



Induction of IL-22 protein and IL-22-producing cells in rainbow trout *Oncorhynchus mykiss*



Yehfang Hu, Yamila Carpio, Callum Scott, Ayham Alnabulsi, Abdo Alnabulsi, Tingyu Wang, Fuguo Liu, Milena Monte, Tiehui Wang and Christopher J. Secombes

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Interleukin (IL)-22

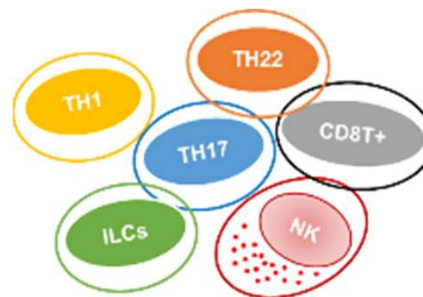
- Belongs to **IL-10 family**

IL-10 subfamily

IL-10	IL-22
IL-19	IL-24
IL-20	IL-26

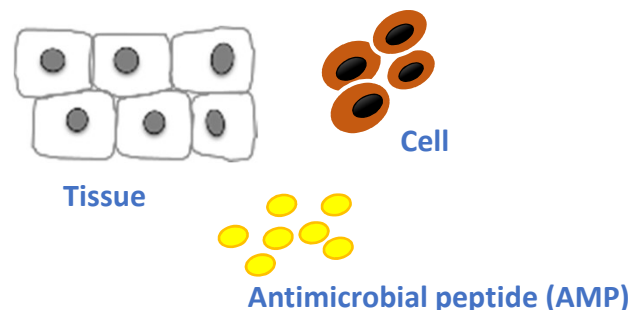
Anti-inflammatory

- Mainly produced by **T cells**, **NK cells** and **innate lymphoid cells (ILCs)**



- **Mucosal defence and tissue protection**

- Tissue regeneration
- Cell proliferation
- Antimicrobial molecule induction

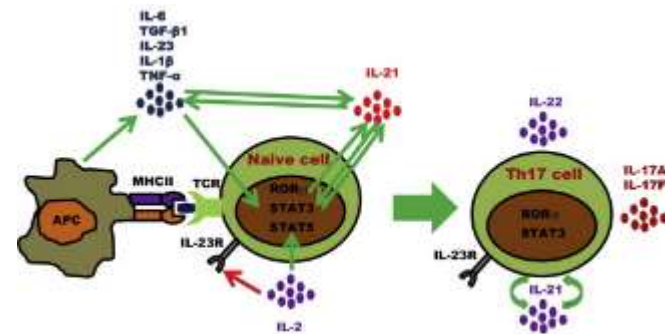


Fish IL-22



- Fish IL-22 has been identified
- Similar functions to mammalian IL-22 as mucosal barrier
- High transcripts in mucosal tissues (gill, intestine, fin, skin)

Mammalian Th17 immune network



(Tiehui Wang and Christopher J. Secombes. 2013)

Transcript IL-22
expression

Protein IL-22
secretion

Monoclonal antibody



Monoclonal antibody (mAb)

