Revista de Ciências Agroveterinárias
Universidade do Estado de Santa Catarina (UDESC)

Research Article

Comparative Effectiveness of Mineral Trioxide Aggregate (MTA) and Calcium Hydroxide in Partial Biopulpectomies of *Mus musculus* teeth

Eficácia comparativa do agregado de trióxido mineral (MTA) e do hidróxido de cálcio em biopulpectomias parciais de dentes de Mus musculus

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Submission: March 05th, 2025 | Acceptance: April 21th, 2025

ABSTRACT

This study compared the effectiveness of Calcium Hydroxide and Mineral Trioxide Aggregate (MTA) in partial biopulpectomies of *Mus musculus* teeth. A total of 16 rats (n=8 per group) were used, evaluating inflammation (levels 1: very mild to 4: severe), necrosis, and granulation tissue formation at 7 and 21 days. Using non-parametric tests (Fisher's exact test, McNemar, Wilcoxon, and Mann-Whitney), no significant intergroup differences were found in inflammation ($p > 0.05$ at 7 days; $p > 0.05$ at 21 days) or necrosis ($p > 0.05$ at 7 days; $p > 0.05$ at 21 days). However, MTA exhibited superior clinical trends, achieving complete necrosis resolution (0% vs. 25% in Calcium Hydroxide) and granulation tissue formation (100% vs. 87.5%) at 21 days, with significant intragroup reductions in granulation ($p < 0.05$ vs. $p < 0.05$). The wide confidence intervals (e.g., necrosis in MTA at 21 days: 0.0–36.9%) and moderate effect sizes (φ=0.42; r=-0.45) reflected sample size limitations, highlighting the need for larger studies. While both materials demonstrated biocompatibility, MTA emerges as a preferred option in contexts prioritizing comprehensive tissue repair, supported by its profile without post-operative complications. These findings provide preliminary evidence to guide future multidisciplinary investigations with greater statistical power.

KEYWORDS: Inflammatory cellular response. Presence of Necrosis Areas. Granulation tissue formation. Therapeutic dentistry.

RESUMO

Este estudo comparou a eficácia do hidróxido de cálcio e do agregado de trióxido mineral (MTA) em biopulpectomias parciais de dentes de Mus musculus. Um total de 16 ratos (n=8 por grupo) foi usado, avaliando a inflamação (níveis 1: muito leve a 4: grave), a necrose e a formação de tecido de granulação em 7 e 21 dias. Usando testes não paramétricos (teste exato de Fisher, McNemar, Wilcoxon e Mann-Whitney), não foram encontradas diferenças significativas entre os grupos na inflamação ($p > 0,05$ em 7 dias; $p > 0,05$ em 21 dias) ou necrose ($p > 0,05$ em 7 dias; $p > 0,05$ em 21 dias). No entanto, o MTA apresentou tendências clínicas superiores, alcançando resolução completa da necrose (0% vs. 25% no hidróxido de cálcio) e formação de tecido de granulação (100% vs. 87,5%) em 21 dias, com reduções significativas intragrupo na granulação ($p < 0,05$ vs. $p < 0,05$). Os amplos intervalos de confiança (por exemplo, necrose no MTA aos 21 dias: 0,0-36,9%) e tamanhos de efeito moderados (φ=0,42; r=-0,45) refletiram as limitações do tamanho da amostra, destacando a necessidade de estudos maiores. Embora ambos os materiais tenham demonstrado biocompatibilidade, o MTA surge como uma opção preferida em contextos que priorizam o reparo abrangente do tecido, apoiado por seu perfil sem complicações pós-operatórias. Esses achados fornecem evidências preliminares para orientar futuras investigações multidisciplinares com maior poder estatístico.

PALAVRAS-CHAVE: Resposta celular inflamatoria. Presença de áreas de necrose. Formação de tecido de granulação. Odontologia terapêutica.

