

Research Article

Parathyroid hormone is associated with prostate cancer

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ABSTRACT

Background: The present study investigated the association of serum parathyroid hormone (PTH), vitamin D, and calcium levels with prostate cancer (CaP).

Methods: The study population consisted of an experimental group [459 patients including 216 patients with CaP and 243 patients with benign prostate hyperplasia (BPH)] and a prostatectomy group (47 patients who underwent radical prostatectomy). Patients with serum creatinine levels >1.4 mg/dl, parathyroid disease, and/or PTH levels <10 pg/ml were excluded. Patients with CaP and patients with BPH were compared, and the correlation between serum parameters and clinical data was determined. Preoperative and postoperative PTH levels were compared in the prostatectomy group.

Results: Mean PTH levels were 41.67 ± 28.82 and 27.06 ± 17.32 pg/ml in the CaP and BPH groups, respectively ($p < 0.001$). When patients were divided into two groups as per prostate-specific antigen levels (≤ 20 or > 20 ng/ml), Gleason score (≤ 7 or ≥ 8), and stage ($\leq T3$ or $\geq T4$), there was no significant difference in PTH levels between the two groups. Mean postoperative PTH levels (26.93 ± 13.58 pg/ml) were significantly lower than preoperative PTH levels (36.71 ± 21.04 pg/ml) in the same patients who underwent radical prostatectomy.

Conclusion: Serum PTH levels were higher in patients with CaP than in patients with BPH and decreased significantly after radical prostatectomy. The present results suggest an association between serum PTH and CaP. Further large cohort studies are necessary to validate the present data.

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1. Introduction

Prostate cancer (CaP) is the most common malignancy among men in the United States, with an estimated 174,650 new cases diagnosed in 2019.¹ However, the incidence of CaP is lower in Asian countries than in Western countries.² In Korea, CaP was the fourth most common cancer among men in 2016, although its incidence is rapidly increasing.³ The Korea National Cancer Incidence Database

showed that 1437 cases were diagnosed in 1999 and 8952 cases were diagnosed in 2011.⁴

The only established risk factors for CaP are age and race. Vitamin D deficiency was suggested as a risk factor based on an inverse correlation between sun exposure and CaP mortality.⁵ In addition, vitamin D metabolites inhibit the proliferation and invasion of human CaP cells *in vitro*.⁶ A recent nested case–control study showed that high vitamin D levels are associated with an increased risk of CaP.⁷ The association between vitamin D deficiency and CaP remains controversial.

Parathyroid hormone (PTH) is closely related to both vitamin D and serum calcium levels. Under normal physiologic conditions, serum vitamin D levels are inversely correlated with serum PTH levels, and serum PTH regulates serum calcium concentration. An

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in vitro study on CaP cells suggested that PTH plays an integral role in the regulation of prostate growth and metastasis.⁸ In addition, high serum calcium or high serum PTH may increase the risk of fatal CaP.⁹ However, a prospective nested case–control study reported no association between prediagnostic PTH and CaP incidence.⁷ The association between increased serum PTH and CaP therefore remains controversial.

The aim of the present study was to determine whether serum PTH, vitamin D, and calcium levels are associated with CaP.

2. Methods

2.1. Study population

The study population consisted of an experimental group [459 patients, including 216 patients with CaP and 243 patients with benign prostate hyperplasia (BPH)] and a prostatectomy group (47 patients who underwent radical prostatectomy) (Fig. 1). CaP cases were recruited from patients with histologically confirmed primary adenocarcinoma of the prostate at our institute. In the prostatectomy group, Gleason score (GS) and pathologic stage were measured from specimens obtained during radical prostatectomy. In the nonprostatectomy group, GS was measured from specimens obtained by prostate biopsy, and clinical stage was measured by magnetic resonance imaging or computed tomography. Bone metastasis was assessed by whole-body bone scan. Patients with BPH were selected from the database of patients who underwent transurethral resection of the prostate (TURP). BPH cases with serum prostate-specific antigen (PSA) levels >3 ng/ml underwent transrectal prostate biopsy before TURP to rule out the presence of cancer, and those with PSA levels >10 ng/ml were excluded from the study to rule out the possibility of CaP. To prevent the inclusion of patients with chronic kidney disease, patients with creatinine levels >1.4 mg/dl were excluded. In addition, patients with serum PTH levels <10 pg/ml, and those with a history of parathyroid disease including primary hyperparathyroidism, osteoporosis, or any medications that affect PTH levels, were excluded.

