



Research Article

Development and Validation of Fertility Index for Turkey Toms Based on Semen Parameters from Nigerian Local Turkey

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ABSTRACT

Fertility assessment in turkey toms is important for optimizing reproductive efficiency, especially in commercial breeding programs. To optimize reproductive efficiency in turkey breeding, we developed and validated a fertility index for turkey toms using semen parameters, with sperm penetration holes as the response variable. Semen samples from 100 toms were analyzed for six parameters, revealing sperm mobility index ($r = 0.864$) and progressive motility ($r = 0.851$) as the strongest predictors of fertility. A simplified linear regression model ($R^2 = 0.851$) using semen volume, sperm concentration, and sperm mobility index was selected to create the Turkey Tom Fertility Index. The scaled 0-100 index demonstrated 77.0% overall classification accuracy and 86.7% test set accuracy. This practical tool offers a standardized method for predicting fertility potential, enabling improved breeding efficiency and reproductive outcomes in commercial turkey production.

Keywords: Breeding soundness evaluation; Fertility index; Semen parameters; Sperm penetration holes; Turkey toms

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INTRODUCTION

Reproductive efficiency is a critical factor in commercial turkey production, with male fertility being a significant determinant of overall flock productivity (Lake, 1989). Turkey toms contribute half of the genetic material to offspring, yet their fertility assessment remains challenging due to the complex interplay of various semen quality parameters (Bakst & Dymond, 2013).

Unlike mammalian species where direct fertilization can be observed, avian reproduction involves unique physiological mechanisms that complicate fertility evaluation (Blesbois & Brillard, 2007; Brillard, 2003). The assessment of turkey tom fertility has traditionally relied on various semen parameters measured individually, including semen volume, sperm

concentration, motility, viability, and morphology (Iaffaldano *et al.*, 2018). However, these individual parameters often provide an incomplete picture of a tom's actual fertility potential. The relationship between standard semen quality measurements and actual fertility outcomes remains poorly understood, leading to potential inefficiencies in breeding programs (Bakst, 1993; Christensen *et al.*, 2005).

In turkey reproduction, sperm penetration holes in the inner perivitelline layer of eggs have emerged as a reliable indicator of actual fertility potential (Bramwell *et al.*, 1995; Wishart, 1987)). These holes represent successful sperm penetration attempts and correlate strongly with fertilization rates. Although in the *in vivo* assay, this measurement can only be assessed after eggs are laid, making it a retrospective rather than predictive measure of fertility (Bakst *et al.*, 2010), the *in vitro* version can be considered predictive. The development of a predictive fertility index that combines multiple semen parameters to estimate potential fertility, as measured by sperm penetration holes, would provide significant advantages to the turkey breeding industry. Such an index would allow for early identification of high-fertility toms before breeding, more efficient allocation of breeding resources, improved selection decisions in breeding programs, standardized assessment protocols across production systems and potential increases in overall flock fertility and productivity.

Previous attempts to develop fertility indices in poultry have shown promise but have often been limited by small sample sizes, focus on single parameters, or lack of validation against actual fertility outcomes (Donoghue, 1999; King *et al.*, 2000). Other studies have suggested that multivariate approaches incorporating several semen quality parameters may provide more accurate fertility predictions (Bowling *et al.*, 2003; Neuman *et al.*, 2002).

The present study aims to address these limitations by developing and validating a comprehensive fertility index for turkey toms based on multiple semen parameters. We, specifically, sought to identify the key semen parameters most strongly associated with sperm penetration holes; develop a predictive model that accurately estimates fertility potential; create a practical, standardized fertility index for use in commercial and research settings; validate the index against actual fertility outcomes and establish classification thresholds for practical application in breeding programs. By creating a reliable fertility index, this research contributes to the broader goal of improving reproductive efficiency in turkey production systems while providing a valuable tool for both

researchers and industry professionals involved in turkey breeding and reproduction.

MATERIALS AND METHODS

Study design and data collection

This study was conducted at the Department of Theriogenology and Production, Faculty of Veterinary Medicine, Ahmadu Bello University Zaria. Dataset from 100 sexually matured turkey toms was utilized for the research. Semen samples were collected from each tom using the abdominal massage method (Burrows & Quinn 1937). Collections were performed by trained technicians under standardized conditions to minimize variation in collection technique. All toms were maintained under identical environmental conditions (13L:11D photoperiod, 18-34°C ambient temperature) under intensive system throughout the study period.

