Bionatura
Latin American Journal of Biotechnology and Life Sciences

Bionatura Issue 4 Vol 8 No 1 2023

Article

The effect of Zinc Oxide nanoparticles with sodium fluoride in remineralization of enamel caries

*Farah Alaa Abdulkareem [1], Athraa Alwaheb [2,] **
[1] Master student. Department of Pedodontics and Preventive Dentistry, College of Dentistry, University of Baghdad, Baghdad, Iraq.
[2] Professor, Department of Pedodontic and Preventive Dentistry, College of Dentistry, University of Baghdad, Baghdad, Iraq
**Correspondence: farah.alaa.farah@gmail.com ; Tel(009647705857992)*
Available from: http://dx.doi.org/10.21931/RB/CSS/2023.08.04.73

ABSTRACT

Background**:** the most prevalent chronic condition in the world is caries; the revolution in minimally invasive dentistry clarified the necessity for clinically effective methods to allow remineralization of newly formed enamel caries, and Nanotechnology provides new approaches. Nano-sized ZnO In addition to the individualized antifungal and antibacterial properties, it has been used in fluoride precipitation from aqueous solutions; it acts as an adsorbent to remove F ions. The study aimed to test the effect of applying Zinc Oxide NP with Sodium Fluoride solution on the microhardness of artificially induced dental caries on the enamel surface compared to Sodium Fluoride alone. Materials and methods: thirteen upper first premolars extracted for orthodontic treatment were used.results: the nanoparticles increase the surface remineralization more than sodium fluoride alone and have more excellent microhardness scores. Conclusion: Using zinc Oxide nanoparticles with sodium fluoride is better than using sodium fluoride alone.
Keywords: Dental caries, Noninvasive dentistry, Nanoparticles, Zinc Oxide Nanoparticles, Reminerlization.

INTRODUCTION

Dental caries occur in over a third of the world's population [1,2]. It is a dynamic process described by rotatory terms of demineralization and remineralization. Caries development or reversal will lay on the balance between demineralization pathological agents (fermentable carbohydrates, cariogenic bacteria, salivary dysfunction) and the defensive agents (remineralizing ions, antibacterial agents and sufficient saliva) that drag the balance on the way toward remineralization [3].
The minimally invasive dentistry revolution declared that the main aim is to initiate remineralization of new enamel caries lesions. Fluoride, nevertheless the most vital agent used in caries management strategies; a range of new remineralization methods have been developed to promote remineralization of deeper lesions, decreasing

the possible danger related to oral care products of high-fluoride contents and facilitating caries management over a lifetime[4,5]. Nanotechnology provides new approaches for the therapeutic prevention of oral disease, mainly dental caries and periodontal disease. Some Nanoparticles endorse antimicrobial and remineralizing traits that are magnificent and manageable to prevent biofilm formation, slow down demineralization, remineralize tooth structure, and fight caries-related microorganisms[6]. Nano-sized Zinc Oxide (ZnO NP) has prompted research into its potential as a novel antibacterial agent.

Along with its unique antibacterial and antifungal qualities, ZnO-NPs possess excessive catalytic and photochemical activities [7,8]. They have been used in fluoride precipitation from aqueous solutions. They act as an adsorbent to remove F ions [9]. This work would study the remineralization effect and the ability of zinc oxide nanoparticles to increase enamel microhardness with sodium fluoride compared to sodium fluoride alone.

MATERIALS AND METHODS

The teeth used were 30 maxillary first premolars extracted from 15-20-year-old patients for orthodontic purposes, collected from private dental clinics. The study involved two parts; the first was to prepare and test the extracted material. The second part involved teeth sample preparation and Vickers microhardness measurement.

