**Research**

# A real-world study of active vitamin D as a prognostic marker in patients with sarcoma

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## Abstract

**Purpose**  The assessment of sarcoma during clinical practice is primarily based on imaging examination, with no effective biomarkers available. Although it has been established that $1,25(OH)_2D_3$ is abnormally expressed in patients with sarcoma, it remains unclear whether $1,25(OH)_2D_3$ level could be used as an evaluation marker in these patient population.
**Methods**  This real-world study investigated $1,25(OH)_2D_3$ level and its association with clinical features in sarcoma patients. Data on $1,25(OH)_2D_3$, parathyroid hormone, calcium, and calcitonin were collected from 331 patients with sarcoma, while the imaging results and the variation in $1,25(OH)_2D_3$ among 213 patients with sarcoma before and after treatment was further analyzed.
**Results**  We found that the serum $1,25(OH)_2D_3$ level was predominantly decreased in patients with sarcoma, with a mean of 45.68 nmol/L. $1,25(OH)_2D_3$ was significantly correlated with the gender and age of sarcoma patients, with more substantial reductions in women and younger patients. Among sarcoma patients, those with progressive disease exhibited a 7.08 nmol/L (−13.73%) decrease in serum $1,25(OH)_2D_3$ levels compared to baseline, while patients with non-progressive disease showed a 1.11 nmol/L (+ 7.0%) increase.
**Conclusion**  The variation of serum $1,25(OH)_2D_3$ can predict the disease status of patients with sarcoma. Decreased serum $1,25(OH)_2D_3$ levels are indicative of disease progression in sarcoma patients, suggesting its potential for application as a prognostic marker for disease assessment in this patient population.

## 1  Introduction

Sarcomas, rare but highly aggressive tumors, encompass a heterogeneous group of mesenchymal neoplasms traditionally divided into two main groups: soft tissue sarcomas and bone sarcomas [1–3]. Currently, the response of anti-cancer therapy in sarcomas are primarily evaluated by imaging without specific and reliable biomarkers. However, it should be borne in mind that repeated imaging examinations appears to bedetrimental to the body, expensive, and inconvenient. Thus, it is essential to find useful biological markers that can identify which patients with sarcoma acquire better response to anti-cancer therapy and guide the optimal timing for imaging evaluation.

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Lingyun Zhang and Wei Li have contributed equally.

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Discover

An increasing body of evidence suggests that vitamin D deficiency might increase the incidence and mortality risk for many kinds of cancer, including colorectal, prostate, breast, and lung cancer [4–9]. The active hormonal form of vitamin D is 1,25-dihydroxyvitamin D ($1,25(OH)_2D_3$), also known as calcitriol, which has been reported to regulate gene expression and play an essential role in cell differentiation and growth [10]. Current evidence suggests that the half-lives of vitamin D and $1,25(OH)_2D_3$ are 24 h and 4 h respectively, which indicates that $1,25(OH)_2D_3$ may represent the actual functional level of vitamin D, for the reason that $1,25(OH)_2D_3$ have shorter half-lives and are the active hormonal form of vitamin D. $1,25(OH)_2D_3$ is a kind of hormone with anti-cancer and anti-inflammatory activity [11]. However, the status of $1,25(OH)_2D_3$ in patients with sarcoma has been largely underinvestigated, warranting further research.

During clinical practice, decreased serum $1,25(OH)_2D_3$ level is commonly observed in many patients with sarcoma. However, whether the change of $1,25(OH)_2D_3$ is related to the disease development of patients with sarcoma has not been reported. Therefore, this study analyzed the expression of $1,25(OH)_2D_3$ in sarcoma patients at our hospital and further explored whether the change in serum $1,25(OH)_2D_3$ level could predict sarcoma prognosis.

## 2 Materials and methods

### 2.1 Study design and patients

This retrospective study included sarcoma patients treated between January 2021 and November 2022. This study protocol was approved by the Ethics Committee (Approval No. B2020-338). Given that this real-world study involved retrospectively collected data, the requirement for individual consent was waived by the Ethics Committee of Zhongshan Hospital of Fudan University. All data were anonymized before data processing.

Inclusion criteria: (1) sarcoma confirmed by histology; (2) blood test results are available, including serum $1,25(OH)_2D_3$, parathyroid hormone (PTH), calcium, and calcitonin; (3) no synchronous or metachronous cancer; (4) no severe liver and kidney insufficiency.

### 2.2 Data collection

Data were collected on various demographic and clinical parameters, including gender, age, stage, pathology, sarcoma location, and treatment phase. Serum $1,25(OH)_2D_3$, PTH, calcium, and calcitonin were further collected and analyzed before and after disease evaluation. The disease assessment was executed by the Response Evaluation Criteria in Solid Tumors version 1.1. The normal concentration range for $1,25(OH)_2D_3$ was > 50 nmol/L [12]. The normal concentration range for calcium was between 2.15 to 2.55 mmol/L. The normal concentration range for PTH was 15 to 65 pg/ml, while for calcitonin, it was 0 to 6.4 pg/ml.