The methods used for sample collection and analysis were approved by the Ethics Committee of Chungbuk National University Hospital. All participants provided written informed consent (IRB approval number: 2010-12-010).

2.2. Samples and laboratory tests

On the morning of the day of operation, serum was collected from patients and stored at -80°C until use. To measure postoperative serum PTH levels in the prostatectomy group, serum was collected at the outpatient clinic 3 months after prostatectomy. Serum PSA levels were measured using a quantified monoclonal Immunoradiometric assay (IRMA) radioimmunoassay (Izotop, Budapest, Hungary). PTH and vitamin D levels were measured using an Elecsys 2010 autoanalyzer (Roche Diagnostics, Indianapolis, IN, USA) in accordance with electrochemiluminescence immunoassay principles. Intact PTH levels were measured by a method

based on intact PTH-specific monoclonal antibodies and the sandwich test principle. The vitamin D assay was based on the competitive test principle and a polyclonal vitamin D–specific antibody. All assays were performed in accordance with the manufacturer's instructions. Serum calcium and PSA levels were obtained by preoperative lab. But serum PTH and vitamin D levels was measured later using -80°C –stored serum on the morning of the day of operation.

2.3. Statistical analysis

The baseline characteristics of the patients were compared between CaP and BPH cases using independent *t* tests and Mann–Whitney U test. Correlations between serum PSA levels or prostate size and other clinicolaboratory parameters were assessed by bivariate correlation analysis in patients with CaP and patients with BPH. In patients with CaP, correlations between serum PSA levels, GS, or stage and other clinicolaboratory parameters were assessed. The patients were separated into two groups as per PSA levels (≤ 20 or >20 ng/ml), GS (≤ 7 or ≥ 8), stage ($\leq T3$ or $\geq T4$), or metastasis (nonmetastasis or bone metastasis), and the mean serum PTH levels were compared using independent *t* tests. Comparison of serum PTH, vitamin D, calcium, and PSA in patients with CaP based on bone metastasis was assessed by independent test and Mann–Whitney U test. In patients who underwent radical prostatectomy, preoperative and postoperative serum PTH levels were compared using paired *t* tests. Statistical analyses were performed using the Statistical Package for Social Sciences, version 23, software (SPSS, Inc., Chicago, IL, USA). A *P*-value <0.05 was considered statistically significant.

3. Results

3.1. Baseline characteristics of the test set patients

The baseline characteristics of all patients are shown in Table 1. The mean PTH levels were 41.67 ± 28.82 and 27.06 ± 17.32 pg/ml in patients with CaP and patients with BPH, respectively. The mean PTH levels were significantly higher in patients with CaP than in patients with BPH ($p < 0.001$).

3.2. Correlations between PSA or prostate size and clinicolaboratory parameters in patients with CaP and patients with BPH

As shown in Table 2, PSA levels were correlated with age and serum calcium levels ($r = 0.163$, $p = 0.016$ and $r = -0.367$, $p < 0.001$, respectively) and prostate size was correlated with age and vitamin D levels ($r = 0.164$, $p = 0.017$ and $r = -0.137$, $p = 0.047$, respectively) in patients with CaP. In patients with BPH, PSA levels were correlated with age and prostate size ($r = 0.238$, $p < 0.001$ and $r = 0.543$,

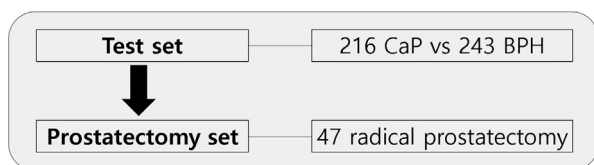


Fig. 1. Schematic shows the flow of the study design. BPH, benign prostate hyperplasia; CaP, prostate cancer.

