

ADDITION EFFECT OF PINEAPPLE, ORANGE AND BEET JUICES ON THE MITOCHONDRIAL MEMBRANE POTENTIAL OF RAM SPERM

EFEITO DA ADIÇÃO DE SUCOS DE ABACAXI, LARANJA E BETERRABA NO POTENCIAL DE MEMBRANA MITOCONDRIAL DE ESPERMATOZOIDE DE CARNEIRO

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Sperm cells are specialized for the transport of gametes, requiring a great amount of energy to move about, and mitochondrial membrane potential is an important parameter when evaluating mitochondrial activity in a cell. Plants have thousands of phytochemical compounds in their roots, leaves, fruits, flowers and stems, which have several biological effects, such as anti-oxidant, anti-inflammatory, anti-carcinogenic and anti-inflammatory action. The objective of this work was to evaluate the effects of adding pineapple, beetroot, and orange juices individually and in different combinations at concentration of 10% in the cryopreserved semen extender of rams, through the mitochondrial membrane potential of sperm cells. At the Institute of Zootechnics in Nova Odessa, SP, five semen collections from five rams were performed during a two-week period with the use of an artificial vagina. After collection, the semen was analyzed with a microscope following the recommendations and standards of sperm kinetics according to the CBRA. On each day, after the collection and analysis of the semen of all rams, a pool was formed to minimize the influence of individual traits of the animals. The sperm concentration was corrected to 800.10^6 spz / ml with addition of the BotuBov® semen extender and subsequently the juices, after the fruits and beetroots were washed with distilled water, peeled, cut into pieces and squeezed in nylon mesh, were added to the semen extender, obtaining the treatments: pineapple (P), beetroot (B), orange (O), pineapple and orange (P + O), pineapple and beetroot (P + B), beetroot and orange (B + O) and pineapple, beetroot and orange (P + B + O), at a concentration of 10 %, and the control group. The samples were packed in 0.25 ml straws with a concentration of 100.10^6 spz per straw, submitted to cryopreservation with a negative curve of -0.25 °C and stored in liquid nitrogen for further analysis. The analyses were performed at USP Pirassununga and the Accuri® flow cytometry system was used to measure low mitochondrial membrane potential (LMP) and high mitochondrial membrane potential (HMP) through a JC-1 fluorescence probe. In the LMP parameter, treatments did not differ ($p > 0.05$), with values of $27.31\% \pm 19.36$, $21.62\% \pm 10.55$, $43.43\% \pm 21.6$, $31.72\% \pm 29.23$, $19.73\% \pm 5.05$, $31.34\% \pm 24.37$, $33.67\% \pm 23.94$ and $21.78\% \pm 7.25$ for control group, P, B, O, P + B, P + O, B + O and P + B + O respectively. In the HMP parameter, the P + B + O treatment caused the highest percentage of high mitochondrial membrane potential, with a value of $21.15\% \pm 9.89$, greater ($p < 0.05$) than the control group, B, O and B + O, with values of $9.23\% \pm 1.34$, $7.38\% \pm 2.24$, $9.81\% \pm 2.89$ and $9.06\% \pm 2.29$, respectively, which did not differ from each other ($p > 0.05$). Interestingly, the P + B + O treatment did not differ ($p > 0.05$) from treatments P, P + B and P + L, with values of $14.73\% \pm 6.19$, $17.94\% \pm 9.32$ and $14.74\% \pm 4.56$, respectively, which did not present differences ($p > 0.05$) from the other treatments, except for B, which was lower than P + B. The addition of pineapple juice in the different treatments culminated in a gain in the percentage of sperm with high mitochondrial membrane potential, showing a possible agonist effect of compounds present in this fruit.

Keywords: phytochemicals, mitochondria, semen.