INTRODUCTION

Partial biopulpectomy, also known as pulpotomy, is a fundamental therapeutic procedure in conservative dentistry aimed at preserving pulp vitality in teeth with exposure or irreversible inflammation of the coronal pulp (BOUTSIOUKI et al. 2021, BASTOS et al. 2024). This approach involves the surgical amputation of the affected pulp tissue (OZTURK & GENC SEN 2024), followed by the application of a medicated agent on the remaining radicular pulp to promote its healing and the formation of a protective dentin barrier (LEONARDO et al. 1980). Its clinical relevance lies in the possibility of avoiding more invasive treatments, such as total pulpectomy or tooth extraction, especially in young teeth with immature apices, where preserving pulp vitality is critical for root development and the homeostasis of the dentin-pulp complex (ABU-TAHUN & TORABINEJAD 2010, COOPER et al. 2021).

Among the materials used in these therapies, calcium hydroxide has historically been the agent of choice due to its ability to neutralize the pH of the environment (SOLIS et al. 2024), exert an antibacterial effect, and stimulate the formation of tertiary dentin (SILVA et al. 2023). However, its use presents limitations, such as solubility, marginal microleakage, and inconsistency in forming a homogeneous dentin bridge. In contrast, Mineral Trioxide Aggregate (MTA) has emerged as an innovative alternative, standing out for its biocompatibility, hermetic sealing, and ability to induce cellular proliferation and odontoblast differentiation (VERMA et al. 2021), attributes that position it as a promising material in regenerative pulp therapies (GOMES-FILHO et al. 2012).

The preclinical evaluation of these materials in animal models, such as *Mus musculus*, offers unique methodological advantages (STANDLEY et al. 2024), including the standardization of experimental conditions, genetic homogeneity, and the ability to analyze early histological responses (AUGUSTSSON et al. 2005). These models not only replicate key biological processes observed in humans, such as pulp inflammation and mineralized tissue formation, but also allow for the exploration of molecular mechanisms underlying pulp repair, which is essential for validating the safety and efficacy of new treatments before their translation into clinical practice. Previous studies in dental research have increasingly employed *Mus musculus* as a model organism due to its well-documented genetic homogeneity, favorable cost-to-benefit ratio, and anatomical suitability for standardized pulpal interventions (SHIN et al. 2014, LINHARES et al. 2018). Its molar structure allows for precise operative access and consistent histopathological analysis, particularly in early-phase healing. Compared to larger models such as Wistar rats, *Mus musculus* also facilitates refined microsurgical techniques and is especially advantageous in studies aiming to evaluate cellular responses to biomaterials within a controlled experimental timeframe.

In this context, the present study focuses on comparing the effectiveness of calcium hydroxide and MTA in partial biopulpectomies performed on *Mus musculus* teeth, evaluating critical parameters such as reparative dentin formation, inflammatory

response, and the presence of necrosis or granulation tissue. The findings of this research aim to provide robust scientific evidence to optimize therapeutic strategies in endodontics, prioritizing the preservation of pulp vitality and reducing the need for invasive interventions in pediatric and young adult patients.

MATERIALS AND METHODS

The procedures and ethical aspects of this research were based on the "Code of Ethics for Scientific Research." Additionally, the study was conducted following international and national guidelines for the care and use of animals in research, in accordance with the "Directive 2010/63/EU of the European Union on the Protection of Animals Used for Scientific Purposes." These regulations were supported by the letter “N° 003-GRJ-DRA-AAC-PERÚ-2024” that ensured the welfare of the specimens and compliance with ethical standards throughout the experimental process.

Study Area

The present study was conducted at two specialized institutions: the San Pablo Veterinary Clinic and the Huancayo Dental Care Clinic, both located in the city of Huancayo, Huancayo district, Huancayo province, Junín department, at an altitude of 3,270 meters above sea level (SENAMHI – PERÚ 2022). The San Pablo Veterinary Clinic provided the facilities and necessary care for the *Mus musculus* specimens used in the research, ensuring optimal biosecurity conditions and animal welfare. Meanwhile, the Huancayo Dental Care Clinic supplied the equipment and materials required for performing the partial biopulpectomy procedures and the clinical follow-up of the cases. Data collection took place between June and August 2024, following strict ethical and methodological protocols.

