Unleashing the power of inflammasomes and trained immunity: promising strategies in the fight against Staphylococcus aureus infection

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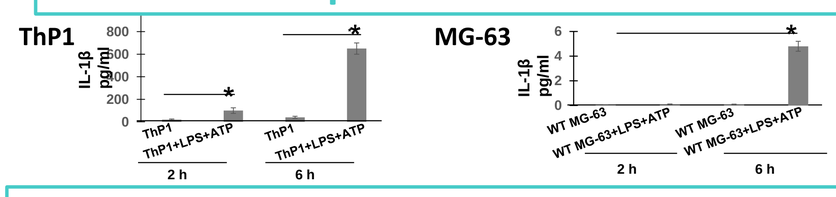
CONTEXT & AIM

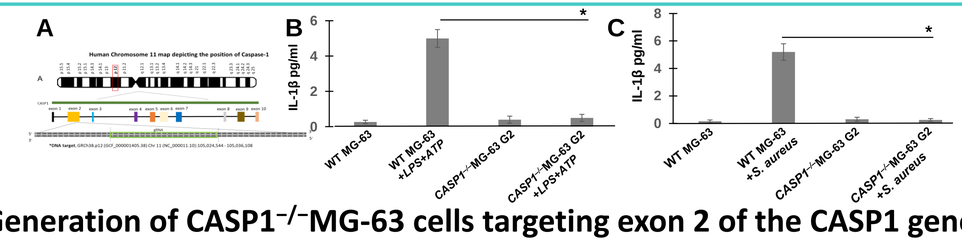
- The inflammasome is a signaling platform that assembles upon danger signal or pathogen recognition, activating downstream proteases like Caspase-1 and Caspase-11 to mature pro-IL-1β, pro-IL-18, and pro-IL-33.
- *Staphylococcus aureus* is a gram-positive bacterium that can cause several fatal infections, which are prone to recurrence. The quorum-sensing system in *S. aureus* (Agr) regulates the expression of PSMs encoding genes.
- Trained immunity (TI) enhances the immune response to subsequent unrelated challenges through epigenetic reprogramming of transcriptional pathways and alteration of cell metabolism.
- Our objectives include a comparison of IL-1β production by monocyte–like ThP1 cells vs osteoblast-like MG-63 cells and comprehending the role of inflammasomes effector, caspase-1, investigating the development of TI in non-immune cells.

