

Predictors of ovarian sensitivity to gonadotropins: a multicentric retrospective investigation of 1,942 women candidates to ART

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No competing interests reported.

Table 1 to 3 are available in the Supplementary Files section.

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2 **1,942 women candidates to ART**

3

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29 **Background:** Ovarian sensitivity to exogenous gonadotropin represents a matter of interest in the
30 reproductive field. Follicle to Oocytes Index and Follicle output rate were proposed to better assess
31 this phenomenon better reflecting the dynamic nature of ovarian sensitivity. So far, few are known
32 regarding the pattern of ovarian sensitivity and its relationship with anthropometric, chronological,
33 and biological age. The aim of the present paper is to assess relevant predictors of ovarian
34 sensitivity measured by FORT and FOI in women undergoing IVF/ICSI cycles

35 **Methods:** This retrospective study was carried out through 81,547 IVF/ICSI cycles conducted from
36 January 2023 to July 2024 at a tertiary IVF center. The following inclusion criteria were adopted: age
37 between 21- 40 years old; daily gonadotropin dosage between 225-300 IU; antral follicle count based
38 between 5 and 25; Body Mass Index (BMI) between 18.5 and 30 kg/m²; antagonist protocol.
39 Exclusion criteria were: oocyte donor cycle; dual trigger or GnRH agonist triggering. Ovarian
40 stimulation used recombinant and urinary gonadotropins with a flexible antagonist protocol; hCG
41 trigger was given when ≥ 3 follicles reached 16–17 mm.

42 **Results:** Chronological age is negatively correlated with both FOI ($\rho = -0.145$, p value < 0.001 , 95%
43 CI from -0.189 to -0.100) and FORT ($\rho = -0.084$, p value < 0.001 , 95% CI from -0.129 to -0.038).
44 Conversely, AMH positively correlated with FOI ($\rho = 0.141$, p value < 0.001 , 95% CI from 0.096,
45 0.186) and FORT ($\rho = 0.136$, p value < 0.001 , 95% CI from 0.091 to 0.181). A significant negative
46 association between FOI and BMI ($\rho = -0.045$, p value = 0.048, 95 CI from -0.090 to 0.001) was also
47 observed. A multivariable regression analysis considering FOI as a dependent variable confirmed that
48 Age and AMH are respectively negative and positive predictors of FOI.

49 **Conclusion:** This study suggests that age and AMH may be the most important predictors of FOI and
50 FORT which could be the best way to assess the ovarian sensitivity to exogenous gonadotropin. To
51 confirm our findings, larger analyses involving different ethnicities and multiple predictors will be
52 required in the future.

53

54 **Keywords:** AMH, FORT, FOI, Ovarian Sensitivity, IVF, ICSI, ART

55 **Introduction**

56 Ovarian response to exogenous gonadotropin represents a matter of interest in reproductive field. To
57 assess it the “crude” number of oocytes retrieved after ovarian stimulation (OS) represent the primary
58 endpoint most used worldwide. There is a robust evidence that clearly demonstrated the relationship
59 between number of eggs collected and cumulative or live birth rate [1,2]. Thus, the higher the number
60 of oocytes retrieved, the better are the chance to become pregnant. Apart from the number of eggs
61 collected, even ovarian reserve markers such as Antral Follicle Count and Anti-Mullerian hormone
62 are widely used to predict the ovarian response [3,4]. Based on their measurements starting dose is
63 established with the aim to avoid hyper or poor responsiveness [5–7]

64 Despite the indisputable usefulness both the number of oocytes and ovarian reserve markers their use
65 does not appropriately reflect the ovarian responsiveness of women that mainly depend on the
66 integrated evaluation of both parameters. In clinical practice, it is not infrequent to manage women
67 that retrieved less than expected oocytes at the end of stimulation despite the optimal ovarian reserve.
68 Such discrepancy was also defined as hypo-response [8] that is characterized by a low prognosis to
69 IVF [9]. According to a “real world data” including over 13,000 IVF women with different ethnicity,
70 the overall prevalence of “hypo-responders” women ranges between 44.2% and 36.1% [10] To better
71 express the concept of “hypo-response”, Alviggi and Conforti on behalf of Poseidon group introduced
72 the “Follicle to Oocytes index” (FOI) that consists in the ratio between the number of oocytes
73 collected at the ovum pick up and the number of antral follicles at the beginning of ovarian stimulation
74 [11]. This novel tool was also proposed as key performance indicator by SIFES [12] to evaluate the
75 efficiency of ovarian stimulation. When the ovarian stimulation is efficient, the ovarian reserve has
76 been adequately exploited conducting an “efficient” stimulation in terms of starting dose, dose
77 adjustment and triggering. It means that the number of oocytes retrieved is consistent with ovarian
78 reserve. Conversely, if a reduced amount of antral follicle count has been exploited it means that
79 something “wrong” happened during ovarian stimulation. Apart from FOI index, even “follicle output

