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Abstract

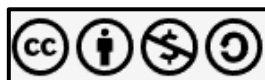
This study focused on evaluating the motility rate of semen from Large White and Duroc breeds using organic semen extenders—specifically coconut water and honey—compared to a commercial extender used as the control. It explored the composition of these organic extenders, the morphology of boar semen based on normal fraction, and motility rate, which was assessed using a mobile computer-assisted sperm analyzer (mCASA). This study employed a Multiple Time Series Design (MTSD), wherein evaluations were both performed before processing, with the sperm temperature at 35 °C, and after processing. The samples were monitored at 18 °C from 8 a.m. to 8 p.m. at 2-hour intervals. Monitoring occurred after 2, 4, 6, 8, 10, and 12 hours across all treatments, including the control. Results showed that honey-based Treatment 4 best preserved motility in Large White boar semen for up to 12 hours, outperforming all other treatments, including the commercial extender. In contrast, coconut water-based Treatment 1 maintained high motility in Duroc semen for up to 48 hours, exceeding both commercial and organic alternatives. These findings suggest breed-specific responses to extenders and highlight the potential of Treatments 4 and 1 as effective, organic option. Future studies should consider genotype-specific traits to optimize semen preservation.

Keywords: artificial insemination, boar semen, coconut water, Duroc, honey, large white, mCASA, organic semen extender, sperm motility, time-series design

Introduction

Artificial insemination (AI) is a strategically important tool for improving swine genetics, enhancing reproductive efficiency, disease control, and genetic progress (Waberski et al., 2019). However, AI success depends on semen viability and quality during storage (Colenbrander et al., 2020). While semen extenders are essential for sperm survival, synthetic options present cost challenges (Odrada et al., 2023). This has driven interest in natural, locally available organic alternatives (Machebe et al., 2015; Luther et al., 2023), which offer affordability, accessibility, and beneficial antioxidant and antimicrobial properties that reduce sperm damage, extend shelf life, and maintain motility for improved semen quality in swine production.

Artificial insemination improves genetic diversity, productivity, and breed preservation by overcoming natural mating limitations, but its effectiveness depends on semen quality (Pardede et al., 2021). Proper handling and thawing are then essential to maintain this quality (Diwan, 2021). Semen extenders, whether liquid or powder, support sperm survival by supplying nutrients, maintaining ideal conditions, and preventing early motility (Chapman, 2016). Coconut water, which is rich in sugars, nutrients, and antioxidants, shows potential as a natural semen extender by supporting sperm motility and



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protecting against oxidative stress (Wynn, 2017; Banday et al., 2017). Similarly, honey enhances motility and reduces abnormalities in freeze-thawed semen due to its sugar and antioxidant content, which helps minimize ice crystal formation (Zaghloul, 2017). These properties may influence the response of Large White and Duroc boars to such extenders.

Coconut water and honey, due to their natural composition and local availability, are promising organic alternatives to synthetic semen extenders in the Philippines. Their suitability for small, resource-limited swine farms in tropical areas makes them ideal for sustainable and cost-effective use—coconut water supports sperm viability, while honey enhances preservation through its osmotic and antibacterial properties.

This study explores cost-effective and eco-friendly semen preservation methods suitable for developing countries like the Philippines, where synthetic extenders are expensive. By using locally available coconut water and honey, it supports sustainable and organic farming. The research addresses a gap in breed-specific studies on organic extenders, focusing on their components and effects on semen motility over 48 hours, contributing to swine artificial insemination practices.

Objectives of the Study

This study aimed to determine the motility of Large White and Duroc semen using various formulations of coconut water and honey as organic semen extenders. Specifically, it aimed to:

1. Analyze the biochemical composition of coconut water and honey used as semen extenders, including:
 - 1.1. Moisture;
 - 1.2. Bicarbonate;
 - 1.3. Carbonate;
 - 1.4. Phosphate;
 - 1.5. Total Sugar;
 - 1.6. Crude Protein;
 - 1.7. Calcium;
 - 1.8. Salt (NaCl);
 - 1.9. Sodium; and
 - 1.10. Potassium
2. Evaluate baseline semen characteristics of Large White and Duroc boars, including:
 - 2.1. Ejaculate volume;
 - 2.2. Morphology based on normal fraction;
 - 2.3. Motility based on
 - 2.3.1. Static;
 - 2.3.2. Progressive;
 - 2.3.3. Motile;
 - 2.3.4. Slow; and
 - 2.3.5. Total counts
3. Evaluate the motility rate of semen of Large White and Duroc before and immediately after the addition of organic extenders, and
4. Compare the motility rate of processed semen of Large White and Duroc using organic extender and commercially available semen extender across multiple post-processing time points, including:
 - 4.1. After processing
 - 4.2. Two hours after processing (10 a.m.)
 - 4.3. Four hours after processing (12 noon)
 - 4.4. Six hours after processing (2 p.m.)

- 4.5. Eight hours after processing (4 p.m.)
- 4.6. Ten hours after processing (6 p.m.)
- 4.7. Twelve hours after processing (8 p.m.)

Hypothesis

This study tested the hypothesis that there is no significant difference in the motility rate of processed semen of Large White and Duroc boars using organic extenders (coconut water and honey) and a commercial extender. The semen was evaluated at different time intervals after processing, and after two, four, six, eight, ten, and twelve hours.