Animals, data colection

Sixteen 8-week-old rodents of the *Mus musculus* species were used, selected as an experimental model due to their biological similarity to human pulp processes and their widespread use in dental research. These animals were biologically, nutritionally and genetically similar, thus supporting the use of that number of animals. At the veterinary clinic, they were housed in an animal facility under controlled conditions, with an ambient temperature ranging from 18 °C to 25 °C, 12-hour light-dark cycles, and ad libitum access to food and water. The rodents were anesthetized using 2% lidocaine with epinephrine to ensure a painless procedure. Absolute isolation of the operative field was performed to prevent contamination and ensure treatment precision.

Subsequently, a partial biopulpectomy was carried out, which involved the removal of carious tissue and the opening of the pulp chamber using high-speed burst. The coronal pulp was amputated with sharp curettes, leaving the radicular pulp intact, and hemorrhage was controlled using sterile cotton swabs soaked in saline solution.

In half of the specimens (8 rodents), calcium hydroxide was applied to the remaining radicular pulp, while in the other half (8 rodents), Mineral Trioxide Aggregate (MTA) was used as the therapeutic agent. Both materials were placed directly on the pulp tissue and covered with a layer of glass ionomer to prevent microleakage. Clinical and radiographic evaluations were performed at 7- and 21-days post-treatment, assessing parameters such as tertiary dentin formation, cellular inflammatory

response, the presence of necrotic areas, and granulation tissue formation.

The inflammatory response was classified into four ordinal levels: very mild (1), characterized by minimal cellular infiltrates; mild (2), with moderate infiltration without functional involvement; moderate (3), associated with edema and localized tissue damage; and severe (4), with extensive necrosis and organ dysfunction. This scale, adapted from standardized histopathological criteria, allowed a reproducible and quantitative assessment of inflammation, as recommended by the MASSOUD et al. (2023) tissue assessment guide.

To ensure animal welfare and minimize postprocedural discomfort, a postoperative analgesic protocol was implemented, consisting of subcutaneous administration of meloxicam at a dose of 1 mg/kg every 24 hours for 3 consecutive days following the intervention (OLIVERA-CALDERON et al. 2025). This regimen was selected based on its proven efficacy and safety in rodent models of oral surgery, contributing to ethical compliance and reducing the potential influence of pain-related confounders on the inflammatory outcomes (CARHUAS et al. 2024).

Statistical analysis

Statistical analysis was performed using R-Studio software version 4.4.2 (R CORE TEAM 2022) with the package agricolae and ggplot2, nonparametric tests according to the nature of the variables.ordinal nature of the data. To compare the groups (calcium hydroxide vs. MTA), Fisher's exact test was used for necrosis and granulation proportions, and the Mann-Whitney U test for ordinal variables. Temporal changes were evaluated with McNemar's test and the Wilcoxon test for paired samples (KASSAMBARA & MUNDT 2020).

RESULTS

Inflammatory Cellular Response

Table 1 shows differences in the inflammatory response at 7 days between the groups treated with Calcium Hydroxide (n=8) and Mineral Trioxide Aggregate (MTA) (n=8). While 37.5% of cases treated with Calcium Hydroxide exhibited very mild responses (3/8) and 12.5% severe responses (1/8, the only severe case in the study), the MTA group demonstrated a higher proportion of very mild responses (50%, 4/8) and an absence of severe cases. Both groups presented similar frequencies of mild (37.5% each) and moderate (12.5% each) responses. However, considering the overall sample (n=16), very mild (43.8%, 7/16) and mild (37.5%, 6/16) responses were predominant, suggesting a less severe inflammatory profile in the MTA group.

Table 2 reflects the progression of the inflammatory response 21 days post-treatment. In the Calcium Hydroxide group (n=8), 75% of cases (6/8) exhibited very mild responses, while 25% (2/8) showed mild responses. In contrast, in the MTA group (n=8), 87.5% (7/8) demonstrated very mild responses, and only 12.5% (1/8) had mild responses. At the overall level (n=16), very mild responses (81.3%, 13/16) were predominant, with the MTA group accounting for the highest proportion within this category (53.8% of all very mild responses). Both treatments showed a reduction in inflammatory severity compared to the first evaluation; however, MTA exhibited a more pronounced trend toward milder responses.

