Evaluation of Ocimum Santum as MMP Inhibitors in Human Dentin: an In Vitro Study

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Abstract:Matrix metalloproteinases (MMPs) play a significant role in the degradation of the dentin collagen matrix, contributing to dental caries progression. Inhibiting MMPs can preserve dentin integrity and improve the longevity of dental restorations. This study evaluates the potential of *Ocimum sanctum*, known for its anti-inflammatory, antioxidant, and antimicrobial properties, as a natural MMP inhibitor. Active compounds such as eugenol, ursolic acid, and rosmarinic acid may inhibit MMP activity, protecting dentin from collagen degradation. Fifteen freshly extracted, healthy human teeth were collected and stored at 4°C until further processing. To prepare the dentin extract, the roots were separated, and both the enamel and any remaining pulp tissue were carefully removed. The cleaned teeth were then pulverized in liquid nitrogen using a minimal volume of 50 mM phosphate buffer to obtain a dentin powder extract, which served as the source of matrix metalloproteinases (MMPs).This dentin powder extract was subsequently treated with a solution of *Ocimum sanctum* to evaluate its inhibitory effect on MMP activity. The treated samples were incubated under controlled conditions, and enzyme inhibition was assessed using enzyme-linked immunosorbent assay (ELISA) to quantify the extent of MMP suppression.There is dose dependant decrease in MMP activity. Maximum inhibitory activity was observed at maximum concentration*Ocimum sanctum* has a significant decrease in MMP activity in treated samples compared to controls suggesting its potential use as a natural agent in dental treatments to enhance the longevity of restorations

**Key words:** *Ocimum sanctum*, MMP inhibitors, human dentin, in vitro study

# Introduction

Matrix metalloproteinases are enzymes which are linked to morphogenesis, tissue repair, and wound healing. They are important players in the breakdown of extracellular matrix and bone remodeling.[(Nagase et al., 2006; Visse & Nagase, 2003)](https://paperpile.com/c/eTxaNf/hKTmh+oyOpA) MMPs are activated by acid generation by cariogenic bacteria and are crucial in the breakdown of dentine during the formation of caries[(Ajay et al., 2023; Chokkattu et al., 2023; Padarthi et al., 2023)](https://paperpile.com/c/eTxaNf/6TgT8+64tb1+BbcIO). There are 23 types of MMPs each known to have different catalytic characteristics[(Keerthana & Ramesh, 2021; Murugesan, 2021; Tiwari & Jain, 2021)](https://paperpile.com/c/eTxaNf/TtzLS+xQvcE+Thp3c). They are divided into six groups: membrane-type MMPs, collagenases, gelatinases, stromelysins, matrilysins, and other MMPs, based on homology and substrate specificity. [(Sulkalam et al., 2007)](https://paperpile.com/c/eTxaNf/yRVSO) [(Mazzoni et al., 2007)](https://paperpile.com/c/eTxaNf/4kaVx) The resin-dentin bonding is primarily responsible for the clinical results and durability of composite restorations[(Ramakrishnan et al., 2023; Shenoy & Maiti, 2023; J. S. Sindhu et al., 2023)](https://paperpile.com/c/eTxaNf/XeFbr+dtoQQ+1Yevy). The hybrid layer is an exclusive mixture of mineralized dentin with resin infiltration and demineralized dentin combined with adhesives [(Pranati et al., 2021; Sakthi et al.,2021)](https://paperpile.com/c/eTxaNf/CyhTS+6QR2Z). The activation of natural collagenolytic enzymes contained in the collagen matrix destroys the exposed collagen matrix found in the hybrid layer. [(Tjäderhane et al., 2001)](https://paperpile.com/c/eTxaNf/0H3h)This enzymatic breakdown weakens the connection, compromising the long-term success of restorative procedures[(Kasabwala et al., 2021; Rajeshkumar & Lakshmi, 2021; Varghese et al., 2023)](https://paperpile.com/c/eTxaNf/6LNcT+SIm2a+qPAX2).The exploration of *Ocimum sanctum* as a natural MMP inhibitor could offer an alternative or complementary approach to conventional dental treatment [(Keerthana & Ramesh, 2021; Murugesan, 2021; Subramanian et al., 2021; Tiwari & Jain, 2021)](https://paperpile.com/c/eTxaNf/TtzLS+xQvcE+Thp3c+2PZ2J). *Ocimum sanctum* possesses a wide range of beneficial properties, including antioxidant, antidiabetic, and antiulcer effects[(Dharman et al., 2023; S. Sindhu et al., 2023; Sreenivasagan et al., 2023)](https://paperpile.com/c/eTxaNf/F2Fjp+xYYuU+UOMrE). It also exhibits potent anticancer, antibacterial, and antifungal activities. [(Utispan et al., 2020)](https://paperpile.com/c/eTxaNf/QKDiN) This study aims to evaluate the inhibitory effects of *Ocimum sanctum* extracts on MMPs in human dentin, providing insights into its possible therapeutic applications in dentistry.

