International Research Journal of Advanced Engineering and Science
ISSN (Online): 2455-9024

The Effect of Aluminium Foil Packaging and Storage Temperature on Local Sheep Sperm Quality

Setyo Utomo[1], Nur Rasminati[2], Ajat Sudrajat[3]
[1, 2, 3]Department Animal Husbandry, Mercu Buana University of Yogyakarta, Yogyakarta, Indonesia-55752
Corresponding author : [1]+62 821 3845 4463 / Email : esutama_set @ yahoo.com

Abstract**—** *This study was designed to evaluate and produce methods for storing sheep spermatozoa with a refrigerator temperature of 5 °C and -5 °C and liquid nitrogen N2 at -196 °C. The study used sperm from selected males, then diluted and divided it into two treatment treatments, packaging and three storage temperature treatments. The variables observed were motility and the method used was a 2 x 3 factorial completely randomized design with three replications. The data obtained will be analyzed statistically with analysis of variance. The results showed that sperm packed with aluminum foil at a storage temperature of 5 °C had motility of 63.3% and lasted up to ten days, the same thing happened to storage -196 °C, which could last up to ten days with only 23.3% motility. Whereas sperm stored at -5 °C only survived on the third day, namely, the motility was 65%. The analysis also showed an interaction between the packaging model and the storage temperature on the difference in decreasing motility for ten days. The concluded that aluminum foil packaging at 5 °C resulted in 63.3% motility with the lowest daily motility decrease.*

Keywords**—** *Aluminum foil, motility, sheep sperm, storage.*

I. INTRODUCTION

The problems faced in the development of livestock nurseries are the low number of superior livestock breeds, the not optimal breeders' cooperation in providing livestock seeds in the number, type, and quality according to needs (Bahri, 2006). Artificial insemination (AI) activities accompanied by the strict selection of mother sheep support the utilization and improvement of livestock genetic resources. The conservation of genetic resources is carried out by considering the increasing demand for livestock products, and FAO predicts that there will be two-fold demand for meat by 2030 (Subandriyo, 2006).

In the development of national nurseries, the problem faced is how to increase the number of livestock seedlings in the right quantity and quality (Bahri, 2006). Research that has been carried out on etawah crossbreed goat semen at a storage temperature of 3–5°C with Tris diluent with the addition of fructose or lactose in the diluent can improve the quality of liquid cement

(Kostaman et al., 2001). Activity is Spermatozoa influenced by temperature or environmental temperature factors (Utomo, and Rasminati, 2013). The average spermatozoa activity occurs at 37°C and will increase twofold at about 46°C, likewise at low temperatures, and the activity will decrease (Partodihardjo, 1992). This decrease in activity is the basic idea for spermatozoa storage both in the short and long term.

Long-term spermatozoa storage at -196°C uses liquid nitrogen as a refrigerant, and this method has been the guideline for long-term sperm storage activities. The constraints of long-term storage of sperm using liquid nitrogen are the processing technique and the enormous cost required, especially the price of liquid nitrogen, which is very high, so it is not affordable for practitioners such as farmers and breeders.

The function of diluents is to provide nutrients as a source of energy for spermatozoa, protect spermatozoa from cold shock, buffer, maintain isotonic conditions, prevent germ growth and increase volume. Meanwhile, the requirements for sperm thinner are cheap, simple, practical but have high preservation power, contain elements that are almost the same physical and chemical properties as cement, and do not contain toxic substances or are good for the life of spermatozoa and ovum (does not inhibit fertilization), must remain isotonic. The diluent must still ensure ease in testing the quality of semen (Feradis, 2010). A good diluent must have isotonic osmotic pressure and be able to maintain it during storage, provide the mineral balance needed by spermatozoa, provide food for spermatozoa for their metabolic processes, have lipoprotein or lecithin to protect spermatozoa against cold shock, provide buffer, are a source of reducing material to protect cellular enzymes containing sulfhydryl, free from germs. Diluents commonly used for cement dilution include diluent tris, lactose, and milk (Salisbury and Vandemark, 1985).

