Bratislava Medical Journal
https://doi.org/10.1007/s44411-025-00092-x

RESEARCH

Investigation of Changes in Blood Choline Levels in Individuals Diagnosed with Osteoporosis: A Case–Control Study

Volga Ozturk[1] · **Elif Baris**[2]

Received: 4 January 2025 / Revised: 24 February 2025 / Accepted: 4 March 2025

Abstract

Objectives This study aimed to compare blood choline levels and its correlations with bone mineral density (BMD) calcium, vitamin D, and parathyroid hormone (PTH) in patients with osteoporosis and healthy controls.

Background Osteoporosis is a condition marked by reduced bone density and a higher risk of fractures, posing a major health concern. Cholinergic activity promotes bone formation, while its inhibition may contribute to bone loss.

Methods Blood choline levels in 64 female participants, 38 with osteoporosis and 26 healthy controls. were measured using ELISA, and BMD was assessed via Dual-Energy X-Ray Absorptiometry (DEXA). Additional biochemical markers, including calcium, vitamin D, and PTH, were analyzed.

Results Blood choline levels were significantly lower in the osteoporosis group compared to controls. Moderate correlations were observed between choline levels and BMD, as well as weak correlations with calcium, vitamin D, and PTH in patients with osteoporosis.

Conclusion Lower choline levels in osteoporosis patients suggest a potential link between cholinergic deficiency and the disease, emphasizing the need for further research into cholinergic therapies for osteoporosis.

Keywords Choline · Osteoporosis · Bone mineral density · Calcium · Vitamin D · Parathyroid hormone

1 Introduction

Adrenergic and cholinergic stimulation, components of the autonomic nervous system, have functional roles in bone homeostasis and remodeling. Adrenergic stimulation leads to bone loss, whereas cholinergic activity has been shown to support bone formation. In vitro studies indicate that cholinergic activity stimulates the proliferation and differentiation of bone cells. It has been demonstrated that suppression of cholinergic activity in bone tissue and the central nervous system leads to bone loss, while its increase results in bone mass gain [1].

Choline is an essential nutrient involved in numerous physiological processes, including acetylcholine synthesis, membrane structure maintenance, and methyl group donation. In humans, choline can be obtained from dietary sources or synthesized endogenously primarily in the liver. Choline is metabolized into phosphocholine, glycerophosphocholine, and betaine, which play critical roles in cell membrane integrity, neurotransmission, and methylation processes. These metabolic pathways are crucial for maintaining systemic homeostasis, including potential interactions with bone metabolism [2, 3]. Research has shown that osteoblasts and osteoclasts have nicotinic receptors for acetylcholine, suggesting a potential direct role of the parasympathetic system in bone regulation [4]. Additionally, bone morphogenetic proteins (BMPs) are associated with various functions in the developing nervous system, indicating a complex interaction between the nervous system and bone development [5]. Furthermore, choline plays a more prominent role in acetylcholine synthesis and metabolism in the peripheral nervous system compared to the brain, where it directly contributes to acetylcholine production and acts as an agonist at cholinergic receptors [6, 7]. As a result, the

✉ Elif Baris
elif.baris@ieu.edu.tr

[1] Department of Orthopaedics and Traumatology, Turgutlu State Hospital, Subaşı, Son Sk. No:18, Turkey 45400 Turgutlu, Manisa

[2] Medical Pharmacology, Faculty of Medicine, Izmir University of Economics, Fevzi Çakmak, Sakarya Cd. No:156, Türkiye 35330 Balçova, İzmir

Published online: 17 March 2025

parasympathetic system and parasympathomimetic agents, including choline and acetylcholine, play essential roles in various physiological processes, including bone metabolism.

The potential anabolic effect of increasing cholinergic activity on bone formation has been investigated with acetylcholinesterase inhibitors (AChEI). AChEI drugs inhibit the acetylcholine (ACh) degrading enzyme, increasing ACh levels and thereby enhancing cholinergic transmission through cholinergic receptors [8]. Among the cholinergic receptors, alpha 7 nicotinic acetylcholine receptor (α7nAChR) agonists have been shown to balance the OB/OC ratio by stimulating OB proliferation and differentiation [9]. Given the limited clinical effects and side effects of preventive and therapeutic strategies used in osteoporosis treatment (e.g., calcium and vitamin D supplements, bisphosphonates, parathyroid hormone), there is an emphasized need for new approaches to control or treat the disease. Enhancing cholinergic transmission has demonstrated anti-inflammatory, anti-catabolic, and anti-resorptive properties on bone and cartilage, indicating that increasing α7nAChR-mediated cholinergic transmission with pharmacological agents, like choline or ACh, could be a new method for treating bone related disorders like osteoporosis [10, 11].

Current osteoporosis treatments, though effective, often pose concerns for long-term use. While bisphosphonates are widely used to increase bone density, their long-term administration has been associated with rare but serious complications such as atypical femur fractures. Thus, a long-term, safe solution remains essential based on the understanding on pathophysiology of osteoporosis. The complex relationship between the parasympathetic system and osteoporosis requires further investigation. The observed association between lower choline levels and osteoporosis contributes to the understanding of the pathophysiology of osteoporosis and provides a foundation for future research into potential therapeutic approaches [12].

The primary aim of the study is to compare blood choline/ACh levels in individuals clinically diagnosed with osteoporosis to those in healthy controls. The secondary aim of the study is to determine the correlation between blood choline/ACh levels with bone mineral density (BMD), biochemical measurements (calcium, vitamin D, parathyroid hormone) in patients with osteoporosis.

2 Materials and Methods

This case–control study conducted in accordance with the Declaration of Helsinki, Good Clinical Practice Guidelines, and relevant regulations, in collaboration between the Orthopedics and Traumatology department of Turgutlu State Hospital and the Department of Pharmacology at İzmir University of Economics Faculty of Medicine. The ethical approval obtained from the Clinical Research Ethics Committee at İzmir University of Economics (B.30.2.İEUSB.0.05.05-20-300).

3 Participants

The inclusion criteria targeted postmenopausal females aged 40–70 years who were newly diagnosed with osteoporosis (T-score ≤ − 2.5 by DEXA), drug-naïve at the time of enrollment, or healthy controls. A T-score of − 1.0 and above was considered normal, values between − 1.0 and − 2.5 were classified as osteopenia (low bone density), and a T-score of − 2.5 and below indicated osteoporosis. The control group consisted of individuals who did not have a diagnosis of osteoporosis and did not meet the exclusion criteria.

Exclusion criteria consisted of cancer patients, pregnant women, breastfeeding women, individuals with acute infection symptoms, and those with severe cardiovascular diseases, severe respiratory problems, epilepsy, diabetes, and decompensated endocrine diseases and non-smokers and individuals using drugs that could modify parasympathetic activity. No interventions conducted on the volunteers participating in the study.

A patient record form was filled out for all volunteers, containing information on height, weight, age, gender and comorbidities. All participants were clinically evaluated by clinician, based on BMD results and clinical findings.

4 Diagnosis

The patients who have in the history of widespread bone pain who did not respond to conservative treatment were examined in terms of clinical and laboratory values underwent DEXA due to laboratory findings that may indicate abnormal bone mineral density. The majority of these patients had family history, significant height loss, and early menopause among their clinical complaints.

5 Laboratory Analyses

Fasting blood samples, 5 mL of blood was drawn from the volunteers during routine blood collection procedures, were collected from participants between 9:00 and 11:00 a.m. to control for diurnal variation in biochemical parameters. Samples were drawn into serum separator tubes, which were then centrifuged at 3000–3500 rpm for 10 min. The separated serum was stored at − 20 °C in a refrigerator with an uninterrupted power supply until analysis (Figs. 1 and 2).

Serum choline/ACh levels were measured using the commercial kit according to the instructions (Biovision K615,

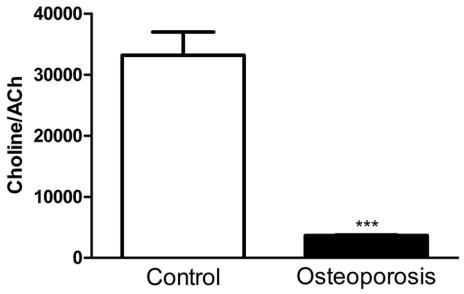

Fig. 1 Serum total choline levels of groups. Shown are serum total levels in control and FMS groups. Mann Whitney U test used for comparison of two groups. ***: $p < 0.001$ versus control

sensitivity: > 0.01 nmol/well). The kit employs a colorimetric method (λ = 570 nm) to quantify choline levels based on the oxidation of free choline via the intermediate betaine aldehyde to betaine. The reaction generates products that interact with the OxiRed Probe to produce a measurable signal. ACh levels were derived by including the acetylcholinesterase enzyme in the reaction mix, enabling measurement of total choline (free choline + ACh). All samples, standards, and controls were run in duplicate to ensure accuracy and consistency. Standard curves were freshly prepared for each assay, and the linearity of the response was confirmed within the assay measurement range [13].

The levels of biochemical parameters, including calcium, vitamin D, and parathyroid hormone, evaluated in hospital. Biochemical parameters, including calcium (CA), 25-hydroxyvitamin D (25VD), and parathyroid hormone (PTH), were measured using standardized immunoassay and colorimetric methods on Siemens Atellica analyzers.

Calcium levels were determined using the Arsenazo III method on the Siemens Atellica CH Analyzer. This method involves a colorimetric assay, where the binding of calcium to the Arsenazo III dye forms a complex measurable at a specific wavelength. The results were expressed in milligrams per deciliter (mg/dL). The concentration of 25-hydroxyvitamin D was assessed using a chemiluminescence-based immunoassay on the Siemens Atellica IM Analyzer. This method detects the total 25-hydroxyvitamin D levels (D2 and D3). Results were recorded in nanograms per milliliter (ng/mL). PTH levels were measured using the same chemiluminescence-based immunoassay on the Siemens Atellica IM Analyzer. This method quantifies intact PTH levels, which were expressed in picograms per milliliter (pg/mL).

All assays were calibrated and validated according to the manufacturer's instructions using Siemens Atellica reagents. Internal and external quality controls were performed regularly to ensure accuracy and precision. The instruments were maintained according to the manufacturer's protocols, and reagent lot-to-lot variability was monitored.

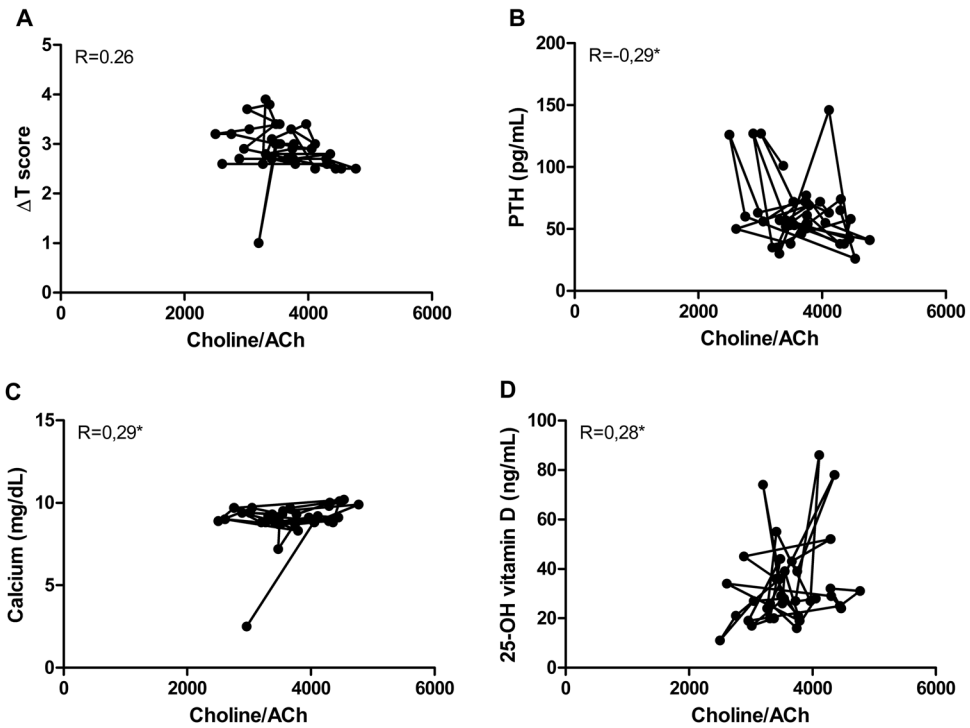

Fig. 2 Correlation between serum total choline levels, BMD and biochemical parameters. Shown are correlation of serum total levels with T score (**A**), PTH (**B**), Calcium (**C**) and 25-OH Vitamin D (**D**). Pearson correlation analysis used for statistical evaluation. *: $p < 0.05$, **: $p < 0.01$

6 BMD Measurement

BMD measurement was conducted to diagnose osteoporosis using a dual-energy X-ray absorptiometry (DEXA) device. Imaging was performed with the DMS Stratos DR device (DMS Imaging, Montpellier, France). All BMD levels were measured at Turgutlu State Hospital. Individuals with a T-score of -2.5 and below were included in the study.

7 Statistical Analysis

Statistical analysis was conducted using GraphPad Prism software (Prism 5, La Jolla, CA). Quantitative variables were presented as percentage distributions or means and standard deviations. Student's t-test was used to determine statistical significance between two groups. The correlation between choline and other measurement parameters was examined using Pearson correlation analysis as the measurements normally distributed and multivariable multiple regression analysis and $p < 0.05$ was considered significant. The final statistical power was calculated to ensure sufficient sensitivity in detecting differences in choline levels, confirming the study's ability to detect meaningful differences of our findings.

8 Results

All volunteers were female in both groups. As the interim analysis results reached significance, the study was concluded with a total of 64 female participants, consisting of 38 newly diagnosed, drug-naïve individuals with osteoporosis and 26 controls. Rigorous exclusion criteria for primary osteoporosis (e.g., younger age with risk factors such as fractures after age 50, rheumatoid arthritis, chronic kidney disease, or specific lifestyle habits like smoking and alcohol consumption) reduced the patient pool available. (11, 12). The data were normally distributed and mean age of the volunteers was 55 ± 2 (46–72 years) in the control group and 59 ± 1 (41–72 years) in the osteoporosis group ($p > 0.05$). The mean body mass index (BMI) was 30.3 ± 1.1 (kg/m^2) in the control group and 29.3 ± 0.7 (kg/m^2) in the osteoporosis group, and there was no significant difference between the groups ($p > 0.05$). In the osteoporosis group, 15 participants had at least one chronic disease, with some having more than one condition. Specifically, 7 participants had diabetes mellitus, 10 had hypertension, 3 had asthma, and 1 had Crohn's disease, all of which were under control. In the control group, 4 participants had at least one chronic disease, including 2 with diabetes mellitus, 3 with hypertension, and 1 with asthma, also under control.

All volunteers in the control group were pain-free and did not possess any clinical features of osteoporosis like pain, abnormal blood test, pathological fracture history and chronic metabolic disorders. The mean BMD (T-score) was -2.9 ± 0.07 in the osteoporosis group and -0.4 ± 0.09 in the control group ($p < 0.001$). (Table 1).

9 Laboratory Analyses

Serum choline levels were significantly lower in osteoporosis group (3659 ± 93.0 µmol/mL and) compared to control group (33,175 ± 3820 pg/mL µmol/mL; $p < 0.0001$). Blood PTH levels were 63.4 ± 4.6 pg/mL, calcium levels 8.9 ± 0.2 mg/dL and 25-OH Vitamin D levels 34.9 ± 3.1 ng/mL in the osteoporosis group. Calcium levels were 9.8 ± 0.1 mg/dL and 25-OH Vitamin D levels 30.1 ± 2.0 ng/mL in the control group. Parathyroid hormone (PTH) levels are not routinely measured in individuals who do not exhibit any findings indicative of an osteoporosis diagnosis during regular controls.

There was a positive correlation between total choline/ACh levels and BMD scores in patients with osteoporosis ($R = 0.26$, $p = 0.057$). There was a low positive correlation between total choline/ACh levels and calcium levels ($R = 0.30$, $p = 0.038$) and 25-OH vitamin D ($R = 0.28$, $p = 0.046$) in patients with osteoporosis. There was a low

Table 1 Demographic characteristics of volunteers (osteoporosis, BMI: body mass index, PTH: parathyroid hormone)

	Control (n = 26)	Osteoporosis (n = 38)	p
Age (year, mean ± SD)	55.2 ± 1.9	59.7 ± 1.2	0.062
Age range	46–74	41–64	
Gender	Female (26)	Female (38)	
BMI (kg/m^2)	30.3 ± 1.1	29.3 ± 0.7	0.892
Calcium	9.8 ± 0.1	8.8 ± 0.2	< 0.001***
25-OH vitamin D	30.1 ± 2.0	34.3 ± 3.1	0.848
T score	− 0.4 ± 0.09	− 2.9 ± 0.07	< 0.001***

Student's t test and Mann Whitney U test used for comparison of two groups

*** $p < 0.001$ versus control

negative correlation between total choline/ACh levels and PTH (R = − 0.29, p = 0.041) in patients with osteoporosis (Figs. 1 and 2).

