



NCX1 disturbs calcium homeostasis and promotes RANKL-induced osteoclast differentiation by regulating JNK/c-Fos/NFATc1 signaling pathway in multiple myeloma

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Abstract

Although several types of calcium channels abnormalities have been shown to promote myeloma bone disease (MBD), the relationship between Na⁺/Ca²⁺ exchanger 1 (NCX1) and MBD remains unexplored. Here, we examined the role of NCX1 in the development of multiple myeloma (MM), with a special focus on the underlying effects involved osteoclast differentiation. Firstly, we detected NCX1 protein highly expressed in BM tissues of MM patients, and its expression was positively correlated with serum calcium and the percentage of BM CD138⁺ cells. In vitro, NCX1 suppression with the inhibitor KB-R7943 reduced cell viability of MM cells and caused apoptosis. Extracellular high Ca²⁺ environment increased the level of intracellular Ca²⁺ in MM cells through gating the calcium influx, with subsequently promoting the expression of NCX1 and osteoclastogenesis-related genes (receptor activator of nuclear factor-κB (RANKL), nuclear factor of activated T cell cytoplasmic 1 (NFATc1), and proto-oncogene Fos (c-Fos). This phenomenon could be reversed by KB-R7943 or calcium chelation. Furthermore, NCX1 overexpression in MM cells accelerated osteoclastogenesis, while NCX1 knockdown or suppression resulted in the opposite effect. Mechanistically, we further investigated the related mechanisms of NCX1 regulating osteoclast differentiation using RNA sequencing, western blotting and Enzyme linked immunosorbent assay, and found that NCX1 modulated osteoclast differentiation in MM through JNK/c-Fos/NFATc1 signaling pathway. In conclusion, the Ca²⁺/NCX1-mediated signaling participates in the osteoclasts-myeloma cell interactions, which represents a promising target for future therapeutic intervention in MBD.

Keywords NCX1 channel · Calcium · Multiple myeloma · Osteoclast differentiation

Abbreviations

BM Bone marrow

CM Conditioned medium

[Ca²⁺]_o Extracellular calcium

DAB Diaminobenzidine

F-actin Fibrous actin

GSEA Gene Set Enrichment Analysis

IHC Immunohistochemistry

M-CSF Macrophage-colony stimulating factor

NCX Sodium/calcium exchanger

NES Normalized enrichment scores

PBL Peripheral blood

RNA-seq RNA sequencing

TNF Tumor necrosis factor

CCK-8 Cell Counting Assay Kit-8

[Ca²⁺]_i Intracellular calcium

c-Fos Proto-oncogene Fos

DAPI 4',6-Diamidino-2-phenylindole dihydrochloride

GEP Gene expression profiling

HBSS D-Hanks Balanced Salt Solution

MBD Myeloma bone disease

MM Multiple myeloma

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