Table 1. Contingency of the first 7-day check-up.

		1st control				
		Mild	**Moderate**	**Severe**	**Very mild**	**Total**
Medication	Calcium hydroxide	3	1	1	3	8
		37.5%	12.5%	12.5%	37.5%	100.0%
		50.0%	50.0%	100.0%	42.9%	50.0%
		18.8%	6.3%	6.3%	18.8%	50.0%
	Mineral trioxide aggregate	3	1	0	4	8
		37.5%	12.5%	0.0%	50.0%	100.0%
		50.0%	50.0%	0.0%	57.1%	50.0%
		18.8%	6.3%	0.0%	25.0%	50.0%
Total		6	2	1	7	16
		37.5%	12.5%	6.3%	43.8%	100.0%

Table 2. Contingency of the second control after 21 days.

		2nd control		
		Mild	**Very mild**	**Total**
Medication	Calcium hydroxide	2	6	8
		25.0%	75.0%	100.0%
		66.7%	46.2%	50.0%
		12.5%	37.5%	50.0%
	Mineral trioxide aggregate	1	7	8
		12.5%	87.5%	100.0%
		33.3%	53.8%	50.0%
		6.3%	43.8%	50.0%
Total		3	13	16
		18.8%	81.3%	100.0%

From Table 3, intragroup comparisons using the Wilcoxon test for paired samples did not reveal statistically significant differences in inflammation levels between the first and second evaluations, neither for the Calcium Hydroxide group (p=0.233; effect size r=−0.42) nor for the Mineral Trioxide Aggregate (MTA) group (p=0.203; r=−0.45). Although the negative effect sizes suggest a trend toward inflammation reduction in both groups, this change did not reach statistical significance under conventional criteria (α=0.05). On the other hand, intergroup comparisons using the Mann-Whitney U test showed no significant differences either in the first evaluation (p=1.000) or the second evaluation (p=0.587), indicating similar inflammatory profiles between both treatments at each time point. These findings, obtained from a sample of 8 subjects per group, may be influenced by the limited statistical power associated with the small sample size. Overall, the results do not support the superiority of one treatment over the other, highlighting the need for future studies with larger cohorts to explore potential clinically relevant differences.

Table 3. Statistical methods for comparisons between groups.

Comparison	P-value	Size-effect	Method
CH: C1 vs C2	0.2330380	-0.421637	Wilcoxon
CH: C1 vs C2	0.2030918	-0.450000	Wilcoxon
CH vs MTA (C1)	1.0000000	NA	Mann-Whitney
CH vs MTA (C2)	0.5873695	NA	Mann-Whitney

CH: Calcium hydroxide; MTA: Mineral trioxide aggregate.

Presence of Necrosis Areas

From Table 4, statistical analysis using Fisher's exact test showed no significant differences in necrosis incidence between the Calcium Hydroxide and Mineral Trioxide Aggregate (MTA) groups, neither at 7 days (Calcium Hydroxide: 5/8 [62.5%, 95% CI: 24.5–91.5%] vs. MTA: 4/8 [50.0%, 95% CI: 15.2–84.8%]; p=0.999) nor at 21 days (Calcium Hydroxide: 2/8 [25.0%, 95% CI: 3.2–65.1%] vs. MTA: 0/8 [0.0%, 95% CI: 0.0–36.9%]; p=0.478). However, clinically relevant trends were observed: the MTA group completely eliminated necrosis cases by the end of the follow-up period (0/8), while the Calcium Hydroxide group reduced its prevalence from 62.5% to 25.0%, although without statistical significance (p=0.683, McNemar's test). In the MTA group, the reduction from 50.0% to 0.0% showed a trend toward significance (p=0.125), supported by a moderate effect size (φ = 0.42). However, the limited statistical power (n=8 per group) and the wide confidence intervals reflected uncertainty in the estimates. At 21 days, the absolute risk difference between groups was 25.0% (95% CI: -10.5–60.5%), highlighting the favorable profile of MTA.

