CHARACTERIZATION OF TRANSGENE INTEGRATION SITES OF GENETICALLY MODIFIED FIBROBLASTS.

Lisauskas, S; Oliveira, RR; Carvalho, DM; Melo, EO; Rech, EL; Aragão, FJL
EMBRAPA Recursos Genéticos e Biotecnologia, Parque Estação Biológica, Brasília-DF, CEP 70.770-900.
sharon@cenargen.embrapa.br
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The transference of genetic material manipulated by recombinant DNA technology is an innovative method designed to produce transgenic dairy animals with an altered genotypes, containing useful traits or producing proteins of pharmaceutical importance. An alternative to generate transgenic animals is the use of transfected fibroblasts lines as a source of donor nucleus by nuclear transfer into an enucleated oocyte. The development of an efficient fibroblast transfected system utilizing liposomes is the tool used in this experiment to produce the scFv fragment deriving from anti Tn 83 D4 monoclonal antibody under control of ?-lactoglobulina promoter. Transgenic cells were selected in culture medium with appropriated antibiotic (G418: 400?g/ml). Since the transgene integration into bovine genome is randomly, just a small number of events will generate transgenic animals expressing the protein in high levels. In order to turn the process more efficient and predictable, studies of the integration loci has been carried out. The approach was based in the plasmid rescue technique and several integrations loci have been characterized. Our data suggest that single-locus transgenic lines have multiple copies of the transgenes without or with very short DNA linker between the integrated plasmids.

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