Published in final edited form as:

Endocr Res. 2018 February ; 43(1): 21–28. doi:10.1080/07435800.2017.1369432.

Effect of parathyroidectomy on osteopontin and undercarboxylated osteocalcin in patients with primary hyperparathyroidism

Raelene E. Maser^{a,b}, M. James Lenhard^{b,c}, Ryan T. Pohlig^d, P. Babu Balagopal^e, and Raafat Abdel-Misih^f

^aDepartment of Medical Laboratory Sciences, University of Delaware, Newark, DE, USA

^bDiabetes and Metabolic Research Center, Christiana Care Health System, Newark, DE, USA

^cDiabetes and Metabolic Diseases Center, Christiana Care Health System, Wilmington, DE, USA

^dBiostatistics Core Facility, University of Delaware, Newark, DE, USA

^eBiomedical Research & Analysis Laboratory, Nemours Children's Specialty Care & Mayo Clinic College of Medicine, Jacksonville, FL, USA

^fDepartment of Surgery, Christiana Care Health System, Newark, DE, USA

Abstract

Purpose: Surgical treatment for primary hyperparathyroidism (PHPT) improves bone metabolism. Osteocalcin (OC) and its undercarboxylated form (ucOC) are associated with bone and energy metabolism. Osteopontin (OPN), a multifunctional protein expressed in bone, is involved in resorption, along with β -carboxyl-terminal cross-linking telopeptide of type 1 collagen (β -CTX), and osteoprotegerin (OPG). Our aim was to investigate these biomarkers of bone metabolism in patients with PHPT.

Methods: We examined 30 individuals with PHPT, in a clinical research facility, before and one month following parathyroidectomy. Circulating levels of OC, ucOC, OPN, β -CTX, and OPG were examined as bone biomarkers along with inflammatory markers (e.g., interleukin-6 [IL-6], lipocalin-2), insulin resistance (i.e., homeostasis model assessment for insulin resistance [HOMA-IR]), adiposity (i.e., leptin, adiponectin), PTH, calcium, 25-hydroxyvitamin D, creatinine, and demographics.

Results: Participants (27 females/3 males) were 60 ± 9 (mean $\pm$ SD) years old. There was a significant reduction of ucOC (7.9 ± 5.1 [median $\pm$ SIQR] vs. 6.6 ± 3.7 ng/mL, $p=0.022$) and OPN (75.4 ± 14.5 vs. 54.5 ± 9.2 ng/mL, $p<0.001$) pre- vs. post-parathyroidectomy. There were no univariate differences postoperatively for IL-6, HOMA-IR, leptin or adiponectin. Regression analysis showed that postoperative levels of adiponectin, IL-6, and OPN were significantly associated with ucOC, while adjusting for PTH and albumin corrected calcium levels (model

Corresponding author: Raelene E. Maser, Ph.D., Department of Medical Laboratory Sciences, 305F Willard Hall Education Building, University of Delaware, Newark, DE 19716, USA, rmaser@udel.edu.

Declaration of interest

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

$R^2=0.610$, $p=0.001$). With OPN as the dependent variable, higher adiponectin and lower ucOC were significantly associated with lower OPN levels postoperatively (model $R^2=0.505$, $p=0.010$).

Conclusion: The lower one-month postoperative OPN and ucOC levels in PHPT indicate reduced bone resorption. Decreased ucOC levels may also suggest lower energy demands postoperatively.

Keywords

Primary hyperparathyroidism; Parathyroidectomy; Undercarboxylated osteocalcin; Osteopontin

Introduction

Primary hyperparathyroidism (PHPT), a common endocrine disorder, is characterized by alterations in serum calcium and PTH (1). Increased secretion of PTH leads to enhanced bone resorption and formation resulting in an increased risk of fracture (2). Surgical removal of the adenoma or multiple hypercellular glands is the only curative treatment (1).

