Case Study

ASSESSMENT OF BONE TURNOVER MARKERS PRIOR TO DENTAL IMPLANT PLACEMENT FOR OSTEOPOROSIS PATIENT- A CASE-CONTROL STUDY

Nirubama Karunakaran[1], Subhabrata Maiti[2], Selvaraj Jayaraman[3], Jessy Paulraj[4]*

[1]Saveetha Dental College and Hospitals, Saveetha Institute of Medical and Technical Sciences (SIMATS), Saveetha University, Chennai-600077, Tamil Nadu, India.
[2]Department of Prosthodontics and Implantology, Saveetha Dental College and Hospitals, Saveetha Institute of Medical and Technical Sciences (SIMATS), Saveetha University, Chennai-600077, Tamil Nadu, India. drsubhoprostho@gmail.com
[3]Department of Biochemistry, Saveetha Dental College and Hospitals, Saveetha Institute of Medical and Technical Sciences (SIMATS), Saveetha University, Chennai-600077, Tamil Nadu, India.
[4]Department of Pedodontics, Saveetha Dental College and Hospitals, Saveetha Institute of Medical and Technical Sciences (SIMATS), Saveetha University, Chennai-600077, Tamil Nadu, India.

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ABSTRACT

Understanding bone biology before the placement of an implant is a key factor in successful implant survival. The purpose of the study is to assess bone turnover markers as a diagnostic tool for the evaluation of bone quality in patients undergoing dental implant surgery prior to the treatment. The present study is a single-center, case–control, cross-sectional study done in a private institution, chennai. Patients with single edentulous space for the past 6 months were selected for the study. BTM (Bone Turnover Markers) levels from saliva samples collected concurrently with regularly scheduled blood tests before implant surgery were evaluated with Enzyme-linked immunosorbent assay ELISA. The values were compared in SPSS software using an independent t-test. Bone turnover markers such as BALP, Osteocalcin, CTX-1, and NTX-1 were increased in osteoporotic patients ($p<0.05$) which showed an increased chance of failure of the dental implant. Assessing bone turnover markers from saliva can serve as a successful tool in assessing the bone level for placement of dental implants and aids treatment planning. The difference in bone turnover markers serves as a promising biological tool in evaluating the jawbone condition in addition to the radiological examination for implant placement.

Key words: Innovation, Bone turnover marker, Dental implant, Osteoporosis patient.

Introduction

Bone turnover markers are responsible for bone resorption and growth which indicates the state of bone remodeling [1]. It is the main mechanism responsible for osteoporosis. Osteoporosis is a generalized skeletal disorder in which the condition of bone becomes brittle and fragile due to the loss of tissues and leads to an increased risk of bone fracture. According to WHO, osteoporosis refers to a reduction in bone formation by about 25% whereas osteopenia refers to a decrease in physiological bone mineral density by about 10-25%. Bone is a living tissue that is constantly being replenished and degenerating [2]. Previous studies investigating muscle biopsy samples from individuals suffering from osteoporosis showed shrinkage of muscle fibers type II, the level of fiber atrophy being proportional to the degree of bone mineral density loss [3].

When new bone cannot form, osteoporosis sets in. or compensate for the loss of old bone [4]. Osteoporosis can occur due to lack of calcium intake, malnutrition, postmenopausal estrogen deficiency, genetic disorder, and patients undergoing gastrointestinal surgery [5, 6]. People of all races can develop osteoporosis. White and Asian women, particularly older women who have experienced menopause, are at the highest risk. Patients with osteoporosis do not show any symptoms in the early stage, but once the bone begins to become brittle symptoms are observed. Back discomfort results from a shattered or collapsed vertebra. Eventually losing height, hunch over, and break bones far more readily than usual [7]. Weight-bearing activity, a balanced diet, and treatments can improve already fragile bones or stop bone loss [8, 9].

