Supplemental_Material_&_Methods

Animals, infections, and *in vivo* cell depletion

Six- to twelve-week-old C57BL/6 (male) (RRID:IMSR_JAX:000664) and XCR1-DTRvenus mice (male and female)(Yamazaki et al. 2013) were infected intraperitoneally (i.p.) with either a low dose (LD; $2*10^2$ plaque-forming units) or a high dose (HD; $2*10^6$ plaque-forming units) of the strain Docile of LCMV ($LCMV_{Doc}$) to induce an acute or chronic infection, respectively. Viral titers from spleens of infected mice were determined with MC57 cells using focus-forming assay as described previously (Battegay et al. 1991). For depletion of $XCR1^+$ dendritic cells, heterozygote XCR1-DTRvenus mice were infected with a high dose of LCMV to induce a chronic infection, and Diphtheria Toxin (DT; 25ng/g body weight) was administered at days 7, 10 and 13 postinfection. Physiological serum was used as control vehicle.

Flow cytometry, sorting, and immunohistochemistry

For Flow Cytometry analysis and sorting of cells, spleens were harvested and single-cell suspensions were generated. Biotinylated MHC class I monomer (MBL International Corporation) was tetramerized using PhycoPro R-PE conjugated to Streptavidin (Prozyme). For determination of IFNG- or TNFA-producing T cells, splenocytes were stimulated for 5 hours with GP33 or GP61 peptides in the presence of Brefeldin A (BFA) before Intracellular Cytokine Staining (ICS). For determination of IL6-producing monocytes/macrophages, splenocytes from half of each spleen were directly put into media containing BFA. ICS was performed without additional stimulation. All antibodies were purchased from either BD Biosciences, eBioscience, Biolegend or R&D Systems. Data were analyzed using FlowJo 10.1 software. A LSR Fortessa (BD Biosciences) was used for flow cytometry. Monocytes/macrophages and $DbGP33\text{-}41^+$ $CD8^+$ T cells were sorted with a FACSAria II SORP (BD Biosciences). All samples were kept at 4ºC during the sorting and sort purity was >95% for all populations. Spleen samples for immunohistochemistry were embedded in paraffin after an overnight fixation with 4% buffered formaldehyde. Three micrometer thick tissue sections were deparaffinized and rehydrated before staining with Mayer's Hematoxylin and Eosin. Extra sections were obtained and were incubated with DAKO EnVision+ System HRP-labelled polymer with anti-Mouse IgG for immunohistochemical labelling and semiquantification of IgG-positive cells. The reaction was visualized using hydrogen peroxide and 3-3'-diaminobenzidine as a chromogen substrate. A set of scores was defined from 0 (absence of immunolabeled cells in the studied sections) to 10 (100% IgG labelled cells in the studied sections). For semiquantification of fibrosis, collagen fibers in spleen sections were stained with Masson's Trichrome stain. For each spleen a transversal section of one tip of the organ (1/4) was evaluated along with two longitudinal sections of the remaining spleen (3/4) thus enabling a longitudinal and transversal inspection of the organ. Per animal, all three sections were evaluated. A semiquantitative fibrosis score from 0 to 10 was defined. It takes both the thickness of the capsule and the presence of interstitial fibrosis within the splenic parenchyma into account. A score of 0 represents normality (a certain amount of blue stained collagen is to be observed in the splenic capsule and trabeculae), while 10 would correspond to a spleen in which the parenchyma has totally been replaced by

connective tissue. Each time point analyzed consisted of four animals of low dose and four animals of high dose LCMV infection

RNA-seq library preparation and sequencing

Total RNA from spleens and sorted cells ($5*10^4$ cells per sample) was isolated according to the manufacturer's instructions using Qiagen RNeasy Mini kit and Qiagen RNeasy Micro kit (Qiagen), respectively. RNA was submitted for sequencing to the Genomics Unit of Centre for Genomic Regulation (CGR, PRBB). The quality and concentration of RNA were determined by an Agilent Bioanalyzer. Sequencing libraries were obtained after removing ribosomal RNA by a Ribo-Zero kit (Illumina). cDNA was synthesized and tagged by addition of barcoded Truseq adapters. Libraries were quantified using the KAPA Library Quantification Kit (KapaBiosystems) prior to amplification with Illumina's cBot. Four libraries were pooled and sequenced (single strand, 50nts) on an Illumina HiSeq2000 sequencer to obtain 50-60 million reads per sample.