RESULTS

1 Monocyte-like ThP1 cells produced a higher amount of IL-1β vs osteoblast-like MG-63 cells

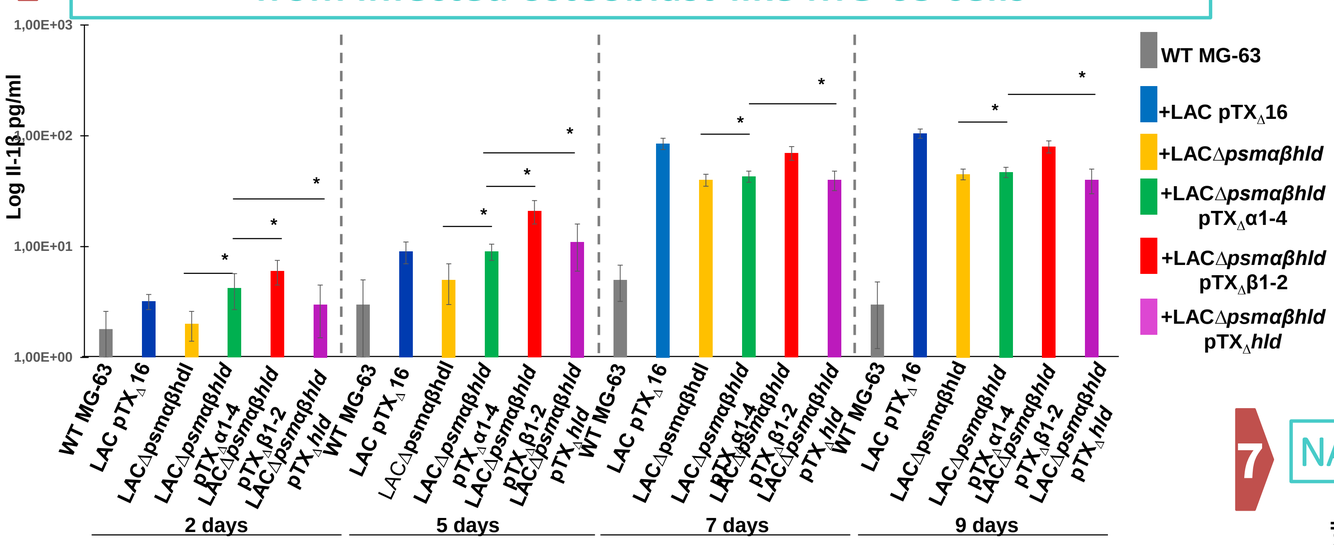


2 Generation of CASP1$^{-/-}$ MG-63 cells using the CRISPR-Cas9



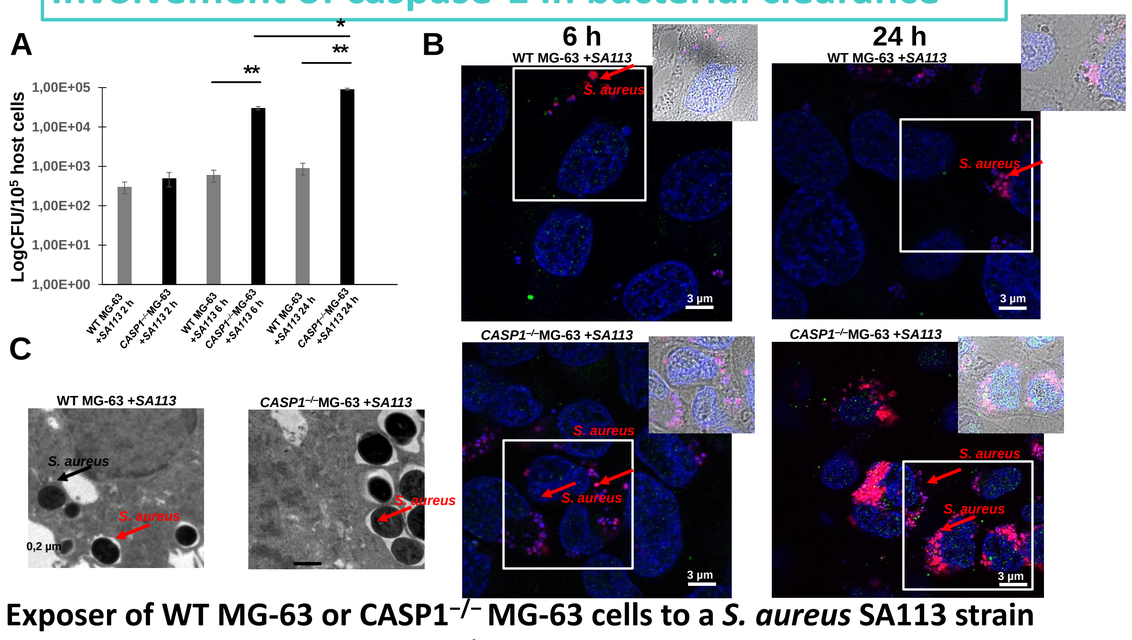
A. Generation of CASP1$^{-/-}$MG-63 cells targeting exon 2 of the CASP1 gene
B. Assessment of NLRP3 mRNA levels in WT MG-63 and CASP1$^{-/-}$ MG-63 cells (RT-qPCR).
D. Assessment of the level of IL-1β in cell supernatants (ELISA)

3 *S. aureus* phenol-soluble modulins stimulate IL-1β release from infected osteoblast-like MG-63 cells



