Original article

BIOMEDICINSKA ISTRAŽIVANJA

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Analysis of clinical parameters of white spots on young permanent teeth after their treatment with different dental varnishes

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Summary

Introduction. Detection of early carious lesion and its remineralization is the most effective way for caries treatment. The aim of this study is to compare effectiveness of remineralization of the early carious lesions with different fluoride varnishes.

Method. The research included 30 children, girls and boys, aged 12 and 13, in whom the presence of white lesions on at least two vestibular surfaces was detected on 60 permanent teeth during a clinical examination by visual method. Gingival status was determined based on bleeding or non-bleeding after probing, activity of the white lesion was visually examined and the size of the lesion was measured. After the measurement, the application of varnishes was performed by the procedure that two different dental varnishes were applied to two different teeth with white lesion in the same person: group I – fluoride varnish enriched with minerals with 5% sodium fluoride. Activity, lesion dimensions and gingival status were examined after four, eight and twelve weeks when new varnishes were also applied.

Results. Inactivation of white lesions was recorded in the eighth week, but statistically significant difference was recorded after twelve weeks with 56.2% inactive lesions in the group I and 57.5% in the group II. There was no recorded difference in the lesion activity of the varnishes. Changes in the gingivo-incisal diameter during the visits amounted to: 3.47 mm/3.59 mm/3.53 mm/3.46 mm for the group I and 3.21 mm/3.19 mm/3.18 mm/3.20 mm for the group II. Changes in mesio-distal diameter amounted to: group I: 2.94 mm/2.81 mm/2.84 mm/2.4 mm; group II: 2.94 mm/2.87 mm/2.89 mm/2.90 mm. Statistically significant difference in both diameters was recorded between the first and second visit in the group I (p = 0.0046). There was no statistically significant difference recorded in the group I during other visits. In the group II statistically significant differences in the changes of lesion dimension were not recorded. Statistically significant differences p< 0.001 were recorded in the presence of gingival bleeding between the examined groups at the first visit, where significantly more lesions with healthy gingiva were observed in group I (84.4%).

Conclusion. Dental fluoride varnishes cause remineralization of early carious lesion after twelve weeks without significant difference between classic and enriched varnishes.

Key words: caries, white lesion, fluoride, remineralization

Introduction

Caries, as one of the most frequent chronical diseases, remains a very significant public health problem in our country as well as in the world. In the previous decades new scientific knowledge was developed and it influenced the paradigm shift in caries prevention and treatment. Modern principle of minimum intervention dentistry is focused on the early prevention and diagnosis of caries in order to restore the tooth integrity by secondary prevention measures, enabling thus significant savings in comparison to classic restoration treatment [1]. This principle implies: determination of the caries risk, dental plaque control, application of a certain hygiene and diet regime, as well as remineralization of the early carious lesions. The early carious lesion or white spot (white spot lesion - WSL) represents the earliest clinical stage of disease which manifests itself in histopathological changes within crystal bar of the surface intact enamel where "ad integrum" recovery can still be used. Clinically the lesion manifests itself as a white matte surface and in this stage of disease it has reversible character. At this stage it is possible to stop further demineralization and cavity formation process by using certain preparations and stimulate remineralization eliminating thus the necessity for preparation and restoration [2]. The existing evidence clearly shows that caries is the consequence of the disturbed balance between permanently present remineralization and demineralization processes caused by the presence of active cariogenic biofilm and continuous presence of dissolvable carbohydrates. Longer enamel exposure to acids produced by cariogenic bacteria leads to more extensive mineral extraction from hydroxyapatite crystal with increase in the volume of intercrystal areas. In case of further demineralization this phenomenon becomes clinically visible as turbidity of the bright enamel surface already around the fourteenth day and this is marked as the early carious lesion, or "white spot" [1, 3]. The possibility to control caries by stimulating remineralization process is a significant achievement of modern dentistry. Fluorides have been used for a longer period of time as a standard procedure not only in prevention, but also in remineralization of early lesions. It is recommended to use preparation with high fluoride concentration, whereby application of dental varnishes has proved to be most efficient and secure choice in children of school and preschool age [4]. Besides the high prevention effects achieved by fluoride varnishes recent research increasingly point to the necessity of the presence of the building elements for restoration of the crystal apatite [5]. One of those - very popular one is dental fluoride varnish enriched by amorphous calcium phosphate, stabilized by milk protein casein phosphopeptide (CPP-ACP) [6].