80 rate” (FORT) was proposed as tool to evaluate the ovarian responsiveness [13] that is defined by the
81 ratio between the number of follicles at the time of *pick up* and antral follicle count.

82 Despite growing interest in functional ovarian response indices, the determinants of ovarian
83 sensitivity in relation to chronological age, ovarian reserve markers, and anthropometric parameters
84 remain poorly characterized. The present study sought to evaluate the predictors of ovarian sensitivity
85 using FOI and FORT.

86

87 **Material and Method**

88 *Study population*

89 This retrospective study was carried out at an Indian tertiary care (Indira IVF Hospital Ltd) centre
90 and was based on clinical data obtained from women who underwent IVF/ICSI treatment between
91 January 2023 and July 2024. The data was taken from centralised system of Hospital. The study was
92 conducted in compliance with the principles outlined in the Declaration of Helsinki. Written informed
93 consent was obtained from all participants for the anonymized use of their data. Ethical clearance for
94 the study was approved by the Institutional Ethics Committee (IIHPL Institutional Ethics Committee;
95 IIVF-UA00084). The following inclusion criteria were considered: age between 21- 40 years old;
96 daily gonadotropin dosage between 225-300 IU; antral follicle count based according to Broekman
97 criteria [14] between 5 and 25; Body Mass Index (BMI) between 18.5 and 30 Kg/m²; antagonist
98 protocol. Exclusion criteria were FOI and FORT above 1.2; dual trigger or GnRH agonist triggering;
99 oocyte donors cycle.

100

101 *Ovarian stimulation protocol*

Ovarian stimulation was conducted tailoring gonadotropin starting dosage according to the ovarian reserve [6]. Both recombinant and urinary formulations have been used to carry out ovarian stimulation. Flexible antagonist protocol for pituitary suppression [15] and hCG triggering at the dosage between 5.000 IU and 10.000 IU were adopted when at least 3 follicles $\geq$ 16-17 mm were observed at ultrasound monitoring. Embryo transfer was performed 5 days after oocytes retrieval. Luteal phase support was sustained using oral progesterone. In case of frozen embryo transfer the endometrial preparation was carried out adopting natural modified cycle or HRT, as reported elsewhere [16].

110

111 *Statistical analysis*

Continuous variables were summarized using mean $\pm$ standard deviation (SD), median with interquartile range (IQR), and range (minimum-maximum). Categorical variables were presented as frequencies and percentages. Comparisons of Follicle-to-Oocyte Index (FOI) and Follicular Output Rate (FORT) with Age and Anti-Müllerian Hormone (AMH) levels were conducted using one-way ANOVA or Kruskal-Wallis tests, as appropriate. Pairwise comparisons were adjusted using Bonferroni correction. Spearman's rank correlation coefficient (ρ) was used to assess the relationship between FOI/FORT and Age, AMH, and Body Mass Index (BMI). The 95% confidence intervals (CIs) for correlation coefficients were computed, and significance was determined using p-values. Univariate linear regression models were used to analyze the effect of independent variables (Age, AMH, BMI, Height, and Weight) on FOI and FORT. Significant predictors from univariate analysis were further examined in multivariable linear regression using stepwise selection. The model coefficients (β), standard errors (SE), t-values, and p-values were reported, along with 95% confidence intervals. Statistical analyses were performed using IBM SPSS (Statistical Package for Social science, Version 22) and R studio (RStudio: Integrated Development Environment for R. Posit

126 Software, PBC, Boston, MA. URL <http://www.posit.co/>. version 12.1). A p-value <0.05 was
127 considered statistically significant.

128

129 **Results**

130 A total of 81,547 women underwent IVF/ICSI cycle. Among them 1,942 women fitted the inclusion
131 and exclusion criteria (Figure 1). Characteristics of the study population included are reported in
132 Table 1. Women involved in the analysis were clustered in different ages (21-30 years; 31-35 years;
133 above 35 years) and Anti-Mullerian hormone values (below or equal to 0.49 ng/ml; 0.50-1.19 ng/ml;
134 1.2-3.5 ng/ml; above 3.5 ng/ml).