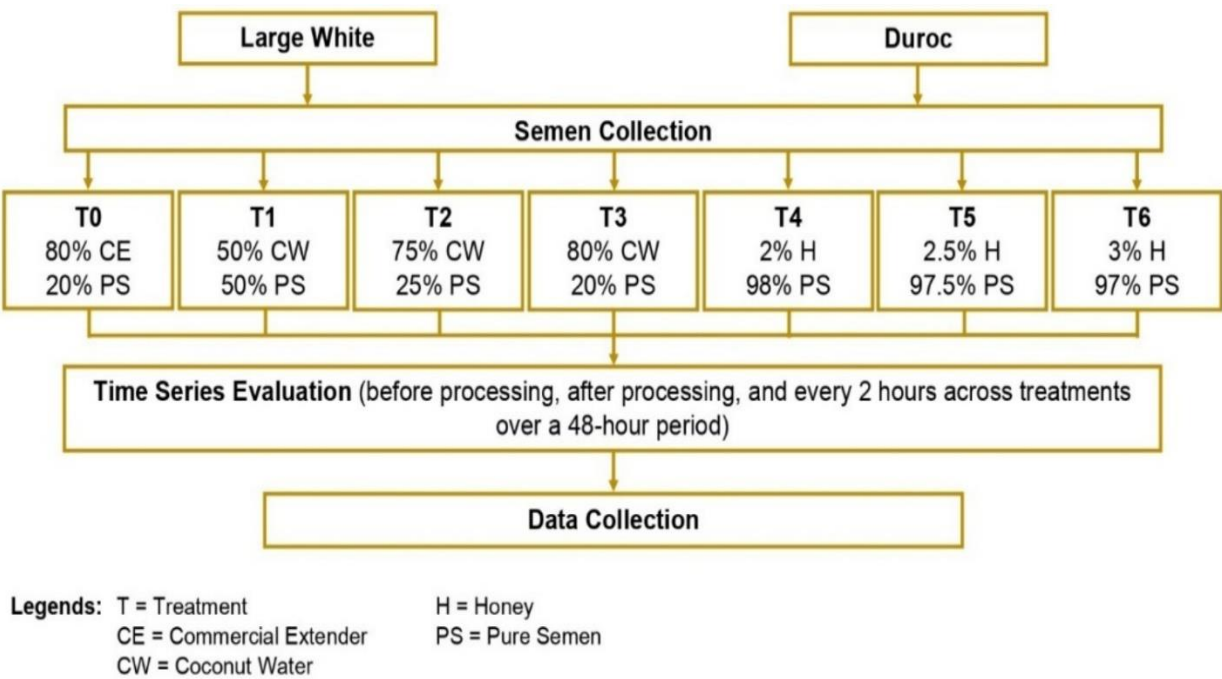
Materials and Methods

Research Design

This study used a multiple time series design (MTSD) to evaluate the motility of Large White and Duroc semen with different organic extender formulations. MTSD, as described by Phan and Ngu (2016) and Colicev and Pauwels (2018), is ideal for studies where true experiments are not feasible, as it involves repeated measurements over time across treatment and control groups (Schweizer et al., 2016). Tshabalala et al. (2021) and Gerzilov and Andreeva (2021) demonstrated this approach by assessing semen quality at various time points using different extenders, which aligned with this study’s tracking of motility over 48 hours. Other applications of MTSD, such as those by Monger et al. (2021) and Patrick et al. (2023), underscore its value in revealing temporal trends. Similarly, this study monitored semen motility from 8 a.m. on Day 1 to 8 a.m. on Day 3, omitting the 10 p.m. to 6 a.m. period, to examine the effects of extender type and storage time.

Figure 1

Researcher-Made Model for Semen Evaluation Using MTSD



The researcher-made model presented in Figure 1 outlines the experimental setup used to assess the impact of organic extenders (coconut water and honey) on semen motility in Large White and Duroc breeds. Semen samples were divided into seven treatments (T0–T6), each replicated thrice: T0 used a

commercial extender, T1–T3 used coconut water, and T4–T6 used honey. Motility was assessed before and after processing, then monitored from 8 a.m. to 8 p.m. at 2-hour intervals. Monitoring occurred after 2, 4, 6, 8, 10, and 12 hours across all treatments, including the control, and was assessed using the iSperm CASA device. Treatment protocols, adapted with minor changes from Rodriguez (2016), are detailed in Table 1.

Table 1

Summary of Different Semen Extenders for Large White and Duroc Semen

Legend	Treatment Combination
Treatment 0	80% Commercial Extender + 20% Pure Semen
Treatment 1	50% Coconut Water + 50% Pure Semen
Treatment 2	75% Coconut Water + 25% Pure Semen
Treatment 3	80% Coconut Water + 20% Pure Semen
Treatment 4	2% Honey + 98% Pure Semen
Treatment 5	2.5% Honey + 97.5% Pure Semen
Treatment 6	3% Honey + 97% Pure Semen

Each treatment group (T0–T6) consisted of 20 mL samples. T0 consisted of 80% commercial extender (MS DiluFert Silver® in 1 L distilled water) with 20% semen. T1–T3 used 50%, 75%, and 80% coconut water, while T4–T6 contained 2%, 2.5%, and 3% honey, all mixed with 20% semen. These ratios were based on Rodriguez (2016), who used a 3:1 ratio, with added variations for comparison. Lower honey concentrations were chosen based on prior studies and preliminary trials showing their efficacy even at minimal levels (Machebe et al., 2015; Castro et al., 2020; Balogun et al., 2021).

