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## Effects of Extenders on Fresh and Freezing Semen of Boer Goat

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### Abstract

The aim of this study was to evaluate the effect of soybean lecithin and egg yolk on semen quality of Boer goats. Semen of the three Boer bucks was collected using artificial vagina. In the first experiment, the semen samples were preserved in liquid state and subsequently were diluted with Tris-fructose-citric acid (TFC), Tris-fructose-citric acid + 1.5% soybean lecithin (TFCSL) and Tris-fructose-citric acid + 2.5% egg yolk (TFCEY). In the second experiment, the semen samples were cryopreserved in various freezing media as the same treatment with experiment 1. The results from the first experiment indicated that sperm motility and viability in TFCEY were significantly greater than those of TFCSL ( $P < 0.05$ ). In the second experiment, there was no difference in post-thawing quality of semen in TFCEY and TFCSL. In conclusion, extender containing 1.5% soybean lecithin is an alternative for the preparation for freezing goat semen.

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## 1. Introduction

Semen cryopreservation is an effective technology for improving goat breeding program. Post-thaw sperm motility and membrane integrity are reduced due to cold shock and osmotic stress during cryopreservation process (Salamon and Maxwell, 2000). Currently, egg yolk is the most common component of most semen preservation extenders to protect spermatozoa from damaging during the freezing–thawing process (Forouzanfar et al., 2010). However, egg yolk has some problems as it increases the risk of microbial contamination which may lead to endotoxin production that can reduce the fertilizing of spermatozoa, and increase the risk of disease transmission in the international exchange of stored semen (Beccaglia et al., 2009). Furthermore, the problem about the extenders containing egg yolk in goat semen has been attributed to egg yolk coagulating enzyme (EYCE) which can be harmful to the sperm cells (Leboeuf et al., 2000; Purdy, 2006). An alternative to replace the components of animal's origin in semen extenders is the soybean lecithin, a natural mixture of phosphatidylcholine and several fatty acids such as stearic, oleic, and palmitic. Such fatty acids are prevailing phospholipids in most of mammalian biological membranes and are known to confer structural stability to cells (Oke et al., 2010). The efficiency of soy lecithin as a primary source of lipoproteins in semen extenders had been proven (Papa et al., 2010). Previous studies also suggested that addition of soybean lecithin (SL) to semen extender improved post thawing sperm motility, viability, acrosome integrity and sperm membrane structure in human (Reed et al., 2009), ram (Sharafi et al., 2009; Forouzanfar et al., 2010), cat (Vick et al., 2010), dog (Kmenta et al., 2011), and goat (Jiménez-Rabadán et al., 2012; Salmani et al., 2014). The aim of the present research was to study the effect of three different extenders with washing seminal plasma on the *in vitro* survival of Boer goat spermatozoa.

## 2. Materials and Methods

### 2.1 Experimental animals, Semen collection and handling

Three sexually mature Boer goats aged 12–18 months, (body weighed over 50.0 kg and 2.5 body condition score, scale of 1–5, with 1 being emaciated and 5 being grossly over condition) were used in the study. All goats were healthy and clinically freed of internal and external parasites. The animals were kept under natural photoperiod and balanced nutritional status (the concentrate diet contains 21% Crude Protein and 75% Total Digestible Nutrients). The rations offered to goats adjusted to meet the requirements according to NRC (1981). The goats also received mineral block and fresh water during the whole experimental period.

Semen samples were collected twice a week using artificial vagina after stimulating with an oestrus doe. The bucks were exposed to teaser for a period of 5 min. Libido was recorded at a scale of 0–3, with 0 being the complete absence of sexual desire and 3 being the highest level of sexual desire giving minimal time to jump over the teaser animal (Qureshi et al, 2012). After collection, semen was placed in a water bath at 37°C and transferred to the laboratory for semen evaluation in 15 min. Ejaculates were evaluated for volume (ml), colour, pH, density (D–4D), mass movement (0–5), sperm motility (%), sperm abnormality (%) and sperm viability (%) using eosin–nigrosin staining and sperm concentration ( $n \times 10^9$  sperm/ml) by haemocytometer. Only ejaculates between 1.0 and 2.0 ml volume with a concentration of greater than  $2.5 \times 10^9$  sperm/ml having >75% sperm motile and >85% normality sperm were selected and pooled for cryopreservation (Salmani et al., 2014).

### 2.2 Semen extenders

The basic extender as Tris fructose citric glycerol; TFC (control treatment) used in this study was composed of 2.4 g Tris, 1 g fructose, 1.4 g citric acid, 1.4% glycerol (v/v), 100 mg/ml Streptomycin and 100 µg/ml penicillin G, respectively (Qureshi et al, 2012). The osmotic pressure and pH were set at 420 mOsm/kg and 6.8, respectively. Two different extenders (TFC + 1.5% soybean lecithin; TFCSL and TFC + 2.5% egg yolk; TFCEY) were prepared by the addition of 1.5%, soybean lecithin and 2.5% chicken egg yolk (Table 1).

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