135

136 *FOI and FORT and chronological age*

137 Correlation analysis revealed that FOI ($\rho = -0.145$, p value = <0.001, 95% CI from -0.189 to -0.100)
138 and FORT ($\rho = -0.084$, p value <0.001, 95% CI from -0.129 to -0.038) are negatively associated with
139 aging (Figure 2a-b). Considering the distribution of FOI and FORT values according to age clustering,
140 significantly higher values were observed in younger women versus the older counterpart (Table 2).

141

142 *FOI and FORT and AMH*

143 Correlation analysis revealed that FOI ($\rho = 0.141$, p value <0.001, 95% CI from 0.096, 0.186) and
144 FORT ($\rho = 0.136$, p value <0.001, 95% CI from 0.091 to 0.181) are positively associated with AMH
145 (Figure 3a-b). Considering the distribution of FOI and FORT values according to AMH clustering,
146 significantly higher values were observed in women with higher AMH levels (Table 3).

147

148 *FOI and FORT and anthropometric values*

149 Correlation analysis showed a significant negative association between FOI and BMI ($\rho = -0.045$, p
150 value = 0.048, 95 CI from -0.090 to 0.001) (Figure 4). On the other hand, not a significant correlation
151 between FORT and BMI was observed. No significant effect was observed between FOI/FORT and
152 Height and Weight.

153

154 *Multivariate regression analysis FOI and FORT*

155 This multivariable linear regression analysis was carried out to establish the relationship between the
156 dependent variable FOI/FORT and the independent variables AMH, Age using a stepwise regression
157 method. Considering FOI, in this initial model, only AMH is included as a predictor of FOI. The
158 positive coefficient for AMH (0.042) is statistically significant ($p < 0.001$), suggesting a strong
159 positive relationship between AMH levels and FOI, where higher AMH is associated with higher FOI.
160 In the second model, both AMH and Age are included. AMH remains a significant positive predictor
161 of FOI ($p < 0.001$), though the coefficient has decreased slightly from Model 1 (0.036). Age is added
162 as a significant negative predictor ($p < 0.001$), indicating that as age increases, FOI decreases. Both
163 predictors significantly contribute to the model.

164 On the other hand, when dependent variable considered was FORT only, AMH was retained in the
165 final model because age did not improve the predictive power of the model when AMH was already
166 included.

167

168 **Discussion**

169 This study aimed to assess the influence of several demographic and clinical predictors, including
170 height, weight, BMI, age, and AMH, on ovarian sensitivity indices, namely FOI and FORT.

171 Multivariate regression analysis identified age and AMH as the only significant predictors, with
172 increasing age negatively affecting ovarian sensitivity, whereas higher AMH levels were positively
173 associated with improved FOI and FORT.

174 Thus, with respect to ovarian sensitivity, our findings indicate that *biological* ovarian age, as reflected
175 by AMH levels, is a more relevant determinant than chronological age. Notably, across all age
176 categories, the positive association between AMH and FOI remained consistent. An additional
177 important observation is that chronological age was associated with FOI but not with FORT. This
178 discrepancy may be explained by the greater susceptibility of FOI to mechanisms related to oocyte
179 competence, such as follicular recruitment and triggering dynamics, whereas FORT appears to be less
180 influenced by age-related qualitative changes [17,18]

181 These findings highlight the importance of both age and AMH in predicting ovarian response, which
182 is consistent with previous research indicating that AMH is a valid marker of ovarian reserve and age
183 is a well-established markers of ovarian reserve [19]. In this paper we observed that AMH is positively
184 associated with FORT and FOI. This aspect is consistent with the fact that AMH positively correlated
185 with number of oocytes retrieved and antral follicle count [19]. Indeed, several studies have found
186 that greater AMH levels are related with improved ovarian reserve and increased likelihood of
187 successful oocyte retrieval [20]. In our study, AMH has emerged as a strong positive predictor of FOI
188 and FORT. From clinical point of view this implicated that in women with elevated AMH is more
189 feasible to collect number of oocytes consistent with ovarian reserve. The mean value observed in
190 our records is consistent with competence values proposed by SIFES [12]

191 The negative relationship between age and outcome is consistent with the concept that fertility falls
192 with age, owing to a decrease in both the quantity and quality of eggs [21–23]. As individuals age,
193 the likelihood of having desirable reproductive outcomes increases. This finding supports numerous
194 studies [24] showing that fertility declines with age due to the natural aging process of the ovaries,
195 with a decline in oocyte quantity, chromosomal integrity, and embryo development [25]. As a result,

196 Age has been recognized as one of the strongest predictors of reproductive outcomes. This study
197 provides unique evidence from an Indian population linking age to ovarian sensitivity, as
198 measured by the follicle–oocyte index (FOI).

