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Cathepsin K in treatment monitoring following intravenous zoledronic acid

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Abstract. Cathepsin K (CatK) is mainly expressed by osteoclasts and plays an important role in bone resorption. As CatK is expressed and secreted by osteoclasts during active bone resorption, it may be a useful and specific biochemical marker of osteoclastic activity. Therefore, CatK serum levels were studied for monitoring the treatment of females with postmenopausal osteoporosis by zoledronic acid. The serum CatK levels were determined in nine postmenopausal females before and after 3, 6 and 12 months of treatment. The levels were significantly reduced after 3 and 6 months ($P<0.05$), whereas they returned to baseline after 1 year. Taken together, the serum level of CatK may be suitable for monitoring anti-osteoporotic therapy in association with treatment response.

Introduction

Osteoporosis is a systemic skeletal disease characterized by low bone mass and a deterioration of trabecular microarchitecture, resulting in an increase in bone fragility and a tendency to fractures (1). Osteoporosis is standardly diagnosed via the measurement of bone mineral density (BMD) by dual-energy X-ray absorptiometry (DXA). The operational World Health Organization definition of osteoporosis is based on a T-score of ≤-2.5 (2). Bone turnover markers (BTM) do not establish the diagnosis of osteoporosis. While DXA reflects changes in BMD 1-2 years after anti-osteoporotic treatment, BTM levels were altered after 3-6 months of therapy so they may provide earlier information on the response to treatment (3-7). In the present study, the clinical potential of cathepsin K (CatK) serum levels for monitoring the treatment of osteoporosis with zoledronic acid is studied.

The prevalence of osteoporosis in Germany among persons aged ≥50, as revealed by diagnoses of osteoporosis or osteoporotic fractures, or by the prescription of medication for osteoporosis, was found to be 14% in the year 2009. The gender-specific prevalence was 24% in women and 6% in men (8).

CatK is mainly expressed in mature osteoclasts, cleaves both helical and telopeptide regions of collagen I and is therefore essential for bone resorption (9). This cysteine protease was first described by Inaoka *et al* (10). Deficiency of CatK results in pyknodysostosis (Toulouse Lautrec syndrome), an autosomal recessive osteosclerotic skeletal dysplasia with impaired bone resorption, which is characterized by decreased bone turnover and an accumulation of undigested collagen fibrils (11). Saftig *et al* (12) showed that in CatK-deficient mice impaired osteoclastic bone resorption leads to osteopetrosis. Due to the fact that CatK is expressed and secreted by osteoclasts during active bone resorption, it may be a useful and specific biochemical marker of osteoclastic activity. Henriksen *et al* (13) postulated that circulating levels of CatK are proportional to the number of osteoclasts, and thus can be used as a surrogate marker of osteoclast number.

Materials and methods

Study subjects. In total, nine postmenopausal females aged 62-75 years were included. Of these, seven females could be monitored completely with DXA before and 1 year after anti-osteoporotic therapy with intravenous infusion of zoledronic acid, and furthermore, the levels of serum CatK before and 3, 6 and 12 months after therapy could be assessed. Two individuals had missing observations after 1 year. The indication for anti-osteoporotic treatment was according to the guidelines of the Dachverband Osteologie (DVO) by the lowest T-score in DXA and associated risk factors (14). Surgery or fracture ≤12 months, malignant tumor, ovariectomy and the intake of drugs prior to the beginning of treatment, including

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cortisone, strontium, fluorides, bisphosphonates, selective estrogen receptor modulators, estrogens and steroids, were exclusion criteria.

Ethical approval. The study was conducted according to the ethical guidelines of the Declaration of Helsinki as revised in 1989 and approved by the Ethical Committee of the Otto von Guericke University (91/08) and the German Office of Radiation Protection in Salzgitter (no. Z5-22462/2-2009-022).

BMD measurements. All the female patients underwent bone density measurements before and 1 year after treatment. Radiographs of the spine were not performed, as there were no references for vertebral fractures, such as back pain and loss of height, in the medical history or examination. The measurement of BMD was performed at the lumbar spine (L1-L4), femoral neck and total hip with DXA as described previously (15).

Laboratory. Serum parameters, according to the DVO guidelines, were examined to exclude secondary osteoporosis, and furthermore, 25-(OH)-vitamin D. The serum samples were aliquoted and stored at -80˚C until analysis. The measurement of CatK was performed before and 3, 6 and 12 months after the beginning of treatment [ELISA; Biomedica, Wien, Austria; intra-assay coefficient of variance (CV) 4-6%, inter-assay CV 6-8%, detection level 1.1 pmol/l] as described previously (15).

Anti-osteoporotic therapy. A total of 100 ml (5 mg) zoledronic acid (Aclasta®; Novartis, Basel, Switzerland) was infused intravenously within 15 min. In all females, calcium-supplementing was completed with 500 mg daily. Individually, a daily supplementation of 400-2,000 U of vitamin D3 was carried out.