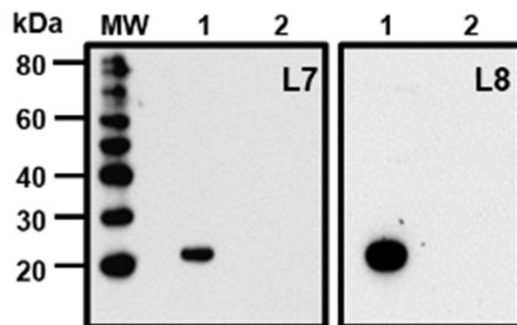


A. mAb production



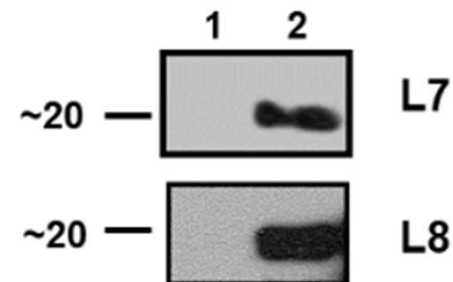
B. mAb characterisation

Specific to peptide immunogen and rIL-22



(1) 100 µg rIL-22
(2) 100 µg rIL-2B

Recognise induced native IL-22



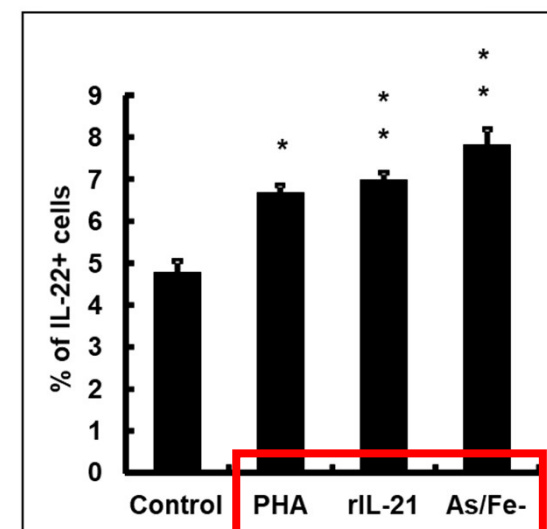
(1) HK cells + PBS 72 h
(2) HK cells + **rIL21** 72 h



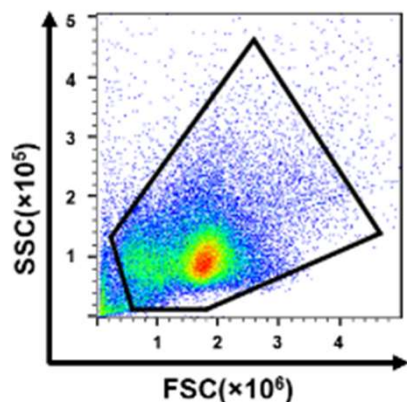
IL-22 producing cells

- Hypotonic PBL- rapid and high quantity (Hu et al., 2018)
- IL-22 transcript inducing stimulants