The disease is linked to several factors, and growing evidence suggests that it may also be linked to oral health issues like periodontal disease, decreased jaw bone density, and tooth loss [10]. Osteoporosis affects osseointegration which determines the success rate of implant placement. The rate of bone production and resorption can be determined using markers for bone turnover [11]. Recently, a variety of biomarkers have been employed to sensitively and precisely quantify bone growth and bone resorption [12]. The biomarkers for bone formation are alkaline phosphatase (ALP), procollagen type 1 N-terminal propeptide (P1NP), bone-specific alkaline phosphatase (BALP), osteocalcin (OC), and procollagen type 1 C-terminal propeptide (P1CP). Tarrate-resistant acid phosphatase 5b, hydroxyproline (HYP), hydroxylysine (HYL), deoxypyridinoline (DPD), and pyridinoline (PYD) are the biomarkers for bone resorption (TRAP 5b). Bone resorption and formation are regulated by several proteins,

such as RANKL, osteoprotegerin (OPG), dickkopf-1 (DDK-1), and sclerostin [13-15].

Saliva was proposed as a diagnostic tool for systemic diseases. Bone turnover markers were evaluated from patients' saliva in healthy and osteoporotic individuals in the study. These biomarkers help provide an early evaluation of bone turnover rate, which DXA can provide with accuracy. Due to the extensive knowledge and research skills of our staff, we have produced publications of the highest caliber [16-31]. The success rate of dental implants is determined by osseointegration; the primary goal of the study is to employ bone turnover indicators as a diagnostic tool for evaluating the quality of the bone in patients having dental implant surgery before the procedure.

Materials and Methods

Patient selection

In the prosthodontics department of a university hospital, a case-control study was conducted. G power software was used to estimate the sample size, and the sample included 80 patients (40 with known osteoporosis and 40 Nonsign of osteoporosis, the duration of one to two years) with a single edentulous space were included. They had never received osteoporosis treatment and did not exhibit any subjective or objective symptoms, such as back pain brought on by a fractured or collapsed vertebra, height deterioration over time, or stooped posture **(Figure 1)**. Multiple missing, long-term edentulous areas, long-span edentulism, and periodontally compromised patients were excluded. All of the chosen patients were told of the study and provided with voluntarily informed written permission.

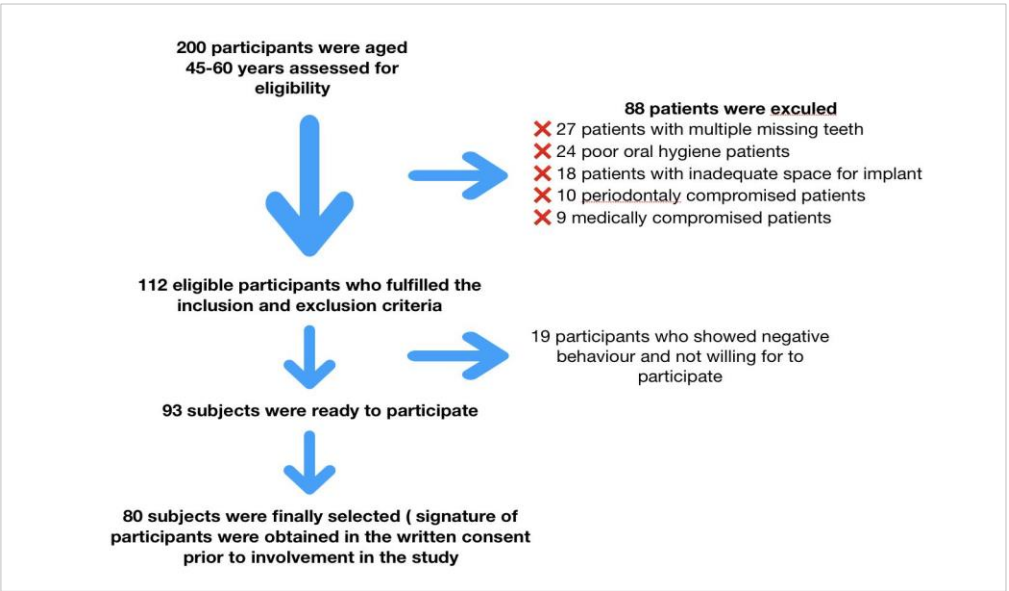

Figure 1. Patient Selection Criteria

Study design

Case-control study

Measurement of bone turnover markers

Enzyme-linked immunosorbent assay was used to detect BTM levels in saliva samples collected concurrently with regular blood tests before implant surgery (ELISA). A morning sample was taken from participants who had not exercised for 24 hours as advised and were fasting. Until analysis, saliva samples were kept at 70 °C. The bone-specific indicators of bone development were osteocalcin (OC) and osteopontin.