Aluminum foil has a thickness of about 0.025 mm and is air and watertight. Aluminum foil functions as a protection from light, oxygen (prevents the oxidation of fat from turning rancid), keeps the aroma (odor), moisture, and anti-bacterial. In society, aluminum foil is easy to obtain and cheap. By using aluminum foil as a protector of sperm whose activity is reduced through low temperatures in the refrigerator (refrigerator), the life of the sperm can be extended. Aluminum foil has one side that positively reflects light, and the other side retains heat so that the temperature is stable. Aluminum foil has been successfully used as a wrapper for tubes containing sperm preserved at room temperature in Australia (Pangestu, 2001). This study aimed to obtain a storage model for sheep sperm using aluminum foil packaging at 5^{o}C, -5^{o}C (in the refrigerator), and -196^{o}C in liquid nitrogen.

Setyo Utomo, Nur Rasminati, and Ajat Sudrajat, "The Effect of Aluminium Foil Packaging and Storage Temperature on Local Sheep Sperm Quality," *International Research Journal of Advanced Engineering and Science*, Volume 6, Issue 1, pp. 298-300, 2021.

International Research Journal of Advanced Engineering and Science
ISSN (Online): 2455-9024

II. Materials and Methods

A. Materials and Tools

This study's materials include fresh sperm from rams, Tris diluent egg yolk with glycerol, aluminum foil, penicillin, and streptomycin. The tools include an artificial vagina with equipment, female teasers, laboratory equipment for sperm quality testing, equipment for cooling such as refrigerators, test tubes, mini straws, water baths, cool tops, microscopes, measuring cups, PGF2α, breeders, refrigerators, and a set. artificial insemination.

B. Method

The research used one superior male selected from three tails prepared as a source of sperm, then carried out using an artificial vagina and carried out a sperm quality test before being treated. From the test results, the sperm quality was selected and then diluted using egg yolk tris diluent. Furthermore, the sperm were divided into treatments, namely two packaging models and three storage temperature factors with three units of each replication. The basic design used was a 2x3 factorial, completely randomized design. The data observed were sperm motility (%). Observations were made every day until the motility approached the minimum acceptable limit for artificial insemination, namely 60% for liquid sperm and a minimum of 40% for frozen sperm. Data analysis used analysis of variance (Gill, 1981).

III. Result and Discussion

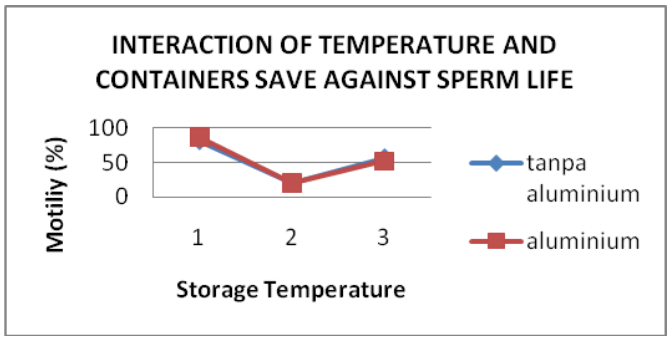
The results showed that sperm packed with aluminum foil at a storage temperature of 5°C until day 10 had motility of 63.3%. At a storage temperature of -196°C, the motility was 23.3%, and a storage temperature of -5 ° C sperm motility only lasted until day 3. possible artificial insemination was 65%, and on the 4th day, the motility decreased drastically to 20%.