Sample Preparation

Every tooth was cleaned using a cumin scaler, then prophylaxis was accomplished using a pumice and a rubber cup, then saved for two weeks in a thymol solution[10]. Enamel is scrutinized to omit any cavities, stains, or enamel flaws. Then, the buccal surface was polished using Sof-Lex Disks (3M ESPE, USA), starting with the coarse, then medium and fine, ending with the terrific fine, using a contra-angle slow-speed hand-piece. A circular window of uncovered enamel 2 × 2 mm (in dimension) was accomplished by applying acid-resistant nail varnish to the remaining buccal surface of each tooth. Followed by soaking Each tooth alone in the demineralizing solution for 4 successive days (96 h) to create artificial caries-like lesions on the enamel surface [11]. Then, it was washed cautiously and stored in storage with artificial saliva and divided into three groups, Group A: untreated (demineralization followed by soaking in artificial saliva as control group unfavorable); Group B: demineralization followed by soaking in zinc oxide Nanoparticles (99.8%, 10-30nm, 0.7% w) with 5%NaF solution for 4 minutes, then stored in artificial saliva and Group C: demineralization followed by soaking in 5% sodium fluoride solution for 4 minutes and stored in artificial saliva, as control positive. Every Sample was stored independently by immersion in every group in artificial saliva that was changed daily until the surface microhardness test (SMH) [10]. thirty teeth (ten teeth from each treatment group) were used to measure SMH at baseline (sound enamel surface), after the demineralization procedure, and after remineralization. The measurements have been executed with a digital Vickers microhardness tester with a diamond indenter in the laboratory of metal checking, Department of Metallurgy and Manufacturing Engineering, University of Technology. The measurements were gathered via the utility of a five-hundred-gram load for thirty seconds vertically toward the enamel's surface. The identical calibrated device and examiner were used to collect all measurements. The hardness value for each specimen was determined by taking the average of three indentations during each analysis [(12)].

Statistical Analysis

Data description, evaluation and presentation were performed using Statistical Package for Social Science (SPSS version 21, Chicago, Illinois, USA)
Level of significance as Not significant P>0.05, Significant P<0.05.

RESULTS

Normality test of Standardized Residual of phases among groups:

Variables	Groups	Shapiro-Wilk		
		Statistic	df	P value
Standardized Residual for Baseline	Control	0.903	10	0.235
	ZnONP+5% NaF	0.863	10	0.084
	5%NAF	0.850	10	0.058
Standardized Residual for Demin	Control	0.854	10	0.066
	ZnONP+5% NaF	0.943	10	0.586
	5%NAF	0.891	10	**0.**173
Standardized Residual for Remin	Control	0.939	10	0.537
	ZnONP+5% NaF	0.961	10	0.798
	5%NAF	0.870	10	0.100

Table 1. Normality test of Standardized Residual of phases among groups

The results above show that the Standardized Residual of microhardness in all phases is usually distributed among groups using the Shapiro-Wilk test at p>0.05.

3.2 Multiple Pairwise Comparisons of surface microhardness among phases by groups:

Multiple Pairwise Comparisons of SMH among phases by groups.				
Groups	(I) Phases	(J) Phases	Mean difference	p-value
Group A	Baseline	Demin.	174.213	**0.00000**
		Remin.	173.313	**0.00000**
	Demin.	Remin.	-0.900	0.93559
Group B	Baseline	Demin.	181.599	**0.00000**
		Remin.	55.286	**0.00160**
	Demin.	Remin.	-126.312	**0.00000**
Group C	Baseline	Demin.	137.112	**0.00000**
		Remin.	104.910	**0.00000**
	Demin.	Remin.	-32.202	**0.00701**

Table (2) Compares the phases of each group. NS=not significant at p>0.05., *=significant at p<0.05.

In group A:

A- There is a significant difference between the baseline phase and each demineralization and remineralization phase of the SMH test (the baseline records of SMH are higher).

B- There is no significant difference between the demineralization and remineralization phases of the SMH test.