### 2.3 Statistical analysis

Categorical data were displayed in numbers and further examined by the chi-square test. The values of continuous variables were presented as means ± SEM (standard error of the mean). The results were compared using unpaired t-tests and ordinary one-way ANOVA tests.

## 3 Results

### 3.1 Characteristics of the patients

From January 2021 to November 2022, 331 patients with a mean age of 46 (14 to 83 years) met the inclusion criteria, exhibiting female predominance (n = 183, 55.3%). A comprehensive data collection was conducted for all patients (Table 1). Most sarcoma patients were in the 40 to 60 years old group. The pathological types of sarcomas mainly include leiomyosarcoma (LMS), liposarcoma (LPS), and undifferentiated pleomorphic sarcoma (UPS), etc. LMS was the most common pathological type. More than 70% of patients (n = 242) with sarcoma presented with stage IV disease. Among the patients, 56.19% (n = 186) were undergoing first-line treatment, while 15.41% (n = 51) were receiving second-line treatment, and 14.80% (n = 49) were receiving adjuvant treatment. Interestingly, the abdomen and limbs were the most

**Table 1** The characteristics of patients with sarcoma

| Variable | Number | Percent (%) |
|---|---|---|
| Gender | | |
| Male | 148 | 44.71 |
| Female | 183 | 56.29 |
| Age | | |
| ≤ 20 | 20 | 6.04 |
| ≤ 40 | 97 | 29.31 |
| ≤ 60 | 150 | 45.32 |
| > 60 | 64 | 19.34 |
| Pathology | | |
| LMS | 69 | 20.85 |
| LPS | 55 | 16.62 |
| UPS | 34 | 10.27 |
| Angiosarcoma | 26 | 7.86 |
| DSRCT | 22 | 6.65 |
| SS | 21 | 6.34 |
| Osteosarcoma | 21 | 6.34 |
| Fibrosarcoma | 20 | 6.04 |
| RMS | 14 | 4.23 |
| Ewing | 10 | 3.02 |
| Other | 39 | 11.78 |
| Stage | | |
| I | 22 | 6.65 |
| II | 15 | 4.53 |
| III | 52 | 15.71 |
| IV | 242 | 73.11 |
| Treatment | | |
| Adjuvant | 49 | 14.80 |
| First | 186 | 56.19 |
| Second | 51 | 15.41 |
| Posterior | 31 | 9.37 |
| Neoadjuvant | 14 | 4.23 |
| Site | | |
| Head/neck | 15 | 4.53 |
| Chest | 51 | 15.41 |
| Abdomen | 133 | 40.18 |
| Pelvic | 59 | 17.82 |
| Limbs | 73 | 22.05 |
| Total | 331 | 100.00 |

*LMS* leiomyosarcoma, *LPS* liposarcoma, *RMS* Rhabdomyosarcoma, *SS* synovial sarcoma, *UPS* undifferentiated pleomorphic sarcoma, *AS* angiosarcoma, *DCSRT* desmoplastic small round cell tumor and small round cell sarcoma, *Ewing* non-bone Ewing sarcoma, *FS* fibrosarcoma, *PTH* parathyroid hormone

commonly affected sites, accounting for 40.18% and 22.05%, respectively. Notably, among patients with abdominal involvement, the retroperitoneum accounted for the majority of those cases (n = 96, 29%).

### 3.2  Relationship between 1,25(OH)$_2$D$_3$ status and clinical outcomes

The mean serum 1,25(OH)$_2$D$_3$ level in these patients with sarcoma was 45.68 ± 1.06 nmol/L, lower than the standard normal level (> 50 nmol/L). However, the median serum PTH, calcium, and calcitonin were all within the normal range. The mean serum level of 1,25(OH)$_2$D$_3$ was 50.65 ± 1.68 and 41.67 ± 1.28 nmol/L for male and female sarcoma patients, respectively. The concentration of 1,25(OH)$_2$D$_3$ in females was significantly lower than in males ($P < 0.01$, Table 2). The

concentration of PTH in females was slightly higher than in males (43.14 vs. 37.66 pg/ml), with a significant difference ($P = 0.024$). Furthermore, there were slight variations in $1,25(OH)_2D_3$ levels based on age in sarcoma patients. Patients below 60 years old exhibited lower levels of $1,25(OH)_2D_3$, which were below the normal range. However, among patients above 60 years old, the levels of $1,25(OH)_2D_3$ were within the normal range. In this respect, among patients younger than 20 years old age, the serum concentration of $1,25(OH)_2D_3$ was the lowest, with a mean value of approximately $33.80 \pm 2.18$ nmol/L.