# Materials and methods

## Preparation of *Ocimum sanctum* extract

To avoid contamination, dried leaves of *Ocimum sanctum* were collected, cleaned, and allowed to air dry in a controlled setting. After that, the dried leaves have been crushed into a fine powder. The leaf powder was macerated for. 48 hours at room temperature with continuous stirring using 70% ethanol as the extraction solvent After filtering, the ethanol extract was concentrated under low pressure in a rotary evaporator and kept cold (-20°C). The extract was reconstituted in dimethyl sulfoxide (DMSO) to create working solutions with the appropriate concentrations prior to use in the experiments.

## Preparation of Human Dentin Powder

Informed consent and ethical approval were obtained before any human premolars were removed for orthodontic purposes. To separate the dentin from the rest of the tooth, they were cleaned and sectioned. To prevent heat damage, the dentin was then ground into a fine powder using a cryogenic mill. By treating the dentin powder with 10% phosphoric acid for 24 hours, the organic matrix was made visible and the mineral content was eliminated. This process is known as demineralization. Following demineralization, the dentin powder was cleaned with 1 M sodium hydroxide (NaOH) until the pH reached a neutral level. Before being used again, the neutralized dentin powder was washed with phosphate-buffered saline (PBS) and kept at 4°C.

## Experimental group

To evaluate the impact of ocimum sanctum extract on MMP-9 activity, the study comprised four groups:

Group 1: Control (PBS-treated): PBS-treated dentin powder was used as the standard for measuring MMP-9 activity.

Group 2: Negative Control (DMSO-treated): To account for potential effects of the solvent used to reconstitute the ocimum sanctum extract, dentin powder was treated with DMSO alone.

Group 3: Positive Control (Doxycycline-treated): To serve as a baseline for comparison, dentin powder was treated with doxycycline, a recognized synthetic MMP inhibitor.

Group 4: Test Groups (100 μg and 200 μg ocimum sanctum extract): Dentin powder treated with two distinct ocimum sanctum extract concentrations (100 μg and 200 μg) in order to assess the extract's dose-dependent impact on MMP-9 activity.

## Incubation protocol

Dentin powder was divided into groups and treated with PBS, DMSO, doxycycline, or *Ocimum sanctum* extract for the duration of 24 hours at 37°C. The approaches were able to interact with the dentin matrix and possibly suppress MMP-9 activity during the incubation period.

## Measurement of MMP-9 activity

Following incubation, an enzyme-linked immunosorbent assay (ELISA) kit designed specifically for human MMP-9 was used to quantify the MMP-9 activity in each sample. The ability of the enzyme to cleave a particular substrate that is attached to a colorimetric reporter is how the ELISA kit determines the concentration of active MMP-9. Using a microplate reader, the absorbance of the reaction product was determined at 450 nm. The MMP-9 activity in the samples was directly correlated with the absorbance values.