Despite small volumes, honey has been effective in preserving motility and vitality in Large White and Duroc semen, which are sensitive to thermal and osmotic stress (Pezo et al., 2018). To avoid early exhaustion and keep sperm alive longer, their metabolic activity must be slowed using inhibitors or cooling. Therefore, semen should be extended shortly after its collection (Lopez Rodriguez et al., 2017; Business Queensland, 2022; Knox, 2016).

Experimental Procedures

This study was conducted in a laboratory setting, specifically at the AI Laboratory of the International Training Center on Pig Husbandry (ITCPH), using semen samples from Large White and Duroc breeds. The sperm samples underwent seven treatments, including the control, and each treatment was replicated three times to ensure the reliability of the data.

Extender Preparation

The study used a commercial extender (MS DiluFert Silver®) and two organic extenders, coconut water and honey. The commercial extender came from MS Schippers Europe; standardized coconut water of the same maturity level came from a trusted store; and pure honey (THE BEEHIVE Farm and Kitchen) came from a reputable honeybee farm in Lipa City, Batangas.

The researcher considered MS DiluFert Silver®, which can preserve sperm for up to 3 days, as the control. Coconut water and honey, on the other hand, were sent to Mach Union Laboratories Inc. and Lipa Quality Control Center, Inc. for analysis and testing to assess key components of the samples. To ensure compatibility, extenders were prepared at 35°C, matching the semen's temperature (Crowell & Flowers, 2018). A tolerance of $\pm 1^\circ\text{C}$ was allowed if exact matching was not possible (Magapor, 2019).

Collection of Semen Samples

Semen samples used in the study were collected from 1-year-and-2-month-old Large White and Duroc boars, both common breeds in Philippine swine production. Semen collection began at 8:00 a.m. following the cleaning of each boar's penis with distilled water and tissue. Ejaculation was stimulated using a dummy sow, and semen was collected carefully to avoid urine contamination.

The semen was filtered to remove gel and debris, then evaluated using a full CASA system. Initial motility served as the baseline, with greater than 70% considered ideal (Chapman, 2016). It is also important to note that one Large White boar produced 200 mL and one Duroc 250 mL, which are sufficient for all treatments. Their semen temperature (35°C) was used to standardize extender preparation. Samples were stored in sterile, labeled squeeze bottles by breed.

Processing and Evaluation of Semen Samples

After initial evaluation, semen samples were processed using both commercial and organic extenders at 35 °C, with three replications per setup. Each treatment used no more than 20 mL of semen, adhering to the proportions shown in Table 1.

For Treatment 0. Before processing, 1 liter of distilled water was heated to 35°C and mixed with MS DiluFert Silver®, a 3-day commercial extender from MS Schippers Europe. A 20 mL solution was then prepared, consisting of 80% extender and 20% pure semen.

For Treatments 1, 2, and 3. Coconut water, of the same maturity level and standardized across all treatments, was sourced from a trusted store, filtered with nylon to remove solid particles, and kept at 35 °C. For T1, a 20 mL pure coconut water–semen solution was prepared, containing 50% coconut water and 50% pure semen. For T2, a 20 mL coconut water–semen solution was prepared, containing 75% coconut water and 25% pure semen. For T3, a 20 mL coconut water–semen solution was prepared, containing 80% coconut water and 20% pure semen.

For Treatments 4, 5, and 6. A 20 mL pure honey (THE BEEHIVE Farm and Kitchen) from a reputable Batangas farm was mixed with semen to form the honey–semen solution. The honey was maintained at 35 °C. For T4, the solution contains 2% pure honey and 98% pure semen. For T5, the solution contains 2.5% pure honey and 97.5% pure semen. For T6, the solution contains 3% pure honey and 97% pure semen.

Semen motility was assessed immediately using iSperm, a mobile analyzer from Aidmics Biotechnology (Domain et al., 2022), through the dipping method, which involves immersing the chip 5 mm into the sample. After analysis, samples were stored at 18 °C and monitored every 2 hours for 2 days, including the control group.