For each tom, the following semen parameters were measured; semen volume, which was measured directly in graduated collection tubes and recorded in milliliters (mL), Sperm concentration, which was determined using a Neubauer hemacytometer, with results expressed as billion sperm cells per milliliter (billion/mL); progressive motility, which was assessed by placing a drop of diluted semen on a pre-warmed slide and examining under phase-contrast microscope at 40x magnification. The percentage of sperm showing forward progressive movement was recorded; sperm mobility index, which was measured using the Accudenz® procedure as described by Umar *et al.*, (2021), it involved layering diluted semen over 6% Accudenz® solution, incubating at 41°C for 5 minutes, and measuring absorbance at 550 nm. Results were expressed as a percentage; sperm viability, which was determined using the eosin-nigrosin staining technique. A minimum of 200 sperm cells were counted per sample, with the percentage of live (unstained) sperm recorded and normal morphology, which was assessed using phase-contrast microscopy at 400x and 1000x magnification. A minimum of 200 sperm cells were evaluated per sample, with the percentage of morphologically normal sperm recorded (Partyka *et al.*, 2012).

The response variable, "Sperm penetration holes", was measured using two different methods; *in vivo* and *in vitro* sperm penetration assays, both described by Bask and Long (2010).

Insemination of the Hens

All hens were assessed to confirm the absence of spermatozoa in their sperm storage tubules by collecting a full clutch from each hen and incubating for 28 days. Two unfertilized eggs were then reserved from each hen for the *in vitro* sperm egg assay. The hens were then inseminated with doses containing 100 million

sperm cells, according to the method described by Sotirov (2002). The hens were turned upside down, pressure applied to the right side of the abdomen until the vent everted (venting) and a poultry insemination gun containing the fresh diluted semen which was obtained from the toms was inserted to a depth of about 1.5-2 cm into the cloaca, and semen containing 100×10^6 spermatozoa was deposited in the vagina.

***In vivo* sperm-egg assay**

The oviposited eggs (from inseminated hens) were broken and the albumen separated from the yolk. The yolk was then placed in a pan with the blastoderm positioned upwards. Excess albumen was removed by blotting with paper towel and 2% NaCl solution was added; an area of 3.0 mm^2 of the perivitelline layer (PL) over the blastoderm was carefully cut and immediately rinsed in phosphate buffered saline (PBS) to remove excess yolk material. The perivitelline layer was then placed on a glass slide and 4 drops of 3% formalin were added to fix the membrane and immediately decanted. The perivitelline layer was finally stained with Schiff's reagent. The holes were then counted using dark phase microscopy (Datyson Biological microscope) at a magnification of X 40.

***In vitro* sperm-egg assay**

In this tests, ten (10) μL of fresh semen was diluted in 1000 μL of NaCl-TES the aliquots were stored at 40°C in a shaking water bath for 45 min before assay. A fragment of the isolated inner perivitelline (IPL) approximately $0.5 \text{ cm} \times 0.5 \text{ cm}$ square was cut and added to a vial containing 100 μL of NaCl-TES diluted semen in 1 ml Dulbecco's Modified Eagle Medium (DMEM). The mixture was incubated at 40°C for 5 min in a shaking water bath. The incubated IPL was removed from the vial and washed in 1% NaCl. The IPL was carefully spread onto a microscope slide using forceps avoiding wrinkles and a cover-slip was applied. The slide was then viewed immediately using dark phase microscopy at X 10 and X 40 objectives. The number of holes per 3 mm^2 were counted according to the method described by Baskt and Long (2010).

Data Analysis

Exploratory data analysis

Initial data exploration was conducted to examine the distribution of each variable and identify potential outliers. Descriptive statistics including mean, standard deviation, minimum, maximum, and quartiles were calculated for all parameters. The Shapiro-Wilk test was used to assess normality of distributions. Correlation analysis was performed to examine relationships between predictor variables and the response variable, with Pearson's correlation coefficients calculated for all pairs of variables.

Multicollinearity among predictor variables was assessed using variance inflation factors (VIF), with values above 10 considered indicative of problematic multicollinearity. Feature importance was evaluated using F-scores from univariate regression analyses to identify the most significant predictors of sperm penetration holes.