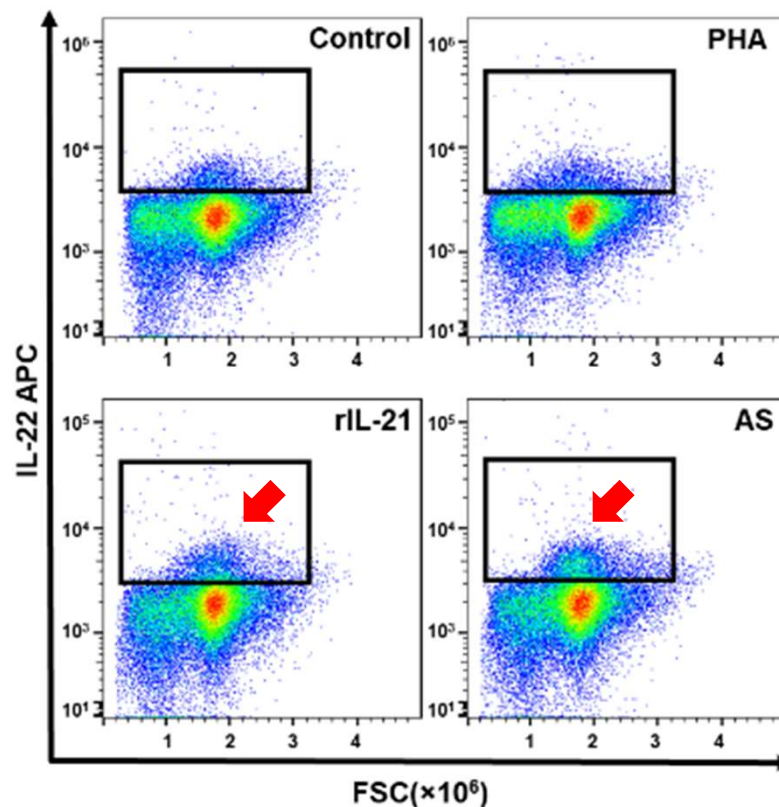
C. Frequency of IL-22+



A. Total PBLs gating



B. IL-22+ gating



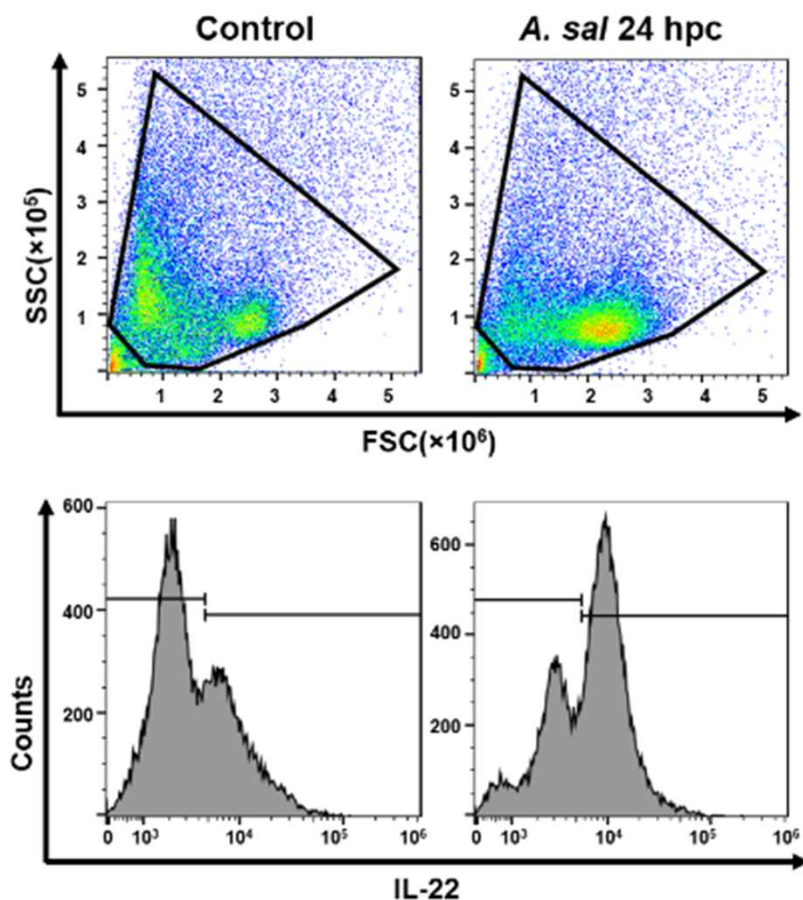
- Native IL-22 is detectable by intracellular staining
- Number of IL-22+ cells increased by IL-22 inducing stimulants



