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Original Article

The Association Between Mature Oocyte Proportion and IVF Success: A Retrospective Analysis of 2,565 ICSI Cycles

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Abstract

Background: IVF/ICSI success depends on the proportion of fertilizable metaphase II (MII) oocytes, yet direct evidence linking mature-oocyte rate to clinical outcomes remains limited. This study aimed to assess the impact of mature oocyte rate on clinical pregnancy outcomes in a large retrospective cohort of women undergoing ICSI.

Methods: A total of 2,565 women who underwent ICSI cycles at a single IVF center in Türkiye were retrospectively analyzed. Patients were stratified into three groups according to mature oocyte proportion: Group 1 (0-50%), Group 2 (51-75%), and Group 3 (76-100%). Data were analyzed per initiated ICSI cycle. Embryo transfer was performed on day 3 or day 5 post-fertilization, depending on embryo quality and patient characteristics. Logistic regression analysis was performed to assess the independent effect of mature oocyte rate on pregnancy outcomes. Statistical significance was set at $p<0.05$.

Results: Clinical pregnancy rates were significantly lower in Group 1 (10.23%) compared to Group 2 (28.96%) and Group 3 (29.99%) ($p<0.001$). Live birth and implantation rates increased in the higher maturity groups (7.5% *vs.* 21.5% *vs*. 22.9% and 12.1% *vs*. 24.3% *vs*. 26.0%, respectively), whereas miscarriage rates decreased (18.9% *vs*. 13.4% *vs*. 12.8%). Logistic regression analysis confirmed that a higher mature oocyte proportion was an independent predictor of clinical preg-nancy (Group 2: OR=3.4, 95%CI: 2.5-4.6; Group 3: OR=3.5, 95%CI: 2.6-4.7; $p<0.001$).

Conclusion: This large-scale retrospective cohort analysis demonstrates that mature oocyte proportion is an important prognostic factor of IVF success. Mature oocyte proportion should be considered an essential parameter in clinical practice and patient counseling.

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Introduction

Infertility is a growing public health concern with substantial medical, psychological, and social effects worldwide (1–4). In assisted reproductive technology (ART), particularly in vitro fertilization (IVF) and intracytoplasmic sperm injection (ICSI), clinical success ultimately depends on the generation of high-quality embryos with adequate implantation potential (5). A central upstream driver of embryo competence is oocyte maturation. Only oocytes that complete

meiotic maturation to the metaphase II (MII) stage are normally fertilizable, whereas germinal vesicle (GV) and metaphase I (MI) oocytes display inferior developmental potential (6–11).

Much of the literature has focused on the number of oocytes retrieved, reporting variable relationships with fertilization, embryo development, and live birth (12–20). However, larger oocyte cohorts do not invariably translate into higher pregnancy rates, underscoring that quantity alone is insufficient to predict success (12, 17–20). A parameter that better captures the quality dimension is the proportion of MII oocytes within a cycle. From a biological standpoint, incomplete oocyte maturation leads to cytoplasmic immaturity, including suboptimal organelle redistribution, mitochondrial function, and mRNA/protein stores, which compromises fertilization and embryo develop-ment, and this becomes particularly evident in individuals yielding a lower proportion of mature oocytes (23–27).

Prior studies have suggested that oocyte maturity is associated with improved IVF outcomes, yet important limitations temper the strength and generalizability of existing evidence. These include small sample sizes, single-center or highly selected cohorts, heterogeneous stimulation/ transfer strategies, and, in several reports, the absence of multivariable modeling (*e.g.*, logistic regression) to test whether the mature oocyte proportion exerts an independent effect on outcomes after accounting for key confounders (8, 9, 21, 22). Moreover, some findings remain inconsistent across studies, which further justifies evaluation in larger datasets using standardized analytic approaches.