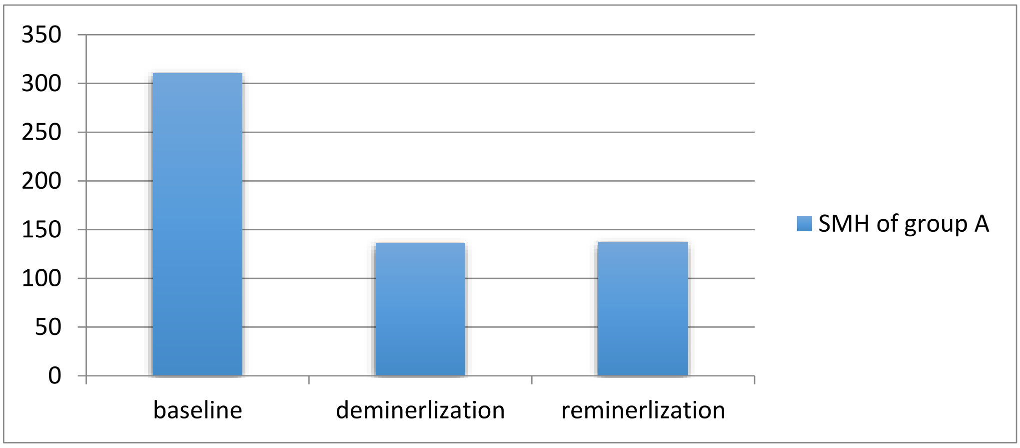

Figure 1. Surface microhardness between phases of group A.

In group B:

A- There is a significant difference between the baseline phase and each SMH demineralization and remineralization phase.

B- There is a significant difference between the demineralization and remineralization phases of the SMH test (SMH is higher in the remineralization phase)

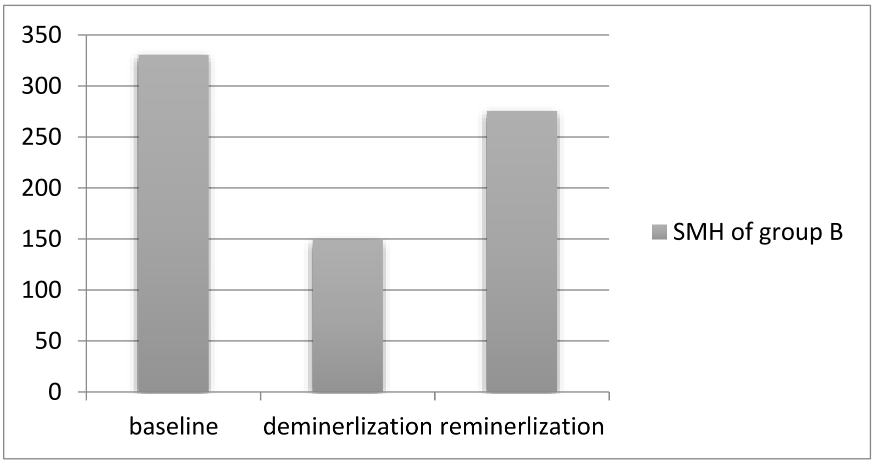

Figure 2. Surface microhardness between phases of group B.

In group C

A- There is a significant difference between the baseline phase and each SMH demineralization and remineralization phase.

B- There is a significant difference between the demineralization and remineralization phases of the SMH test (SMH is higher in the remineralization phase)

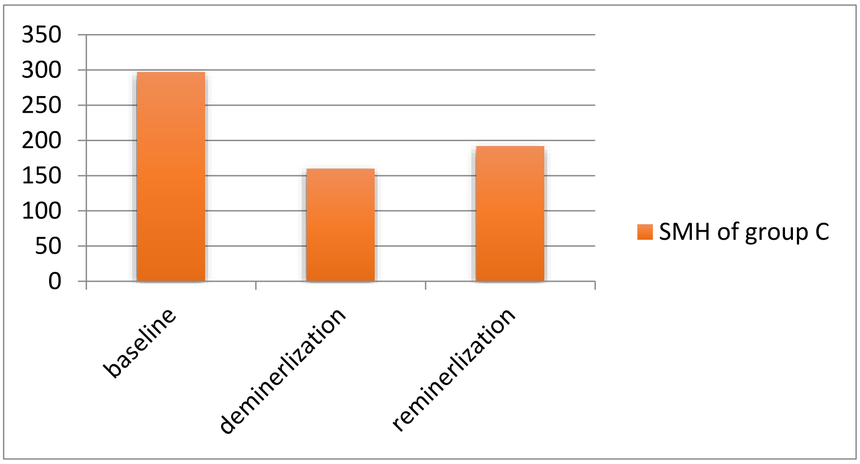

Figure 3. Surface microhardness between phases of group C.

