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Inflammasome as a Novel Regulator of Human Mesenchymal Stem Cell Function

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Introduction: Inflammasome is a multimeric protein complex which senses inflammatory stimuli. Mesenchymal stem cells (MSCs) are promising tool for regenerative medicine and immune-related diseases. Previous reports demonstrate that MSC function can be altered by stimuli derived from innate and adaptive immunity. In the present study, we investigated the expression and functional regulation of inflammasomes in human umbilical cord blood-derived MSCs (hUCB-MSCs).

Materials and Methods: The expression of inflammasome complex genes in hUCB-MSCs was detected by RT-PCR. Change in the expression of surface markers after inflammasome activation was analyzed by flow cytometry. Proliferation, pyroptosis differentiation and immunomodulation of hUCB-MSCs after inflammasome activation were assessed. Therapeutic efficacy of inflammasome-activated hUCB-MSCs was determined in mouse colitis model induced by dextran sulfate sodium.

Results: The hUCB-MSCs expressed the components of inflammasomes. Among several types of inflammasomes, NLRP activation did not alter the characteristics of hUCB-MSCs nor induced the pyroptosis of MSCs. Surprisingly, NLRP activation promoted the proliferation and osteogenic differentiation of hUCB-MSCs. Moreover, the immunomodulatory effects of MSCs on T cell proliferation, dendritic cell (DC) and regulatory T cell were changed in response to NLRP activation. In addition, the expression levels of immunomodulatory factors were elevated in hUCB-MSC after NLRP stimulation.

Conclusions: In conclusion, for the first time, our data suggest that NLRP, one of the inflammasome family, is expressed

in hUCB-MSCs and its activation can regulate the functions of hUCB-MSCs along with the up-regulation of multiple soluble factors.

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Evaluation of a SYBR Green Real-Time PCR Assay for Quantification of Major Cytokines in PBMC from Bovine

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Introduction: Cytokines are important in host responses to infection, immune responses, inflammation, trauma, sepsis, cancer, and reproduction. Currently, SYBR Green quantitative real time PCR (qRT-PCR) is the most general method used to investigate the expression pattern of cytokine genes in ruminants [1]. The objective of this study was to determine the expression level of systemic inflammatory markers in peripheral blood mononuclear cells (PBMC) from bovine [2].

Materials and Methods: PBMC were isolated from heparinized whole blood by density gradient centrifugation. Total RNA was extracted and converted to complementary DNA for qRT-PCR analysis.

Results: Primer sets were designed to quantify cytokines, their related genes and reference genes in bovine. All the primer sets were compartmented to exon-exon boundaries and used the similar hybridization temperature. And also, we investigated total relative expression of cytokine and their-related genes in PBMC from bovine. Results of these experiments indicated that expression levels of pro-inflammatory markers such as interleukin-8, 13, 17, 23 and CCL-8 increased in PBMC isolated from bovine, excluding tumor necrosis factor- α , interleukin-2, 12(p40), 16, CCL-3 and interferon- γ .

Conclusions: Based on the above findings, it could be concluded that the experiment technique was a method to measure that the relative quantification of cytokine genes expression compared with precise normalization by reference genes. We speculated that qRT-PCR assay could be used to investigate ruminant immune responses, and also it would be widely achievable to the veterinary investigation community.

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