Quantitative Real-Time PCR

cDNA templates were generated using SuperScript III reverse transcriptase (Thermo Fisher Scientific). Quantitative real-time PCR was performed in a Quantstudio 12K flex (Thermo Fisher Scientific). Expression of all target genes was normalized to expression of the housekeeping gene *Gapdh*. Primer sequences are as follows: *Gapdh* forward, 5'-CCA GTA TGA CTC CAC TCA CG-3' and reverse, 5'-GAC TCC ACG ACA TAC TCA GC-3'; *Xcl1* forward, 5'-TTT GTC ACC AAA CGA GGA CTA AA-3' and reverse, 5'-CCA GTC AGG GTT ATC GCT GTG-3'; *Il6* forward, 5'-TGG GAC TGA TGC TGG TGA CA-3' and reverse, 5'-TTT CCA CGA TTT CCC AGA GAA-3'; *Il1b* forward, 5'-GGA GCT CCC TTT TCG TGA ATG-3' and reverse, 5'-TCT TGG CCG AGG ACT AAG GA-3', *Nos2* forward, 5'-ACT CTT CAC CAC AAG GCC ACA T-3' and reverse, 5'-GTT GAT GAA CTC AAT GGC ATG AG-3'; *Itgb2* forward, 5'-AGC CAC CGA TGT GTG AGG AT-3' and reverse, 5'-AAG GGA TAG TCT GCA GCG TCA T-3'; *Ccr1* forward, 5'-CTC ATG CAG CAT AGG AGG CTT-3' and reverse, 5'-ACA TGG CAT CAC CAA AAA TCC A-3', *C5ar1* forward, 5'-TAC CAT TAG TGC CGA CCG TTT-3' and reverse, 5'- CCG GTA CAC GAA GGA TGG AAT-3', *Itgam* forward, 5'-GCA CCA AAA CTG CAA GGA GAA-3' and reverse, 5'-CCG GAG CCA TCA ATC AAG AA-3'; *Zap70* forward, 5'- TCG GCA CTA TGC CAA GAT CA -3', and reverse, 5'-TCA CTG CGG CTG GAG AAC TT -3'; *Cd3d* forward. 5'-GTG GAA GGA TGG TTT GCA AA-3' and reverse, 5'- CAC ACA GTT CTG GCA CAT TCG -3'; *Cd3e* forward, 5'-CCA GCG GGA CCT GTA TTC TG -3', and reverse, 5'- AAC AAG GAG TAG CAG GGT GC -3'; *Fut8* forward, 5'- GTT ATT GGA GTC CAT GTC AG -3', and reverse, 5'- TTG GAG TAC TTT GTC TTT GC -3'; *Ighg2c* forward, 5'- TGA TTG TGC AGA CCC TCG TG -3', and reverse, 5'- CTG TGG ACT GGA CCA GCA AT -3'; *Cd247* forward, 5'- GCT GGA TCC CAA ACT CTG CT -3', and reverse, 5'- GCT GTT TGC CTC CCA TCT CT -3'; *Arg2* forward, 5'- ACC AGG AAC TGG CTG AAG TG 3', and reverse, 5'- TGA GCA TCA ACC CAG ATG AC -3'.

Supplemental_References

Battegay M, Cooper S, Althage A, Bänziger J, Hengartner H, Zinkernagel RM. 1991. Quantification of lymphocytic choriomeningitis virus with an immunological focus assay in 24- or 96-well plates. *J Virol Methods* **33**: 191–198.

Yamazaki C, Sugiyama M, Ohta T, Hemmi H, Hamada E, Sasaki I, Fukuda Y, Yano T, Nobuoka M, Hirashima T, et al. 2013. Critical roles of a dendritic cell subset expressing a chemokine receptor, XCR1. *J Immunol* **190**: 6071–6082.