# Statistical analysis

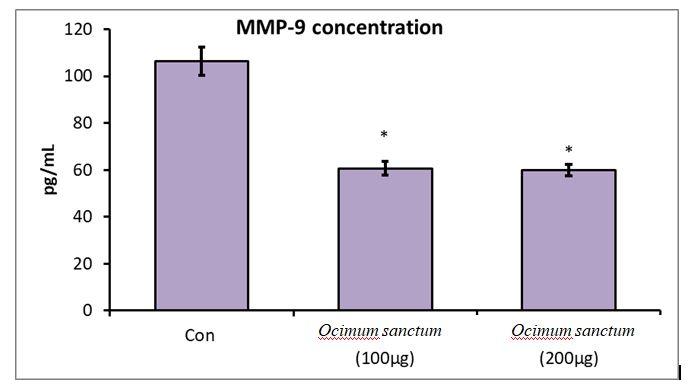
After analysis, the data were shown as mean ± standard deviation (SD). One-way analysis of variance (ANOVA) was used to compare the groups statistically, and post hoc testing was used to identify any significant differences between the groups. A statistically significant p-value of less than 0.05 meant that the observed differences were not the result of chance. Version 25.0 of SPSS software was utilized for all statistical analyses.

# Results

*Ocimum sanctum* on MMP-9 concentration, demonstrating a progressive decline in MMP-9 levels as the administered dose increases from 100 µg to 200 µg. (Table 1) A dose of 200 µg demonstrates superior inhibition of MMP-9, indicating that a higher concentration enhances therapeutic potential. This suggests that the bioactive compounds present in *Ocimum sanctum* interact with MMP-9, potentially inhibiting its enzymatic activity or down regulating its expression. The consistently low SEM values across all experimental groups, suggests high reliability and reproducibility of the observed data(Saadh et al., 2024). (Table 2) Low SEM values indicate minimal variation around the mean, reinforcing the statistical significance of the results. (Graph 1) This precision strengthens confidence in the inhibitory effect of *Ocimum sanctum* on MMP-9 and supports the hypothesis that its bioactive constituents contribute to enzymatic modulation in a controlled and quantifiable manner (Almatrafi et al., 2024).

**Table-1:** The table presents OD values and corresponding MMP-9 concentrations (pg/mL) for three different treatment groups: control, O*cimum sanctum* (100 µg), and (200 µg) *Ocimum sanctum*

| [**Sample. no**](http://sample.no/) |  | **OD Value** | **pg/ml** |
| --- | --- | --- | --- |
| 1 | Control | 0.159 | 112.9 |
| 1 | Control | 0.141 | 100.1 |
| 2 | ocimum sanctum (100µg) | 0.082 | 58.2 |
| 2 | ocimum sanctum (100µg) | 0.089 | 63.2 |
| 3 | ocimum sanctum (200µg) | 0.076 | 54.0 |
| 3 | ocimum sanctum (200µg) | 0.0673 | 47.8 |



**Figure 1**: The graph shows the standard error of the mean (SEM) and the mean concentration of MMP-9 (in pg/mL) for each treatment group.

**Table 2:** The table provides data on the mean concentration of MMP-9 (in pg/mL) and the standard error of the mean (SEM) for different treatment groups

|  | Conc | *Ocimum sanctum*  (100µg) | *Ocimum sanctum*  (200µg) |
| --- | --- | --- | --- |
| Mean | 106.5 | 60.7 | 59.9 |
| S.E.M | 6 | 3 | 2.5 |

