

SPERMATOZOA MOTILITY IN BOER BUCK LIQUID SEMEN WITH ADDITION OF SWEET ORANGE ESSENTIAL OIL AND PENICILLIN

Sukma Aditya Sitepu, Julia Marisa*, Aditiya Amanda Rianto

Agrotechnology Department, Universitas Pembangunan Panca Budi, Indonesia

Corresponding author: juliamarisa@pancabudi.ac.id

ABSTRACT

One of the obstacles to implementing the goat's Artificial Insemination program is the low durability of liquid semen of Boer Goats during storage. An important parameter in assessing the quality of liquid semen is the value of motility. Liquid semen is used with a motility percentage value of at least 40%. This study aimed to determine the durability of Boer Buck semen by knowing the percentage value of motility in the semen added with penicillin and sweet orange essential oil in egg yolk tris diluent. This research hypothesizes that adding sweet orange essential oil and penicillin to tris yolk extender can increase and maintain the percentage of spermatozoa motility in semen during storage. The materials used in this study were fresh Boer Buck semen, tris yolk extender, penicillin, and sweet orange essential oil. The experimental design used in this study was a non-factorial, Completely Randomized Design with five treatments and five replications. The treatment given was the addition of sweet orange essential oil P0 0%, P1 0.25%, P2 0.5%, P3 0.75% and P4 1% which were stored for 0-15 hours. The results showed that at 9 hours of storage, P0 had a motility percentage value of 30%, P1 = 30%, P2 = 35%, P3 = 35% and P4 = 40%. The study concludes that based on the motility value of spermatozoa, liquid semen can be used for up to 9 hours in the P4 treatment because it has a motility value of 40%.

Keywords: Essential Oil, Liquid Semen, Motility, Penicillin, Sweet Orange

INTRODUCTION

One of the livestock commodities that can develop as a meat producer is a goat, but local goats have low body weight, and the population is still limited. Improving the genetic quality of local goats/sheep can be done through the Artificial Insemination program. The Artificial Insemination Program for goats can use liquid semen from good quality goats, such as Boer Goats. Boer goats have several advantages, including high body weight and fast growth. To obtain good quality semen, liquid semen for goats requires proper handling and selection of liquid semen extender.

The growth and contamination of nuisance microorganisms such as bacteria found in liquid semen can cause low semen quality. Bacterial contamination in the liquid semen of goats/sheep comes from the male reproductive tract, environment, handling during the semen dilution process, and diluents, especially egg yolks (Toelihere, 1993). Egg yolks are susceptible to attack by bacteria such as salmonella and staphylococcus (Yuwanta, 2010), and this will affect the resistance of semen that uses egg yolk as a diluent. Reducing the bacteria in frozen semen can be done by adding antibiotics to the diluent.

The bacteria in the liquid semen can reduce motility because it inhibits the killing movement and makes spermatozoa abnormal. Motility is the movement of spermatozoa to reach the egg, so this parameter is the most important in microscopic observations to determine the quality of liquid semen. In addition, the quality of semen knows from the percentage of live spermatozoa (viability). The more sperm that live, the higher the success of fertility. Abnormal spermatozoa are less likely to fertilize an egg. The lower the percentage of abnormalities in semen.

Penicillin works effectively against gram-positive bacteria (Herawati and Irawati, 2014). The sweet orange essential oil contains limonene and linalool, which are toxic to bacteria (Fisher and Phillips, 2006). Research on the use of sweet orange essential oil on the durability of Boer Goat semen has been carried out (Sitepu et al., 2018), and it is proven to be able to maintain the quality of Boer Goat liquid semen for a period of up to 9 hours at room temperature and 48 hours at refrigeration temperature. In this study, the combination treatment of penicillin and sweet orange peel essential oil to inhibit bacterial growth further and improve the quality of Boer Goat liquid semen during storage, including the percentage of motility, viability, and abnormalities of spermatozoa.

