

A Preliminary Study for Sperm Sexing by Using Sucrose Density

Gradients in Jersey Bull at Artificial Insemination Centre at

Thirunelvely (Northern Province of Sri Lanka)

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Abstract

Sperm sexing rouses great interest due to extensive application in animal production and new separation techniques which present both better accuracy and low costs are necessary. Thus the present study was experimented to separate X and Y-bearing bovine sperm using density gradient. To prepare discontinuous sucrose density gradient, sucrose solutions 35%, 30%, 25%, 20% and 15% were layered upon one another respectively into the eppendorf tube. Finally 20 μ L semen sample was loaded on the top layer. Then it was centrifuged at 500 x g for 12 minutes at room temperature. After elution of fractions and centrifugation (at 700 x g for 5 minutes), sperm was used for quality control by 0.4% Trypan Blue and 0.75% Giemsa stain. Other part of the pellet was stained with 2% orcein red or 1% eosin for 30 minutes to establish karyotypes. The percentages of female sperms were counted at different layers and statistically compared with the Paired-T test. Results have shown that means of Percentage of X chromosomes increased from top layer (27.01 \pm 7.501 %) to bottom layer (36.93 \pm 3.316 %). But the difference are not statistically significant (P>0.05). However it needs to perform further studies to obtain appropriate density gradient model. Our preliminary study demonstrated that the discontinuous sucrose density gradients can be considered as low cost tool for sperm sexing of bovine semen.