Combination of transduction enhancers with Ecotropic-MLV pseudotyped lentiviral vectors enables highly efficient transfer of chimeric antigen receptors into murine T effector and T regulatory lymphocytes

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Gene transfer of Chimeric Antigen Receptors (CARs) into T regulatory (Treg) lymphocytes using HIV-1-based lentiviral vectors (LVs) has several therapeutic applications ranging from inflammatory diseases to autoimmunity. The development of these new therapeutic approaches requires the use of murine animal models. However, the transduction of murine lymphocytes with LVs pseudotyped with the broadly used vesicular stomatitis virus envelope glycoprotein (VSV-G) is known to be inefficient. To target more specifically murine cells, it is possible to pseudotype LVs with the ecotropic murine leukemia virus envelope glycoprotein (Eco-LV). Furthermore, numerous peptide derived-transduction enhancers have been developed to improve LV entry into target cells, a rate limiting step. We have evaluated a combination of these two technologies to improve the transduction of murine T lymphocytes. Among the family of transduction enhancers tested, Vectofusin-1® (also called LAH4-A4 peptide) and Protransduzin-A® (also called EF-C peptide) showed a very significant improvement of murine T cell transduction (from 50 to 80%) with Eco-LVs. Under optimal transduction conditions, the viability of the target cells was not affected and the expansion of the transduced cells was not reduced. In conclusion, the strong increase of lentiviral transduction into murine Teff or Treg lymphocytes, by 1) Ecotropic pseudotyping, 2) the use of high titers and 3) the use of transduction enhancers, allowed us to bypass the strong lentiviral restrictions described in murine cells. It opens the way to the implementation of robust protocols for CAR lentiviral gene transfer into murine animal models.