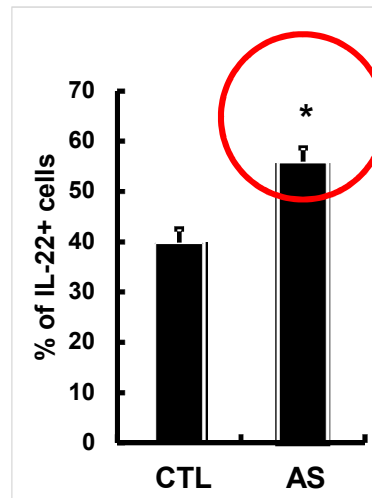
Gill IL-22 post *A. sal* infection

- Mucosal tissue-gill
- Bacterial infection

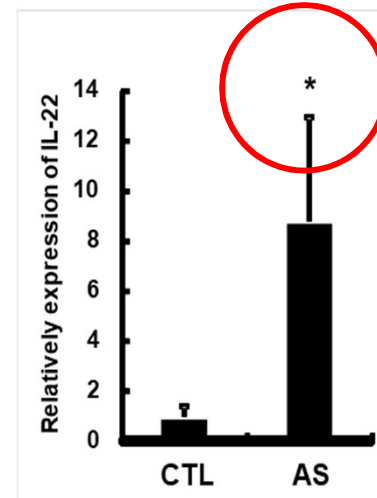
A. Total leucocytes/IL-22+ gating



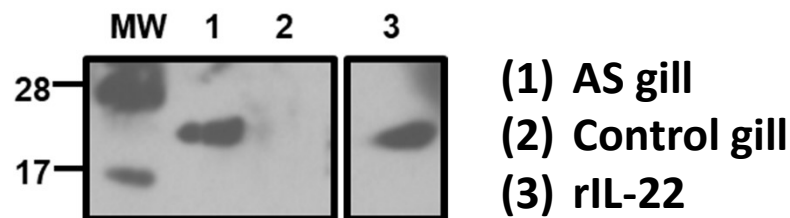
B. Frequency of IL-22+



C. IL-22 transcript



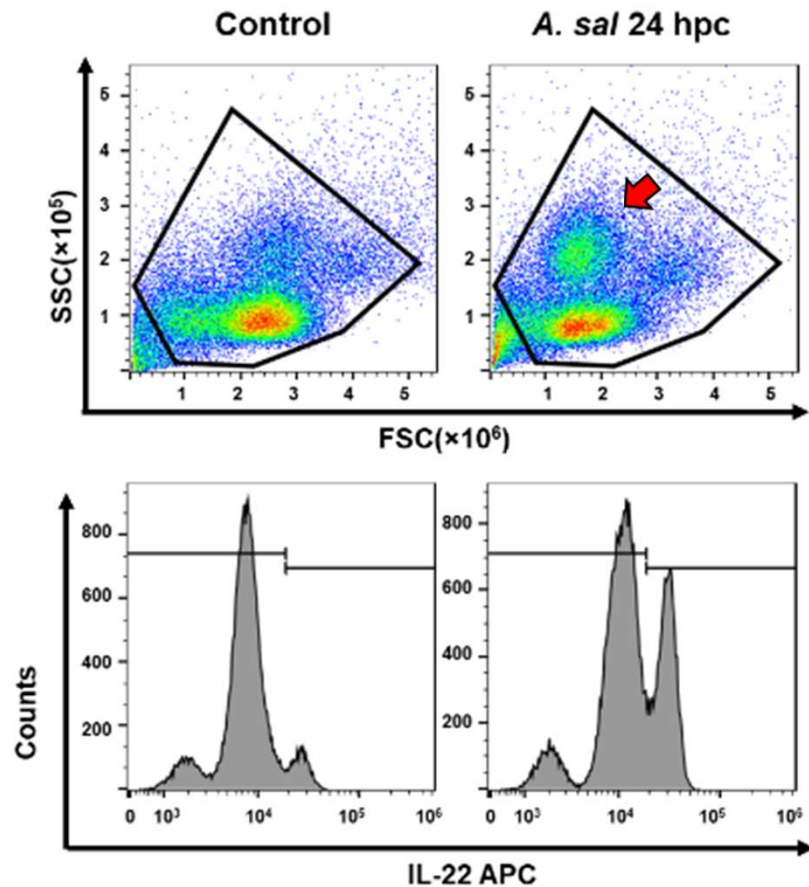
D. IL-22 protein expression



