Effects of the cholinergic signalling pathway on NLRP3 inflammasome stimulation in human primary cells

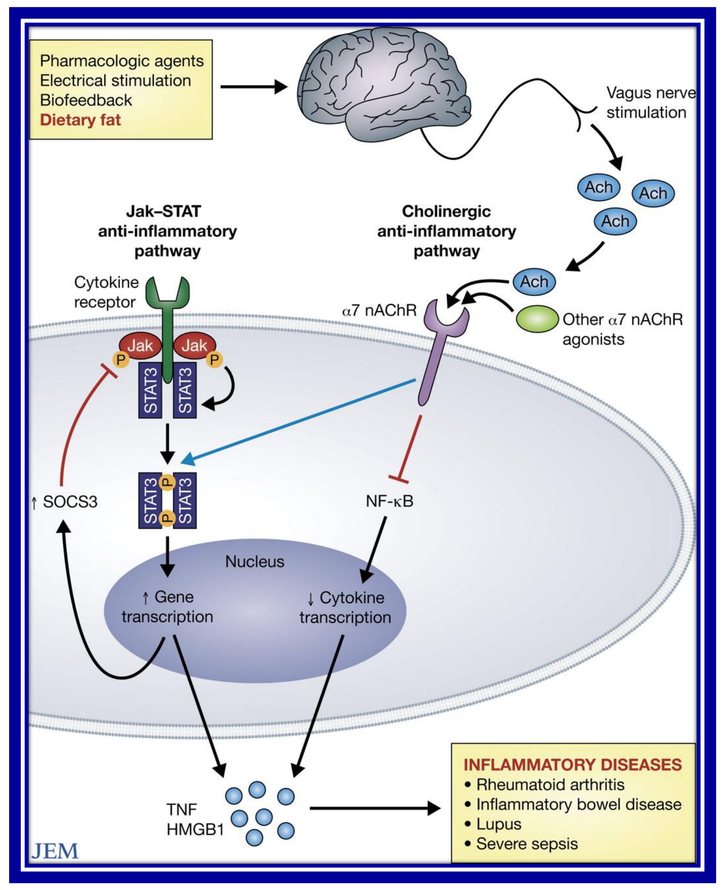
H.W. Grievink*, R. Alvarez-Jimenez§, M. Moerland§, J. Burggraaf§, G.J. Groeneveld§, A.W.J.W. Tepper▪, A.P.H. Scheefhals▪, C. Kluft*, K.E. Malone*

*Good Biomarker Sciences, Leiden, the Netherlands §Centre for Human Drug Research, Leiden, the Netherlands ▪Crossbeta Biosciences, Utrecht, the Netherlands

INTRODUCTION & AIM

Acetylcholine is a neurotransmitter, which can be released after vagal nerve stimulation. Acetylcholine agonizes (amongst others) the α7 nicotinic acetylcholine receptor (α7nAchR), which leads to NF-κB inhibition. This process, known as the cholinergic anti-inflammatory pathway, suppresses cytokine release. Previous work by our lab and others indicates this reflex has a strong influence on IL-1β secretion. Release of IL-1β and IL-18 is now recognized to be a primary result of inflammasome stimulation.

Therefore, we investigated the effect of the cholinergic anti-inflammatory reflex on NLRP3 inflammasome stimulation, using choline as a α7nAchR agonist and different NLRP3 stimuli.



Tracey K.J. J Exp Med 2005;202:1017-1021

METHODS

- Matrices: whole blood, THP-1 cell line
- Preincubation with choline (15 min.)
- Inflammasome stimulation with LPS or LPS + a secondary trigger (eATP, aluminiumhydroxide, β amyloid 1-42 oligomers (Crossbeta))
- Readout: IL-1β and IL-18

RESULTS

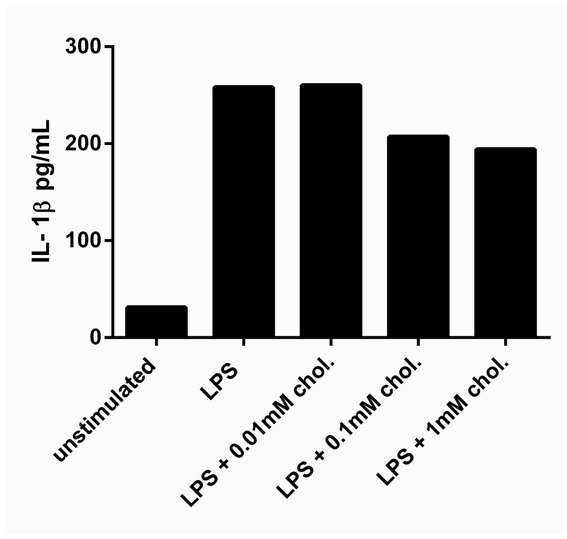


Fig. 1: effect of choline on LPS stimulation in whole blood, 3h incubation.

IL-1β release decreases as much as 25% when whole blood is incubated with 1mM choline before LPS stimulation.

