IL-15-dependent NKp46^{+} cells control intestinal inflammation via CCL3-CCR1-dependent recruitment of inflammatory monocytes

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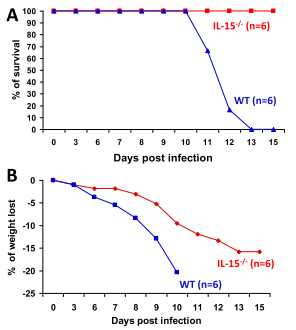
INTRODUCTION

Interleukin 15 (IL-15) is a pleiotropic cytokine with a large range of functions at the interface between innate and adaptive immunity. An essential role in the differentiation, survival and/or activation of NK, NKT cells, TCRγδ intraepithelial lymphocytes and CD8 memory T cells has been firmly established in mice lacking IL-15 or IL-15Ra (Kennedy et al., 2000 ; Lodolce et al., 1998). In humans, IL-15 might contribute to the pathogenesis of a spectrum of inflammatory or autoimmune diseases and different mechanisms have been propounded. Herein we have analysed how IL-15 might promote acute intestinal inflammation using the model of acute ileitis induced by *Toxoplasma gondii* in C57BL/6 mice.

METHODS

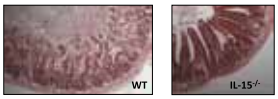
We have used a model where C57BL/6 mice (WT) are orally infected with *Toxoplasma gondii* cysts. These mice develop a severe ileitis on day 7 leading to animal death by day 13.

Survival curve (A), weight loss curve (B) and histology (C) of infected IL-15$^{-/-}$ and WT mice



This survival curve shows that all infected WT mice died between days 10 and 13 while all IL-15$^{-/-}$ mice survived to the acute ileitis.

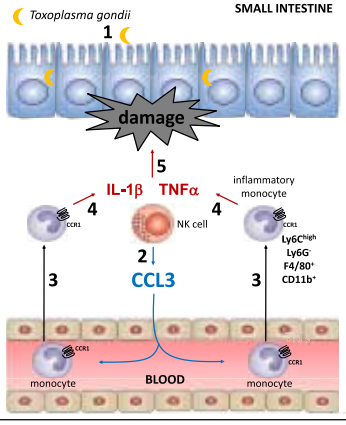
C



Ileitis is not abolished in IL-15$^{-/-}$ mice but is less severe : IL-15$^{-/-}$ mice lost less weight than WT mice and histological lesions were less severe **indicating that IL-15 controls the severity *T. gondii* ileitis**. No change in parasitic load was observed (data not shown).

CONCLUSION

During *T. gondii*-induced ileitis, IL-15-dependent NK cells produce CCL3 which stimulates the recruitment of CCR1^{+} inflammatory monocytes.



LEGEND

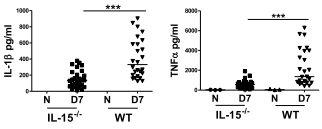
1. Intestinal infection by *T. gondii*.
2. IL-15-dependent NK cells produce CCL3.
3. CCL3 recruits CCR1 inflammatory monocytes.
4. Inflammatory monocytes produce inflammatory cytokines that participate in intestinal damage (5).

This mechanism might operate in Crohn's disease. Blocking CCL3/CCR1 interactions could be a novel therapeutic option.

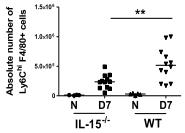
RESULTS

QUESTION : Is there any proinflammatory role of IL-15 ?

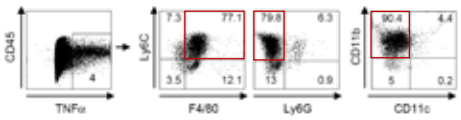
1. IL-15 enhances the secretion of inflammatory cytokines during *T. gondii*-induced ileitis



3. IL-15 stimulates the recruitment of IM in LP of *T. gondii*-infected mice



2. Inflammatory cytokines are produced by lamina propria (LP) inflammatory monocytes (IM)



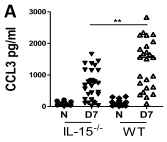
Ly6C^{hi} F4/80^{+} Ly6G^{-} CD11b^{+} = Inflammatory monocytes (IM)

IL-15 stimulates the recruitment of IM during *T. gondii* induced ileitis.

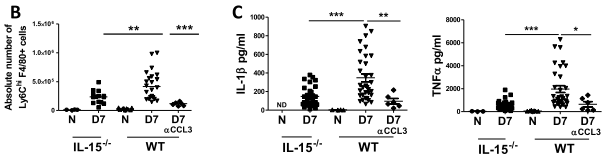
QUESTION : What is the mechanism of IL-15-dependent recruitment of IM ?
HYPOTHESIS : IL-15 controls the induction of chemokine(s) that recruit(s) IM

1. qPCR was used to assess *ccl2*, *ccl3*, *ccl4* and *ccl5* mRNA in isolated LP cells. Only *ccl3* mRNA were significantly increased in LP cells on day 7 post-infection (PI) in WT compared to IL-15$^{-/-}$ mice. Increased CCL3 secretion by isolated LP cells on day 7 PI was confirmed by ELISA (A).

A



2. To test the role of CCL3, WT mice were injected with a blocking anti-CCL3 antibody and recruitment of IM (B) and secretion of inflammatory cytokines (C) were evaluated.

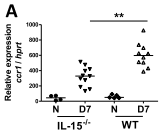


CCL3 participes to the recruitment of IM during *T. gondii*-induced ileitis.

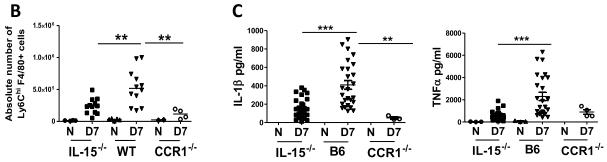
QUESTION : Which chemokine receptor allows the recruitment of IM via CCL3 ?
HYPOTHESIS : CCL3 binds two receptors : CCR1 and CCR5. One of these receptors ?

1. qPCR indicated that only ccr1 mRNA were significantly increased in isolated LP cells on day 7 PI in WT compared to IL-15$^{-/-}$ mice (A).

A



2. To test the role of CCR1, CCR1$^{-/-}$ mice were infected and recruitment of IM (B) and secretion of inflammatory cytokines (C) were evaluated.

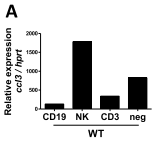


CCR1 plays a key role in the recruitment of IM during *T. gondii*-ileitis.

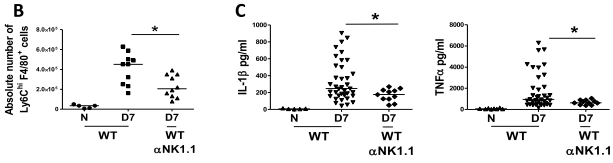
QUESTION : What is the cellular source of CCL3 allowing the recruitment of IM ?

1. During infection, *ccl3* mRNA were induced in LP cells but not in enterocytes (not shown). qPCR on sorted LP cell subsets on on day 7 PI indicated that *ccl3* mRNA were predominantly expressed in NK1.1^{+} CD3^{-} cells , a subset of IL-15 dependent intestinal NK cells (A).

A



2. To test the role of IL-15-dependent NK cells, WT mice were injected with a depleting anti-NK1.1 antibody and the recruitment of IM (B) and the secretion of inflammatory cytokines (C) were evaluated.



IL-15-dependent NK cells are necessary for IM recruitment during ileitis.