



# Effect of nutrient deficiencies on *in vitro* cytokine response of peripheral blood mononuclear cells to *Plasmodium falciparum* infection

Erasto V Mbugi,<sup>1</sup> Marjolein Meijerink,<sup>1,5</sup> Jacobien Veenemans,<sup>1</sup> Matthew McCall,<sup>2</sup> Raimos M Olomi,<sup>3</sup> John F Shao,<sup>3</sup> Jaffu O Chilongola,<sup>3</sup> Jos van der Wielen,<sup>4</sup> Hans Verhoef,<sup>1,6</sup> Huub FJ Savelkoul<sup>1</sup>

<sup>1</sup> Cell Biology and Immunology Group, Wageningen University<sup>1</sup>, <sup>2</sup>Medical Microbiology, Radboud University, Nijmegen, <sup>4</sup>Meander Medical Centre, Amersfoort, Host-Microbe Interactomics, Wageningen University; **The Netherlands**, Kilimanjaro Christian Medical Centre<sup>3</sup>, Moshi; **Tanzania**; <sup>6</sup>LSHTM, Nutrition and Public Health Intervention Research Unit; **England**



# Objectives

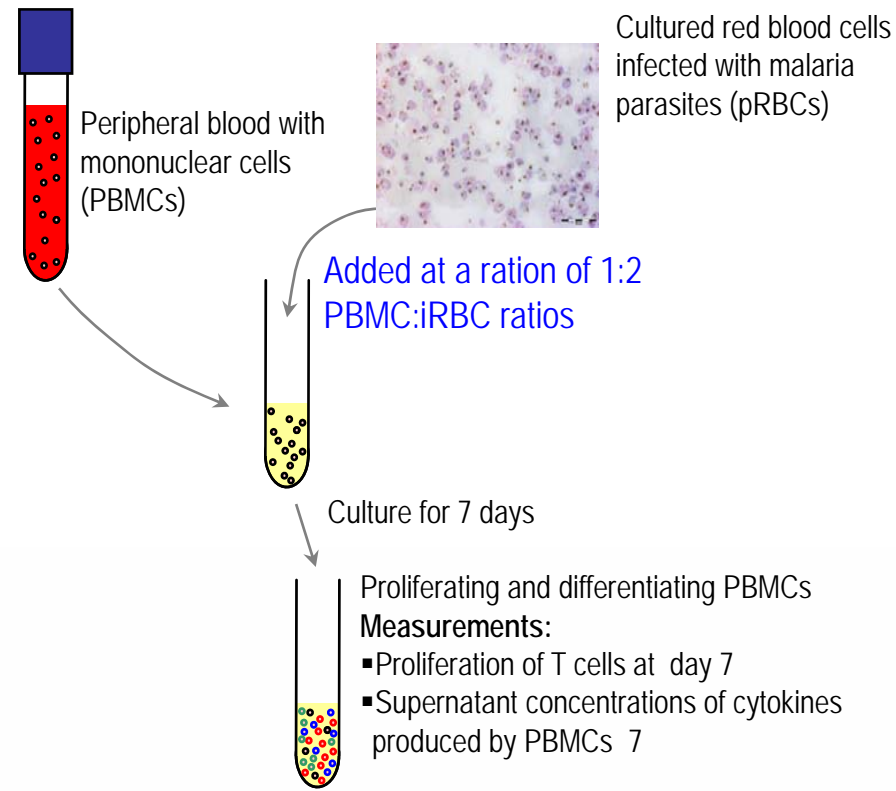
- To assess the effects of nutrient deficiencies, particularly zinc, magnesium and conditions of Iron deficiency anaemia (IDA) on T-cell responses to malaria parasite infected erythrocyte – stimulated PBMCs of Tanzanian children
- **Hypothesis:** Efficient immune response to *Plasmodium falciparum* is influenced by nutrient deficiencies resulting into an imbalance in Th1 and Th2 cells responses; consequently affecting the adaptive immune protection
- Signature cytokines: IFN-gamma, TNF-alpha, IL-1beta and IL-12

# Methods

- 304 PBMC samples Tanzania children aged 6 -72 months
- Stimulation of PBMCs with infected erythrocytes

## Outcome indicators:

- T cell proliferation
- Cell death
- Surface markers
- Cytokine concentration
- Intracellular staining