There are few studies that followed dynamics of the clinical changes of the white spot. The aim of the research was to determine to what extent it is possible to track by visual-tactile method clinical recovery of the white spot on enamel and whether the type of varnish influences changes in clinical parameters of the remineralization process.

Method

Research was conducted with 30 children (12 boys and 18 girls), ages 12 and 13. Out of each respondent two permanent teeth were chosen which, during clinical examination by visual method, showed white spots on at least two vestibular surfaces. The research included only healthy children without local or common diseases and/or allergies.

After professional removal of the soft tooth deposits, the tooth was air-dried for 5 seconds. First the gingival status of the examined teeth was determined using WHO periodontal probe with the rounded tip, whereby after probing sulcus in the white spot area it was monitored whether there would be bleeding. We decided to use gingival bleeding as a parameter of oral hygiene practice, because determination of plaque index is often not a reliable sign, as children are used to brush their teeth immediately before the dental check-up. Then the lesion size was determined in millimeters using graded periodontal probe, whereby gingivo-incisal and mesio-distal diameters were measured. Visual lesion examination implied observation of the lesion, which depending on whether it was rough and matte or smooth and shiny could be characterized as active or inactive. After the gingival status had been determined, the lesion was measured and visual examination was conducted, varnish was applied so that two different varnishes were used with the same person and on two different teeth:

Group I (classic varnish) – fluoride varnish (Fluor Protector S[®], Vivadent, Lichenstein) with 1.5% non-organic ammonium fluoride (7700 ppmF u 1 ml) in base with water and alcohol and with alcohol as a solvent which has 4 times higher concentration after bonding,

Group II (enriched varnish) – fluoride varnish enriched with minerals (MI varnish[®], GC, Tokyo, Japan) with 5% sodium fluoride (22.600 ppmF u 1 ml) containing also a bioavailable calcium phosphate in from of casein-phosphopeptide RECALDENTTM (ACP-CPP).

Application of varnish was conducted by special applicators (brushes) according to standard procedure and strictly adhering to producer's instruction. After both treated teeth had been painted from all sides, respondents and their parents were advised not to drink and eat for the next 60 minutes and not to brush teeth until the evening. All participants were educated how to brush their teeth properly (with a toothpaste containing 1450 ppm fluoride). During the next two visits, after four weeks (second visit) and eight weeks (third visit) from the first application of varnish, soft teeth deposits were removed with the rotating brush and toothpaste without fluoride. Gingival status, lesion activity, diameter in mesio-distale and gingival-incisal direction were recorded in the monitored teeth and varnishes were applied again. After twelve weeks (fourth visit), all measurements were conducted again.

The results regarding changes in white spots dimensions were shown in form of average value of all individual measures and by determining median and standard deviation. Regression, stagnation or progression of the initial lesion were recorded and the results from the visits and treatment groups were compared. Statistical processing of results was done by the program SPSS 16.0 Windows and by using the program MS Office Word and MS Office Excel. Test for independent samples or non-parameter Mann-Whitney U test was used depending on the normality of characteristic. Normal distribution of the observed characteristics was tested by Kolmogorov-Smirnov test and Pearson's chi-Squared test. For changes in related samples Wilcoxon signed-rank test was used with the significance limit of at least p = 0.05.

Results

During the first visit the examined lesions were active. Clinical inactivation of lesions was noticed during the third visit, after eight weeks (group I 15.6%; group II 15.2%). Half of the observed lesions were characterized as inactive after twelve (12) weeks during the fourth visit (56.2% group I and 57.5% group II; p< 0.0001). No significant lesion activities were recorded between the examined varnishes (Table 1).