10 Final Statistical Power Calculation

The study anticipated a medium effect size (d = 0.50) in choline level differences between osteoporosis cases and controls, leading to an initial sample size calculation of 64 participants per group to achieve 80% power at a 95% confidence level. However, due to interim analyses we concluded the study with 38 participants in osteoporosis and 26 participants in controls. For the final sample size of 38 participants per group, we recalculated the power specifically for detecting differences in choline and leptin levels. The recalculated power for detecting a medium effect size (d = 0.50) was found to be 100% for choline levels, confirming that despite the reduced sample size. This indicates that, despite the reduction in sample size, the study retained sufficient power to detect statistically significant differences between the groups for these key variables [14, 15].

11 Discussion

Adrenergic stimulation results in bone loss, while cholinergic activity has been shown to promote bone formation. In vitro studies suggest that cholinergic activity stimulates the proliferation and differentiation of bone cells. Suppression of cholinergic activity in bone tissue and the central nervous system has been demonstrated to cause bone loss [1]. Although the function of nicotinic acetylcholine receptors (nAChR) in bone metabolism is not fully understood, in a collagen-induced arthritis mouse model where α7nAChR expression was transcriptionally suppressed (knockdown, KD), significant joint destruction was observed [16]. Activation of α7nAChR, a receptor coupled with calcium-permeable ion channels, increases intracellular calcium concentration [17]. The potential anabolic effect of enhancing cholinergic activity on bone formation has been investigated using acetylcholinesterase inhibitors (AChEI), drugs that inhibit the AChE enzyme, leading to increased acetylcholine levels and thereby enhancing cholinergic transmission through cholinergic receptors [8]. In vivo studies have shown that pyridostigmine, an AChEI, increases bone mass [18]. Similarly, AChEI used in the treatment of Alzheimer's disease has been associated with reduced hip fracture risk and improved healing of osteoporotic fractures in this patient group [12]. Cholinomimetic α7nAChR agonists have been shown to balance the OB/OC ratio by stimulating OB proliferation and differentiation [9]. The selective α7nAChR agonist, GTS-21, has been shown to prevent bone damage by reducing inflammatory cytokines in a collagen-induced in vivo arthritis model, suggesting it may be a promising agent in treatment [19]. These findings suggest that cholinergic agents might play a role in bone metabolism. Moreover, targeting nAChRs with agonists and positive allosteric modulators has emerged as a promising therapeutic strategy for managing neuroinflammatory and neuropathic pain conditions [2, 21].

Choline, as an essential nutrient and precursor to the neurotransmitter acetylcholine (ACh), contributes to acetylcholine synthesis and stimulates nicotinic acetylcholine receptors (nAChRs) directly, mediating analgesic effects in both acute and chronic pain models across animals and humans when administered in high doses [20–22]. Despite these insights, there is limited understanding of how blood choline/ACh levels change in patients with osteoporosis. In this study, serum choline/ACh levels were found to be significantly lower in the osteoporosis group compared to the control group. Supporting role of the cholinergic system's role in bone metabolism has been suggested through studies investigating acetylcholinesterase inhibitors (AChEIs), increasing ACh levels and thereby enhancing cholinergic transmission via cholinergic receptors [8]. For example, in vivo studies indicate that pyridostigmine, an AChEI, increases bone mass [18]. Similarly, AChEIs used in Alzheimer's disease treatment have been linked to reduced hip fracture risk and improved healing of osteoporotic fractures in affected patients [12]. α7nAChR agonists, such as GTS-21, have been demonstrated to restore balance in osteoblast (OB) and osteoclast (OC) activity by promoting OB proliferation and differentiation. Furthermore, GTS-21 has been shown to prevent bone damage by reducing inflammatory cytokine levels in collagen-induced arthritis models, highlighting its potential as a therapeutic agent for bone-related conditions [19]. These findings collectively suggest that cholinergic agents may play a significant role in bone metabolism, offering new avenues for understanding and treating osteoporosis. This study suggests that serum choline/ACh levels significantly low in patients with osteoporosis emphasizing that choline metabolism might play a role in bone metabolism in osteoporosis.

Bone mineral density (BMD) is a critical parameter for assessing bone health, reflecting the amount of minerals per unit of bone volume [23]. Various factors influence BMD, including genetic predisposition, lifestyle choices, and different medical conditions. Additionally, Low plasma choline was associated with low BMD in both sexes and increased the risk of hip fracture in elderly women [24]. Our findings suggest that choline levels are positively correlated with BMD in patients with osteoporosis and suggest that choline deficiency may have a role in the pathogenesis of osteoporosis.