Human ALP ELISA kit was purchased from Abbkine company with catalog number KTE63711. The assay was done according to the user manual instructions. The kit uses a competitive inhibition principle enzyme immunoassay technique. The microtiter plate was precoated with antibodies. The standards and samples were run simultaneously. The secondary antibody conjugated with horseradish peroxidase was added and incubated. Then substrate specific to the enzyme was added and the color development was noted. ELISA plate reader at 450 nm was used to gauge the color's intensity. The same analysis was done on other biomarkers such as NTX, osteopontin, and osteocalcin.

Alkaline phosphatase (BALP); crosslinked type I collagen N-telopeptide (NTX). Each BTM marker's reference range was calibrated to fall within the accepted mean for healthy people (mean 1.96 SD) [17]. BAP, 3.7-20.9 ng/ml for males and 3.9-14.5 ng/ml in women, and NTX, 9.5-17.7 ng/ml for men and 7.5-16.5 ng/ml for women, are the typical ranges for BTM readings. At least one BTM result outside of the usual range was considered to be part of the abnormal group.

Statistical analysis

Statistical analysis was done using SPSS software

(Statistical Package for Social Science for Windows Versions, 20.0). The comparison in bone turnover markers of healthy and osteoporotic patients was analyzed. Data were entered into Microsoft Excel 2016 (Office 10) before being exported and statistically analyzed in SPSS (Statistical Package for Social Science for Windows Versions, 20.0), SPSS Inc. (Chicago IU, USA). The data were tabulated after being statistically analyzed using the Independent T-test ($p<0.05$).

Results and Discussion

All markers for healthy individuals were showing the normal value (within the limit) whereas osteoporosis individuals were showing a higher value of markers, the difference in markers value for healthy and osteoporotic individuals was statistically significant ($p<0.05$) **(Table 1)**.

Table 1. Comparison of bone turnover markers for normal and osteoporosis patients.

Markers	Group	Mean±Sd	Standard Error	95% Ci (Upper)	95% (Lower)	T Value	P Value
BALP	Healthy (n=40)	22.92±3.75	0.594	-11.77	-15.214	-15.598	0.00*
	Osteoporotic (n=40)	36.42±3.97	0.628	-11.77	-15.214	-15.598	0.00*
OSTEOCALCIN	Healthy (n=40)	7.43±3.61	3.618	-13.42	-15.957	-23.131	0.00*
	Osteoporotic (n=40)	22.12±1.74	1.744	-13.42	-15.964	-23.131	0.00*
OSTEOPONTIN	Healthy (n=40)	12.58±2.92	0.463	-7.4	-9.47	-16.2	0.00*
	Osteoporotic (n=40)	21.02±1.50	1.507	-7.39	-9.48	-16.2	0.00*
NTX-1	Healthy (n=40)	12.9±2.69	2.692	-8.07	-10.21	-17.043	0.00*
	Osteoporotic (n=40)	22.04±2.06	2.066	-8.07	-10.21	-17.043	0.00*

*significant at $p<0.05$; p-value was derived from an independent t-test.