199 It means that age in some way regulates ovarian response to exogenous medication. One possible
200 explanation behind this observation is that aging per se represent a condition related with
201 gonadotropin deficiency [26]. Especially, luteinizing activity and consequently androgen production
202 seems to be impaired in advanced reproductive age [27]. These aspects could in some way affect
203 ovarian sensitivity thereby reducing FOI and FORT in women with advanced maternal age, although
204 most of the mechanisms involved have yet to be clarified.

205 Previous studies confirmed that women with increase BMI could show a resistance to exogenous
206 gonadotropin [28,29]. In detail higher amount of gonadotropins and fewer oocytes retrieved are
207 usually observed in women with obesity compared with normal-weight controls [30]. In this analysis
208 for the first time, we observed a negative association between BMI and FOI confirming that metabolic
209 disorders could affect ovarian sensitivity.

210 The main limitation of this study resides in the retrospective design that renders our findings more
211 prone to bias. Nonetheless, the elevated sample size allows a proper evaluation of several main factors
212 related to ovarian sensitivity, not explored elsewhere. Another important limitation is the fact only
213 few independent variables have been measured. This implies that we are still far from totally
214 elucidating all factors that could be implicated in the ovarian response. Finally, only the most widely
215 used triggering strategy (hCG triggering) has been adopted in our population study. Hence, the impact
216 of different triggering strategies on ovarian output (especially FOI assessment) needs to be evaluated
217 in future studies. Despite these limitations major strength lies in the high degree of uniformity
218 maintained across all centres via similar Protocol, gonadotropin dose utilization triggering agent ,
219 training of the consultants involved , which ensures exceptional methodological consistency.

220 In conclusion, our retrospective studies suggest that age and AMH are the most important predictors
221 of FOI and FORT that represent a trustful way to assess the ovarian sensitivity to exogenous
222 gonadotropin. Our findings might encourage further investigations regarding this topic, and need to
223 be corroborated in future by more robust studies.

224 **Declarations**

225

226 *Ethics approval and consent to participate:* Written informed consent was obtained from all
227 participants for the anonymized use of their data. Ethical clearance for the study was approved by the
228 Indira IVF Hospital Institutional Ethics Committee (IIVF-UA00084).

229 *Consent for publication:* Not applicable.

230 *Availability of data and materials:* The datasets used and/or analyzed during the current study are
231 available from the corresponding author on reasonable request.

232 *Competing interests:* Alessandro Conforti and Carlo Alviggi declared personal fee outside the
233 submitted paper. The author authors do not have any conflict of interest.

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235 *Author contribution:* AC, CA, NRB, SS, VC: Conceptualization, Methodology; VC, KM, NM: Data
236 curation, resources; AC, SS: Writing- Original draft preparation. SS: Visualization, Investigation. AC,
237 CA, SS: Supervision.: WM: Software, Validation; AC, IS: Writing- Reviewing and Editing. All
238 authors read and approved the final manuscript.

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241

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247

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251 Table 3. Comparison of FOI/FORT with AMH