Table 1

Baseline characteristics of the patients with prostate cancer and patients with BPH

Variables	Prostate cancer (n = 216)	BPH (n = 243)	<i>P</i> -value
Age (years)	68.19 ± 7.12	68.79 ± 7.07	0.369
BMI (kg/m ²)	24.11 ± 2.75	23.71 ± 3.53	0.228
PSA (ng/ml) ^{a)}	14.2 (7.19–37.77)	2.30 (1.10–4.30)	<0.001
Prostate size (g)	37.16 ± 19.52	42.50 ± 23.24	0.008
Vitamin D (pg/ml)	19.27 ± 7.06	18.23 ± 8.10	0.146
PTH (pg/ml)	41.67 ± 28.82	27.06 ± 17.32	<0.001
Serum calcium (mg/dl)	9.25 ± 0.55	9.32 ± 2.07	0.621

BMI, body mass index; BPH, benign prostate hyperplasia; PSA, prostate-specific antigen; PTH, parathyroid hormone.

^{a)} Median (25th–75th percentile).

Table 2
Correlations between PSA or prostate size and clinicolaboratory parameters in patients with CaP and patients with BPH

Variables	PSA		Prostate size		PSA		Prostate size	
	Correlation (r)	P-value	Correlation (r)	P-value	Correlation (r)	P-value	Correlation (r)	P-value
	Prostate cancer				BPH			
Age (years)	0.163	0.016	0.164	0.017	0.238	<0.001	0.100	0.125
BMI (kg/m ²)	-0.127	0.079	0.056	0.440	-0.112	0.145	0.049	0.515
PTH (pg/ml)	-0.044	0.525	-0.077	0.266	0.050	0.450	0.038	0.564
Vitamin D (pg/ml)	-0.028	0.678	-0.137	0.047	0.009	0.895	-0.002	0.980
Calcium (mg/dl)	-0.367	<0.001	-0.085	0.223	0.114	0.083	0.028	0.675
Prostate size (g)	0.103	0.137	-	-	0.543	<0.001	-	-
PSA (ng/ml)	-	-	0.103	0.137	-	-	0.543	<0.001
	Prostate cancer without bone metastasis				Prostate cancer with bone metastasis			
Age (years)	0.144	0.050	0.083	0.262	0.171	0.376	0.040	0.845
BMI (kg/m ²)	-0.038	0.622	0.077	0.321	-0.320	0.136	0.215	0.324
PTH (pg/ml)	-0.112	0.126	-0.033	0.656	-0.080	0.678	-0.266	0.190
Vitamin D (pg/ml)	-0.147	0.045	-0.101	0.171	0.040	0.835	-0.082	0.692
Calcium (mg/dl)	-0.032	0.659	-0.045	0.544	-0.576	<0.001	0.071	0.735
Prostate size (g)	0.252	0.001	-	-	-0.009	0.966	-	-
PSA (ng/ml)	-	-	0.252	0.001	-	-	-0.009	0.966

CaP, prostate cancer; BMI, body mass index; BPH, benign prostate hyperplasia; PSA, prostate-specific antigen; PTH, parathyroid hormone.

$p < 0.001$, respectively) and prostate size was correlated with PSA levels ($r = 0.543$, $p < 0.001$).

3.3. Correlations between PSA, GS, or stage and clinicolaboratory parameters in patients with CaP

In patients with CaP, PSA levels were correlated with age, serum calcium levels, and stage ($r = 0.163$, $p = 0.016$; $r = -0.367$, $p < 0.001$; and $r = 0.188$, $p = 0.006$, respectively). GS was correlated with stage ($r = 0.333$, $p < 0.001$), and stage was correlated with age, serum calcium levels, prostate size, PSA, and GS ($r = 0.347$, $p < 0.001$; $r = -0.189$, $p = 0.006$; $r = 0.214$, $p = 0.002$; $r = 0.188$, $p = 0.006$; and $r = 0.333$, $p < 0.001$, respectively) (Table S1).

In patients who underwent prostatectomy, PSA levels were correlated with prostate size, GS, and stage ($r = 0.235$, $p = 0.003$; $r = -0.340$, $p < 0.001$; and $r = 0.397$, $p < 0.001$, respectively). GS was correlated with PSA levels and stage ($r = 0.340$, $p < 0.001$ and $r = 0.244$, $p = 0.002$, respectively), and stage was correlated with PSA and GS ($r = 0.397$, $p < 0.001$ and $r = 0.244$, $p = 0.002$, respectively) (Table S2).