# Discussion

The results of this study demonstrate that *Ocimum sanctum* effectively inhibits MMP-9 activity in a dose-dependent manner, with higher doses (200 µg) producing a greater reduction in MMP-9 concentration compared to lower doses (100 µg). This trend suggests that the bioactive compounds in *Ocimum sanctum* may interfere with MMP-9 function through direct enzyme inhibition or transcriptional down regulation. Given that MMP-9 plays a key role in extracellular matrix degradation, inflammation, and pathological tissue remodeling, these findings highlight the potential therapeutic value of *Ocimum sanctum*. The consistently low SEM values observed across all groups indicate high precision in the measurements, reinforcing the reliability of the data. The observed dose-dependent response aligns with classical pharmacokinetic and pharmacodynamic principles, where enzyme inhibition increases with higher concentrations of the bioactive compound. This supports the notion that *Ocimum sanctum* contains phytochemicals capable of modulating MMP-9 activity, possibly through mechanisms such as zinc chelation, enzyme binding, or suppression of MMP-9 gene expression. Further studies are necessary to isolate and characterize the specific active constituents responsible for this inhibitory effect.Chlorhexidine and other MMP inhibitors have been proven effective in reducing MMP activity through various application methods.[(Breschi et al., 2009; Loguercio et al., 2009)](https://paperpile.com/c/eTxaNf/i7EwA+hwubQ) Gendron et al. demonstrated that chlorhexidine effectively inhibits MMP-2, MMP-8, and MMP-9, while additional studies have shown that concentrations of 0.2% and 2% enhance stability, increase durability, and reduce bond strength deterioration over time.[(Gendron et al., 1999)](https://paperpile.com/c/eTxaNf/6X9lo) Proanthocyanidin, a naturally occurring bioactive compound, is another widely used MMP inhibitor with dual functionality. In addition to suppressing MMP activity, it acts as a collagen crosslinker within human dentin, strengthening its mechanical properties. [(Hass, Luque-Martinez, Gutierrez, et al., 2016; Hass, Luque-Martinez, Muñoz, et al., 2016; Kiuru et al., 2021; Lee et al., 2008)](https://paperpile.com/c/eTxaNf/6x9Pl+lsnyc+RmBok+rXk1K)From a clinical perspective, the findings suggest that *Ocimum sanctum* could serve as a natural alternative for regulating MMP-9 activity, potentially contributing to therapies aimed at preventing matrix degradation in various pathological conditions.[(Kim et al., 2010; Kwak et al., 2014)](https://paperpile.com/c/eTxaNf/kuDob+amWBv) However, while in vitro results provide valuable mechanistic insights, additional in vivo studies and clinical trials are required to validate these effects in physiological conditions. Future research should also explore optimal dosages, bioavailability, and possible synergistic effects with existing therapeutic agents to maximize the clinical potential of *Ocimum sanctum* in MMP-9-associated diseases.

# Conclusion

This study demonstrates that *Ocimum sanctum* inhibits MMP-9 concentration in a dose-dependent manner, with higher doses (200 µg) showing greater efficacy than lower doses (100 µg). The consistently low SEM values indicate high measurement precision, reinforcing the reliability of the observed inhibitory effect.

# References

1. Almatrafi, T. A., Almohaimeed, H. M., Chakravarthi, S., Amin, A. H., Jafer, A., & Akhavan-Sigari, R. (2024). Reducing metastasis ability of gastric cancer cell line by targeting MMP16 using miR-193a-5p and 5-FU. Advances in Medical Sciences, 69(2), 463-473.
2. [Ajay, R., JafarAbdulla, M. U., Sivakumar, J. S., Baburajan, K., Rakshagan, V., & Eyeswarya, J. (2023). Dental alloy adhesive primers and bond strength at alloy-resin interface: A systematic review and meta-analyses. *The Journal of Contemporary Dental Practice*, *24*(8), 521–544.](http://paperpile.com/b/eTxaNf/64tb1)
3. [Breschi, L., Cammelli, F., Visintini, E., Mazzoni, A., Vita, F., Carrilho, M., Cadenaro, M., Foulger, S., Mazzoti, G., Tay, F. R., Di Lenarda, R., & Pashley, D. (2009). Influence of chlorhexidine concentration on the durability of etch-and-rinse dentin bonds: a 12-month in vitro study. *The Journal of Adhesive Dentistry*, *11*(3), 191–198.](http://paperpile.com/b/eTxaNf/hwubQ)
4. [Chokkattu, J. J., Mary, D. J., Shanmugam, R., & Neeharika, S. (2023). Evaluation clove ginger-mediated titanium oxide nanoparticles-based dental varnish against Streptococcus mutans Lactobacillus Species: vitro study. *World J Dent*, *14*(3), 233–237.](http://paperpile.com/b/eTxaNf/BbcIO)
5. [Dharman, S., Maragathavalli, G., Shanmugam, R., & Shanmugasundaram, K. (2023). Curcumin mediated gold nanoparticles analysis its antioxidant, anti-inflammatory, antimicrobial activity against oral pathogens. *Pesquisa Brasileira Em Odontopediatria E Clínica Integrada*, *23*.](http://paperpile.com/b/eTxaNf/xYYuU)
6. [Gendron, R., Grenier, D., Sorsa, T., & Mayrand, D. (1999). Inhibition of the activities of matrix metalloproteinases 2, 8, and 9 by chlorhexidine. *Clinical and Diagnostic Laboratory Immunology*, *6*(3), 437–439.](http://paperpile.com/b/eTxaNf/6X9lo)
7. [Hass, V., Luque-Martinez, I., Muñoz, M. A., Reyes, M. F. G., Abuna, G., Sinhoreti, M. A. C., Liu, A. Y., Loguercio, A. D., Wang, Y., & Reis, A. (2016). The effect of proanthocyanidin-containing 10% phosphoric acid on bonding properties and MMP inhibition. *Dental Materials: Official Publication of the Academy of Dental Materials*, *32*(3), 468–475.](http://paperpile.com/b/eTxaNf/lsnyc)
8. [Hass, V., Luque-Martinez, I. V., Gutierrez, M. F., Moreira, C. G., Gotti, V. B., Feitosa, V. P., Koller, G., Otuki, M. F., Loguercio, A. D., & Reis, A. (2016). Collagen cross-linkers on dentin bonding: Stability of the adhesive interfaces, degree of conversion of the adhesive, cytotoxicity and in situ MMP inhibition. *Dental Materials: Official Publication of the Academy of Dental Materials*, *32*(6), 732–741.](http://paperpile.com/b/eTxaNf/rXk1K)
9. [Kasabwala, H., Nallaswamy, D., Subhashree, R., & Ahmed, N. (2021). Evaluation Of Overall Marginal Accuracy Of DMLS Copings Fabricated Using 3 Different DMLS Printing Machines. *Int J Dentistry Oral Sci*, *8*(7), 3335–3340.](http://paperpile.com/b/eTxaNf/SIm2a)
10. [Keerthana, T., & Ramesh, S. (2021). Knowledge, attitude and practice survey on awareness of the association between diet and dental erosion. *International Journal of Dentistry and Oral Science*, *8*(2), 1533–1540.](http://paperpile.com/b/eTxaNf/xQvcE)
