Increase of ICSI Efficacy with Hyaluronic Acid-Binding Sperm for Low Aneuploidy Frequency in Pig

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Hyaluronic acid (HA)-binding sites have been shown the diagnostic potential for assessment of sperm maturity, which is related to male fertility. This study was designed to evaluate chromosomal patterns in porcine embryos produced by in vitro fertilization (IVF) and intracytoplasmic sperm injection (ICSI) with non- or HA-binding sperm (HABS). For binding of sperm with HA, sperm incubated in 10 μℓ drop containing HA (0.8 mg/ml)-agarose (0.8%) mixture for 15 min. IVF and ICSI with non- or HA-bound sperm examined with matured oocytes at 44 hr after in vitro maturation. Embryos were cultured in 50 μl of NCSU 23 containing 0.5% BSA for 5 days and then in 50 $\mu\ell$ of NCSU 23 containing 10% FBS for 2 days. For the evaluation of chromosomal aneuploidies, chromosome 1 sub-metacentric specific probe was used in sperm and embryos by fluorescence in situ hybridization (FISH). The frequency of an euploidy sperm for chromosome 1 was 6.25%. The significant differences following IVF and ICSI with non- or HA-bound sperm were not observed in blastocyst formation rates (18.6, 23.5, and 23.8%) and cell number (61.8 \pm 12.5, 55.5 \pm 7.3, and 59.3 ± 9.6). Moreover, the percentage of diploidy in 4-cell stage embryos was 57.1% (IVF), 68.8% (ICSI), and 76.3% (ICSI-HABS). These results suggest that HA-binding sites may be a material for selection of normal sperm for ICSI. Therefore HA selection of normal sperm may be reduce the loss to embryonic mortality prior to embryo transfer in pig.

Key words) Hyaluronic acid, Sperm, ICSI, FISH, Pig