# Number of circulating CD14-positive cells and the serum levels of TNF-$\alpha$ are raised in acute charcot foot.

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OBSERVATIONS

# Number of Circulating CD14-Positive Cells and the Serum Levels of TNF-α Are Raised in Acute Charcot Foot

Charcot neuro-osteoarthropathy (CNO) is a devastating complication of diabetes characterized by increased local bone resorption mediated by osteoclasts. Often CNO leads to multiple fractures and joint destruction resulting in severe deformity of the foot. Osteoclasts are multinucleated cells that have a common precursor with monocytes/macrophages (1). Previously, we reported that the number of osteoclasts generated in vitro from circulating cells was higher in acute CNO patients compared with diabetic control subjects and was partially independent from receptor activator for NF-κB (RANK)-RANK ligand (RANKL), the main osteoclastogenic mediator (2). One hypothesis could be that the number of circulating osteoclast precursors is increased in CNO. Among all monocytes subpopulations, CD14-positive cells are the most potent to transform into bone-resorbing osteoclasts (3).

We studied 11 diabetic patients with recent onset of acute CNO, 10 diabetic patients with no previous history of CNO, and 6 healthy control participants. All patients were matched for age, sex, and duration of diabetes. Mean serum creatinine and vibration perception threshold on the apex of the hallux were not different between CNO and diabetic patients. Patients with bone diseases (osteoporosis, rheumatoid arthritis, Paget's disease) were excluded from this study. This study had research ethics committee approval and was carried out in accordance with the Declaration of Helsinki. Peripheral blood mononuclear cells were isolated and the number of CD14-positive monocytes was determined by flow cytometry. Serum concentration of tumor necrosis factor (TNF)-α, interleukin-1β, and LIGHT were determined by ELISA. Data were compared using Mann-Whitney *U* test and linear regression analysis.

The percentage of CD14-positive cells in CNO was significantly increased by 1.7–fold (9.06 ± 3.64 vs. 4.98 ± 2.68%, $P = 0.012$) and 2.1–fold (9.06 ± 3.64 vs. 4.0 ± 2.0%, $P = 0.016$) compared with diabetic patients and healthy participants, respectively. Compared with diabetic patients and healthy participants, in CNO serum levels of TNF-α were significantly increased by 1.7–fold (4.3 ± 0.9 vs. 2.43 ± 0.3 pg/mL, $P = 0.014$) and 2.2–fold (4.3 ± 0.9 vs. 1.93 ± 0.8 pg/mL, $P = 0.009$). In CNO, a strong correlation was encountered between the percentage of CD14-positive cells and the serum levels of TNF-α ($R = 0.78$). LIGHT was only detectable in the serum of six acute CNO patients but was increased significantly compared with diabetic patients ($P = 0.018$) and healthy participants ($P = 0.018$). A poor correlation ($R = 0.02$) between the percentage of CD14-positive cells and the serum levels of LIGHT was observed in CNO. On the other hand, although interleukin-1β was detectable in eight acute CNO patients, six diabetic patients, and two healthy volunteers, these levels were not significantly different between the three groups of patients, and no correlation was observed between the number of CD14-positive cells and serum levels of this mediator.

Our results suggest that the excessive bone resorption in CNO could potentially be linked to increases in circulating osteoclast precursors and serum levels of TNF-α. With the recent development of anti-TNF biological therapies and based on our findings, it is plausible to suggest the use of this therapeutic approach for controlling CNO.

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**References**

1. Udagawa N, Takahashi N, Akatsu T, et al. Origin of osteoclasts: mature monocytes and macrophages are capable of differentiating into osteoclasts under a suitable microenvironment prepared by bone marrow-derived stromal cells. Proc Natl Acad Sci USA 1990;87:7260–7264
2. Mabilleau G, Petrova NL, Edmonds ME, Sabokbar A. Increased osteoclastic activity in acute Charcot's osteoarthropathy: the role of receptor activator of nuclear factor-kappaB ligand. Diabetologia 2008;51:1035–1040
3. Husheem M, Nyman JK, Vääräniemi J, Vaananen HK, Hentunen TA. Characterization of circulating human osteoclast progenitors: development of in vitro resorption assay. Calcif Tissue Int 2005;76:222–230