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# Disruption of Cardiac $Na^+$-$Ca^{2+}$ Exchanger Gene in Mice

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$Na^+$-$Ca^{2+}$ Exchanger is known to play a critical role in the regulation of intracellular $Ca^{2+}$ in many tissues and cells. In heart, the $Na^+$-$Ca^{2+}$ exchange is the principal $Ca^{2+}$ extrusion mechanism and affects cardiac excitation-contraction coupling. To understand the functional role of cardiac $Na^+$-$Ca^{2+}$ exchanger (NCX1) *in vivo*, we tried to ablate the cardiac $Na^+$-$Ca^{2+}$ exchanger gene locus by the use of the gene targeting technologies.

The targeting vector was constructed to disrupt the first cording exon of NCX1 gene. For gene disruption and positive selection, neomycin resistant gene (*neo*) was inserted into the first cording exon of the NCX1 gene and viral thymidine kinase gene (*tk*) was inserted at the end of the 3′ homology region as a negative selection marker. The disruption of exon 2 should inactivate the NCX1 gene. The resultant targeting vector was transfected into 129/sv J1 ES cells, and homologous recombination was assessed by genomic Southern blot analysis with probes flanking the targeted DNA. Two clones carrying the targeted allele were microinjected into C57BL/6J mouse blastocysts, and resulting chimeric male were bred to C57BL/6J female mice. The germline transmitted F1 offspring was obtained from one clone. These offsprings were genotyped by either Southern blotting or PCR. The heterozygous offsprings will be intercrossed to give rise to homozygous mutant mice.