REPORTER GENE EXPRESSION FOLLOWING REPEAT ADMINISTRATION OF A HIV-1 LENTIVIRAL VECTOR IN MICE

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BACKGROUND: Repeat dosing of a gene vector may be necessary to ensure long term correction for cystic fibrosis airway disease. Using [illegible] (Luc) bioluminescence imaging, we have examined the persistence of gene expression in individual animals over [illegible] lentiviral gene vector maintained gene expres[illegible] airways [illegible]mal mice.

METHODS: Three groups of normal C57Bl/6 female mice (n=8/group) received a single VSG-G pseudotyped LV gene vector delivered intranasally (4µl of 0.3% lysophosphatidylcholine) followed 1 hour later by a 20µl bolus of the LV vector containing the Luc gene. Four weeks later, one group of mice was re-dosed with a different reporter gene (LacZ), wh[illegible] another group was re-dosed with the [illegible]

RESULTS: Nasal bioluminescence was the same across all 3 groups at 5 weeks post initial LV instillation (Fig. 2., n.s., ANOVA). Those mice that received a different transgene at re-dose (i.e. LacZ) displayed similar gene ex[illegible] 8 and 12 weeks (Fig. 3., i.e. 2 months after the 2nd dose), mice that received two doses of Luc showed significantly less gene expression compared those given a single dose of Luc ($p<0.001$, ANOVA).
LacZ gene transduction was only detected in those mice that received the different [illegible] second dose (Fig. 4., $p<0.05$, ANOVA).
Circulating neutralising antibodies to both the transgene a[illegible] the LV [illegible] detected in the sera, regardless of the number of doses given.

CONCLUSION: These results indicate that re-administration of our LV vector is possible. However re-adm[illegible] the same LV vector transgene after 4 weeks can reduce subsequent gene expression. This effect is likely to be primarily due to a cell-mediated immune response directed against the specific transgene.

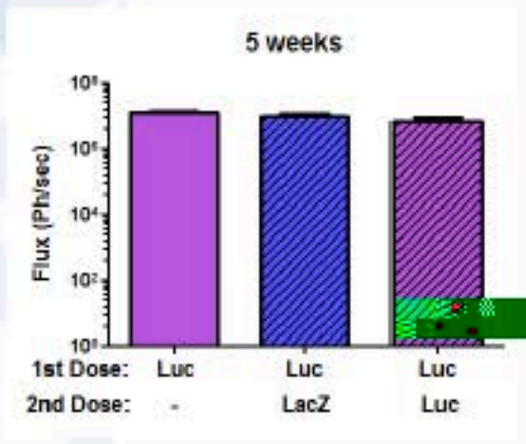


Fig. 2. No significant difference in nasal bioluminescence between all three groups at 5 weeks (ANOVA).

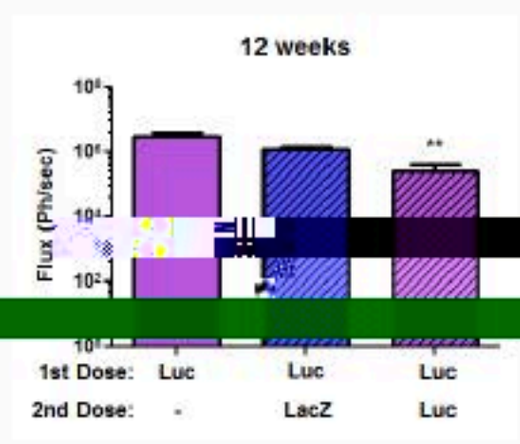


Fig. [illegible] bioluminescence in those mice [illegible] two doses of Luc ($p<0.001$, ANOVA).

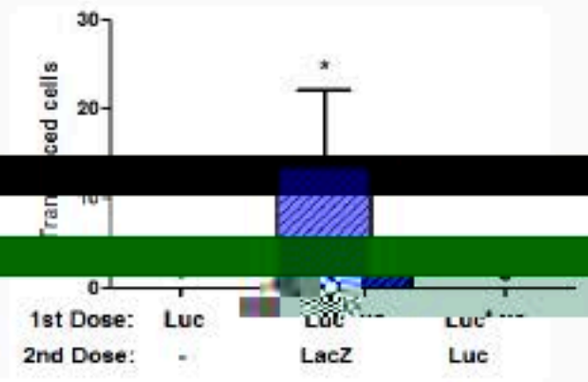


Fig. 4. Mice that received LacZ as the second [illegible] expression at week 12 ($p<0.05$, ANOVA; range: 0-66 blue transduced cells, 2/8 = zero counts).

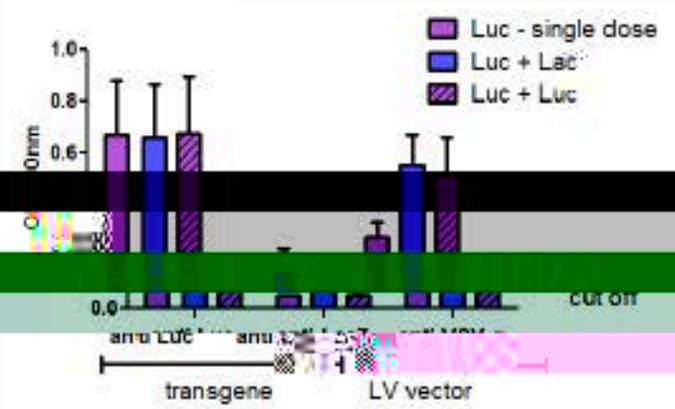
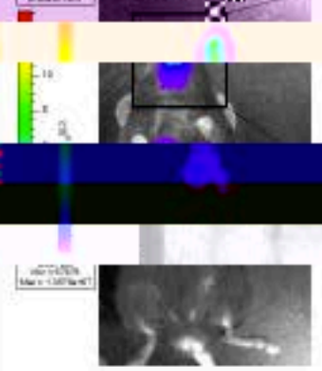
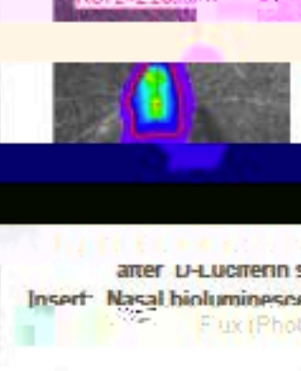
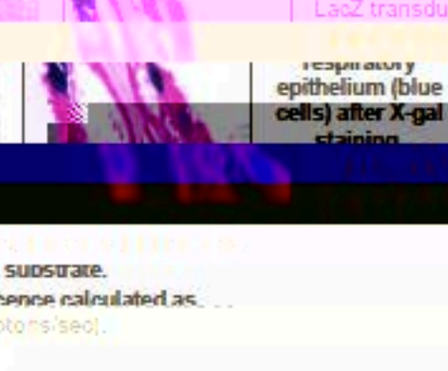


Fig. 5. Presence of circulating neutralising [illegible] envelope, in sera at week 12.







after D-Luciferin substrate.
Insert: Nasal bioluminescence calculated as Flux (Photons/sec).

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