Treatment Packaging (S)	Storage Temperature (A)			Average A
	S1	S2	S3	
A1	2:41^{c}	9:07^{a}	5.74^{b}	5.74
A2	2.96^{c}	10.88^{a}	5.93^{b}	6:59
Average S	2.68	9.98	5.84	

At a storage temperature of -196°C, aluminum foil packaging was only able to maintain artificial insemination until the eighth day, which was 41.67%, after which the motility decreased drastically until the tenth day was 23.3%. Aluminum foil can maintain a stable temperature inside the straw to maximize the work glycerol as *cryoprotectant* drop in temperature of 5 °C. Aluminum foil is commonly used to wrap or coat food, drinks, and medicines so that it is protected from the light because it can break down fat, odor, moisture, and bacteria. Aluminum foil has one side that is highly reflective of light, and the other side retains heat so that the temperature is stable. Aluminum foil has been used successfully to wrap tubes containing sperm preserved at room temperature in Australia (Pangestu, 2001).

This study indicates that aluminum foil cannot maintain freezing conditions of -5 °C for more than three days and eight days on liquid nitrogen. This is presumably because the presence of aluminum foil is not able to prevent sperm from freezing or the work of glycerol is not optimal, as well as the storage of -196 °C aluminum foil packaging makes the temperature in liquid nitrogen liquid not reach optimal -196 °C so that the sperm experiences *cold shock*.

In the treatment without aluminum foil, it showed that storage at 5 °C was still able to maintain sperm so that it could be used with motility of 63.3%, but had a higher decrease than sperm packed in aluminum foil, which was 2.96% per day, although it was not statistically significant. However, numerically, aluminum packaging at a storage temperature of 5 °C is better. This is because aluminum foil packaging has a positive effect on the life of the sheep's sperm stored at 5 °C. The positive effect is maintaining humidity and stable storage temperature so that glycerol will work optimally as a *cryoprotectant* against sperm cells (Feradis., 2010).



At -5 °C, storage without aluminum packaging showed that sheep sperm suitable for use with artificial insemination only lasted until the third day, which had 61.67% and 20% motility on the fourth day. This is due to glycerol's inability to protect sperm at freezing -5 °C resulting in *cold shock*. At storage -196 °C, a viable AI motility of 40% was achieved on the 8th day and subsequently decreased until the 10th day with 26.7% motility. This decrease was due to *cold shock* caused by low levels of glycerol so that the work of glycerol was not optimal, so that sperm could not survive at -196 °C. Insistence at storage temperature can be caused by the ram factor or the presence of certain conditions that cause sperm not to withstand freezing in liquid nitrogen (Djanuar et al., 1988).

The diluent function provides nutrients as an energy source for spermatozoa, protects the spermatozoa from cold shock, provides buffer material, maintains isotonic conditions, prevents sperm growth of germs, and increases the volume (Feradis, 2010).

Based on the analysis of variance on changes in daily motility during storage, it showed significant differences ($P < 0.05$) in packaging treatment and storing temperature, and there was a significant interaction ($P < 0.05$) intertreatment factors to the decreased sperm motility (Table 1 and Fig. 1). The highest average decrease in motility occurred at -5° C without aluminum foil which was 10.88% per day, but not significantly different from sperm packaged in aluminum foil at the same temperature, 9.07% per day.

Setyo Utomo, Nur Rasminati, and Ajat Sudrajat, "The Effect of Aluminium Foil Packaging and Storage Temperature on Local Sheep Sperm Quality," *International Research Journal of Advanced Engineering and Science*, Volume 6, Issue 1, pp. 298-300, 2021.

The lowest decrease in motility occurred in sperm packaged with aluminum foil at a temperature of 5° C, which was 2.41% / day better than sperm packaged without aluminum foil which was 2.96% / day. In the storing temperature of -196 ° C, the results of the analysis of variance showed a significant decrease in motility between sperm cells packaged with aluminum foil and without aluminum foil (5.74% / day vs. 5.93% / day). Overall, the results of the analysis of variance also showed that sperm storage at 5°C had the best motility decrease of 2.68%/day, at -196°C was 5.84%/ day, and at -5°C was 9.98%/day.