Model development

Multiple regression models were developed to predict sperm penetration holes based on semen parameters. The dataset was split into training (80%) and testing (20%) sets using random sampling stratified by the response variable. The following models were evaluated. 1. Multiple Linear Regression: A standard linear model including all predictor variables. 2. Ridge Regression: Linear regression with L2 regularization to address potential multicollinearity. 3. Lasso Regression: Linear regression with L1 regularization for feature selection. 4. Random Forest Regression: An ensemble method using multiple decision trees. 5. Gradient Boosting Regression: An ensemble method that builds trees sequentially. 6. Support Vector Regression: A non-linear approach using radial basis function kernel. 7. Simplified Linear Regression: Using only the top two predictors identified in feature importance analysis. 8. Simplified Linear Regression: Using only the top three predictors identified in feature importance analysis. Model performance was evaluated using multiple metrics i. R^2 (coefficient of determination) ii. Root Mean Square Error (RMSE) iii. Mean Absolute Error (MAE) iv. Cross-validation R^2 using 5-fold cross-validation.

Fertility index development

Based on the best-performing model, a fertility index was developed following these steps: 1. formula creation, the regression coefficients from the selected model were used to create a mathematical formula for calculating a raw fertility index, 2. Scaling; the raw index was scaled to a 0-100 range using min-max normalization.

(Scaled Index = $((\text{Raw Index} - \text{Min}) / (\text{Max} - \text{Min})) \times 100$) where Min and Max represent the minimum and maximum values of the raw index in the dataset and 3. Classification thresholds, thresholds for categorizing toms into fertility classes were established based on the distribution of scaled index values (Low Fertility, < 25th percentile, Medium Fertility: 25th to 75th percentile and High Fertility: > 75th percentile)

Validation methods. We validated the generated index using four different approaches, i. Classification accuracy, the agreement between predicted fertility classifications and actual classifications based on sperm penetration holes was assessed using confusion matrices, accuracy, precision, recall, and F1-scores, ii.

Cross-validation, the index was evaluated on a held-out test set not used during model development to assess generalizability, iii. Practical usability, the index was tested with example cases representing the full range of potential fertility levels to verify practical application and iv. Sensitivity analysis: The response of the index to variations in input parameters was examined by systematically varying each parameter while holding others constant.

All statistical analyses were performed using Python 3.10 with the following libraries: pandas (1.5.3), numpy (1.24.3), scikit-learn (1.2.2), statsmodels (0.14.0), and matplotlib (3.7.1) for visualization. A significance level of $\alpha = 0.05$ was used for all statistical tests.

RESULTS

The analysis included semen samples from 100 turkey toms. Descriptive statistics for all measured parameters are presented in Table 1. The mean semen volume was 0.34 ± 0.09 mL, with values ranging from 0.20 to 0.50 mL. Sperm concentration averaged 7.40 ± 0.89 billion/mL, with a range of 5.50 to 9.70 billion/mL. Progressive motility showed a mean of $82.9 \pm 8.2\%$, while the sperm mobility index averaged $42.0 \pm 8.9\%$. Sperm viability and normal morphology had means of $82.9 \pm 6.3\%$ and $83.2 \pm 5.8\%$, respectively. The response variable, sperm penetration holes, averaged $158.4 \pm$

15.2 holes per sample, with values ranging from 130 to 197 holes.

Correlation analysis

Correlation analysis revealed significant relationships between several semen parameters and sperm penetration holes (Table 2). The strongest correlations with sperm penetration holes were observed for sperm mobility index ($r = 0.864$, $p < 0.001$) and progressive motility ($r = 0.851$, $p < 0.001$). Normal morphology and sperm viability showed weak positive correlations ($r = 0.183$, $p = 0.068$ and $r = 0.155$, $p = 0.123$, respectively), while sperm concentration demonstrated a very weak positive correlation ($r = 0.116$, $p = 0.249$). Semen volume showed a negligible negative correlation ($r = -0.042$, $p = 0.677$) with sperm penetration holes.

Notable correlations were also observed between certain predictor variables. Sperm mobility index and progressive motility showed a strong positive correlation ($r = 0.943$, $p < 0.001$), indicating potential multicollinearity. Similarly, normal morphology and sperm viability were strongly correlated ($r = 0.796$, $p < 0.001$). The variance inflation factor (VIF) analysis confirmed multicollinearity concerns for progressive motility (VIF = 9.76) and sperm mobility index (VIF = 9.59), while other parameters showed acceptable VIF values (< 3.0). The relationship between top predictor variables is demonstrated in Figure 1.