252

253 **Reference**

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Figure 1. Flow chart

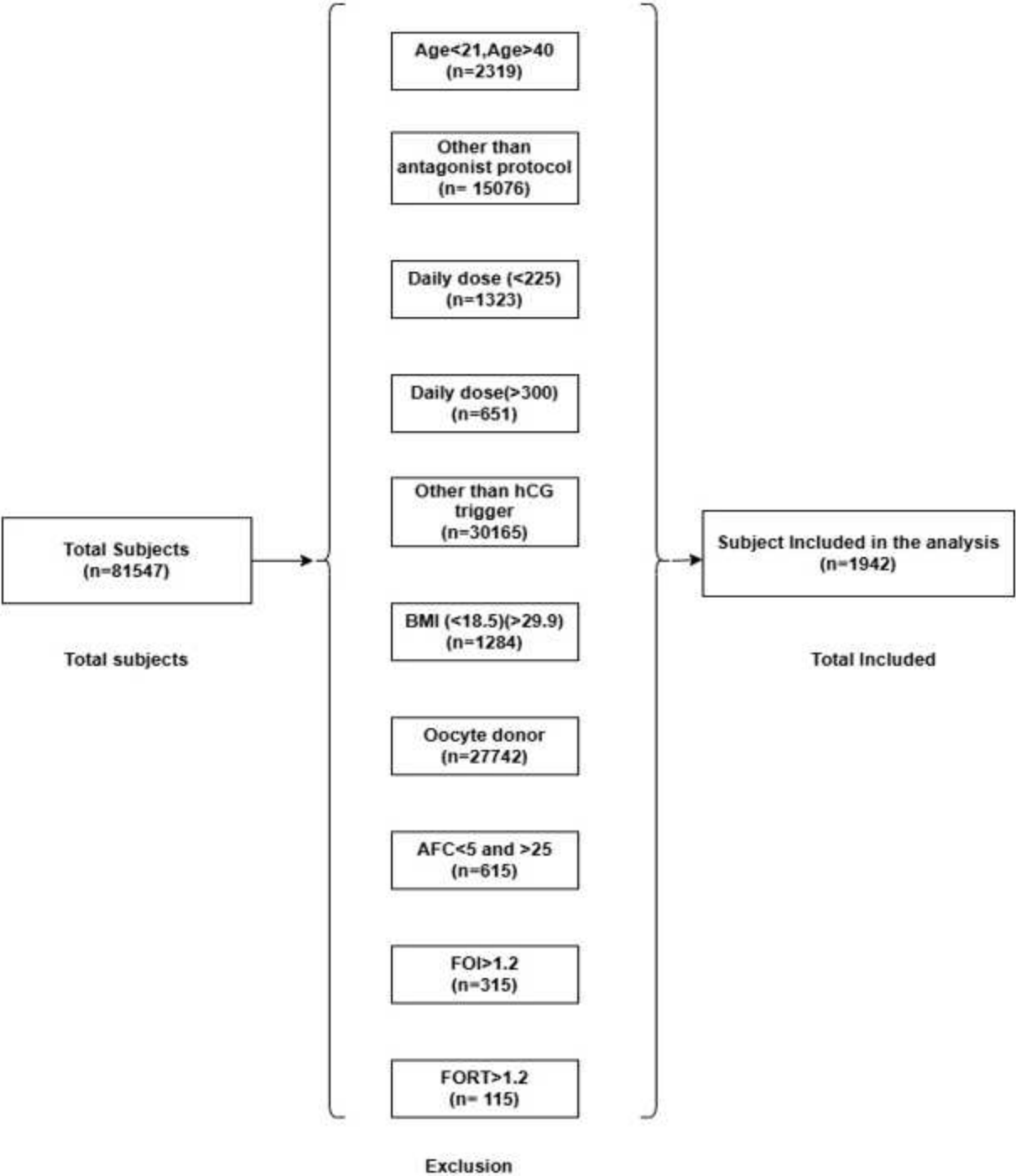


Figure 2a. Correlation between patient age and follicle to Oocyte Index

