IL-27-producing CD14+ cells infiltrate inflamed joints of rheumatoid arthritis and regulate inflammation and chemotactic migration

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<Introduction>
Rheumatoid arthritis (RA) is a chronic inflammatory autoimmune disease, and has systemic inflammatory arthritis with hypertrophy of the synovium followed by the destruction of cartilage, bone, and joint structures. Several in vivo experimental autoimmune animal models and in vitro human studies have suggested that interleukin (IL)-17-secreting helper T (Th17) cells can be considered a critical mediator of RA with respect to tissue inflammation or bone resorption. Th17 cells specifically express chemokine receptor CCR6, and its ligand CCL20 recruits Th17 cells. IL-27 is a recently identified cytokine, which is structurally related to IL-12 as a heterodimeric cytokine, similar to IL-23 and IL-35. IL-27 is composed of Epstein–Barr virus-induced gene 3 (EBI3), a p40-related molecule, and IL-27Rβ, a p35-related molecule.[1] IL-27 is produced by antigen presenting cells (APCs) including dendritic cells (DCs) and monocytes, and is secreted as a heterodimer. IL-27 is reported to be expressed at chronic inflammatory sites, such as synovial tissues in RA, skin lesions in psoriasis, inflamed intestine in Crohn’s disease, and granulomas in tuberculosis or sarcoidosis. IL-27 suppresses the development of Th17 cells and the production of cytokines including IL-17A from activated CD4+ T cells by blocking RAR-related orphan receptor C (RORC) expression in humans and in several experimental animal models, while it induces Th1 differentiation. IL-27 receptor (IL-27R) is also a heterodimer of WSX-1 (IL-27Rα), a homologue of IL-12R β2, and gp130, a component of the IL-6 receptor. Both components are coexpressed in various human cell types, and are required for the signal transduction of IL-27. In a marine collagen-induced arthritis (CIA) model, administration of IL-27 reduces the amount of serum IL-6, cellular infiltration to the joints, synovial hyperplasia and joint erosion at the onset of the disease. IL-27 also inhibits human receptor of NF-κB ligand (RANKL)-mediated osteoclastogenesis. These reports suggest protective roles for IL-27 in the pathogenesis of arthritis. In contrast, it is also reported that IL-27 possesses pathogenic roles in RA. In several experimental animal models, IL-27 is critically involved with the development of arthritis by inducing the differentiation of naive T cells into interferon (IFN)-γ-producing Th1 cells. In humans, high concentrations of IL-27 induce the production of IL-6 and inflammatory chemokines from FLSs of established RA.[2] These conflicting reports reflect the complex functions of IL-27 in human immunology. In this study, we investigated the source and role of IL-27 in RA.

<Material and methods>
Ethics approval for this study was granted by the ethics committee of Kyoto University Graduate School and Faculty of Medicine, and written consent was obtained for every sample. Informed consent was obtained from 69 patients (28 with RA, 26 with knee osteoarthritis (OA), and 15 as healthy volunteers (HV)); RA and OA were diagnosed according to the criteria of the American College of Rheumatology. Peripheral blood and synovial fluid samples of these patients were collected. After centrifugation, the supernatants were used for ELISA, and MNCs, which were isolated from the pellets, were cultured and used for ELISA or flow cytometry. Human synovial tissues were also obtained during total knee replacement surgery from Kyoto University Hospital from RA and OA patients. A piece of synovial tissues were frozen, and FLSs were prepared as previously reported.[3] Frozen synovial tissues and FLSs were used for immunohistochemical staining. The supernatants of cultured FLSs were collected for ELISA.

<Results>
1. The IL-27 level in synovial fluid of RA patients (mean 0.13 ng/ml; range 0.017–0.37 ng/ml) was significantly higher (P < 0.0005) that in OA patients (mean 0.003 ng/ml; range 0–0.033 ng/ml). However, the concentrations of plasma IL-27 in RA patients (mean 0.4 ng/ml; range 0.17–0.86 ng/ml), OA patients (mean 0.34 ng/ml; range 0.017–0.83 ng/ml) and HV (mean 0.48 ng/ml; range 0.09–1.0 ng/ml) were similar.
2. RA FLSs stimulated with proinflammatory cytokines such as TNF-α, IL-1β, IL-6 or IL-17A failed to produce detectable IL-27. Stimulated MNCs from synovial fluid and peripheral blood produced IL-27 protein. Interestingly, the production of IL-27 by MNCs in RA synovial fluid was significantly lower than that by MNCs from peripheral blood from RA and OA patients.
3. We stained MNCs with anti-CD4, -CD8, -CD14, or -CD19, and intracellular anti-IL-27. Only CD14+ cells were IL-27 positive.

<Discussion>
Circulating IL-27-producing CD14+ cells significantly infiltrate into inflamed regions such as RA synovium and have anti-inflammatory effects in several ways: both directly through the reduction of IL-6 production, and possibly through the induction of Th1 development and the suppression of Th17 development; and indirectly by regulation of recruitment of CCR6+ cells, such as Th17 cells, through the suppression of CCL20 production. Our results suggest that such a serial negative feedback system could be applied to RA.

<References>