During subgroup analysis according to pathological type and tumor clinical stage, $1,25(OH)_2D_3$ serum concentration was not significantly different. Although the serum concentration of $1,25(OH)_2D_3$ was higher in the patients with adjuvant

**Table 2** Serum level of $1,25(OH)_2D_3$ and PTH in patients with sarcoma

| Variable | $1,25(OH)_2D_3$ | | | PTH | | |
|---|---|---|---|---|---|---|
| | Mean | SEM | P-value | Mean | SEM | P-value |
| Gender | | | <0.01[a] | | | 0.03[a] |
| Male | 50.65 | 1.68 | | 37.66 | 1.77 | |
| Female | 41.67 | 1.28 | | 43.14 | 1.66 | |
| Age | | | <0.01 | | | 0.83 |
| ≤20 | 33.80 | 2.18 | | 33.91 | 3.71 | |
| ≤40 | 39.07 | 1.60 | | 40.64 | 2.11 | |
| ≤60 | 47.82 | 1.55 | | 42.01 | 2.09 | |
| >60 | 54.41 | 2.75 | | 39.79 | 2.05 | |
| Pathology | | | 0.37 | | | 0.11 |
| LMS | 45.85 | 2.08 | | 45.36 | 2.85 | |
| LPS | 48.60 | 2.97 | | 45.50 | 3.37 | |
| UPS | 45.16 | 3.54 | | 28.52 | 3.50 | |
| AS | 41.47 | 4.56 | | 34.91 | 3.29 | |
| DSRCT | 51.05 | 3.34 | | 37.20 | 3.06 | |
| SS | 48.73 | 3.54 | | 44.10 | 5.93 | |
| Osteo | 48.33 | 2.66 | | 37.27 | 1.93 | |
| FS | 39.44 | 2.03 | | 34.11 | 2.18 | |
| RMS | 41.35 | 4.01 | | 43.17 | 5.21 | |
| Ewing | 41.52 | 4.16 | | 36.67 | 3.12 | |
| Other | 42.12 | 3.08 | | 40.78 | 3.83 | |
| Stage | | | 0.05 | | | 0.04 |
| I | 43.90 | 3.64 | | 32.80 | 2.65 | |
| II | 49.76 | 4.72 | | 29.41 | 2.68 | |
| III | 52.01 | 3.07 | | 39.40 | 2.69 | |
| IV | 44.23 | 1.20 | | 42.38 | 1.52 | |
| Treatment | | | 0.55 | | | <0.01 |
| Adjuvant | 48.84 | 2.69 | | 31.12 | 1.69 | |
| First | 45.38 | 1.39 | | 40.73 | 1.60 | |
| Second | 43.20 | 2.79 | | 47.44 | 4.01 | |
| Posterior | 46.30 | 3.01 | | 46.86 | 3.99 | |
| Neoadjuvant | 46.24 | 7.20 | | 35.40 | 3.98 | |
| Site | | | 0.28 | | | 0.15 |
| Head/neck | 41.25 | 4.03 | | 38.21 | 3.47 | |
| Chest | 48.51 | 2.92 | | 41.82 | 3.63 | |
| Abdomen | 44.08 | 1.63 | | 42.59 | 1.94 | |
| Pelvic | 44.06 | 2.19 | | 43.13 | 3.27 | |
| Limbs | 48.84 | 2.47 | | 34.97 | 1.95 | |
| Total | 45.68 | 1.06 | | 40.69 | 1.22 | |

*AS* angiosarcoma, *DCSRT* desmoplastic small round cell tumor and small round cell sarcoma, *Ewing* Ewing sarcoma, *FS* fibrosarcoma, *LMS* leiomyosarcoma, *LPS* liposarcoma, *Osteo* Osteosarcoma, *PTH* parathyroid hormone, *RMS* Rhabdomyosarcoma, *SS* synovial sarcoma, *UPS* undifferentiated pleomorphic sarcoma

[a]Unpaired t test

therapy (48.84 ± 2.69 nmol/L), the difference was not significant. Besides, there was no correlation between the level of $1,25(OH)_2D_3$ and tumor sites (Table 2). The serum concentration of PTH was slightly different after stratification according to gender, tumor stages, and tumor treatment phase but remained within the normal range (Table 2). We also analyzed calcium and calcitonin in sarcoma patients (Table 3). The concentrations of calcium and calcitonin were both within the normal range. Further stratification according to gender revealed that calcium and calcitonin were significantly different, indicating calcium and calcitonin levels were higher in male patients. Additionally, the concentration of calcitonin was slightly different based on the sarcoma treatment phase, and calcitonin levels were different based on tumor sites, but all values remained within the normal range.