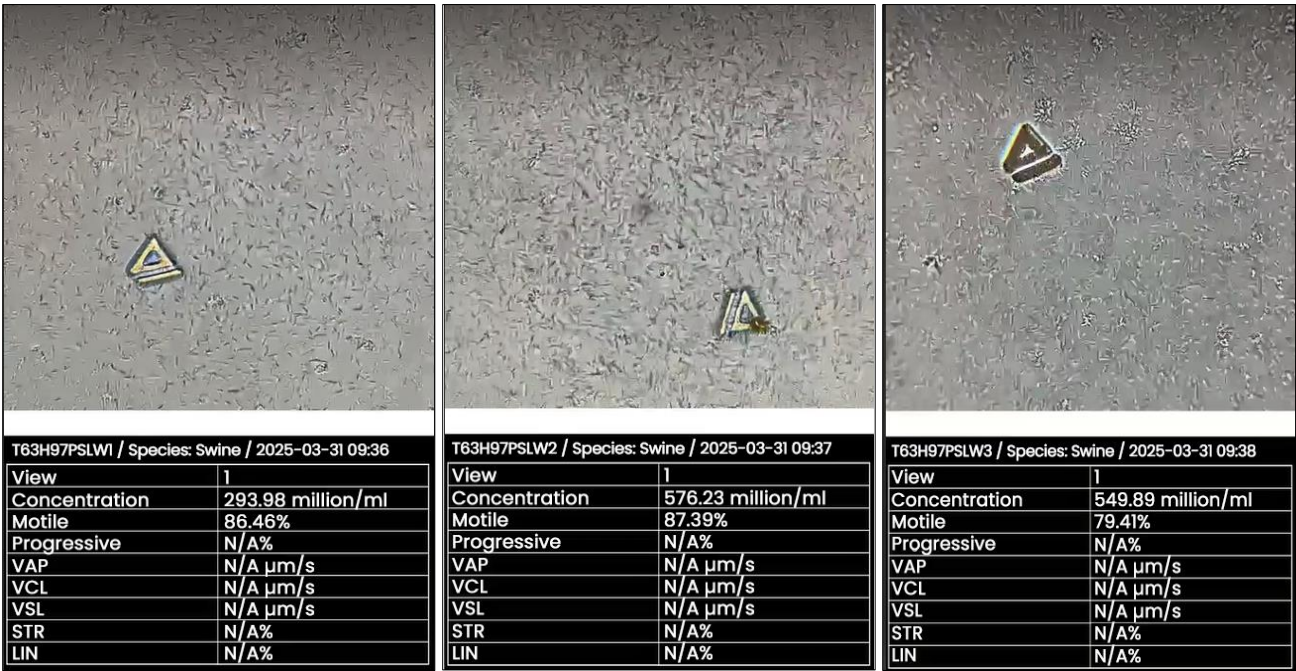
Monitoring of Semen Samples

After the initial evaluations (i.e., before and after processing), semen samples were monitored at 2, 4, 6, 8, 10, 12, and 48 hours across all treatments during daytime hours over two days. Motility was measured using the iSperm device, and proper handling and storage were maintained to ensure accuracy.

Figure 2 shows screenshots from the iSperm device, displaying swine semen under the microscope along with metadata like treatment, species, date, time, and motility percentage, the latter being the study's main focus.

Figure 2

iSperm Screenshots Showing Swine Semen and Motility Data



Time and Place of the Study

The study was conducted at the Artificial Insemination (AI) Laboratory of the International Training Center on Pig Husbandry (ITCPH) in Marawoy, Lipa City, Batangas, from March 27 to April 2, 2025. ITCPH, operated by the Department of Agriculture–Agricultural Training Institute (DA-ATI), is the only swine-specialized training center in Asia, known for its advanced facilities and expert staff in swine production. Treatments for Large White boars were carried out from March 27–29, and for Duroc boars from March 31–April 2, with no activity on March 30 due to the center’s Sunday closure. Each experimental setup lasted 48 hours, from 8 a.m. on the first day to 8 a.m. on the third.

Materials and Instruments

Table 2

List of Materials and Instruments

Materials and Instruments	Purpose
1. Large White and Duroc Semen	Essential samples for the study
2. Coconut Water and Honey	Organic components for semen extenders
3. Commercial Extender	Standard benchmark for comparison with organic extenders
4. iSperm	For sperm analysis, video capture, and extender calculation.
5. CASA	For sperm morphology analysis
6. Bio Refrigerator (18 °C)	For storing extended semen samples
7. Distilled Water (35 °C)	An addition to both commercial and organic extenders
8. Dummy Sow	For stimulating boars during semen collection
9. Nylon Filter	For filtering semen samples
10. Squeeze Bottle	For storage of processed semen samples
11. Measuring Cup	Used for the precise measurement of solutions and samples
12. Timer	For monitoring the duration of semen evaluation and processing
13. Sterile Gloves and Lab gown	For aseptic handling and preventing contamination

Data Gathering Procedure

The study systematically collected and analyzed data on the composition and performance of both organic and commercial semen extenders, along with the motility rate of boar semen. The coconut water and honey samples were analyzed for carbonate, bicarbonate, total sugar, and phosphate (as P) at Mach Union Laboratories Inc., while other components were evaluated at Lipa Quality Control Center, Inc. The semen of Large White and Duroc boars was initially evaluated at the International Training Center on Pig Husbandry (ITCPH) using a full CASA system, measuring ejaculate volume, morphology, and motility rates—including static, progressive, motile, slow, and total counts. Motility rates before and after the addition of organic semen extenders were assessed using iSperm. Seven treatments (T0 to T6), with three replications each, were compared to the baseline motility of pure semen for both breeds. Additionally, the motility rates of processed semen using both organic and commercial extenders were monitored using iSperm at various time intervals: after processing and at 2, 4, 6, 8, 10, 12, and 48 hours.