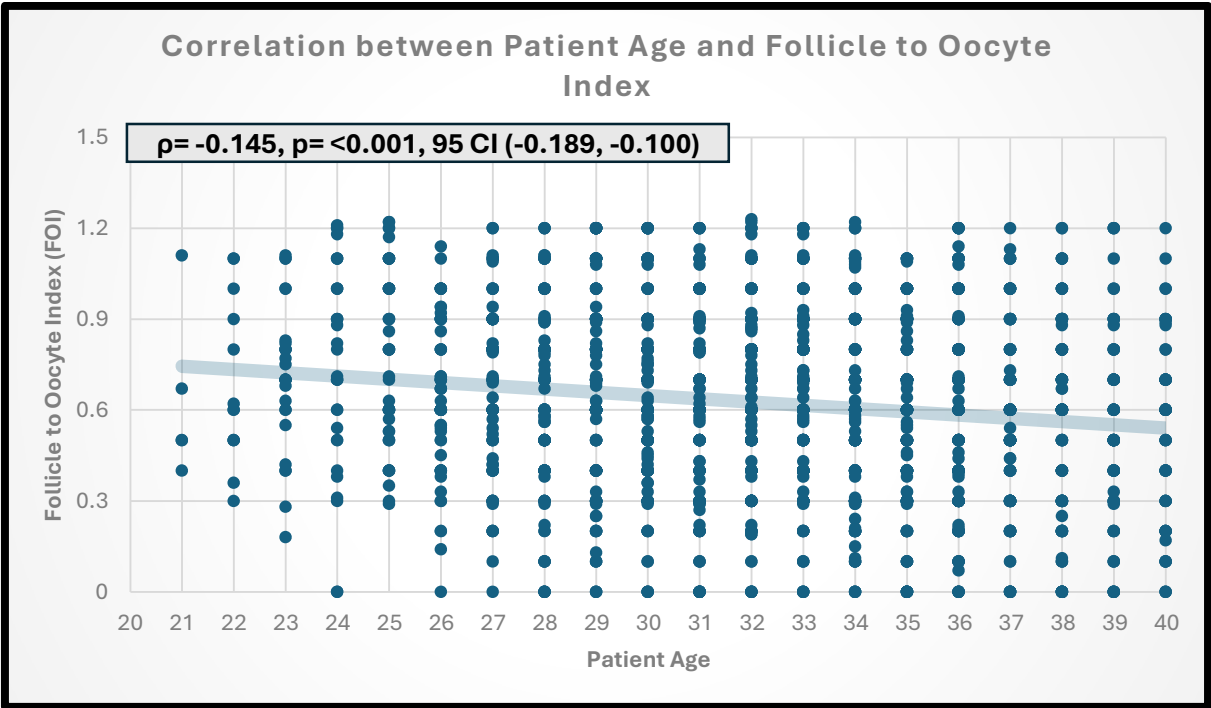


Figure 2b. Correlation between patient age and follicle output rate

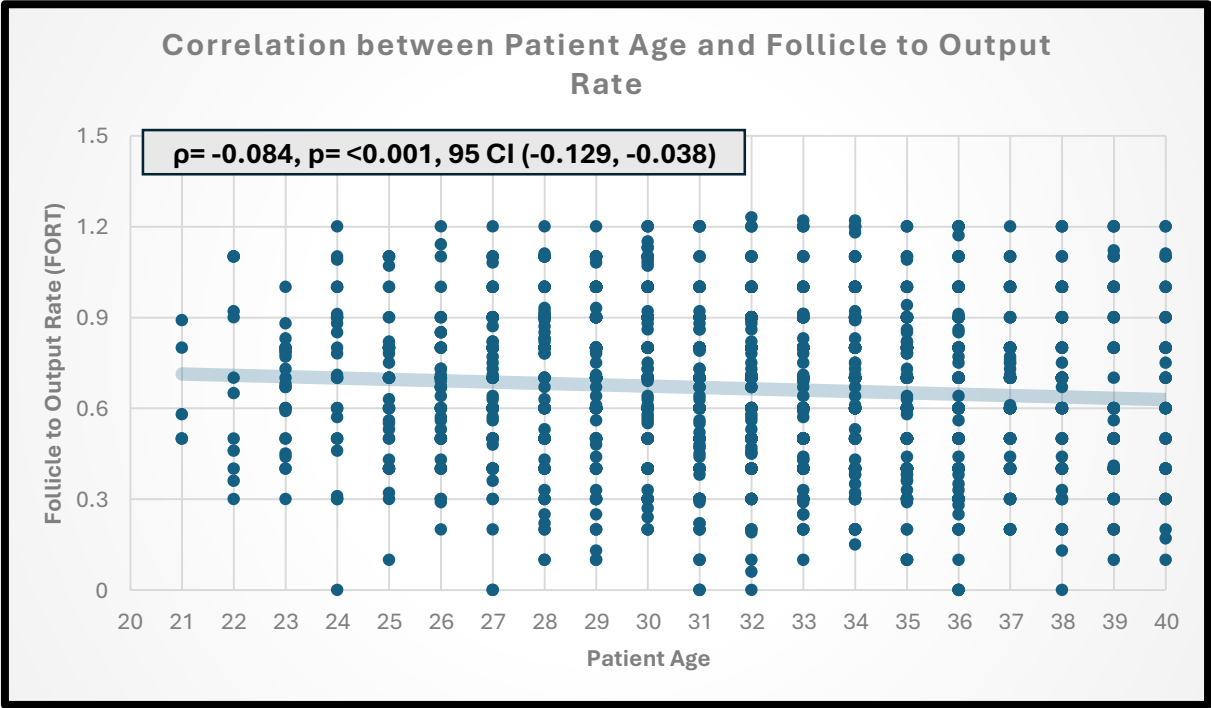


Figure 3a -b: Correlation between anti-Mullerian Hormone and Follicle to oocyte index

