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THU0201 ANALYSIS OF GUT MICROBIOTA DIVERSITY IN PATIENTS WITH RHEUMATOID ARTHRITIS AND BENEFICIAL EFFECTS OF PROBIOTICS ON INTESTINAL FLORA

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Background: Rheumatoid arthritis (RA) is a progressive, irreversible disease characterized by autoimmune imbalance. Recent studies have found that the number and type of intestinal flora in RA patients were changed (called disorders), which in turn leads to immune imbalance in RA patients. The breath test can indirectly assess the growth of bacteria in the intestine. This study aimed to investigate the intestinal flora imbalance in RA patients and the effect of probiotics on the intestinal flora of RA patients using methane and hydrogen breath test.

Objectives: To study the beneficial effects of probiotics on the intestinal flora of RA patients by detecting the concentrations of methane and hydrogen and the microbial populations in patients with rheumatoid arthritis (RA) before and after probiotic treatment.

Methods: The lactulose methane and hydrogen breath technique were used to detect and compare the concentration of methane and hydrogen in 36 patients with RA before and after probiotic treatment (mainly *Bifidobacterium*, *Clostridium butyricum*, *Lactobacillus*) for 3 weeks. At the same time, the 16S rRNA V3 region in stool samples of 17 patients with RA was qualitatively analyzed by Roche/45 high-throughput sequencing platform and compared with 9 healthy adult stool samples.

Results: Compared with pre-intervention, after 3 weeks of probiotic treatment, the exhaled methane concentration was significantly increased in RA patients at 30 minutes, 60 minutes, and 90 minutes ($P < 0.05$). The sum of methane and hydrogen concentrations increased at 30 minutes ($P = 0.041$). There was no significant difference in hydrogen concentration before and after intervention ($P > 0.05$) (Table 1). Compared with healthy adults, the fecal diversity index (Shannon) of RA patients increased ($P < 0.05$) and the species uniformity index (flatness) also increases.

Figure 1: Compared with healthy subjects (n=9), RA patients (n=17) increased the fecal diversity index, $P < 0.05$.

Abstract THU0201 – Figure 1

Conclusion: Patients with RA had a significant difference in the structure of intestinal flora compared with that of healthy controls, which may represent perturbations of microbial communities and contribute to the pathogenesis and condition of RA. Probiotics can restore the composition of the gut microbiome and introduce beneficial functions to gut microbial communities through diet, probiotics or fecal transplantation, providing a promising prospect for clinical treatment of RA.

Figure 2: Compared with healthy people (n=9), RA patients (n=17) The average index of feces increased, $P < 0.003$.

Abstract THU0201 – Figure 2

REFERENCES:

- [1] Schett I B M a G, *The pathogenesis of rheumatoid arthritis*. The New England Journal of Medicine, 2011. 365(23): p. Number of 2205–2219.
- [2] Edwards C J, *Commensal gut bacteria and the etiopathogenesis of rheumatoid arthritis*. J Rheumatol, 2008. 35(8): p. Number of 1477–1479.
- [3] Horta-Baas G, Romero-Figueroa MDS, Montiel-Jarquín AJ, Pizano-Zárate ML, García-Mena J, Ramírez-Durán N. Intestinal Dysbiosis and Rheumatoid Arthritis: A Link between Gut Microbiota and the Pathogenesis of Rheumatoid Arthritis. J Immunol Res. 2017;2017:4835189

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SLE, Sjögren's and APS – etiology, pathogenesis and animal models

THU0202 INTERFERON STIMULATED GENE 15 PROTECTS REGULATORY T CELL OF SYSTEMIC LUPUS ERYTHEMATOSUS PATIENTS FROM INTERFERON ALPHA-MEDIATED DEPLETION

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Background: Data on T regulatory (T reg) cells number and functions in patients with Systemic Lupus Erythematosus (SLE) are conflicting (1). Type I interferon (IFN) decreases Treg proliferation and induces Treg apoptosis (2). IFN stimulated gene 15 (ISG15) is an IFN-induced protein with a negative effect on IFN pathway.

Objectives: Aim of the study was to evaluate the effect of IFN alpha and ISG15 on Treg number and on STAT1 phosphorylation in patients with SLE.

Methods: We recruited patients with SLE classified according to 1997 ACR criteria. We collected demographic and clinical data including disease duration, disease activity scored according to *Systemic Lupus Erythematosus Disease Activity Index* (SLEDAI) 2K. Peripheral blood mononuclear cells (PBMC) were isolated from whole blood samples. Intracellular phosphorylated STAT1 (pSTAT) levels were evaluated in Treg (CD4+FOXP3+CD127 low cells) and conventional T cell (Tconv, CD4+FOXP3- cells) by multiparametric flow cytometry, *ex vivo* or after stimulation with recombinant IFN alpha. PBMC were cultured for 48 hours with anti-CD3 or anti-CD3 and IFN alpha; the frequency of Treg was analysed by multiparametric flow cytometry. ISG15 mRNA expression was evaluated by RT-PCR on PBMC from SLE patients. Data were expressed as mean $\pm$ standard deviation or median (interquartile range) according to the distribution; Mann-Whitney and Spearman tests were applied. P value < 0.05 were considered statistically significant.

Results: We enrolled 21 SLE patients [F:M 20:1; mean age 45.2 ± 12.9 years; mean disease duration 157.5 ± 103 months; median SLEDAI 2(4)]. Overall, median baseline ISG15 mRNA expression was 0.06 (IQR 0.16); patients were divided into ISG15-high (n=9) and ISG15-low (n=12), according to the mean mRNA expression. *Ex vivo*, after short-term treatment with IFN alpha (15 and 25 minutes), we observed a significant increase in STAT1 phosphorylation compared to baseline both in Treg and Tconv ($p < 0.00001$), in ISG15-high and ISG15-low patients (Figure 1 A and B); however, high ISG15 levels protect both Treg and Tconv from STAT1 phosphorylation (Figure 1 C). After 48 of culture with IFN alpha, we observed a decrease in Treg and Tconv number, significantly greater in ISG15-low cells (Figure 1D). Treg frequency and ISG15 expression in Treg were higher in active vs inactive SLE patients [SLEDAI > 4 vs SLEDAI < 4] ($p < 0.05$).

Conclusion: ISG15 protects T conventional cells from STAT1 phosphorylation and induces resistance to T regulatory cells depletion. The results of the study suggest that ISG15, a protein induced by IFN in the acute phase of the disease, exerts a negative feedback, allowing the recovery of T reg.