

THE EFFECT OF TWO INTRAVAGINAL PROGESTERONE DOSES AND OVUM PICK-UP OUTCOMES IN BROWN SWISS COWS: A BAYESIAN HIERARCHICAL ANALYSIS

Carlos QUISPE-EULOGIO¹ , Neale SANTIAGO-INGA¹ , Edith ANCCO-GOMEZ¹ , Jordan NINAHUANCA²  ,
Ide UNCHUPAICO-PAYANO² , Milagros GARCÍA-HOSPINAL¹ , and Cecil RIVERA-PALOMINO¹ 

¹Facultad de Ciencias de la Salud, Universidad Peruana Los Andes, Huancayo, Junín, Perú

²Facultad de Zootecnia, Universidad Nacional del Centro del Perú, Huancayo, Perú

 Email: jordan.nina96@gmail.com

 Supporting Information



ABSTRACT: This study evaluated the effect of two intravaginal progesterone doses on follicular recruitment, oocyte recovery, and oocyte viability in Brown Swiss cows undergoing ovum pick-up. Eight dry cows were allocated to two treatments: T1 (0.6 g progesterone) and T2 (1.2 g progesterone), with four cows per treatment and five repeated sessions per animal. The responses analyzed were the numbers of 2–4 mm follicles, 4–8 mm follicles, total recovered oocytes, recovery probability, and oocyte viability. Count data were analyzed with negative binomial models, whereas recovery and viability were modeled as binomial responses, with cow included as a random effect. Overall, T1 showed higher adjusted posterior means for the number of 2–4 mm follicles per ovum pick-up (OPU) session (7.62 vs. 5.36 follicles), 4–8 mm follicles per OPU session (4.17 vs. 3.27 follicles), and total recovered oocytes per OPU session (4.75 vs. 3.56 oocytes) compared with T2. Oocyte viability, expressed as an adjusted posterior probability, was also higher in T1 than in T2 (0.750 vs. 0.566, equivalent to 75.0% vs. 56.6%). In contrast, the adjusted posterior probability of oocyte recovery was nearly identical between treatments (0.403 vs. 0.402, equivalent to 40.3% vs. 40.2%). Among the evaluated outcomes, oocyte viability showed the clearest treatment difference, with an odds ratio of 2.50 and a posterior probability of superiority of 0.989 in favor of T1. Under the conditions of this study, the 0.6 g progesterone protocol was associated with a more favorable follicular response and better oocyte viability, while oocyte recovery efficiency remained essentially unchanged.

Keywords: Cumulus-oocyte complexes; Follicular synchronization; Oocyte competence; Posterior inference; Reproductive hormones.

INTRODUCTION

In vitro embryo production is an important reproductive tool for accelerating genetic progress in cattle (Chinarov, 2024; Dogan and Yenilmez, 2024). Within this process, ultrasound-guided transvaginal follicular aspiration, or ovum pick-up (OPU), plays a central role because it determines the number of oocytes available for later stages such as maturation, fertilization, and embryo development (Sakaguchi, 2025; Unchupaico-Payano et al., 2025). The success of OPU depends not only on how many oocytes are recovered, but also on their biological competence, since both quantity and quality influence the final efficiency of in vitro embryo production (Jemal, 2022).

Follicular wave synchronization and pre-OPU hormonal treatments are commonly used to improve the ovarian response of donor cows by creating a more favorable endocrine environment (Hayden et al., 2025). Previous studies have shown that synchronization may influence not only the number of follicles available for aspiration, but also oocyte recovery and the developmental competence of cumulus-oocyte complexes, depending on the hormonal combination used, including progesterone, estradiol, gonadotropin-releasing hormone (GnRH), follicle-stimulating hormone (FSH), or equine chorionic gonadotropin (eCG) (Payano et al., 2024; Unchupaico-Payano et al., 2025). Current evidence also indicates that both the number and the quality of recovered oocytes remain central to the success of ovum pick-up and in vitro production (OPU-IVP) programs, which explains the continued interest in improving endocrine protocols before aspiration (Salek et al., 2025; Sakaguchi, 2025). Even so, an important question remains unresolved. Although progesterone-based protocols are widely applied in practice, the specific effect of changing the intravaginal progesterone dose on follicular recruitment, oocyte recovery, and oocyte viability is still not well defined. This distinction matters because a protocol may increase follicle numbers or oocyte yield without improving the biological quality of the recovered cumulus-oocyte complexes (COCs), whereas in other cases the main benefit may be reflected in oocyte competence (Lemseffer et al., 2022). For this reason, evaluating only total yield is not sufficient; it is also necessary to examine how treatment affects each biological stage that precedes embryo production.

An additional point concerns the statistical treatment of OPU data. These responses are not simple Gaussian outcomes (Perez et al., 2017), but rather counts and conditional variables measured repeatedly in the same cows (Motta

RESEARCH ARTICLE
Pii: S222877012600025-16
Received: March 30, 2026
Revised: July 15, 2026
Accepted: July 17, 2026

et al., 2024). Despite this, studies in bovine reproduction have often relied on linear methods or conventional comparisons between treatments (Ciftci and Dinc, 2023). A hierarchical Bayesian framework is better suited to this type of data, since it allows each outcome to be analyzed according to its distributional structure, incorporates between-cow heterogeneity, and estimates treatment effects together with their posterior uncertainty (Feldes et al., 2024). For this reason, the present study examined the effect of two intravaginal progesterone doses on follicular recruitment, oocyte recovery, and oocyte viability in Brown Swiss cows undergoing OPU through a hierarchical Bayesian approach.

MATERIALS AND METHODS

Study area

The study was carried out at the cattle unit of the Faculty of Zootechnics, Universidad Nacional del Centro del Perú, located on the left bank of the Mantaro Valley, in the district of El Mantaro, Jauja province, Junín, Peru. The experimental herd consisted of Brown Swiss cows raised under highland conditions at approximately 3,320 m above sea level, about 36.5 km from the city of Huancayo (Senamhi, 2025). Throughout the experimental period, the animals were maintained under an exclusive alfalfa grazing system. Overall, this Andean production setting, together with an alfalfa-based feeding system, represents a suitable context for evaluating reproductive protocols in dairy cattle under the environmental and management conditions of the central Peruvian highlands.