Table 1. Descriptive statistics of semen parameters and sperm penetration holes in turkey toms (n=100)

Parameter	Mean \pm SD	Minimum	25th Percentile	Median	75th Percentile	Maximum
Semen Volume (mL)	0.34 ± 0.09	0.20	0.26	0.34	0.42	0.50
Sperm Concentration (10^9 /mL)	7.40 ± 0.89	5.50	6.80	7.40	8.10	9.70
Progressive Motility (%)	82.9 ± 8.2	60.0	78.0	83.0	89.0	95.0
Sperm Mobility Index (%)	42.0 ± 8.9	25.0	35.0	42.0	50.0	60.0
Sperm Viability (%)	82.9 ± 6.3	70.0	78.0	83.0	88.0	95.0
Normal Morphology (%)	83.2 ± 5.8	71.0	79.0	83.0	88.0	94.0
Sperm Penetration Holes (/m ²)	158.4 ± 15.2	130.0	145.8	158.0	170.0	197.0

All parameters showed normal distributions as confirmed by Shapiro-Wilk tests ($p > 0.05$), with no extreme outliers identified

Table 2. Correlation coefficients between semen parameters and sperm penetration holes

Parameter	Correlation with Sperm Penetration Holes	p-value
Sperm Mobility Index	0.864	< 0.001
Progressive Motility	0.851	< 0.001
Normal Morphology	0.183	0.068
Sperm Viability	0.155	0.123
Sperm Concentration	0.116	0.249
Semen Volume	-0.042	0.677

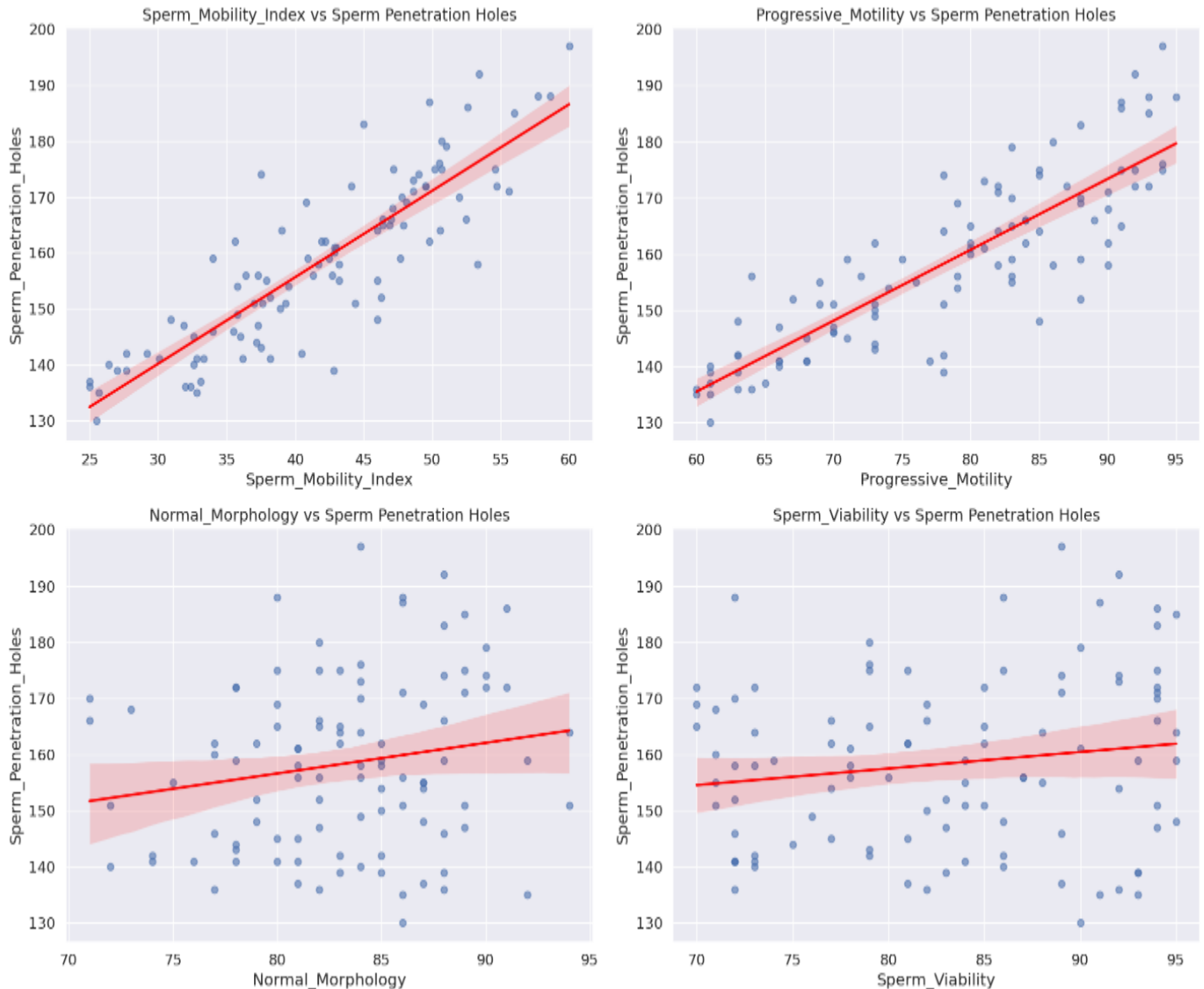


Figure 1. The relationships between top predictor variables and sperm penetration holes