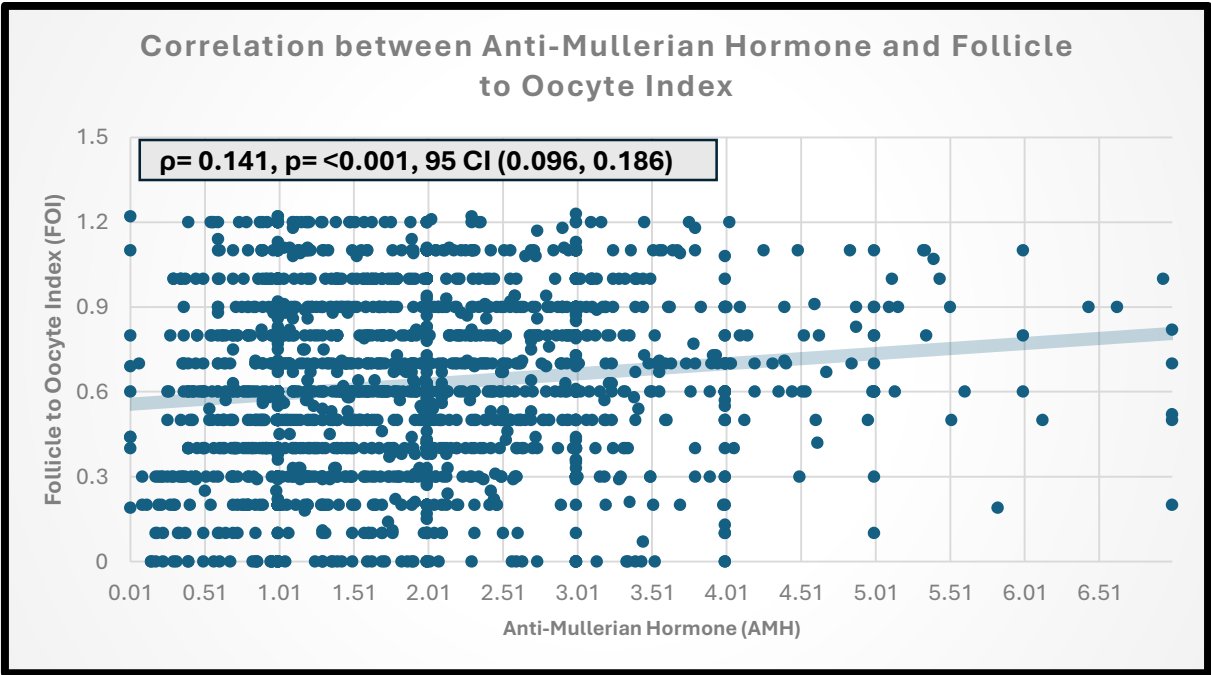


Figure 3b

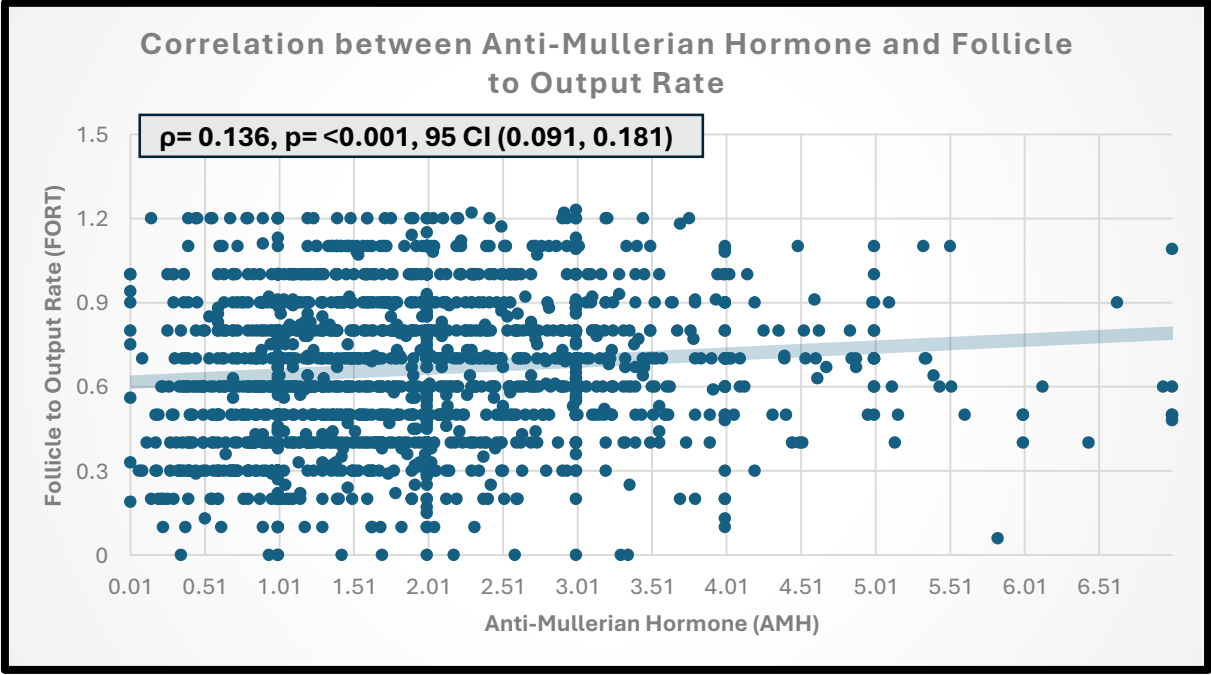
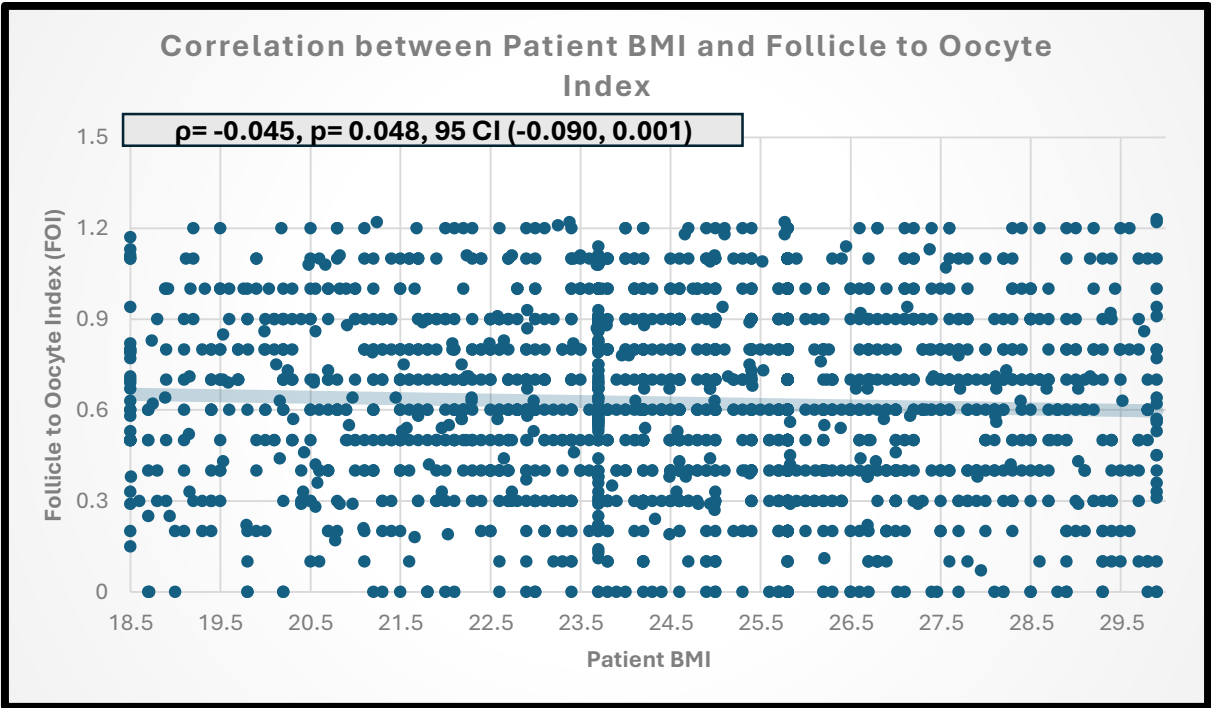


Figure 4. Correlation between BMI and FOI



Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- [Table1.Basalcharacteristics.docx](#)
- [Table2.docx](#)
- [Table3.docx](#)