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Macroscopic Quality Of Pig Semen (Sus Scrofa) In A Combination Of *Aloe Vera*, Soybean Juice (*Glycine Max*), And Lontar Fruit Water (*Borassus Flabellifer*)

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Abstract

Artificial insemination (AI) in pig farming requires good quality liquid semen that can be maintained during storage. *The purpose of this study was to determine the macroscopic quality of pig semen after the addition of natural diluting ingredients from the combination of* aloe vera juice (LB), soy juice (SK), and palm fruit water (ABL). This study used a complete random design with 6 treatments and 4 replicates. The treatment consists of P0 = no treatment, P1 = 1% LB: 5% SK: 94% ABL, P2 = 2% LB: 10% SK: 88% ABL, P3 = 3% LB: 15% SK: 82% ABL, P4 = 4% LB: 20% SK: 76% ALB, and P+ = Diltec Semen Energy. The observed parameters included volume, color, odor, pH, and cement consistency. The results showed that all treatments showed a semen volume of 11 mL, milky white color, dilute consistency, and pH of 6.6-6.8. The difference was only observed in the smell of cement, where the treatment containing palm fruit water showed a distinctive aroma of palm fruit compared to the typical smell of cement/fishy in the control group that used *Diltec Cement Energy* odorless. Conclusion: The combination of these diluting ingredients did not negatively alter the macroscopic parameters of pigs.

Keywords: palm fruit water; artificial insemination; macroscopic quality; aloe vera; soy juice

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INTRODUCTION

Pig farming is one of the livestock commodities that has advantages in the form of a fast growth rate and a high number of children per birth, so it has great potential as a source of meat production to meet the animal protein needs of the community (Oroh et al., 2023). The success of pig livestock development is largely determined by the efficiency of the reproductive system, where artificial insemination (IB) is widely chosen because it is able to accelerate the genetic spread of superior males, reduce the risk of transmission of reproductive diseases and reduce the cost of maintaining males (Umbu *et al.,* 2025).

The quality of liquid cement is a key factor in IB's success. The cement used must be able to maintain the motility, viability and integrity of the membrane during the storage period before being applied to the female (Renaldi et al., 2024). During the storage process, spermatozoa are highly susceptible to oxidative stress, pH changes, and lack of energy sources so that the quality of semen is easily degraded. Cement thinners play an important role in providing nutrients, maintaining pH and osmotic pressure, and protecting spermatozoa from damage due to temperature changes (Foeh and Gaina, 2017; Riwu *et al.,* 2022).

Commercial diluents such as BTS and Durasperm have been widely used and proven to be effective in maintaining the quality of pig semen in short-term storage (Krause et al., 2019; Banamtuan *et al.,* 2021). However, relatively high prices and limited distribution make it difficult for smallholder farmers in areas such as East Nusa Tenggara to access them (Foeh and Gaina, 2017). This condition encourages the development of natural diluents based on local ingredients that are cheap, easy to obtain, and still able to maintain cement quality (Diana et al., 2017; Silvestre *et al.,* 2021).

Palm fruit water (Borassus flabellifer) is one of the local ingredients that has the potential to be used as a natural diluent. Palm fruit water contains simple carbohydrates as a source of energy, minerals, and phenolic compounds that are antioxidants (Mere et al., 2019; Dzigbor et al., 2025). Foeh and Gaina (2017) reported that palm fruit juice was able to maintain the quality of pig spermatozoa at room temperature storage, while Banamtuan *et al.* (2021) showed that palm fruit water supplementation in Durasperm diluent effectively maintained the motility and viability of Duroc pig liquid semen for up to 64 hours.

In addition to lontar fruit water, aloe vera (Aloe vera) and soy juice (Glycine max) also have the potential to be diluting components. Aloe vera gel contains vitamins A, C and E, polysaccharides, and phenolic compounds that act as antioxidants and cell membrane protectors (Renaldi *et al.,* 2024). Soy juice contains protein, lecithin and isoflavones; Lecithin protects

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spermatozoan membranes from cold shock, while isoflavones act as antioxidants that suppress lipid peroxidation (Winarsi *et al.,* 2010).

Evaluation of the macroscopic quality of cement, including volume, color, odor, consistency and pH, is a crucial first step before cement is microscopically assessed and used for IB (Riwu et al., 2022). Negative changes in macroscopic parameters can describe the presence of contamination, inappropriate diluent composition, or metabolic disorders that have the potential to degrade fertility (Setiadi *et al.,* 2022). Based on this, this study was conducted to evaluate the macroscopic quality of pig liquid semen in a natural diluent combination of palm fruit water, aloe vera juice and soybean juice in several ratios.