Osteocalcin (OC), one of the most abundant noncollagen proteins in bone, is a specific product of osteoblasts (3). In the circulation, OC levels are considered a measure of bone formation (3). During bone resorption, OC is released from the bone matrix suggesting that OC is a putative marker of bone turnover as well (4). OC contains 3 residues of gamma-carboxyglutamic acid. Post-translational modification results in the majority of OC being completely carboxylated and deposited into the bone matrix or released into the circulation (5). Some OC, however, is only partially carboxylated (i.e., undercarboxylated OC [ucOC]). ucOC avidity for hydroxyapatite is lower, providing for its leakage into the circulation (5). Factors that regulate the addition of carboxyl moieties to OC in humans are poorly understood. Results from animal studies have suggested that ucOC may act as a regulator of energy metabolism affecting insulin secretion as well as insulin sensitivity in peripheral tissues (6, 7). Although various *in vitro* and *in vivo* studies suggest that ucOC is the form of OC responsible for endocrine functions (8), not all studies of ucOC in humans (9) have confirmed this notion. A recent meta-analysis (10) suggested that both OC and ucOC could serve as biomarkers associated with glucose metabolism.

Osteopontin (OPN), osteoprotegerin (OPG), and β -carboxyl-terminal cross-linking telopeptide of type 1 collagen (β -CTX) are additional bone metabolism biomarkers involved in bone resorption (11–13). Like OC, OPN is an abundant noncollagenous bone matrix protein, expressed in osteoblasts and osteoclasts (14). OPN's expression is strongly influenced by PTH (14). Furthermore, the expression of OPN is upregulated by proinflammatory cytokines and OPN is involved in adipose tissue inflammation and insulin resistance (15, 16). OPG has been shown to decrease osteoclast numbers but has no direct influence on osteoblasts (12). PTH inhibits the expression of OPG messenger RNA in cultured murine bone marrow and osteoblasts (17). β -CTX is a marker of osteoclast activity (18). Serum β -CTX and PTH have also been shown to be positively correlated in PHPT (18).

In the current study, we investigated various biomarkers of bone resorption and formation such as OC, ucOC, OPN, OPG, and β -CTX in patients with PHPT before and one month

after parathyroidectomy. We also examined additional mediators of glucose homeostasis plus markers of inflammation such as interleukin-6 (IL-6), lipocalin-2, leptin, adiponectin, and homeostasis model assessment for insulin resistance (HOMA-IR). We hypothesized that elevated PTH levels and increased energy needs required for bone resorption and formation occurring in PHPT would be associated with incomplete carboxylation of OC (i.e., increased levels of ucOC) and that ucOC levels would be reduced following successful parathyroidectomy.

Methods

Subjects

Thirty-two participants, who volunteered to participate in this study, were evaluated at the Diabetes and Metabolic Research Center, Christiana Care Health System, Newark, DE, USA. This study had approval of the Institutional Review Board of Christiana Care Corporation and each person gave written informed consent before taking part in the study. Participants were eligible for the study if they were ≥18 years old with PHPT. The diagnosis of PHPT was established according to laboratory data characterized by the persistence of high concentrations of PTH and total calcium. All participants underwent preoperative localization by Tc-99m sestamibi imaging. Histopathological postoperative reports confirmed a single parathyroid adenoma for all participants, with the exception of two individuals that had a double parathyroid adenoma. Exclusion criteria for the study included: (a) history of known cardiovascular disease or acute myocardial ischemia; (b) dose changes 2 months prior to enrollment for chronic prescription medications (e.g., antihypertensive); and (c) chronic kidney disease stage 3b. It should be noted that two individuals that were enrolled in the study had to be excluded. One individual decided not to undergo surgery and the other person had four parathyroid gland hyperplasia. Thus after exclusion of these individuals, the results for 30 individuals were utilized. Participants were re-examined approximately 1 month (median=32 days) after parathyroidectomy.

Clinical measurements

Weight and height were measured using a stadiometer. Body mass index (BMI) was calculated as body weight divided by height squared (kg/m^2). Blood pressure was monitored electronically in the supine posture using an oscillometric automatic recorder. Twenty-four participants performed a 24-hour urine collection for calcium. Results from an earlier 24-hour collection, performed prior to enrollment in this study, were utilized for 15 participants. The median time of collection before the baseline visit was approximately 2.8 months. No participants had hypocalciuria. Specimens were not obtained for 6 participants due to refusal, insufficient time before surgery, or incomplete urine collection. The mean calcium level was 5.0 ± 2.1 (mean $\pm$ SD) mg/kg/24 hours.