Table 4. Comparisons between the two groups for necrosis area.

Group (n=8)	7 days	21 days	Difference (Post-Pre)	Method	P-value
CH	5/8 (62.5 %)	2/8 (25.0 %)	-3	McNemar	0.683
MTA	4/8 (5.0 %)	0/8 (0 %)	-4	McNemar	0.125
FET (P-value)	0.999	0.478			

CH: Calcium hydroxide; MTA: Mineral trioxide aggregate; Fet: Fisher's exact test.

Granulation tissue formation

From Figure 1, the evaluation of granulation tissue formation revealed that at 7 days post-treatment, the Calcium Hydroxide group presented granulation tissue in 25.0% of cases (2/8; 95% CI: 3.2–65.1%), while the Mineral Trioxide Aggregate (MTA) group showed a proportion of 37.5% (3/8; 95% CI: 8.5–75.5%), with no significant differences between the two (Fisher's exact test, p=0.999).

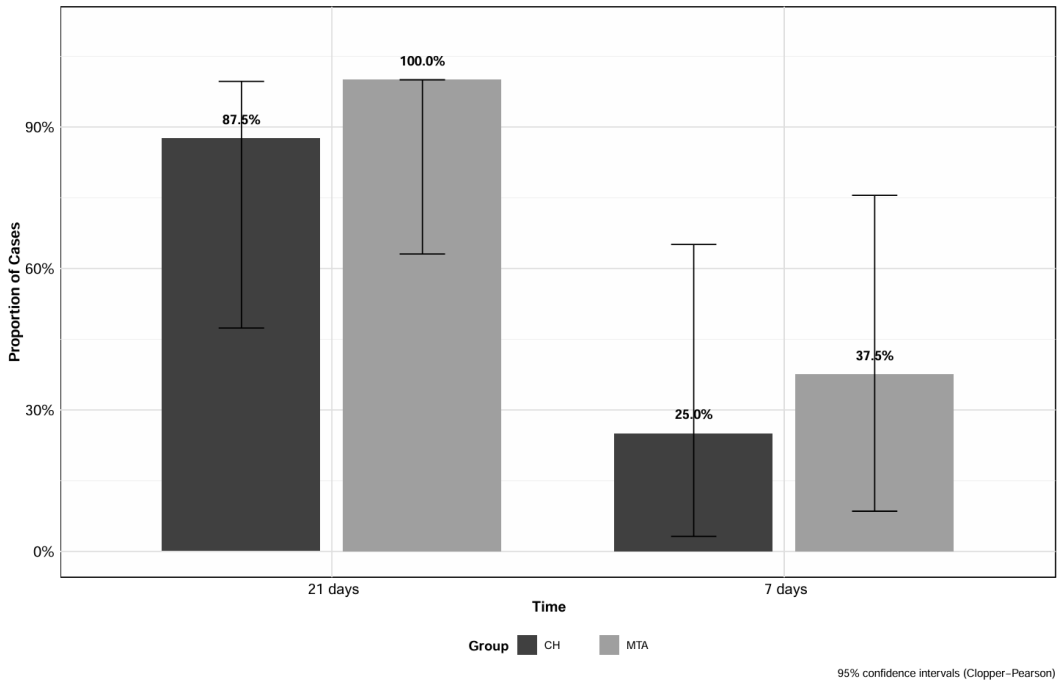

Figure 1. Temporal Evolution of Granulation Tissue Formation.

At 21 days, both groups exhibited a statistically significant increase in granulation tissue formation (McNemar's test: Calcium Hydroxide p=0.046, MTA p=0.025), reaching 87.5% (7/8; 95% CI: 47.3–99.7%) in the Calcium Hydroxide group and 100% (8/8; 95% CI: 63.1–100.0%) in the MTA group. Although no intergroup differences were detected at any time point (p=0.999 at 21 days), the MTA group stood out for achieving complete resolution (100%) of granulation tissue formation.