THEORETICAL INSIGHT

1. Pig Liquid Cement

Semen is a male reproductive excretion that is generally channeled by female reproductive organs through copulation, but semen can also be collected for the purpose of Artificial Insemination (IB). The composition of semen includes two main components: spermatozoa (male seed cells) suspended in a liquid or semi-gelatin medium known as cement plasma, similar to the structure of blood consisting of cells and plasma.

Pig semen has voluminous properties, which means that pig semen has a high volume of 150-200 ml and a low spermatozoa concentration of 200-300 x 106 cells/ml. Pig semen can only be taken twice a day for a period of several days, if more than that is taken, the quality of sperm in semen will decrease. Pig cement can only be stored while maintaining its quality in the temperature range of 15-20°C (Sumardani *et al*., 2008).

2. Cement Thinners

Pengencer Semen is a substance or solution that is added to liquid or frozen semen from animals to maintain the fertility of spermatozoa during storage so that it can be used in the Artificial Insemination (IB) process. Natural materials are more recommended as the base material for cement thinners. Especially from vegetable sources, which contain various important components such as carbohydrates (glucose and fructose), minerals, vitamins, amino acids, as well as bioactive compounds and antioxidants that play a role in maintaining the survival of spermatozoa. Natural cement thinners are used as a substitute alternative or as a combination with synthetic and commercial thinners, with the aim of reducing production costs, improving the efficiency of cement use, and utilizing local resources that are easily available (Diana *et al*., 2017).

Physiologically, spermatozoa are cells that are very sensitive to environmental changes,

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especially during the storage process outside the body. Natural cement thinners play an important role in providing a source of energy for spermatozoa through simple sugar content, maintaining pH balance and osmotic pressure, and protecting cell membranes from damage due to oxidative stress and temperature changes. The content of vitamins and antioxidant compounds in natural ingredients is known to be able to suppress the formation of free radicals that can damage membrane structure and reduce spermatozoa motility and viability (Sylvester *et al*., 2021). However, the effectiveness of natural cement thinners is greatly influenced by the type of material, concentration, as well as processing and storage methods. Variations in nutrient composition between natural ingredients can cause differences in spermatozoa responses, so proper formulation and treatment are needed for natural diluents to provide optimal results

3. Aloe *Vera* Juice

Aloe vera is a medicinal plant known by the scientific name Aloe vera. This plant belongs to the Liliaceae family and is native to Africa, but is now widely cultivated in tropical areas such as Indonesia. In general, aloe vera is a thick-fleshed plant (succulent) that has long, green leaves, and contains a clear gel inside (Marhaeni, 2004).

Aloe vera extract is a liquid taken from the gel part of the leaves of the Aloe vera plant. This gel is widely used because it contains various active substances such as vitamins (including vitamins A, C, and E), flavonoids, phenolic compounds, and polysaccharides, all of which play a role in warding off free radicals. It is this antioxidant content that makes aloe vera often studied in many studies because of its ability to help protect cells from oxidative damage. Vitamin E, as one of the main antioxidant components in aloe vera, is known to have a strong ability to maintain cell membrane stability by inhibiting lipid peroxidation processes triggered by free radicals (Renaldi et al., 2024).

The manufacturing process usually begins with cutting aloe vera leaves, then part of the gel is taken and soaked with a solvent such as water or ethanol so that the active substances come out into the liquid. After that, the liquid is filtered so that a clean and ready to use aloe vera extract is produced. This extract is often used in various products because it has antimicrobial and antioxidant properties that help inhibit the growth of bacteria and maintain the stability of the product.

4. Air Buah Lontar (*Birassus flabellifer*)

Palm fruit water is a natural liquid derived from the fruit of the palm plant or siwalan with the scientific name Borassus flabellifer. Young palm fruit has a sweet taste because it contains quite high natural sugar. The sugar content is included in the group of simple carbohydrates such

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as glucose and fructose which act as a source of energy. In addition to carbohydrates, palm fruit also contains various important minerals such as potassium, calcium, and phosphorus that support electrolyte balance and physiological functions of the body (Mere et al., 2019). Palm fruit is also known to contain bioactive compounds such as phenolics and flavonoids that act as natural antioxidants. These compounds help ward off free radicals and protect cells from oxidative damage (Dzigbor et al., 2025).

In the field of livestock reproduction, materials containing simple sugars are often used as a source of energy for spermatozoa because they can help maintain motility and viability during storage. The mineral content in natural materials also plays a role in maintaining osmotic balance and pH stability of cement diluent solutions (Mere et al., 2019). Based on this nutritional content, palm fruit water has the potential to be used as an alternative to natural diluents that are more economical and easy to obtain.