Animals and distribution

The experiment included eight clinically healthy Brown Swiss cows in the dry period from the cattle unit of the Faculty of Zootechnics, Universidad Nacional del Centro del Perú. The animals were distributed into two treatments, with 4 cows per treatment. Treatment 1 corresponded to the bovine intravaginal device (DIB) with 0.6 g of progesterone, while treatment 2 corresponded to the DIB with 1.2 g of progesterone. Five repetitions were performed on each cow, obtaining a total of 20 repetitions per treatment and 40 experimental observations in total. For follicular wave synchronization, on day 0 the corresponding DIB was inserted, and 1.2 mg of estradiol benzoate and 0.524 mg of sodium cloprostenol were administered (Payano et al., 2024). Subsequently, on day 5, oocyte collection was performed via ovum pick-up (OPU) using an EsaoteVet® MyLabOneVet ultrasound scanner, equipped with a 7.5 MHz microconvex transducer, employing a WTA® Brazil follicular aspiration guide and a pressure of 90 mmHg. The evaluated variables were the number of 2 to 4 mm follicles, the number of 4 to 8 mm follicles, the total number of recovered oocytes, and oocyte viability (Unchupaico-Payano et al., 2025).

Data collection

The study variables were: number of 2 to 4 mm follicles, number of 4 to 8 mm follicles, total number of recovered oocytes, and oocyte viability, following the methodology used by Payano et al. (2024). Data collection was performed during each ovum pick-up (OPU) session. Prior to aspiration, through transvaginal ultrasound evaluation, the follicles present in both ovaries were identified and counted, classifying them according to their diameter into two categories: 2–4 mm follicles and 4–8 mm follicles. Subsequently, during follicular aspiration, cumulus-oocyte complexes (COCs) were recovered, which were quantified to obtain the total number of recovered oocytes per repetition. Oocyte viability was determined immediately after recovery, through observation under a stereoscope, considering the morphological characteristics of the cumulus and the cytoplasm. The COCs were classified into quality categories, and for the analysis, they were grouped into viable and non-viable oocytes. COCs with adequate morphology and structure compatible with biological competence were considered viable, while those with deficient morphology or signs of degeneration were classified as non-viable. All records were obtained individually per cow and per repetition, thus generating the database for statistical analysis.

Statistical analysis

The statistical analysis was performed under a Bayesian hierarchical approach, considering the repeated structure of the data, with 5 repetitions per cow and 4 cows per treatment, for a total of 40 experimental observations. The treatment was included as a fixed effect, with two levels: T1 (0.6 g of progesterone) and T2 (1.2 g of progesterone), while the cow was incorporated as a random intercept to capture the biological heterogeneity among animals.

The count variables, corresponding to the number of 2–4 mm follicles, number of 4–8 mm follicles, and total number of recovered oocytes, were modeled using negative binomial distributions due to the discrete nature of the data and the possible presence of overdispersion. For these responses, a logarithmic link function was used, so that, for each observation i in cow j , a model of the form was specified:

$$Y_{ij} \sim \text{NegBin}(\mu_{ij}, \phi), \log(\mu_{ij}) = \beta_0 + \beta_1 \text{Treatment}_{ij} + u_j$$

where β_0 represents the intercept, β_1 the treatment effect and u_j the random effect associated with cow j , with $u_j \sim N(0, \sigma_u^2)$.

The probability of oocyte recovery was analyzed as a binomial response conditional on the total number of observed follicles, whereas oocyte viability was modeled as a binomial response conditional on the total number of recovered oocytes. In both cases, a logit link function was used. Thus, the models were formulated as:

$$R_{ij} \sim \text{Binomial}(F_{ij}, p_{ij}), \text{logit}(p_{ij}) = \alpha_0 + \alpha_1 \text{Treatment}_{ij} + u_j$$

for recovery, where R_{ij} is the number of recovered oocytes and F_{ij} the total number of follicles; and:

$$V_{ij} \sim \text{Binomial}(O_{ij}, q_{ij}), \text{logit}(q_{ij}) = \gamma_0 + \gamma_1 \text{Treatment}_{ij} + u_j$$

for viability, where V_{ij} is the number of viable oocytes and O_{ij} the total number of recovered oocytes.

Inference was performed using Markov chain Monte Carlo (MCMC) sampling implemented in Stan through the brms package in R. Weakly informative prior distributions were used for the fixed coefficients and variance components in order to regularize estimation without imposing strong restrictions on the parameters. Model convergence was assessed through the number of divergent transitions, the $\hat{R}$ statistic, and the effective sample size for the bulk and tails of the posterior distribution. Additionally, trace plots of the chains and the marginal posterior distributions of the treatment effects were graphically inspected.

From the posterior distributions, posterior means, medians, 95% credible intervals, posterior probabilities of superiority $P(T1 > T2)$, rate ratios for the count variables, and odds ratios for the binomial variables were obtained. Likewise, population-level adjusted values for each treatment and posterior absolute contrasts between T1 and T2 were calculated in order to interpret the results both on the relative scale and on the original biological scale. R Studio version 4.5.2 was used.

RESULTS

Data structure

Table 1 summarizes the descriptive results of the variables evaluated in each treatment group. Both T1 and T2 included 20 observations, indicating a balance dataset. In T1 (0.6 g), the means and standard deviations were 4.75 ± 2.34 for recovered oocytes, 3.60 ± 2.30 for viable oocytes, 1.15 ± 0.88 for non-viable oocytes, 7.60 ± 2.76 for 2–4 mm follicles, 4.15 ± 2.08 for 4–8 mm follicles, and 11.75 ± 2.67 for total follicles. In T2 (1.2 g), the corresponding values were 3.55 ± 2.33 , 2.0 ± 1.65 , 1.55 ± 1.05 , 5.55 ± 3.39 , 3.30 ± 2.08 , and 8.85 ± 3.33 , respectively. For the variables derived, recovery was very similar in both treatments, reaching 0.410 in T1 and 0.417 in T2. Oocyte viability, however, was higher in T1 (0.72) than in T2 (0.48). These descriptive values offer a preliminary overview of the response pattern in each before the statistical analysis.

Table 1 - Structure of the analyzed dataset by treatment and cow

Variable	T1 (0.6 g)	T2 (1.2 g)	p-value
Number of observations	20	20	—
Recovered oocytes, n/session	4.75 ± 2.34	3.55 ± 2.33	0.112
Viable oocytes, n/session	3.60 ± 2.30^a	2.00 ± 1.65^b	0.016
Non-viable oocytes, n/session	1.15 ± 0.88	1.55 ± 1.05	0.200
2–4 mm follicles, n/session	7.60 ± 2.76^a	5.55 ± 3.39^b	0.043
4–8 mm follicles, n/session	4.15 ± 2.08	3.30 ± 2.08	0.204
Follicles count	11.75 ± 2.67^a	8.85 ± 3.33^b	0.004
Oocyte recovery, %	41.0	41.7	0.949
Oocyte viability, %	75.8 ^a	56.3 ^b	0.008

Values are presented as mean $\pm$ standard deviation for count variables and as percentages for oocyte recovery and viability. For count variables, p-values were calculated using Welch's two-sample t-test. For oocyte recovery and viability, p-values were calculated using a two-proportion z-test based on aggregated counts. Different superscript letters within the same row indicate statistically significant differences between treatments ($p < 0.05$). When treatments share the same letter, no statistically significant difference was detected. OPU: ovum pick-up.