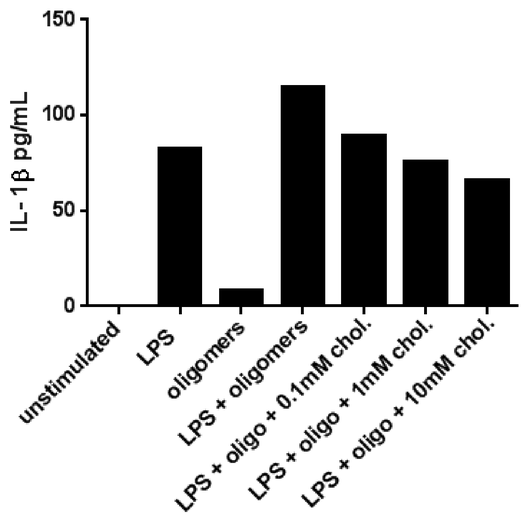


Fig. 2: effect of choline on LPS + β amyloid1-42 oligomer stimulation in THP-1 cells (24h incubation).

Preincubation with choline dose dependently inhibits IL-1β release to a level lower than LPS only.

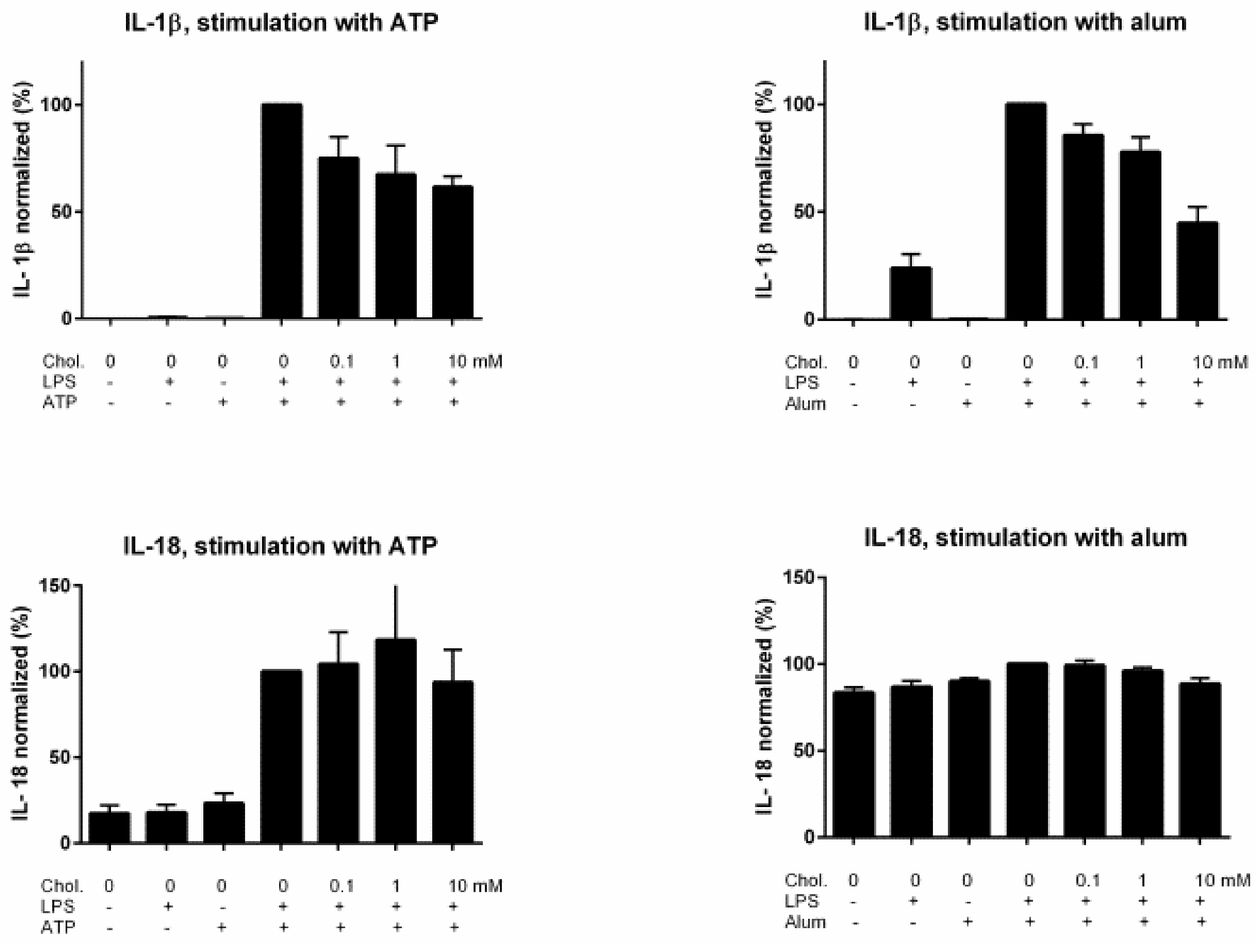


Fig .3: effect of choline on LPS + ATP/alum stimulation in whole blood. Normalization of 3 subjects, 3h incubation.

Choline dose-dependently reduces IL-1β release but not IL-18 in the context of extracellular ATP or particulate exposure.

CONCLUSIONS

These results support a direct mechanism for cholinergic anti-inflammatory activity at the level of NLRP3 inflammasome activation, as determined by IL-1β release. Notably these are the first data to suggest differences in the ability of the cholinergic reflex to modulate the regulation of IL-1β secretion as compared to the related cytokine IL-18, specifically in human cells.
Modulation of cholinergic signaling may be of therapeutic value for diseases where inflammasome activation is implicated, such as cardiovascular disease, diabetes, rheumatoid arthritis and Alzheimer's disease.

Good Biomarker Sciences, Zernikedreef 8, 2333 CL, Leiden, the Netherlands
www.gbsleiden.nl