Statistical analysis. The statistical analyses were performed with SPSS (version 21; SPSS, Inc., Chicago, IL, USA) using non-parametric tests, as graphical inspection indicated deviations from the normal distribution. Data for the BMD were evaluated with the Wilcoxon signed rank test due to the small random sample. The serum parameters with four time measurements were analysed by Friedman test. The individual time points were compared to the baseline value with the Wilcoxon matched-pairs signed rank test. A two-sided P-value of 0.05 was set to be the level of significance and was considered to indicate a statistically significant difference. Bonferroni adjustment was performed to control for family-wise error rate. The results are illustrated in median, minimum and maximum.

Results

CatK serum levels. Overall, no significant changes in the lowest T-Score were observed 1 year after anti-osteoporotic treatment (P=0.523). A significant improvement was only observed in the lumbar spine (P=0.008). Serum CatK presented a significant change over the four visits (P=0.003). In particular, there was a significant decrease in the first 3 (P=0.023) and 6 months (P=0.012) after the start of therapy. One year after the initiation of treatment, the CatK levels almost reached the base level (P=0.469) (Fig. 1). Individual serum CatK levels at 3, 6 and 12 months after antiresorptive therapy are shown in Fig. 2.

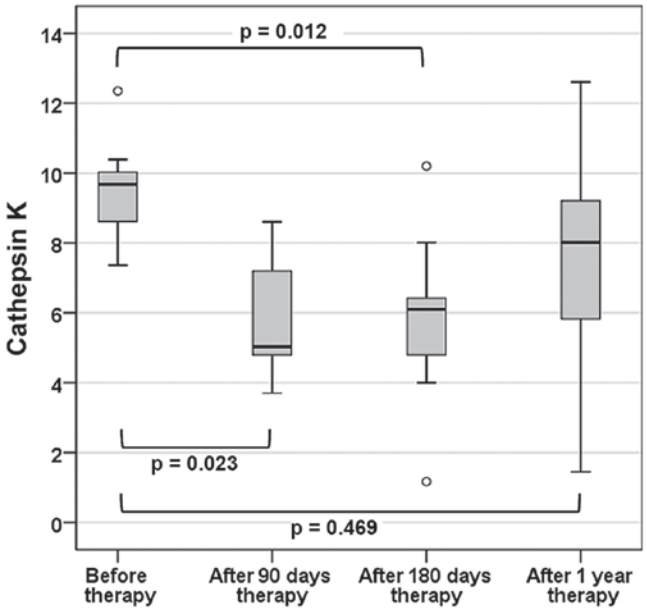

Figure 1. Cathepsin K (pmol/l) levels in the course of one year for all females (n=9). Boxplot representation for whole group (˚circles representing outliers).

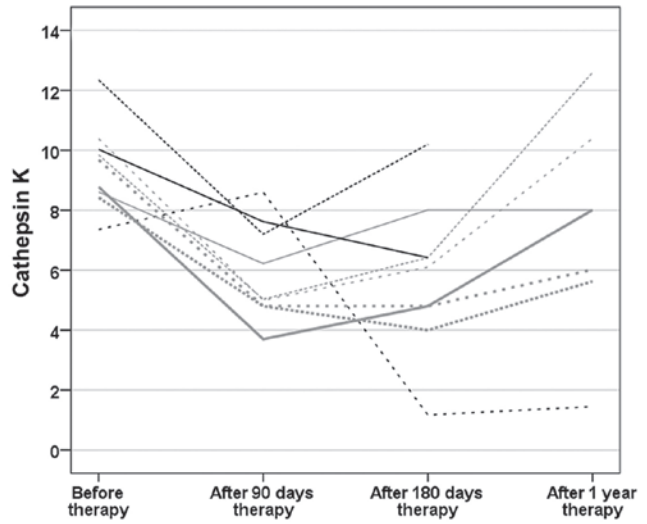

Figure 2. Individual courses of therapy and cathepsin K levels (pmol/l).

The strongest decrease in the majority of cases was observed after 3 months. In one case, the decrease set on 6 months after therapy was started, with low residual values. In the other females the CatK serum levels increased after 1 year following intravenous zoledronic acid.

Discussion

The study by Holzer *et al* (16) monitored serum CatK concentrations of 18 patients for 14.25±11.57 months (range, 1-48 months) after the start of osteoporosis treatment (oral bisphosphonate plus a combination of calcium and vitamin D). Serum CatK concentrations clearly decreased (mean at initial examination, 7.10±3.8 pmol/l; after treatment, 5.9±3.4 pmol/l), but did not demonstrate significance (t=1.362, df=17, P=0.191). Meier *et al* (17) examined longitudinally 21 females with

postmenopausal osteoporosis and 10 patients with Paget's disease of bone. All the patients started on oral (alendronate or risedronate) or intravenous bisphosphonate (pamidronate or zoledronate) treatment and were followed prospectively over 6 months. In postmenopausal osteoporotic females, oral and intravenous bisphosphonate treatment resulted in a significant reduction in serum CatK levels (P=0.03) with the majority of the effect occurring after 1 month (mean % change, -33%). In patients with mild Paget's disease, serum CatK levels decreased during bisphosphonate treatment. The study by Munoz-Torres *et al* (18) included 46 postmenopausal females with osteoporosis, which were treated with oral bisphosphonate (alendronat) plus calcium and vitamin D. The serum CatK levels gradually decreased after alendronate treatment (17, 22 and 41% at 3, 6 and 12 months, respectively; P<0.01). According to the current literature and the data of the present explorative analysis, serum CatK appears to be a putative marker for monitoring and controlling anti-osteoporotic treatment. The fast decrease of serum levels in the majority of the study subjects, already after 3 months, may provide early information on the response to treatment. Future studies are required to explore whether this information can be used for early allocation of patients to different treatments.