Regarding changes in the lesion dimension in the group I, a statistically significant change was noticed in both directions: mesio-distale and gingival-incisale between the first and second visit (p = 0.046) (Table 2 and Table 3). During other visits (third and fourth visit) no statistically significant differences in change of

	Group I N=32 N (%) A I		Group II N=33 N (%) A I		р
1 visit	32 (100.0)	0 (0.0)	33 (100.0)	0 (0)	p=0.03
2 visit	32 (100.0)	0 (0.0)	33 (100.0)	0 (0)	p=0.03
3 visit	27 (84.4)	5 (15.6)	28 (84.8)	5 (15.2)	p=0.03
4 visit	14 (43.8)	18 (56.2)	14 (42.4)	19 (57.5)	p=0.03
p (result for related sample)	p<0.0001		p<0.0001		

Table 1. Lesion activity in the examined groups during visit

A = active lesion; I = inactive lesion

mesio-distale and gingivo-incisal lesion diameter were recorded in the group I (Table 2 and Table 3). There were no statistically significant changes in lesion dimension in both directions mesio-distale and gingivo-incisal in the group II. The value of gingivo-incisal changes in dimension between visits in the group I amounted to 3.47 mm/3.59 mm/3.53 mm/3.46 mm, and in the group II 3.21 mm/3.19 mm/3.18mm/3.20 mm. Regarding mesio-distal diameter the values were 2.94 mm/2.81 mm/2.84 mm/2.91 mm for the group I and 3.21 mm/3.19 mm/3.18 mm/3.20 mm for the group II.

Determination of gingival health by presence or absence of bleeding through probing points out to the level of hygiene routine. During the first check-up of the group I, a significantly greater number of surfaces with healthy gingiva was noticed (84.4%), whereas the condition of the group II was much worse (48.5%). During the second visit there was a mild decrease in healthy segments in the group I (there was no statistical significance) and increase in healthy segments in the group II, so that both groups were getting equal. This continued during further monitoring until the check-up after twelve weeks when there was no difference (93.8% group I; 87.9% group II, p> 0.05). The treated lesions were surrounded with healthy gingiva (Table 4).

	Group I N=32 X±SD (mm)	Group II N=33 X±SD (mm)	р
1 visit	2.94±0.98	2.94±0.99	p=0.971/ NS
2 visit	2.81±0.97	2.87±0.92	p=0.622/ NS
3 visit	2.84±0.95	2.89±0.93	p=0.736/ NS
4 visit	2.91±0.97	2.90±1.1	p=0.736/ NS
2 vs 1 visit 3 vs 1 visit 4 vs 1 visit	p=0.046 p=0.083/ NS p=0.180/ NS	p=1.000/ NS p=1.000/ NS p=1.000/ NS	

Table 2. Changes in the size of lesion in mesio-distal direction in the examined groups during visits

	Group I N=32 X±SD (mm)	Group II N=33 X±SD (mm)	р
1 visit	3.47±1.08	3.21±1,013	p=0.455 / NS
2 visit	3.59±1.01	3.19±1,01	p=0.150 / NS
3 visit	3.53±1.09	3.18±1,0	p=0.239 / NS
4 visit	3.46±1.07	3.20±1,01	p=0.338 / NS
2 vs 1 visit 3 vs 1 visit 4 vs 1 visit	p=0.046 p=0.157 / NS p=1.000/ NS	p=0.157/ NS p=0.157/ NS p=0.157/ NS	

Table 3. Changes in	the size of lesion in	gingiyo-incisal	direction in the examine	d groups during visits
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Table 4. Presence of gingival bleeding around the treated lesions in the examined groups during visits

	Group I N=32 N (%)		Group II N=33 N (%)		р
	Bleeding	Without bleeding	Bleeding	Without bleeding	Ĩ
1 visit	5 (15.6)	27 (84.4)	17 (51.5)	16 (48.5)	p=0.001
2 visit	8 (25.0)	24 (75.0)	5 (15.2)	28 (84.8)	p=0.26
3 visit	1 (3.1)	31 (96.9)	5 (15.2)	28 (84.8)	p=0.26
4 visit	2 (6.2)	30 (93.8)	4 (12.1)	29 (87.9)	p=0.26
p (result for ralared sample)	p=0.001		p=0.001		