Statistical Analysis

The data were arranged in Microsoft Excel (2021) and analyzed using SPSS version 20. Descriptive statistics (mean and percentage) were used to assess coconut water and honey parameters, including moisture, bicarbonate, carbonate, phosphate, total sugar, crude protein, calcium, salt (NaCl), sodium, and potassium. Analysis of Variance (ANOVA) was conducted to identify significant differences in semen motility across seven treatments (T0–T6) at a significance level of 0.005, following tests for normality and homogeneity of variance. Each treatment had three replicates ($n = 3$), and Tukey's HSD served for post hoc tests. Neither effect sizes nor confidence intervals were computed.

Ethical Consideration

This study adhered to the Animal Welfare Act of 1998, DA Administrative Order No. 40 (s.1999), and the PALAS Code of Practice for the Care and Use of Laboratory Animals. Boars used in this study were housed in standard-compliant facilities, and managed by a licensed veterinarian and trained handlers. Semen collection was conducted humanely, and any signs of animal distress were addressed promptly. Moreover, institutional and government ethical policies were strictly observed. Research data were handled securely, findings were reported with transparency, and any ethical issues were disclosed.

Results and Discussion

Composition of Coconut Water and Honey as Organic Semen Extenders

Analysis from Lipa Quality Control Center and Mach Union Laboratories revealed key compositional differences between coconut water and honey. Mature coconut water had higher moisture content (94.76%) and bicarbonate levels (66.8 mg/100 g vs. 0.022 mg/100 g), making it more suitable for hydration and pH buffering. In contrast, pure honey had higher total sugar content (53.6 g/100 g vs. 3.7 g/100 g), providing energy for sperm cells.

These findings align with Hussain et al. (2018) and Baiee et al. (2017), who highlighted the importance of extender composition in improving sperm viability, motility, and fertilization. Likewise, Pezo et al. (2018) and Bustani and Baiee (2021) stressed the need for extenders to shield sperm from freeze shock and oxidative stress by ensuring proper pH and antioxidant levels.

Table 3*Composition of Coconut Water and Honey as Semen Extenders*

Compositions	Coconut Water	Honey
Moisture (%)	94.76	22.01
Bicarbonate (mg/100g)	66.8	0.022
Carbonate (mg/100g)	0	0
Phosphate (mg/100g)	13.3	16.7
Total Sugar (g/100g)	3.7	53.6
Crude Protein (%)	0.43	1.19
Calcium (%; mg/kg)	0.05; 481.21	0.02; 226.26
Salt (NaCl) (%; mg/kg)	0.05; 479.95	0.12; 1,265.58
Sodium (%; mg/kg)	0.02; 195.89	0.05; 474.27
Potassium (%; mg/kg)	0.18; 1,764.10	0.29; 2,947.67

Semen Evaluation of Large White and Duroc Before Processing

The International Training Center on Pig Husbandry (ITCPH) found baseline differences in semen quality between pure Large White and Duroc boars. Duroc boars had a higher ejaculate volume (250 mL vs. 200 mL) and total motility (99% vs. 98%), while Large White boars had greater progressive motility (80.90% vs. 76%). Morphology was nearly identical (71.50% vs. 71.60%).

The observed differences support Czubaszek et al. (2019), who noted intra-ejaculate variability in morphology, though values remained above the 70% threshold. Duroc's higher volume aligns with Gorski et al. (2017), reflecting its known semen quality. Both breeds met standards for morphology and motility, confirming their suitability for organic extender trials.

Table 4*Evaluation of Large White and Duroc Semen Before Processing*

Composition	Large White		Duroc	
	Count	Percent of Total	Count	Percent of Total
Static	4.00	2.00	2	1.00
Progressive	165.00	80.90	219	76.00
Motile	200.00	98.00	285	99.00
Slow	21.00	10.30	36	12.50
Total	204.00	100.00	288	100.00
Morphology	71.50%		71.60%	
Ejaculate volume	200.00		250.00	

Note. Motile, progressive, and slow are mutually exclusive and were analyzed separately. Morphology is based on the normal fraction, which refers to the percentage of sperm with no visible defects in head, midpiece, or tail structure.

Motility Rate of Large White and Duroc Semen Before and After the Addition of Organic Semen Extenders

Pure Large White and Duroc semen maintained high motility with honey-based extenders, particularly in Treatments 4 and 6, while coconut water-based extenders resulted in lower and more variable motility, with Treatment 3 being the least effective. Overall, honey-based extenders outperformed both coconut-based and commercial extenders in preserving sperm motility.

These results are parallel with studies on bull semen, wherein honey improved cryopreservation outcomes. Malik (2018) reported enhanced motility and reduced abnormalities with honey-treated bull

semen, Yimer et al. (2015) found 2.5% honey in Tris extender outperformed Bioxcell, and Chung et al. (2019) observed breed-specific effects, with Jersey bulls showing the highest sperm quality.