In patients who did not undergo prostatectomy, PSA levels were correlated with serum calcium levels ($r = -0.484$, $p < 0.001$). GS was correlated with stage ($r = 0.320$, $p = 0.012$), and stage was correlated with GS ($r = 0.320$, $p = 0.012$) (Table S3).

3.4. Comparisons of PTH levels in patients with CaP divided based on PSA, GS, and stage

As shown in Table 3, when the patients were divided into two groups based on PSA levels (≤ 20 or > 20 ng/ml), GS (≤ 7 or ≥ 8), and

Table 3
Comparison of PTH levels in patients with CaP divided based on PSA, GS, and stage

Variables	Based on PSA			Based on GS			Based on stage		
	PSA (≤ 20)	PSA (> 20)	P-value	GS (≤ 7)	GS (≥ 8)	P-value	Stage ($\leq T3$)	Stage ($\geq T4$)	P-value
PTH (pg/ml)	42.5 ± 25.4	40.5 ± 33.1	0.602	43.6 ± 28.3	39.6 ± 29.4	0.313	42.4 ± 29.7	38.3 ± 24.5	0.424
Vitamin D (pg/ml)	20.1 ± 6.7	18.1 ± 7.4	0.034	19.9 ± 6.7	18.6 ± 7.4	0.154	19.5 ± 6.7	18.4 ± 8.6	0.449
Calcium (mg/dl)	9.3 ± 0.5	9.2 ± 0.7	0.072	9.2 ± 0.5	9.2 ± 0.6	0.914	9.3 ± 0.5	9.0 ± 0.8	0.056

CaP, prostate cancer; PSA, prostate-specific antigen; GS, Gleason score; PTH, parathyroid hormone.

stage ($\leq T3$ or $\geq T4$), there was no significant difference in PTH levels between the two groups.

3.5. Comparisons of serum PTH levels, vitamin D, and calcium in patients with CaP based on bone metastasis

The mean PTH level in patients with CaP with nonmetastatic disease was 42.10 ± 29.15 pg/ml, whereas that in patients with bone metastasis was 38.87 ± 26.89 pg/ml ($p = 0.576$). The mean serum

Table 4
Comparison of serum PTH, vitamin D, calcium, and PSA in patients with CaP based on bone metastasis

Variables	Nonmetastasis (n = 187)	Bone metastasis (n = 29)	P-value
PTH (pg/ml)	42.10 ± 29.15	38.87 ± 26.89	0.576
Vitamin D (pg/ml)	19.62 ± 6.67	16.97 ± 9.02	0.138
Calcium (mg/dl)	9.30 ± 0.48	8.89 ± 0.82	0.015
PSA (ng/ml) ^{a)}	11.20 (6.68–25.76)	127.70 (71.91–396.50)	<0.001

CaP, prostate cancer; PTH, parathyroid hormone; PSA, prostate-specific antigen.

^{a)} Median (25th–75th percentile).

Table 5
Comparisons between preoperative and postoperative PTH in patients who underwent radical prostatectomy

Variables	Preoperative	Postoperative	P-value
PTH (pg/ml)	36.71 ± 21.04	26.93 ± 13.58	0.001

PTH, parathyroid hormone.

