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DNA uptake by embryo could be one of the causes for creation transgenic animals through SMGT

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Abstract

The uses of live sperm as carrier of the transgene into oocytes were idea that was predicted to be able to make a great development in transgenesis. However, due to its very low reproducibility, heretofore this technique has not been confirmed in scientific society for transgenesis and discusses about it continues. The aim of this study was assessment of chicken embryo ability in DNA uptake as one of the causes of creation transgenic animals in sperm mediated gene transfer (so-called SMGT) and following injection of transgene in the vicinity of embryonic cells. Four treatments including: 2µg pDB2 plasmid mixed with 3µl 0.5 M EDTA (PE); 2µg pDB2 plasmid Combined with Lipofectamine 2000 at 1 to 3 ratio (PL); 2µg pDB2 plasmid Combined with Lipofectamine 2000 at 1 to 3 ratio at presence of 3µl 0.5 M EDTA (PLE) and depc water as a control group (C) were considered. Injection was performed in the vicinity of embryo by opening a window with 10–15 mm at the sharp edge of egg shell in the stage X. In each group, 50 fertilized eggs were used. During the incubation period, tissue samples from different parts of the embryo collected and after examining embryos under the light green fluorescent, DNA extracted and PCR was performed for pDB2 transgene. Results obtained from light green fluorescent and PCR were positive for embryos that belonging to PLE group. In conclusion, transgene could be uptake by chicken embryo under physiological condition and incubation of only non-viral vector is insufficient for in vivo transgenesis of chicken embryo.

Keywords: Transgenesis, Chicken embryo, DNA uptake, SMGT.