



BONE RESORPTION MARKER BETA-CROSSLAPS FOR EARLY MONITORING OF OSTEOPOROSIS TREATMENT WITH DENOSUMAB

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SUMMARY:

Antiresorptive long-term treatment for osteoporosis increases bone mineral density. Due to the long duration, poor compliance of patients to medication therapy is a big challenge. Bone mineral density measurements are recommended for the monitoring of treatment, but detectable changes may take up to 2-3 years. Bone turnover markers can indicate changes in bone turnover rates earlier, and assessment their levels may be effective in enhancing compliance and clinical efficacy.

Purpose: Our study aimed to assess whether the dynamics in serum levels of bone resorption marker beta-CrossLaps have a real practical benefit for early monitoring of postmenopausal osteoporosis treatment.

Materials and methods: 21 Bulgarian women in menopause with newly diagnosed osteoporosis were studied. All participants hadn't been under treatment. Serum beta-CrossLaps levels were measured before and six months after subcutaneous administration of Denosumab – Injection 60 mg.

Results: Serum concentrations before and six months after starting treatment were respectively 0.589 ± 0.266 ng/ml (0.06 – 1.2) and 0.166 ± 0.139 ng/ml (0.05 – 0.59). The beta-CrossLaps pre-treatment serum levels were within the reference range for the commercial kit used. After six months of treatment, there was a significant decrease in serum concentrations of about 72% from baseline. In our study, although pre-treatment levels were within the reference range, a significant decrease in concentrations was observed.

Conclusions: The results show that the dynamics of beta-CrossLaps may be useful in the early monitoring of osteoporosis treatment.

Keywords: postmenopausal osteoporosis, early monitoring treatment, beta-CrossLaps, bone resorption markers, Denosumab,

INTRODUCTION

The treatment of postmenopausal osteoporosis lasts for years. Currently available antiresorptive drugs increase bone mineral density (BMD), reduce new fractures and related morbidity. However, these effects occur slowly and the first few years of treatment is difficult to be measured objectively with the established methods. Measurement of BMD is not useful as a singular tool of treatment response because changes may be slow or minimal and recommended retesting interval for BMDscan at the spine and hip is every 1 to 3 years [1]. Long-term therapy is a big challenge, especially for asymptomatic patients and those who start with oral antiresorptive drugs. This may reduce patient's compliance and lead to premature discontinuation. Another problem with the initial treatment of osteoporosis are the nonresponders that must be identified early enough for a change in treatment strategy. Therefore early monitoring of the effect of treatment is very important for clinicians. Due to the limitations of BMD measurements, the development of useful tools in this direction is still ongoing.

Measurement of bone turnover markers (BTM) can offer a convenient way to monitor the treatment, and adherence of the patients, however, their potentially beneficial role is still being established. Measurement of BTMs in a blood or urine sample is easy and non-invasive. Recently efforts have been focused on standardizing their use in practice. Reference intervals studies and examination of appropriate decision thresholds, reference change values and treatment targets are still ongoing [2].

The International Osteoporosis Foundation and the International Federation of Clinical Chemistry and Laboratory Medicine recommend (serum C-terminal cross-linking telopeptide of type I collagen, s-CTX, beta-CrossLaps) to be used as a reference bone resorption marker and measured by standardized assays in observational and intervention studies in order to enlarge the international experience of its application to clinical medicine[3].

Reference values of beta-CrossLaps in healthy women and patients with osteoporosis in different countries are still being determined due to the lack of sufficient published data. The use of beta-CrossLaps in monitoring osteoporosis treatment is currently recommended in several guidelines all over the world. According to the recommendations, serum beta-CrossLaps, today is used to monitor adherence to oral bisphosphonate treatment [4].

Denosumab is a fully human monoclonal antibody to the receptor activator of the nuclear factor-kappaB ligand, which is a key mediator of the resorptive phase of bone remodeling [5]. Denosumab could be administered once in 6 months by subcutaneous injection of 60 mg each time [6]. Such a therapeutic regimen is easy to follow and significantly improves patient's adherence and it is most commonly used for osteoporosis treatment in recent years. There are no sufficient data about the use of beta-CrossLaps for early monitoring of Denosumab treatment.

In our country, there are no published data about the levels of beta-CrossLaps in healthy menopausal women and in patients with osteoporosis. Data about its use in early monitoring of osteoporosis treatment are also lacking.