11. [Kim, S.-C., Magesh, V., Jeong, S.-J., Lee, H.-J., Ahn, K.-S., Lee, H.-J., Lee, E.-O., Kim, S.-H., Lee, M.-H., Kim, J. H., & Kim, S.-H. (2010). Ethanol extract of Ocimum sanctum exerts anti-metastatic activity through inactivation of matrix metalloproteinase-9 and enhancement of anti-oxidant enzymes. *Food and Chemical Toxicology: An International Journal Published for the British Industrial Biological Research Association*, *48*(6), 1478–1482.](http://paperpile.com/b/eTxaNf/kuDob)
12. [Kiuru, O., Sinervo, J., Vähänikkilä, H., Anttonen, V., & Tjäderhane, L. (2021). MMP inhibitors and dentin bonding: Systematic review and meta-analysis. *International Journal of Dentistry*, *2021*, 9949699.](http://paperpile.com/b/eTxaNf/6x9Pl)
13. [Kwak, T.-K., Sohn, E. J., Kim, S., Won, G., Choi, J.-U., Jeong, K., Jeong, M., Kwon, O. S., & Kim, S.-H. (2014). Inhibitory effect of ethanol extract of Ocimum sanctum on osteopontin mediated metastasis of NCI-H460 non-small cell lung cancer cells. *BMC Complementary and Alternative Medicine*, *14*(1). https://doi.org/](http://paperpile.com/b/eTxaNf/amWBv)[10.1186/1472-6882-14-419](http://dx.doi.org/10.1186/1472-6882-14-419)
14. [Lee, K. W., Kang, N. J., Oak, M.-H., Hwang, M. K., Kim, J. H., Schini-Kerth, V. B., & Lee, H. J. (2008). Cocoa procyanidins inhibit expression and activation of MMP-2 in vascular smooth muscle cells by direct inhibition of MEK and MT1-MMP activities. *Cardiovascular Research*, *79*(1), 34–41.](http://paperpile.com/b/eTxaNf/RmBok)
15. [Loguercio, A. D., Stanislawczuk, R., Polli, L. G., Costa, J. A., Michel, M. D., & Reis, A. (2009). Influence of chlorhexidine digluconate concentration and application time on resin-dentin bond strength durability. *European Journal of Oral Sciences*, *117*(5), 587–596.](http://paperpile.com/b/eTxaNf/i7EwA)
16. [Mazzoni, A., Mannello, F., & Tay, F. R. (2007). Zymographic analysis and characterization ofMMP-2 and -9 forms in human sound dentin. *J Dent Res*, *86*(05), 436–440.](http://paperpile.com/b/eTxaNf/4kaVx)
17. [Murugesan, A. (2021). Saravana Dinesh SP evaluation of shear bond strength of ceramic brackets with two different base designs: An in-vitro study. *Int J Dentistry Oral Sci*.](http://paperpile.com/b/eTxaNf/Thp3c) <https://www.academia.edu/download/72981941/IJDOS_2377_8075_08_304.pdf>
18. [Nagase, H., Visse, R., & Murphy, G. (2006). Structure and function of matrix metalloproteinases and TIMPs. *Cardiovascular Research*, *69*(3), 562–573.](http://paperpile.com/b/eTxaNf/oyOpA)
19. [Padarthi, L. C., Anumula, L., Chinni, S. K., Sannapureddy, S., & Govula, K. (2023). Evaluation Composite Restoration Posterior Teeth Proanthocyanidin Pretreatment Liner Using Fédération Dentaire Internationale Criteria: Split-mouth Randomized Controlled Trial. *International Journal Prosthodontics Restorative Dentistry*, *13*(4), 191–200.](http://paperpile.com/b/eTxaNf/6TgT8)
20. [Pranati, T., Ranjan, M., & Sandeep, A. H. (2021). Marginal adaptability custom made cast post made different techniques-a literature review. *Int J Dentistry Oral Sci*, *8*(8), 3954–3959.](http://paperpile.com/b/eTxaNf/6QR2Z)
21. [Rajeshkumar, S., & Lakshmi, T. (2021). Biomedical potential of zinc oxide nanoparticles synthesized using plant extracts. *Int J Dent Oral Sci*, *8*, 4160–4163.](http://paperpile.com/b/eTxaNf/qPAX2)
22. [Ramakrishnan, M., Shanmugam, R., Neeharika, S., Chokkattu, J. J., Thangavelu, L., & Khanna, N. (2023). Anti-inflammatory activity and cytotoxic effect of ginger and Rosemary-mediated titanium oxide nanoparticles-based dental varnish. *World Journal of Dentistry*, *14*(9), 761–765.](http://paperpile.com/b/eTxaNf/dtoQQ)