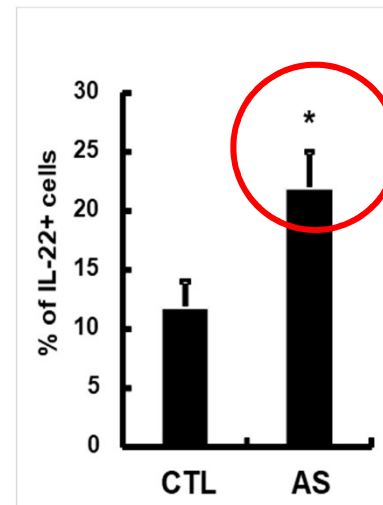
PBL IL-22 post *A. sal* infection



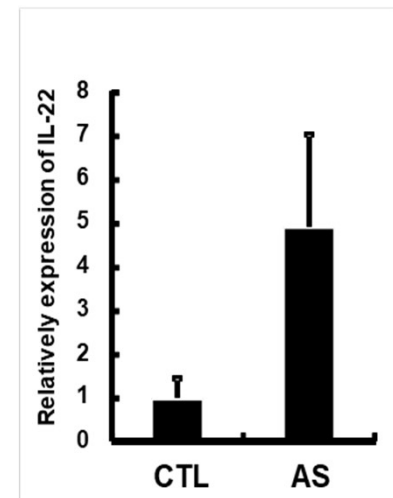
A. Total leucocytes/IL-22+ gating



B. Frequency of IL-22+



C. IL-22 transcript

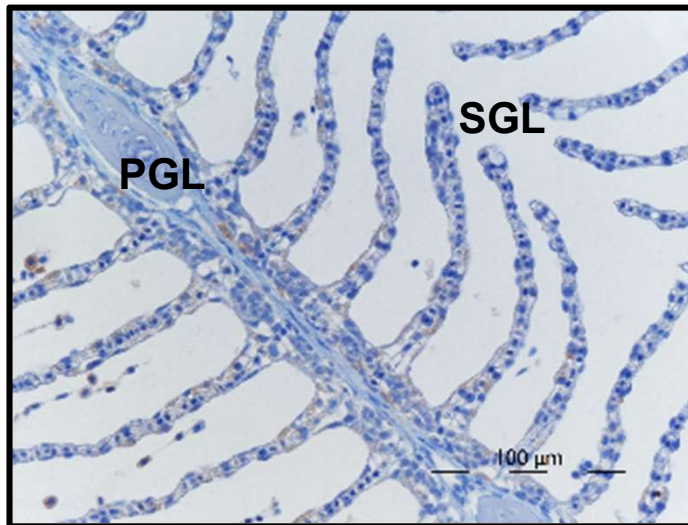


- IL-22 transcript and protein are increased post *A. sal* infection
- IL-22 producing neutrophils?