Blood analytes

Blood samples were drawn after an overnight fast, with the exception of one participant who had fasted for 5 hours in the morning. Samples at baseline and follow-up were drawn between 8:45 AM and 12:15 PM. Levels of PTH (reference range 15–65 ng/L), 25-hydroxyvitamin D (reference range >72 nmol/L), and insulin (reference range 2.6–24.9

μIU/mL) were determined by an electrochemiluminescence immunoassay while glucose (reference range <5.55 mmol/L) measurements were performed using an enzymatic method. Insulin resistance was calculated using the HOMA-IR online calculator downloaded from <http://www.dtu.ox.ac.uk> (19). Fructosamine levels (reference range 200–285 μmol/L) were determined by a colorimetric rate reaction using a Roche Cobas c501 chemistry analyzer (Roche Diagnostics Corporation, Indianapolis, IN, USA). Serum creatinine levels (reference range 44–115 μmol/L) were measured by an enzymatic colorimetric assay. Calcium levels (reference range 2.1–2.575 mmol/L) were determined using the Vitros Ca slide method on the Vitros 5,1 FS chemistry system (Ortho-Clinical Diagnostics, Rochester, NY, USA). Albumin levels (reference range 38–51 g/L) were determined using a bromocresol green colorimetric method on the AU5800 clinical chemistry analyzer (Beckman Coulter, Brea, CA, USA). Calcium levels were corrected for albumin levels using the equation: corrected calcium (mmol/L) = total calcium (mmol/L) + 0.02 × (40 - albumin (g/L)). Leptin and adiponectin levels were measured by radioimmunoassay (Millipore Corporation, Billerica, MA, USA). Specific enzyme-linked immunosorbent assays (ELISA) were used for the measurement of OC, OPG (ALPCO, Salem, NH, USA), ucOC (Takara Bio, Inc., Otsu, Shiga, Japan), OPN, IL-6, lipocalin-2 (R&D Systems, Minneapolis, MN, USA), and β-CTX (MyBioSource, San Diego, CA, USA). Samples for OC, ucOC, OPG, OPN, β-CTX, IL-6, lipocalin-2, leptin, and adiponectin were analyzed at the Nemours Biomedical Research & Analysis Laboratory, Jacksonville, FL, USA. It should be noted that OPN and β-CTX levels were missing for one participant due to insufficient sample.

Statistical analyses

Summary statistics are presented as mean±SD for those variables that are normally distributed and median±semi-interquartile ranges are reported for those variables that are non-normally distributed. Comparisons of demographic and metabolic parameters before and after parathyroidectomy were made with the paired t-test or Wilcoxon signed-rank test, where appropriate. Linear regression analyses, where the dependent variables were bone biomarkers (i.e., ucOC, OPN) that had significantly changed postsurgery, were performed to assess for independent associations of postsurgical metabolic parameters, while adjusting for PTH and albumin corrected calcium levels. Normality was tested and if violated, a nonparametric test was used.

Results

Table 1 provides participants' physical characteristics and metabolic parameters before and one month following parathyroidectomy. As expected PTH and calcium levels were significantly lower following surgery. In addition OPN, ucOC, and the ucOC/OC ratio, were also significantly reduced postsurgery. OC, β-CTX, and OPG levels, however, did not change significantly whereas lipocalin-2 concentrations were higher. Of note, two participants had a previous history of resolved malignant disease that did not require current treatment. Three of the females in this study were pre-menopausal. With the exception of one female that used Premarin vaginally twice per week, none of the participants indicated that they were currently taking bisphosphonates, estrogen, or raloxifene. Four participants had a history of type 2 diabetes mellitus (T2DM) that was treated with metformin (n=4) and

a sulfonylurea (n=2) but none were using a thiazolidinedione. One participant had a previous history of gastric bypass surgery. Because of the characteristics of the study cohort, data analysis was repeated excluding (a) premenopausal women (n=3), (b) individuals with T2DM (n=4), and (c) the individual with bariatric surgery. The univariate statistically significant results for OPN, ucOC, and lipocalin-2 pre- vs. post-surgery remained after reanalyses, despite the exclusion of these individuals (data not shown).