Descriptive and statistical test of Surface microhardness (SMH) among groups and phases:

Groups		Baseline	Demineralization	Remineralization	F	P value
Group A	Min.	252.800	48.950	48.950	68.407	**0.00000***
	Max.	371.800	165.820	166.820		
	Mean	310.540	136.327	137.227		
	±SD	50.692	37.637	37.311		
Group B	Min.	309.600	105.820	205.600	97.772	**0.00000***
	Max.	370.700	200.950	320.769		
	Mean	330.326	148.727	275.039		
	±SD	22.674	25.777	41.886		
Group C	Min.	252.800	100.680	120.200	38.007	**0.00000***
	Max.	351.800	200.000	260.400		
	Mean	296.440	159.328	191.530		
	±SD	36.182	29.547	38.343		
F		1.979	1.346	31.315		
P value		0.158 NS	0.277 NS	**0.00000***		

Table 3. Descriptive and statistical test of Surface microhardness (SMH) among groups and phases. NS=not significant at p>0.05., *=significant at p<0.05.

The table above shows the comparison between enamel microhardness between phases of each group (baseline, demineralization and remineralization) and between each phase of the four groups:

1- There is a significant difference in surface microhardness between the three phases of each group ($p<0.05$), with the highest SMH in baseline records and the lowest at the demineralization phase.

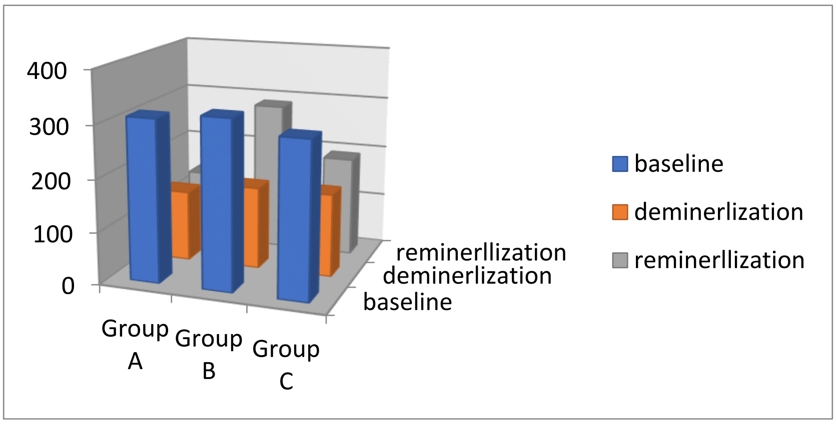

Figure 4. The difference in surface microhardness between the three phases of each croup.

There is no significant difference in surface microhardness in the baseline phase between the three groups (p=0.158 NS)

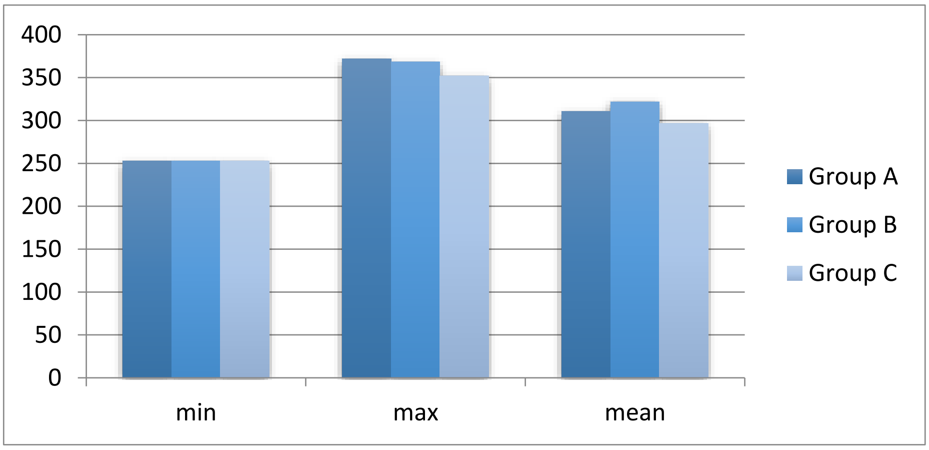

Figure 5. the difference in surface microhardness in the baseline phase between the three groups