DISCUSSION

Inflammatory Cellular Response

The results of this study, which evaluate the cellular inflammatory response to Calcium Hydroxide and Mineral Trioxide Aggregate (MTA), reveal no significant differences both within and between groups, despite a trend toward inflammation reduction in both treatments. These findings align with previous studies that emphasize the biocompatibility of both materials, though with some nuances. For instance, PARIROKH & TORABINEJAD (2010) reported that MTA induces a transient inflammatory response, followed by rapid tissue regeneration, which could explain the trend observed in our study toward reduced inflammation (effect size r = -0.45). However, the lack of statistical significance ($\alpha = 0.05$) highlights that differences in inflammatory response between materials may be subtle and require larger sample sizes for detection (CAMILLERI et al. 2013).

The similarity in inflammatory profiles between groups ($p = 1.000$ in the first evaluation; $p = 0.587$ in the second) supports the hypothesis that both materials modulate inflammation comparably in early stages. This contrasts with studies such as JANANI et al. (2020), where MTA showed a significant reduction in pro-inflammatory mediators compared to Calcium Hydroxide. This discrepancy could be attributed to methodological differences, such as the use of animal versus human models or the evaluation of specific biomarkers not included in our study design. The limited statistical power (n = 8 per group) emerges as a critical limitation, a common issue in preclinical studies with small sample sizes (FESTING & ALTMAN 2002). This factor may mask clinically relevant differences, especially in ordinal variables like inflammation levels, where individual variability is high.

Additionally, the negative effect sizes ($r = -0.42$ to -0.45), though moderate, suggest that the direction of inflammatory reduction warrants attention in future research. As proposed by FAUL et al. (2007), an effect size of $|r| > 0.5$ is considered large, indicating that our results may reflect a real but underestimated trend. Taken together, these findings underscore the need for studies with greater statistical power, ideally using sample sizes calculated a priori through power analysis, as recommended by BUTTON et al. (2013). Additionally, the incorporation of complementary techniques (histomorphometry, cytokine quantification) would allow for a more precise characterization of the inflammatory response, correlating TNF-α levels with histological severity (CHEN et al. 2024). Although our data do not support the superiority of MTA, its non-inferior inflammatory profile compared to Calcium Hydroxide, combined with its effectiveness in other parameters (granulation tissue formation), positions it as a viable alternative in clinical settings that prioritize comprehensive tissue repair.

Presence of Necrosis Areas

The incidence of necrosis in tissues treated with Calcium Hydroxide and Mineral Trioxide Aggregate (MTA) revealed no statistically significant differences between the groups at the evaluated time points. However, the observed trends warrant a critical interpretation in light of existing literature. The complete elimination of necrosis in the MTA group by the end of the follow-up period (0/8) contrasts with previous studies highlighting its ability to promote tissue regeneration through the inhibition of apoptotic mediators, such as caspase-3, and the stimulation of vascular growth factors (PARIROKH & TORABINEJAD 2010). Although the analysis did not reach statistical significance ($p=0.125$), the observed effect size ($\phi=0.42$) suggests a clinically relevant reduction, aligning with the findings of CAMILLERI et al. (2013), who reported a lower expression of TNF-α in tissues treated with MTA compared to Calcium Hydroxide.

The lack of intergroup differences ($p=0.478$ at 21 days) could be attributed to the limited statistical power ($n=8$ per group), compounded by wide confidence intervals. In fact, the absence of necrosis in the MTA group at 21 days aligns with studies such as TANOMARU-FILHO et al. (2006), which attribute this effect to its ability to maintain a prolonged alkaline pH, thereby inhibiting bacterial proliferation and promoting tissue repair. The non-significant reduction in necrosis in the Calcium Hydroxide group (62.5% to 25.0%; $p=0.683$) may be explained by its initial antibacterial effect, although its inability to sustain a long-term bioactive environment (MOHAMMADI et al. 2012). In contrast, MTA, by inducing a more modulated and persistent inflammatory response, could facilitate a more efficient transition into the repair phase, as observed in bone regeneration models (CHEN et al. 2024).

