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Progress of Inter-species Somatic Cell Nuclear Transfer in Bovidae and Felidae Family

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It has been shown that advanced reproductive biotechnologies such as artificial insemination (AI), *in vitro* fertilization (IVF) and embryo transfer (ET) are the important tools for conservation of endangered species. Since the birth of first cloned mammalian animal using somatic cell nuclear transfer (SCNT) was announced in 1997, several live cloned domestic, laboratory and companion species have been produced using homologous oocytes. In endangered species, there are extremely lack of homologous oocytes and recipients as perform in traditional SCNT. Interspecies SCNT (iSCNT) is an alternative technique for producing embryos and offspring of endangered species. The iSCNT procedure need the oocytes of animals which are the same genus of individual that we want to propagate. The advanced of iSCNT already demonstrated in bovidae and felidae families. Gaur (*Bos gaurus*) and banteng (*Bos javanicus*) are member of the bovidae family and seriously threatened.In iSCNT procedure, we can use oocytes from domestic cattle (*Bos taurus* and *Bos indicus*) reconstruct with karyoplasts of gaur or banteng to produce iSCNT embryos. The potential offspring will be born after transfer embryos to uterine horn of domestic cattle. To date, only one live offspring iSCNT gaur has previous been reported, unfortunately, the calf died two days after birth. Additionally, two live calves iSCNT banteng were born and only one still survive. Recently, we produced live birth iSCNT gaur calf, unfortunately, it died 12 h after birth. In case of endangered felids, the birth of iSCNT African Wildcat (AWC, *Felis silvestris lybrica*) kittens has been reported using domestic cat (*Felis silvestris catus*) oocytes and recipients in the processes. It has been exiting proven that male and female iSCNT AWC could naturally mate and deliver normal kittens. This is one of the demonstration that iSCNT could save and propagate endangered felids. Beside AWC, our group and other scientists are trying to produce iSCNT felid embryos and offspring including leopard cat (*Prionailurus benga-*

lensis), black-footed cat (*Felis nigripes*), flat headed cat (*Prionailurus planiceps*), rusty spotted cat (*Prionailurus rubiginosus*) and marbled cat (*Pardofelis marmorata*). So far the success rate of embryo development and implantation remainlow and live offspring in these felid species has not yet been reported. We had been produced iSCNT marbled cat embryos using domestic cat cytoplasts but could not get embryos develop beyond morula stage. From our studied, the transcription level of Oct4, DNMT1, DNMT3a, DNMT3b, HAT1 and HDAC1 genes were found to be aberrant in the iSCNT marbled cat embryos which may lead to the failure of further development and incomplete reprogramming of marbled cat nuclei in the domestic cat cytoplast. The cloned domestic and leopard cats after their reconstruction with domestic cat cytoplasts were able to develop to the blastocyst stage, and their development rate and cell number were similar to the IVF derived embryos. Although cloned domestic and leopard cat embryos have normal developmental rates, the transcription level of pluripotent genes showed a similar pattern to the IVF embryos but the cloned embryos showed the higher level of DNA passive demethylation and de novo methylation and lower level of histone acetylation transcripts.

Even though live offspring iSCNT animals have been produced in bovidae and felidae families, there are numerous technical and biological factors affecting the success of SCNT. The low frequency of successful has been associated with abnormal nuclear and epigenetic reprogramming, mitochondrial heteroplasmy as well as incompatibilities between the nucleus and cytoplasm that could possibility impair embryo development, high abortion rate, anatomical and physiological abnormalities of fetus, offspring died immediately before and after birth. Although iSCNT has potential application for conservation of endangered species, numerous factors which involve in low efficiency of embryo production and survival of live birth need to be examined. This work was supported by National Centerfor Genetic Engineering and Biotechnology (BIOTEC) and Suranaree University of Technology.