Linear regression models with postoperative levels for ucOC and OPN as the dependent variables were used to test for independent postsurgical associations of metabolic parameters while adjusting for PTH and albumin corrected calcium levels (Table 2). We examined ucOC and OPN as the dependent variables of interest because these were the only two bone biomarkers that were significantly reduced following parathyroidectomy. The metabolic parameters included in the models as potential independent variables were HOMA-IR, adiponectin, and IL-6. With ucOC as the dependent variable, lower adiponectin, IL-6, and OPN levels were significantly associated with lower ucOC levels, $F(6,22)=5.73$, $p=0.001$, model $R^2=0.610$, and $\text{adjusted } R^2=0.503$ (Table 2). With OPN as the dependent variable, higher adiponectin and lower ucOC were significantly associated with lower OPN levels, $F(6,22)=3.74$, $p=0.010$, model $R^2=0.505$, and $\text{adjusted } R^2=0.370$. The data were reanalyzed excluding (a) premenopausal women, (b) individuals with T2DM, and (c) one individual with a past history of gastric bypass surgery. The significant association between OPN and ucOC in these models persisted, despite the exclusion of these individuals (data not shown).

Discussion

In this study, we investigated various biomarkers of bone metabolism such as OC, ucOC, OPN, β -CTX, and OPG along with markers of insulin resistance and inflammation in patients with PHPT before and one month postsurgery. The data demonstrated significant reduction in the circulating levels of ucOC and OPN but no significant changes in the levels of OC, β -CTX, or OPG one month following parathyroidectomy.

The coupling of bone formation and resorption maintains bone mass in a steady state under normal conditions (3). In humans, OC in bone and serum may be incompletely carboxylated (i.e., ucOC) (3). We speculated that elevated PTH levels, as a result of PHPT, may potentially affect the carboxylation of OC resulting in increased circulating levels of ucOC. Experiments from animal models also suggest that bone resorption affects circulating levels of ucOC (20, 21). The reported mechanism is acidification of the extracellular compartment which decarboxylates OC into ucOC with release of ucOC into the circulation (20, 21). Given the potential role of ucOC in energy metabolism as shown in animal studies (7), it is possible that ucOC directs energy to bones and muscles during increased bone turnover occurring in PHPT. Following parathyroidectomy with reduced bone resorption, ucOC levels decrease presumably as a result of reduced osteoclast vacuolar proton pump acidification of the bone matrix and perhaps due to the reduction in PTH levels. Further, there may also be reduced energy demand.

OPN, a secreted glycoprotein, is a prominent bone matrix protein that promotes the adhesion of osteoclasts to bone matrix regulating the synthesis and resorption of bone (22). OPN is

also an inflammatory cytokine expressed in adipose tissue (23). Recent data from animal models, however, showed that OPN did not induce inflammatory macrophages but in the presence of OPN, macrophages have enhanced endocytic activity (23). We believe that the postsurgical reduction in levels of OPN observed in the current study was due to a slowing down of bone resorption rather than an indicator of changes in inflammation, as evidenced by a lack of significant reduction of IL-6 (Table 1). It is also possible that the reduced OPN levels were a direct result of the reduction in PTH levels as PTH regulates the expression of OPN (14). Results of linear regression modeling appear to indicate that ucOC and OPN may track together in patients with PHPT following parathyroidectomy. That is, both OPN and ucOC could be indicating the reduction in bone resorption following surgery while it is also possible that reduced ucOC is an indication of decreased energy aid being required.

OPG, another regulator of bone resorption, suppresses osteoclast numbers and activation (12). Previous in vitro studies have shown that PTH decreased the expression of OPG messenger RNA (17). In our study at one month postsurgery, we did not, however, observe a significant change in serum OPG levels. Likewise, other studies failed to show a change in serum OPG levels 12 months after parathyroidectomy (2, 24).

