RESEARCH ARTICLE

Editorial Process: Submission:03/23/2025 Acceptance:08/15/2025 Published:08/22/2025

Phosphoprotein Profile of Ameloblastoma

Sirima Sanguansin[1], Sudaporn Kengkarn[2], Boworn Klongnoi[3], Supachai Topanurak[4], Sittirak Roytrakul[5], Suthipong Chujan[6,7], Nakarin Kitkumthorn[1]*

Abstract

Objective: Ameloblastoma (AM) is a well-known benign odontogenic tumor recognized for its aggressive nature, believed to originate from tooth-forming tissue or the dental follicle (DF). Phosphoproteins are crucial for cellular signaling, enabling intracellular communication and regulating various physiological processes. In cancer, phosphoproteins are fundamental to both pathogenesis and pathophysiology. However, studies on phosphoproteins in AM are still limited. This study aimed to compare phosphoprotein profile and identify the crucial phosphoproteins between AM and DF. **Methods:** The phosphoprotein profiles of seven AM and five DF were discovered using mass spectrometry, and their associated phosphosites were examined by Netphos 3.1. Biological functions were analyzed by Metascape database. **Results:** Thirteen significant phosphoproteins were found in AM, and six in DF, all of which have phosphorylation sites. For example, among the proteins uniquely identified in AM were SENP1 (Sentrin-specific protease 1), DDX42 (ATP-dependent RNA helicase DDX42), LMBR1L (Protein LMBR1L), Cathepsin H (CATH), and Retinoblastoma-binding protein 5 (RBBP5), whereas those unique to DF included GC-rich sequence DNA-binding factor (GCF), Plexin-C1 (PLXC1), and proline/serine-rich coiled-coil protein 1 (PSRC1), PTHD3 (Patched domain-containing protein 3), and TPC6B (Trafficking protein particle complex subunit 6B). For biological analysis, the enriched terms included processing of capped intron-containing pre-mRNA, signaling by rho GTPases, establishment of organelle localization, signaling by receptor tyrosine kinases and cell morphogenesis. **Conclusion:** These phosphoproteomic findings provide essential insights into the pathogenesis of AM and warrant further investigation. This is crucial for advancing our understanding of AM biology and identifying potential therapeutic targets.

Keywords: Phosphoprotein- Ameloblastoma- Dental follicle- LCMS/MS

Asian Pac J Cancer Prev, **26 (8)**, 3085-3091

Introduction

Ameloblastoma (AM), the second most common odontogenic tumor of the jaw, originated from tooth-originating tissue or the dental follicle (DF). Most AM occurs in mandible and locally invasive, slow-growing tumor causes painless jaw enlargement. The optimum treatment for AM is vigorous en bloc resection and concomitant repair. The high recurrence rate and significant tissue abnormalities have been longstanding concerns in treating AM [1, 2]. Recent molecular findings strongly indicate that targeted therapies, particularly BRAF V600E inhibitors, have the potential to improve clinical outcomes in AM [3]. A comprehensive investigation of AM using modern molecular approaches has yielded discoveries in genomes, transcriptomics, and proteomics [4-6].

Phosphoproteins, which are proteins containing phosphorylated residues, are crucial in the regulation of the cell cycle, gene expression, and signal transduction. Dysregulated phosphorylation is linked to the pathogenesis of several diseases, including cancer, neurological disorders, and metabolic syndromes. [7, 8]. In AM, phosphoproteins involved in the MAPK pathway, particularly ERK1/2, play a critical role in tumor development. Mutations in upstream regulators like BRAF (especially V600E) lead to persistent MAPK activation, promoting tumor growth and invasiveness.

*[1]Department of Oral Biology, Faculty of Dentistry, Mahidol University, Bangkok, Yothi Road, Rajthewi District, Bangkok 10400, Thailand. [2]Department of Hematology, Faculty of Medical Technology, Rangsit University, 52/347 Muang-Ake, Phaholyothin Road, Lak-Hok, Muang Pathumthani, Pathumthani, 12000, Thailand. [3]Department of Oral and Maxillofacial Surgery, Faculty of Dentistry, Mahidol University, Bangkok, Yothi Road, Rajthewi District, Bangkok 10400, Thailand. [4]Department of Molecular Tropical Medicine and Genetics, Faculty of Tropical Medicine, Mahidol University, Bangkok, 10400, Thailand. [5]Functional Proteomics Technology Laboratory, National Center for Genetic Engineering and Biotechnology, National Science and Technology Development Agency, 113 Thailand Science Park, Phahonyotin Road, Klong 1, Khlong Luang, 12120, Pathum Thani, Thailand. [6]Laboratory of Pharmacology, Chulabhorn Research Institute, Bangkok, 10210, Thailand. [7]Center of Excellence on Environmental Health and Toxicology (EHT), Office of the Permanent Secretary (OPS), Ministry of Higher Education, Science, Research and Innovation (MHESI), Bangkok, 10400, Thailand. *For Correspondence: Nakarinkit@gmail.com*

These phosphorylated proteins serve as both markers of pathway activation and potential therapeutic targets, underscoring the value of phosphoproteomic profiling in elucidating AM pathogenesis [9-11]. Understanding AM phosphoproteins will offer novel therapeutic approaches. Both kinase inhibitors and phosphatase activators can target abnormal phosphorylation events. Due to the limited research on phosphoproteins, the differences in phosphoproteomic profiles between AM and DF remain unclear.

Recent advances in mass spectrometry have revolutionized phosphoproteomics by enabling the simultaneous identification of hundreds of phosphorylation events [8, 12]. The aim of this study was to use mass spectrometry to elucidate the phosphoprotein profiles in AM compared to DF. Understanding the differential phosphoprotein expression between these lesions may uncover key signaling pathways involved in AM pathogenesis, particularly those driving its locally aggressive behavior. These insights hold promise for the identification of novel diagnostic biomarkers and therapeutic targets, particularly in the development of personalized treatment strategies for AM. Ultimately, the findings from this study may enhance the classification and clinical management of the disease. However, they also underscore the need for further research in this area.

Materials and Methods

Sample recruitment

This investigation used twelve tissue samples as shown in Table 1. Seven AM tissues were taken from patients who had a mandibulectomy for AM. Five DF tissues were derived from patients who had their wisdom teeth removed. All tissue samples were collected by the Department of Oral and Maxillofacial Surgery, Faculty of Dentistry, Mahidol University, Bangkok, Thailand. The study included only AM cases with at least 80% tumor content. One half of each specimen was sent for histopathology confirmation. The present study was approved by the Institutional Review Board of the Faculty of Dentistry/Faculty of Pharmacology, Mahidol University, Bangkok, Thailand (approval no. COA. NO.MU DT/PY IRB 2021/034.3003).

Protein preparation and liquid chromatography-tandem mass spectrometry (LC/MS-MS)

The frozen tissue (0.5×0.5×0.5 cm^3) was collected at -80°C and stored in a 1.5-ml tube. It was later ground in liquid nitrogen using an Axygen™ Tissue Grinder (Thermo Fisher Scientific) and solubilized with liquid nitrogen and detergent lysis buffer [50 mM Tris-HCl (pH 7.2), 1% SDS, and 20 mM DTT], followed by mixing at room temperature for 10 min. After two rounds of sonication (5 sec each, 80% amplitude), the lysed tissue was heated at 72°C for 3 min and centrifuged at 12,000 × g for 30 min. The protein solutions were stored at -20°C until analysis.

Each sample's protein concentration was determined and enriched phosphoprotein by the phosphoprotein enrichment process (Phosphoprotein Enrichment Kit, Pierce, IL, USA). The enriched phosphoprotein was then concentrated using a 9-kDa cut-off membrane column (Phosphoprotein Enrichment Kit, Pierce, IL, USA) and subsequently desalted using gel filtration (Thermo Scientific, Rockford, IL, USA). Then, the samples were prepared for tryptic peptide by being mixed with 50 ng/µl of sequencing-grade trypsin (1:20 ratio), and incubated overnight at 37°C. Dry and protonate digested samples with 0.1 % formic acid before putting them into an Ultimate3000 Nano/Capillary Liquid Chromatography System (Thermo Scientific, UK) with a hybrid quadrupole Q-Tof Impact IITM (Bruker Daltonics; Bruker Corporation) and nano-captive spray ion source. Ionization was done with CaptiveSpray at 1.6 kV. Compass 1.9 (Bruker Daltonics; Bruker Corporation) was used to acquire positive-ion mass spectra (MS) and MS/MS spectra from 150–2,200 m/z. A triplicate LC-MS analysis was performed on each sample.

Protein interpretation and bioinformatic analysis

Individual sample proteins were measured using MaxQuant 1.6.6.0. [13]. Using the Andromeda search engine, MS/MS spectra were correlated to the Uniprot Homo sapiens database (https://www.uniprot.org) [14]. Protein identification requires seven amino acids and one unique peptide per peptide. Protein FDR was evaluated using reversed search sequences at 1%. Perseus version 1.6.6.0 was utilized to import the ProteinGroups.txt file obtained from MaxQuant [15]. The max intensities were log2 transformed, and t-tests were conducted to compare conditions. Venn diagrams were employed to depict the variations between protein lists obtained from different differential analyses. The Seaborn Python heatmap program visualizes protein levels [16]. The phosphosites of selected proteins were proved by NetPhos - 3.1 (https://services.healthtech.dtu.dk/services/NetPhos-3.1/) [17].

Biological alterations analysis by using MCODE modular clustering

To investigate biological functions of significant proteins, Metascape database (http://metascape.org) was used to perform the enrichment of biological alterations analysis [18]. Metascape is a freely available online analysis tool that applies bioinformatics methods in batch gene or protein to get a more detailed understanding of biological alterations.

Results

The Venn diagram was performed with all identified proteins, of which 26 and 13 were exclusively identified in AM and DF, respectively (Figure 1A). Among the 39 phosphoproteins, some were found in low frequency, prompting us to focus on proteins present in more than 70% of cases. Ultimately, there were 13 phosphoproteins in AM and 6 in DF. The heatmap data is shown in Figure 1B, while the protein names and detailed data are listed in Tables 2, Supplement Table1, and 2.

In AM, the phosphoproteins that were highlighted were sentrin-specific protease 1 (SENP1), ATP-dependent RNA helicase DDX42 (DDX42), cathepsin H (CATH),

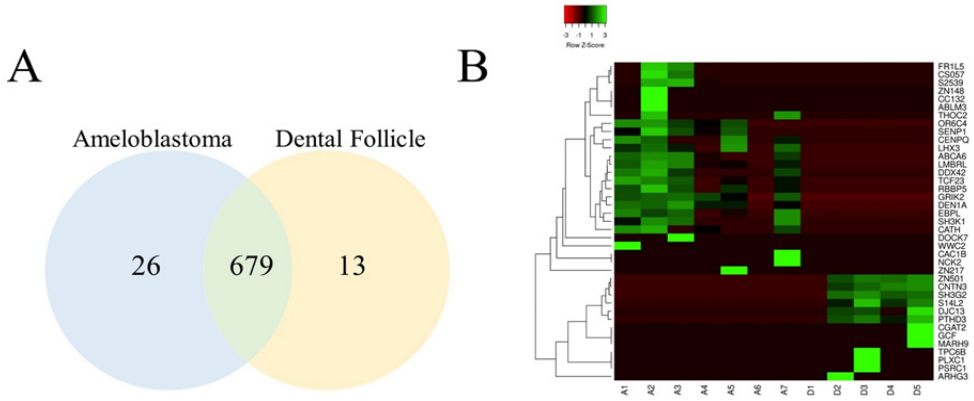

Figure 1. Differential Distribution Phosphoprotein between AM and DF. (A) Venn diagram demonstrated 13 phosphoproteins that are unique to ameloblastoma and 6 phosphoproteins that are specific to dental follicle. (B) Heatmap of unique proteins between. A, ameloblastoma; D, dental follicle.

Table 1. Detailed data of ameloblastoma and dental follicle

Type of tissue	Dental follicle	Ameloblastoma
No.	5	7
Sex (M,F)	2.3	3.4
Age (y, median (range)	21 (17.25)	42 (13.66)
Location	38=2, 48=3	Q3=4, Q4=3
Histological subtype	N/A	Follicular =4, Plexiform=3

N/A, not applicable; Q3, left posterior mandible; Q4, right posterior mandible

protein LMBR1L (LMBRL), retinoblastoma-binding protein 5 (RBBP5), Transcription factor 23 (TCF23), ATP-binding cassette sub-family A member 6 (ABCA6), DENN domain-containing protein 1A (DEN1A), Olfactory receptor 6C4 (OR6C4), uncharacterized protein C19orf57 (CS057), SH3 domain-containing kinase-binding protein 1 (SH3K1), solute carrier family 25 member 39 (S2539), and glutamate receptor, ionotropic kainate 2 (GRIK2). Conversely, in DF, the unique phosphoproteins were plexin-C1 (PLXC1), GC-rich sequence DNA-binding factor (GCF), patched domain-containing protein 3 (PTHD3), trafficking protein particle complex subunit 6B (TPC6B), chondroitin sulfate N-acetylgalactosaminyltransferase 2 (CGAT2), and proline/serine-rich coiled-coil protein 1 (PSRC1).

For biological alteration analysis, the enriched terms included processing of capped intron-containing pre-mRNA, signaling by rho GTPases, establishment of organelle localization, signaling by receptor tyrosine kinases and cell morphogenesis as shown in Figure 2.

Next, the phosphopeptide was confirmed using the Netphos program. Table 2 shows that all unique phosphopeptides had phosphosites at the amino acids' serine, threonine, and tyrosine. demonstrates the predicted phosphorylation enzymes and their scores at each phosphosite. Various enzymes were involved in phosphorylation, including - DNA-activated protein

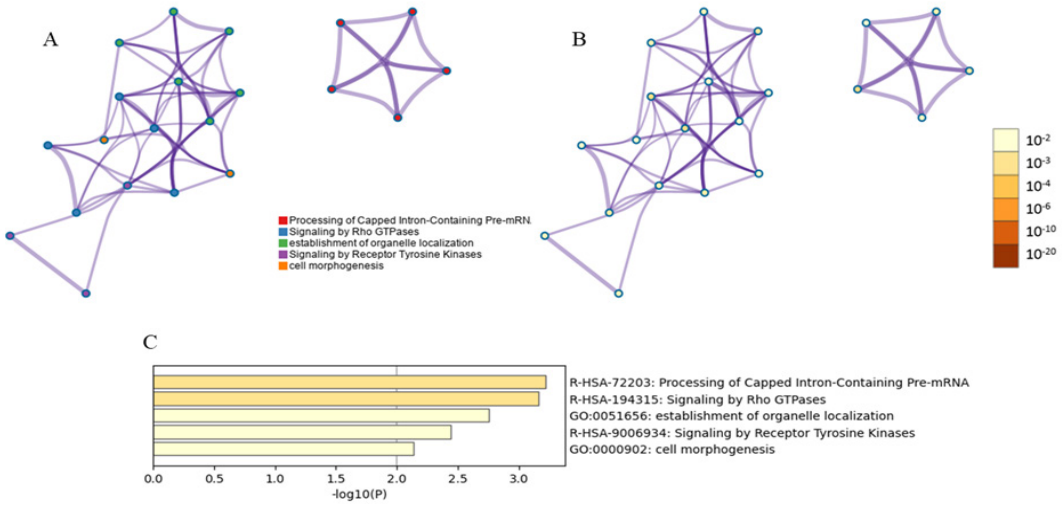

Figure 2. The Enrichment Analysis of Biological Alterations by Using Metascape. (A) The network of enriched terms (coloring by cluster ID). (B) The network of enriched terms (coloring by P-value). (C) A bar graph of enriched terms of the significant proteins

Table 2. Unique Phosphoprotein and Phosphosites in AM and DF

	Protein symbol	AC	Protein name	Peptide	Phosphosite
Ameloblastoma	SENP1	Q9P0U3	Sentrin-specific protease 1	NSTPSSSSSLQK	101S, 102T, 104S, 105S, 106S, 107S, 108S
	DDX42	Q86XP3	ATP-dependent RNA helicase DDX42	QTLLFSATFRK	440T
	CATH	P09668	Cathepsin H	KINAHNNGNHTFK	74T
	LMBRL	Q6UX01	Protein LMBR1L	QVLALQTQR	279T
	RBBP5	Q15291	Retinoblastoma-binding protein 5	TDSQDLVASFR	182S, 188S
	TCF23	Q7RTU1	Transcription factor 23	ERSRVRTLR	87S, 91T
	ABCA6	Q8N139	ATP-binding cassette sub-family A member 6	MNMKQKSVYQQTK	7S, 9Y
	DEN1A	Q8TEH3	DENN domain-containing protein 1A	DSILNPSDK	679S, 684S
	OR6C4	Q8NGE1	Olfactory receptor 6C4	FLTSMTTGNK	81T
	CS057	Q0VDD7	Uncharacterized protein C19orf57	KKLRTSGEGLCPPKPLK	10S
	SH3K1	Q96B97	SH3 domain-containing kinase-binding protein 1	KEMKKDPLTNK	66T
	S2539	Q9BZJ4	Solute carrier family 25 member 39	LQSQRPSMASELMPSSR	41S, 45S
	GRIK2	Q13002	Glutamate receptor, ionotropic kainate 2	TLLPNTTLTYDTQK	69T, 77T, 80T
Dental follicle	PLXC1	O60486	Plexin-C1	SQPNRTCTCSIPTR	550S, 562T
	GCF	P16383	GC-rich sequence DNA-binding factor	AADSSDSDGAEESPAEPGAPR	17S, 19S, 25S
	PTHD3	Q3KNS1	Patched domain-containing protein 3	MSNVYSK	480S,483Y, 484S
	TPC6B	Q86SZ2	Trafficking protein particle complex subunit 6B	SAEQGEVENGRCITK	21S, 34T
	CGAT2	Q8N6G5	Chondroitin sulfate N-acetylgalactosaminyltransferase 2	NVGANGIGYQSNK	109Y, 111S
	PSRC1	Q6PGN9	Proline/serine-rich coiled-coil protein 1	AAKLRVSGSGEFVGLTLK	223S

Denoted, AC, Accession number in UniProtKB entry

kinase (DNAPK), protein kinase C (PKC), protein kinase G (PKG), protein kinase A (PKA), cyclin-dependent kinase inhibitor (CKI), casein kinase II (CKII), cyclin-dependent kinase 1 (CDK1) cyclin-dependent kinase 5 (cdk5), glycogen synthase kinase 3 (GSK3), ribosomal S6 kinase (RSK), Ataxia-telangiectasia mutated (ATM) and insulin receptor (INSR)

Discussion

To our knowledge, although this is a preliminary study, it is the first study of phosphoproteomics in AM. This profile will reveal key insights into the molecular landscape of cancer. In this study, we observed 13 unique phosphoproteins in AM and 6 in DF. Unfortunately, all of these proteins have not been previously reported in AM and odontogenesis. Other than that, some AM-specific phosphoproteins have been associated to faster tumor cell growth. For example, SENP1 is involved in the SUMOylation pathway. It acts as a cysteine protease involved in the deSUMOylation of target proteins, which affects various cellular processes, including transcription, proliferation, apoptosis, angiogenesis, invasion, metastasis, DNA repair, and cell cycle progression [19, 20].

DDX42, a DEAD-box RNA helicase, is involved in RNA splicing, translation initiation, and ribosome assembly. While its direct role in cancer is unclear, its interaction with SF3B1 suggests potential involvement in cancer-related RNA splicing alterations. SF3B1 mutations are linked to aberrant splicing in cancer, and DDX42 may influence these processes. Additionally, other DEAD-box helicases contribute to cancer progression by affecting proliferation, migration, and apoptosis [21, 22].

DEN1A, is involved in the deneddylation process, regulating the activity of cullin-RING ligases. DENND1A has been implicated in cancer through its role as a guanine nucleotide exchange factor (GEF) for the small GTPase RAB35, which is involved in endocytic trafficking and receptor turnover. DEN1A activates RAB35, which has been shown to promote cancer cell migration in gastric cancer, indicating its involvement in tumor progression [23]. RBBP5 is part of the MLL/SET methyltransferase complex, influencing histone modification and gene expression. Dysregulation of epigenetic modifiers like RBBP5 can lead to aberrant gene expression profiles in cancers. its role in chromatin remodeling suggests potential involvement in tumor biology [24, 25]. Given the lack of direct information connecting these finding phosphoproteins with AM, further research or related research may offer potential connections.

AM pathogenesis is a complicated process and various signaling have been identified in each stage of the ameloblast life cycle and regulate the differentiation processes [26]. Rho GTPase has been identified as a regulatory mechanism of cytokinesis, migration, polarization, cell adhesion, and cell cycle [27]. Rho GTPases including RhoA, Rac1, and Cdc42 play an important role in many cellular events such as transcription regulation, membrane trafficking, cell proliferation, embryonic development, and reactive oxygen species production [28]. According to our analyses, we also found the enrichment of rho GTPases, establishment of organelle localization and cell morphogenesis. Previous study has demonstrated a strong correlation between RhoA and its downstream effector ROCK with ameloblast differentiation. Additionally, RhoA is present in the enamel organ during the initiation and morphogenesis of the tooth germ [29]. A prior study using immunohistochemistry found that RhoA and RhoB were present in a large number of cells and exhibited greater intensity in nonpolarized cells across follicular, plexiform, and unicystic AMs [30]. Overall, these findings provide evidence of the significant role of Rho GTPase in cytoskeletal rearrangement, cell-cell adhesion, cell proliferation, and the regulation of gene transcription in AM.

Our study identified potential phosphosites on a discovered peptide (Table 2). Such phosphosites can modulate oncogene or tumor suppressor activity. In cancer, aberrant phosphorylation resulting from dysregulated kinase signaling promotes proliferation, resistance to apoptosis, angiogenesis, and metastasis [31]. Phosphoproteomic analyses have identified tumor-specific phosphorylation sites, uncovering key signaling networks involved in cancer [32]. These sites may serve as both biomarkers and therapeutic targets, as exemplified by the clinical success of kinase inhibitors [33]. In summary, the characterization of cancer-associated phosphosites enhances our mechanistic understanding of tumorigenesis and advances the field of precision oncology [34].

A limitation of this study is the absence of key kinases, such as those in the MAPK pathway. These signaling proteins are typically expressed at much lower levels than abundant proteins like cytoskeletal components or metabolic enzymes [35, 36]. During mass spectrometry, high-abundance proteins are preferentially ionized and detected, often masking low-abundance phosphoproteins. Additionally, phosphopeptide enrichment is technically challenging; phosphorylated peptides have lower ionization efficiency under standard positive-ion MS conditions, making detection difficult [37, 38]. These challenges are further compounded when working with large clinical sample sets, where high variability and complexity persist despite limited protein quantities. As a result, some key phosphoproteins may be underrepresented or missed entirely, and our findings reflect only the subset of proteins that could be enriched and identified within these limitations.

Another limitation lies in its limited number and exclusive focus on mandibular AM. Furthermore, in cancer biology, the investigation of phosphoproteins is complicated by the intricate signaling networks, the temporal dynamics of phosphoproteins, and post-translational modifications extending beyond phosphorylation. Despite these limitations, continuous research and technological progress strive to overcome these challenges and improve the usefulness of phosphoproteins in cancer research and clinical applications. Current treatments include kinase inhibitors, monoclonal antibodies, and combination therapy to disrupt abnormal phosphorylation and inhibit cancer growth [7, 8]. In summary, this study presents a significant phosphoprotein profile of AM,

driving additional research into these molecules to better diagnostics and targeted treatments to AM.

Author Contribution Statement

SS and NK conceived the study, designed the experiments, confirm the authenticity of all the raw data, analyzed the data and wrote the manuscript. SS, SK, SR, ST, and SC performed experiments and interpreted data. NK and BK recruited the patients and collected the clinical samples. All authors have read and approved the final version of the manuscript.

Acknowledgements

The authors would like to express their gratitude to the Functional Proteomics Technology Laboratory, National Center for Genetic Engineering and Biotechnology (Pathumthani, Thailand) for providing the facilities and laboratory equipment.

Funding Statement

The present study was supported by Mahidol University, Faculty of Dentistry Grant (grant no. DTRS-EG-2021-04).

Ethical Declaration

The present study was approved by the Institutional Review Board of the Faculty of Dentistry/Faculty of Pharmacology, Mahidol University, Bangkok, Thailand (approval no. COA. NO.MU DT/PY IRB 2021/034.3003) and was conducted in accordance with the Declaration of Helsinki (1975), as revised in 2013. Written informed consent was obtained from all patients.

Data Availability

The data that support the findings of this study are available from the corresponding author upon reasonable request.

Conflict of Interest

The authors declare that there was no conflict of interest.

References

1. WHO. Classification of Tumours Editorial Board. Head and neck tumours. Lyon: International Agency for Research on Cancer; 2022.
2. Ghai S. Ameloblastoma: an updated narrative review of an enigmatic tumor. Cureus. 2022;14(8):e28236. https://doi.org/10.7759/cureus.28236.
3. Malakar A, Kumar VR, Yadav P, Bhardwaj V, Barua CG, Bhardwaj G. The role of BRAF inhibitors in the management of ameloblastoma: a literature review. Cureus. 2023;15(10):e47682. https://doi.org/10.7759/cureus.47682.
4. Duarte-Andrade FF, Vitório JG, Pereira T, Gomes CC, Gomez RS. A review of the molecular profile of benign and malignant odontogenic lesions. Oral Surg Oral Med Oral Pathol Oral Radiol. 2020;129(4):357-68. https://doi.org/10.1016/j.oooo.2019.12.017
5. Pongpanich M, Sanguansin S, Kengkarn S, Chaiwongkot A, Klongnoi B, Kitkumthorn N. An integrative analysis of genome-wide methylation and expression in ameloblastoma: a pilot study. Oral Dis. 2021;27(6):1455-67. https://doi.org/10.1111/odi.13666.
6. Sanguansin S, Kengkarn S, Klongnoi B, Chujan S, Roytrakul S, Kitkumthorn N. Exploring protein profiles and hub genes in ameloblastoma. Biomed Rep. 2024;20(1):64. https://doi.org/10.3892/br.2024.1752.
7. Ardito F, Giuliani M, Perrone D, Troiano G, Lo Muzio L. The crucial role of protein phosphorylation in cell signaling and its use as targeted therapy. Int J Mol Med. 2017;40(2):271-80. https://doi.org/10.3892/ijmm.2017.3036.
8. Higgins L, Gerdes H, Cutillas PR. Principles of phosphoproteomics and applications in cancer research. Biochem J. 2023;480(6):403-20. https://doi.org/10.1042/bcj20220220
9. Kurppa KJ, Catón J, Morgan PR, Ristimäki A, Ruhin B, Kellokoski J, et al. High frequency of braf v600e mutations in ameloblastoma. J Pathol. 2014;232(5):492-8. https://doi.org/10.1002/path.4317.
10. Brown NA, Rolland D, McHugh JB, Weigelin HC, Zhao L, Lim MS, et al. Activating FGFR2–RAS–BRAF mutations in ameloblastoma. Clin Cancer Res. 2014;20(21):5517–26. https://doi.org/10.1158/1078-0432.CCR-14-1069.
11. Tun KM, Lapthanasupkul P, Iamaroon A, Thosaporn W, Klanrit P, Kintarak S, et al. A multi-center cross-sectional investigation of BRAF V600E mutation in Ameloblastoma. PeerJ. 2025;13:e19137. https://doi.org/10.7717/peerj.19137
12. Wu Z, Tiambeng TN, Cai W, Chen B, Lin Z, Gregorich ZR, et al. Impact of phosphorylation on the mass spectrometry quantification of intact phosphoproteins. Anal Chem. 2018;90(8):4935-9. https://doi.org/10.1021/acs.analchem.7b05246.
13. Cox J, Mann M. MaxQuant enables high peptide identification rates, individualized ppb-range mass accuracies and proteome-wide protein quantification. Nat Biotechnol. 2008;26(12):1367-72.
14. UniProt Consortium. UniProt: the universal protein knowledgebase. Nucleic Acids Res. 2021;49(D1):D480-D489. https://doi.org/10.1093/nar/gkaa1100.
15. Tyanova S, Temu T, Sinitcyn P, Carlson A, Hein MY, Geiger T, et al. The Perseus computational platform for comprehensive analysis of (prote) omics data. Nature methods. 2016 Sep;13(9):731-40
16. Waskom M. Seaborn: Statistical data visualization. J Open Source Softw. 2021;6:3021. https://doi.org/10.21105/joss.03021
17. Blom N, Sicheritz-Pontén T, Gupta R, et al. NetPhos 3.1: prediction of phosphorylation sites in eukaryotic proteins. Bioinformatics. 2004;20(17):3179-3180. https://doi:10.1093/bioinformatics/bth403.
18. Zhou Y, Zhou B, Pache L, et al. Metascape: understanding the molecular mechanisms of complex diseases through integrated pathway enrichment analysis. Nucleic Acids Res. 2019;47(W1):W566-W574.
19. Lin M, Zhang M, Yi B, Chen J, Wen S, Chen R, Chen T, Li Z. Emerging role of SENP1 in tumorigenesis and cancer therapy. Front. Pharmacol. 2024;15:1354323.
20. Sheng Z, Luo S, Huang L, Deng YN, Zhang N, Luo Y, et al. SENP1-mediated deSUMOylation of YBX1 promotes colorectal cancer development through the SENP1-YBX1-AKT signaling axis. Oncogene. 2025;23:1-4.
21. Zhao B, Li Z, Qian R, Liu G, Fan M, Liang Z, et al. Cancer-associated mutations in sf3b1 disrupt the interaction between sf3b1 and ddx42. J Biochem. 2022;172(2):117-26. https://doi.org/10.1093/jb/mvac049

22. Yang F, Bian T, Zhan X, Chen Z, Xing Z, Larsen NA, et al. Mechanisms of the rna helicases ddx42 and ddx46 in human u2 snrnp assembly. Nat Commun. 2023;14(1):897. https://doi.org/10.1038/s41467-023-36489-x
23. Zhang L, Lin L, Zhang J, et al. Epidermal growth factor stimulates DENND1A-mediated Rab35 activation, promoting gastric cancer cell migration. Oncol Rep. 2018;40(2):1209-1216
24. Liu Y, Zhai H, Chen H, et al. Upregulation of RBBP5 promotes cell cycle progression and proliferation in hepatocellular carcinoma. Oncol Lett. 2019;18(5):5094-5102.
25. Zhang Q, Gao L, Yang X, et al. RBBP5 functions as a tumor suppressor in melanoma by promoting H3K4me3 expression, thereby inhibiting cell proliferation, migration, and invasion. Oncol Lett. 2022;23(3):95.
26. Bei M. Molecular genetics of ameloblast cell lineage. J Exp Zool B Mol Dev Evol. 2009;312B(5):437-44. https://doi.org/10.1002/jez.b.21261.
27. Jordan SN, Canman JC. Rho GTPases in animal cell cytokinesis: an occupation by the one percent. Cytoskeleton (Hoboken). 2012;69(11):919-30. https://doi.org/10.1002/cm.21071.
28. Hervé JC, Bourmeyster N. Rho GTPases at the crossroad of signaling networks in mammals. Small GTPases. 2015;6(2):43-8. https://doi.org/10.1080/21541248.2015.1044811.
29. Otsu K, Harada H. Rho GTPases in ameloblast differentiation. Jpn Dent Sci Rev. 2016;52(2):32-40. https://doi.org/10.1016/j.jdsr.2015.09.001.
30. Modolo F, Biz MT, de Sousa SM, Fachinelli Rde L, Crema VO. Immunohistochemical expression of Rho GTPases in ameloblastomas. J Oral Pathol Med. 2012;41(5):400-7. https://doi.org/10.1111/j.1600-0714.2011.01108.x.
31. Blume-Jensen P, Hunter T. Oncogenic kinase signalling. Nature. 2001;411(6835):355–65. https://doi.org/10.1038/35077225.
32. Olsen JV, Blagoev B, Gnad F, Macek B, Kumar C, Mortensen P, Mann M. Global, in vivo, and site-specific phosphorylation dynamics in signaling networks. Cell. 2006;127(3):635–48. https://doi.org/10.1016/j.cell.2006.09.026.
33. Zhang J, Yang PL, Gray NS. Targeting cancer with small molecule kinase inhibitors. Nat Rev Cancer. 2009;9(1):28–39. https://doi.org/10.1038/nrc2559.
34. Mertins P, Mani DR, Ruggles KV, Gillette MA, Clauser KR, Wang P, et al. Proteogenomics connects somatic mutations to signalling in breast cancer. Nature. 2016;534(7605):55–62. https://doi.org/10.1038/nature18003.
35. Aebersold R, Mann M. Mass spectrometry-based proteomics. Nature. 2003;422(6928):198–207. https://doi.org/.1038/nature01511.
36. Cox J, Mann M. Quantitative, high-resolution proteomics for data-driven systems biology. Annu Rev Biochem. 2011;80:273–99. https://doi.org/10.1146/annurev-biochem-061308-093216.
37. Thingholm TE, Jensen ON, Larsen MR. Analytical strategies for phosphoproteomics. Proteomics. 2009;9(6):1451–68. https://doi.org/10.1002/pmic.200800454.
38. Mertins P, Qiao JW, Patel J, Udeshi ND, Clauser KR, Mani DR, et al. Integrated proteomic analysis of post-translational modifications by serial enrichment. Nat Methods. 2013;10(7):634–7. https://doi.org/10.1038/nmeth.2518.