**Table 3** Serum calcium and calcitonin concentration in patients with sarcoma

| Variable | Calcium | | | Calcitonin | | |
|---|---|---|---|---|---|---|
| | Mean | SEM | *P*-value | Mean | SEM | *P*-value |
| Gender | | | 0.02[a] | | | < 0.01[a] |
| Male | 2.34 | 0.01 | | 4.09 | 0.30 | |
| Female | 2.31 | 0.01 | | 1.25 | 0.10 | |
| Age | | | 0.13 | | | 0.52 |
| ≤ 20 | 2.37 | 0.02 | | 1.78 | 0.30 | |
| ≤ 40 | 2.34 | 0.01 | | 2.71 | 0.36 | |
| ≤ 60 | 2.32 | 0.01 | | 2.39 | 0.21 | |
| > 60 | 2.32 | 0.01 | | 2.75 | 0.39 | |
| Pathology | | | 0.16 | | | 0.38 |
| LMS | 2.32 | 0.01 | | 1.87 | 0.20 | |
| LPS | 2.30 | 0.02 | | 2.08 | 0.33 | |
| RMS | 2.39 | 0.02 | | 2.25 | 0.64 | |
| SS | 2.32 | 0.03 | | 2.96 | 0.61 | |
| UPS | 2.30 | 0.02 | | 2.78 | 0.40 | |
| AS | 2.34 | 0.02 | | 2.20 | 0.28 | |
| DSRCT | 2.34 | 0.01 | | 2.81 | 0.28 | |
| Osteo | 2.36 | 0.02 | | 2.96 | 0.72 | |
| Ewing | 2.30 | 0.02 | | 2.44 | 0.84 | |
| FS | 2.30 | 0.03 | | 3.62 | 1.07 | |
| Other | 2.35 | 0.02 | | 3.16 | 0.54 | |
| Stage | | | 0.31 | | | 0.48 |
| I | 2.35 | 0.02 | | 2.21 | 0.43 | |
| II | 2.35 | 0.03 | | 2.83 | 0.54 | |
| III | 2.35 | 0.02 | | 3.07 | 0.47 | |
| IV | 2.32 | 0.01 | | 2.41 | 0.19 | |
| Treatment | | | 0.01 | | | 0.45 |
| Adjuvant | 2.37 | 0.02 | | 2.56 | 0.39 | |
| First | 2.32 | 0.01 | | 2.59 | 0.24 | |
| Second | 2.32 | 0.02 | | 2.20 | 0.29 | |
| Posterior | 2.28 | 0.02 | | 2.23 | 0.35 | |
| Neoadjuvant | 2.31 | 0.02 | | 3.24 | 0.88 | |
| Site | | | 0.19 | | | < 0.01 |
| Head/neck | 2.37 | 0.02 | | 2.58 | 0.69 | |
| Chest | 2.33 | 0.01 | | 2.62 | 0.33 | |
| Abdomen | 2.31 | 0.01 | | 2.13 | 0.19 | |
| Pelvic | 2.31 | 0.01 | | 1.94 | 0.29 | |
| Limbs | 2.34 | 0.02 | | 3.60 | 0.52 | |
| Total | 2.33 | 0.01 | | 2.52 | 0.16 | |

[a]Unpaired t test

### 3.3 Characteristics of patients stratified according to 1,25(OH)$_2$D$_3$

To further analyze the potential value of 1,25(OH)$_2$D$_3$ in sarcoma patients, we stratified sarcoma patients according to 1,25(OH)$_2$D$_3$ serum concentration and divided them into normal-level (> 50 nmol/L) and low-level (≤ 50 nmol/L) groups. The results showed that 1,25(OH)$_2$D$_3$ expression was lower than normal in 208 (62.8%) patients with sarcoma and remained within the normal range in 123 (37.2%) patients (Supplementary Table 1). The average 1,25(OH)$_2$D$_3$ concentration was 66.25 nmol/L in the normal-level group and 33.52 nmol/L in the low-level group (Table 4). The serum concentration of 1,25(OH)$_2$D$_3$ was correlated with sex in both normal-level and low-level groups ($P < 0.05$). The reduction of 1,25(OH)$_2$D$_3$ was more likely in female patients (n = 128) than in male patients (n = 80). The levels of 1,25(OH)$_2$D$_3$