β -CTX levels were not significantly changed one-month post-surgery in the current study. This is in contrast to other studies where a significant decrease in β -CTX was demonstrated at 3 months (24) and approximately 12 months (2) following parathyroidectomy. Our results also differ from a study where 24-hour urinary N-terminal telopeptide of type I collagen levels were reduced one-month post-parathyroidectomy (25). The exact reason for this discrepancy is not obvious from the current study. However, differences in patient cohorts, sample size, varying durations of follow-up after surgery, and the type of assays and substrates used (e.g., blood vs. urine) may contribute to the conflicting results observed in our study. It is also possible the β -CTX is not as sensitive a marker as ucOC and OPN at one-month post-parathyroidectomy. Obviously, these possibilities need to be clarified in future studies.

Patients with PHPT manifest signs of subclinical inflammation (26). Animal models have shown that IL-6 is involved in osteoclast formation and may therefore contribute to increased bone resorption (27). Thus it is possible that IL-6 may be involved in bone loss of patients with PHPT (28). In the current study linear regression modeling with ucOC as the dependent variable showed that higher IL-6 levels were associated with higher ucOC levels postoperatively, when all participants were included in the model. Thus, the higher IL-6 levels may be a response to tissue damage, which potentially might have occurred as a result of surgery (26). IL-6 may also be tracking with ucOC in that higher energy needs are still required in those individuals with greater degree of inflammation.

We also examined lipocalin-2, another modulator of inflammation acting on adipocytes and macrophages (15, 29). In vitro studies indicate that lipocalin-2 plays an important role in bone metabolism, indicating a potential inhibitory role in osteoclast formation (30). Lipocalin-2 levels, in the current study, were increased postsurgery and therefore may be another indicator of reduced bone resorption. It should be noted, however, that when

lipocalin-2 was included as a potential independent variable in the regression models, it was not independently associated with either ucOC or OPN (data not shown).

Reports with regard to the effect of parathyroidectomy on insulin resistance have been conflicting. Most studies, like ours, did not demonstrate improvement in insulin resistance after surgery when surrogate indices such as HOMA-IR were used (31). Similar to other studies adipokines (32), such as leptin and adiponectin that can affect insulin sensitivity, were not significantly different after surgery in our study. Nonetheless, higher post-operative adiponectin was associated with lower OPN levels in regression analysis. It is well known that adiponectin exerts anti-inflammatory effects which may be what is represented here. Adiponectin also enhances insulin sensitivity (33) and the positive association shown in the regression model with ucOC as the dependent variable indicates that both adiponectin and ucOC appear to be functioning in parallel. Thus, the pathways involved in directing energy and substrate to the energy intensive process of bone remodeling following parathyroidectomy appear to be multi-factorial.

Our study has several strengths including simultaneous assessment of OC, ucOC, OPN, β -CTX, and OPG in the same cohort of patients with PHPT along with the measurement of inflammatory markers, insulin resistance, and adipokines. The magnitude of the explained variability was high for the regression models (e.g., $R^2 = 0.610$ for ucOC model). This study also has some potential limitations that deserve mention. The post-operative follow-up period was intentionally short in order to examine changes in ucOC. Additional information at 3, 6, and 12 months would have been interesting. There was no matched control group who did not undergo parathyroidectomy. Medications that participants were taking could have affected results. A few individuals had medication changes during follow-up, such as the use of pain medication, antibiotics, and a dosage change in antihypertensive medication. We were unable to control for changes in calcium and vitamin supplements. The majority of the participants were females and therefore it is uncertain whether the results are applicable to men as well. Studies of bone mineral density and a comprehensive study of other biomarkers of bone biology were not available in this study, thus we were not able to examine for other potential associations with OPN and ucOC. We did not measure serum vitamin K levels, although this might affect carboxylation of OC and serum ucOC levels. Conversely, vitamin K deficiency in adults is uncommon except for those with gastrointestinal or liver disease. It should be noted that individuals in this study did not have clinically significant abnormal liver function tests ($n=30$) or a known history of chronic malabsorptive bowel disorders ($n=29$), with the exception of one individual who had bariatric surgery.