Discussion

Fluoride treatment is a usual prevention method used in order to reduce enamel demineralization by Fluoride concentration in saliva and plaques efficient in preventing the process of demineralization and activating the one of remineralization. Organic acids produced by cariogenic bacteria cause reduction in the plaque pH value, which consequently leads to fluoride diffusion from plaque and saliva into enamel and formation of fluoridated hydroxyapatite. This new crystal form is more resistant to acids [7]. The work group formed in 2018 by the American Dental Association (ADA) and Center for Dentistry based on evidence recommends the use of 5% varnish based on fluoride for either blocking or recovery of the inactivated carious lesion on smooth surfaces of permanent teeth [8]. In the systematic analysis of the published studies, Urquhart et al. point out that by application of 5% fluoride varnish chances for either blocking or recovery of lesion on milk or permanent teeth are 2 to 3 times higher when compared to diseased enamel surfaces treated only by oral hygiene [9]. Researches show prolonged fluoride release from 5% varnishes (contain 22.000 ppmF) in the first six months after application and with the highest release in the first three months [10].

We decided for gingival bleeding as a parameter for defining the level of hygiene practice as determination of the plaque index is often not a reliable marker due to the fact that children brush their teeth immediately before dental check-up. Alavi and Yagarhi found out that the teeth treated by fluorine showed decrease in plaque index and reduced gingival inflammation and that fluorine treatment prevented development of white spots in children wearing fixed braces [11].

Although there was a statistically significant difference in gingival bleeding around treated lesions between the examined groups before the research (15.6% group I and 51.5% group II), it disappeared already during the subsequent visits, which points out to unevenness of the examined groups before the application of varnishes. However, the initial differences did not impact the remineralization process. Decrease in gingival reaction was gradual and started occurring only after two months (third visit), which points out to the significance of frequent remotivation for the establishment of healthy habits [12]. Wiegand et al. emphasize that application of varnishes can maintain balance on enamel surface even in patients who do not follow advice regarding anticariogenic nutrition and regular hygiene practice with fluoride toothpaste [13].

For our research we used specimen of classic and enriched fluoride dental varnish being among the rare ones registered in B&H, which points out to low dentists' interest in profilactic and secondary prevention intervention. Certain researches show a slightly higher effectiveness of combination of fluoride and remineralised biovelent ions [14, 15].

There was no significant difference between classical and enriched dental varnish in our research, which is in accordance with other studies [16, 17].

The most interesting aspect of the research was the result of statistically significant dimensional changes in both directions: mesio-distal and gingivo-incisal between the first and second visit in the examined teeth of the group I. It is confusing that there was a decrease in mesio-distal diameter and that there was even an increase in lesion dimension in gingivo-incisal. Possible explanation for higher decrease in mesio-distal diameter could lie in more aggressive teeth brushing after given instruction or demineralization level was more expressed with respondents showing gingivitis, so that oral hygiene and varnish effect led to an intensified remineralization in the first run. There is so far no adequate explanation for significant increase in gingivo-incisae lesion diameter, although it was minimum and there are also no requests for research using different methodology for monitoring remineralization process. Some studies which recorded mineral density using DIA-GOdent apparatus have not discovered such variations [18, 19], so that this finding could be ascribed to inaccuracy in measurement.

The results demonstrate that in the period of three months there was no significant reduction in white spots dimensions in both groups where we applied varnish two times, which at first sight could point out to low effectiveness of dental varnishes. However, by further monitoring of the activity after three months, significant increase in clinically inactive lesions was noticed as a clear sign of recovery. Statistically significant decrease in the number of active lesions in the twelveth week from the beginning of the research is still only partial, which indicates that this period is insufficient for recovery. Rahmi et al. showed that a more significant level of clinical reduction of the "white spot" was to be expected only after six months, and that even afterwards there could remain a whitish "scar" [20, 21].

Deficiency of this study was the fact that the analysis did not include other important parameters for demineralization reduction different from mere oral hygiene, such as presence of free sugars, whose effect, when consumed frequently, cannot be neutralised by fluoride. [22]. No significant difference in effectiveness of the examined dental varnishes was noticed. Certain researches published results about more favourable reminalisation features of varnishes containing kazein phosphopeptide-amorphium calcium phosphate (CPP-ACP), whereas other prioritized solely classic varnishes [23–25].