Against this background, the present study does not seek to introduce a novel concept but rather to confirm and strengthen previous observations by analyzing a large single-center cohort of 2,565 ICSI cycles, with a primary focus on the mature oocyte proportion. The association of this proportion with clinical pregnancy, implantation, and live birth was examined, and logistic regression analysis was used to assess whether the mature oocyte rate served as an independent predictor of clinical outcomes. In interpreting these relationships, established prognostic factors including maternal age, anti-Müllerian hormone (AMH) levels, antral follicle count (AFC), number of oocytes retrieved, day of embryo transfer, and embryo quality were also taken into account as these variables are known to influence IVF success and are essential for a comprehensive appraisal of treatment prognosis (12–16, 25–27).

Methods

Study population and design: This retrospective cohort study included 2,565 women who underwent ICSI cycles at the Istanbul Kadıköy Florence Nightingale Hospital In Vitro Fertilization Center between January 2013 and December 2020. Eligible patients were women aged 20–42 years, with 1 to 32 oocytes retrieved per cycle. Only cases of female factor infertility were included in order to minimize potential confounding. Etiologies included tubal factor, unexplained infertility, and diminished ovarian reserve. Exclusion criteria were smoking, poor responders according to ESHRE consensus, polycystic ovary syndrome (Rotterdam criteria), endometriosis, BMI <19 or >26 *kg/m²*, and any cycles involving preimplantation genetic testing, coasting, assisted hatching, or empirical techniques. Male factor infertility cases were also excluded. Baseline characteristics such as number of previous IVF cycles were recorded.

The ethics committee approval (approval number: E-30785963-020-237953) was received from the Malatya Turgut Özal University Non-invasive Clinical Research Ethics Committee. All procedures were conducted in accordance with the principles of the Helsinki Declaration. The data used in the study were anonymized, keeping patient identities confidential.

Controlled ovarian stimulation protocols: Ovarian stimulation was performed using either a long GnRH agonist protocol and the GnRH antagonist protocol. In the long agonist protocol, a GnRH agonist was started on day 21 of the menstrual cycle for pituitary downregulation. After suppression (E2 <50 *pg/ml*), recombinant FSH (recFSH; 125–600 *IU/day*) was initiated. In the antagonist protocol, recFSH stimulation began on day 3, with a GnRH antagonist (Cetrorelix or Ganirelix, 0.25 *mg/day*) added from stimulation day 5 onward.

When three or more follicles reached a diameter of at least 18 *mm*, ovulation was triggered with 5000 *IU* hCG (Ovitrelle; Merck Serono, Germany). Oocyte retrieval was performed 35.5 *hr* later.

Details of the stimulation protocols, including the allocation of long agonist versus antagonist cycles, the starting and total gonadotropin doses, and any dose adjustments, were recorded. Protocols were individualized based on age, ovarian

reserve (AMH, AFC), and clinical profile, with drug doses modified in response to ovarian stimulation. All data related to oocyte quality assessment and stimulation were prospectively recorded in the IVF center database.

Oocyte assessment and grouping: Following retrieval, oocytes were enzymatically denuded of their cumulus cells. Maturity assessment was performed immediately after cumulus removal, based on the presence of the first polar body (MII stage). Oocytes were classified as GV, MI, or MII following standardized morphological criteria routinely applied at our IVF center.

For each patient, the mature oocyte rate was calculated as: (MII oocyte count/total oocyte count) ×100.

Patients were stratified into three pre-specified groups based on literature and clinical relevance: Group 1 included those with 0–50% mature oocytes, Group 2 included 51–75% mature oocytes, and Group 3 included 76–100% mature oocytes.

Laboratory procedures and embryo transfer: Intracytoplasmic sperm injection was performed on MII oocytes using micromanipulation systems (Optimas, Turkey). Embryos were cultured in LifeGlobal medium and assessed daily. Only fresh embryo transfers were included. Transfers were performed on either day 3 or day 5, depending on embryo quality and patient profile. The number of embryos transferred per cycle was recorded.

Pregnancy assessment: Pregnancy was confirmed by serum β-hCG ≥50 *IU/L*, measured 16 days after embryo transfer, followed by ultrasound visualization of a gestational sac. Live birth was defined as delivery of a viable infant after 24 weeks of gestation. Pregnancy and live birth outcomes were assessed by independent clinicians blinded to patient grouping to ensure impartiality.

