Sorting sperm by sex chromosome

Abstract

The ability to select the sex of offspring is highly desirable in a number of domestic mammals. One way of achieving this is by sorting sperm on the basis of their sex chromosome before fertilisation occurs. There are several different methods for achieving this, and this project looks at a technology to sort ram sperm by activating TLR7/8 receptors, which are only found in sex cells with an X chromosome. Activation of these receptors reduces the motility of X-containing sperm and they sink to the bottom of the test tube, making it possible to physically separate them from Y-chromosome sperm. The aim of the project is to test this method on ram sperm and to verify the separation of cells by RT-PCR (real-time PCR).

Cells from 4 different animals were used for the experiment. After evaluating their motility and morphology and obtaining a spermogram, they were mixed with a solution of the ligand R848, a TLR7/8 receptor agonist. After separation of the two fractions, their purity was assessed by RT-PCR. This procedure allowed to determine the ratio of X and Y chromosomes in the obtained fractions. After sorting, the motility and morphology of the cells were again measured to ensure that they were capable of fertilization.

Spermogram results showed good cell parameters both before and after sorting. Isolation of Y-containing spermatozoa was more successful, with fraction purity averaging 77%. When isolating X-containing spermatozoa, the average purity was about 64%. To improve the process, it is possible to re-mix each of the samples with R848 and divide again into two fractions, which will provide an even higher proportion of sperm with the desired chromosome.