

# Effect of Quality Difference Concentration of Tris Yolk on Duck Eggs Spermatozoa Padjadjaran Sheep

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## ABSTRACT

This study aims to determine the effect of different concentrations of Yellow Tris Egg Duck on the quality of sheep spermatozoa. The method used for cement shelter with an artificial vagina is then diluted with Tris Yellow Duck Egg at different concentrations from 10%, 20%, 30%, 40%, 50% and stored at 5 °C. The quality parameters of spermatozoa observed were motility, morphology, and viability of spermatozoa up to 2 days. The design used was a one-way complete randomized design (RAL) and continued with the Tukey-W-Procedure test with SPSS 16. The results showed concentrations of 50% Tris duck egg yolks have high quality spermatozoa with motility 76.67 + 5.164, viability 54.00 + 14.629, and abnormality 8.93 + 7.535.

**Keywords:** sheep, extender, spermatozoa quality

## I. INTRODUCTION

Indonesian people raise livestock as a side business, which is more about savings. Farmers will sell their livestock when they need money, so sheep are the right choice. Caring for sheep has the advantage of short reproduction, easy feeding and adaptability. Many people in West Java choose male sheep as a fighting sheep, so that they have a high economic value. Sheep that win in every competition will be very unfortunate if there is an accident and even death, so there needs to be a method of dilution and storage of sheep sperm so that it can be used for artificial insemination (IB) later.

Artificial Insemination (IB) is one way to improve the genetic quality of livestock, with IB the community can choose the type of male they like and can reduce diseases caused by natural marriage. The success of IB is strongly supported by inseminator skills and also the quality of frozen semen. The process at freezing is highly recommended as a diluent used in the process of diluting fresh semen.

The thinner material that is often used is Tris (hydroxymethyl) aminomethane. Tris as a buffer has the ability to be a good buffer with low toxicity, and eggs contain lipoprotein and lecithin which function to maintain and protect the integrity of the spermatozoa plasma membrane. Eggs are easily available ingredients and affordable prices can be found anywhere, so eggs are used as sperm diluents that are often used.

## II. METHODS

### Time and Place of Research

This practicum was held from November to December 2015, in the Cattle Sheep of Padjadjaran University as a sheep sperm shelter and the Animal Reproduction Laboratory of the Faculty of Animal Husbandry, Padjadjaran University as a place to analyze the quality of spermatozoa microscopically.

This study was conducted with three replications and each replication was analyzed for 2 days.

### Research Material

The material used in this study were two rams which were taken sperm alternately each shelter with a second age of 2 years, duck eggs, 1% NaCl, eosin solution, glucose solution, and aquabides water, tissue, aluminum foil, and label stickers. The tools used are artificial vagina, thermos carrying sperm, light microscope, and biocamera, erythrocyte pipette, haemocytometer, test tube, 0.1 ml, and 1 ml, micropipette, counter, glass object, glass cover, pH meter, refrigerator cupboard, pipette, tissue, aluminum foil, and label sticker

### Research methods

The methods carried out in this study include:

#### Tris Eggs Yolk

Egg yolk which has been separated from egg white, egg yolks are then placed in suction paper until there is absolutely no sticking albumin then move it back in a clean suction paper puncturing the vitellus membrane so that the yolk can come out flowing through suction paper and accommodating in glass cups. Tris solution made from 3.643 gr tris, 0.5 glucose, 1.99 citric acid and added 100 ml aquabides, then stirring until it dissolves and mixes all. Treat with egg yolk concentration 10%, 20%, 30%, 40% and 50%. Repeated 3 times then microscopic analysis for 2 days included motility, morphology, and viability. The results of sperm collection were then analyzed microscopically by motility, concentration, and viability of spermatozoa.

#### Sperm collection

Sheep sperm shelter is used using an artificial vagina and shelter is carried out in the morning at 08.00 WIB 3 times, once a week once.

**Data analysis**

Characteristic sperm value data identified, namely spermatozoa quality parameter data observed were pH, motility, morphology, and viability of spermatozoa up to 2 day analyzed by one-way analysis of variance (ANOVA). If there are significant differences ( $P < 0.05$ ) or very real ( $P < 0.01$ ), then the Tukey-W-Procedure test with SPSS 16.

**Table 1. Average percentage and standard deviation of quality spermatozoa.**

| (%) | pH              | Motility (%)      | Abnormality (%)  | Viability (%)     |
|-----|-----------------|-------------------|------------------|-------------------|
| 10  | $7.00 \pm 0.00$ | $60.83 \pm 16.25$ | $7.33 \pm 6.99$  | $47.00 \pm 10.53$ |
| 20  | $7.00 \pm 0.00$ | $60.00 \pm 13.78$ | $10.25 \pm 8.44$ | $45.83 \pm 11.99$ |
| 30  | $7.00 \pm 0.00$ | $65.00 \pm 13.42$ | $7.83 \pm 8.53$  | $44.03 \pm 12.19$ |
| 40  | $6.67 \pm 0.52$ | $75.83 \pm 4.92$  | $9.58 \pm 8.62$  | $49.17 \pm 13.81$ |
| 50  | $6.67 \pm 0.52$ | $76.67 \pm 5.16$  | $8.93 \pm 7.54$  | $54.00 \pm 14.63$ |

Description: = average and standard deviation Tris-YED (Tris-Yellow Egg Duck)

**pH spermatozoa**

The pH results in sperm with a low dilution concentration of 10%, 20% and 30% have a neutral pH and will decrease further at high concentrations of 40% and 50%, this can be caused by the more energy obtained from egg yolk thinners, the more increases with storage time. One of the factors that influence the decrease in pH is the length of storage of spermatozoa that affects the metabolism used. Spermatozoa stored in a relatively short time such as at the sperm pH stored for 2 days will have an average pH  $6.47 \pm 0.17$  in frozen sperm [1]. This is of course due to the influence of storage temperatures and different days, the longer the sperm is stored, the pH tends to decrease because the metabolism of spermatozoa increases because there is an increase in lactic acid in large amounts in its metabolism. Acidic pH can also be caused by a decrease in metabolic rate (MR) in the spermatozoa while at neutral pH MR will increase. The use of energy is very influential on the speed of metabolism, in anaerobic conditions namely conditions in storage will affect the decrease in pH and lactic acid [2].