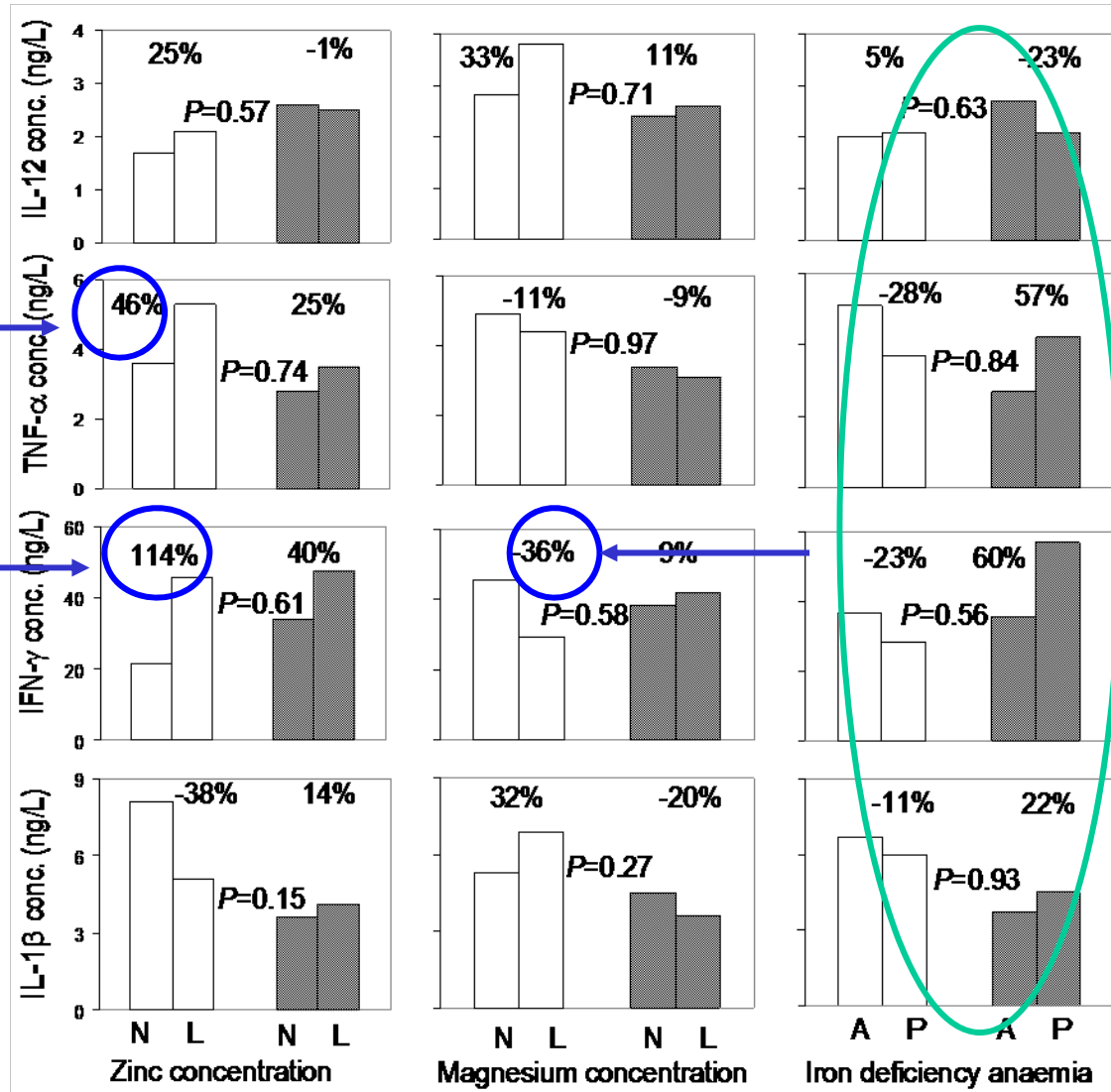




# Experiment: set-up

- Optimal PBMC:pRBCs ratio – 1:2
- Culture time: 7 days
- Negative control: uninfected erythrocytes
- Positive control: maximum response with anti-CD3/ anti-CD28