23. [Sakthi, S., (2021). Thymus vulgaris mediated selenium nanoparticles, characterization and its antimicrobial activity - an in vitro study. *International Journal of Dentistry and Oral Science*, 3516–3521.](http://paperpile.com/b/eTxaNf/CyhTS)
24. [Shenoy, N. D., & Maiti, S. (2023). Evaluation marginal fit CAD/CAM crowns using CBCT digital scanners. *Annals Dental Specialty*, *11*(3-2023), 37–44.](http://paperpile.com/b/eTxaNf/XeFbr)
25. Saadh, M. J., Rasulova, I., Khalil, M., Farahim, F., Sârbu, I., Ciongradi, C. I. (2024). Natural killer cell-mediated immune surveillance in cancer: Role of tumor microenvironment. Pathology-Research and Practice, 254, 155120.
26. [Sindhu, J. S., Maiti, S., & Nallaswamy, D. (2023). Comparative analysis on efficiency and accuracy of parallel confocal microscopy and three-dimensional in motion video with triangulation technology-based intraoral scanner under influence of moisture and mouth opening - A crossover clinical trial. *Journal of Indian Prosthodontic Society*, *23*(3), 234–243.](http://paperpile.com/b/eTxaNf/1Yevy)
27. [Sindhu, S., Maiti, S., & Nallaswamy, D. (2023). Factors affecting accuracy intraoral scanners-a systematic review. *Annals Dental Specialty*, *11*(1-2023), 40–52.](http://paperpile.com/b/eTxaNf/F2Fjp)
28. [Sreenivasagan, S., Subramanian, A. K., Mohanraj, K. G., & Kumar, R. S. (2023). Assessment of toxicity of Green Synthesized Silver Nanoparticle-coated Titanium Mini-implants with Uncoated Mini-implants: Comparison in an Animal Model Study. *The Journal of Contemporary Dental Practice*, *24*(12), 944–950.](http://paperpile.com/b/eTxaNf/UOMrE)
29. [Subramanian, E., Ravindran, V., & Jeevanandan, G. (2021). Comparison of amount of tooth reduction in primary first molar for stainless steel, zirconia and fibre-glass crowns–in-vitro study. *International Journal of Dentistry and Oral Science*, *8*(7), 3427–3430.](http://paperpile.com/b/eTxaNf/2PZ2J)
30. [Sulkalam, T. T., Sorsa, T., Larmas, M., Salo, T., & Tjäderhane, L. (2007). Matrix metalloproteinase-8 (MMP-8) is the major collagenase in human dentin. *Arch Oral Biol*, *52*(02), 121–127.](http://paperpile.com/b/eTxaNf/yRVSO)
31. [Tiwari, A., & Jain, R. K. (2021). The effect of motivational and reminder therapy on the compliance of patients wearing fixed appliances. *Int J Dent Oral Sci*, *8*(7), 3303–3305.](http://paperpile.com/b/eTxaNf/TtzLS)
32. [Tjäderhane, L., Palosaari, H., Wahlgren, J., Larmas, M., Sorsa, T., & Salo, T. (2001). Human odontoblast culture method: the expression of collagen and matrix metalloproteinases (MMPs). *Advances in Dental Research*, *15*(1), 55–58.](http://paperpile.com/b/eTxaNf/0H3h)
33. [Utispan, K., Niyomtham, N., Yingyongnarongkul, B.-E., & Koontongkaew, S. (2020). Ethanolic extract of Ocimum sanctum leaves reduced invasion and matrix metalloproteinase activity of head and neck cancer cell lines. *Asian Pacific Journal of Cancer Prevention: APJCP*, *21*(2), 363–370.](http://paperpile.com/b/eTxaNf/QKDiN)
34. [Varghese, R., Maliael, M., & Subramanian, A. (2023). Antibacterial activity of nanoparticle-coated orthodontic archwires: A systematic review. *Journal of International Oral Health: JIOH*, *15*(1), 1.](http://paperpile.com/b/eTxaNf/6LNcT)
35. [Visse, R., & Nagase, H. (2003). Matrix metalloproteinases and tissue inhibitors of metalloproteinases: structure, function, and biochemistry. *Circulation Research*, *92*(8), 827–839.](http://paperpile.com/b/eTxaNf/hKTmh)