5. Soybeans (*Glycine max*)

Soybeans (*Glycine max*) are leguminosa plants that are widely used as a source of vegetable protein and raw materials for various food and non-food products. In addition to its use in the food sector, soybeans also have potential in the field of livestock reproduction, especially as a basic material for cement diluents. This is due to the content of nutrients and bioactive compounds in it that are able to support survival and protect spermatozoa during the storage process. Soybean juice is known to contain protein, fat, vitamins, lecithin, and natural antioxidant compounds such as isoflavones which play an important role in maintaining cell stability (Fadilla, Z. J., 2024).

One of the main components in soybeans that plays a role in cement dilution is isoflavones. Isoflavones are phenolic compounds that have high antioxidant activity. In semen storage, spermatozoa are prone to increased *Reactive Oxygen Species* (ROS), which is a free radical molecule that can damage the plasma membrane through the process of lipid peroxidation. This damage has an impact on decreased motility, disruption of membrane integrity, and even cell death. The isoflavones in soybean juice work by neutralizing free radicals so that they can reduce oxidative stress and maintain the viability of spermatozoa during storage (Winarsi *et al*., 2010).

Another meaningful component in soybeans is phospholipid-containing lecithin, for example phosphatidyl choline. The phospholipids have a structure arrangement similar to the main material of the plasma membrane of spermatozoa, which is useful for maintaining the stability and fluidity of the membrane during the cooling process. Cold shock or damage due to sharp temperature changes is often the dominant factor that reduces the quality of liquid cement.

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Based on the report, the lecithin level in soybean juice is in the range of 1.488%–3.08%. Lecithin acts as a membrane protector by stabilizing lipid composition and inhibiting the leakage of intracellular substances, so that spermatozoa quality is better maintained during storage at low temperatures (Tethool et al., 2022).

Soybeans also have proteins and fats that act as nutritional providers for spermatozoa. Proteins provide amino acids necessary in the metabolism and enzymatic activity of cells, while fats sustain the stability of the plasma membrane structure which is predominantly composed of lipid layers. Fat can also serve as an additional source of energy to maintain spermatozoa motility. Thanks to the presence of these nutrients, spermatozoa obtain more optimal metabolic support to maintain their mobility and vitality during the storage of liquid semen. Studies on the use of soybean juice in cement dilution revealed that at certain concentrations it can yield the best results for spermatozoa quality. The 25% concentration is reported to be able to maintain motility, increase viability, while suppressing the level of abnormalities during storage. Levels that are too low are less effective, while levels that are too high have the potential to increase osmotic pressure and trigger stress in spermatozoa (Klas *et al.,* 2025).

MATERIALS AND METHODS

1. Research Location

The research location was carried out at the Animal Husbandry Laboratory Unit of the Kupang State Agricultural Polytechnic.

2. Research Materials

The diluting materials used in this study are natural ingredients consisting of aloe vera (LB), soy juice (SK) and palm fruit water (ABL), as well as Diltec Semen Energy's commercial diluent as a positive control.

3. Research Methods

a. Cement Collection and Evaluation

Fresh semen was accommodated with the help of *dummy sow* using *the Glove hand method* in 1.5-year-old male Landrace pigs. The cement is stored in a container whose top is filtered to separate it from the gelatin. Fresh cement is taken to the laboratory to be evaluated for its macroscopic quality including: Volume, color, pH, and consistency.

b. Research Design

The study was conducted experimentally with a Complete Random Design (RAL), consisting of 6 treatments with 4 repetitions, namely P0 = no treatment, P1 = 1%LB : 5%SK : 94%ABL, P2 =

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2%LB : 10%SK : 88%ABL, P3 = 3%LB : 15%SK : 82%ABL, P4 = 4%LB : 20%SK : 76% ALB, P+ = Diltec Semen Energy. Each treatment was repeated 4 times with an average of relief from the first to the fourth week.

c. Data Analysis

The data were analyzed using *analysis of variance* (ANOVA) and there were differences followed by Duncan's follow-up test (Steel and Torrie, 1995).

RESULTS AND DISCUSSION

The results of the examination of spermatozoa volume, color, pH, and consistency in pigs are presented in Table 1.

Table 1. Macroscopic quality of pig liquid cement on a variety of natural diluents

Treatment	Volume (mL)	Color	Construction/Aroma	Consistency	pH
P0	11	Milky white	Typical of cement/fishy	Ink	6,0
P1	11	Milky white	Aroma of palm fruit	Ink	6,6
P2	11	Milky white	Aroma of palm fruit	Ink	6,8
P3	11	Milky white	Aroma of palm fruit	Ink	6,8
P4	11	Milky white	Aroma of palm fruit	Ink	6,8
P+	11	Milky white	Odorless/faint	Ink	6,6

The volume of liquid cement produced in all treatments was the same which was 11 mL. This uniformity shows that variations in diluent composition do not affect the final volume because the dilution ratio used is the same in each treatment. These results are in line with the reports of Foeh and Gaina (2017) and Nafi et al. (2021) which stated that the use of natural diluents based on palm fruit water and coconut water did not cause changes in cement volume during the dilution comparison was controlled.