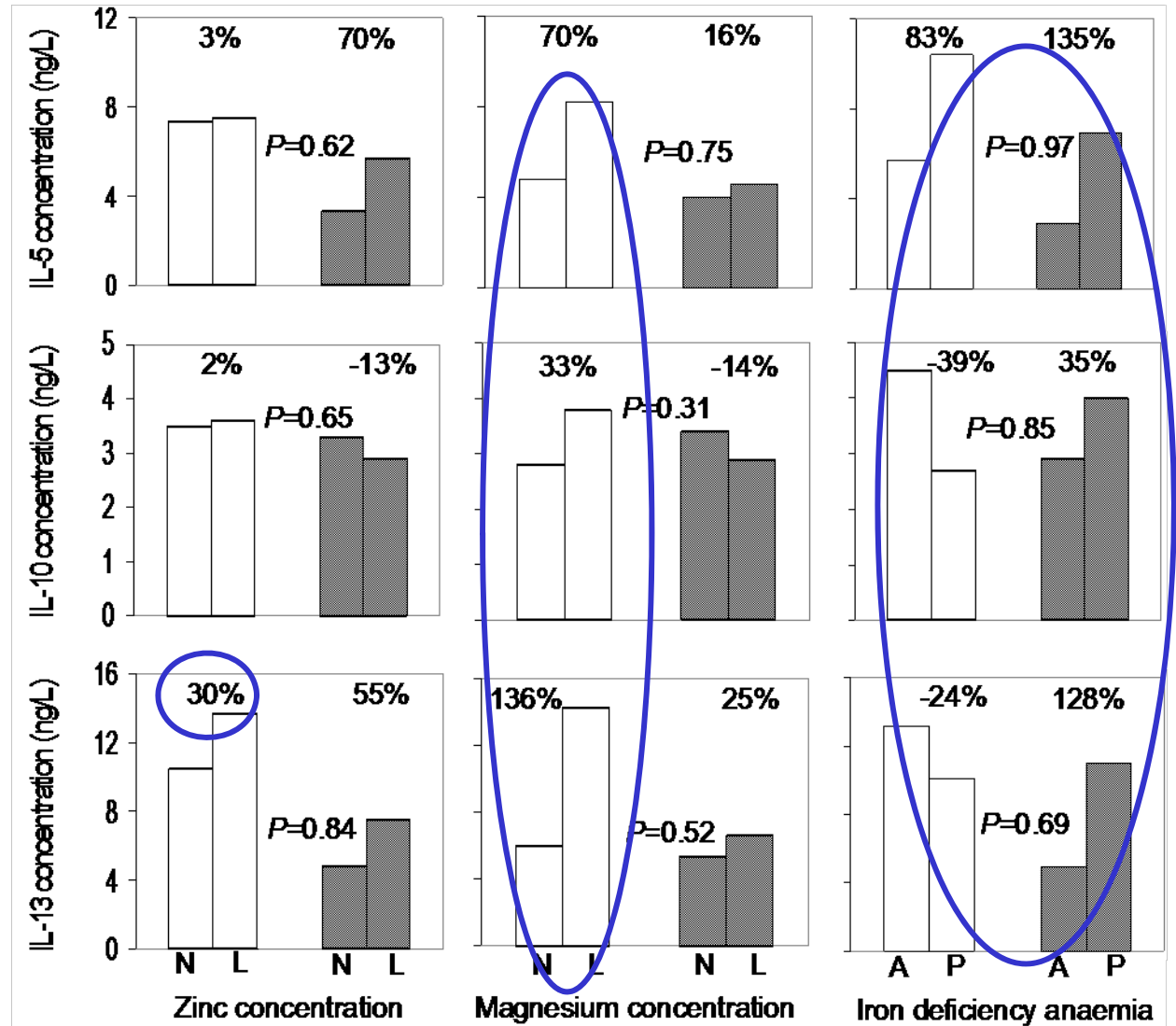
N = Normal conc.  
 L = Low conc.  
 A = Absent  
 P = Present

**Figure 1:** Associations between micronutrient status and supernatant type I cytokine concentrations following 7 days of stimulation

