient's age, the basal FSH, the basal LH, the AMH, the trigger day E2 levels together with the number of follicles sized ≥17 mm on the trigger day. The oocyte count decreased with increasing age. In the group with the lowest oocyte number (the <30% group), the basal FSH and LH levels were found to be the highest among groups. The group with the lowest AMH value had also the lowest oocyte output. The group with the lowest trigger day E2 level was the group in which the least number of oocytes were retrieved.

Limitations

In this study, some of the variables that might have affected the oocyte output from the follicle were investigated, and other causes such as the OPU experience of the clinician, the embryologist's experience, and the OPU technique were not investigated. The unexpected results observed in the oocyte output groups might have originated due to such a reason. Therefore, our oocyte prediction was limited with the investigated parameters.

## CONCLUSION

Our study demonstrated that the AUC of 0.734for AMH, the AUC of 0.768 for trigger day E2, and the AUC of 0.721 for the basal LH were strongly predictive, whereas the basal E2, FSH, and PRL were poorly predictive. The predictivities of the routinely used tests to maximize the oocyte output from appropriately sized follicles formed by COH in IVF treatment are not optimal. Further studies should be conducted for determining tests with more powerful predictivity.

DISCLOSURE

Authors have declared no conflict of interest.

AUTHORS' CONTRIBUTIONS

Levent Dikbaş, MD: Study design, writing, redaction, statistical analysis, corresponding

Güler Mammadli, MD: Data collection, Ethics Committee procedures

Yavuz Emre Şükür, MD: Redaction

Batuhan Özmen, MD: Management of project

Others: Data producers

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