**Table 4** Serum level of 1,25(OH)$_2$D$_3$ in patients with sarcoma cut-off by 50 nmol/L

| 1,25(OH)$_2$D$_3$ | Normal-level | | | Low-level | | |
|---|---|---|---|---|---|---|
| | Mean | SEM | *P*-value | Mean | SEM | *P*-value |
| Gender | | | 0.03[a] | | | 0.02[a] |
| Male | 68.62 | 1.76 | | 35.37 | 1.03 | |
| Female | 63.33 | 1.61 | | 32.36 | 0.81 | |
| Age | | | 0.03 | | | 0.06 |
| ≤ 20 | 57.95 | 2.51 | | 31.11 | 1.34 | |
| ≤ 40 | 60.56 | 1.68 | | 31.61 | 1.15 | |
| ≤ 60 | 66.27 | 1.79 | | 35.17 | 0.97 | |
| > 60 | 70.77 | 2.52 | | 34.67 | 1.73 | |
| Pathology | | | 0.21 | | | 0.31 |
| LMS | 64.96 | 2.64 | | 35.66 | 1.26 | |
| LPS | 69.07 | 3.59 | | 33.89 | 1.79 | |
| UPS | 64.48 | 2.08 | | 37.44 | 0.82 | |
| AS | 64.79 | 6.18 | | 29.81 | 2.84 | |
| DSRCT | 67.24 | 2.53 | | 32.84 | 1.79 | |
| SS | 64.93 | 3.36 | | 34.86 | 1.26 | |
| Osteo | 68.61 | 6.70 | | 34.29 | 2.21 | |
| FS | 63.72 | 2.60 | | 34.11 | 2.18 | |
| RMS | 62.40 | 5.30 | | 31.85 | 2.23 | |
| Ewing | 63.97 | 3.64 | | 36.09 | 2.46 | |
| Other | 66.02 | 2.63 | | 29.42 | 2.25 | |
| Stage | | | 0.20 | | | 0.59 |
| I | 62.31 | 3.56 | | 33.39 | 2.60 | |
| II | 68.35 | 5.50 | | 37.37 | 2.38 | |
| III | 71.15 | 3.02 | | 34.28 | 1.71 | |
| IV | 65.02 | 1.44 | | 33.18 | 0.75 | |
| Treatment | | | 0.54 | | | 0.40 |
| Adjuvant | 65.44 | 2.50 | | 34.17 | 1.76 | |
| First | 66.86 | 1.69 | | 34.12 | 0.82 | |
| Second | 65.34 | 3.11 | | 31.13 | 1.77 | |
| Posterior | 62.68 | 2.91 | | 34.47 | 2.00 | |
| Neoadjuvant | 74.90 | 10.76 | | 30.32 | 3.30 | |
| Site | | | 0.95 | | | 0.66 |
| Head/neck | 63.75 | 4.62 | | 33.06 | 1.88 | |
| Chest | 67.88 | 2.48 | | 32.59 | 2.00 | |
| Abdomen | 65.63 | 2.11 | | 33.07 | 0.93 | |
| Pelvic | 65.24 | 2.45 | | 35.49 | 1.54 | |
| Limbs | 66.79 | 2.73 | | 33.19 | 1.47 | |
| Total | 66.25 | 1.24 | | 33.52 | 0.65 | < 0.01 |

[a]Unpaired t test

were higher in male patients with sarcoma than in female patients in both normal-level and low-level groups. In the normal-level group, $1,25(OH)_2D_3$ was also upregulated with increased age ($P < 0.05$). However, there was no correlation between $1,25(OH)_2D_3$ and age in the low-level group. After stratification by serum levels of $1,25(OH)_2D_3$, we analyzed the correlation between $1,25(OH)_2D_3$ and the pathological characteristics, clinical stage, disease progression stage, and location of sarcoma. However, there was no significant correlation between them.

Next, we analyzed the differences in PTH, calcium, and calcitonin between the normal-level and low-level groups (Supplementary Table 2). PTH was slightly higher in the $1,25(OH)_2D_3$ low-level group than in the high-level group (42.81 vs. 37.10 nmol/L, Supplementary Table 2). PTH exhibited differences with age and treatment phase in the $1,25(OH)_2D_3$ normal-level group. However, there was no difference in PTH with age in the $1,25(OH)_2D_3$ low-level group. Calcitonin was also lower in the $1,25(OH)_2D_3$ low-level group, but there was no statistical difference (Supplementary Table 3). Calcitonin was highly expressed in male patients with sarcoma in both low-level and normal-level groups. Calcium concentration was also lower in the $1,25(OH)_2D_3$ low-level group (Supplementary Table 4). Calcium was associated with age and treatment phase in the $1,25(OH)_2D_3$ low-level group. However, it is worth noting that the mean values of PTH, calcitonin, and calcium in all sarcoma patients were within the normal range, irrespective of grouping.

### 3.4  $1,25(OH)_2D_3$ and survival analysis

Among the 331 patients with sarcoma, 213 underwent baseline and post-treatment imaging assessments and the serum data of $1,25(OH)_2D_3$, calcium, calcitonin, and PTH. The disease assessment was executed by the Response Evaluation Criteria in Solid Tumors version 1.1 (RECIST 1.1). 90 patients had progressive disease (PD), while 213 were classified with non-progressive disease (non-PD, including complete remission, partial remission, and disease stabilization).

Further analysis showed that the serum level of $1,25(OH)_2D_3$ decreased by 7.08 nmol/L in the PD group, while it increased by 1.11 nmol/L in the non-PD group, compared with baseline in sarcoma patients ($P < 0.01$). $1,25(OH)_2D_3$ was decreased by 13.74% in the PD group, while it increased by 6.98% in the non-PD group ($P < 0.01$). The serum concentration of PTH increased in both non-PD and PD groups. Although the increase in PTH levels was higher in the PD group compared to the non-PD group, there was no significant difference. Furthermore, there were no significant differences in calcium and calcitonin levels between the non-PD and PD groups (Table 5).

