CONCLUSIONS

Retroviral vectors are transcriptionally unstable in mammalian cells. The ASLV-derived vectors are the most affected by silencing. We modified the vector by insertion of the CpG island inner element (IE) into the vector LTR in three different positions in either sense or antisense orientation. Each vector variant exhibited a certain extent of stabilization. The vector with insertion of a tandem of two IEs between the enhancer and the promoter was the most stable and exhibited almost no silencing after four months of cultivation in rodent and human cells. The IE comprises two high-affinity Sp1 binding sites. The presence of Sp1 binding sites is important for the protective effect of IE, but at least a part of the entire anti-silencing capacity is maintained in IE with mutated Sp1 sites.

We identified the Tvc receptor of ASLV. The *tvc* gene encodes a 488-amino-acid protein most closely related to mammalian butyrophilins, members of the immunoglobulin protein family. To confirm the identification of the Tvc receptor, we disrupted both *tvc* alleles in normally susceptible DT40 cells. The DT40 $tvc^{-/-}$ clone was resistant to the ASLV(C) infection.

We identified the mutation that results in decreased susceptibility to infection by ASLV subgroups B and D and resistance to ASLV subgroup E of line M chickens. The *tvb* gene in line M, tvb^{r2}, encodes a mutant Tvb^{S1} receptor protein with substitution of a serine for a cysteine at position 125 (C125S). The C125S substitution significantly reduces the binding affinity of the Tvb^{S1} receptor for the subgroup B, D, and E ASLV envelope glycoproteins. These are the first results that demonstrate a possible role of the cysteine-rich domain 3 in the function of the Tvb receptors.

The MLV-based retroviral vectors expressing the v-*src* and kinase-dead double Y416F-K295N mutant were constructed. They were used for the preparation of stable cell lines expressing one of the variants of the *src* gene in order to assess the effect of these mutants on the c-*src* activity. We found that expression of the active v-*src* induced activation of endogenous c-s*rc* and increased general protein-tyrosine phosphorylation in the infected cells. Expression of the kinase-dead mutant induced hypophosphorylation of Tyr416 of the endogenous c-s*rc*.

We prepared a MLV-based VSV-G pseudotyped vector capable of efficiently transducing spermatogonial stem cells (SSC). The explanted SSCs were infected with the

vector *in vitro* and subsequently transplanted into the testes of recipient cockerels sterilized by repeated γ-irradiation. The transduced reporter gene encoding the green fluorescent protein was detected in the sperms of recipient cockerels with restored spermatogenesis.