In summary, bone resorption and formation is an energy intensive process that involves turnover of various bone parameters and facilitates cross talk between organs. The current study adds valuable information to the literature in this area and to the best of our knowledge the roles of OPN and ucOC following parathyroidectomy in individuals with PHPT are novel. The lower one- month postoperative OPN and ucOC levels indicate reduced bone resorption, while decreased ucOC may also suggest lower energy demands postoperatively. Further studies that address the limitations of the current study are needed to confirm these relationships.

Acknowledgments

The authors thank members of the Department of Nuclear Medicine, Christiana Care Health System, Newark, DE, USA, for their help in recruiting participants for this study.

Funding: This work was supported by the Clinical Research Committee, Department of Medicine, Christiana Care Health System, Newark, DE, USA. The statistical analysis only was partially supported by an Institutional Development Award (IDeA) from the National Institute of General Medical Sciences of the National Institutes of Health under grant number U54-GM104941.

References

1. Shindo M , Lee JA , Lubitz CC , et al. The changing landscape of primary, secondary, and tertiary hyperparathyroidism: highlights from the American College of Surgeons Panel, “What’s new for the surgeon caring for patients with hyperparathyroidism”. *J Am Coll Surg.* 2016;222:1240–1250.27107975
2. Stilgren LS , Hegedus LM , Beck-Nielsen H , et al. Osteoprotegerin levels in primary hyperparathyroidism: effect of parathyroidectomy and association with bone metabolism. *Calcif Tissue Int.* 2003;73:210–216.14667132
3. Booth SL , Centi A , Smith SR , et al. The role of osteocalcin in human glucose metabolism: marker or mediator? *Nat Rev Endocrinol.* 2013;9:43–55.23147574
4. Ivaska KK , Hentunen TA , Vaaraniemi J , et al. Release of intact and fragmented osteocalcin molecules from bone matrix during bone resorption in vitro. *J Biol Chem.* 2004;279:18361–18369.14970229
5. Li J , Zhang H , Yang C , et al. An overview of osteocalcin progress. *J Bone Miner Metab.* 2016;34:367–379.26747614
6. Kim YS , Paik IY , Rhie YJ , et al. Integrative physiology: defined novel metabolic roles of osteocalcin. *J Korean Med Sci.* 2010;25:985–991.20592887
7. Lee NK , Sowa H , Hinoi E , et al. Endocrine regulation of energy metabolism by the skeleton. *Cell.* 2007;130:456–469.17693256
8. Lacombe J , Ferron M . Gamma-carboxylation regulates osteocalcin function. *Oncotarget.* 2015;6:19924–19925.26343369
9. Wang Q , Zhang B , Xu Y , et al. The relationship between serum osteocalcin concentration and glucose metabolism in patients with type 2 diabetes mellitus. *Int J Endocrinol.* 2013;2013:842598.23533407
10. Liu DM , Guo XZ , Tong HJ , et al. Association between osteocalcin and glucose metabolism: a meta-analysis. *Osteoporos Int.* 2015;26:2823–2833.26089135
11. Subraman V , Thiagarajan M , Malathi N , et al. OPN -revisited. *J Clin Diagn Res.* 2015;9:ZE10–ZE13.
12. Kostenuik PJ . Osteoprotegerin and RANKL regulate bone resorption, density, geometry and strength. *Curr Opin Pharmacol.* 2005;5:618–625.16188502
13. Rosenquist C , Fledelius C , Christgau S , et al. Serum CrossLaps one step ELISA. First application of monoclonal antibodies for measurement in serum of bone-related degradation products from C-terminal telopeptides of type I collagen. *Clin Chem.* 1998;44:2281–2289.9799755
14. Kitahara K , Ishijima M , Rittling SR , et al. Osteopontin deficiency induces parathyroid hormone enhancement of cortical bone formation. *Endocrinology.* 2003;144:2132–2140.12697722
15. Catalan V , Gomez-Ambrosi J , Rodriguez A , et al. Peripheral mononuclear blood cells contribute to the obesity-associated inflammatory state independently of glycemic status: involvement of the novel proinflammatory adipokines chemerin, chitinase-3-like protein 1, lipocalin-2 and osteopontin. *Genes Nutr.* 2015;10:11.
16. Lancha A , Rodriguez A , Catalan V , et al. Osteopontin deletion prevents the development of obesity and hepatic steatosis via impaired adipose tissue matrix remodeling and reduced inflammation and fibrosis in adipose tissue and liver in mice. *PLoS One.* 2014;9:e98398.24871103