The color of the semen throughout the treatment is milky white, which is a normal characteristic of pig semen with a fairly high concentration of spermatozoa (Klass *et al.,* 2025). No color change towards reddish or greenish indicates the absence of blood or pus contamination. The consistency of diluted semen in all treatments is also in accordance with the natural properties of pig semen which is indeed relatively dilute compared to ruminant semen (Riwu et al., 2022). The color stability and consistency indicate that the combination of palm fruit water, aloe vera gel, and soy juice is compatible with the biological composition of pig semen and does not cause aggregation or clumping.

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The main difference that stood out most in this study lay in the smell. In the P0 (BTS) treatment, the cement showed a typical cement/fishy smell, while in the P1–P4 and P+ treatment, the semen smell changed to a typical aroma of lontar. This change in aroma is a consequence of the addition of palm fruit water that contains volatile aromatic compounds, not an indicator of cement damage (Foeh & Gaina, 2017; Mere et al., 2019). Research by Foeh and Gaina (2017) and Banamtuan et al. (2021) also reported the presence of a distinctive aroma of palm oil in cement diluted with palm juice, but this change did not correlate with a decrease in the quality of motility, viability or pH.

Ethically, changes in smell towards the aroma of fruit are acceptable as long as they are not accompanied by a bad smell that indicates bacterial activity or decay processes (Pork Information Gateway, 2015). In this study, cement with palm dilution did not show a bad odor so it can be concluded that the change in aroma is physiological and does not interfere with the feasibility of cement for IB.

The pH range of cement throughout the treatment was 6.0–7.0. This range still corresponds to the optimal pH to maintain the viability of pig spermatozoa during storage (Foeh & Gaina, 2017; Riwu et al., 2022). Lontar fruit water is known to have a fairly good *buffering* ability because it contains bicarbonate and minerals, thus helping to maintain the pH stability of the diluent (Mere et al., 2019; Dzigbor et al., 2025). In addition, the protein and phospholipid components of soy juice and polysaccharides from aloe vera gel can also contribute to stabilizing pH and protecting spermatozoa membranes against environmental changes (Winarsi et al., 2010; Tethool et al., 2022).

Research by Banamtuan et al. (2021) reported that palm fruit water supplementation in Durasperm diluent did not cause any noticeable pH changes during the 64 hours of storage of Duroc pig semen. Similarly, Nalley et al. (2024) showed that the addition of palm fiber extract in Tris or BTS diluents was able to maintain the pH of boar semen close to neutral. Based on the results of the study, the results of this study show that the water-based diluent of palm fruit has an adequate pH buffer capacity.

The results of this study show that the combination of palm fruit, aloe vera juice, and soybean juice does not cause negative changes to the macroscopic parameters of pig liquid semen. All natural ingredient-based treatments exhibit the same character as BTS controls, so macroscopically they are worthy of consideration as alternative diluents. This is in line with various studies that state that the use of natural fruit and plant-based diluents is able to maintain the quality of pig semen well (Foeh & Gaina, 2017; Nafi et al., 2021; Banamtuan et al., 2021; Klaas,

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2025).

The use of local ingredients such as palm oil and soybeans has the potential to reduce the cost of cement dilution and increase the independence of farmers in the implementation of IB in areas where it is difficult to access commercial diluents. In addition, this approach is in line with the principles of local resource utilization and the sustainability of the livestock system.

However, this study only highlights the macroscopic aspect. To obtain stronger recommendations, it is necessary to conduct further studies on microscopic parameters (motility, viability, abnormality), durability during storage, and *in vivo* fertility tests in incubated females (Banamtuan et al., 2021; Nalley et al., 2024).

CONCLUSIONS AND RECOMMENDATIONS

The addition of lemon juice, aloe vera juice and soy juice as diluting components did not cause negative changes in the macroscopic quality of pig liquid semen. The entire treatment showed a volume of 11 mL, milky white color, dilute consistency and pH of 6.0–7.0 which was in accordance with the standard of semen suitable for artificial insemination. The difference is only seen in the odor parameters, where the treatment containing palm fruit water shows a physiological aroma of palm fruit. Thus, palm fruit water-based thinners have the potential to be developed as a more economical and easily obtainable commercial diluent alternative, especially in the East Nusa Tenggara region.

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