Objectives

• To investigate the role of NALRP3 inflammasome in interleukin (IL)-1β production by peripheral mononuclear cells (PBMC’s) isolated from patients with inflammatory bowel disease (IBD)

• To compare IL-1β, IL-6 and TNFα production by PBMC’s of IBD patients and healthy controls after LPS stimulation.

METHODS: PBMCs were isolated from 11 healthy controls, 12 patients with Crohn disease (CD) and 10 patients with Ulcerative Colitis (UC) after gradient centrifugation of heparinised whole blood over Ficoll. PBMCs were stimulated with the TLR4 ligand lipopolysaccharide (LPS) and with the NLRP3 ligand monosodium urate (MSU). After 24-hour incubation, cytokine concentrations were measured in supernatants; after 4-hour incubation PBMCs were lysed for measurement of mRNA transcripts of interleukin (IL)-1β by RT-PCR.

RESULTS: As shown in the table, cytokine production was lower among patients with IBD than healthy controls (indicates significant differences compared to controls) and this was most prominent in CD. Activation of NLRP3 is expressed as the % increase of IL-1β production in the presence of MSU. Mean relative IL-1β copies in PBMC of controls, of UC and of CD were 3.12, 16.3 and 55.3 (pNS).

CONCLUSION: Cytokine production by PBMCs is significantly modulated in IBD. Main characteristics are down-regulation after LPS stimulation and activation of NLRP3 inflammasome.

Twelve patients with Crohn’s disease, 10 patients with Ulcerative Colitis and 11 age matched controls were asked to donate blood. The participants were recruited from the population attending the IBD outpatient clinic of ATTikon University Hospital and also from hospitalized patients. The study protocol was approved by the Ethics Committee of the ATTikon University Hospital and participants were asked to give their written informed consent. The inclusion criteria were:

• Patients with diagnosis of Crohn’s disease or Ulcerative Colitis with clinical, endoscopic and histological documentation.

• Production of IL-1β, IL-6 and TNFα was significantly lower in patients with IBD comparing to controls (figure 1).

• Activation of NALRP3 inflammasome, expressed as the % increase of IL-1β production in the presence of MSU, is observed in PBMC’s of patients with ulcerative colitis but not in PBMC’s of patients with Crohn’s disease (figure 1).

• Mean relative IL-1β copies in PBMC’s of controls, of UC and of CD were 3.12, 16.3 and 55.3 (pNS).

Conclusions

• Activation of NALRP3 inflammasome in PBMC’s of patients with ulcerative colitis is significantly higher than controls.

• Production of IL-1β, IL-6 and TNFα by LPS – stimulated PBMC’s is down-regulated in patients with IBD.

References