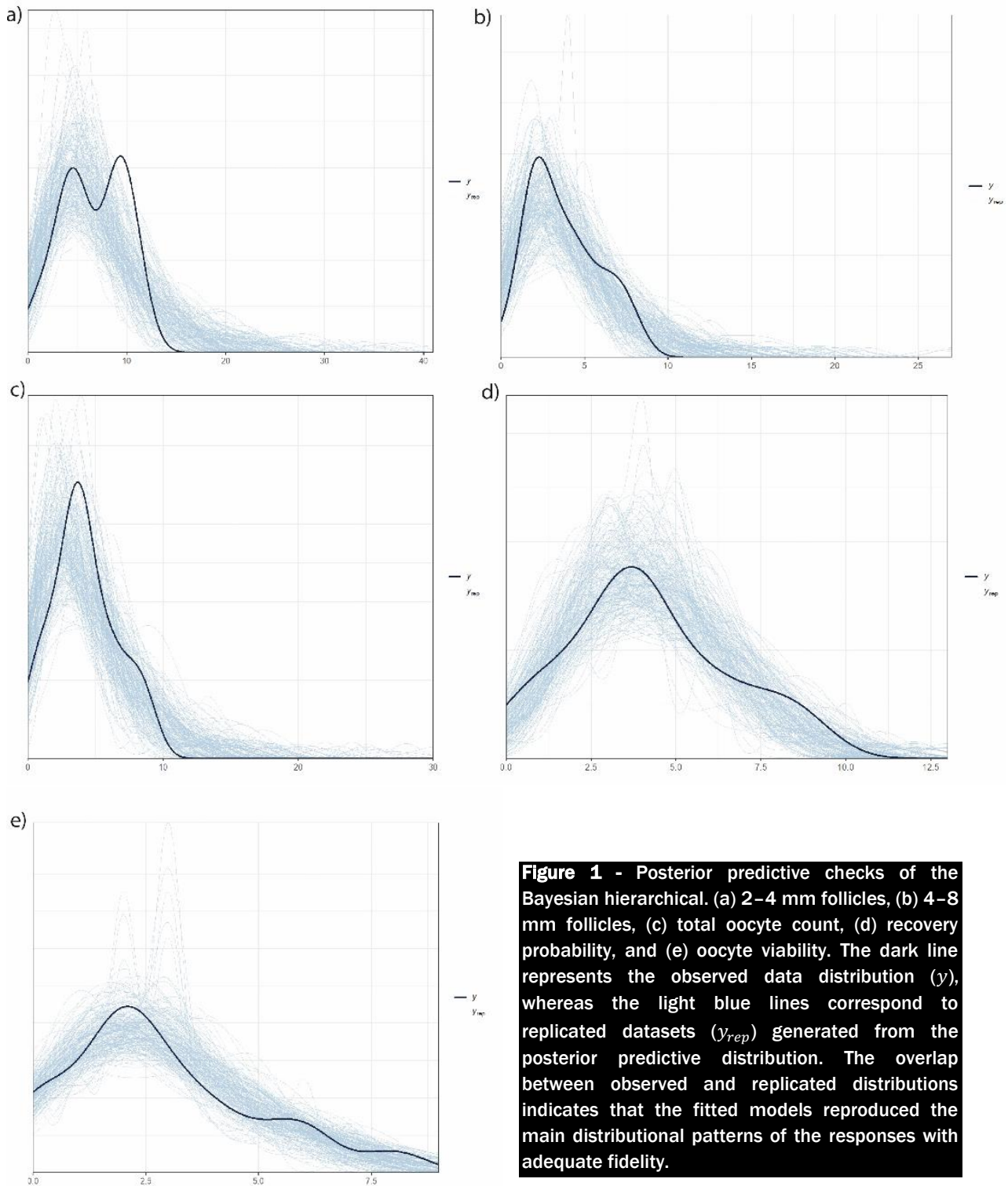
Convergence diagnostics

Table 2 presents the convergence diagnostics for the hierarchical Bayesian models. No divergent transitions were detected in any of the fitted models, which suggests stable posterior sampling. In all cases, the maximum R value was 1.00, indicating good convergence and adequate mixing of the chains. The minimum effective sample sizes were also satisfactory, ranging from 2024 to 3648 for the bulk of posterior distribution and from 2711 to 3937 for the tails. Overall, these results support the computational stability of the models and the reliability of the posterior estimates for treatment effects. Posterior predictive checks further showed that the fitted models were able to reproduce the general shape of the observed distributions for the five responses analyzed (Figure 1).

Table 2 - Convergence diagnostics of the Bayesian hierarchical models fitted for each response variable.

Response variable	n_divergent	Max rhat	Min bulk ESS	Min tail ESS
2–4 mm follicles	0	1.00	2185	2711
4–8 mm follicles	0	1.00	3648	3937
Oocyte recovery	0	1.00	2024	2805
Oocyte viability	0	1.00	3418	3844
Ovocitos count	0	1.00	3514	3739

Divergent transitions indicate the number of problematic MCMC transitions. max Rhat represents the maximum potential scale reduction factor; values close to 1.00 indicate adequate convergence. min bulk ESS and min tail ESS indicate the minimum effective sample sizes for the central and tail regions of the posterior distribution, respectively.



Posterior treatment effects

Table 3 shows the posterior effect of T1 (0.6 g) relative to T2 (1.2 g) for each response variable. On the model's linear scale, the posterior mean was positive for 2-4 mm follicles (0.353), 4-8 mm follicles (0.244), oocyte viability (0.845), and total oocyte count (0.290), whereas the effect on recovery was essentially zero (0.000692). The posterior probability that the effect of T1 was greater than zero was 0.931 for 2-4 mm follicles, 0.842 for 4-8 mm follicles, 0.989 for oocyte viability, and 0.878 for total oocyte count.

By contrast, the corresponding value for recovery probability was 0.502, which suggests no clear directional difference between treatments. For ease of interpretation, count outcomes were expressed as rate ratios and binomial outcomes as odds ratios. For 2-4 mm follicles, the mean posterior rate ratio was 1.47, with a 95% credible interval between 0.874 and 2.36; for 4-8 mm follicles, the rate ratio was 1.32 (0.782-2.07); and for total oocytes, the rate ratio was 1.38 (0.797-2.21).