## 4  Discussion

Sarcomas comprise several heterogeneous subtypes of mesenchymal tumors, posing challenges in their assessment due to reliance on imaging tests and a lack of specific tumor biomarkers. In clinical practice, abnormal $1,25(OH)_2D_3$ levels have been observed in patients with a wide range of sarcoma. However, few studies have been conducted on the correlation between $1,25(OH)_2D_3$ level and the progonosis and clinical features of sarcoma, especially on the potential value of $1,25(OH)_2D_3$ as a prognostic marker in patients with sarcoma. In this study, we comprehensively analyzed the clinical characteristics of sarcoma patients in our hospital and uncovered that LMS and LPS were the most common types. These sickness have the special age distribution. Data were collected from 331 Chinese patients with a mean age of 46 years old at baseline and an age range from 40 to 60 years. In addition, the most common site of sarcoma in our present study is abdomen, especially the retroperitoneum, which is slightly inconsistent with the published research [2].

A past study has documented that $1,25(OH)_2D_3$ deficiency was discovered in 28% of patients with sarcoma [12]. However, our study found that $1,25(OH)_2D_3$ level was decreased in more than 60% of patients with sarcoma. Our study found that $1,25(OH)_2D_3$ level was lower in females with sarcoma than males with sarcoma. However, the

**Table 5** Comparison of variables in patients with sarcoma with tumor evaluation

| Variables | Concentration | | | Percent (%) | | |
|---|---|---|---|---|---|---|
| | Non-PD | PD | *P*-value | Non-PD | PD | *P-value* |
| $1,25(OH)_2D$ | 1.11 | −7.08 | 0.03 | 6.98 | −13.74 | < 0.01 |
| PTH | 3.98 | 10.92 | 0.08 | 18.77 | 62.61 | 0.05 |
| Calcium | −0.06 | −0.07 | 0.78 | −2.35 | −2.70 | 0.84 |
| Calcitonin | −0.28 | −0.40 | 0.70 | 2.92 | −0.01 | 0.65 |

*Non-PD* non- progressive disease, *PD* progressive disease

reference [12] showed the results that the mean $1,25(OH)_2D_3$ concentration was $101.6 \pm 8.7$ nmol/l for females with sarcoma than $82.4 \pm 5.9$ males with sarcoma, which has no significant value. We noted the sample number of that study was very small, which only 15 males and 10 females. And there was a significant difference of age between female sarcoma patients and male sarcoma patients. So we conduct the difference in sample size and age may be one of the reasons for the different results. Extensive studies during the past few decades revealed that $1,25(OH)_2D_3$ is the major regulator of calcium homeostasis and protects the organism from calcium deficiency via effects on the intestine, kidney, parathyroid gland, and bone [13]. Therefore, patients with renal insufficiency and thyroid dysfunction were excluded from our data. Vitamin D has a complicated role in bone, which stimulates matrix formation and bone maturation, enhances osteoclast activity, and may affect the cell differentiation of osteocyte precursors [14]. During stratification according to pathological types, we also analyzed patients with osteosarcoma and chondrosarcoma. However, we found there is no correlation between $1,25(OH)_2D_3$ and pathological types in sarcomas, which suggests that the difference in pathological types was not responsible for the abnormal changes in $1,25(OH)_2D_3$ level. Next, we analyed the relationship between $1,25(OH)_2D_3$ and sarcoma patients' clinical features and found that serum concentration of $1,25(OH)_2D_3$ was not associated with clinical features, such as tumor stage, site, and treatment phase. However, serum $1,25(OH)_2D_3$ level correlated with patient gender and age, suggesting that systemic factors were the main factor influencing the serum $1,25(OH)_2D_3$ level.

It has been established that multiple factors influence $1,25(OH)_2D_3$ metabolism. The production of $1,25(OH)_2D_3$ is tightly regulated by serum PTH, calcium, and calcitonin [15]. Thus, to clarify the abnormal expression of $1,25(OH)_2D_3$ in sarcoma, we also analyzed serum PTH, calcium, and calcitonin levels. PTH levels were within the normal range (15–65 pg/ml) for 85% of sarcoma patients. The calcium and calcitonin levels of more than 90% sarcoma patients were in the normal region. Taken together, these data revealed no significant alterations in PTH, calcium, or calcitonin among patients with sarcoma, which indicates that $1,25(OH)_2D_3$ variation in patients with sarcoma might be independent of PTH, calcium, and calcitonin. Compared to PTH, calcium, and calcitonin, $1,25(OH)_2D_3$ was more closely associated with disease development in patients with sarcoma.

We found that serum $1,25(OH)_2D_3$ level was downregulated in more than half of patients with sarcoma. To further understand the clinical characteristics of sarcoma patients based on their $1,25(OH)_2D_3$ levels, we divided them into a low-level group and a normal-level group. Our results revealed that female patients and younger patients were more likely to have a lower concentration of $1,25(OH)_2D_3$. However, there was no significant difference in $1,25(OH)_2D_3$ levels after stratification according to pathological types, tumor stage, treatment phase, or tumor site.

