Electromagnetic Fields modulate HLA-G Molecules in Osteoarthritic Synovial Fibroblasts

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ABSTRACT INTRODUCTION:
Several studies suggest that a certain degree of inflammation and altered synovial fibroblasts (SFs) activities including secretion of pro-inflammatory mediators, such as cytokines and lipid mediators of inflammation, contribute to the development of osteoarthritis (OA). HLA-G antigens are non-classical HLA-class I molecules characterized by a low allelic polymorphism, a limited tissue distribution in non-pathological conditions and the presence of membrane bound and soluble isoforms. The HLA-G antigens were firstly detected in cytotrophoblast cells at the feto-maternal interface where they maintain a tolerogenic status between the mother and the semiallogenic fetus. Besides, several studies point out to a broader immunoregulatory role for this molecule and HLA-G expression has been associated to different inflammatory and autoimmune processes including rheumatoid arthritis. Further, recently both membrane and soluble HLA-G molecules expression has been reported in OASFs [1]. In vitro, ex vivo and in vivo data suggest that electromagnetic fields (EMFs) may represent a potential therapeutical approach to limit cartilage degradation and to control inflammation associated to OA, by modulating both chondrocyte activities as well as inflammatory properties [2]. From this background, the first aim of this study was to investigate if EMFs might modulate HLA-G expression in human OASFs. Furthermore, we investigated if the possible changes in HLA-G levels might be related to changes in interleukin 10 (IL-10) expression, an anti-inflammatory cytokine mainly involved in HLA-G upregulation and which is modulated by EMFs (submitted article).

METHODS:
Patients and OASFs cultures. Synovial tissue was obtained from 6 patients (2 males and 4 females, mean age 70±10 years, range, 59-82 years) with end-stage OA undergoing total hip joint replacement surgery. Cells were isolated from fresh synovial tissue and characterized, as previously described [1]. Synovial cells at the 3rd passage were used in the experiments. OASFs were unexposed (control) or exposed to EMF exposure (75 Hz, 1.5 mT) (Igea, Carpi, Italy), for 24 or 48 hours. HLA-G detection. HLA-G expression in OASFs was detected by immunofluorescence [1]. The levels of soluble HLA-G were analyzed by immunoenzymatic assay (ELISA) in the OASFs culture supernatants. IL-10 detection. IL-10 concentrations were determined by ELISA using the commercially available Human IL-10 BioSource Immunoassay Kit (Human IL-10 US; BioSource, Camarillo, CA).

RESULTS SECTION:
The presence of HLA-G molecules in our OASF cultures was confirmed by immunofluorescence, as shown in Fig. 1. When the cells were exposed to EMF, the soluble HLA-G (sHLA-G) levels in the OASFs culture supernatants increased of 15% and 59% respect to the unexposed control cells, after 24 and 48 hours exposure respectively. As IL-10 is one of the main up-modulator of sHLA-G production, we evaluated also the levels of IL-10 in our EMF exposed and unexposed OASF cultures. We found that IL-10 production was higher in EMF exposed cells of 13% and 40% after 24 and 48 hours exposure, compared to control cells, similarly to what observed for HLA-G molecules.

DISCUSSION:
In this study we evaluated the effects of EMF exposure on the modulation of HLA-G molecules in OASFs.

As recently reported, the expression and production of membrane and soluble HLA-G molecules is higher in OASFs in comparison with control SFs [1]. In our experimental conditions, 24 and 48 hours EMF exposure increased the HLA-G levels in the OASFs culture supernatants in a time-dependent fashion. It has been shown that EMFs display anti-inflammatory effects in human OASFs, and that they are in part mediated by the inhibition of proinflammatory mediators such as IL-6, IL-8 and prostaglandine E2 and by the stimulation of IL-10 release. The results of this study show a role of EMF in increasing IL-10 levels in OASFs. As IL-10 is a pleiotropic cytokine with anti-inflammatory properties, negatively correlated with joint destruction and it represents the main up-modulator of sHLA-G production, we speculate that EMF might modulate HLA-G production via IL-10. The results of these data add new information concerning the EMF anti-inflammatory activities.

SIGNIFICANCE:
EMF exposure increase the production of HLA-G molecules in OASFs. HLA-G levels are modulated by the cytokine IL-10. The anti-inflammatory activity of EMF is related to the enhancement of both IL-10 and HLA-G levels in OASFs, suggesting a possible new mechanism to counteract the synovial joint inflammation in OA.

REFERENCES: