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ORIGINAL ARTICLE

Biomimetic hydroxyapatite paste for molar–incisor hypomineralization: A randomized clinical trial

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Abstract

Objectives: The aim of this randomized clinical trial was to evaluate the desensitizing and remineralizing effect of a new zinc-hydroxyapatite-based paste in sites affected by molar–incisor hypomineralization (MIH), by assessing dental sensitivity, tooth wear, and periodontal indexes.

Materials and Methods: Twenty-five patients with presence of 1 enamel demineralization of permanent molars and incisors in two different quadrants were recruited. After professional dental hygiene, a domiciliary hydroxyapatite-based paste was assigned and recommended to be applied on 2 MIH teeth in one random quadrant (test group), while the 2 contralateral MIH teeth did not undergo paste application (control group). The following primary outcomes were assessed: Plaque Control Record (PCR), Bleeding Index (BI), MIH Treatment Need Index (MIH-TNI), and Schiff Air Index (SAI).

Results: No significant inter- and intragroup differences were found for PI and BI, except for both intragroup T0–T1. For MIH-TNI, significant intergroup differences were detectable in the test group after 9 months of treatment. For SAI values, no significant differences were found in the control group, while in the test group, significant lower values were found after 1 and 3 months since baseline, respectively.

Conclusions and Clinical Relevance: Biomimetic zinc-hydroxyapatite showed a desensitizing effect when used to treat MIH.

KEYWORDS
biomimetic hydroxyapatite, demineralization, enamel, molar–incisor hypomineralization, paste, randomized clinical trial

1 | INTRODUCTION

Molar–incisor hypomineralization (MIH) is a systemic origin defect present in at least one permanent first molar, in association or not with the permanent incisors (Weerheijm et al., 2001). However, MIH can also show up on other dental elements such as permanent second molars, permanent canines, and deciduous molars. Hypomineralized second primary molars (HSPMs) can be considered a predictor of MIH (Giuca et al., 2020). The defect will be more severe as the number of affected molars and incisors increases (Reis et al., 2021).

The mean global prevalence of MIH is reported to be 14.2% (18% in South America and 10.9% in Africa). MIH prevalence in children

≤10 years of age is higher (15.1%) than in children >10 years of age (12.1%; Dave & Taylor, 2018).

Multiple factors may contribute to the onset of MIH; however, their association is still unclear (Sundfeld et al., 2020). Recent studies have proposed a genetic predisposition to MIH in association with one or more etiological factors, identifying some genetic variants related to amelogenesis such as ENAM, AMELX, and MMP20 genes and immune response-related genes (Lygidakis et al., 2021). In addition, it has been suggested that epigenetics may regulate systemic factors influencing the function of enamel proteins involved in MIH (Hočevar et al., 2020).

The early use of antibiotics, in particular amoxicillin, during the first years of life could modify the immunological and inflammatory response of the child by altering several growth factors levels expressed by ameloblasts, thus interfering in amelogenesis and contributing to the development of MIH (Giuca et al., 2018).

MIH can be associated with etiologic factors such as prenatal factors (Lee et al., 2020), perinatal factors (Butera, Maiorani, et al., 2021), fever in early childhood, when associated with other symptoms such as chest infections and/or ear infections (Ghanim et al., 2013). In addition, viral infections (Sönmez et al., 2013), kidney disease (Kusku et al., 2008), gastrointestinal disease (Sönmez et al., 2013), and upper airway diseases (Ghanim et al., 2013) have been implicated in the etiology of MIH.

Clinically, MIH shows up as opacities at the occlusal and vestibular surfaces of the crown of the dental elements. The color may range from white to yellow or brownish, due to alterations in hydroxyapatite crystals, enamel prisms, and interprismatic space, and the size may be negligible (if <1 mm) or can involve most of the crown, increasing the risk of fracture and dentinal exposure with the onset of carious lesions (Bezamat et al., 2021; Elhennawy et al., 2017). Differential diagnosis must be performed to rule out the presence of other conditions such as amelogenesis imperfecta, enamel hypoplasia, fluorosis, and white spots (Ghanim et al., 2017).

Treatment approaches for MIH can range from early diagnosis, prevention of caries, and timely administration of remineralizing products (William et al., 2006) for the management of hypersensitivity or pain, restorative treatments, and finally, premature extraction of affected tooth elements (Weerheijm, 2004). The treatment plan for MIH should include a short- and long-term approach, including frequent follow-ups over time (Rodd et al., 2021).

MIH treatments can be fluoride varnish applications or resin infiltration (Nogueira et al., 2021), arginine pastes (Da Cunha Coelho et al., 2019), glass ionomer cements restorations (Fragelli et al., 2015), casein phosphopeptide and amorphous calcium phosphate CPP-ACP (Pasini et al., 2018), resin or silver diamine fluoride adhesive and sealant application (Ballikaya et al., 2022), composite restorations (Lygidakis et al., 2021), indirect restorations (Davidovich et al., 2020), stainless steel crowns (Wadia, 2021), and, finally, tooth extraction (Elhennawy & Schwendicke, 2016).

The aim of the present study is to evaluate the desensitizing and remineralizing effect of a new hydroxyapatite-based paste use in MIH-affected sites, compared to MIH non-treated control teeth, by assessing Plaque Control Record (PCR) and Bleeding Index (BI), MIH Treatment Need Index (MIH-TNI), and dental sensitivity through Schiff Air Index (SAI), after 9 months of treatment.

The first null hypothesis is that there are no differences between oral hygiene status of the patients in terms of PCR and BI. The second null hypothesis is that there are no differences between the two groups as regard MIH-TNI. The third null hypothesis of the study is that there are no differences between the two groups for hypersensitivity assessed by SAI.

2 | MATERIALS AND METHODS

2.1 | Study design

This was a split-mouth randomized controlled clinical trial. The study protocol was approved by the Unit Internal Review Board (approval 2021–0303) and registered on clinicaltrials.gov (registration number: NCT04808180).

The study was conducted according to the Principles of the Declaration of Helsinki on experimentation involving human subjects. CONSORT statement was followed for this trial. Written informed consent was obtained from parents/guardians of the subjects involved.

2.2 | Participants

Patients' enrollment, data collection, and statistical analyses were conducted at the Unit of Dental Hygiene, Section of Dentistry, Department of Clinical, Surgical, Diagnostic and Pediatric Sciences, University of Pavia, 27100 Pavia, Italy.

2.2.1 | Eligibility criteria for participants

Inclusion criteria:

- presence of at least 2 MIH teeth (permanent molars and incisors) responding to the following scores of Steffen classification MIH-TNI (Steffen et al., 2017): (1) MIH without hypersensitivity, without defect; (2) MIH without hypersensitivity, with defect; (3) MIH with hypersensitivity, without defect; (4) MIH with hypersensitivity, with defect; the same number of MIH teeth had to belong to contralateral quadrants
- among teeth with enamel defects were chosen those with <1/3 of defect extension (scores 2a and 4a)
- good general health (absence of systemic diseases)

Exclusion criteria:

- patients undergoing orthodontic therapy
- patients taking drugs (chlorhexidine-based gels and mouthwashes)

2.3 | Interventions

2.3.1 | Clinical procedure

At the baseline (T0), patients were visited and instructed to proper oral hygiene domiciliary procedures. Professional dental hygiene was performed with a piezoelectric instrument (Multipiezo; Mectron S.p.a) and Gracey curettes (Hu-Friedy). For all the duration of the study, patients had to perform oral hygiene procedures with Oral-B Kids® electric toothbrush (Procter & Gamble) and a hydroxyapatite-free toothpaste twice a day. Instructions were given to patients and their guardians for the use of Biorepair Desensitizing Enamel-Repair Shock Treatment® paste. Its application had to be performed for one MIH molar in one random quadrant (test group), while the contralateral MIH molar did not undergo paste application (control group). The same protocol was applied for MIH incisors. The application was performed filling a bite given by the manufacturer in the corresponding tooth side; the bite was kept for 10 min inside the oral cavity and the procedure was repeated for 1 week per month until the end of the study (9 months in total; Scribante et al., 2020).

The composition of the paste is shown in Table 1.

2.4 | Outcome measures

The following primary outcomes were assessed: Plaque Control Record (PCR—percentage of sites with plaque with respect of total dental sites; O'Leary et al., 1972), Bleeding Index (BI—percentage of sites showing bleeding with respect of total probed sites; Butler et al., 2011), MIH Treatment Need Index (MIH-TNI from 0 to 4, according to a combination of absence/presence of sensitivity, presence/absence of enamel defect and extension of the defect; Steffen et al., 2017) and Schiff Air Index (SAI—scores from 0 to 3 after evaporative air stimulus; Pepelassi et al., 2015).

The clinical parameters were collected at the baseline (T0) and after 1 month (T1), 2 (T2), 3 (T3), 6 (T6), and 9 months (T9) by means of a manual periodontal probe (UNC probe 15; Hu-Friedy, Chicago, IL, USA) and visual examination.

2.5 | Sample size calculation

Sample size calculation (Alpha 0.05; Power = 95%) for two independent study groups and a continuous primary endpoint was performed concerning the variable "Schiff Air Index".

The following mathematical formula was used for sample size calculation:

$$\text{Sample size} = \frac{Z^2_{\left(1-\frac{\alpha}{2}\right)} p(1-p)}{d^2}$$

where $z_{\left(1-\frac{\alpha}{2}\right)}$ is the standard normal variate corresponding to 1.96 at 5% type 1 error, p is the expected proportion in population expressed as decimal and based on previous studies, and finally, d is the confidence level decided by the researcher and expressed as decimal too.

An expected value of 1.19 was hypothesized, and the expected difference between the means was supposed to be 0.91 with a standard deviation of 0.9 (Bekes et al., 2017); therefore, 50 teeth were required per group; and therefore, 25 patients had to be enrolled for the study.

2.6 | Randomization

Using a block randomization table, the data analyst generated a randomization sequence, considering a permuted block of 25 participants. Based on previously prepared sequentially numbered, opaque, sealed envelopes (SNOSE), the care provider randomized the teeth of the patients and instructed patients and their guardians to domiciliary oral hygiene procedures.

2.7 | Blinding

For the domiciliary oral hygiene procedures, the paste was concealed. The care provider was not blinded, together with participants, while investigator and outcome assessor were blinded. The data analyst was blinded for the allocation and for the data acquisition.

2.8 | Statistical analysis

Statistical analysis of data was performed with R Software (R version 3.1.3, R Development Core Team, R Foundation for Statistical Computing, Wien, Austria). For each group and variable, descriptive statistics (mean and standard deviation) were calculated. PCR and BI were calculated in percentages. Data normality was assessed with Kolmogorov–Smirnov test. For each variable, intragroup comparisons were performed using ANOVA test with Tukey test as post hoc. For intergroup comparisons, Student's *t*-test was performed. A

TABLE 1 Composition of the tested paste

Paste	Manufacturer	Composition
Biorepair® Desensitizing Enamel-Repair Shock Treatment®	Coswell S.p.A., Funo di Argelato, BO, Italy	Aqua, ZincHydroxyapatite (30% w/v), HydratedSilica, Silica, Sodium Myristoyl Sarcosinate, Sodium Methyl Cocoyl Taurate, Sodium Bicarbonate, Aroma, Sodium Saccharin, Phenoxyethanol, Benzyl Alcohol, Sodium Benzoate, Citric Acid, Menthol.

letter-based comparison has been adopted to detect significant differences (Piepho, 2004).

MIH-TNI and SAI were calculated as pure value; chi-square test and McNemar test were used to calculate intragroup and intergroup differences.

Significance was predetermined for $p < 0.05$ for all statistical tests.

3 | RESULTS

3.1 | Participants, demographic characteristics, and recruitment

After screening, 25 patients fulfilling the inclusion criteria were recruited. None of them withdraw from the study. Recruitment started in March 2021 and ended in May 2021. The study ended in February 2022. No harms related to any of the two interventions were recorded.

The demographic characteristics of the study sample are shown in Table 2. 76% were female, and the overall mean age was 8.6 ± 1.22 years (range, 6–10 years). Figure 1 shows the flow chart of the study.

TABLE 2 Baseline demographic characteristics

	Total	Males	Females
n	25 (100%)	6 (24%)	19 (76%)
Age (years ± SD)	8.6 ± 1.22	8.83 ± 1.6	8.53 ± 1.22

Fifty teeth were included in the study, 32 incisors (81.3% upper and 18.7% lower) and 18 molars 88.9% upper and 11.1% lower (Table 3).

3.2 | Outcomes

For both groups analyzing PCR values (Table 4), T0 differs from all other periods ($p < 0.05$). For the control group, no statistical differences were verified for all intragroup periods, except when compared T1–T9. Accordingly, for the test group, no statistical differences were verified for intragroup comparisons except for T1–T9 ($p < 0.05$). No significant intergroup differences occurred ($p > 0.05$).

As regard BI values (Table 5), T0 differs from all other periods ($p < 0.05$). For the control group, a significant difference occurred for T1–T9 time frames ($p < 0.05$). For the trial group, no significant differences were found between T1–T9 ($p > 0.05$). Moreover, no significant intergroup comparisons were found ($p > 0.05$).

Regarding MIH-TNI values (Table 6), a significant higher frequency of scores 3 and 4a was found at T0 for both the Groups ($p < 0.05$). A significant higher frequency of scores 3 and 4a was found in the control group at T9 ($p < 0.05$), instead of the trial group, with a significant higher frequency of scores 1 and 2 ($p < 0.05$).

For SAI values (Table 7), a significant higher frequency of score 3 was found at T0 for both the Groups ($p < 0.05$). A significant higher frequency of scores 1 and 2 was found in the control group at T9 ($p < 0.05$). In the trial group, a significant higher frequency of score 0 was found ($p < 0.05$).

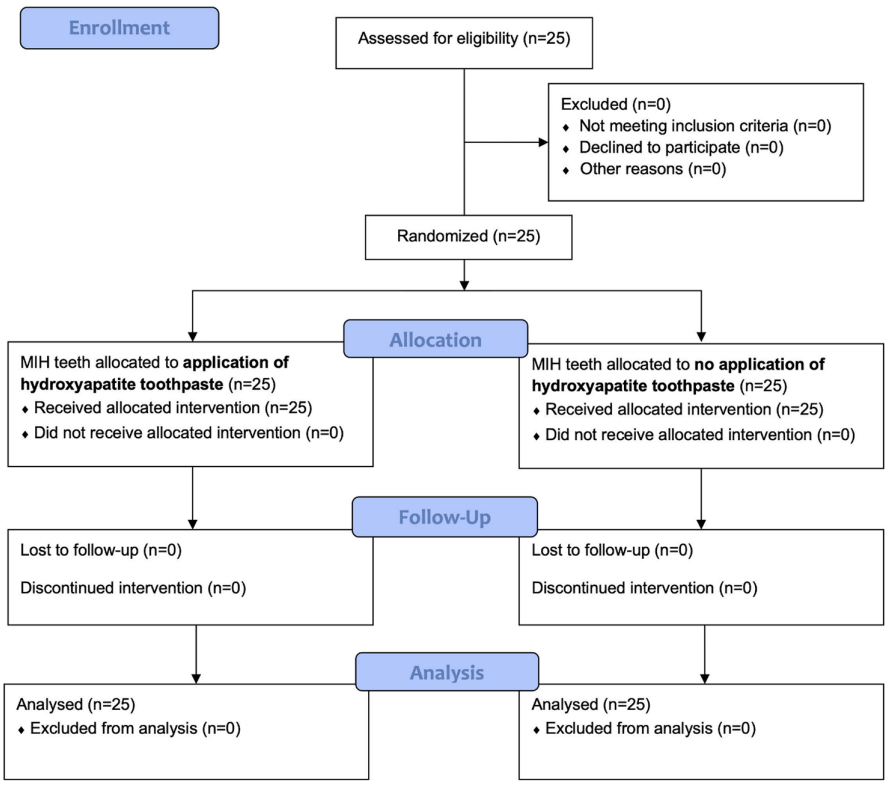

FIGURE 1 CONSORT flow chart.

TABLE 3 Number of MIH affected teeth per type

Teeth	Total	Upper	Lower
Molars	18	16	2
1.6	8		
2.6	8		
3.6	1		
4.6	1		
Incisors	32	26	6
1.1	8		
1.2	5		
2.1	8		
2.2	5		
4.1	2		
4.2	1		
3.1	2		
3.2	1	6	
Total	50	42	8

4 | DISCUSSION

Molar–incisor hypomineralization (MIH) is a delimited qualitative developmental defect leading to several dental complications, like hypersensitive teeth, increased susceptibility to caries, impaired chewing due to rapid tooth loss, and aesthetic defects; this condition may significantly affect patients' quality of life, so a correct diagnosis and treatment plan are strictly required (Solinas et al., 2021). To date, different clinical trials have been conducted to evaluate the efficacy of remineralizing agents like fluoride in different formulations, casein-based mousses and amorphous calcium phosphate (CPP-ACP), and hydroxyapatite (Bakkal et al., 2017; Biondi et al., 2017; Restrepo et al., 2016). This approach is based on the concept that increasing the mineral component in teeth affected by MIH can lead to an improvement of the physical structure of the enamel together with its aesthetic appearance, and to the decrease of dental sensitivity (Solinas et al., 2021).

Fluoride products are among the most popular devices for remineralizing purposes. Fluoride ions interact with dental hydroxyapatite crystals, forming the less insoluble fluoridated hydroxyapatite or fluorapatite, which are more resistant to the acid erosion (Lussi et al., 2019). However, the efficacy of fluoride is limited to a partial substitution of the hydroxyl groups with fluoride ions in natural hydroxyapatite, with no deposition of an additional mineral content. Conversely, biomimetic hydroxyapatite even leads to a mineral deposition, thus forming a real coating on enamel and dental surfaces (Lelli et al., 2014) together with composite resins (Butera, Pascadopoli, et al., 2021). Hydroxyapatite toothpastes have been shown superior remineralizing effect to fluoridated toothpastes, in post orthodontic debonding patients (Verma & Muthuswamy, 2021). Furthermore, fluoride may be linked to a risk of toxicity in case of high dosage intake, with fluorosis in children and bone diseases in the elderly (Pajor et al., 2019).

On the basis of these considerations, the aim of the present study was to evaluate the desensitizing and remineralizing effect of a new hydroxyapatite-based paste in MIH-affected sites, by assessing dental sensitivity, tooth wear, and periodontal indexes, after 9 months of treatment.

The first null hypothesis was accepted. In fact, for PCR values (Table 3), no statistically significant inter- and intragroup differences were found, except for both intragroup T0-T1 ($p < 0.05$). The same happened for BI (Table 4). Several studies reviewed the relationship between MIH and oral hygiene, showing that disease severity was related to increased hypersensitivity and poorer oral hygiene: Teeth with MIH showed increased sensitivity to cold stimuli and surface roughness, lowering the ability to maintain proper oral hygiene and favoring the onset of caries processes risk (Ebel et al., 2018). Recent study by Fütterer et al. (2020) showed that the improvement of oral hygiene in patients with MIH through the application of fluoride varnish, fissure sealant, filling, and stainless-steel crowns ranged according to the severity of the disease: Patients with high-severity MIH had significantly improved oral hygiene after therapy; in contrast, patients with low-severity or medium-severity MIH did not show great changes in oral hygiene.

The second null hypothesis was rejected. In fact, as regard MIH-TNI values (Table 5), significant intergroup differences were detectable in the test group after 9 months of treatment. Intragroup differences were not significant in the control group, whereas in the test group, a gradual decrease was found, with a significant difference between T0 and T1. Previous in vitro research (Scribante et al., 2020) tested biomimetic zinc-hydroxyapatite using the same application protocol adopted in the present research. Results showed a significant reduction of demineralization area extension. Accordingly, in the present report, MIH-TNI seems to be affected by the tested paste with a decrease of the index among the time frames of the trial group and a significant difference at T9. The tested paste could have played a role in the remineralization of enamel surface. However, this hypothesis that cannot be supported directly by the present results. In fact, further research is needed, involving scanning electron microscopy evaluation, ultrastructural analysis, and future clinical trials.

Several previous clinical trials showed the remineralizing effect of different agents on MIH-affected dental elements, increasing the inorganic mineral component and improving both mechanical resistance to stress and aesthetic aspect: 4% titanium tetrafluoride varnish (Mendonça et al., 2022), casein phosphopeptide and amorphous calcium phosphate (CPP-ACP) and CPP-ACP with fluoride (CPP-ACFP; Olgen et al., 2022), 8% arginine and calcium carbonate with 14,450 ppm fluoride toothpastes (Bekes et al., 2017), and 10% microcrystalline hydroxyapatite remineralizing toothpastes (Ehlers et al., 2021).

The third null hypothesis was partially rejected. In fact, in the control group SAI frequencies (Table 7) resulted not significant, while in the trial group significant lower frequencies were found after 1 and 3 months since the baseline, respectively. Significant intergroup differences were also found comparing the end of the follow-up in the

TABLE 4 PCR scores

Group	Time	Mean	SD	Intragroup (T0–T9)	Intergroup (control-test)
Control ($N = 25$)	T0	73.40	21.10	A	a
	T1	42.00	21.36	B	a
	T2	35.65	13.76	B, C	a
	T3	30.00	12.25	B, C	a
	T6	25.45	12.43	B, C	a
	T9	18.50	6.51	C	a
Test ($N = 25$)	T0	70.60	24.93	A	a
	T1	36.40	23.61	B	a
	T2	30.43	19.36	B, C	a
	T3	24.09	17.09	B, C	a
	T6	21.36	17.20	B, C	a
	T9	13.75	9.44	C	a

Note: Means with the same letters are not significantly different ($p > 0.05$).

TABLE 5 BI scores

Group	Time	Mean	SD	Intragroup (T0–T9)	Intergroup (control-test)
Control ($N = 25$)	T0	38.84	20.23	A	a
	T1	24.68	14.09	B	a
	T2	20.17	7.66	B, C	a
	T3	16.05	5.05	B, C	a
	T6	13.05	5.22	C	a
	T9	11.00	3.84	C	a
Test ($N = 25$)	T0	30.28	20.59	A	a
	T1	17.44	11.24	B	a
	T2	13.17	8.57	B	a
	T3	10.00	6.55	B	a
	T6	9.27	7.42	B	a
	T9	7.78	4.61	B	a

Note: Means with the same letters are not significantly different ($p > 0.05$).

control group and the third month in the trial one. Accordingly, the use of the experimental paste based on zinc-hydroxyapatite resulted to be effective in decreasing dental hypersensitivity, a frequent complaint in children affected by MIH. Previous studies have tried to evaluate the benefit of hydroxyapatite on hypersensitivity caused by MIH. In particular, Ehlers et al. (2021) conducted a randomized controlled trial aimed to evaluate the non-inferiority of a toothpaste containing microcrystalline hydroxyapatite compared to amine fluoride in hypersensitivity relief of children with MIH. In this study, the primary outcome was pain sensation in response to tactile stimulus (Wong-Baker FACES Pain Rating Scale) 56 days after randomization. The results of the trial did not show the non-inferiority in hypersensitivity relief of the toothpaste containing hydroxyapatite compared to amine fluoride; however, the hydroxyapatite group tended to be less hypersensitive. In a pediatric sample, it was demonstrated that worse oral health quality of life is associated with MIH as regard oral symptoms (Portella et al., 2019) Therefore, a reduction in hypersensitivity with domiciliary treatment could surely bring to an improvement in MIH children's quality of life, as it is one of the major concerns during both daily life and dental treatment (Mendonça et al., 2022).

In their recent case report, Solinas et al. (2021) described the case of a 4-years-old female patient complaining of hypersensitivity to heat and cold, even during normal breathing. The patient presented hypomineralization of the enamel on permanent upper and lower incisors and left lower first permanent molar. The patient underwent home remineralization treatment with a biomimetic nano-hydroxyapatite mousse for 6 months, at alternating periods, with parents' compliance. Painful perception of the lesions was measured by the Wong-Baker rating scale. 1 year after diagnosis, all affected teeth had no further painful symptoms.

Several critical points emerged during the preparation, conduct, and processing of the data for this study. It would be interesting to compare the remineralizing and desensitizing effect of the tested

TABLE 6 MIH-TNI scores

Group	Time	Scores			
		1 (%)	2a (%)	3 (%)	4a (%)
Control (*N* = 25)	T0	2 (8)	4 (16)	11 (44)	8 (32)
	T1	2 (8)	5 (20)	11 (44)	7 (28)
	T2	2 (8)	5 (20)	11 (44)	7 (28)
	T3	3 (12)	5 (20)	10 (40)	7 (28)
	T6	3 (12)	6 (24)	10 (40)	6 (24)
	T9	4 (16)	4 (16)	9 (36)	6 (24)
Test (*N* = 25)	T0	0 (0)	2 (8)	15 (60)	8 (32)
	T1	0 (0)	5 (20)	15 (60)	5 (20)
	T2	3 (12)	7 (28)	12 (48)	3 (12)
	T3	5 (20)	10 (40)	10 (40)	0 (0)
	T6	8 (32)	10 (40)	7 (28)	0 (0)
	T9	11 (44)	10 (40)	4 (16)	0 (0)

TABLE 7 SAI scores

Group	Time	Scores			
		0 (%)	1 (%)	2 (%)	3 (%)
Control (*N* = 25)	T0	3 (12)	2 (8)	6 (24)	14 (56)
	T1	4 (16)	1 (4)	13 (52)	7 (28)
	T2	4 (16)	1 (4)	16 (64)	4 (16)
	T3	4 (16)	2 (8)	18 (72)	1 (4)
	T6	4 (16)	9 (36)	12 (48)	0 (0)
	T9	4 (16)	11 (44)	10 (40)	0 (0)
Test (*N* = 25)	T0	2 (8)	4 (16)	6 (24)	13 (52)
	T1	6 (24)	11 (44)	6 (24)	2 (8)
	T2	12 (48)	9 (36)	2 (8)	2 (8)
	T3	17 (68)	8 (32)	0 (0)	0 (0)
	T6	21 (84)	4 (16)	0 (0)	0 (0)
	T9	21 (84)	4 (16)	0 (0)	0 (0)

paste compared to another biomimetic hydroxyapatite paste or fluoride varnishes, casein phosphopeptide and amorphous calcium phosphate (CPP-ACP), and arginine with calcium carbonate and fluoride pastes.

The tested paste was applied 1 week per month for 9 months, as indicated by the manufacturer; however, it would be interesting to evaluate the remineralizing and desensitizing effects in prolonged application time periods. Other evaluations of MIH remineralization sites could include ICDAS criteria and quantitative light-induced fluorescence (QLF). In addition, it would be interesting to evaluate enamel ultrastructure and mineral contents by means of field emission scanning microscope (FESEM) and energy-dispersive spectroscopy (EDS).

Due to the lower reliability of visual indexes in the evaluation of demineralized enamel compared to microscopic analysis, it would be interesting to evaluate the remineralizing effect of this tested paste on exfoliating deciduous teeth or on permanent elements to be extracted through microscopic evaluations.

The present report evaluated MIH defects with extension <1/3 of enamel. The performing of randomized clinical trials enrolling MIH teeth with higher TNI scores would be desirable, as the effects of the tested paste were assessed only for teeth with small defect extension.

As the application of the paste was performed at home, bias in the procedure could have been present due to patients' and parents' compliance in following the study protocol.

Further studies are needed to evaluate whether one or more excipients contained in the paste employed in the present study may have influenced the results obtained and to assess the superiority of the application of hydroxyapatite, alone or in combination with other treatments, for the management of MIH, despite the high hydroxyapatite content.

5 | CONCLUSIONS

Molar-incisor hypomineralization (MIH) results to be a more and more frequent condition encountered in clinical practice. In the present randomized clinical trial, biomimetic zinc-hydroxyapatite applied in form of paste showed a desensitizing effect, along with a slight reduction of MIH-TNI scores, in teeth affected from mild MIH defects. Considering the ease of use and the short application time of the tested paste, its domiciliary use for MIH patients is recommended to reduce dental sensitivity and improve enamel integrity.

AUTHOR CONTRIBUTIONS

Andrea Butera: Conceptualization; funding acquisition; project administration; supervision; validation. **Maurizio Pascadopoli:** Resources; validation; visualization; writing – original draft. **Matteo Pellegrini:** Resources; writing – original draft. **Benedetto Trapani:** Data curation; investigation. **Simone Gallo:** Supervision; visualization; writing – original draft. **Monica Radu:** Data curation; investigation. **Andrea Scribante:** Conceptualization; data curation; formal analysis; methodology; project administration; software; supervision; validation; visualization; writing – review and editing.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

PEER REVIEW

The peer review history for this article is available at https://publons.com/publon/10.1111/odi.14388.

DATA AVAILABILITY STATEMENT

The data sets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

INFORMED CONSENT

Informed consent was obtained from parents/guardians of all individual participants included in the study.

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REFERENCES

Bakkal, M., Abbasoglu, Z., & Kargul, B. (2017). The effect of casein phosphopeptide-amorphous calcium phosphate on molar-incisor hypomineralisation: A pilot study. *Oral Health & Preventive Dentistry*, *15*, 163–167. https://doi.org/10.3290/j.ohpd.a37928

Ballikaya, E., Ünverdi, G. E., & Cehreli, Z. C. (2022). Management of initial carious lesions of hypomineralized molars (MIH) with silver diamine fluoride or silver-modified atraumatic restorative treatment (SMART): 1-year results of a prospective, randomized clinical trial. *Clinical Oral Investigations*, *26*, 2197–2205. https://doi.org/10.1007/s00784-021-04236-5

Bekes, K., Heinzelmann, K., Lettner, S., & Schaller, H. G. (2017). Efficacy of desensitizing products containing 8% arginine and calcium carbonate for hypersensitivity relief in MIH-affected molars: An 8-week clinical study. *Clinical Oral Investigations*, *21*, 2311–2317.

Bezamat, M., Souza, J. F., Silva, F. M. F., Corrêa, E. G., Fatturi, A. L., Brancher, J. A., Carvalho, F. M., Cavallari, T., Bertolazo, L., Machado-Souza, C., Koruyucu, M., Bayram, M., Racic, A., Harrison, B. M., Sweat, Y. Y., Letra, A., Studen-Pavlovich, D., Seymen, F., Amendt, B., … Vieira, A. R. (2021). Gene-environment interaction in molar-incisor hypomineralization. *PLoS One*, *16*, e0241898. https://doi.org/10.1371/journal.pone.0241898

Biondi, A. M., Cortese, S. G., Babino, L., & Fridman, D. E. (2017). Comparison of mineral density in molar incisor hypomineralization applying fluoride varnishes and casein phosphopeptide-amorphous calcium phosphate. *Acta Odontológica Latinoamericana*, *30*, 118–123.

Butera, A., Maiorani, C., Morandini, A., Simonini, M., Morittu, S., Barbieri, S., Bruni, A., Sinesi, A., Ricci, M., Trombini, J., Aina, E., Piloni, D., Fusaro, B., Colnaghi, A., Pepe, E., Cimarossa, R., & Scribante, A. (2021). Assessment of genetical, pre, peri and post Natal risk factors of deciduous molar hypomineralization (DMH), hypomineralized second primary molar (HSPM) and molar incisor Hypomineralization (MIH): A narrative review. *Children*, *8*, 432. https://doi.org/10.3390/children8060432

Butera, A., Pascadopoli, M., Gallo, S., Lelli, M., Tarterini, F., Giglia, F., & Scribante, A. (2021). SEM/EDS evaluation of the mineral deposition on a polymeric composite resin of a toothpaste containing biomimetic Zn-carbonate hydroxyapatite (microRepair®) in Oral environment: A randomized clinical trial. *Polymers (Basel)*, *13*, 2740. https://doi.org/10.3390/polym13162740

Butler, A., Targett, D., & Bosma, M. L. (2011). Maintenance of gingival health–ameasurebasedonclinicalindices.*InternationalDentalJournal*, *61*, 28–32. https://doi.org/10.1111/j.1875-595X.2011.00046.x

Da Cunha Coelho, A. S. E., Mata, P. C. M., Lino, C. A., Macho, V. M. P., Pereira Areias, C. M. F. G., Peixoto Norton, A. P. M. A., & Macedo Augusto, A. P. C. (2019). Dental hypomineralization treatment: A systematic review. *Journal of Esthetic and Restorative Dentistry*, *31*, 26–39. https://doi.org/10.1111/jerd.12420

Dave, M., & Taylor, G. (2018). Global prevalence of molar incisor hypomineralisation. *Evidence-Based Dentistry*, *19*, 78–79. https://doi.org/10.1038/sj.ebd.6401324

Davidovich, E., Dagon, S., Tamari, I., Etinger, M., & Mijiritsky, E. (2020). An innovative treatment approach using digital workflow and CAD-CAM part 2: The restoration of molar incisor hypomineralization in children. *International Journal of Environmental Research and Public Health*, *17*, 1499. https://doi.org/10.3390/ijerph17051499

Ebel, M., Bekes, K., Klode, C., & Hirsch, C. (2018). The severity and degree of hypomineralisation in teeth and its influence on oral hygiene and caries prevalence in children. *International Journal of Paediatric Dentistry*, *28*, 648–657. https://doi.org/10.1111/ipd.12425

Ehlers, V., Reuter, A. K., Kehl, E.-B., Enax, J., Meyer, F., Schlecht, J., Schmidtmann, I., & Deschner, J. (2021). Efficacy of a toothpaste based on microcrystalline hydroxyapatite on children with hypersensitivity caused by MIH: A randomised controlled trial. *Oral Health & Preventive Dentistry*, *19*, 647–658. https://doi.org/10.3290/j.ohpd.b2403649

Elhennawy, K., Maton, D. J., Crombie, F., Zaslansky, P., Radlanski, R. J., Jost-Brinkmann, P.-G., & Schwendicke, F. (2017).