The marker level rose from 22.923.75 to 36.423.97 when comparing the average value of bone alkaline phosphatase between normal and osteoporotic individuals. In people with normal liver function, blood levels of ALP are created from bone to a degree of around 50%. The mean readings for males and premenopausal women, respectively, are 24.97.0 U/L and 19.75.6 U/L, while the detection limit for BALP is 0.7 U/L. Similar to the current study, BALP is employed as a marker of osteoblastic activity in the treatment of osteoporosis in premenopausal and postmenopausal women [32]. Women older than 59 who had osteoporosis have been found to have greater BALP activity, which supports the study's findings [33]. Osteocalcin levels rose from 7.43 3.61 to 22.12 1.74 in those with osteoporosis. Osteocalcin, also known as bone gamma-carboxyglutamic acid-containing protein, is a tiny protein of 49 amino acids. Controlling metabolism, bone mineralization, and calcium ion balance all depend on the OC produced by osteoblasts [34]. It has been demonstrated that the growth in BMD following osteoporosis therapy with bone-forming medicines is highly associated with the amount of serum OC. Serum OC has been found as a particular biomarker of osteoblast activity for measuring the rate of bone resorption in osteoporosis. Numerous studies have demonstrated the importance of osteocalcin as a biomarker for assessing how well a medicine affects bone growth [35]. In the current investigation, the osteopontin level was determined to be 12.582.92 in healthy individuals and 21.021.50 in osteoporotic patients. Bone cells, T-lymphocytes that have been activated, specialized epithelial cells, macrophages, and altered cells all release OP, a phosphorylated glycoprotein [36]. According to a recent study, women with OP over-expression were less resistant to postmenopausal osteoporosis than women with normal OP levels [37]. In order to evaluate the effectiveness of intermittent parathyroid hormone treatment for menopausal osteoporosis, plasma OP concentrations may be employed as a biomarker. In a normal patient, the market value of NTX-1 was seen to be 12.92.69, but in an osteoporotic patient, it was observed to be 22.042.06. Type 1 collagen, which develops in bone from procollagen type 1, makes up more than 90% of the organic bone matrix.

When compared to the general healthy population, there is a discernible increase in the levels of bone turnover indicators in osteoporosis patients. Primary implant stability is necessary for successful peri-implant healing. In order to get positive outcomes and promote the growth of the tissues around the implant, a stable implant requires less micromotion between the bone and implant (e.g., angiogenesis and osteogenesis). The number and distribution of dental implants in the arch, periodontal health, occlusion, and biting forces are only a few examples of the local variables that might affect the success or failure of implant placement in addition to systemic ones. Few studies have actively investigated how alterations in mandibular bone metabolism and their links to systemic bone metabolic disease statuses are affected by the implantation of endosseous implants and the insertion of overdentures supported by implants. Studies investigating the link between skeletal and oral osteoporosis and loss of dental implants associated with low bone quality and quantity found no association between systemic BMD status, mandibular BMD status, bone quality, or implant

loss. Becker *et al*. have suggested that Through radiographic bone quality testing, which was more useful than peripheral bone density examination, only the visual assessment of bone quality can be optimal for inserting an implant [38]. In the current study, a more straightforward approach to evaluating BMP may be carried out utilizing salivary biomarkers, which are similarly effective as radiographic techniques. In addition, von Wowern and Gotfresden [39]. have done research on the osteoporotic edentulous jaw's marginal bone loss around dental implants. Endosseous implants were still regarded as an effective therapy for osteoporotic patients although people with a greater osteoporosis state had more marginal bone loss. Since the BTM represents bone turnover, which has been measured using ELISA from salivary indicators, we predicted that the BMD decreased as a result of the greater bone turnover.

Bone quality is complex and difficult to classify since it varies from patient to patient. The primary focus of the study was on bone turnover markers, one of the clinical indications of bone health. Elevation of a BTM was not shown to be a reliable indicator of fracture in a prospective investigation [40]. To diagnose the patient's jaw bone quality, it may be helpful to combine radiographic and biological methods, such as examining bone turnover indicators. The cancellous bone density of patients with high BTM values was considerably lower than that of patients with normal BTM values, demonstrating that the quality of the cancellous bone in the abnormal group was worse at baseline than that in the normal group. BTMs and cancellous bone density must both be assessed when assessing bone strength in female patients who will undergo implant procedures.

To yet, only radiographic evaluations have been utilized to determine the quality of jaw bones. The bone structure has not yet been accurately evaluated. The use of salivary bone turnover indicators for evaluating bone quality can meet this need. To properly treat patients and prevent osteoporosis, especially in women, this aids in forecasting the prevalence of the ailment in its early stages.

Conclusion

Measurement of bone turnover markers can be a valuable clinical biomarker prior to implant placement. Further animal trials and clinical trials have to proceed. The difference in bone turnover markers serves as a promising biological tool in evaluating the jawbone condition in addition to the radiological examination for implant placement.

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