These findings emphasize the importance of interpreting results beyond statistical significance, incorporating effect sizes and confidence intervals (FAUL et al. 2007). While the data do not confirm the superiority of MTA, its necrosis-free profile at the end of the study, along with its effectiveness in other parameters (granulation tissue formation), supports its use in clinical scenarios that prioritize the prevention of necrotic complications. Future studies should increase sample size, conduct detailed histopathological analyses, and evaluate specific biomarkers (IL-1β, TGF-β) to elucidate the underlying mechanisms of these trends, as recommended by the ARRIVE 2.0 guidelines for enhancing rigor in preclinical research (PERCIE DU SERT et al. 2020).

Granulation tissue formation

The formation of granulation tissue following the use of Calcium Hydroxide and Mineral Trioxide Aggregate (MTA) revealed two key findings: (1) the absence of significant differences between groups at both evaluated time points, and (2) a significant temporal improvement in granulation formation, particularly pronounced in the MTA group, which achieved complete resolution (100%) at 21 days. Although the lack of intergroup differences ($p=0.999$) could initially be attributed to the equivalence in the efficacy of both materials, the observed trend toward a faster and more complete resolution with MTA warrants a critical analysis in the context of existing literature. The absence of significant differences at seven days (Calcium Hydroxide: 25.0% vs. MTA: 37.5%) aligns with studies attributing biocompatible properties to both materials, enabling them to modulate early inflammation and promote granulation, though

through different mechanisms. MTA induces a sustained alkaline environment, which favors stem cell activity and angiogenesis (CAMILLERI et al. 2013).

In contrast, Calcium Hydroxide, while effective in its initial antibacterial action, can cause pH fluctuations that may delay the proliferative phase (MOHAMMADI et al. 2012). This divergence in mechanisms of action could explain the numerical trend in favor of MTA, even though it did not reach statistical significance in this study. The significant increase in granulation formation at 21 days in both groups ($p=0.046$ for Calcium Hydroxide; $p=0.025$ for MTA) reflects an expected tissue repair process, but the complete resolution (100%) in the MTA group stands out compared to 87.5% in the Calcium Hydroxide group. This finding aligns with studies such as JOSHI et al. (2020), which reported that MTA promotes a more efficient transition from the inflammatory to the reparative phase, mediated by the regulation of anti-inflammatory cytokines such as TGF-β. Although the intergroup difference was not significant ($p=0.999$), the absence of residual cases in the MTA group suggests a potential advantage of the profile clinically, supported by its ability to maintain a stable microenvironment.

CONCLUSION

Although no statistically significant differences were found between Calcium Hydroxide and MTA in inflammatory response or granulation tissue formation, MTA demonstrated a more promising profile by completely eliminating necrosis (0% at 21 days) and achieving 100% resolution in granulation tissue formation. These trends, supported by moderate effect sizes, suggest a biological advantage of MTA in advanced repair phases, positioning it as a preferred option in clinical settings seeking optimal outcomes.

The absence of residual necrosis and complete granulation resolution with MTA highlight its potential for clinical applications prioritizing complication prevention and comprehensive tissue regeneration. Although Calcium Hydroxide remains a viable alternative, the findings support preferential use of MTA in complex scenarios, especially when minimizing long-term risks is a priority.

AUTHOR CONTRIBUTIONS

Conceptualization, methodology, and formal analysis**, Williams Olivera-Acuña, Daniel Felen Hinostroza and Jane Hospinal P. Escajadillo**; software and validation**, Jordan Ninahuanca;** investigation**, Jorge Calderón Fernández**; resources and data curation**, Jordan Ninahuanca**; writing-original draft preparation**, James Anticona Gonzales**; writing-review and editing**, Edwin Tovar Sedano**. All authors have read and agreed to the published version of the manuscript.

FUNDING

This work was supported by the Universidad Peruana los Andes.

INFORMED CONSENT STATEMENT

Not applicable because this study did not involve humans.

DATA AVAILABILITY STATEMENT

The data can be made available upon request by emailing the corresponding author.

ACKNOWLEDGEMENTS

We would like to thank the staff of the Clinics and Universidad Peruana Los Andes for the facilities provided.

CONFLICTS OF INTEREST

There is no conflict of interest for any of the authors.

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