Our study aims to assess whether the dynamics of the marker for bone resorption beta-CrossLaps has a real practical benefit in the early monitoring of the treatment of patients with postmenopausal osteoporosis. Our research will contribute to the enrichment of the available information.

MATERIALS AND METHODS

1. Participants:

We studied 21 menopausal women aged between 50-81 years with newly diagnosed osteoporosis. The diagnosis was made by measuring the BMD of the lumbar spine via Dual-energy X-ray absorptiometry (DXA) and interpreted following standards of the World Health Organization - T-score ≤ -2.5 SD. All participants hadn't been undertreatment. All participants have signed informed consent. Treatment was started with Denosumab – Injection 60 mg, subcutaneously once every 6 months. Venous blood samples were taken before treatment and after six months.

2. Blood samples collecting:

Adherence to accepted blood sampling protocols for beta-CrossLaps testing is crucial to avoid pre-analytical variations in serum concentrations. The variability of beta-CrossLaps concentrations was minimized by assessing the morning fasting serum. According to the recommendations in our study, blood samples were collected in the morning after overnight fasting at the time interval of 8.00-09.30 a.m. They were examined without being frozen. Quantitative immunological analysis of beta-CrossLaps was performed by electro-chemiluminescent immunoassay with Cobas E 411 immunoassay analyzer, Roche Diagnostics. Elecsys β -CrossLaps®, Roche Diagnostics kit with sensitivity 0.010 - 6.00 ng/ml and referent values for women in menopause are 0 - 1.008 ng/ml was used.

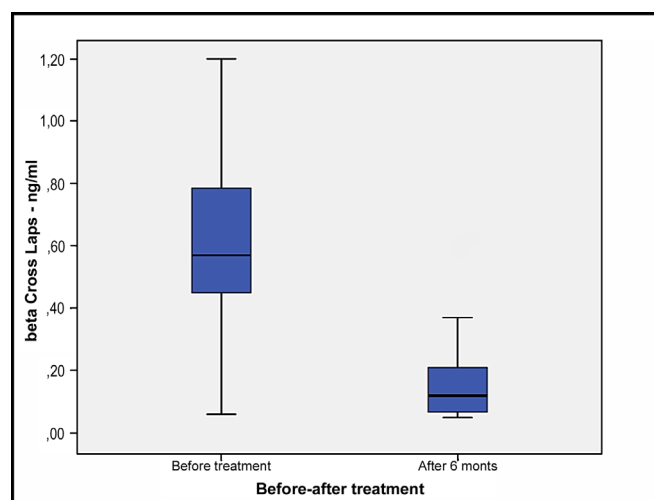
3. Statistical analysis

The statistical processing of our data was made by SPSS v23.0, Microsoft Office Excel 2013.

RESULTS

The results were expressed as mean \pm SD, minimum and maximum in brackets. The serum concentrations of the bone resorption marker beta-CrossLaps before treatment were 0.589 ± 0.266 (0.06-1.2) ng/ml. Six months after starting the treatment, serum concentrations were 0.166 ± 0.139 (0.05-0.59) ng/ml. (Fig.1)

Fig. 1. Serum concentrations of beta-CrossLaps in ng/ml before and after treatment.



The beta-CrossLaps serum levels of a pre-treatment study are within the reference range for the commercial kit. After six months of treatment, there was a significant decrease in serum concentrations of about 72% from baseline. In our study, although pre-treatment levels were within the reference range, a significant decrease in concentrations was observed.

DISCUSSION

The automated assays for serum beta-CrossLaps are precise and convenient to use. Manufacturers have developed their own reference values, but normal levels for different countries or even regions still need to be defined and compared [7]. Individual studies use country-specific reference ranges to analyze their data, with beta-CrossLaps reference values obtained using different cohorts in Europe, Asia, and North America. They all show similar values, suggesting that they may be universal, but more research is needed [8].

Most studies are focused on determining the reference values of beta-CrossLaps in healthy premenopausal women. There are currently insufficient data about the levels in menopausal women and patients with osteoporosis.

Serum levels of beta-CrossLaps show significant variation in various publications both in healthy menopausal women and in patients with osteoporosis. Garnero

et al. [7] and Boudou et al. [9] reported values of 0.556 ± 0.226 ng/ml and $0.13-0.60$ ng/ml, respectively in healthy French postmenopausal women.