Parathyroid hormone (PTH) levels are crucial in various medical conditions, particularly those related to calcium metabolism. Elevated PTH levels are commonly associated with primary hyperparathyroidism, characterized by hypercalcemia [25, 26]. The relationship between calcium and PTH levels is intricate, with PTH secretion being triggered when serum calcium levels are low. Furthermore, the dynamics of PTH secretion are influenced by factors like phosphorus and vitamin D, which modulate both basal PTH levels and the response to hypocalcemia [27, 28]. Our findings indicate that choline levels are negatively correlated with PTH and positively correlated with calcium and vitamin D in patients with osteoporosis. This suggests that choline deficiency may play a role in the pathogenesis of osteoporosis.

12 Limitations

This study has several limitations that should be considered when interpreting the results. First, PTH concentrations for the control group were not included in the analyses, as PTH levels are not routinely measured in healthy individuals without clinical indications. Consequently, the analysis of PTH and its relationship with choline levels was performed exclusively for the osteoporosis group, which may limit the ability to generalize these findings. This limitation has been explicitly noted. Additionally, the study focused solely on female participants aged 40–70 years, limiting applicability to males or younger individuals. Future research should include diverse populations to enhance the understanding of choline's role in bone metabolism across different demographics. Despite these limitations, the study provides valuable insights into the association between choline levels and osteoporosis, paving the way for further research.

13 Conclusion

This study observed significantly lower serum choline and acetylcholine levels in patients with osteoporosis compared to healthy controls. Correlations between choline levels and bone mineral density, calcium, vitamin D, and PTH were identified, suggesting a potential association between choline metabolism and bone health. These findings provide initial evidence supporting the role of choline as a potential biomarker for bone-related conditions. Further research is needed to investigate the mechanistic role of choline in osteoporosis and explore its potential clinical and therapeutic implications.

Author Contribution E.B. nd V.O. wrote the main manuscript text and E.B. prepared figures and tables. V.O. collected the participants and E.B. conducted the biochemical analyses. All authors reviewed the manuscript.

Funding Open access funding provided by the Scientific and Technological Research Council of Türkiye (TÜBİTAK).

Data Availability No datasets were generated or analysed during the current study.

Declarations

Conflict of Interest Authors Volga Ozturk and Elif Baris declare that they have no conflict of interest.

Grant support NA.

References

1. Eimar H, Tamimi I, Murshed M, Tamimi F. Cholinergic regulation of bone. J Musculoskelet Neuronal Interact. 2013;13:124–32.
2. Colović MB, Krstić DZ, Lazarević-Pašti TD, et al. Acetylcholinesterase inhibitors: pharmacology and toxicology. Curr Neuropharmacol. 2013;11:315–35. https://doi.org/10.2174/1570159X11311030006.
3. Ma F, Luo X, Ma J, et al. The effect of the α7nAChR agonist on Wnt/β-catenin signaling in osteoporosis. Int J Clin Exp Pathol. 2019;12:2867–74.
4. Luo X, Lauwers M, Layer PG, Wen C. Non-neuronal role of acetylcholinesterase in bone development and degeneration. Front cell Dev Biol. 2020;8: 620543. https://doi.org/10.3389/fcell.2020.620543.
5. Lauwers M, Courties A, Sellam J, Wen C. The cholinergic system in joint health and osteoarthritis: a narrative-review. Osteoarthr Cartil. 2021;29:643–53. https://doi.org/10.1016/j.joca.2021.02.005.
6. Vernejoul MC De (2013) More on bone and nerves. IBMS Bonekey 10. https://doi.org/10.1038/bonekey.2013.47
7. López-Coviella I, Berse B, Krauss R et al. Induction and maintenance of the neuronal cholinergic phenotype in the central nervous system by BMP-9. Science 2000;289:313–16. https://doi.org/10.1126/science.289.5477.313
8. Crivello NA, Blusztajn JK, Joseph JA, et al. Short-term nutritional folate deficiency in rats has a greater effect on choline and acetylcholine metabolism in the peripheral nervous system than in the brain, and this effect escalates with age. Nutr Res. 2010;30:722–30. https://doi.org/10.1016/j.nutres.2010.09.008.

