

# Establishment and Application of the Intracytoplasmic Sperm Injection in Pigs

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Recently, pigs have been categorized as domesticated livestock but now expected also to be large experimental animals, where many pig strains have been established. Simultaneously, the importance of technology development of preserving the germplasm from genetic resources that have useful economic traits and show genetic diversity increases because they are now exposed to the menace of animal disease.

Intracytoplasmic sperm injection (ICSI) of a non-motile sperm into the ooplasm for assisted fertilization is a highly-specialized procedure for producing the next generation. This assisted reproductive technology is very beneficial also for pigs because polyspermy often occurs in pig *in vitro* fertilization, and the frozen and stored boar sperm sometimes show great loss of motility or immotility after thawing, depending on the individual from which the spermatozoa were collected. We have already demonstrated that oocytes injected with sperm and transferred to recipients could develop into viable piglets [1]. However, the efficiency of successful fertilization and embryonic development was too low. Therefore, we have tried to make clear of the cause of the failure in fertilization and embryonic development after ICSI in pigs. We have already demonstrated that a key factor in success of ICSI is not involved in the sperm membrane and/or nuclear status, but in the state of oocyte activation [2].

Furthermore, we also have tried to develop new methods for preservation and utilization of male genetic resource such as freeze-drying of boar sperm and producing of boar sperm from ectopic xenografted testis tissues. We then have demonstrated clearly that the oocytes injected with freeze-dried sperm or xenogeneic testicular sperm show the competence to grow to fetus or offspring [3], respectively. These results of our will contribute to preserve and utilize efficiently of male genetic resource in pigs.

The above knowledge can be applied in other large domestic animals, such as cattle and horses. Therefore, our research will support the preservation and application the genetic resources, and further contribute to the development of livestock industry or bioindustry in Japan.

## References

- [1] Nakai M., Kashiwazaki N., Takizawa A., Hayashi Y., Nakatsukasa E., Fuchimoto D., Noguchi J., Kaneko H. and Kikuchi K.: Biol Reprod 68: 1003-1008 (2003).
- [2] Nakai M., Kashiwazaki N., Takizawa A., Maedomari N., Ozawa M., Noguchi J., Kaneko H., Shino M. and Kikuchi K.: Reproduction 131: 603-611 (2006).
- [3] Nakai M., Kaneko H., Somfai T., Maedomari N., Ozawa M., Noguchi J., Ito J., Kashiwazaki N., and Kikuchi K.: Reproduction 139: 331-335 (2010).