The interaction between aluminum foil packaging and storing temperature. The highest average decrease in motility occurs at -5°C without aluminum foil (10.88% / day), but not significantly different from the aluminum foil packaging at the same temperature (9.07% / day). The lowest motility decrease occurs at 5° C with aluminum foil packaging and without aluminum foil (2.41 vs. 2.96% / day). Although the statistical tests results show no significant difference, sperm with aluminum foil packaging shows lower motility decreases. This is due to the function of aluminum foil, which can maintain the effects of temperature and humidity in sperm storage (Pangestu, 2001).

The graph also shows a decrease in sperm motility that is getting higher with decreased storage temperature. This is caused by spermatozoa's ability to the cooling effect that varies between types of livestock (Djanuar *et al.*, 1988).

IV. CONCLUSION

The packaging of aluminum foil on sheep sperm storage of 5^{o}C can maintain proper insemination until the ten-day storage duration with the motility of 63.3% with the lowest daily decreased motility level of 2.41%.

ACKNOWLEDGMENT

The author's gratitude goes to the Institute for Research and Community Service (LPPM) the Mercu Buana University of Yogyakarta. The author would also like to convey to the Directorate of Research and Development of the Ministry of Education and Culture of Indonesia to realize this research.

REFERENCES

[1] Bahri, S. "National Breeding Policy Relation to Utilization of Livestock Germplasm". Breeding Director of Dirjennak. Indonesian National Germplasm Conservation and Development Meeting. Faculty of Animal Science, Universitas Gadjah Mada. Yogyakarta. 2016.

[2] Subandriyo. "Conservation of Livestock Genetic Resources through Regional and Community-Based Approaches". Balitnak Indonesian Germplasm National Conservation and Development Meeting. Yogyakarta. Indonesia. 2006.

[3] *Basics of Artificial Insemination in Cattle*. Physiology and Reproduction Laboratory of Universitas Jendral Soedirman. Purwokerto. 1988.

[4] Feradis. 2010. Reproductive Biotechnology in Livestock. *Alfabeta Publisher*. Bandung. Indonesia.

[5] Toelihere, M. . 1985. Artificial Insemination in Livestock. *Angkasa Publisher*. Bandung. Indonesia.

[6] Salisbury, G. W., dan L. N. Van Demark. 1985. Reproductive Physiology and Artificial Insemination in Cows. *Gadjah Mada University Press*. Yogyakarta. Indonesia.

[7] Gill, J. L. 1981. *Design and Analysis of Experiments in Animal and Medical Sciences Vol* 2 : 1. Ames Iowa USA: The Iowa State University Press.

[8] Kostaman, T., P, S., and Sutama, I. K.. The Ability of Life of Etawah Breed Goat Spermatozoa on Various Types of Diluents. *UNSOED National Seminar*. Purwokerto. Indonesia. 2001.

[9] Pangestu, M. 2001. The inventor of simple cement freezing from Indonesia. Kompas. http://www.monash.edu.au/pubs/monmemo/old_archive/issue01-01/

[10] Partodihardjo, S. 1992. Animal Reproduction Science. *Mutiara Publisher*. Bandung. Indonesia.

[11] Salamon, S., and W.M.C., Maxwel. "Frozen Storage of Ram Semen I. Processing, Freezing, Thawing and Fertility after Cervical Insemination". *Anim. Reprod, Sci, 37*, 185–249. 1995.

[12] Utomo, S. "The Influence of the Difference in Place Height on the Results of Artificial Insemination on Ettawa Breed Goats:. *Animal Husbandry Science Journal*, *11*(1), 34–42. 2013.

[13] Utomo, S., dan Rasminati, N. 2012. Application of Applied Artificial Insemination Technology (IB) on Etawa (PE) Goat Goats in Coastal Areas. *Procedings of National Seminar on the Optimization of Research and Service in Building Characterists*. LPPM UNY, 755–764.