

Supplemental figure 1: At high MOI, MV induces activation of pDCs without IL3 pDCs were cultured with IL3, IL3+MV (MOI=10) or with MV alone at a MOI=50. The expression of surface markers by the indicated cells was determined by flow cytometry. IFN- α secretion was measured by ELISA. Results are expressed as the mean±SEM of three independent experiments.



Supplemental figure 2: MV induces granzyme B secretion by pDCs

pDCs were cultured with IL3, IL3+MV or the TLR7 agonist R837. Granzyme B secretion was measured by ELISA. Results are expressed as the mean±SEM of two independent experiments



Supplemental figure 3: Tumor cells have to be infected by MV to induce IFN-α secretion and TRAIL expression by DCs

pDCs were cultured with IL3, with non-infected or MV-infected M18 melanoma cells (A). CD1c⁺ DCs were cultured alone (-), with non-infected or MV-infected M18 melanoma cells (B). IFN- α secretion was measured by ELISA. #, values are below the limit of detection of the kit (7 pg/mL). The expression of TRAIL by the indicated cells was determined by flow cytometry. Results are expressed as the mean ±SEM of two independent experiments.



Supplemental figure 4: The RLR inhibitor MRT67307 impairs the TLR7 and type I IFN signaling pathways

pDCs were pretreated or not with MRT67307 and then cultured with the TLR7 agonist R837 (A). pDCs were pretreated or not with MRT67307 or Ruxolitinib and then cultured with type I IFNs (IFN- α and IFN- β) (B). IFN- α secretion was measured by ELISA. #, values are below the limit of detection of the kit (7 pg/mL). The expression of TRAIL by pDCs was determined by flow cytometry. Results are expressed as the mean±SEM of two independent experiments.



Supplemental figure 5: shRNA-mediated knockdown of RIG-I and TLR7 in Gen2.2 cells

(A, B) Wild-type (wt) Gen2.2 cells, or modified Gen2.2 cell lines tranduced with lentiviruses expressing Scrambled shRNA, RIG-I- (A) or TLR7-(B) specific shRNAs were analyzed by Western blot using antibodies against indicated proteins. Histograms display the quantitative analysis of the extent of shRNA-mediated knockdown of RIG-I and TLR7. Gen2.2 cells were lyzed in a buffer containing 50 mM Tris-HCl pH 7.5, 150 mM sodium chloride, 0.5% (v/v) Igepal, and protease inhibitor cocktail (Complete Mini EDTA-free tablets, Roche Diagnostic). Protein concentration was determined using the PierceTM 660nm Protein Assay (Thermo-Scientific). SDS gel electrophoresis was performed using precast Bis-Tris 4–12% gradient polyacrylamide gels, in the MOPS buffer system (Life Technologies). Proteins were transferred to nitrocellulose membranes (Trans-Blot Turbo Midi Nitrocellulose Transfer Packs, Biorad) then probed with the indicated primary antibodies and appropriate IR dye-conjugated secondary. Rabbit anti-TLR7 antibody was from Abcam. Mouse anti-RIG-I (clone Alme-1) was from Adipogen. Mouse β -Actin (AC-15) was from Sigma-Aldrich. The membranes were scanned using the Odyssey IR imager (LI-COR®). Relative abundance of protein was determined by quantitative densitometry using ImageJ software (NIH, MD, U.S.A.). Densitometry data were normalized to β -actin.