Statistical analysis: Analyses were performed with SPSS version 22.0 (IBM, USA). Continuous variables were expressed as mean±SD and categorical variables as percentages. Comparisons among groups were performed using chi-square test for categorical variables and ANOVA or Kruskal–Wallis test for continuous variables. Post hoc analyses included Tukey's test for parametric and Dunn's test for non-parametric data.

Logistic regression analysis was used to assess whether mature oocyte proportion was an independent predictor of clinical pregnancy, adjusting for potential confounders such as maternal age, infertility etiology, and baseline characteristics. A $p<0.05$ was considered statistically significant.

Results

The study included a total of 2,565 patients, divided into three groups according to the proportion of mature oocytes: Group 1 (0–50%, n=518), Group 2 (51–75%, n=770), and Group 3 (76–100%, n=1,277).

Patient characteristics: The baseline demographic characteristics, including mean age, BMI, and duration of infertility were comparable among the three groups ($p>0.05$) (Table 1). The distribution of infertility etiology (tubal factor, unexplained infertility, diminished ovarian reserve), the number of previous IVF attempts, as well as baseline hormonal markers (AMH and FSH) showed no significant intergroup differences.

Oocyte and fertilization outcomes: The mean number of retrieved oocytes differed significantly across groups, with higher values observed in Group 3 compared to Group 1 (Table 2). Fertilization rates also increased significantly with a higher mature oocyte proportion (Group 1: 52.1%, Group 2: 68.3%, and Group 3: 70.5%; $p<0.001$). The mean number of embryos formed and the proportion of good-quality embryos were significantly higher in Groups 2 and 3 compared with Group 1 ($p<0.001$).

In addition, the number of embryos formed on day 3 and day 5 of culture and the percentage of

Table 1. Baseline characteristics of the study population

Characteristics	Group 1: 0–50% (n=518)	Group 2: 51–75% (n=770)	Group 3: 76–100% (n=1277)	p-value
Age (years)	34.2±4.8	33.9±5.1	34.1±4.7	0.621
Infertility duration (years)	6.1±3.2	5.9±2.9	6.0±3.1	0.738
BMI (kg/m^2)	25.1±3.9	24.8±4.2	25.0±4.0	0.574
AMH (*ng/ml*)	2.1±0.8	2.2±0.9	2.2±0.9	0.482
FSH (*IU/L*)	7.4±2.1	7.3±2.3	7.2±2.0	0.655
Previous IVF cycles (n)	1.3±0.6	1.4±0.7	1.4±0.6	0.413

good-quality embryos were recorded and found to be significantly greater in higher maturity groups (Table 2).

Clinical pregnancy outcomes: Clinical pregnancy rates were 10.2% (53 patients) in Group 1, 28.9% (223 patients) in Group 2, and 29.9% (383 patients) in Group 3. The difference among groups was statistically significant (p<0.001). No significant difference was observed between Groups 2 and 3.

The number of embryos transferred per cycle was significantly higher in Group 1 compared to the other groups, yet pregnancy outcomes were poorer. Only fresh embryo transfers were included, performed on either day 3 or day 5, depending on embryo quality and patient characteristics. The impact of transfer stage was analyzed separately, and higher pregnancy rates were observed in day 5 transfers, although the difference did not reach statistical significance.

Additional clinical outcomes: The live birth rate increased progressively with the mature oocyte proportion (7.5% in Group 1, 21.5% in Group 2, and 22.9% in Group 3; p<0.001). The number of twin births was also reported and was higher in Groups 2 and 3.

The implantation rate increased significantly across groups (12.1%, 24.3%, and 26.0%, respectively; p<0.01). Miscarriage rates showed a declining trend (18.9%, 13.4%, and 12.8%), though the differences were not statistically significant. The lack of significance may be related to limited subgroup sizes and multifac-torial causes of miscarriage beyond oocyte maturity (Table 3). Additionally, chemical preg-nancies (gestational sac without further progres-sion) were documented in all groups and are presented in table 3.