There is no significant difference in surface microhardness in the demineralization phase between the four groups (p=0.277 NS)

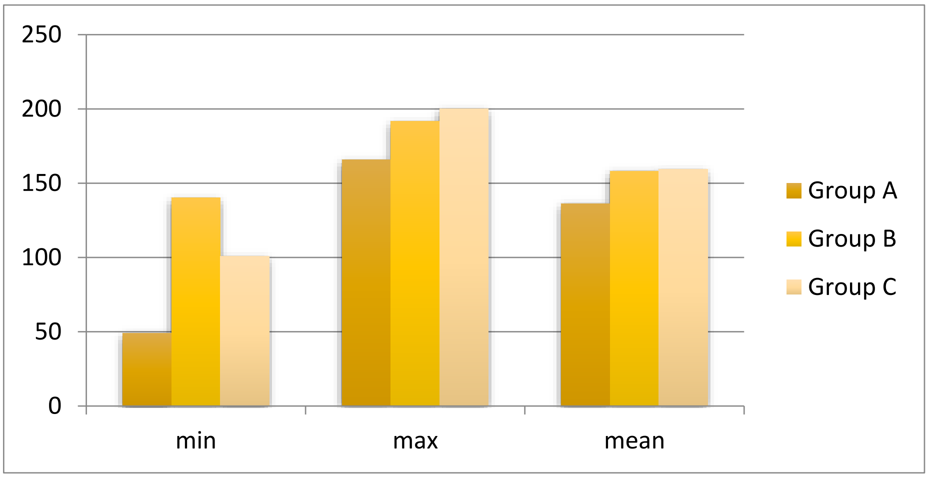

Figure 6. Difference in Surface microhardness in the demineralization phase between the three groups.

There is a significant difference in surface microhardness in the remineralization phase between the three groups ($p<0.05$), with the lowest SMH in group A and the highest in group B.

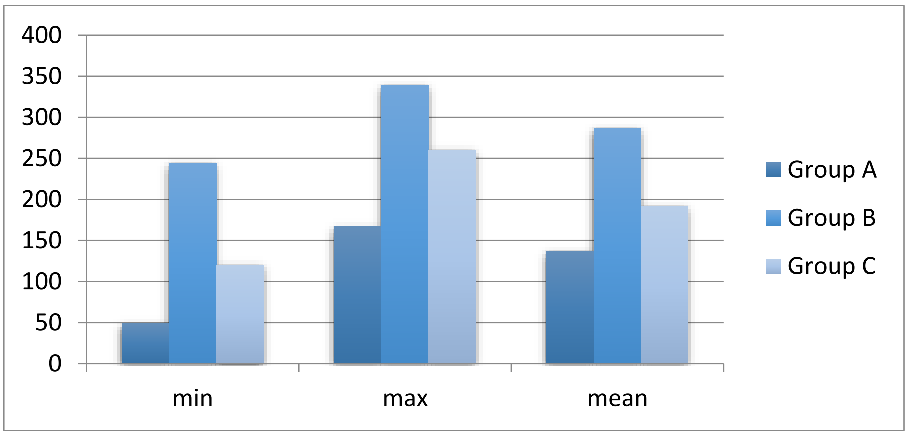

Figure 7. Difference in Surface microhardness in the remineralization phase between the three groups

Multiple pairwise comparisons of SMH among groups in the third phase: -

The table below shows the difference in SMH among groups in the remineralization phase:

There is a significant difference between group A and the other groups in SMH (group A has lower SMH).

Group B has a significant difference in SMH with Group C.

	(I) Groups	(J) Groups	Mead difference	**P value**
	Group A	Group B	-137.812	**0.00000**
		Group C	-54.303	**0.00454**
	Group B	Group C	83.509	**0.00006**

Table 4. Multiple pairwise comparisons of SMH among groups in the third phase.*=significant at p<0.05

DISCUSSION

Many methods have It has been evolved for the curing of initial carious Lesions that would fill the hole in the cure between preventive And. Restorative procedures [(13,14)]. In this in vitro revision, the effects of Zinc Oxide nanoparticles with Sodium fluoride solution were studied on permanent teeth. They were compared to sodium fluoride solution alone by using a microhardness tester.

A demineralization procedure was followed for in vitro initiation of artificial enamel caries that was previously used in other studies [11,15]. Soaking every Sample alone for 4 consecutive days (96 hours) in the demineralizing solution to produce artificial caries-like lesions on the enamel surface, and then carefully rinsed and stored in storage with deionized water. Different methods are utilized to offer evidence for mineral addition or thrashing. The most commonly used one is Surface microhardness analyses(SMH), which are rapid, easy and simple to gauge in a non-destructive pattern. Its method of Action depends on quantifying The confrontation of materials' surfaces is contrary to plastic deformation from a standard source, so the measurements can be taken from the same Sample frequently over an episode, decreasing the trial variation 16. Vickers technique was used that also provided successful results by previous studies [17,18,19,20,21,22,23]. Enamel microhardness was measured for sound enamel after demineralization and following treatment with the selected agents.

Enamel surface has more minerals Than enamel subsurface. So, polished teeth were used to decrease the normal variation of enamel surface among the samples that may respond differently to acidic dissolution [24]. Specimens were fixed in a metal mold. The samples were prepared because any tilting or non-flat surface might produce a very long indentation and, hence, a lesser hardness value. Consequently, a flat specimen's surface was essential in this test [25,26].

The outcome of this study revealed a statistically major reduction in microhardness of enamel surface after demineralization for all groups that indicate enamel caries because an acidic environment results in outward movement of the tooth minerals, mostly calcium and phosphate parting behind micro pores and decreasing solidity [27].

Study Groups:

Group A (control negative): - There is a significant difference between the baseline phase and each demineralization and remineralization phase of the SMH test (the baseline records of SMH is higher), but there is no significant change in microhardness between the demineralization and remineralization phase. This matches up with Somani *et al*. (2014) and Yahya (2020)[15,28] studies. Who discovered the control group revealed the lowest remineralization levels. This might be related to the fact that the artificial saliva utilized in this investigation was made without fluoride ions [29].

Group B (Zinc Oxide nanoparticles): - In SMH, the baseline records had the highest scores, and they decreased significantly after demineralization and then returned to a high score after adding the remineralization agent.

Nano-sized ZnO has led to exploring its use as a new antibacterial and antifungal agent. Many studies supported its antibacterial actions in controlling dental caries, like Bindhu et al. 2020 and Rajadorai et al. 2020 [30,31].

There are no studies about using zinc oxide nanoparticles to increase fluoride precipitation on the enamel surface and enhance remineralization. However, it has been used in fluoride precipitation from aqueous solutions; it acts as an adsorbent to remove F ions [8].

GroupC (NaF):- The SMH is greatest during the baseline phase and is degreased significantly after demineralization, then increases again after remineralization but remains less than SMH after treatment with group B.

Compression between groups:-

The SMH results are the same in the baseline and the demineralization phase for all groups, but the results differ in the remineralization phase, with the lowest results for group A then increasing significantly in group C and the greatest results in group B, so the use of ZnO NP with NaF increase the efficiency of caries remineralization than sodium fluoride ions alone and result in more homogenous surface.

CONCLUSION

Adding Zinc Oxide nanoparticles to 5% Sodium Fluoride is more efficient than sodium fluoride alone in the remineralization of enamel caries, and the SMH test scores were much higher, so it is better for use in the prevention of dental caries.

Ethical Clearance: the research ethical committee at scientific research by ethical approval of both environmental and health and higher education and scientific research ministries in Iraq (480, 19-1-2022)

Conflict of interest: the authors declare no conflict of interest.

Funding self-funding

Reference:

1. Vos T.; Flaxman A.D.; Naghavi M. et al. Years lived with disability (YLDs) for 1160 sequelae of 289 diseases and injuries 1990-2010: a systematic analysis for the Global Burden of Disease Study 2010 [published correction appears in Lancet, 2013;381(9867):628]. Lancet. 2012;380(9859):2163-2196.
2. Marcenes W.; Kassebaum N.J.; Bernabe E. et al. Global burden of oral conditions in 1990-2010: a systematic analysis. J Dent Res, 2013; 92(7):592---597
3. Featherstone, J. & Chaffee, B. The evidence for caries management by risk assessment (CAMBRA®). Advances in dental research, 2018; 29, 9-14.
4. Philip, N. State of the Art Enamel Remineralization Systems: The Next Frontier in Caries Management. Caries Res, 2019; 53, 284-295.
5. Swadi AK, Jassim ZW, Kadum MU, Ali MR. Influence of Fluoride Addition on Hydroxyapatite Prepared for Medical Applications. Baghdad Science Journal. 2012;9(3):541-6.
6.
7. 6. Mary A. S. Melo ; Sarah F. F.; Guedes. Hockin H. K. Xu. Lidiany K. A. Rodrigues. Nanotechnology Based Restorative Materials for dental caries management, 2013.
8.
9. 7. Padmavathy, N. and Vijayaraghavan, R. Enhanced bioactivity of ZnO nanoparticles—an antimicrobial study. Science and technology of advanced materials, 2008.
10.
11. 8. Song, Z.; Kelf, T.A.; Sanchez, W.H.; Roberts, M.S.;Rička, J.;Frenz, M. and Zvyagin, A.V. Characterization of optical properties of ZnO nanoparticles for quantitative imaging of transdermal transport. Biomedical optics express, 2011; 2(12), pp.3321-3333.
12.
13. 9. Mahvi, A.H.; Rahdar, A.; Igwegbe, C.A.; Rahdar, S. and Ahmadi, S. Fluoride removal from aqueous solutions by zinc oxide nanoparticles. Fluoride, 2020; 53(3), pp.531-541.
14.
15. 10. Elkassas, D. & Arafa, A. Remineralizing efficacy of different calcium-phosphate and fluoride based delivery vehicles on artificial caries like enamel lesions. Journal of Dentistry, 2014; 42, 466-474.
16.
17. 11. Itthagarun, A.; Verma, S.; Lalloo, R.; King, N.; Wefel, J. & Nair, R. Effects of fluoridated milk on artificial enamel carious lesions: a pH cycling study. Journal of Dentistry,2011; 39, 817- 824.
18.
19. 12. Kamal, D., Hassanein, H., Elkassas, D. & Hamza, H. Comparative evaluation of remineralizing efficacy of biomimetic self-assembling peptide on artificially induced enamel lesions: An in vitro study. Journal of conservative dentistry: JCD, 2018; 21, 536.
20.
21. 13. Fearherstone, J. Remineralization, the natural caries repair process—the need for new approaches. Advances in dental research, 2009; 21, 4-7.
22.
23. 14. Fisher, J.; Johnston, S.; Hewson, N.; Van Dijk, W.; Reich, E.; Eisele, J. L. & Bourgeois, D. FDI G lobal C aries I
24. nitiative; implementing a paradigm shift in dental practice and the global policy context. International dental journal, 2012; 62, 169-174.
25. 15. Yahya mohammed yahya. Remineralizing Efficacy of Biomimetic Self-Assembling Peptide on Artificially Induced Enamel Lesions (a Comparative in Vitro Study), 2020.
26.
27. 16. Oliveira, G. M., Ritter, A. V., Heymann, H. O., Swift JR, E., Donovan, T., Brock, G. & Wright, T. Remineralization effect of CPP-ACP and fluoride for white spot lesions vitro. Journal of Dentistry, 2014; 42, 1592-1602.
28.
29. 17. Al-sayyab, M. The potential effect of combined CO2 laser and fluoride on acid resistance of human dental enamel and root surface in vitro. PhD thesis, college of Dentistry, University of Baghdad, 2000.
30.
31. 18. Al-anni, M. Effect of selected metal salts on the microhardness and microscopic feature of initial carious lesion of permanent teeth. A master thesis, College of Dentistry, University of Baghdad.2005.

32.
33. 19. Al-aubaidi, R. Effect of water extract of black and green tea on the microhardness and microscopic feature of initial caries-like lesion of permanent teeth, compared to fluoridated agent. A Master thesis, College of Dentistry, University of Baghdad,2007.
34.
35. 20. Shaker, N. Effect of water extracts of cinnamon on the microhardness and microscopic feature of initial caries-like lesion of permanent teeth, compared to fluoridated agents. A Master thesis, College of Dentistry, University of Baghdad.2008.
36.
37. 21. Fahad, A. Effect of casein phosphopeptide-amorphous calcium phosphate on the microhardness and microscopic features of the sound enamel and initial caries-like lesion of permanent teeth, compared to fluoridated agents. College of Dentistry, University of Baghdad,2010.
38.
39. 22. Al-lami, A. Effect of water Clove extract on the microhardness and microscopic features of initial caries-like lesion of permanent teeth, compared to fluoridated agent. J Bagh Coll Dent. , 2011; 23: 110-113.
40.
41. 23. Al-bazzaz, F. The effect of CO2 laser, hydroxyapatite and iron oxide nanoparticles on human dental enamel and streptococcus mutans (In Vitro study). PhD. thesis, College of Dentistry,University of Baghdad, 2017.
42.
43. 24. Adebayo, O.; Burrow, M.; Tyas, M. An SEM evaluation of conditioned and bonded enamel following carbamide peroxide bleaching and casein phosphopeptide-amorphous calcium phosphate (CPP-ACP) treatment. J Dent, 2009; 37(4): 297-306.
44.
45. 25. Kinney, J.; Balooch, M.; Marshall, S.; Marshall J, G. & Weihs, T. Hardness and Young's modulus of human peritubula and intertubular dentine. Arch Oral Biol, 1996; 41(1): 9-13.
46.
47. 26. Gutierrez-Salazar, M. & Reyes-Gasga, J. Microhardness and chemical composition of human tooth. Mater Res. 2003; 6(3): 367-373.
48.
49. 27. Li, X.; Wang, J.; Joiner, A. & Chang, J. The remineralization of enamel: a review of the literature. J Dent. , 2014; 42(1): S12-S20.
50.
51. 28. Somani, R.; Jaidka, S.; Singh, D. J. & Arora, V. Remineralizing potential of various agents on dental erosion. journal of oral biology and craniofacial research, 2014; 4, 104-108.
52.
53. 29. Memarpour, M.; Fakhraei, E.; Dadein, S. & Vossoughi, M. Efficacy of fluoride varnish and casein phosphopeptideamorphous calcium phosphate for remineralization of primary teeth: a randomized clinical trial. Medical Principles and Practice,2015; 24, 231-237.
54.
55. 30. Rajadorai, S.; Harris, G.; Robinson, A.; Miles, E.J.; Roberts, J.M.; Cooper, L.; Abou Neel, E.A.; Higham, S.M.; Flannigan, N.L. and Valappil, S.P. Antibacterial, remineralizing zinc oxide-doped phosphate-based glasses. Materials Letters, 2022; 306, p.130813.
56. 31. Bindhu, M.R.; Ancy, K.; Umadevi, M.; Esmail, GA; Al-Dhabi, N.A. and Arasu, M.V., Synthesis and characterization of zinc oxide nanostructures and its assessment on enhanced bacterial inhibition and photocatalytic degradation. Journal of Photochemistry and Photobiology B: Biology, 2020; 210, p.111965.

Received: May 15, 2023/ Accepted: June 10, 2023 / Published: June 15, 2023
Citation: Abdulkareem, F.A.; Alwaheb, A. The effect of Zinc Oxide nanoparticles with sodium fluoride in remineralization of enamel caries. Revista Bionatura 2023;8 (2) 63. http://dx.doi.org/10.21931/RB/CSS/2023.08.04.73