In the case of recovery, the mean posterior odds ratio was 1.03 (0.595-1.67), reflecting a response centered very close to the null value. In contrast, viability showed the most pronounced relative difference between treatments, with a mean posterior odds ratio of 2.50 and a 95% credible interval from 1.10 to 4.92, providing the clearest posterior evidence of a treatment effect among all evaluated responses.

Figure 2 presents the population-posterior adjusted means and 95 % credible intervals for each treatment on the original scale of the response variable. T1 (0.6 g) showed higher adjusted means than T2 (1.2 g) for 2-4 mm follicles, 4-8 mm follicles, total recovered oocytes, and oocyte viability. By contrast, recovery probability was virtually the same in both treatments. These results indicate that the main differences between protocols were concentrated in follicular response, total oocyte yield, and especially viability, while recovery remained stable.

Figure 3 presents the posterior absolute contrasts between treatments, expressed as T1 (0.6 g) minus T2 (1.2 g), on the original scale of each variable. Accordingly, the results can be interpreted not only by the direction of the difference, but also by its practical magnitude in counts or percentage points. In the count variables, the contrasts were located at positive values for 2-4 mm follicles, 4-8 mm follicles, and total oocytes, indicating that the adjusted posterior means of these responses tended to be higher in T1 than in T2.

Among them, the greatest absolute shift was observed in 2-4 mm follicles, followed by the total number of oocytes and, to a lesser extent, by 4-8 mm follicles. In the variables expressed as probability, the contrast for recovery remained very close to zero, reflecting an almost null absolute difference between treatments. In contrast, oocyte viability showed the greatest shift on the probability scale, with a clearly wider positive contrast than the rest of the variables, indicating a substantial difference in percentage points in favor of T1.

Joint posterior patterns among biological responses

Figure 4 summarizes the joint structure of the posterior distributions of the treatment effects on pairs of key biological responses, expressed on the transformed scale as rate ratios or odds ratios of T1 (0.6 g) relative to T2 (1.2 g). In all panels, the dotted lines at 1 delimit the null value, so that the regions of greatest density located to the right and/or above that point indicate posterior shifts compatible with a greater relative response under T1. This representation makes it possible to evaluate simultaneously not only the direction of the treatment effect on each variable, but also the way in which these responses tend to shift jointly within the posterior space.

In panel a), corresponding to the joint density between the rate ratio for 2-4 mm follicles and the rate ratio for total oocytes, the greatest posterior concentration is located in the upper-right quadrant, with both axes above the null value. This pattern suggests that the highest posterior density was concentrated in the region where T1 was associated with both a greater availability of small follicles and a higher total oocyte yield.

In panel (b), which compares the odds ratios for recovery and viability, a different pattern emerged: posterior density remained centered near the null value for recovery but was clearly shifted above 1 for viability. In biological terms, this reflects that the joint posterior signal is concentrated on a practically stable recovery between treatments, accompanied by relatively greater oocyte viability under T1.

Panel c), corresponding to the joint density between the rate ratio for 4-8 mm follicles and the rate ratio for total oocytes, also presents its main posterior core in the upper-right region, although with a somewhat wider dispersion than that observed for 2-4 mm follicles. This suggests that the joint shift between medium follicles and oocyte yield was positive, but with greater relative uncertainty than that observed for the early follicular response.

Finally, in panel d), which relates the rate ratio for 2-4 mm follicles to the odds ratio of viability, the posterior mass is again concentrated above the null value on both axes, showing that the shift toward a greater presence of small follicles tends to coexist with a favorable shift in the viability of the recovered oocytes. Taken together, the figure reinforces that the most consistent posterior treatment patterns were located in the combination of early follicular recruitment, oocyte production, and especially oocyte viability, whereas recovery remained comparatively more stable in the joint response space.

Table 3 - Posterior treatment effects of T1 (0.6 g progesterone) relative to T2 (1.2 g progesterone).

Response variable	Posterior effect, mean $\pm$ SD	95% CrI	Pr(T1 > T2)	Transformed effect	95% CrI	Interpretation
2–4 mm follicles	0.353 $\pm$ 0.247	-0.134 to 0.857	0.931	Rate ratio = 1.47	0.874 to 2.36	Tendency in favor of T1
4–8 mm follicles	0.244 $\pm$ 0.250	-0.245 to 0.726	0.842	Rate ratio = 1.32	0.782 to 2.07	Tendency in favor of T1
Oocyte recovery probability	0.00692 $\pm$ 0.258	-0.519 to 0.511	0.502	Odds ratio = 1.03	0.595 to 1.67	No relevant difference
Oocyte viability	0.845 $\pm$ 0.380	0.0985 to 1.59	0.989	Odds ratio = 2.50	1.10 to 4.92	Clear evidence in favor of T1
Total recovered oocytes	0.290 $\pm$ 0.259	-0.227 to 0.792	0.878	Rate ratio = 1.38	0.797 to 2.21	Tendency in favor of T1

SD: posterior standard deviation; CrI: credible interval; Pr(T1 > T2): posterior probability that the treatment effect favors T1 over T2. Rate ratios are reported for count outcomes, whereas odds ratios are reported for binomial outcomes. For transformed effects, the null value is 1.00. In this Bayesian framework, posterior probabilities and 95% credible intervals provide the inferential evidence for treatment comparison.