In the Camargo cohort study, Martinez et al. [10] found values of 0.387 ± 0.197 ng/ml in Spanish postmenopausal women. Trento et al. [11], in 200 Italian postmenopausal women (54.6 ± 6.1 years) found values of 0.45 ± 0.10 ng/ml and 0.47 ± 0.12 ng/ml in women with normal and osteopenic BMD values respectively.

There are also different data about the serum levels of beta-CrossLaps in women with osteoporosis and osteopenia. Elevated serum and urine levels have been reported in women with osteopenia and osteoporosis compared to premenopausal or normal menopausal women [12, 13].

Publications from various studies show a weak or moderate inverse correlation between BMD and BTMs. In the TRIO study, only 20% of menopausal women diagnosed with osteoporosis by DXA had serum concentrations of beta-CrossLaps above the upper limit of normal for healthy menopausal women [14]. Another study conducted among Pakistani women showed no significant differences in serum concentrations in healthy, osteopenic and osteoporotic menopausal women [15].

BTMs cannot be used to diagnose osteoporosis because of low sensitivity and specificity [16]. Accumulating data on beta-CrossLaps levels in postmenopausal women with osteoporosis confirm the currently accepted position. In our study, the diagnosis was made by DXA, but the assessment of pre-treatment beta-CrossLaps levels showed values within the reference range for the commercial kit. The interpretation of the obtained results is further complicated by the lack of any published data about the levels of the marker in both healthy and osteoporotic patients in Bulgaria. Our results are similar to those published for healthy French menopausal women, but serum levels are higher than the reported ones for Italian and Spanish menopausal women without osteoporosis. This probably confirms the geographical variations and the need to set the reference values in accordance with the geographical regions.

Current scientific literature shows promise for BTMs to function as useful clinical tools in osteoporosis treatment management. BTMs quickly respond to changes in bone physiology. This can help physicians to ensure an adequate response, identify those patients needing to be trialed on another therapy in the event of suboptimal response, and confirm patients compliance to the therapeutic regimen [8]. Denosumab is one of the most potent inhibitors of bone resorption, which decreases rapidly serum levels of beta-CrossLaps. This marker has been reported to decrease within 24 hours after Denosumab administration [17].

In the FREEDOM study, the dynamics of several BTMs were measured at baseline and at 1, 6, 12, 24, and 36 months after subcutaneous administration of Denosumab (60 mg) or placebo injections. One month after in-

jection, CTX-1 levels in all denosumab-treated subjects decreased to levels below the premenopausal reference interval [18].

Aslan et al. [19] compared the changes in beta-CrossLaps levels and BMD in women with osteoporosis before and after 6 months of treatment. They report that there was no change in BMD, however, serum beta-CrossLaps levels decrease by 70% in all cases.

In Bulgaria, successful treatment is considered to be a decrease in beta-CrossLaps values of more than 56% [20]. Our results after six months of treatment with Denosumab showed a decrease in serum concentrations of beta-CrossLaps by more than 70% from baseline.

Today lack of sufficient data about beta-CrossLaps levels in patients with postmenopausal osteoporosis should not discourage clinicians because there is still not a consensus about universal reference ranges and this makes it difficult to determine bone turnover levels as low, normal or high. At the moment, the ability to perform serial beta-CrossLaps blood testing of one patient will be more indicative and useful. Measuring morning blood samples from fasting patients is easy and inexpensive. This should be the preferred way to assess the early therapeutic response. In our study pre-treatment levels were within the reference range, but a significant decrease in concentrations was observed in all patients. A disadvantage is the small number of patients studied. Also, it is impossible to compare obtained results due to the lack of published data about beta-CrossLaps levels in women with osteoporosis in Bulgaria. The results show that the dynamics of beta-CrossLaps may have a potential role in the early monitoring of the therapeutic effect of treatment. More data needs to be accumulated.

CONCLUSION

Recent data for early reduction of serum beta-CrossLaps levels after initiation of antiresorptive treatment allow us to assume that the marker can be useful in the management of osteoporosis much earlier than the measurement of the BMD. The decrease in beta-CrossLaps level after the start of treatment indicates the presence of pharmacological effects on bone cells, confirms the patient's compliance and suggests the success of the treatment. This is especially important in the first year when the visible change in BMD is indistinguishable by DXA scanning. The availability of a reliable tool to confirm the therapeutic effect may motivate patients to adhere to treatment. It is advisable to measure beta-CrossLaps before starting pharmacological treatment and again between the 3rd and 6th month of treatment. Our results support the data for the potential role of this marker for bone resorption in the early monitoring of osteoporosis treatment.

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