Fig S1: Percentage of GFP (surrogate of CXCR6 transcriptional expression) on T<sub>RM</sub> and effector T cells in Cxcr6<sup>gfp/+</sup> after IN vaccination.

Cxcr6<sup>gfp/+</sup> mice (n = 4) were grafted with TC-1 cells (5x10<sup>4</sup>) in the tongue at D0, then vaccinated by IN route with STxB-E7 and poly-ICLC at D5 and D10, and sacrificed at D15. GFP expression on T<sub>RM</sub> and effector CD8<sup>+</sup> T cells (Teff) was analyzed by flow cytometry in the tumor, lung parenchyma (CD8a<sup>iv</sup>-) and BAL (n=3 mice/group). Results are representative of two experiments. Mean ±sem is shown.*P < 0.05; ** P < 0.01
Fig S2: *Cxcr6<sup>Δp/fΔp</sup>* and C57BL/6 mice have similar levels of H-2D<sup>b</sup> E7 tetramer in the spleen after IN vaccination

(A-B) C57BL/6 or *Cxcr6<sup>Δp/fΔp</sup>* mice were vaccinated with STxB-E7 by IN route at day 0 and 14, then sacrificed at day 21. Representative flow plots (A) and percentage of specific H-2D<sup>b</sup> E7 tetramer gated on CD8<sup>+</sup> T cells (B) in the spleen. Mean ±sem. n=16 mice/group.

(C) *Cxcr6<sup>Δp/fΔp</sup>* mice were grafted with TC-1 cells (5x10<sup>6</sup>) in the tongue at D0, then vaccinated via the IN route with STxB-E7 and poly-ICLC at D5 and D10, and mice were sacrificed at D15. Representative flow plots showing expression of CXCR6 (GFP and surface) among H-2D<sup>b</sup> E7 tetramer in the spleen. D : C57BL/6 mice (n = 5) were immunized with STxB-E7 by IN or IM route at day 0 and 14, then sacrificed at day 21. Percentage and absolute number are shown. n=5 mice/group. Mean ±sem. Mann-Whitney t-test *p<0.05 **p<0.01
Fig S3 : Role of CXCR6 on T cells in the decrease number of specific CD8^+ T_{RM} in the lung.

A : Description of the experimental design of competitive adoptive transfer of T cells derived from CXCR6 deficient or non-deficient mice congenic for CD45

B : T cells isolated from spleen of naive C57BL/6 (CD45.1) and Cxcr6^{gfp/gfp} (CD45.2) were expanded in vitro on anti-CD3 coated plates + IL-2 (4ng/ml) for 5 days in RPMI complete medium, then incubated an additional 2 days in the presence of IL-2 (2 ng/ml).

In vitro activated T cells were harvested, washed, mixed at equal numbers in PBS, then injected i.v into CD3 KO recipient mice which were then immunized with STxB-E7 by IN route at day 1 and 21 and sacrificed at day 28. Absolute number of H-2D^b E7_{49-57} tetramer and specific T_{RM} were analyzed in BAL and lung parenchyma.
Figure S4: Cxcr6 deficiency does not impair the functionality of antigen-specific CD8+ T cells

A-B: C57Bl/6 and Cxcr6<sup>gfp/gfp</sup> mice were vaccinated with STxB-E7 via the IN route at day 0 and 14, then sacrificed at day 21. C: C57BL/6 or Cxcr6<sup>gfp/gfp</sup> mice were grafted in the submucosal lining cheek (IC: intra-cheek) with TC-1 tumor cells, then vaccinated with STxB-E7 by IN route at day 7 and 14, and sacrificed at day 20.

Lymphocytes from BAL (A), lung parenchyma (B) or from the tumor after cell dissociation (C) were stimulated with the E7<sub>49-57</sub> peptide (10 µg/mL) (A, B) or the TC1 tumor for 5 h, in the presence of anti-CD107a/b antibody, Golgistop® (monensin) and Golgiplug® (Brefeldin A). Percentages of CD107a/b, GrzB and IFN<sub>γ</sub> among E7<sub>49-57</sub> tetramer in BAL (A) and lung parenchyma (B) or E7 spéific T<sub>RM</sub> (C) were analyzed by flow cytometry.

Mean ±sem. n=3-5 mice/group. Data are representative of 2 independent experiments. n.s = not significant.
Figure S5: Other chemokines are transiently modulated after IN vaccination.
C57BL/6 mice were vaccinated intranasally with STxB-E7 (20 µg)+poly-ICLC (10 µg). Chemokines CCL3, CCL5, CCL4, CXCL10 and CCL20 were measured by Luminex assay in lysate from lung at the indicated times after immunization. Data are representative of 2 independent experiments with 4 mice/group. Mean ±sem. Mann-Whitney t-test. *P < 0.05; ** P < 0.01; *** P < 0.001; ****P < 0.0001.
**Fig S6 : CXCL16 is expressed by epithelial cells derived from the mucosal lining cheek and lung.**

Frozen section from mucosal lining cheek (A) and lung (B) derived from mice previously injected with Poly:IC were stained with antibodies against epithelial cells (Epcam) or CXCL16. Isotype control antibodies were included in each experiment. Nuclei were highlighted using DAPI solution (1ug/mL, Sigma D9542), slides were mounted with mounting medium (DAKO, S3023). Images were acquired x20 on Vectra® 3 automated microscope and analyzed with inForm® Image Analysis Software. Scale bars are 50 µm.