A meta-analysis of observational studies revealed that women with the highest blood levels of $1,25(OH)_2D_3$ experienced a significantly reduced risk of breast cancer [16]. Recent reports suggested that maintaining vitamin D levels within the normal range during anti-PD-1 inhibitor therapy in patients with advanced melanoma may improve clinical outcomes [17]. Vitamin D deficiency was found in Kaposi sarcoma patients but was not associated with tumor response and survival time [18]. However, it remains unclear whether the variation in serum concentration of $1,25(OH)_2D_3$ was associated with the survival of sarcoma patients. Therefore, we then employed the value of serum $1,25(OH)_2D_3$ level before and after treatment in sarcoma patients that underwent imaging evaluation. After further analysis, we found that the variation in serum $1,25(OH)_2D_3$ level was consistent with the outcome of the imaging evaluation. Accordingly, decreased $1,25(OH)_2D_3$ indicated tumor progression, and increased $1,25(OH)_2D_3$ indicated tumor stability. Emerging evidence suggests that lower vitamin D level is associated with a higher cancer incidence [19, 20]. Interestingly, vitamin D supplementation may improve the supportive hazard ratio of metastatic colorectal cancer patitents,- and contributes to longer two months of progression-free survival [21]. Our findings suggest that an increased $1,25(OH)_2D_3$ serum level may predict a better treatment effect. However, whether exogenous $1,25(OH)_2D_3$ supplementation can prolong the survival of sarcoma patients remains to be further investigated.

$1,25(OH)_2D_3$ has been reported to be important in regulating cancer cell proliferation, apoptosis, and angiogenesis [22, 23]. A recent studies have unraveled that $1,25(OH)_2D_3$ induced autophagosome formation, increased autophagy-related protein light chain (LC)3II/LC3I levels, and decreased ubiquitin-binding protein P62 expression [24]. The supplementation of optimal levels of $1,25(OH)_2D_3$ may have the ability to reduce cancer risk [25]. Treatment with $1,25(OH)_2D_3$ combined with active anticancer agents may prevent the onset of cancer and delay its progression [26]. Nonetheless, the mechanism underlying the association between $1,25(OH)_2D_3$ and the prediction of sarcoma patient survival requires further investigation.

## 5 Conclusion

This study uncovered the clinical and biological significance of the potent regulator $1,25(OH)_2D_3$ and suggested that the serum level of $1,25(OH)_2D_3$ may serve as a potential prognostic marker for sarcoma patients. The observed decrease of $1,25(OH)_2D_3$ in the majority of sarcoma patients indicated its potential role in disease progression. Accordingly, monitoring the variation in this biological marker could assist in predicting the development and outcomes of sarcoma.

**Author contributions**  Each author participated sufficiently in the work to take responsibility for appropriate portions of the content. LZ and WL participated in the research design, collected and analyzed the data of patient, and wrote the manuscript. YZ contributed to the conception and study design. XW, YS, and RZ supervised the study, and edited the manuscript. All authors read and approved the final version of the manuscript.

**Data availability**  The data sets generated and/or analyzed during the current study are not publicly available, but are available from the corresponding author on reasonable request.

## Declarations

**Ethics approval and consent to participate**  All procedures have been approved by the appropriate ethics committee and have therefore been performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki and its later amendments. This study was approved by approved by the Institutional Ethics Committee of Zhongshan Hospital of Fudan University (Approval No. B2020-338). All data were anonymized before processing.

**Competing interests**  The authors declared that no competing interest exists.

## References

1. Skubitz KM, D'Adamo DR. Sarcoma. Mayo Clin Proc. 2007;82(11):1409–32.
2. Gilbert NF, Cannon CP, Lin PP, Lewis VO. Soft-tissue sarcoma. J Am Acad Orthop Surg. 2009;17(1):40–7.
3. Sbaraglia M, Bellan E, Dei Tos AP. The 2020 WHO classification of soft tissue tumours: news and perspectives. Pathologica. 2021;113(2):70–84.
4. Gandini S, Boniol M, Haukka J, Byrnes G, Cox B, Sneyd MJ, Mullie P, Autier P. Meta-analysis of observational studies of serum 25-hydroxy-vitamin D levels and colorectal, breast and prostate cancer and colorectal adenoma. Int J Cancer. 2011;128(6):1414–24.
5. Zhou W, Heist RS, Liu G, Asomaning K, Neuberg DS, Hollis BW, Wain JC, Lynch TJ, Giovannucci E, Su L, et al. Circulating 25-hydroxyvitamin D levels predict survival in early-stage non-small-cell lung cancer patients. J Clin Oncol. 2007;25(5):479–85.
6. Giovannucci E. Vitamin D and cancer incidence in the harvard cohorts. Ann Epidemiol. 2009;19(2):84–8.
7. Jeon SM, Shin EA. Exploring vitamin D metabolism and function in cancer. Exp Mol Med. 2018;50(4):1–14.
8. Cruz-Pierard SM, Nestares T, Amaro-Gahete FJ. Vitamin D and calcium as key potential factors related to colorectal cancer prevention and treatment: a systematic review. Nutrients. 2022;14(22):4934.
9. Arayici ME, Basbinar Y, Ellidokuz H. Vitamin D intake, serum 25-hydroxyvitamin-D (25(OH)D) levels, and cancer risk: a comprehensive meta-meta-analysis including meta-analyses of randomized controlled trials and observational epidemiological studies. Nutrients. 2023;15(12):2722.
10. Takeyama K, Kitanaka S, Sato T, Kobori M, Yanagisawa J, Kato S. 25-Hydroxyvitamin D3 1alpha-hydroxylase and vitamin D synthesis. Science. 1997;277(5333):1827–30.