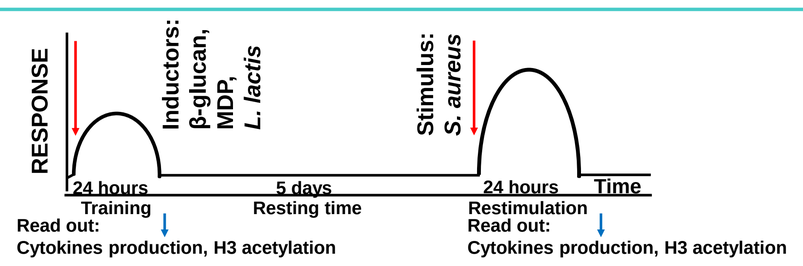
Exposure of MG-63 cells to USA300 LAC (pTXΔ16), which carries the control plasmid, the deletion mutant LACΔpsmαβhld (pTXΔ16), and the complemented strains

4 Involvement of caspase-1 in bacterial clearance



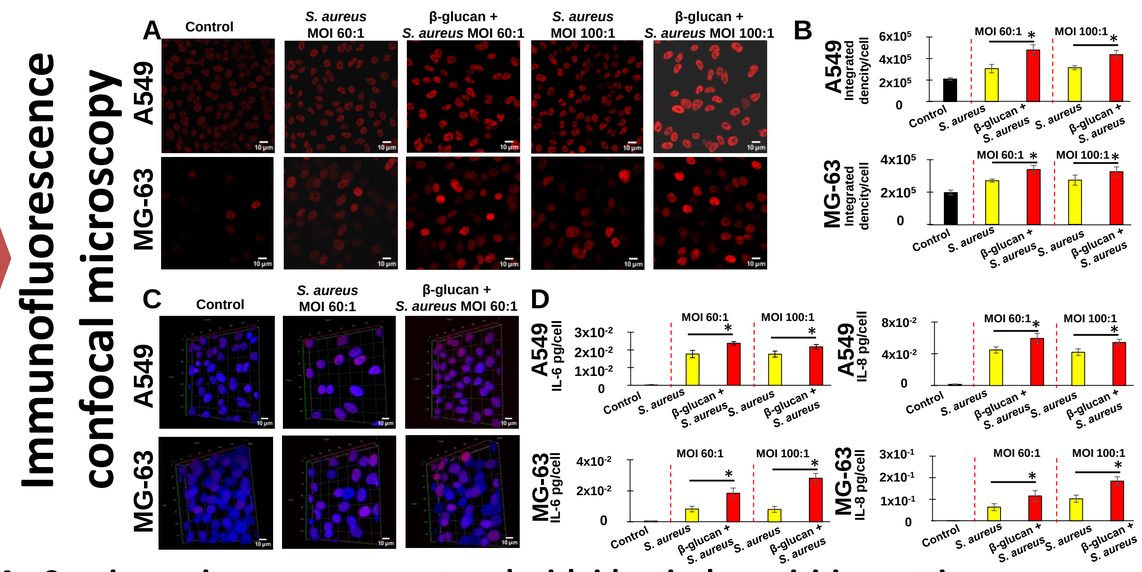
A. Exposer of WT MG-63 or CASP1$^{-/-}$ MG-63 cells to a *S. aureus* SA113 strain
B. Exposer of WT MG-63 or CASP1$^{-/-}$ MG-63 cells to *S. aureus* SA113 (red) Staining with anti-PYCARD antibody (green)
C. Transmission electron micrographs of cells infected with SA113 strain