Logistic regression analysis: Multivariable logistic regression confirmed that a higher mature oocyte proportion was an independent predictor of clinical pregnancy after adjusting for potential confounders including maternal age, AMH, number of oocytes retrieved, stimulation protocol, and embryo quality. Compared with Group 1 (reference), the adjusted odds ratios (OR) for clinical pregnancy were 3.2 (95%CI: 2.3–4.4, p<0.001) in Group 2 and 3.3 (95%CI: 2.4–4.6, p<0.001) in Group 3.In addition, adjusted logistic regression for live birth demonstrated similar findings, with higher odds of live birth in Groups 2 and 3 compared to Group 1 (Figure 1).

Stimulation protocols and treatment details: The distribution of stimulation protocols (long agonist versus antagonist) and total gonadotropin doses are summarized in supplementary tables. Protocol selection was individualized according to age and ovarian reserve. No significant correlation was found between protocol choice and mature oocyte proportion.

Table 2. Oocyte, embryo, and clinical pregnancy outcomes according to mature oocyte proportion groups

Parameters	Group 1: 0–50% (n=518)	Group 2: 51–75% (n=770)	Group 3: 76–100% (n=1277)	p-value
Total oocytes retrieved	8.2±3.7	9.1±3.9	9.4±4.2	0.041
Fertilization rate (%)	52.1	68.3	70.5	<0.001
Embryos formed–Day 3	3.6±1.5	5.1±2.0	5.5±2.3	<0.001
Blastocysts formed–Day 5	1.2±0.7	2.0±1.1	2.4±1.3	<0.001
Good-quality embryos (%)	34.5	51.2	55.4	<0.001
Clinical pregnancy (+), n (%)	53 (10.2)	223 (28.9)	383 (29.9)	<0.001

Table 3. Additional clinical outcomes according to mature oocyte proportion groups

Outcome	Group 1: 0–50% (n=518)	Group 2: 51–75% (n=770)	Group 3: 76–100% (n=1277)	p-value
Mean number of embryos transferred	2.1 ± 0.6	1.9 ± 0.5	1.8 ± 0.4	0.047
Chemical pregnancy (gestational sac, n)	14 (2.7)	28 (3.6)	41 (3.2)	0.327
Implantation rate (%)	12.1	24.3	26.0	0.008
Abortion rate (%)	18.9	13.4	12.8	0.092
Live birth rate (%)	7.5	21.5	22.9	<0.001
Twin births (n)	3	12	18	0.022

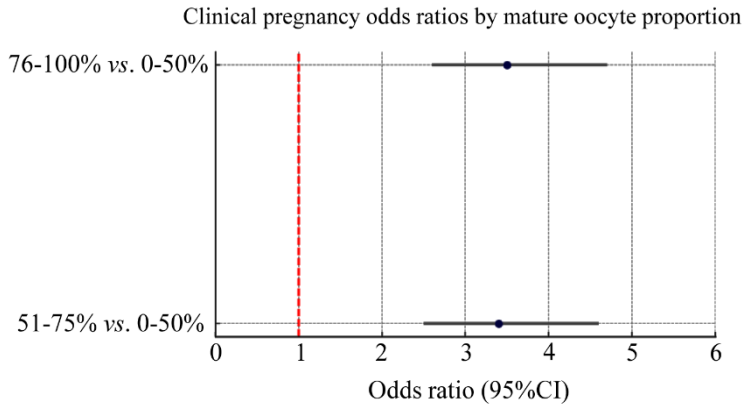

Figure 1. Forest plot illustrating the adjusted odds ratios (ORs) for clinical pregnancy according to mature oocyte proportion groups. Group 1, reference (OR=1.0); Group 2, OR=3.2 (95%CI: 2.3–4.4; p<0.001); and Group 3, OR=3.3 (95%CI: 2.4–4.6; p<0.001). The vertical dashed line indicates the null value (OR=1)

Discussion

In this large retrospective cohort study, the association between the proportion of mature oocytes and IVF/ICSI outcomes was investigated. Our results demonstrated that a higher proportion of MII oocytes was associated with significantly higher fertilization, clinical pregnancy, and live birth rates. These findings confirm and extend previous research, such as that of Sunkara et al. (12), who reported that the proportion of mature oocytes is a stronger prognostic factor for pregnancy outcomes than the total number of oocytes retrieved.

