The sperm-egg interactions are species-specific forms of cell recognition and the binding event which is a necessary prerequisite for fertilization (Park et al. 2002 Anim. Reprod. Sci. 72, 83–94). Glycosidase enzymes that remove carbohydrates could play an important role in the reproductive tract, modulating decisive physiological events mediated by carbohydrates, which play a key role in sperm–oocyte recognition. The aim of this study was to analyze the presence of the glycosidases α-D-mannosidase, α-L-fucosidase, β-D-glucosaminidase, and β-D-galactosaminidase in intact and acrosome-reacted sperm from fertile matured boars. Sperm were washed three times in PBS by centrifugation at 800 × g for 10 min. The pelleted sperm were resuspended in the same buffer to obtain a final concentration of 250 × 10^9 spermatozoa mL⁻¹. The acrosome reaction was induced by incubation of the sperm with 10 μM of calcium ionophore A23187 at 37°C for 30 min. Different enzymes were detected by incubating 8 μL (for α-D-mannosidase) or 80 μL (for the rest of the enzymes) of sperm sample with the corresponding substrate conjugated to 4-methylumbelliferone for 2 h at 37°C in PBS at pH 7.3. Fluorescences were read on a Fluostar Galaxy fluorimeter (BMG LabTech GmbH, Offenburg, Germany), using wavelengths of 340 and 450 nm for excitation and emission, respectively, and were corrected by subtracting tissue and substrate blanks. The results were analyzed using a one way ANOVA. An average of fluorescence units of 9685.86 ± 1018.75, 7394.63 ± 874.29, 3154.17 ± 514.10, and 1666.40 ± 117.86 was detected in the intact sperm sample for the α-D-mannosidase, α-L-fucosidase, β-D-glucosaminidase, and β-D-galactosaminidase, respectively. For the acrosome-reacted sperm sample (60–65% acrosome-reacted sperm in the samples measured by fluorescence microscope), an average of fluorescence units of 9756.14 ± 1011.45, 7026.93 ± 771.48, 1185.70 ± 277.51, and 1111.60 ± 176.70 for α-D-mannosidase, α-L-fucosidase, β-D-glucosaminidase, and β-D-galactosaminidase, respectively. Statistically significant differences (P < 0.05) between intact and acrosome-reacted sperm were detected only for the β-D-glucosaminidase and β-D-galactosaminidase. These results suggest that the four different enzymes detected are mainly present in the sperm plasma membrane. Under the conditions used in this study, α-D-mannosidase is the main enzyme activity present in the sperm. Importantly, β-D-glucosaminidase and β-D-galactosaminidase activity detected in the intact sperm is decreased after the induction of the acrosome reaction.

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224 A COMPARATIVE STUDY BETWEEN WOOD AND PLAINS BISON


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In Canada, brucellosis and tuberculosis threaten an estimated 4500 wood bison (Bison bison athabascae), a species considered at risk by the Committee on the Status of Endangered Wildlife In Canada (COSEWIC). To help rescue this species, our Wood Bison Reproductive Research group proposes to employ advanced reproductive technologies. Unfortunately, little is known about the reproductive physiology of the wood bison, which hinders the application of these reproductive technologies. In order to modify advanced reproductive techniques developed in cattle for use in wood bison, the large amounts of semen, embryos, and oocytes from wood bison required are not available. The purpose of this study was to compare semen collected from the more abundant and closely related plains bison (Bison bison bison) with that of wood bison. Semen from 3 wood and 11 plains bison were collected and evaluated for fertilization potential in vitro. The results indicated that semen from plains bison has a higher percentage of normal sperm than wood bison, which could be due to the larger amount of semen produced in plains bison. These findings suggest that further research is needed to optimize the use of wood bison semen in assisted reproductive technologies.