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Poster Presentations

P-23

Evaluation the effects of vitamin D supplementation of the extender on sperm quality after freeze-thaw process in normozoospermic and asthenozoospermic Holstein bulls

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Background: Asthenozoopermia is a usual male infertility factor, characterized by decreased sperm motility. There is evidence that vitamin D regulates widespread biological function.

Objective: This study aimed to evaluate the effects of vitamin D on sperm kinematics and apoptosis in normozoospermic and asthenozoospermic bulls' semen after the freeze-thaw process.

Materials and Methods: The effect of vitamin D on sperm quality factors such as sperm kinematic, sperm plasma integrity, acrosomal membrane integrity, reactive oxygen species (ROS) and apoptosis statues following freezing and thawing process in asthenozoospermic bulls were examined. For this purpose, 32 semen samples of four Holstein bulls (normozoospermic, progressive motility > 70%) and 32 semen samples of four bulls (asthenozoospermic progressive motility < 40%) were collected. Then, the poll semen samples of each group (normozoospermic and asthenozoospermic) were diluted into four equal aliquots of extender containing different vitamin D concentrations (0, 5, 10, and 50 ng/mL) and aspirated into 0.5 mL straw. Semen straws were frozen in liquid nitrogen. After thawing, sperm kinematics parameters, viability, plasma membrane integrity, acrosome integrity, apoptosis statues, and ROS production levels were evaluated.

Results: The percentage of sperm motility and viability were significantly higher in 50 ng/mL of vitamin D in both groups ($p < 0.05$). Normozoospermic bull semen samples had significantly higher curvilinear and average path velocity levels in 50 ng/mL vitamin D groups compared to the control group ($p < 0.05$). However, no significant differences were observed in post-thaw sperm kinematics parameters in asthenozoopsrmic samples. No significant differences were identified in membrane integrity and acrosome integrity in both normozoospermic and asthenozoospermic samples. The percentage of early-apoptosis ($p = 0.049$) and late-apoptosis ($p = 0.005$) in the asthenozoospermic group were significantly higher than the normozoospermic group. Generally, in the asthenozoospermic group, the level of ROS production was significantly higher ($p = 0.049$) compared to the normozoospermic samples.

Conclusion: According to our results, it can be concluded that the vitamin D supplementation of the asthenozoospermic semen extender had no significant effect on the quality of semen after the freeze-thaw process.

Key words: *Vitamin D, Apoptosis, Sperm kinematic, Asthenozoospermic, Normozoospermic.*