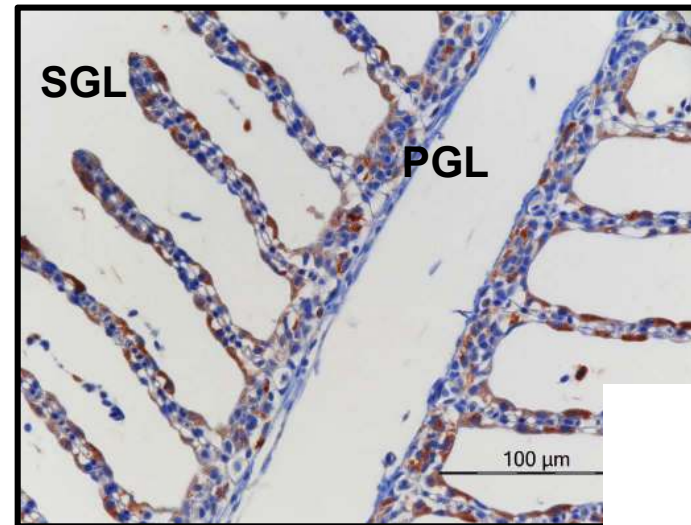
IL-22+ cells in gill



Control

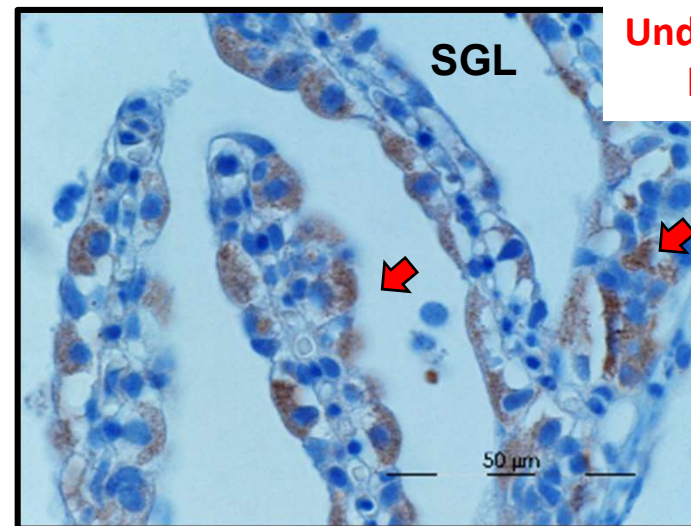
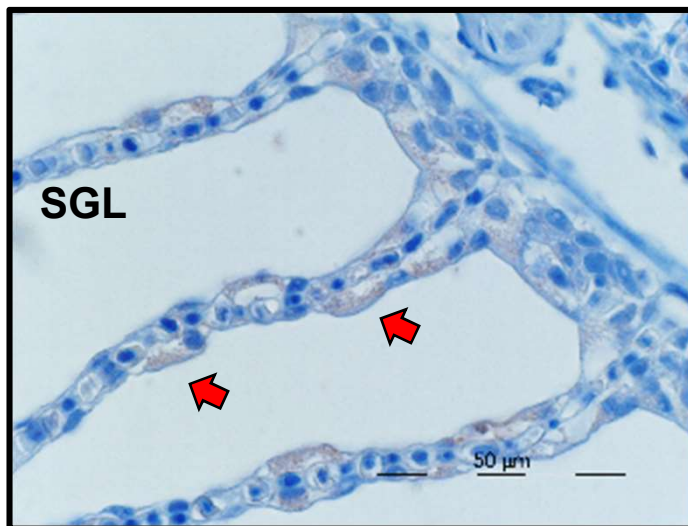