Structural, mechanical and chemical evaluation of molar-incisor hypomineralization-affected enamel: A systematic review. *Archives of Oral Biology*, *83*, 272–281. https://doi.org/10.1016/j.archoralbio.2017.08.008

Elhennawy, K., & Schwendicke, F. (2016). Managing molar–incisor hypomineralization: A systematic review. *Journal of Dentistry*, *55*, 16–24. https://doi.org/10.1016/j.jdent.2016.09.012

Fragelli, C. M. B., de Souza, J. F., Geremia, F., Cordeiro, R. C. L., & Santos-Pinto, L. (2015). Molar incisor hypomineralization (MIH): Conservative treatment management to restore affected teeth. *Brazilian Oral Research*, *29*, 1–7. https://doi.org/10.1590/1807-3107BOR-2015.vol29.0076

Fütterer, J., Ebel, M., Bekes, K., Klode, C., & Hirsch, C. (2020). Influence of customized therapy for molar incisor hypomineralization on children's oral hygiene and quality of life. *Clinical And Experimental Dental Research*, *6*, 33–43. https://doi.org/10.1002/cre2.245

Ghanim, A., Manton, D., Bailey, D., Mariño, R., & Morgan, M. (2013). Risk factors in the occurrence of molar–incisor hypomineralization amongst a group of Iraqi children. *International Journal of Paediatric Dentistry*, *23*, 197–206. https://doi.org/10.1111/j.1365-263X.2012.01244.x

Ghanim, A., Silva, M. J., Elfrink, M. E. C., Lygidakis, N. A., Mariño, R. J., Weerheijm, K. L., & Manton, D. J. (2017). Molar incisor hypomineralisation (MIH) training manual for clinical field surveys and practice. *European Archives of Paediatric Dentistry*, *18*, 225–242. https://doi.org/10.1007/s40368-017-0293-9

Giuca, M. R., Cappè, M., Carli, E., Lardani, L., & Pasini, M. (2018). Investigation of clinical characteristics and etiological factors in children with molar incisor hypomineralization. *International Journal Dentistry*, *2018*, 7584736. https://doi.org/10.1155/2018/7584736

Giuca, M. R., Lardani, L., Pasini, M., Beretta, M., Gallusi, G., & Campanella, V. (2020). State-of-the-art on MIH. Part 1: Definition and aepidemiology. *European Journal of Paediatric Dentistry*, *21*, 80–82. https://doi.org/10.23804/ejpd.2020.21.01.16

Hočevar, L., Kovač, J., Podkrajšek, K. T., Battelino, S., & Pavlič, A. (2020). The possible influence of genetic aetiological factors on molar-incisor hypomineralisation. *Archives of Oral Biology*, *118*, 104848. https://doi.org/10.1016/j.archoralbio.2020.104848

Kusku, O. O., Caglar, E., & Sandalli, N. (2008). The prevalence and aetiology of molar-incisor hypomineralisation in a group of children in Istanbul. *European Journal of Paediatric Dentistry*, *9*, 139–144.

Lee, D.-W., Kim, Y.-J., Kim, S. O., Choi, S. C., Kim, J., Lee, J. H., Kim, H. J., Shin, J., Lee, N.-Y., Kim, S.-M., Ra, J., Kim, J., & Yang, Y.-M. (2020). Factors associated with molar–incisor hypomineralization: A population-based case-control study. *Pediatric Dentistry*, *42*, 134–140.

Lelli, M., Putignano, A., Marchetti, M., Foltran, I., Mangani, F., Procaccini, M., Roveri, N., & Orsini, G. (2014). Remineralization and repair of enamel surface by biomimetic Zn-carbonate hydroxyapatite containing toothpaste: A comparative in vivo study. *Frontiers in Physiology*, *5*, 333. https://doi.org/10.3389/fphys.2014.00333

Lussi, A., Buzalaf, M. A. R., Duangthip, D., Anttonen, V., Ganss, C., João-Souza, S. H., Baumann, T., & Carvalho, T. S. (2019). The use of fluoride for the prevention of dental erosion and erosive tooth wear in children and adolescents. *European Archives of Paediatric Dentistry*, *20*, 517–527. https://doi.org/10.1007/s40368-019-00420-0

Lygidakis, N. A., Garot, E., Somani, C., Taylor, G. D., Rouas, P., & Wong, F. S. L. (2021). Best clinical practice guidance for clinicians dealing with children presenting with molar–incisor-hypomineralisation (MIH): An updated European Academy of Paediatric Dentistry policy document. *European Archives of Paediatric Dentistry*, *23*, 3–21. https://doi.org/10.1007/s40368-021-00668-5

Mendonça, F. L., Di Campli Regnault, F. G., Di Leone, C. C. L., Grizzo, I. C., Bisaia, A., Fragelli, C., Oliveira, T. M., Magalhães, A. C., & Rios, D. (2022). Sensitivity treatments for teeth with molar incisor hypomineralization: Protocol for a randomized controlled trial. *JMIR Research Protocols*, *11*, e27843. https://doi.org/10.2196/27843

Nogueira, V. K. C., Soares, I. P. M., Fragelli, C. M. B., Boldieri, T., Manton, D. J., Bussaneli, D. G., & Cordeiro, R. C. L. (2021). Structural integrity of MIH-affected teeth after treatment with fluoride varnish or resin infiltration: An 18-month randomized clinical trial. *Journal of Dentistry*, *105*, 103570. https://doi.org/10.1016/j.jdent.2020.103570