17. Lee SK , Lorenzo JA . Parathyroid hormone stimulates TRANCE and inhibits osteoprotegerin messenger ribonucleic acid expression in murine bone marrow cultures: correlation with osteoclast-like cell formation. *Endocrinology*. 1999;140:3552–3561.10433211
18. Verheyen N , Fahrleitner-Pammer A , Belyavskiy E , et al. Relationship between bone turnover and left ventricular function in primary hyperparathyroidism: The EPATH trial. *PLoS One*. 2017;12:e0173799.28406904
19. Levy JC , Matthews DR , Hermans MP . Correct homeostasis model assessment (HOMA) evaluation uses the computer program. *Diabetes Care*. 1998;21:2191–2192.9839117
20. Confavreux CB . Bone: from a reservoir of minerals to a regulator of energy metabolism. *Kidney Int Suppl*. 2011;79:S14–S19.
21. Ferron M , Wei J , Yoshizawa T , et al. Insulin signaling in osteoblasts integrates bone remodeling and energy metabolism. *Cell*. 2010;142:296–308.20655470
22. Iwadata H , Kobayashi H , Kanno T , et al. Plasma osteopontin is correlated with bone resorption markers in rheumatoid arthritis patients. *Int J Rheum Dis*. 2014;17:50–56.24472267
23. Schuch K , Wanko B , Ambroz K , et al. Osteopontin affects macrophage polarization promoting endocytic but not inflammatory properties. *Obesity*. 2016;24:1489–1498.27221527
24. Kersch-Schindl K , Riss P , Krestan C , et al. Bone metabolism in patients with primary hyperparathyroidism before and after surgery. *Horm Metab Res*. 2012;44:476–481.22495973
25. Christiansen P , Steiniche T , Brixen K , et al. Primary hyperparathyroidism: short-term changes in bone remodeling and bone mineral density following parathyroidectomy. *Bone*. 1999;25:237–244.10456391
26. Almqvist EG , Bondeson AG , Bondeson L , et al. Increased markers of inflammation and endothelial dysfunction in patients with mild primary hyperparathyroidism. *Scand J Clin Lab Invest*. 2011;71:139–144.21166606
27. Tamura T , Udagawa N , Takahashi N , et al. Soluble interleukin-6 receptor triggers osteoclast formation by interleukin 6. *Proc Natl Acad Sci*. 1993;90:11924–11928.8265649
28. Safley SA , Villinger F , Jackson EH , et al. Interleukin-6 production and secretion by human parathyroids. *Clin Exp Immunol*. 2004;136:145–156.15030526
29. Abella V , Scotece M , Conde J , et al. The potential of lipocalin-2/NGAL as biomarker for inflammatory and metabolic diseases. *Biomarkers*. 2015;20:565–571.26671823
30. Kim HJ , Yoon HJ , Yoon KA , et al. Lipocalin-2 inhibits osteoclast formation by suppressing the proliferation and differentiation of osteoclast lineage cells. *Exp Cell Res*. 2015;334:301–309.25814363
31. Rudman A , Pearson ER , Smith D , et al. Insulin resistance before and after parathyroidectomy in patients with primary hyperparathyroidism--a pilot study. *Endocr Res*. 2010;35:85–93.20408756
32. Bhadada SK , Bhansali A , Shah VN , et al. Changes in serum leptin and adiponectin concentrations and insulin resistance after curative parathyroidectomy in moderate to severe primary hyperparathyroidism. *Singapore Med J*. 2011;52:890–893.22159932
33. Awazawa M , Ueki K , Inabe K , et al. Adiponectin enhances insulin sensitivity by increasing hepatic IRS-2 expression via a macrophage-derived IL-6-dependent pathway. *Cell Metab*. 2011;13:401–412.21459325

Table 1.