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References

1. No authors listed: Consensus Development Conference on Osteoporosis. Hong Kong, April 1-2, 1993. Am J Med 95: S1-S78, 1993.
2. No authors listed: Assessment of fracture risk and its application to screening for postmenopausal osteoporosis. Report of a WHO Study Group. World Health Organ Tech Rep Ser 843: 1-129, 1994.
3. Delmas PD: Markers of bone turnover for monitoring treatment of osteoporosis with antiresorptive drugs. Osteoporos Int 11 (Suppl 6): S66-S76, 2000.
4. Delmas PD, Eastell R, Garnero P, Seibel MJ, Stepan J; Committee of Scientific Advisors of the International Osteoporosis Foundation: The use of biochemical markers of bone turnover in osteoporosis. Committee of Scientific Advisors of the International Osteoporosis Foundation. Osteoporos Int 11 (Suppl 6): S2-S17, 2000.
5. Delmas PD, Hardy P, Garnero P and Dain M: Monitoring individual response to hormone replacement therapy with bone markers. Bone 26: 553-560, 2000.
6. Seibel MJ: Biochemical markers of bone remodeling. Endocrinol Metab Clin North Am 32: 83-113, 2003.
7. Szulc P: The role of bone turnover markers in monitoring treatment in postmenopausal osteoporosis. Clin Biochem 45: 907-919, 2012.
8. Hadji P, Klein S, Gothe H, Häussler B, Kless T, Schmidt T, Steinle T, Verheyen F and Linder R: The epidemiology of osteoporosis - Bone Evaluation Study (BEST): an analysis of routine health insurance data. Dtsch Arztebl Int 110: 52-57, 2013.
9. Troen BR: The role of cathepsin K in normal bone resorption. Drug News Perspect 17: 19-28, 2004.
10. Inaoka T, Bilbe G, Ishibashi O, Tezuka K, Kumegawa M and Kokubo T: Molecular cloning of human cDNA for cathepsin K: novel cystein protease predominantly expressed in bone. Biochem Biophys Res Commun 206: 89-96, 1995.
11. Gelb BD, Shi G, Chapman HA and Desnick RJ: Pycnodysostosis, a lysosomal disease caused by cathepsin K deficiency. Science 273: 1236-1238, 1996.
12. Saftig P, Hunziker E, Wehmeyer O, Jones S, Boyde A, Rommerskirch W, Moritz JD, Schu P and von Figura K: Impaired osteoclastic bone resorption leads to osteopetrosis in cathepsin-K-deficient mice. Proc Natl Acad Sci USA 95: 13453-13458, 1988.
13. Henriksen K, Tanko LB, Qvist P, Delmas PD, Christiansen C and Karsdal MA: Assessment of osteoclast number and function: application in the development of new and impoved treatment modalities for bone diseases. Osteoporos Int 18: 681-685, 2007.
14. Dachverband Osteologie e.v.: DVO-Leitlinie 2009 zur Prophylaxe, Diagnostik und Therapie der Osteoporose bei Erwachsenen. Osteologie 18: 304-328, 2009.
15. Adolf D, Wex T, Jahn O, Riebau C, Halangk W, Klose S, Westphal S, Amthauer H, Winckler S and Piatek S: Serum cathepsin K levels are not suitable to differentiate women with chronic bone disorders such as osteopenia and osteoporosis from healthy pre- and postmenopausal women. Maturitas 71: 169-172, 2012.
16. Holzer G, Noske H, Lang T, Holzer L and Willinger U: Soluble cathepsin K: a novel marker for the prediction of nontraumatic fractures? J Lab Clin Med 146: 13-17, 2005.
17. Meier C, Meinhard U, Greenfield JR, De Winter J, Nguyen TV, Dunstan CR and Seibel MJ: Serum cathepsin K concentrations reflect osteoclastic activity in women with postmenopausal osteoporosis and patients with Paget's disease. Clin Lab 52: 1-10, 2006.
18. Munoz-Torres M, Reyes-Garcia R, Mezquita-Raya P, Fernandes-Garcia D, Alonso G, Luna Jde D, Ruiz-Requena ME and Escobar-Jimenez F: Serum cathepsin K as a marker of bone metabolism in postmenopausal women treated with alendronate. Maturitas 64: 188-192, 2009.