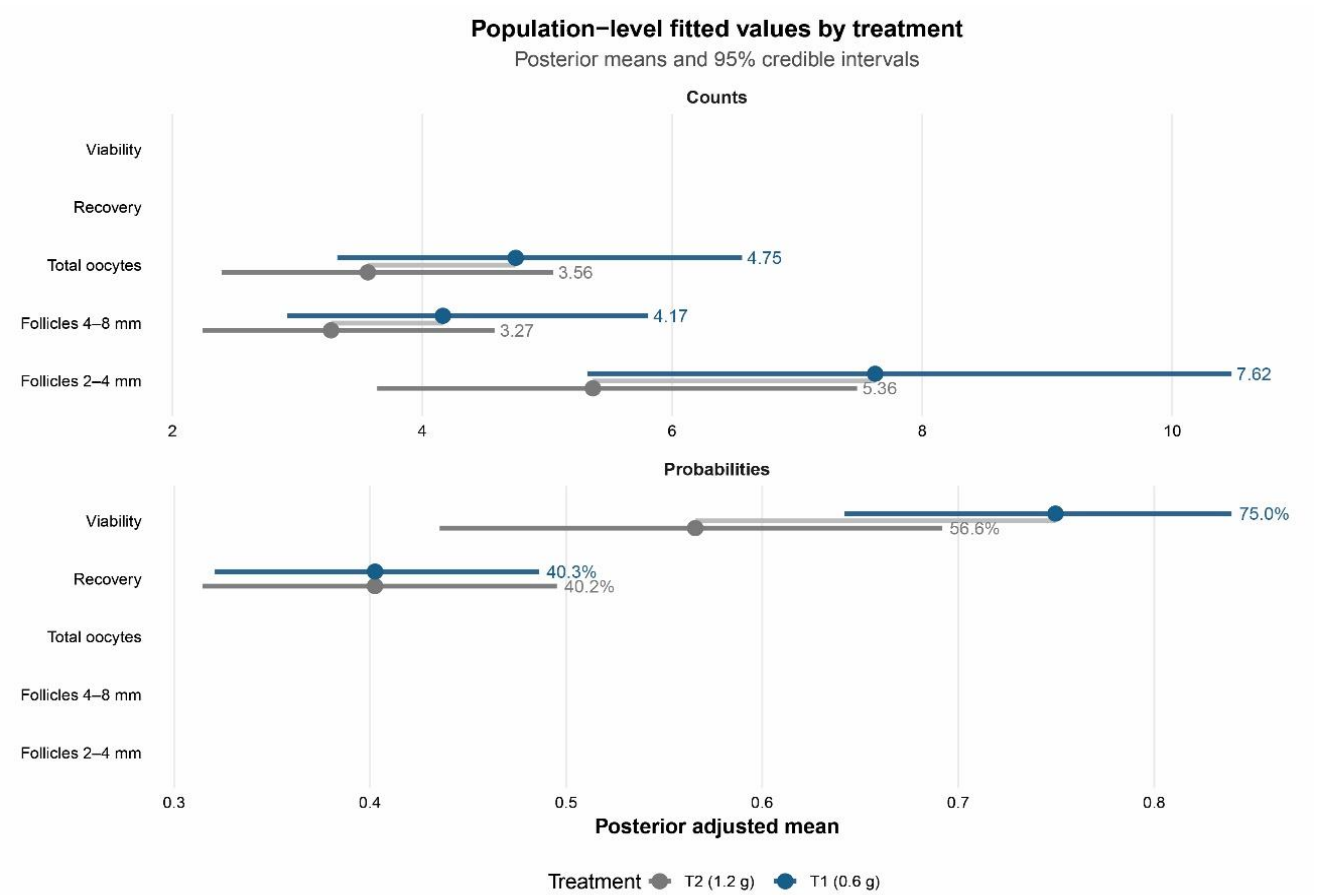


Figure 2 - Population-level fitted values by treatment.

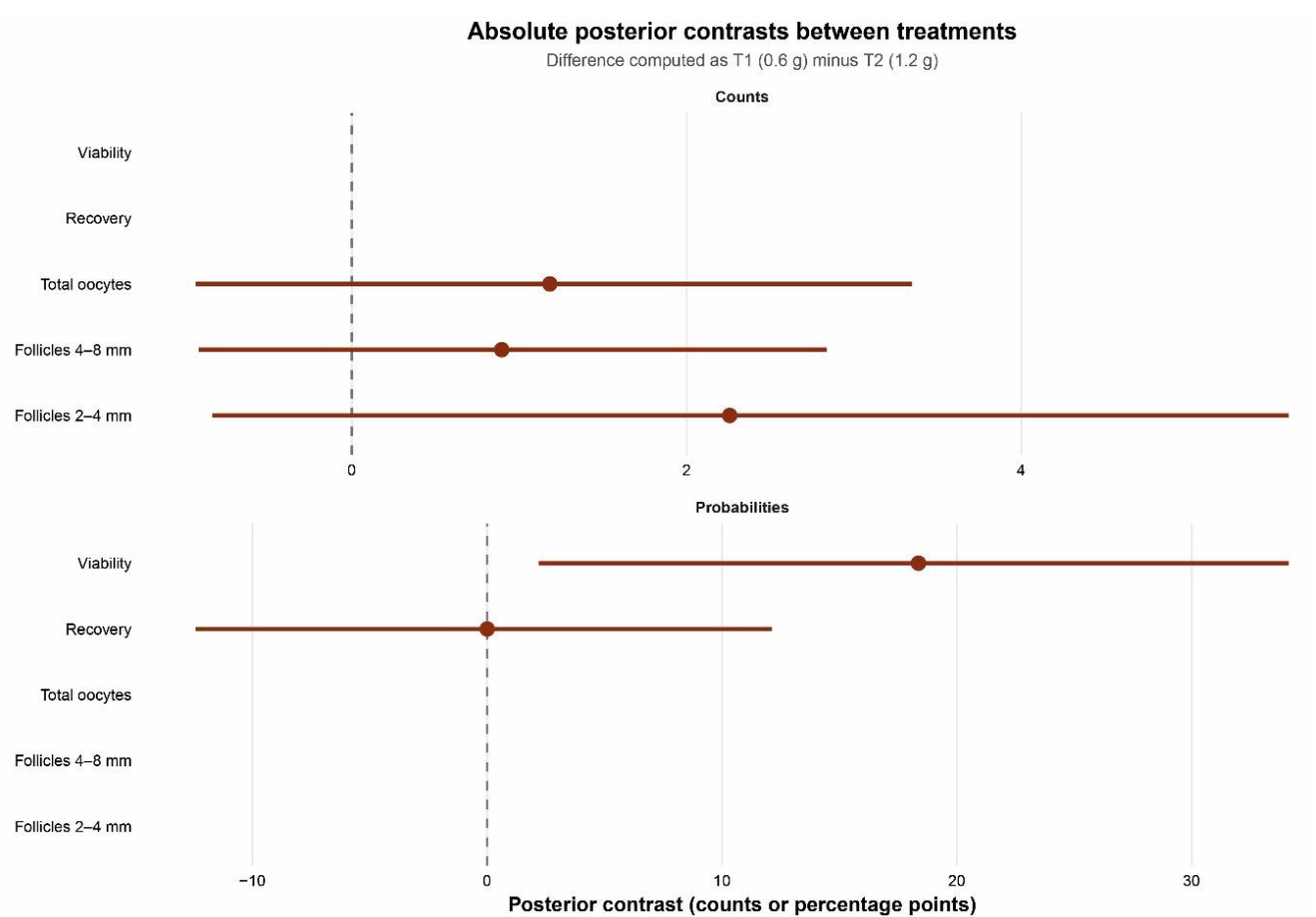


Figure 3 - Absolute posterior contrasts between treatments.

