Supplementary methods

Protozoa: *Neospora caninum* (NC1 strain obtained from ATCC (50843), β-galactosidase expressing NC1 kindly provided by Andrew Hemphill (University of Bern) and TdTomato NC1 and NC1-IL15hRec[18] were grown in Human Foreskin Fibroblast cells (ATCC Hs-27) at 37°C in Dulbecco's Modified Eagle's medium (DMEM, Dutscher) supplemented with 10% FCS and 1% HEPES under an atmosphere containing 5% CO_2. Extracellular protozoa were harvested, centrifuged at 700 g for 10 minutes, washed in endotoxin-free phosphate-buffered saline 1X (PBS, Dutscher) and counted in a hemocytometer. Protein extracts of *N. caninum* were prepared following the same procedure as that for B16F10 cells extracts.

Cellular analysis: Flow cytometry analysis was performed on cell suspension after tumor dissociation using antibody reagents obtained from Miltenyi Biotech: anti-mouse CD3-APC-Vio770 (REA606), CD4-Vioblue (REA 604), CD8a-PE-Vio770 (REA 601), Foxp3-Vio515 (REA 788), NKp46-APC (REA 815), CD11c-PE (REA 754), CD11b-APC-Vio770 (REA 592), Ly6C-Vioblue (REA 796), Ly6G-PE-Vio770 (REA 526), CD38-Vioblue (REA 616), CD86-VioBright515, CD83-APC (REA 304), CD68-APC-Vio770 (REA 835) – or Thermofisher: CD206-PerCP-eFluor710 (MR6F3, Invitrogen). Foxp3, CD68 and CD206 staining was performed using Foxp3 Staining Buffer Set (Miltenyi Biotech).

Antibodies detection: Sera and BALF from mice were analyzed for anti-B16F10 and anti-NC1 immunoglobulins (Ig) A, IgM and IgG by ELISA coating 10µg/well of antigenic extract using alkaline-phosphatase-coupled anti-mouse IgA (A-4937, Sigma), alkaline-phosphatase-coupled anti-mouse IgM (A-9698, Sigma) and alkaline-phosphatase-coupled anti-mouse IgG (A-3438, Sigma).

Protozoan entry assessment: Untreated and desialylated cells were infected with β-galactosidase expressing NC1 strain at a MOI 1 then incubated 4h or 24h at 37°C. Cells were then washed to remove extracellular tachyzoites before permeabilization by 1% Triton X-100 buffer. β-galactosidase was dosed using CPRG substrate at 10µM in 1% HEPES buffer incubated 30 minutes at 37°C.

Macro used for tumor size evaluation with Image J software:

```
macro "melanoma scoring" {
```

```
// Author: mathieu.epardaud@inrae.fr
//   select the blue channel to get total lung
     run("RGB Stack");
     setSlice(3);
 // set threshold
      setThreshold(0, 125);

// measure area and area fraction
   run("Set Measurements...", "area area_fraction limit display redirect=None decimal=3");
   run("Measure");
   selectWindow("Results");

// select the red channel, which has the best contrast
   setSlice(1);
 // set threshold
      setThreshold(0, 75);

// measure area and area fraction
   run("Set Measurements...", "area area_fraction limit display redirect=None decimal=3");
   run("Measure");
   selectWindow("Results");  }
```