(A) Sperm Mobility Index vs. Sperm Penetration Holes, (B) Progressive Motility vs. Sperm Penetration Holes, (C) Sperm Concentration vs. Sperm Penetration Holes, and (D) Semen Volume vs. Sperm Penetration Holes

Feature importance analysis

Feature importance analysis using F-scores from univariate regression further confirmed the significance of sperm mobility index and progressive motility as predictors of sperm penetration holes (fig 2). Sperm mobility index showed the highest F-score (287.74, $p < 0.001$), followed closely by progressive motility (258.24,

$p < 0.001$). The remaining parameters showed substantially lower F-scores, with normal morphology ($F = 3.39$, $p = 0.068$) and sperm viability ($F = 2.42$, $p = 0.123$) demonstrating marginal importance. Sperm concentration ($F = 1.35$, $p = 0.249$) and semen volume ($F = 0.17$, $p = 0.677$) showed minimal predictive value in univariate analysis (Table 3).

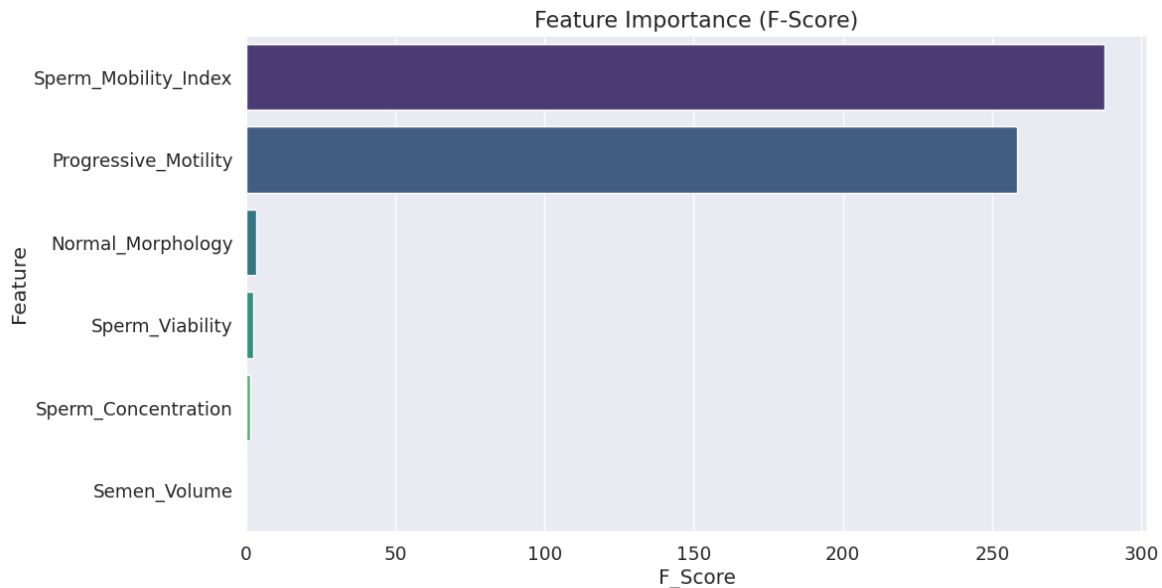


Figure 2. The relative importance of semen parameters for predicting sperm penetration holes based on F-scores from univariate regression analysis

Higher F-scores indicate stronger predictive value.

Table 3: Feature importance analysis for predicting sperm penetration holes

Parameter	F-Score	p-value
Sperm Mobility Index	287.74	<0.001
Progressive Motility	258.24	<0.001
Normal Morphology	3.39	0.068
Sperm Viability	2.42	0.123
Sperm Concentration	1.35	0.249
Semen Volume	0.17	0.677

Predictive model performance

Eight different regression models were evaluated for their ability to predict sperm penetration holes based on semen parameters (Table 4). The multiple linear regression model demonstrated the best performance on the test set with an R^2 of 0.865, indicating that approximately 86.5% of the variance in sperm penetration holes could be explained by the model. This model also showed the lowest test RMSE of 5.84, suggesting high prediction accuracy. Ridge and lasso regression models performed similarly with test R^2 values of 0.864 and 0.863, respectively.