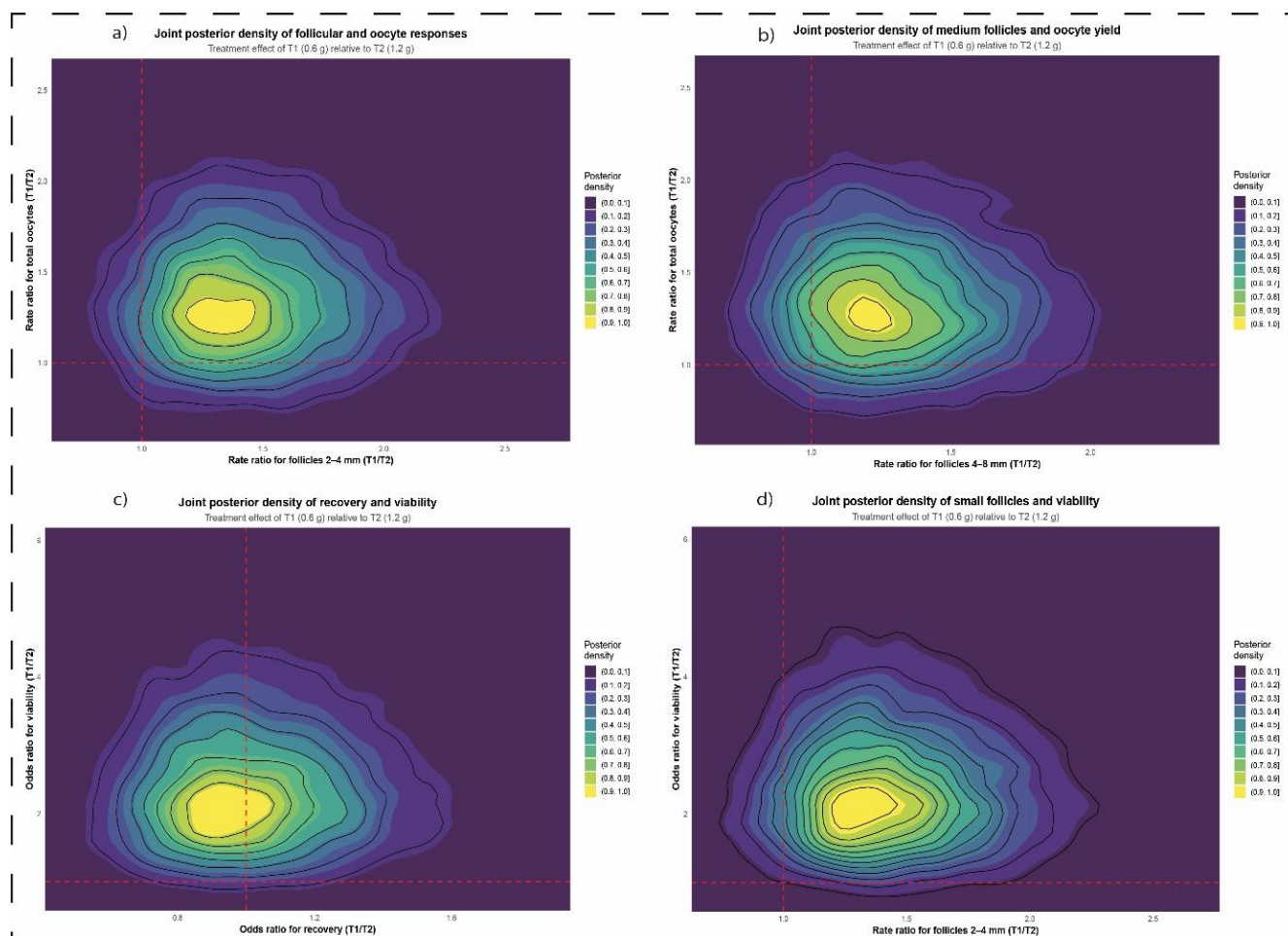


Figure 4 - Joint posterior patterns among follicular, oocyte, recovery, and viability responses under two intravaginal progesterone doses in Brown Swiss cows. (a) Joint posterior density between the rate ratio for 2–4 mm follicles and the rate ratio for total oocytes. (b) Joint posterior density between the rate ratio for 4–8 mm follicles and the rate ratio for total oocytes. (c) Joint posterior density between the odds ratio for recovery and the odds ratio for viability. (d) Joint posterior density between the rate ratio for 2–4 mm follicles and the odds ratio for viability.

DISCUSSION

The main finding of this study was that the protocol with 0.6 g of progesterone shifted the biological response toward higher adjusted values of 2–4 mm follicles, 4–8 mm follicles, and total oocytes, whereas the most marked signal was observed in oocyte viability; in contrast, the probability of recovery remained practically unchanged between treatments. This same structure was repeated in the absolute contrasts and in the joint posterior density maps, where the greatest separation between treatments was concentrated in the follicular response and, above all, in viability, rather than in recovery per follicle.

This pattern is biologically consistent with the literature, as pointed out by [Motta et al. \(2024\)](#), indicating that synchronization of the follicular wave before OPU modifies the distribution of follicular sizes and may improve oocyte competence, although the exact response depends on the protocol and the type of outcome evaluated. In Angus cows subjected to a protocol with an intravaginal progesterone device, estradiol benzoate, and cloprostenol, synchronization increased the number of medium follicles, but did not modify the total number of follicles or the number of recovered COCs, and even reduced the recovery rate ([Anchordoquy et al., 2024](#)). Complementarily, a recent study in Braford cows explicitly proposed that follicular synchronization prior to OPU improves oocyte competence and that the response of cows with a greater taurine component still requires further elucidation ([Unchupaico-Payano et al., 2025](#)). Taken as a whole, these studies suggest that endocrine treatments before OPU do not lead to a uniform improvement in every indicator; rather, their effect may be expressed differently across follicular size, recovery, and oocyte quality.

The results suggest that the lower progesterone dose may have favored an ovarian environment more supportive of early follicular recruitment, especially in the 2–4 mm category, and to a lesser extent, total oocyte yield. This interpretation is consistent with the known role of progesterone in regulating LH pulse Frequency and, together with estradiol, modulating gonadotropin secretion and follicular wave dynamics ([e Silva et al., 2025](#); [Garcia et al., 2025](#)). Previous evidence indicates that lower progesterone concentrations during follicular growth may be associated with increased LH

pulsatility and higher estradiol concentrations, while the combined action of estradiol and progesterone suppresses FSH and LH, induces atresia of the dominant follicle, and synchronizes the emergence of a new wave (Gomez-León et al., 2020). In this sense, a lower progesterone load may have reduced gonadotropic suppression while still maintaining protocol control, which could explain the greater availability of small aspiratable follicles (Hugues and Durnerin, 1998). Still, because progesterone, LH, and estradiol were not directly measured, this mechanism should be viewed as a biologically plausible interpretation rather than a demonstrated effect.

