### Goat semen cryopreservation

Semen is collected from bucks by electroejaculation or artificial vagina and the sample is observed to make sure it is free of urine. The sperm motility, volume, and concentration are determined and the semen is diluted in one step using Evans and Maxwell Tris-egg yolk-glycerol diluent (37 °C) to 400 x 10<sup>6</sup> sperm/mL (Evans and Maxwell, 1987). Next, the samples are loaded into straws (0.25 or 0.5 mL), sealed, and cooled to 5 °C over 2.0 hours. The samples are then frozen in liquid nitrogen vapor (4 cm above liquid nitrogen) for 7 min and plunged into the liquid nitrogen for storage.

### Thawing procedure:

Frozen samples are thawed in a 37 °C water bath for 30 sec.

## Optional seminal plasma removal:

Samples can be collected and held for 24 hours at 5 °C prior to cryopreservation using this diluent described above because the egg yolk concentration is low enough that coagulation should not occur (Mook and Wildeus, 2008). If this is a concern, as may be the situation with bucks which are know reactors to egg yolk, the semen sample can be washed using the same diluent (800 x g for 10 min), but without egg yolk and glycerol, to remove the seminal plasma. After removal of the seminal plasma, the sample can then be cryopreserved using the methodology described above and the diluent containing egg yolk and glycerol.

# Tris-egg yolk-glycerol diluent

Tris	3.62g
Fructose	0.50g
Citric Acid	1.99g
Penicillin G	0.006g
Streptomycin.sulfate	0.100g

Egg yolk 2.5% by volume Glycerol 2.0% by volume

Adjust pH to 7.0 prior to bringing to final volume of 100 ml

### References:

Evans, G., Maxwell, W.M.C., 1987. Salamon's artificial insemination of sheep and goats, Butterworths, Wellington, New Zealand.

Mook, J.L. Wildeus, S. 2008. Effect of egg yolk level, washing and extended pre-freeze equilibration on postthaw motility of buck semen. Southern Section American Society of Animal Science Annual Meeting. Dallas, TX.