The simplified linear regression model using only the top three predictors (sperm mobility index, progressive

motility, and sperm concentration) achieved a test R^2 of 0.851, which was comparable to the full model. This suggests that these three parameters capture most of the predictive information for sperm penetration holes. The cross-validation results showed consistent performance across folds, with mean CV R^2 values ranging from 0.740 to 0.792 across models.

Based on these results, the simplified linear regression model with the top three predictors was selected for fertility index development due to its strong performance and practical simplicity. The coefficients for this model are presented in Table 5.

Table 4. Performance comparison of predictive models for sperm penetration holes

Model	Train R ²	Test R ²	Test RMSE	CV R ² (mean)	CV R ² (std)
Linear Regression	0.820	0.865	5.84	0.792	0.127
Ridge Regression	0.819	0.864	5.85	0.790	0.127
Lasso Regression	0.818	0.863	5.86	0.788	0.127
Linear Regression (Top 3)	0.781	0.851	6.12	0.792	0.127
Gradient Boosting	0.996	0.847	6.20	0.761	0.094
Random Forest	0.967	0.842	6.30	0.740	0.102
SVR	0.889	0.835	6.44	0.781	0.070
Linear Regression (Top 2)	0.761	0.831	6.52	0.792	0.127

Table 5. Coefficients of the simplified linear regression model for fertility index

Parameter	Coefficient
Intercept	63.93
Semen Volume (mL)	13.76
Sperm Concentration (billion/mL)	3.28
Sperm Mobility Index (%)	1.57

The resulting formula for the raw fertility index was: Fertility Index = 63.93 + (13.76 × Semen_Volume) + (3.28 × Sperm Concentration) + (1.57 × Sperm_Mobility_Index)

Fertility Index Development and Validation

The raw fertility index values ranged from 132.12 to 190.12 across the dataset. These values were scaled to a 0-100 range using min-max normalization to create the final Turkey Tom Fertility Index. Classification thresholds were established based on the distribution of scaled index values. Low Fertility: < 28.64 (25th percentile), Medium Fertility: 28.64 - 62.55 (25th to 75th percentile) and High Fertility: > 62.55 (75th

percentile). Thus, the distribution of toms across fertility classifications was 25% in the Low category, 50% in the medium category, and 25% in the High category, reflecting the percentile-based threshold selection.

To validate the fertility index, toms were classified based on both the index and their actual sperm penetration holes values (using the same percentile thresholds). The confusion matrix comparing these classifications is presented in Table 6.

Table 6. Confusion matrix of predicted vs. actual fertility classifications

	Predicted Low	Predicted Medium	Predicted High	Total
Actual Low	19	6	0	25
Actual Medium	6	38	5	49
Actual High	0	6	20	26
Total	25	50	25	100

The overall classification accuracy was 77.0%, with class-specific precision and recall values presented in Table 7.

Table 7: Classification performance metrics for the fertility index

Fertility Class	Precision	Recall	F1-Score	Support
Low	0.76	0.76	0.76	25
Medium	0.76	0.78	0.77	49
High	0.80	0.77	0.78	26
Weighted Average	0.77	0.77	0.77	100

Cross-validation on a held-out test set (30% of the data) demonstrated even stronger performance, with an accuracy of 86.7% and improved precision and recall metrics across all classes (Table 8).

Table 8: Classification performance metrics on the test set

Fertility Class	Precision	Recall	F1-Score	Support
Low	1.00	0.88	0.93	8
Medium	0.77	0.91	0.83	11
High	0.90	0.82	0.86	11
Weighted Average	0.88	0.87	0.87	30

Sensitivity Analysis

Sensitivity analysis was conducted to evaluate the response of the fertility index to changes in input parameters. Each parameter was varied by $\pm 10\%$ and $\pm 20\%$ from a baseline case (Semen Volume = 0.35 mL, Sperm Concentration = 7.5 billion/mL, Sperm Mobility Index = 42.0%), and the resulting changes in the scaled fertility index were recorded (Table 9).

The analysis revealed that the fertility index was most sensitive to changes in sperm mobility index, with a 20% decrease changing the classification from Medium to Low and a 20% increase changing it from Medium to

High. Changes in sperm concentration had a moderate impact on the index value but did not alter the classification in the test case. Semen volume showed the least impact on the index value, with $\pm 20\%$ changes resulting in minimal index changes and no classification changes Figure 3.

The results here confirm that the fertility index responds appropriately to changes in input parameters, with the greatest sensitivity to the parameter (sperm mobility index) that showed the strongest correlation with sperm penetration holes in the initial analysis.