Table 5

Motility Rate Percentage (%) of Large White Pure Semen and After Processing with Organic Extenders

Large White	Replication 1	Replication 2	Replication 3	Mean \pm SD
Pure Semen	82.31	81.87	83.37	82.52 \pm 0.77
Treatment 0	68.88	76.48	73.19	72.85 \pm 3.81 ^a
Treatment 1	82.83	74.60	59.45	72.29 \pm 11.86 ^b
Treatment 2	62.45	66.70	65.57	65.01 \pm 2.25 ^c
Treatment 3	47.36	51.70	48.31	49.12 \pm 2.28 ^{a, b, d, e, f, g}
Treatment 4	86.34	82.02	82.64	83.67 \pm 2.34 ^{d, e}
Treatment 5	86.14	73.85	86.73	82.24 \pm 7.27 ^{d, f}
Treatment 6	86.46	87.39	79.41	84.42 \pm 4.36 ^{c, d, g}

Legend: T0 = 80% Commercial Extender + 20% Pure Semen

T4 = 2% Honey + 98% Pure Semen

T1 = 50% Coconut Water + 50% Pure Semen

T5 = 2.5% Honey + 97.5% Pure Semen

T2 = 75% Coconut Water + 25% Pure Semen

T6 = 3% Honey + 97% Pure Semen

T3 = 80% Coconut Water + 20% Pure Semen

Note: Means sharing the same letter are significantly different ($p < 0.05$) based on post hoc test.

Pure semen samples were evaluated for baseline motility only and were not included in the post-treatment statistical comparisons.

Table 6

Motility Rate Percentage of Duroc Pure Semen and After Processing with Organic Extenders

Duroc	Replication 1	Replication 2	Replication 3	Mean \pm SD
Pure Semen	75.05	82.28	80.47	79.27 \pm 3.76
Treatment 0	80.66	73.42	66.90	73.66 \pm 6.88 ^{a, c, d, e}
Treatment 1	81.23	79.88	75.68	78.93 \pm 2.89 ^{b, c}
Treatment 2	91.51	95.67	97.10	94.76 \pm 2.90 ^{a, b, c, f, g}
Treatment 3	92.87	91.69	94.74	93.10 \pm 1.54 ^{a, d, f, g}
Treatment 4	90.20	89.77	88.36	89.44 \pm 0.96 ^{a, e}
Treatment 5	77.56	76.97	74.91	76.48 \pm 1.39 ^{c, d, f}
Treatment 6	75.46	74.69	77	75.72 \pm 1.18 ^{c, d, g}

Legend: T0 = 80% Commercial Extender + 20% Pure Semen

T4 = 2% Honey + 98% Pure Semen

T1 = 50% Coconut Water + 50% Pure Semen

T5 = 2.5% Honey + 97.5% Pure Semen

T2 = 75% Coconut Water + 25% Pure Semen

T6 = 3% Honey + 97% Pure Semen

T3 = 80% Coconut Water + 20% Pure Semen

Note: Means sharing the same letter are significantly different ($p < 0.05$) based on post hoc test.

The initial motility of pure Duroc semen was high and remained viable with the commercial extender. However, coconut water-based extenders (T2 and T3) yielded even higher motility, sometimes exceeding that of pure semen, while the honey-based extender (T4) also performed well. Though T1, T5, and T6 showed slightly lower values, all treatments maintained acceptable motility, with T2, T3, and T4 showing the most promise. Similar findings were reported in other studies, including those by Tshabalala et al. (2021) and Machebe et al. (2015) on boar semen, and Esguerra et al. (2020) on native chicken semen. Subsequent monitoring over time further revealed breed-specific trends in extender efficacy.

Motility Rates of Processed Large White and Duroc Semen Using Organic and Commercial Extenders Across Time Intervals

Immediately after processing and two hours after, significant differences in sperm motility were observed among Large White semen treatments ($F(6,14) = 13.89$, $p = .001$ and $F(6,14) = 9.67$, $p = .001$,

respectively; $n = 3$ per group), rejecting the null hypothesis in both cases. Treatment 6 (3% honey + 97% semen) showed the highest initial motility at 84.42%, while Treatment 4 (2% honey + 98% semen) had the highest motility after two hours at 83.67%, both outperforming the control groups. These results suggest that honey-based extenders, likely due to their antioxidant and antimicrobial properties, are more effective in maintaining motility than coconut water or commercial alternatives. Conversely, Treatment 3 (80% coconut water + 20% semen) consistently showed the lowest motility—49.12% initially and 42.05% after two hours—implying that high concentrations of coconut water may impair sperm viability, possibly due to osmotic imbalance or suboptimal pH.

Table 7

Motility Rate Percentage of Large White Semen Processed with Organic and Commercial Semen Extenders from 0 to 12 Hours

		Large White						
		T0	T1	T2	T3	T4	T5	T6
After processing	Mean	72.85	72.29	65.01	49.12	83.67	82.24	84.42
	F-value: 13.89							
	p-value: .001							
After 2 hours	Mean	72.18	72.29	66.57	42.05	83.67	82.25	77.50
	F-value: 9.67							
	p-value: .001							
After 4 hours	Mean	64.11	79.05	2.12	0.70	80.05	55.59	48.74
	F-value: 62.78							
	p-value: .001							
After 6 hours	Mean	70.30	35.03	0.33	0.98	66.71	31.57	17.21
	F-value: 53.58							
	p-value: .001							
After 8 hours	Mean	45.47	29.33	0.05	0.32	72.88	25.44	10.58
	F-value: 41.31							
	p-value: .001							
After 10 hours	Mean	39.49	31.63	0.00	0.00	71.52	28.17	13.02
	F-value: 33.89							
	p-value: .001							
After 12 hours	Mean	36.84	44.82	0.00	0.00	69.29	29.28	17.77
	F-value: 23.90							
	p-value: .001							

Note: The p-value is below 0.05, which indicates high significance; thus, the null hypothesis is rejected.