LITERATURE REVIEW

The Boer goat is native to South Africa and has been a registered herd for over 65 years. The word "Boer" means farmer. The Boer goat is the only true meat goat in the world because of its fast growth. This goat can reach a market weight of 35-45 kg at the age of five to six months, with an average body weight gain of 0.02 to 0.04 kg per day. Boer goats can be seen by their broad, deep body, white hair, short legs, convex nose, long hanging ears, and reddish brown or light brown to dark brown heads. Some Boer goats have a white stripe down their face (Pamungkas et al., 2009).

Spermatozoa are male gametes produced by the seminiferous tubules of the testes. The size of the head of spermatozoa in goats varies between species but usually is 8 to 10 μ m long, 4 μ m wide, and 1 μ m thick. For fertilization, spermatozoa must have enough energy for movement, proteins, and other essential compounds. At the same time, in the female genital tract, a suitable plasma membrane so that they can fertilize on time (Purdy et al. 2010). The head of the spermatozoa is generally oval, slightly flattened, and has a nucleus containing chromosomes. The acrosome covers the front end of the head, a thin double-membrane sac containing acrosin, hyaluronidase, and other hydrolytic enzymes that play a role in the penetration of the corona radiata and the zona pellucida in the fertilization process. At the same time, the equatorial section acts as a place that initiates the attachment and incorporation of the spermatozoa membrane with the oocyte membrane during the fertilization process (Bearden & Fuquay 2004).

The content in orange peel is essential or etheric oil, and essential oils are the most significant component of vegetable oils. In principle, essential oils have a thick and volatile form at room temperature, giving off a distinctive aroma. Essential oils are often used as perfume seeds for fragrances in the perfume industry. In addition, this distinctive smelling oil can also be processed into cosmetics, pharmaceutical ingredients, and culinary flavors. In the health sector, essential oils have various benefits, including as a relaxation medium, treating stress, as a conventional antibiotic active against microbes such as bacteria, viruses, and fungi (Rochim, 2009).

Continuous use of antibiotics can cause resistance to certain types of bacteria. The emergence of bacteria resistant to one (anti-microbial resistance) or certain types of antibiotics (multiple drug resistance) dramatically complicates the treatment process. Bacterial resistance to antibiotics has several consequences; for example, infectious diseases caused by bacteria that fail to respond to treatment can result in prolonged illness (Utami, 2012).

METHODS

The research parameter observed was the motility of spermatozoa before and after equilibration. Motility is known as the percentage of spermatozoa that move progressively forward. The evaluation is known by observing spermatozoa in eight fields of view with a light microscope with 400 times magnification.

The population and sample in this study were Boer Goat semen which had added egg yolk Trish diluent, penicillin, and various levels of sweet orange essential oil with the following treatments:

- P0 = Penicillin + Sweet Orange Essential Oil 0%
- P1 = Penicillin + Sweet Orange Essential Oil 0.25%
- P2 = Penicillin + Sweet Orange Essential Oil 0.5%
- P3 = Penicillin + Sweet Orange Essential Oil 0.75%
- P4 = Penicillin + Sweet Orange Essential Oil 1%

RESULTS AND DISCUSSION

Research Result

Table 1. The results of the study on the percentage of spermatozoa motility in goat/sheep semen after equilibration during storage at room temperature

Parameter	Treatment	Observation			
		0 Hours	9 Hours	12 Hours	15 Hours
	0%	60±0.00	20±0.00	5±0.00	0±0.00
	0,25%	65±0.00	30±0.00	10±0.00	0±0.00
Motility	0,5%	70±0.00	35±0.00	10±0.00	0±0.00
	0,75%	70±0.00	35±0.00	10±0.00	0±0.00
	1%	75±0.00	40±0.00	10±0.00	0±0.00

Note: Different superscripts in the column showed very significant differences (P<0.01)

The results of the research on sperm motility test of goat/sheep after semen equilibration showed that the lowest percentage value was without treatment (P0), which was 60%, while the highest was with the addition of 1% sweet orange essential oil (P4), which was 75%. The data obtained show that the addition of sweet orange essential oil increased the percentage value of the sperm motility of goats/sheep after equilibration. The higher the administration level of sweet orange essential oil will further increase the value of the percentage of spermatozoa motility.