11.  Vanoirbeek E, Krishnan A, Eelen G, Verlinden L, Bouillon R, Feldman D, Verstuyf A. The anti-cancer and anti-inflammatory actions of 1,25(OH)(2)D(3). Best Pract Res Clin Endocrinol Metab. 2011;25(4):593–604.
12.  Juzeniene A, Porojnicu AC, Baturaite Z, Lagunova Z, Aksnes L, Bruland OS, Moan J. Vitamin D levels and dietary intake among patients with benign soft tissue tumors and sarcomas. Anticancer Res. 2015;35(2):1171–80.
13.  Dusso AS, Brown AJ, Slatopolsky E. Vitamin D. Am J Physiol Renal Physiol. 2005;289(1):F8-28.
14.  Palmieri GM, Pitcock JA, Brown P, Karas JG, Roen LJ. Effect of calcitonin and vitamin D in osteoporosis. Calcif Tissue Int. 1989;45(3):137–41.
15.  Hughes MR, Brumbaugh PF, Hussler MR, Wergedal JE, Baylink DJ. Regulation of serum 1alpha,25-dihydroxyvitamin D3 by calcium and phosphate in the rat. Science. 1975;190(4214):578–80.
16.  Chen P, Li M, Gu X, Liu Y, Li X, Li C, Wang Y, Xie D, Wang F, Yu C, et al. Higher blood 25(OH)D level may reduce the breast cancer risk: evidence from a Chinese population based case-control study and meta-analysis of the observational studies. PLoS ONE. 2013;8(1): e49312.
17.  Galus L, Michalak M, Lorenz M, Stoinska-Swiniarek R, Tusien Malecka D, Galus A, Kolenda T, Leporowska E, Mackiewicz J. Vitamin D supplementation increases objective response rate and prolongs progression-free time in patients with advanced melanoma undergoing anti-PD1 therapy. Cancer. 2023;129(13):2047–55.
18.  Erlandson KM, Gudza I, Fiorillo S, Ndemera B, Schooley RT, Gwanzura L, Borok M, Campbell TB. Relationship of vitamin D insufficiency to AIDS-associated Kaposi's sarcoma outcomes: retrospective analysis of a prospective clinical trial in Zimbabwe. Int J Infect Dis. 2014;24:6–10.
19.  Garland CF, Garland FC, Gorham ED, Lipkin M, Newmark H, Mohr SB, Holick MF. The role of vitamin D in cancer prevention. Am J Public Health. 2006;96(2):252–61.
20.  Carlberg C, Munoz A. An update on vitamin D signaling and cancer. Semin Cancer Biol. 2022;79:217–30.
21.  Ng K, Nimeiri HS, McCleary NJ, Abrams TA, Yurgelun MB, Cleary JM, Rubinson DA, Schrag D, Miksad R, Bullock AJ, et al. Effect of high-dose vs standard-dose Vitamin D3 supplementation on progression-free survival among patients with advanced or metastatic colorectal cancer: the sunshine randomized clinical trial. JAMA. 2019;321(14):1370–9.
22.  Lopes N, Sousa B, Martins D, Gomes M, Vieira D, Veronese LA, Milanezi F, Paredes J, Costa JL, Schmitt F. Alterations in Vitamin D signalling and metabolic pathways in breast cancer progression: a study of VDR, CYP27B1 and CYP24A1 expression in benign and malignant breast lesions. BMC Cancer. 2010;10:483.
23.  Deeb KK, Trump DL, Johnson CS. Vitamin D signalling pathways in cancer: potential for anticancer therapeutics. Nat Rev Cancer. 2007;7(9):684–700.
24.  Han J, Tang Y, Zhong M, Wu W. Antitumor effects and mechanisms of 1,25(OH)2D3 in the Pfeiffer diffuse large B lymphoma cell line. Mol Med Rep. 2019;20(6):5064–74.
25.  Krishnan AV, Trump DL, Johnson CS, Feldman D. The role of vitamin D in cancer prevention and treatment. Endocrinol Metab Clin North Am. 2010;39(2):401–18.
26.  Luong K, Nguyen LT. The beneficial role of vitamin D and its analogs in cancer treatment and prevention. Crit Rev Oncol Hematol. 2010;73(3):192–201.