Legend: T0 = 80% Commercial Extender + 20% Pure Semen

T4 = 2% Honey + 98% Pure Semen

T1 = 50% Coconut Water + 50% Pure Semen

T5 = 2.5% Honey + 97.5% Pure Semen

T2 = 75% Coconut Water + 25% Pure Semen

T6 = 3% Honey + 97% Pure Semen

T3 = 80% Coconut Water + 20% Pure Semen

Four and six hours after processing, significant differences in motility rates were observed among Large White semen treatments ($F(6,14) = 62.78$, $p = .001$ and $F(6,14) = 53.58$, $p = .001$, respectively; $n = 3$ per group), leading to the rejection of the null hypothesis in both cases. At four hours, Treatment 4 (2% Honey + 98% Semen) showed the highest motility at 80.05%, highlighting honey's potential as an effective organic extender due to its antioxidant and antimicrobial properties. However, high concentrations of coconut water proved detrimental, as seen in Treatment 3 (80% Coconut Water + 20% Semen), which had a motility of only 0.70%. At six hours, although motility declined overall, Treatment 0 (80% commercial extender + 20% semen) recorded the highest rate at 70.30%, confirming its strong preservation capability. Treatment 4 followed with 66.71%, maintaining its effectiveness. Meanwhile, Treatment 2 (75% Coconut

Water + 25% Semen) showed a critically low motility of 0.33%, reinforcing that excessive coconut water compromises sperm viability.

Eight and ten hours after processing, a significant difference in motility rates was observed among Large White semen treatments ($F(6,14) = 41.31$, $p = .001$ and $F(6,14) = 33.89$, $p = .001$, respectively; $n = 3$ per group), both leading to the rejection of the null hypothesis. In both instances, Treatment 4 (2% Honey + 98% Semen) consistently showed the highest motility (72.88% and 71.52%), suggesting that low concentrations of honey effectively preserve sperm motility over time. Conversely, high concentrations of coconut water (75–80%) in Treatments 2 and 3 resulted in drastically reduced or no motility, likely due to osmotic stress or sugar imbalance impairing sperm viability.

Twelve hours after processing, the analysis showed a significant difference in motility rates among Large White semen treatments ($F(6,14) = 23.90$, $p = .001$; $n = 3$ per group), leading to the rejection of the null hypothesis. Treatment 4 (2% Honey + 98% Pure Semen) recorded the highest motility at 69.29%, indicating its effectiveness in preserving sperm viability. In contrast, Treatments 2 and 3, which had high coconut water concentrations, showed complete immotility, suggesting detrimental effects on sperm. By the twelfth hour, the honey-based Treatment 4 proved the most effective extender. Treatment 1 showed inconsistent potential, while coconut water-based Treatments 2 and 3 were ineffective. These results emphasize the critical role of extender composition in artificial insemination success for Large White boars.

Studies highlight honey's potential as a natural extender due to its antimicrobial, antioxidant, and osmo-protective properties, which enhance sperm motility and viability (Arakeri et al., 2020; Machebe et al., 2015; Gonzalez-Castro & Herickhoff, 2022). However, its effectiveness depends on proper formulation and concentration. In contrast, high levels of coconut water can reduce sperm quality due to low buffering capacity and pH imbalance (Sawitri et al., 2021; Tafuli et al., 2024). Organic extenders like honey and coconut water show promise, but their success relies on optimized composition to maintain semen quality over time (Mawin-Ray et al., 2025; Lopez Rodriguez et al., 2017; Reckova et al., 2022).

Immediately after processing ($F(6,14) = 23.78$, $p = .001$), motility analysis revealed a significant difference among Duroc semen treatments, with Treatment 2 (75% Coconut Water + 25% Semen) showing the highest motility at 94.76%, slightly above the baseline and indicating potential extender effects. This suggests that 75% coconut water may be more effective in preserving motility than the commercial extender, as Treatment 0 (80% Commercial Extender + 20% Semen) recorded only 73.66%. Two hours later, a significant difference was again observed ($F(6,14) = 3.54$, $p = .024$), with Treatment 2 maintaining the highest motility at 87.80%, although not statistically different from the control, reinforcing its effectiveness for short-term storage. In contrast, Treatment 6 (3% Honey + 97% Semen) had the lowest motility at 55.61%, emphasizing how extender composition and concentration influence preservation efficacy.

Four and six hours after processing, significant differences in motility rates were observed among Duroc semen treatments ($F(6,14) = 35.31$, $p = .001$ and $F(6,14) = 13.41$, $p = .001$, respectively; $n = 3$ per group), leading to the rejection of the null hypothesis in both cases. At four hours, Treatment 0 (80% Commercial Extender + 20% Semen) had the highest motility at 79.49%, outperforming all organic extenders, while Treatment 3 (80% Coconut Water + 20% Semen) showed minimal viability at 3.23%. At six hours, Treatment 0 again led with 60.51%, followed by Treatment 1 (57.37%) as the most stable organic alternative. Meanwhile, Treatment 3 remained ineffective at 2.49%. These results indicate a general decline in semen quality over time and demonstrate that while commercial extenders perform best, certain organic options may offer moderate midterm preservation.