9. Alkondon M, Pereira EFR, Cartes WS. Choline is a selective agonist of α7 nicotinic acetylcholine receptors in the rat brain neurons. Eur J2006;9:2734–42
10. Ogunwale AN, Colon-Emeric CS, Sloane R et al. Acetylcholinesterase inhibitors are associated with reduced fracture risk among older veterans with dementia. J Bone Miner Res. 2020;35:440–5. https://doi.org/10.1002/jbmr.3916
11. Cohen J. Statistical power ANALYSIS for the behavioral sciences; 1988
12. Portney L, Watkins MP. Foundation of clinical research. Application to Practice; 2009
13. Baris E, Simsek O, Efe H, et al. Effects of CDP-choline and choline on COX pathway in LPS-induced inflammatory response in rats. Int J Pharmacol. 2021;17:84–96. https://doi.org/10.3923/ijp.2021.84.96.
14. Sullivan KM, Dean A, Soe MM. OpenEpi: a web-based epidemiologic and statistical calculator for public health. Public Health Rep. 2009;124:471–4. https://doi.org/10.1177/003335490912400320.
15. Keffer J, Probert L, Cazlaris H, et al. Transgenic mice expressing human tumour necrosis factor: a predictive genetic model of arthritis. EMBO J. 1991;10:4025–31.
16. De Jonge WJ, Ulloa L. The alpha7 nicotinic acetylcholine receptor as a pharmacological target for inflammation. Br J Pharmacol. 2007;151:915–29. https://doi.org/10.1038/sj.bjp.0707264.
17. Bajayo A, Bar A, Denes A, et al. Skeletal parasympathetic innervation communicates central IL-1 signals regulating bone mass accrual. Proc Natl Acad Sci USA. 2012;109:15455–60. https://doi.org/10.1073/pnas.1206061109.
18. Hu Y, Liu R, Li J, et al. Attenuation of collagen-induced arthritis in rat by nicotinic alpha7 receptor partial agonist GTS-21. Biomed Res Int. 2014;2014: 325875. https://doi.org/10.1155/2014/325875.
19. Bartolini A, Ghelardini C, Fantetti L, et al. Role of muscarinic receptor subtypes in central antinociception. Br J Pharmacol. 1992;105:77–82. https://doi.org/10.1111/j.1476-5381.1992.tb14213.x.
20. Hamurtekin E, Gurun M. The antinociceptive effects of centrally administered CDP-choline on acute; 2006
21. Bagdas D, Sonat FA, Hamurtekin E, et al. The antihyperalgesic effect of cytidine-5′-diphosphate-choline in neuropathic and inflammatory pain models. Behav Pharmacol. 2011;22:589–98. https://doi.org/10.1097/FBP.0b013e32834a1efb.
22. Hone AJ, McIntosh JM. Nicotinic acetylcholine receptors: therapeutic targets for novel ligands to treat pain and inflammation. Pharmacol Res. 2023;190: 106715. https://doi.org/10.1016/j.phrs.2023.106715.
23. Mito K, Sato Y, Kobayashi T, et al. The nicotinic acetylcholine receptor α7 subunit is an essential negative regulator of bone mass. Sci Rep. 2017;7:1–12. https://doi.org/10.1038/srep45597.
24. Lü J, Hu L, Guo L, et al. The effects of claw health and bone mineral density on lameness in duroc boars. Animals. 2023;13:1502. https://doi.org/10.3390/ani13091502.
25. Øyen J, Nygård O, Gjesdal CG, et al. Plasma choline, nicotine exposure, and risk of low bone mineral density and hip fracture: the Hordaland health study. J Bone Miner Res. 2013;29:242–50. https://doi.org/10.1002/jbmr.2025.
26. Vybhavi MK, Prashanth V, Muddaiah DVS. A case of right parathyroid adenoma and its management. Int J Otorhinolaryngol Head Neck Surg. 2023;9:677–81. https://doi.org/10.18203/issn.2454-5929.ijohns20232229
27. Prihantono P, Palinggi E, Haryasena H, et al. A case report surgical treatment for parathyroid adenoma: a case report. Open Access Maced J Med Sci. 2019;7:2497–501. https://doi.org/10.3889/oamjms.2019.418.
28. Felsenfeld AJ, Rodri[Combining Acute Accent]guez M, Aguilera-Tejero EAA. Dynamics of parathyroid hormone secretion in health and secondary hyperparathyroidism. Clin J Am Soc Nephrol. 2007;2