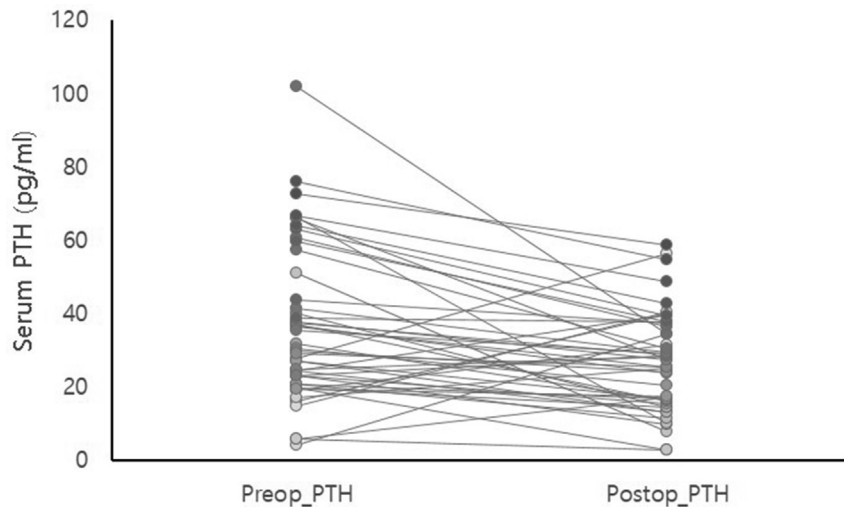


Fig. 2. Changes in preoperative and postoperative PTH levels after radical prostatectomy are shown. PTH, parathyroid hormone.

calcium level of patients with CaP with non-metastatic disease was 9.30 ± 0.48 mg/dl, whereas that of patients with bone metastasis was 8.89 ± 0.82 mg/dl ($p = 0.015$) (Table 4).

3.6. Comparisons between preoperative and postoperative PTH levels in patients who underwent radical prostatectomy

The mean postoperative PTH level (26.93 ± 13.58 pg/ml) was significantly lower than the preoperative PTH level (36.71 ± 21.04 pg/ml) in the same patients who underwent radical prostatectomy ($p = 0.001$) (Table 5, Fig. 2).

4. Discussion

The results of the present study indicated a potential association between serum PTH levels and CaP. In addition, we showed that the mean postoperative PTH level was significantly lower than the preoperative PTH level in the same patients who underwent radical prostatectomy. The present study is the first to show that serum PTH levels decrease after radical prostatectomy and to evaluate the association between serum PTH and CaP in Asian men.

Previous studies suggest that vitamin D deficiency and low vitamin D are associated with CaP.⁵ In addition, vitamin D suppresses the growth and proliferation of CaP cells.⁶ Conversely, a recent study showed that high vitamin D levels in the blood are associated with an increased risk of CaP.^{7,10} A large cohort study conducted in Japan that included 14203 men reported that there was no association between 25-hydroxy vitamin D (25(OH)D) level and the overall incidence of CaP.¹¹ In addition, there was no association between vitamin D and CaP after stratification by intake of fish or calcium. Consistently, no association between vitamin D and CaP was observed in the present study. Therefore, it might be concluded that there is no association between vitamin D and CaP in Asian men.

Vitamin D is closely related to serum calcium. Skinner and Schwartz⁹ reported an approximately threefold increased risk of fatal CaP among men in the upper tertile of the distribution of serum calcium. Brandstedt et al.⁷ reported that only albumin-adjusted calcium was associated with an increased risk of CaP; however, this association was only detected in men aged

55–65 years with a body mass index <25 kg/m². Generally, serum calcium levels in men with advanced CaP are typically normal or low because of the transfer of calcium from the serum to bony lesions.^{12,13} In the present study, we showed that serum calcium was lower in patients with CaP with bone metastasis than in those without bone metastasis. However, there was no association between CaP and serum calcium in the general CaP population in the present study.

Interestingly, Brandstedt et al.⁷ reported that there is no association between PTH and subsequent CaP incidence. However, another study showed that high serum PTH is associated with an increased risk of CaP.⁹ An *in vitro* study suggested that PTH is related to CaP growth.⁸ Most studies of PTH as a risk factor were performed in an advanced/metastatic setting.^{12,13} The present results also showed an association between high PTH levels and CaP in comparison with patients with BPH. However, there was no association between PTH and CaP based on PSA, GS, and stage. The present results showed that PTH levels decreased after radical prostatectomy. This suggests that the relationship between biochemical recurrence (BCR) and decreased PTH levels needs to be evaluated. However, studies on BCR require long-term follow-up; therefore, the relationship between PTH and BCR will be assessed in the future.

In patients with CaP, there was no correlation between GS and PSA. We therefore divided the population into two groups, a prostatectomy group and a nonprostatectomy group, and detected a correlation between GS and PSA in the prostatectomy group. These results might be affected by the large variation in PSA levels in patients who did not undergo prostatectomy.