A. sal 24 hpc



Epithelial
cells

Undifferentiated
basal cells



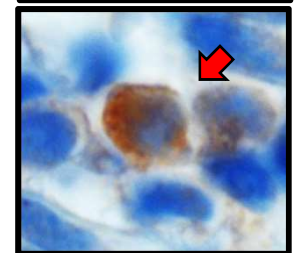
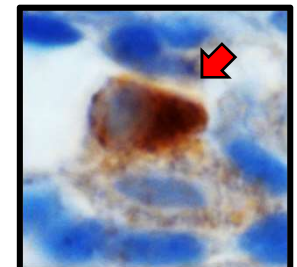
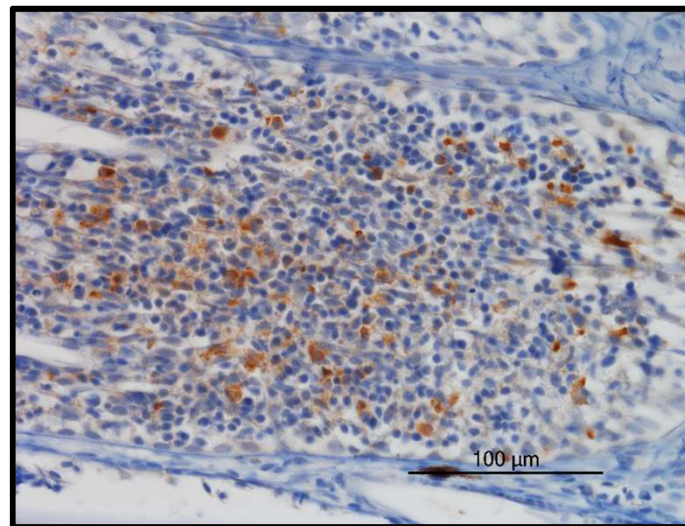
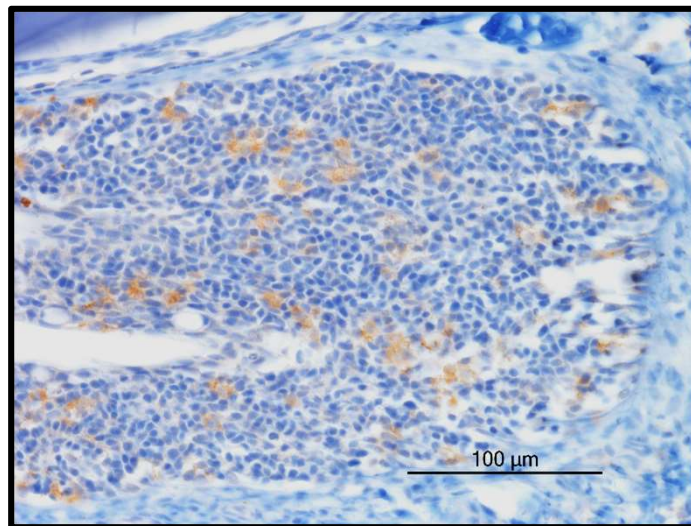
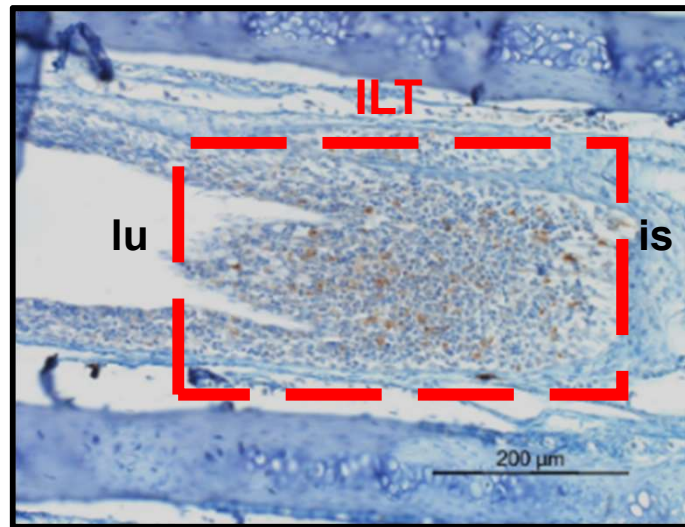
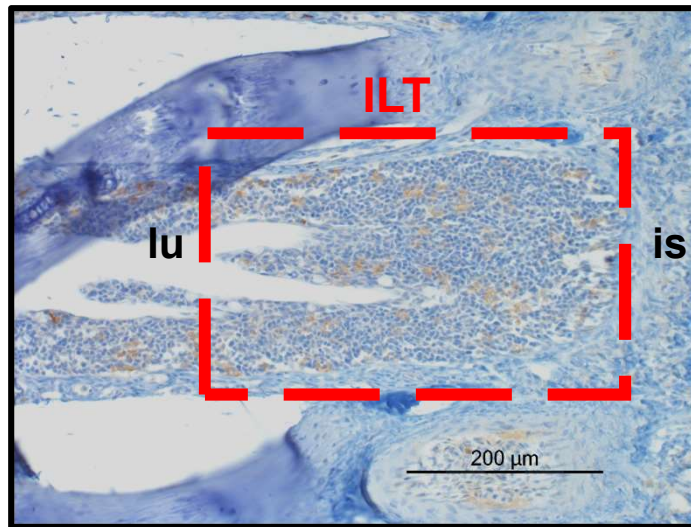
PGL: primary gill lamellae, SGL: secondary gill lamellae

IL-22+ cells in ILT



Control

A. sal 24 hpc



ILT: interbranchial lymphoid tissue, LU: lumen, IS: interbranchial septum



Conclusion



- Two mAbs (L7 & L8) developed against rtIL-22



- IL-22 protein expression is up-regulated in respond to bacterial infection



- Monitoring IL-22 to assess mucosal vaccines

Acknowledgements



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Thanks for your attention