Conclusion

Fluoride-based varnishes lead to remineralization of the early carious lesions, but a period longer than three months is necessary to clinically visible reduction of the "white spots" size. This makes it a very unreliable parameter for monitoring the recovery. Lesion inactivity, i.e. its transition from the stage in which it is rough and matte to the one in which it is smooth and bright is the first visible remineralization sign which precedes reduction in lesion dimension. There is no significant difference in remineralization effectiveness between classic and enriched dental varnishes.

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Ethical approval. The Ethics Committee of the Public Health Institution Dental Institute of the Republic of Srps-ka approved the study and informed consent was obtained

from all individual respondents. The research was conducted according to the Declaration of Helsinki.

Conflicts of interest. The authors declare no conflict of interest.

*The Ethics Committee of the Public Health Institution Dental Institute of the Republic of Srpska gave its consent no: 01-343-3/17 on 15.2.2017. Ministry of Education and Culture of the Republic of Srpska gave its consent no: 07.041/052-7273/17 on 05.10.2017. For each child parents signed their consent which is a part of the medical documentation.

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Analiza kliničkih parametara kod bijelih mrlja mladih stalnih zuba nakon izloženosti različitim dentalnim lakovima

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Uvod. Detekcija početne kariozne lezije i njena remineralizacija je najefikasniji način liječenja karijesa. Cilj rada je upoređivanje efikasnosti remineralizacije početne kariozne lezije različitim fluoridnim lakovima.

Metod. U ispitivanje je bilo uključeno 30 djece oba pola, uzrasta 12 i 13 godina, kod kojih je na 60 stalnih zuba tokom kliničkog pregleda vizuelnom metodom otkriveno prisustvo bijelih mrlja na najmanje dvije vestibularne površine. Utvrđivalo se stanje gingive na osnovu prisustva ili odsustva krvarenja poslije sondiranja, vizuelno se ispitivao aktivitet bijele mrlje i mjerena je veličina lezije. Nakon mjerenja vršena je aplikacija lakova postupkom da su kod iste osobe na dva različita zuba sa bijelom mrljom aplikovana dva različita dentalna laka: I grupa – fluoridni lak sa 1,5% neorganskog amonijum fluorida, II grupa - fluoridni lak obogaćen mineralima sa 5% sodium fluorida. Aktivitet, dimenzije lezije i stanje gingive provjeravani su nakon četiri, osam i dvanaest nedjelja kada su vršene i nove aplikacije lakova.

Rezultati. Inaktivacija bijelih mrlja bilježi se u osmoj nedjelji, ali statistički značajna razlika evidentirana je nakon dvanaest nedjelja sa 56,2% inaktivnih lezija u I grupi i 57,5% u II grupi. Nije zabilježena razlika u aktivitetu lezija između lakova. Promjene gingivo-incizalnog promjera tokom posjeta su iznosile: 3,47 mm/3,59 mm/3,53 mm/3,46 mm za I grupu i 3,21 mm/3,19 mm/3,18 mm/3,20 mm za II grupu. Promjene mezo-distalnog promjera su iznosile: I grupa: 2,94 mm/2,81 mm/,2,84 mm/2,4 mm; II grupa: 2,94 mm/2,87 mm/2,89 mm/2,90 mm. Uočena je statistički značajna razlika oba promjera između prve i druge posjete u I grupi (p = 0,0046). Pri ostalim posjetama u I grupi nije evidentirana statistički značajna razlika. U II grupi nisu uočene statistički značajne razlike promjene promjera dimenzija lezije. Evidentirane su statistički značajne razlike p< 0,001 u prisustvu gingivalnog krvarenja između ispitivanih grupa pri prvoj posjeti, gdje je kod I grupe uočeno značajno više lezija sa zdravom gingivom (84,4%).

Zaključak. Dentalni fluoridni lakovi dovode do remineralizacije početnih karioznih lezija nakon dvanaest nedjelja, bez značajnih razlika između klasičnih i obogaćenih lakova.

Ključne riječi: karijes, bijela mrlja, fluor, remineralizacija