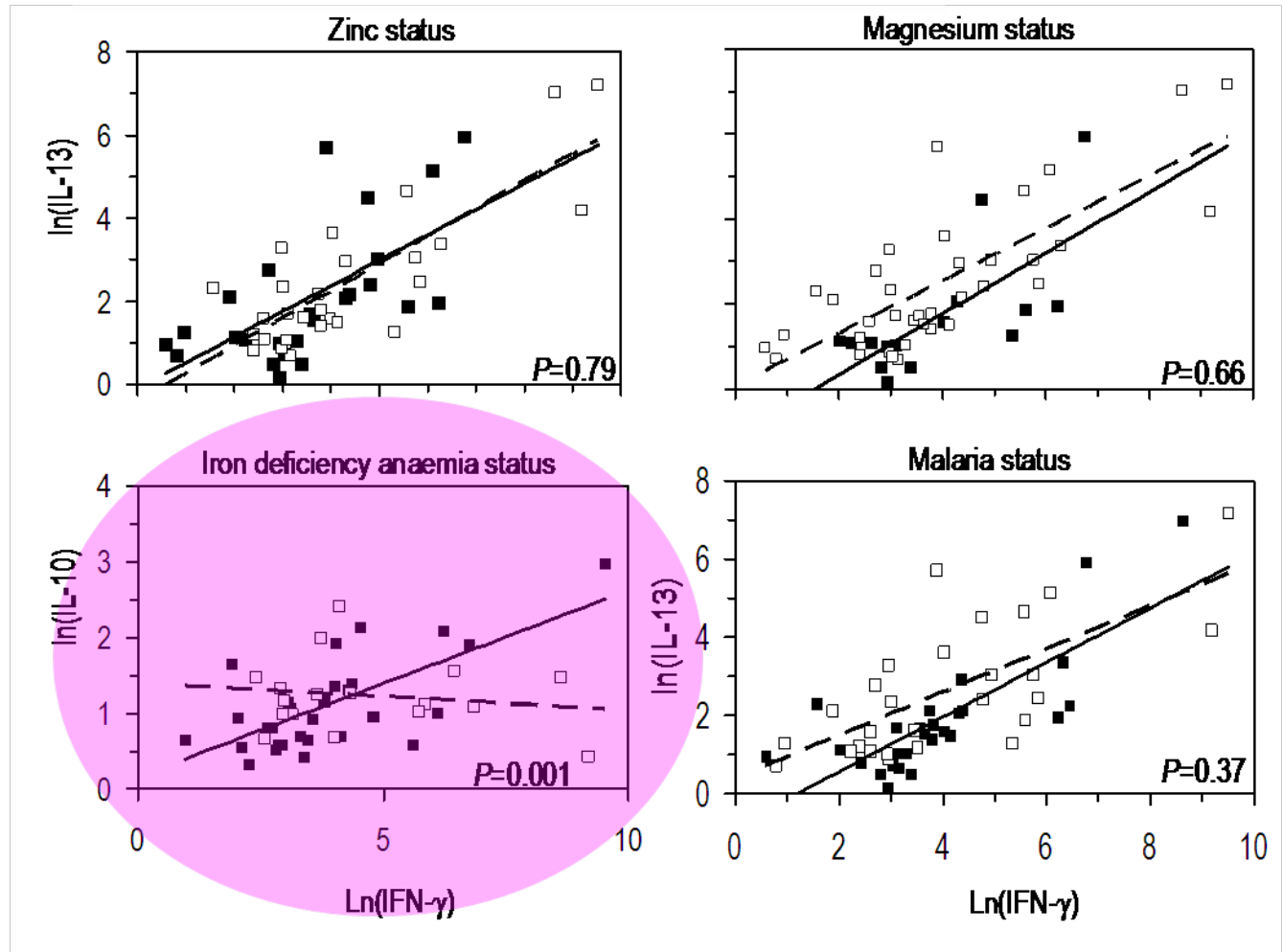


# Results-2

**Figure 2:**  
Associations between nutrient status and supernatant concentrations of type II cytokine



# Results – 3



**Figure 3:** Relationships between supernatant concentrations of IFN- $\gamma$ , IL-13 and IL-10 under different conditions of micronutrient and malaria status



# Discussion

- Zinc deficiency is associated with increased concentrations of TNF-alpha (37%; 95% CI: 14% to 118%) and IFN-gamma (74%; 95% CI: 24% to 297%) – Type I cytokines
- Magnesium deficiency is associated with decreased concentrations of TNF- alpha and IFN-gamma and increase in type II cytokines with strong evidence on the association with IL-13; 80%; 95% CI: 31% to 371%
- IDA is associated with increased concentrations of IL-12 (37%; 95% CI: 1% to 88%)
- Generally, IDA seem to be associated with variable concentration of both type I and II cytokines especially influenced with malaria infection status



# Conclusions

- Nutrients deficiencies may variably be associated with alteration in *in vitro* cytokine production
- Zinc deficiency and iron deficiency anaemia associate with a remarkable increases in type I cytokine production implying a shift in the balance of the cytokine response towards pro-inflammatory and cellular type response
- To reduce the pathological sequel of pro-inflammatory cytokines that may be associated with some nutrients deficiencies, we propose supplementation programs to primarily focus, among other nutrients, zinc, magnesium, and iron
- Proper selection of micronutrients, in right amounts will be ideal for efficient and balanced Th1 and Th2 cytokine responses since deficiencies seem to affect the production and not the association of these important infection protective tools



# Future perspectives

- Accomplish analysis and report the outcomes of the intervention study
- Continue with micronutrients and immune response studies as an approach to fight against malaria through improved body defence
- To mentor with other researchers particularly crop and soil scientists and design the best approach which can cheaply improve these nutrients in the consumables that suits the poor African communities;
- The use of the elements as possible components of fertilizers may be the best idea