Participant demographics and metabolic parameters.

	Pre-operative	Post-operative	P Value
Age (years)	60 ± 9	-----	
Male/female (n)		-----	
Body mass index (kg/m ²)	28.8 ± 5.4	29.0 ± 5.2	0.087
Systolic blood pressure (mmHg)	121 ± 14	120 ± 14	0.794
Diastolic blood pressure (mmHg)	74 ± 8	75 ± 7	0.714
Parathyroid hormone (ng/L)	116 ± 43	63 ± 19	<0.001
Calcium (mmol/L)	2.75 ± 0.09	2.39 ± 0.11	<0.001
Albumin corrected calcium (mmol/L)	2.70 ± 0.08	2.30 ± 0.08	<0.001
Serum creatinine (μmol/L)	69.8 ± 7.9	70.7 ± 8.8	0.465
Fructosamine (mcmol/L)	229 ± 15	230 ± 14	0.957
HOMA-IR	1.4 ± 0.9	1.5 ± 0.6	0.888
Leptin (μg/L)	35.0 ± 23.3	36.8 ± 26.5	0.372
Adiponectin (mg/L)	6.6 ± 2.4	6.8 ± 2.3	0.217
25-hydroxyvitamin D (nmol/L)	59.9 ± 21.2	64.9 ± 22.5	0.818
Interleukin-6 (pg/mL)	1.9 ± 1.3	2.2 ± 1.2	0.166
Lipocalin-2 (ng/mL)	68.1 ± 19.9	75.5 ± 20.2	0.007
β-CTX (pg/mL) ^a	311.9 ± 84.9	309.5 ± 82.5	0.813
Osteopontin (ng/mL) ^a	75.4 ± 14.5	54.5 ± 9.2	<0.001
Osteoprotegerin (pmol/L)	6.1 ± 2.1	5.9 ± 2.2	0.250
Osteocalcin (ng/mL)	20.1 ± 6.4	18.6 ± 6.2	0.280
Undercarboxylated osteocalcin (ng/mL)	7.9 ± 5.1	6.6 ± 3.7	0.022
ucOC/OC ratio	0.45 ± 0.13	0.39 ± 0.10	0.007

HOMA-IR, homeostasis model assessment for insulin resistance

β-CTX, β-carboxyl-terminal cross-linking telopeptide of type 1 collagen

ucOC/OC ratio, undercarboxylated osteocalcin/osteocalcin ratio

Data are presented as mean ± SD for variables that are normally distributed and *median ± semi-interquartile range for variables non-normally distributed*. P-values are from the paired t-test or Wilcoxon signed-rank test, where appropriate.

^aOsteopontin and β-CTX levels were missing for one participant due to insufficient sample.

Table 2.

Linear regression models for biomarkers of bone metabolism following surgery

Dependent Variable	Independent Variables	Regression Coefficient	Standard Error	p-value
ucOC	Parathyroid hormone	0.057	0.016	0.002
	Albumin corrected calcium	5.266	6.895	0.453
	HOMA-IR	-0.232	0.845	0.787
	Adiponectin	0.699	0.255	0.012
	Interleukin-6	1.727	0.753	0.032
	OPN	0.152	0.049	0.005
OPN	Parathyroid hormone	-0.135	0.068	0.060
	Albumin corrected calcium	-25.053	24.93	0.326
	HOMA-IR	-4.582	2.933	0.133
	Adiponectin	-3.066	0.859	0.002
	Interleukin-6	-2.849	3.000	0.353
	ucOC	2.029	0.647	0.005

ucOC, *undercarboxylated osteocalcin*; HOMA-IR, *homeostasis model assessment for insulin resistance*; OPN, *osteopontin*

Linear regression was performed for all those with complete data (n=29).