O'Leary, T. J., Drake, R. B., & Naylor, J. E. (1972). The plaque control record. *Journal of Periodontology*, *43*(1), 38. https://doi.org/10.1902/jop.1972.43.1.38

Olgen, I. C., Sonmez, H., & Bezgin, T. (2022). Effects of different remineralization agents on MIH defects: A randomized clinical study. *Clinical Oral Investigations*, *26*, 3227–3238. https://doi.org/10.1007/s00784-021-04305-9

Pajor, K., Pajchel, L., & Kolmas, J. (2019). Hydroxyapatite and fluorapatite in conservative dentistry and Oral implantology-a review. *Materials (Basel)*, *12*, 2683. https://doi.org/10.3390/ma12172683

Pasini, M., Giuca, M. R., Scatena, M., Gatto, R., & Caruso, S. (2018). Molar incisor hypomineralization treatment with casein phosphopeptide and amorphous calcium phosphate in children. *Minerva Stomatologica*, *67*, 20–25. https://doi.org/10.23736/S0026-4970.17.04086-9

Pepelassi, E., Rahiotis, C., Peponi, E., Kakaboura, A., & Vrotsos, I. (2015). Effectiveness of an in-office arginine-calcium carbonate paste on dentine hypersensitivity in periodontitis patients: A double-blind, randomized controlled trial. *Journal of Clinical Periodontology*, *42*, 37–45.

Piepho, H. (2004). An algorithm for a letter-based representation of all-pairwise comparisons. *Journal of Computational and Graphical Statistics*, *13*, 456–466. https://doi.org/10.1198/1061860043515

Portella, P. D., Menoncin, B. L. V., de Souza, J. F., de Menezes, J. V. N. B., Fraiz, F. C., & Assunção, L. R. D. S. (2019). Impact of molar incisor hypomineralization on quality of life in children with early mixed dentition: A hierarchical approach. *International Journal of Paediatric Dentistry*, *29*(4), 496–506. https://doi.org/10.1111/ipd.12482

Reis, P. P. G., Jorge, R. C., Americano, G. C. A., Pontes, N. S. T., Peres, A. M. A. M., Oliveira, A. G. E. S., & Soviero, V. M. (2021). Prevalence and severity of molar incisor hypomineralization in Brazilian children. *Pediatric Dentistry*, *43*, 270–275.

Restrepo, M., Jeremias, F., Santos-Pinto, L., Cordeiro, R. C., & Zuanon, A. C. (2016). Effect of fluoride varnish on enamel remineralization in anterior teeth with molar incisor hypomineralization. *The Journal of Clinical Pediatric Dentistry*, *40*, 207–210. https://doi.org/10.17796/1053-4628-40.3.207

Rodd, H. D., Graham, A., Tajmehr, N., Timms, L., & Hasmun, N. (2021). Molar incisor hypomineralisation: Current knowledge and practice. *International Dental Journal*, *71*, 285–291. https://doi.org/10.1111/idj.12624

Scribante, A., Dermenaki Farahani, M. R., Marino, G., Matera, C., RRY, B., Lanteri, V., & Butera, A. (2020). Biomimetic effect of Nano-hydroxyapatite in demineralized enamel before orthodontic bonding of brackets and attachments: Visual, adhesion strength, and hardness in in vitro tests. *BioMed Research International*, *2020*, 6747498. https://doi.org/10.1155/2020/6747498

Solinas, G., Grabesu, V., Lattari, M., Strinna, R., Arnould, N., & Amodeo, A. A. (2021). Management of a hypomineralisation of the enamel by applying a remineraliser based on zinc hydroxyapatite (Microrepair). *Case Reports In Dentistry*, *2021*, 5291858. https://doi.org/10.1155/2021/5291858

Sönmez, H., Yıldırım, G., & Bezgin, T. (2013). Putative factors associated with molar incisor hypomineralisation: An epidemiological study. *European Archives of Paediatric Dentistry*, *14*, 375–380. https://doi.org/10.1007/s40368-013-0012-0

Steffen, R., Krämer, N., & Bekes, K. (2017). The Würzburg MIH concept: The MIH treatment need index (MIH TNI): A new index to assess

and plan treatment in patients with molar incisior hypomineralisation (MIH). *European Archives of Paediatric Dentistry*, *18*(5), 355–361.

Sundfeld, D., da Silva, L. M. S., Kluppel, O. J., Santin, G. C., de Oliveira, R. C. G., Pacheco, R. R., & Pini, N. I. P. (2020). Molar incisor hypomineralization: Etiology, clinical aspects, and a restorative treatment case report. *Operative Dentistry*, *45*, 343–351. https://doi.org/10.2341/19-138-T

Verma, P., & Muthuswamy, P. (2021). Bionic effects of nano hydroxyapatite dentifrice on demineralised surface of enamel post orthodontic debonding: In-vivo split mouth study. *Progress in Orthodontics*, *22*, 39. https://doi.org/10.1186/s40510-021-00381-5

Wadia, R. (2021). MIH-stainless steel crown or composite? *British Dental Journal*, *231*, 114. https://doi.org/10.1038/s41415-021-3285-4

Weerheijm, K. L. (2004). Molar incisor hypomineralization (MIH): Clinical presentation, aetiology and management. *Dental Update*, *31*, 9–12. https://doi.org/10.12968/denu.2004.31.1.9

Weerheijm, K. L., Jälevik, B., & Alaluusua, S. (2001). Molar–incisor hypomineralisation. *Caries Research*, *35*, 390–391. https://doi.org/10.1159/000047479

William, V., Messer, L. B., & Burrow, M. F. (2006). Molar incisor hypomineralization: Review and recommendations for clinical management. *Pediatric Dentistry*, *28*, 224–232.

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