The significantly lower fertilization rate in patients with ≤50% mature oocytes reflects the limited developmental competence of GV and MI stage oocytes, consistent with studies showing that only MII oocytes have optimal potential for normal fertilization and embryo development (23, 24). Logistic regression analysis further confirmed that the proportion of mature oocytes is an independent prognostic factor for clinical pregnancy and live birth after adjusting for maternal age, ovarian reserve, embryo quality, and stimulation protocols. Although miscarriage rates were higher in the low maturity group, these differences were not statistically significant. This suggests that miscarriage is multifactorial and not solely dependent on oocyte nuclear maturity.

While our study focused on nuclear maturity, oocyte developmental potential is also critically dependent on cytoplasmic maturation, which regulates mitochondrial redistribution, RNA and protein storage, and spindle organization. Insufficient cytoplasmic competence may contribute to poor embryo quality and implantation failure, even when nuclear maturation appears normal (7, 25–27). This may explain why some patients with apparently high nuclear maturity still experience suboptimal clinical outcomes.

Oocyte maturation influences pregnancy outcomes via multiple mechanisms, including chromosomal segregation, mitochondrial energy supply, and early epigenetic programming. Our results highlight that the proportion of mature oocytes may serve as an adjunct parameter for patient counseling and treatment planning. Specifically, this marker could be useful in adjusting stimulation protocols, setting realistic expectations, and identifying patients who may require tailored interventions. However, it should not be interpreted in isolation, but rather in conjunction with age, AMH, AFC, total number of oocytes, embryo transfer day, and embryo quality.

The absence of a significant difference in miscarriage rates across maturity groups may reflect insufficient subgroup power, as well as the multifactorial etiology of early pregnancy loss. As previous studies have indicated, reduced oocyte quality may increase the risk of chromosomal abnormalities and aneuploidy (28–31). However, since our results did not reach statistical significance, these findings may not be generalizable.

Our findings are consistent with prior reports that a higher proportion of mature oocytes positively influences fertilization and clinical pregnancy rates (23, 24, 32). However, unlike earlier studies that were limited by small cohorts and lacked multivariable analyses, our work benefits from a larger dataset and regression modeling. Emerging data also suggest that stimulation protocols may influence maturation. For instance,

differences between GnRH agonist and antagonist regimens, as well as gonadotropin dose and duration, could modulate the percentage of MII oocytes and thus affect the outcomes (33, 34).

The strengths of our study include the large sample size, standardized embryological assessments, and adjustment for confounders using logistic regression. Nonetheless, limitations must be acknowledged. The retrospective design introduces potential bias, and the single-center setting limits the generalizability of findings. Moreover, certain populations, including smokers, patients with PCOS, and male factor infertility cases were excluded, which further narrows applicability to broader patient populations.

This study demonstrates that the proportion of mature oocytes is an important prognostic factor, though not the sole determinant, of clinical pregnancy and live birth following ICSI. While it strengthens existing evidence, its application should be interpreted in the context of other prognostic markers. Future prospective, multi-center studies are needed to validate these results and explore how cytoplasmic maturation, stimulation protocols, and molecular markers interact with nuclear maturity. Clinically, the proportion of mature oocytes may be a useful adjunct for patient counseling and individualized treatment strategies, provided that its limitations are recognized.

Conclusion

In this retrospective cohort study, the proportion of mature oocytes was found to be associated with fertilization, clinical pregnancy, and live birth outcomes following ICSI. While our findings support previous reports, the results should be interpreted with caution. The mature oocyte proportion may serve as a potential prognostic factor rather than a definitive determinant of pregnancy success. Clinically, it could be considered as an adjunct parameter to guide patient counseling and treatment strategies, though it should always be evalu-ated alongside established predictors such as maternal age, ovarian reserve, embryo quality, and transfer day. Future prospective multicenter studies are required to confirm these observations and further clarify the clinical utility of this para-meter.

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Conflict of Interest

The authors declare that there is no conflict of interest.

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