The clearest finding of this study was the difference observed in oocyte viability. Compared with the other responses, viability showed the strongest separation between treatments on both the absolute and relative scales, and this pattern was also evident in its joint posterior distribution with follicular response. This suggests that the main advantage of the 0.6 g protocol was not a higher mechanical efficiency of aspiration, but an improvement in the biological quality of the recovered oocytes. This interpretation is in line with previous evidence showing that synchronization before OPU may enhance oocyte competence (Garcia et al., 2025), and with studies reporting that exogenous progesterone can improve oocyte quality even when later in vitro outcomes do not change substantially (Payano et al., 2024). In Sahiwal cows, for instance, progesterone treatment increased follicular population, recovery, and the proportion of better-quality oocytes, although it had no effect on nuclear maturation, cleavage, or blastocyst formation (Nawaz et al., 2022). In the same direction, recent work has increasingly viewed synchronization as a strategy to improve competence, not only quantity (Lopes et al., 2019). In contrast, recovery probability remained largely unchanged between treatments. This finding is relevant because it suggests that progesterone dose did not materially affect the proportion of oocytes recovered per aspirated follicle, even though differences were observed in other responses. From both a biological and an operational perspective, this pattern is not unexpected, since oocyte recovery depends not only on the endocrine environment but also on technical and physical factors such as follicle accessibility, aspiration pressure, flow rate, operator handling, and, in some cases, follicular size (Rose, 2014; Anchordoquy et al., 2024). In fact, the literature shows inconsistent findings regarding oocyte recovery when protocols preceding OPU are modified: some studies have reported that synchronization reduced the recovery rate, whereas others have found that recovery may vary more with follicular size or aspiration technique than with the hormonal protocol itself (Seneda et al., 2001; Márquez-Moya et al., 2025). Consequently, the present pattern, stability in recovery together with improvement in viability, reinforces the idea that the main effect of the treatment was more related to oocyte competence than to extraction efficiency.

Within the conditions evaluated in this study, reducing the progesterone load in the intravaginal device from 1.2 g to 0.6 g did not reduce oocyte recovery efficiency. Instead, the lower-dose protocol was associated with a favorable posterior shift toward a greater number of aspiratable follicles, higher total oocyte yield, and improved oocyte viability. From an applied perspective, these findings suggest that the optimization of pre-OPU protocols should not be based only on the total number of recovered structures, but also on how the treatment influences follicular recruitment, oocyte yield, and biological competence (Sarwar et al., 2020). Therefore, the main contribution of the present study is not merely the comparison of two progesterone doses, but the identification of a potential biological advantage of the lower-dose protocol, mainly reflected in the functional quality of the recovered oocytes.

CONCLUSION

Treatment T1 (0.6 g of progesterone) showed a more favorable response than T2 (1.2 g) in follicular recruitment, total number of oocytes, and especially in oocyte viability, the latter being the variable in which the most consistent posterior evidence of biological superiority was observed. No relevant differences were observed between treatments in the probability of oocyte recovery, indicating that both protocols showed similar behavior in the efficiency of obtaining oocytes per aspirated follicle. The hierarchical Bayesian approach made it possible to adequately model the repeated structure of the data, the heterogeneity among cows, and the specific nature of each response, providing more robust and biologically interpretable inference on the effects of the two progesterone doses in Brown Swiss cows subjected to ovum pick-up (OPU).

DECLARATIONS

Corresponding author

Correspondence and requests for materials should be addressed to Jordan Ninahuanca. E-mail: jninahuanca@uncp.edu.pe, ORCID: <https://orcid.org/0000-0002-0137-0631>

Data availability

The datasets generated and/or analyzed during the current study are available from the corresponding author on reasonable request.

Authors' contribution

Carlos Quispe-Eulogio contributed to the study methodology. Neale Santiago-Inga was responsible for data collection. Edith Ancco-Gomez performed the laboratory procedures. Jordan Ninahuanca contributed to manuscript writing and statistical analysis. Ide Unchupaico-Payano and Milagros García-Hospinal contributed to conceptualization and methodological development. Cecil Rivera-Palomino was responsible for the critical revision of the manuscript. All authors reviewed and approved the final version of the manuscript.

Consent to publish

The publication of this manuscript has been approved by all authors.

Acknowledgement

The authors express their gratitude to the staff of the cattle unit of the Faculty of Zootechnics and to all personnel who supported animal handling, field activities, and laboratory procedures during the development of this study.

Funding sources

This study was funded by the Universidad Peruana Los Andes.

Competing Interests

There is no competing interest among the authors regarding the publication of this article.

Ethical approval

The experimental procedures involving animals were conducted in accordance with animal welfare and ethical handling standards applicable to bovine reproductive research. All procedures related to animal handling, hormonal treatment, and ovum pick-up were conducted under veterinary supervision, with special attention to minimizing stress and ensuring proper care of the donor cows throughout the study. The protocol was reviewed and approved by the competent institutional authority under letter N. 006-GRJ-DRAA-AAC-PERU-2025.

REFERENCES

- Anchordoquy JP, Balbi M, Farnetano NA, Giuliodori MJ, Nikoloff N, Carranza-Martín AC, et al. (2024). Effect of follicular wave synchronization using a progesterone-releasing intravaginal device-based protocol on in vitro embryo production in *Bos taurus* cows subjected to 14-day intervals ovum pick-up. *Animal Reproduction Science*, 261: 107397. <https://doi.org/10.1016/j.anireprosci.2023.107397>
- Chinarov RY (2024). Developing the ovum pick-up technology in cattle: State-of-the-art and research directions. *Sel'skokhozyaistvennaya Biologiya* 59: 194-220. <https://doi.org/10.15389/agrobiology.2024.2.194eng>
- Ciftci MF and Dinc DA (2023). The effect of different FSH administration before ovum pick up on superstimulation response and oocyte yield. *Reproduction in Domestic Animals*, 58: 1055-1062. <https://doi.org/10.1111/rda.14394>
- Dogan H and Yenilmez K (2024). Factors influencing ovum pick-up technique results in cattle. *Journal of Istanbul Veterinary Sciences* 8: 13-23. <https://doi.org/10.30704/http-www-ijvs-net.1403722>
- e Silva LO, Cavalcanti PR, Alves RL, Folchini NP, Teixeira NN, Monteiro PJ, et al. (2025). Effect of the follicular stage and circulating progesterone concentrations at the beginning of an estradiol/progesterone-based synchronization protocol on profile of gonadotropins and follicle dynamics in beef heifers. *Theriogenology* 241: 117421. <https://doi.org/10.1016/j.theriogenology.2025.117421>
- Feltes GL, Campos GS, Raidan FSS, Feres LFR, Ribeiro VMP and Cobuci JA (2024). Comparing Bayesian models for the genetic evaluation of oocytes and embryo counts in Dairy Gir cattle. *Journal of Applied Genetics* 65: 591-600. <https://doi.org/10.1007/s13353-024-00862-3>
- Garcia FG, Menezes BMD, Possebon CF, Klaus R, Alborno MS, Trentin JM, et al. (2025). Effect of prior follicular wave synchronization and eCG on ovum pick-up and in vitro embryo production in Braford cows. *Animal Reproduction* 22: e20250020. <https://doi.org/10.1590/1984-3143-ar2025-0020>
- Gomez-León VE, Ginther OJ, Guimarães JD and Wiltbank MC (2020). Hormonal mechanisms regulating follicular wave dynamics II: Progesterone decreases diameter at follicle selection regardless of whether circulating FSH or LH are decreased or elevated. *Theriogenology* 143: 148-156. <https://doi.org/10.1016/j.theriogenology.2019.11.003>
- Hayden CB, Motta JC, Sala RV, Bello NM, da Silva MAC and García-Guerra A (2025). Efficacy of methods to synchronize follicular wave emergence in pregnant heifers. *JDS Communications* 6: 154-159. <https://doi.org/10.3168/jdsc.2024-0629>
- Hugues JN and Durnerin IC (1998). Revisiting gonadotrophin-releasing hormone agonist protocols and management of poor ovarian responses to gonadotrophins. *Human Reproduction Update*, 4: 83-101. <https://doi.org/10.1093/humupd/4.1.83>
- Jemal J (2022). Ovum pick up and in vitro embryo production in Boran and crossbred dairy cattle. *Ethiopian Journal of Agricultural Sciences* 32: 88-104.