5 Schematic overview of the innate immune memory model



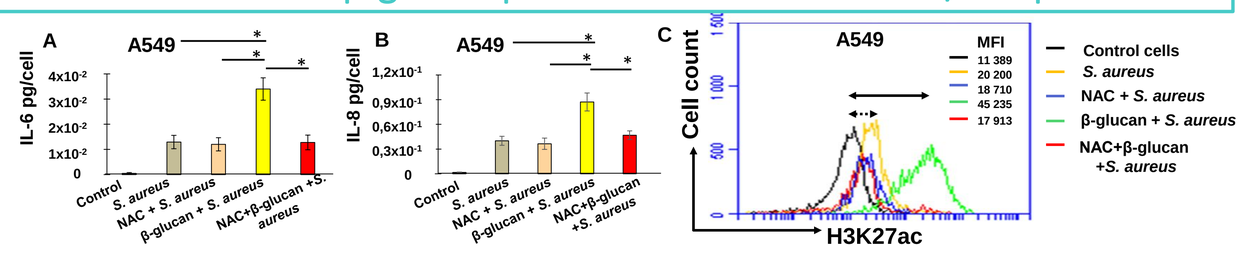
Enhanced H3K27 acetylation in β-glucan-trained cells upon *S. aureus* stimulation, positively correlating with IL-6/IL-8 production

6 Immunofluorescence confocal microscopy



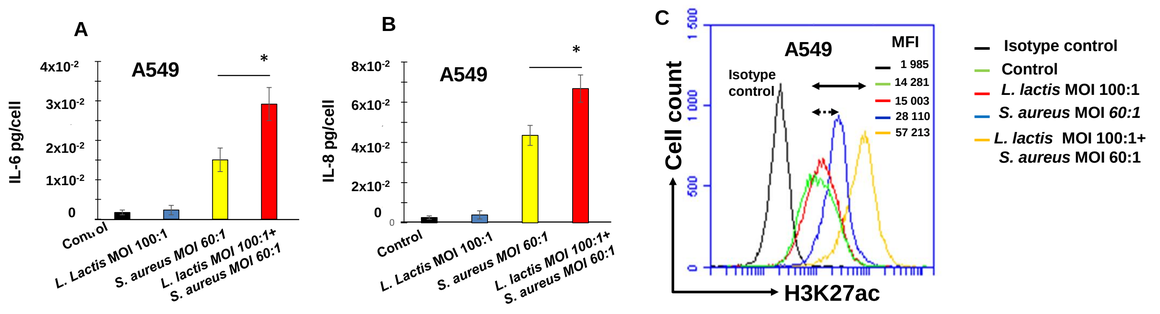
A. Specimens images were captured with identical acquisition settings
B. Normalized Integrated Density was monitored for comparing H3K27 acetylation
C. 3D images were created using Zeiss ZEN software from the Z-stack images
D. Assessment of IL-6 and IL-8 production by ELISA

7 NAC addition before β-glucan-pretreatment reduces IL-6/IL-8 production



A. B. Assessment of IL-6 and IL-8 production by ELISA in NAC-pretreated cells upon *S. aureus* stimulation
C. Flow cytometry analysis of treated cells

8 Cells exposed to *L. lactis* increase IL-6/IL-8 production upon *S. aureus* stimulation, positively correlating with H3K27 acetylation



A. B. Assessment of IL-6 and IL-8 production by ELISA in cells exposed to *L. lactis* before *S. aureus* infection
C. Flow cytometry analysis of treated cells

CONCLUSION

- **Non-immune cells induce an immune response against *S. aureus* through inflammasomes activation and processing of IL-1β**
- **Inflammasomes related IL-1β production by infected osteoblast-like cells appears to be dependent on PSMβ among PSMs**
- **The active caspase-1 restrict intracellular replication of *S. aureus* in non-professional phagocytes**
- **Besides structural functions and tissue homeostasis, non-immune cells contribute to the defense response in infected hosts**
- **The developpment of TI in non-immune cells is partially dependent on ROS production**
- ***L. lactis* may be a potential inducer of the innate immune system in non-immune cells, suggesting the possibility of using this food-grade lactic acid bacterium with probiotic properties before surgery as a preventive measure against staphylococcal infection**

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