Generally, patients with CaP with bone metastasis have increased serum levels of PTH.¹⁴ Although there was no statistically significant difference, the present results showed lower PTH levels in patients with bone metastasis than in patients without bone metastasis. We think these results might be affected by the fewer number of patients with bone metastasis. Therefore, large population study and subgroup analysis (localized, locally advanced, and metastatic group) are necessary.

There were several limitations in this study. First, this study had a fewer number of patients and especially patients with

bone metastasis. Large population study and subgroup analysis are necessary to conclude this results. Second, this study had short-term follow-up. The relationship between BCR and decreased PTH levels needs to be evaluated with long-term follow-up.

To the best of our knowledge, this is the first study comparing preoperative and postoperative serum PTH levels after radical prostatectomy and the first to evaluate the association between serum PTH levels and CaP in Asian men.

5. Conclusions

Serum PTH levels were higher in patients with CaP than in patients with BPH and decreased after radical prostatectomy. This study suggests an association between serum PTH and CaP. Further large cohort studies are necessary to validate the present data.

Conflicts of interest

All authors have no conflict of interest to declare.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.prn.2020.02.002>.

References

1. Siegel RL, Miller KD, Jemal A. Cancer statistics. *CA Cancer J Clin* 2019;69(1): 7–34, 2019.
2. Sim HG, Cheng CW. Changing demography of prostate cancer in Asia. *Eur J Canc* 2005;41(6):834–45.
3. Jung KW, Won YJ, Kong HJ, Lee ES. Cancer Statistics in Korea: Incidence, Mortality, Survival, and Prevalence in 2016. *Cancer Res Treat* 2019;51(2): 417–30.
4. Song W, Jeon HG. Incidence of kidney, bladder, and prostate cancers in Korea: An update. *Korean J Urol* 2015;56(6):422–8.
5. Schwartz GG, Hulka BS. Is vitamin D deficiency a risk factor for prostate cancer? (Hypothesis). *Anticancer Res* 1990;10(5A):1307–11.
6. Schwartz GG, Wang MH, Zang M, Singh RK, Siegal GP. 1 alpha,25-Dihydroxyvitamin D (calcitriol) inhibits the invasiveness of human prostate cancer cells. *Cancer Epidemiol Biomarkers Prev* 1997;6(9):727–32.
7. Brändstedt J, Almquist M, Manjer J, Malm J. Vitamin D, PTH, and calcium and the risk of prostate cancer: a prospective nested case-control study. *Cancer Causes Control* 2012;23(8):1377–85.
8. Ritchie CK, Thomas KG, Andrews LR, Tindall DJ, Fitzpatrick LA. Effects of the calcitrophic peptides calcitonin and parathyroid hormone on prostate cancer growth and chemotaxis. *Prostate* 1997;30(3):183–7.
9. Skinner HG, Schwartz G. Serum calcium and incident and fatal prostate cancer in the National Health and Nutrition Examination Survey. *Cancer Epidemiol Biomarkers Prev* 2008;17(9):2302–5.
10. Albanes D, Mondul AM, Yu K, Parisi D, Horst RL, Virtamo J, et al. Serum 25-hydroxy vitamin D and prostate cancer risk in a large nested case-control study. *Cancer Epidemiol Biomarkers Prev* 2011;20(9):1850–60.
11. Sawada N, Inoue M, Iwasaki M, Yamaji T, Shimazu T, Sasazuki S, et al. Plasma 25-hydroxy vitamin D and subsequent prostate cancer risk in a nested Case-Control study in Japan: The JPHC study. *Eur J Clin Nutr* 2017;71(1):132–6.
12. Schwartz G. Prostate cancer, serum parathyroid hormone, and the progression of skeletal metastases. *Cancer Epidemiol Biomarkers Prev* 2008;17(3):478–83.
13. Murray RM, Grill V, Crinis N, Ho PW, Davison J, Pitt P. Hypocalcemic and normocalcemic hyperparathyroidism in patients with advanced prostatic cancer. *J Clin Endocrinol Metab* 2001;86(9):4133–8.
14. Quirosa Flores S, Varsavsky M, Valle Diaz De La Guardia F, Mijan Ortiz JL, Munoz Torres M, Raya Alvarez E, et al. Secondary hyperthyroidism in advanced prostate cancer. *Endocrinol Nutr* 2010;57(3):100–4.