**Introduction**

- Natural Killer cells (NK cells) are lymphocytes able to recognize and kill tumors for which the expression of Major Histocompatibility Complex (MHC) class I molecules is altered.
- This “missing self” recognition is mediated by the lack of engagement of MHC class I molecules with NK inhibitory receptors that include lectin Ly49 molecules in mice and Killer Immunoglobulin-like Receptors (or KIR) in humans. Blocking interactions between KIR and MHC class I positive tumor cells with monoclonal antibodies (mAbs) constitutes an interesting therapeutic strategy.
- The anti-KIR/HLA1/2/3-specific monoclonal antibody, lirilumab, is a human IgG4 that is being developed for treating both hematologic malignancies and solid tumors.
- The objective of this study was to develop a preclinical model to assess the efficacy of the drug candidate tested in clinical trials, lirilumab.

**The KIR molecules and their ligands**

**Results**

**HLA-C-expressing tumor escapes NK cell control in KIR tg-RAG mice**

- Days post tumoral challenge
- Lirilumab treatment increased mice survival in a dose dependent manner when injected at the same time as the tumor challenge.

**Duration of KIR receptor saturation is lirilumab dose-dependent in KIR tg-RAG mice**

- Lirilumab treatment increased mice survival in a dose dependent manner when injected at the same time as the tumor challenge.

**Preventive lirilumab treatment improves mouse survival in a NK cell-dependent manner...**

- ...and this effect is dependent on KIR saturation duration

**Conclusion**

- Lirilumab treatment increased mice survival in a dose dependent manner when injected at the same time as the tumor challenge.
- This protective effect was NK cell mediated and directly correlated with the duration of KIR saturation.
- Interestingly, lirilumab treatment also improved survival in therapeutic conditions (i.e. when the antibody was injected 5 days after the tumor, also in a NK cell dependent manner).
- In conclusion, this study showed efficacy of lirilumab, even in therapeutic condition, as single agent in a HLA-C3-expressing tumor model and this xenogeneic pre-clinical model will be an excellent tool to investigate the therapeutic benefits of combination treatments.

**Tools**

- tumor cells: the human B lymphoma cell line, 721.221, transduced or not with HLA-C3 molecule, a ligand of KIR2DL3.
- KIR tg-RAG mice expressing the human NK inhibitory KIR2DL3, on the surface of NK cells as a RAG-1 deficient background.

**Therapeutic lirilumab treatment improves mouse survival**

- Lirilumab therapeutic efficacy is mediated by NK cells

---

Caroline Sole1, Fabien CHANUC2, Anne-Thierry3, Nicolas Fusser1, Isabelis Palacios2, Mathieu Blery1, Pascale André4, Eric Victor5, Sophie LOGOLIN2, Robert Grzesiak3, François Krommage1, Régis Broumoux1

1. INNATE PHARMA, MARSEILLE, France
2. Centre d’Immunologie de Marseille-Luminy, MARSEILLE, France
3. Bristol-Myers Squibb, Princeton, NJ

# 493

---

**Fig 3** 721.221 HLA-Cw3 tumor cells (10^6) were injected i.v. to KIR tg2RAG mice. At day 0, mice received lirilumab (mg/kg, day 5) or vehicle (Veh) (100 µg). When indicated, NK cell depletion was achieved by injecting 100 µg of anti-NK1.1 (hIgG4, clone 1wc.5) daily 7 days starting the day before tumor challenge (results of a representative experiment, n=6 mice per group). Differences in median survival were evaluated statistically with logrank (Mantel Cox) test.

---

**Fig 4** 721.221 HLA-Cw3 tumor cells (10^6) were injected i.v. to KIR tg2RAG mice. Lirilumab (1 mg/kg) was injected intravenously once a week starting day 6. When indicated, NK cell depletion was achieved by injecting 100 µg of anti-NK1.1 (hIgG4, clone 1wc.5) daily 7 days starting the day before tumor challenge (results of a representative experiment, n=6 mice per group). Differences in median survival were evaluated statistically with logrank (Mantel Cox) test.

---

Fig 3: 721.221 HLA-Cw3 tumor cell line (10^6) was injected i.v. to KIR tg2RAG mice. At day 0, mice received lirilumab (mg/kg, day 5) or vehicle (Veh) (100 µg). When indicated, NK cell depletion was achieved by injecting 100 µg of anti-NK1.1 (hIgG4, clone 1wc.5) daily 7 days starting the day before tumor challenge (results of a representative experiment, n=6 mice per group). Differences in median survival were evaluated statistically with logrank (Mantel Cox) test.