- Lemseffer Y, Terret ME, Campillo C and Labrune E (2022). Methods for assessing oocyte quality: A review of literature. *Biomedicines*, 10: 2184. <https://doi.org/10.3390/biomedicines10092184>
- Lopes JS, Canha-Gouveia A, París-Oller E and Coy P (2019). Supplementation of bovine follicular fluid during in vitro maturation increases oocyte cumulus expansion, blastocyst developmental kinetics, and blastocyst cell number. *Theriogenology*, 126: 222-229. <https://doi.org/10.1016/j.theriogenology.2018.12.010>
- Márquez-Moya A, Sala-Ayala L, Carreras-Vico N, Martínez-Boví R and Cuervo-Arango J (2025). Factors influencing oocyte recovery during ultrasound-guided follicle aspiration in mares: A postmortem study. *Theriogenology*, 235: 39-45. <https://doi.org/10.1016/j.theriogenology.2024.12.032>
- Motta JC, Hayden CB, Sala RV, Ross PJ and García-Guerra A (2024). Advances in synchronization and superstimulation for OPU/IVF: Optimizing oocyte quantity and quality. *Reproduction, Fertility and Development* 37: RD24143. <https://doi.org/10.1071/RD24143>
- Nawaz M, Saleem M, Ullah F, Shabbir Khan G, Zahoor I, Ahmad N, et al. (2022). Exogenous progesterone-dependent modulation in the follicular dynamics of *Bos indicus* cattle undergoing repeated ovum pick-up sessions. *Reproduction in Domestic Animals*, 57: 55-63. <https://doi.org/10.1111/rda.14028>
- Payano IU, Eulogio CQ, Gómez EA, Carhuas JN, Villar FA, Fermín JU and Sánchez NM (2024). Effects of two types of estrogen on the follicular wave for in vivo oocyte collection in Brown Swiss cows. *World's Veterinary Journal* 14: 366-372. <https://doi.org/10.54203/scil.2024.wvj43>
- Perez BC, Silva FF, Ventura RV, Bruneli FT, Balieiro JCDC and Peixoto MGDC (2017). Count Bayesian models for genetic analysis of in vitro embryo production traits in Guzará cattle. *Animal*, 11: 1440-1448. <https://doi.org/10.1017/S175173111700012X>
- Rose BI (2014). Approaches to oocyte retrieval for advanced reproductive technology cycles planning to utilize in vitro maturation: A review of the many choices to be made. *Journal of Assisted Reproduction and Genetics*, 31: 1409-1419. <https://doi.org/10.1007/s10815-014-0334-9>
- Sakaguchi K (2025). Optimization of ovum pick-up-in vitro fertilization and in vitro growth of immature oocytes in ruminants. *Journal of Reproduction and Development*, 71: 1-9. <https://doi.org/10.1262/jrd.2024-091>
- Salek F, Guest A, Johnson C, Kastelic JP and Thundathil J (2025). Factors affecting the success of ovum pick-up, in vitro production and cryopreservation of embryos in cattle. *Animals*, 15: 344. <https://doi.org/10.3390/ani15030344>
- Sarwar Z, Saad M, Saleem M, Husnain A, Riaz A and Ahmad N (2020). Effect of follicle size on oocytes recovery rate, quality, and in-vitro developmental competence in *Bos indicus* cows. *Animal Reproduction*, 17: e20200011. <https://doi.org/10.1590/1984-3143-ar2020-0011>
- Senamhi (2025). National hydrometeorological data. Servicio Nacional de Meteorología e Hidrología del Perú. Available at: <https://www.senamhi.gob.pe/?p=pronostico-meteorologico> (Accessed 10 July 2025).
- Seneda MM, Esper CR, Garcia JM, de Oliveira JA and Vantini R (2001). Relationship between follicle size and ultrasound-guided transvaginal oocyte recovery. *Animal Reproduction Science*, 67: 37-43. [https://doi.org/10.1016/S0378-4320\(01\)00113-0](https://doi.org/10.1016/S0378-4320(01)00113-0)
- Unchupaico-Payano I, Arauco-Villar F, Ninahuanca-Carhuas J, Quispe-Eulogio C and Ancoco-Gomez E (2025). rbST effects on in vitro embryo production in Andean cattle. *Acta Fytotechnica et Zootechnica* 28: 317-323. <https://doi.org/10.15414/afz.2025.28.04.317-323>

Publisher's note: Scienceline Publication Ltd. remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.



Open Access: This article is licensed under a Creative Commons Attribution 4.0 International License, which permits use, sharing, adaptation, distribution and reproduction in any medium or format, as long as you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons licence, and indicate if changes were made. The images or other third party material in this article are included in the article's Creative Commons licence, unless indicated otherwise in a credit line to the material. If material is not included in the article's Creative Commons licence and your intended use is not permitted by statutory regulation or exceeds the permitted use, you will need to obtain permission directly from the copyright holder. To view a copy of this licence, visit <https://creativecommons.org/licenses/by/4.0/>.

© The Author(s) 2026