Table 9. Sensitivity analysis of the Turkey Tom Fertility Index

Parameter	Variation	Value	Scaled Index	Classification
Baseline	-	-	46.61	Medium
Semen Volume	-20%	0.28	44.95	Medium
Semen Volume	-10%	0.32	45.78	Medium
Semen Volume	+10%	0.39	47.44	Medium
Semen Volume	+20%	0.42	48.27	Medium
Sperm Concentration	-20%	6.0	38.13	Medium
Sperm Concentration	-10%	6.8	42.37	Medium
Sperm Concentration	+10%	8.3	50.85	Medium
Sperm Concentration	+20%	9.0	55.08	Medium
Sperm Mobility Index	-20%	33.6	23.91	Low
Sperm Mobility Index	-10%	37.8	35.26	Medium
Sperm Mobility Index	+10%	46.2	57.96	Medium
Sperm Mobility Index	+20%	50.4	69.30	High

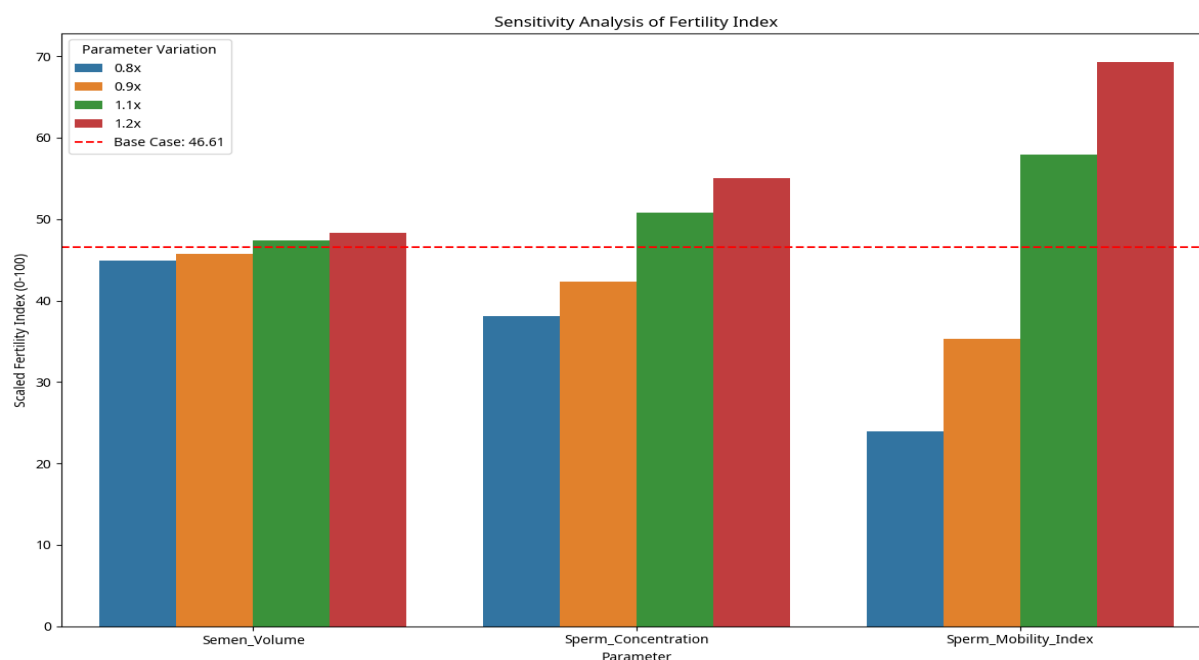


Figure 3. Sensitivity Analysis of Fertility Index

DISCUSSION

This study developed and validated a fertility index for turkey toms based on semen parameters, with sperm penetration holes as the response variable. The results demonstrate that a simplified model using just three semen parameters, semen volume, sperm concentration, and sperm mobility index—can effectively predict fertility potential in turkey toms. The Turkey Tom Fertility Index achieved an overall classification accuracy of 77.0% and a test set accuracy of 86.7%, indicating strong predictive performance.

The strong correlations observed between sperm mobility index ($r = 0.864$) and progressive motility ($r = 0.851$) with sperm penetration holes have been observed in previous research highlighting the importance of sperm motility in avian fertility (Froman *et al.*, 2002; Froman *et al.*, 1999; King *et al.*, 2000). The sperm mobility index, which measures the ability of sperm to penetrate an Accudenz solution, appears to be particularly valuable as a predictor of fertility potential. This finding supports the biological understanding that sperm must be capable of traversing the female reproductive tract and penetrating the inner perivitelline layer for successful fertilization (Froman *et al.*, 2005; Bakst *et al.*, 2010).

