ABSTRACT

In order to investigate the effect of feeding pomegranate seed oil on stallion sperm motion characteristics, viability, membrane integrity, acrosome status and mitochondrial activity and embryos recovery percentage of Arabian mares, two experiments were conducted. In the first experiment, the stallions were fed standard in control groups (n = 4) and the pomegranate seed oil group (n = 4) received daily 200mg of pomegranate oil in addition to the standard diet. Sperm collection was performed on days 0 before feeding and 15, 30, 45, 60, 75 and 90 days after the start of dietary intake. After 90 days, Stallions were reversed across the treatments after a 60-day interval, to remove the possible pomegranate oil residual effects on spermatogenesis in a crossover design, animals were switched across the treatments. Sperm was collected again every 15 days during 90 days. Gel free volume, concentration and abnormal sperm and percentage of live sperm in fresh conditions, and kinetic, viability, membrane integrity, morphology and lipid peroxidation (MDA) of sperm cooled at 5°C for 2, 12 And 24 hours were evaluated. In freezing conditions, frozen-thawed sperm, sperm motility and sperm dynamics using CASA software, acrosome status and mitochondrial activity (using flow cytometry) were determined. To evaluate the fertility rate, 126 mares were inseminated using sperm of two groups after 60 days of experiment. The results showed that the semen volume, sperm concentration, abnormality and live sperm was not affected by dietary treatment ($p<0.05$). The plasma membrane integrity and sperm viability parameters after 24 hours of storage in cold conditions at 5°C showed a significant difference in pomegranate seed oil group compared to control group. The total motility and acrosome status in post-frozen-thawed conditions were significantly different in the pomegranate oil group compared to the control group ($p <0.05$). Percentage of pregnancy rates in the control and treatment groups did not show a significant difference (62.88% and 65.99%) ($p<0.05$). A second experiment was conducted to investigate the effect of feeding pomegranate seed oil before pregnancy and its effect on follicle growth and diameter, ovulation number, uterine condition, endometrial and cervix status using a rectal ultrasound device at the onset of estrus on a daily basis and embryo recovery rates of Arabian mares (n = 12) was performed in 4 cycle. Mares were fed in standard (n = 6) control groups (n = 6) and pomegranate seed oil group (n = 6) received standard 100 ml pomegranate seed oil per day. The use of pomegranate seed oil was performed 20 days before the start of the experiment and during the study (8 weeks). Mares with follicles 35 mm or more were inoculated using artificial insemination technique and embryo collection was performed 7 days after ovulation. The embryos were transferred to the laboratory for qualitative grading. Based on the results of this study, pomegranate seed oil treatment did not affect the rate of embryo collection (follicle growth and diameter, ovulation number, uterine condition, endometrial and cervix status) pomegranate seed oil, 61/76% and control, 60%) in Arabian mares, and a significant difference in measured parameters (in control and Pomegranate oil groups was not observed.

Keywords: Pomegranate seed oil, cryopreservation, Embryo, Spermatozoa, Mitochondrial activity, Punicic acid.

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