Eight and ten hours after processing, a significant difference in motility rates was observed among Duroc semen treatments ($F(6,14) = 27.15$, $p = .001$ and $F(6,14) = 7.27$, $p = .001$, respectively; $n = 3$ per group), leading to the rejection of the null hypothesis at both time points. Treatment 1 (50% coconut water + 50% semen) consistently showed the highest motility—65.26% at 8 hours and 66.23% at 10 hours—demonstrating superior mid- to late-term preservation. Though not the best in earlier assessments, its efficacy became apparent over time, likely due to its sustained metabolic support. In contrast, Treatment 3 (80% coconut water + 20% semen) showed drastic motility decline, reaching just 2.73% at 8 hours and 1.58% at 10 hours, indicating poor extender performance.

Twelve hours after processing, the analysis showed a significant difference in motility rates among Duroc semen treatments ($F(6,14) = 50.98$, $p = .001$; $n = 3$ per group), leading to the rejection of the null hypothesis. Treatment 1 (50% coconut water + 50% pure semen) showed the highest motility rate at 58.34%, outperforming the commercial extender and highlighting its effectiveness as an organic alternative. In contrast, Treatment 3 (80% coconut water + 20% semen) showed complete immotility, indicating poor preservation.

Table 8

Motility Rate Percentage of Duroc Semen Processed with Organic and Commercial Semen Extenders from 0 to 12 Hours

		Large White						
		T0	T1	T2	T3	T4	T5	T6
After processing	Mean F-value: 23.78 p-value: .001	73.66	78.93	94.76	93.10	89.44	76.48	75.72
After 2 hours	Mean F-value: 3.54 p-value: .024	73.66	71.81	87.80	76.11	84.52	67.58	55.61
After 4 hours	Mean F-value: 35.31 p-value: .001	79.49	68.25	36.55	3.23	63.61	29.74	33.75
After 6 hours	Mean F-value: 13.41 p-value: .001	60.51	57.37	14.54	2.49	32.13	21.87	18.41
After 8 hours	Mean F-value: 27.15 p-value: .001	57.76	65.26	12.43	2.73	18.60	13.71	17.60
After 10 hours	Mean F-value: 7.27 p-value: .001	37.88	66.23	13.77	1.58	26.33	9.37	26.03
After 12 hours	Mean F-value: 50.98 p-value: .001	40.61	58.34	27.29	0.00	16.84	12.04	15.67

Note: The p-value is below 0.05, which indicates high significance; thus, the null hypothesis is rejected.

Legend: T0 = 80% Commercial Extender + 20% Pure Semen

T1 = 50% Coconut Water + 50% Pure Semen

T2 = 75% Coconut Water + 25% Pure Semen

T3 = 80% Coconut Water + 20% Pure Semen

T4 = 2% Honey + 98% Pure Semen

T5 = 2.5% Honey + 97.5% Pure Semen

T6 = 3% Honey + 97% Pure Semen

Studies show that coconut water and honey-based extenders can enhance sperm motility, though effectiveness varies by breed, concentration, and storage time (Dziekonska et al., 2017; Lopez Rodriguez et al., 2017; Esguerra et al., 2020). Duroc semen responded better than Large White, indicating breed-

specific sensitivity. While some organic formulations perform well in the short term, their long-term viability is limited (Castro et al., 2020; Reckova et al., 2022).

Extender performance depends on formulation stability, buffering capacity, and antioxidant content rather than the base ingredient alone (Magapor, 2019; Chankitisakul et al., 2023). Treatment 1 consistently outperformed others, including commercial extenders, supporting the importance of optimized natural extenders in preserving boar semen (Rodriguez, 2016; Tshabalala et al., 2021).

Conclusion

Matured coconut water aided hydration and pH balance, while honey provided an energy-rich medium—both influencing semen motility. Duroc boars yielded more ejaculate with higher total motility, whereas Large White boars showed greater progressive motility. Honey-based Treatment 4 preserved Large White semen motility above 70% for 12 hours, while coconut-based Treatment 1 maintained Duroc semen motility for up to 48 hours, reflecting breed-specific responses to extender composition.

Treatment responses varied by breed: coconut-based extenders worked briefly, while honey-based maintained motility only for a short time. Duroc and Large White semen reacted differently, highlighting the importance of breed-specific, balanced formulations. While honey suited Large White and coconut worked better for Duroc, none preserved motility beyond 48 hours, underscoring the need for further studies on antioxidants, buffers, and genotype-specific cryoprotectants.

Recommendations

Based on the study's findings and conclusions, several recommendations are suggested to enhance swine reproduction and artificial insemination practices. Swine farmers and breeders are encouraged to consider honey- and coconut water-based extenders for short- to mid-term semen storage, although these require further validation through multi-site fertility trials and post-AI performance evaluations. AI centers are likewise advised to explore the gradual adoption of organic extenders as sustainable alternatives, particularly in contexts where synthetic supplies are costly or scarce. To support this transition, veterinarians and AI technicians should receive updated training on the preparation and application of organic extenders, with attention to breed-specific protocols, quality control, and dosing accuracy. Finally, future research should aim to develop antioxidant-rich, genotype-specific organic extenders and conduct long-term studies on fertility outcomes, litter size, and overall swine productivity following AI.

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