Semen volume showed a negligible negative correlation with sperm penetration holes ($r = -0.042$) in univariate analysis but emerged as an important predictor in the multivariate model. This suggests that semen volume may interact with other parameters in complex ways that are not apparent in simple correlation analysis. The positive coefficient for semen volume in the final model (13.76) indicates that, when controlling for other factors, larger ejaculate volumes are associated with higher fertility potential. This may reflect overall reproductive health and function in turkey toms.

The sensitivity analysis revealed that the fertility index is most responsive to changes in sperm mobility index, with a 20% decrease changing the classification from Medium to Low and a 20% increase changing it from Medium to High. This finding has practical implications for breeding programs, suggesting that interventions targeting sperm mobility could have the greatest impact on fertility outcomes.

The results of this study are consistent with previous research on turkey reproduction. Donoghue (1999) reported that sperm mobility was highly correlated with fertility in turkeys, with correlation coefficients ranging from 0.68 to 0.87 across different studies. Our observed correlation of 0.864 between sperm mobility index and sperm penetration holes falls at the upper end of this range, confirming the importance of this parameter.

Similarly, our findings on the relative importance of different semen parameters align with those of (Blesbois, 2011; Bowling *et al.* 2003), who found that motility measures were more predictive of fertility than concentration or morphology in turkey toms. However, unlike some previous studies that found significant correlations between sperm morphology and fertility (Hammerstedt, 1996; Christensen *et al.*, 2005), our results showed only a weak relationship between normal morphology and sperm penetration holes ($r = 0.183$). This may be due to differences in methodology or the relatively high and uniform morphology scores in our sample ($83.2 \pm 5.8\%$).

The classification accuracy of our fertility index (77.0% overall, 86.7% on the test set) compares favorably with previous attempts to predict fertility in poultry. Neuman *et al.* (2002) reported classification accuracies of 65-75% for a discriminant analysis model predicting fertility in broiler breeders, while King *et al.* (2000) achieved 72% accuracy using sperm mobility alone to classify turkey toms into fertility categories. Our higher accuracy likely reflects the advantage of combining multiple complementary parameters into a single index.

The Fertility Index offers several advantages over traditional fertility assessment methods; the index provides a prospective assessment of fertility potential before breeding, allowing for more informed selection decisions. It offers simplicity in that it requires only three easily measured parameters, the index is practical for routine use in both research and commercial settings. Additionally, the 0-100 scale and clear classification thresholds provide a standardized framework for comparing toms across different flocks and production systems. The index is based on a model with strong statistical performance ($R^2 = 0.851$) and has been validated through multiple approaches and biological relevance; it incorporates parameters that have clear biological connections to the fertilization process, enhancing its interpretability and credibility. For producers, this index provides a practical tool for selecting high-fertility toms, optimizing breeding pairs, and culling underperforming males, which can lead to significant cost savings and improved flock productivity. For researchers, the index offers a standardized measure for evaluating the impacts of nutritional supplements, management practices, or genetic selection programs on turkey reproductive fitness.

Despite its strengths, several limitations of the current study should be acknowledged, sample characteristics; the dataset included 100 turkey toms, which although substantial, may not capture the full range of variation in commercial flocks. Additionally, all toms were of similar age and maintained under identical conditions,

potentially limiting generalizability, indirect fertility measure; sperm penetration holes, although strongly correlated with fertility, are still an indirect measure; validation against actual hatching results would further strengthen the index, temporal considerations; semen quality can vary over time due to factors such as age, season, and health status. The current study provides a cross-sectional analysis but does not address the stability of the index over time. Breed specificity; the index was developed using data from a single turkey breed/strain. Different genetic lines may show variations in the relationships between semen parameters and fertility and environmental factors; the study did not account for environmental variables such as temperature, nutrition, or stress, which can influence both semen quality and fertility outcomes (Long and Kulkarni, 2004).

CONCLUSION

This study successfully developed and validated a practical fertility index for turkey toms based on semen volume, sperm concentration, and sperm mobility index. The index provides a standardized, quantitative method for predicting fertility potential with strong predictive performance. The simplified formula and clear classification thresholds make it a practical tool for routine use in both research and commercial settings. By enabling a more objective assessment of fertility, the index can help optimize breeding programs, improve resource allocation, and ultimately enhance reproductive efficiency in turkey production. Adoption of this fertility index by breeders and commercial turkey operations could reduce costs, improve flock productivity, and enhance genetic selection programs.

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