Sperm quality and artificial insemination in small ruminants in Indonesia

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Introduction

The discovery of glyserol as a cryoprotectant marked a quantum advance in semen cryopreservation. However, subsequent research has only made relatively small improvements to the basic techniques established in the early 1950s Holt (2000). Nevertheless, the influence of this technology upon the animal breeding industry has been profound. The remarkable success with bull semen has not been matched in other mammals such in sheep and goat. The survival of frozen-thawed ram sperm is affected by many factors extensively reviewed by Salmon *and* Maxwell (1995). These factors include the basic types and concentration of ingredients used in the semen extenders, the concentration of cryoprotectants, packaging, freezing and thawing rate, as well as the quality of semen used for freezing (El Alami *et al.*, 2001). Futhermore, proper insemination technique is a determining factor in a successful AI program in goat and sheep including semen deposition in the female reproductive tract, insemination dose and accurate timing of insemination. There were a number of research groups in Indonesia have studied sperm physiology and processing to support artificial insemination in small ruminant.

Characteristics of ram semen and spermatozoa

A number of parameters such as semen volume, sperm concentration, motility and viability and percentage of abnormal sperm has been used as standard indicators to evaluate the quality of sperm. Findings from our laboratory has indicated a relatively good average sperm quality of Garut ram and Saanen buck undergo three ejaculation time (Tabel 1).

Table 1. Characteristics of semen and spermatozoa in Garut ram and Saanen buck

Parameter	Garut ram*	[illegible]
Volume (ml)	0.98 ± 0.28	0.81 ± 038
Concentration (10^6/ml)	3224 ± 636	2731 ± 716
Motility (%)	76.67 ± 2.36	68.21 ± 3.72
Viability (% life sperm)	87.33 ± 3.40	80.41 ± 4.24
Abnormality (%)	5.47 ± 1.75	10.38 ± 3.09

* Rizal *a* al (2003) ** Tambing (2003)

It was a variation on sperm quality between ejaculates, and among individual ram and buck. Individual variation was observed on the volume of semen. ranging from 0.5 to 2.5 ml in Garut ram. Variation was also noted in the ejaculation frequency as reported by Rizal et al (2003) who

indicated that the second ejaculates consistently higher than the first and third ejaculates. This finding was in contrast with Jenning and McWeeney (1 976) who reported semen volume, sperm concentration and total sperm number per ejaculate significantly declined in the concequtive ejaculates of ram. Tambing (2004) also reported that one collection per session were always better for all parameters compared to two and three collection per session. These findings was in accordance to Toelihere (1993) who reported the range of sperm volume between 0.8-1.2 ml in Bogor local breed ram. This is slightly higher than in Priangan ram (0.78 ml) and Garut ram (0.76 ml) (Inuonu *et al*, 2001). Pamungkas *et al.* (1996) reported extremely lower volume of semen (ranging 0.3-0.7ml) in fat tail ram.

Concentration of sperm of Garut ram vary between individual animals and breed. It can be breed variation with average of $2839x10^6$/ml in Priangan ram and $3785x10^6$/ml in St Croix ram (Feradis, 1999). These finding was slightly lower in average than European type rams.

The variation was also observed in motility rate and viability of sperm. Rizal *at al.* (2003) reported high motility and viability in Garut ram with average of 77.07% (ranging 75-80%) and 87.89% (ranging 80-91%), respectively. These findings was higher than Inuonu er *al.* (2001) who reported the motility rate of 58.08 in Garut ram and between 66-88% in Priangan ram (Wuwuh, 2000). Toelihere (1993) has indicated that good sperm motility has to bc not lower than 75%, while Langford *et al.* (1989) reported the motility of ram kept in temperate zone at the range of 59.4-70.8%. Good motility rate were also reported by Feradis (1999) in St Croix ram (81.67%), Sutama et al.(2001) in Boer ram (72.50%) and Tambing et al (2001) in Ettawah-cross buck (72.79%). Comparable finding has been reported Perez and Mateos (1996) in Verata ram (74.3-76.2%) and Malaquena ram (74.3-76.2%).

Abnormality rate of sperm collected should not more than 14% (Toelihere, 1993). Rizal et al (2004) has reported 5.1% abnormality of sperm while Feradis (1999) has indicated 8.33% abnormality of sperm of St Croix ram. This finding has indicated that heat stess which may affect sperm quality but not specifically generate sperm abnormality.

There is an absolute of plasma membrane inegrity and acrosome intact necessary for high fertility rate. Rizal et al. (2004) reported the intactness of plasma membrane and acrosome cap of Garut ram at the rate of 87.94% and 86.74, respectively. Similar findings was reported in St Croix sperm (Feradis, 1999) who reported plasma membrane integrity and acrosome intact, 86.33 and 94%. Sutama *et al* (2001) the rate of intactness of plasma membrane and acrosome cap at 83.26 and 78.79%, respectively. According to Revell and Mrode (1994) intactness of plasma membrane should not lower than 60%, otherwise undergo infertile,

Semen dilution and processing

Ram sperm are sensitive to extreme changes in temperature during the freeze-thaw process. Quality of frozen semen of Garut ram can be maintained with the addition of various concentrations of cryoprotectants such as lactose and glycerol and antioxidants e.g. glutathione and P-carotene. Rizal *er ol* (2004) reported that the addition of 60 mM lactose and 5% glycerol in Tris extender is the best combination used in frozen semen of Garut ram. However no interaction between lactose and glycerol was doccumented in quality of sperm after dilution, equilibration, and thawing. On the other hand, addition of 0.05-0.10 % glutathione or 0.002% β-carotene in Tris extender is optimum in improving frozen semen quality of Garut ram. Herdis et al (2005) carried out an observation to see the effect of maltose on the quality of frozen semen of Garut rams. They reported that the addition of maltose 1,2 g / 100 ml extender is the optimum dose. Suyadi et al (2005) also reported the use of Tris based extenders combined with egg-yolk to preserve Boer goat sperm for

chiled. It was also reported the suplementation of cictric acid and lactose into Tris egg-yolk used for sperm in the same breed of goat preserved at 5° C for several days (Isnaini and Suyadi, 2005). Our laboratory had studied the use of glutathione as antioxydant in semen preservation. It was confirmed that the quality of frozen-thawed semen of Garut ram improved by supplementation of 0.05-0.10 g glutathione (Rizal, 2003). This sudy was inspired by the fact that tripeptide glutathiones is one of the best antioxydant for human. This substance plays an important role in the defense against oxydative damage mcl toxin. How is the mechanism in reproducutive tract, particularly in seminal tract, remained unclear.

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