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JOURNAL ARTICLE

Column separation of motile sperm from stallion semen

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Subfertility in stallions is common, and methodologies are needed to increase the fertility in these animals. In other species, removal of the dead sperm from semen increases the quality and fertility of semen. With horse semen we evaluated 48 combinations of column separation techniques using micro-spin chromatography columns. The greatest improvement in motility was observed with glass wool, whereas glass beads exhibited the greatest recovery of motile sperm. Although centrifugation time did not influence recovery rate or percent motility, a column length of 2 cm was superior for recovery of motile sperm. In scale-up experiments using 2-cm columns of glass beads in 50-ml syringe barrels, centrifugation proved to be superior to gravity flow, suction, and syringe plunger as extraction methods for drawing semen through the column; however, gravity flow produced acceptable results and may be more suitable for use in a field setting. When the volume of semen for separation was increased from 10 ml to 20 ml, the recovery rate of motile sperm was also increased. Further increasing the volume of semen for separation did not improve the recovery rate, and for volumes greater than 50 ml the column had a tendency to "clog." Thus, a suitable method for column separation of equine sperm utilizes a 2-cm column of glass beads in a 50-ml syringe casing. Centrifugation is the ideal extraction method; however, gravity flow is an acceptable extraction method suited to the field setting, using a maximum semen volume of 50 ml.

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