The percentage value of the motility of goat/sheep spermatozoa at 9 hours of semen storage was lower than that of 0 hours of storage, indicating a decrease in cement quality during the storage process. The highest decrease in the value of the percentage of spermatozoa motility showed in the treatment without the addition of sweet orange essential oil (P0), which was 40%. In comparison, the decrease in the percentage value of the lowest spermatozoa motility showed by adding sweet orange essential oil (P4) 1%, which was 35%.

The analysis of variance showed that the addition of a combination of penicillin with sweet orange peel essential oil as a diluent had a very significant effect (P<0.01) on the motility of spermatozoa after equilibration. The results of the further BNT test showed the highest motility in the P4 treatment, which was 75% after equilibration.

The percentage of spermatozoa motility in goat/sheep semen continued to decrease along with the length of storage at room temperature. The longer the storage time, the lower the percentage of spermatozoa motility. The best results were adding 1% sweet orange essential oil because it had the highest percentage of motility during storage at room temperature.

Discussion

The observation of the motility of fresh semen after it was diluted using tris egg yolk and essential oil and observations after equilibration showed different results for each treatment. The percentage of motility in spermatozoa or the motility of spermatozoa is one of the determinants of the success of spermatozoa in reaching the ovum in the fallopian tube and the simplest way to assess sperm for artificial insemination (Hafez, 1987).

The results showed that the average spermatozoa motility observation in goat/sheep semen met the standards for use as goat/sheep semen for Artificial Insemination because the motility was above 40%, and that figure met the requirements for Artificial Insemination. The requirement for semen for artificial insemination has motility of at least 40% (Evans and Maxwell, 1987).

The best spermatozoa motility that can be used and meet the standards in this study is adding 1% essential oil as a diluent, while the higher the essential oil level given to the diluent, the lower the motility of spermatozoa both after dilution and after equilibration. Cement quality will decrease if storage is not added with the correct diluent (Hafez, 2000). Hydroxynonenal is one of lipid peroxidation which can inhibit glycolysis and motility of spermatozoa. In addition to the damage caused by lipid peroxidation, decreased motility can also occur due to several factors White (1993). According to Toelihere (1993), factors that can reduce motility changes in the pH of the medium, osmotic pressure, and the effects of electrolytes and non-electrolytes. The decrease in spermatozoa motility can be caused by the high use of essential oils in the diluent. Where lipid peroxidation occurs due to the increasing levels of essential oils.

The decreasing percentage of motility was due to reduced food reserves after being stored for four days. Semen contains many compounds needed for the metabolism and movement of spermatozoa. Toelihere (1993) stated that in cow semen, the primary carbohydrate source is fructose, and Spermatozoa will utilize fructose in plasma for their survival and movement. After fructose runs out, GPC (Glyceryl Phosphoryl Choline) will slowly resupply fructose.

Egg yolk contains lecithin, which protects spermatozoa to prevent cold shock, which can cause decreased motility in the preservation process. Toelihere (1993) stated that to reduce spermatozoa cold stress, semen must be added with a protective material before being cooled at 5°C. The protective material for spermatozoa during cold storage is lecithin. Cold stress causes disturbances in motility and metabolism and loss of phospholipids and cations in spermatozoa (Maxwell and Watson, 1996).

The bacteria contained in frozen semen can reduce the quality of semen (Toelihere, 1993), including the value of the percentage of spermatozoa motility. The addition of penicillin antibiotics serves to inhibit the growth of gram-negative bacteria. These Gram-negative bacteria reduce sperm motility through sperm adhesion and agglutination (Wolff et al., 1993; Monga and Roberts, 1994) and cause morphological changes in the plasma membrane and acrosome (Diemer et al., 2000).

CONCLUSION

The results showed that with the addition of penicillin and 1% sweet orange essential oil, the liquid semen could be stored for up to 9 hours and was suitable for artificial goat insemination.

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