**Motility of spermatozoa**

Motility results showed that sperm with Tris-YED thinners had higher motility at all concentration especially at a concentration of 50% ( $76.67 \pm 5.164$ ). this can be due to caloric content, higher protein compared to chicken eggs (race and village), namely 398 kcal vs 361 kcal in egg yolk and 13.1 g vs 12.8 g [3]. With the higher concentration in the thinner with the addition of duck eggs will increase motility because more energy and protein for sperm so that it can maintain storage power at a temperature of  $5^{\circ}\text{C}$ .

**Abnormalities spermatozoa**

Abnormalities are morphological abnormalities experienced by spermatozoa both from primary factors during spermatozoa formation (spermatogenesis) and ripening processes within the epididymis and secondary damage originating from sperm collection or collection and evaluation of sperm. The level of abnormality is an important factor because only normal or intact spermatozoa have a great chance of successful fertilization.

The abnormalities in concentration 10% have a low value , this can occur because the higher the yolk will affect the spermatozoa which is damaged because in the seminal plasma goat or sheep livestock contain phospholipase A

**III. RESULTS AND DISCUSSION****Quality Spermatozoa**

The average percentage of spermatozoa quality between treatments during the study can be seen in Table 1.

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| (%) | pH              | Motility (%)      | Abnormality (%)  | Viability (%)     |
|-----|-----------------|-------------------|------------------|-------------------|
| 10  | $7.00 \pm 0.00$ | $60.83 \pm 16.25$ | $7.33 \pm 6.99$  | $47.00 \pm 10.53$ |
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Description: = average and standard deviation Tris-YED (Tris-Yellow Egg Duck)

enzyme that is responsible for the reduced viability of sperm cells that have been cooled or frozen [4]. Abnormalities of spermatozoa of no more than 20% can still be used for fertilization [5]. Head damage to the postnuclear cap originating from the nuclear membrane is a sign of damage from spermatogenesis. Acrosomal damage to the sperm head which is usually found in Friesian cattle which affects the spermatozoa during ejaculation and makes the livestock sterile. In pigs, head damage will also result in failure of egg fertilization. Damage to the neck can be caused because at hot temperatures or under stressful conditions, the neck can cause damage that causes separation of the head and tail [6]. Separation of the head and tail of the spermatozoa occurs in the epididymal head. More than 60% of the head and tail are cut short 2 to 3  $\mu\text{m}$ . Pseudodroplet is a characteristic of the spermatozoa because the neck diaper is twisted or extended. Abnormalities in the neck or the central part of the spermatozoa of 15 to 50% will cause spermatozoa to die [5].

**Viability spermatozoa**

The results showed that the viability of Tris-EYD ( $54.00 \pm 14.629$ ) at a concentration of 50%, which did not differ greatly from the Miftah study (2008). Average viability of spermatozoa up to a storage time of more than 48 hours using chicken egg yolk at  $51.64 \pm 18.03\%$ , duck egg yolk,  $43\% \pm 15.63\%$  and quail egg yolk  $46.23\% \pm 8.93\%$ . The still high spermatozoa that live because the pH during storage shows no significant changes so that motility also has a good value so the viability of living spermatozoa is also high. The level of abnormalities of spermatozoa is an important factor because with many normal spermatozoa also have a longer viability compared to abnormal spermatozoa and normal spermatozoa have the ability to fertilize before losing motility [5].

**IV. CONCLUSION**

Percentage of 50% Tris- EYD (Tris- Eggs Yolk Duck) have hight quality spermatozoa with motility  $76.67 \pm 5.164$ , viability  $54.00 \pm 14.629$ , and abnormality  $8.93 \pm 7.535$ , because in egg yolk duck contain caloric content, higher protein compared to chicken eggs (race and village) so make value good motility. Abnormality up 50% will cause spermatozoa to die and longer viability compared to abnormal spermatozoa and normal spermatozoa have the ability to fertilize before losing motility.

## REFERENCES

- [1] Kisworo, A.N. 2000. Pengaruh Pencucian dan Suhu Penyimpanan Terhadap Kualitas Sperma Kambing Peranakan Ettawa Yang Diencerkan Dengan Glukosa Sitrat Kuning Telur. Skripsi Universitas Gadjah Mada. Yogyakarta.
- [2] Ismaya, Kustono, Bintara, S., Widayati, D.T. 2008. Teknologi Reproduksi Ternak. Fakultas Peternakan. Universitas Gadjah Mada. Yogyakarta.
- [3] Dinas Kesehatan RI. 2004. Komposisi Gizi per 100g Telur itik dan Telur Ayam.  
<http://suksesdansehat.wordpress.com/2019/11/15/103/>.
- [4] Corteel, J.M. 1981 Collection, processing and artificial insemination of goat semen. In: GALL, C. (Ed). Goat Production. London: Academic Press, p.171-191.
- [5] Hafez, E.S.E. 2000. Reproduction In Farm Animal, 7<sup>th</sup> edition, Lippincott Williams and Wilkins, Philadelphia.
- Evans, G. and W.M.C. Maxwell. 1987. Salomon's Artificial Insemination of Sheep and Goats. Butter Worth. London.