Different effects of whitening treatment on natural teeth and teeth treated with Icon resin: an experimental study in vitro



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Abstract

Introduction It is quite common for white spots to develop on a tooth, due sometimes to a defective formation of the enamel layer, and sometimes to patches of demineralisation as a result of poor oral hygiene during orthodontic treatment with fixed braces.

ICON DMG is currently the only noninvasive treatment for white spots. After a preliminary etching, it infiltrates the enamel, filling the spaces between the prisms with a resinous material that has a refraction coefficient very similar to that of healthy tooth enamel.

The aim of this study was to test the efficacy of professional whitening procedures on teeth previously treated with ICON. The study hypothesis was that infiltration with ICON resin creates a barrier capable of preventing the bleaching action of the whitening agent.

Materials White spots were artificially created on one half of the vestibular surface of 12 human teeth, while the other half was protected with a composite adhesive. The white spots were infiltrated with ICON and the protective adhesive was subsequently removed. A professional teeth whitening procedure was then completed on both halves of the teeth. A statistical analysis was performed to compare spectrophotometric recordings obtained before and after the ICON infiltration and teeth whitening procedures.

Discussion and Conclusions The whitening procedure modified the colour of the teeth on the half not infiltrated with ICON ($p<0.05$), but there was no statistically significant change in colour on the half infiltrated with ICON. The presence of the ICON resin seems to act as a partial barrier to the action of the whitening agent.

KEYWORDS White Spot, Fixed Appliance, Bleaching.

Introduction

In dentistry, the term 'white spot' (WS) is used to describe a number of lesions, including an impaired enamel development and localised areas of demineralisation or caries, that may be due to orthodontic treatments or have other causes [Kidd and Fejerskov, 2004]. From a clinical standpoint, WS looks chalky white and opaque - an optical effect of any demineralisation of the enamel's superficial and deeper layers that is exacerbated with dehydration of the surface area involved [Gorelick et al., 1982]. For a diagnosis of WS, we need to differentiate them from other types of lesions that may look similar clinically but are of a different nature, including fluorosis, hypomineralisation of traumatic or genetic origin, and molar-incisor hypomineralisation. WS originating from caries appear rough and porous, while those due to other causes are generally smooth and shiny [Kidd and Fejerskov, 2004]. The reported prevalence of WS can range from 2% to as high as 97%[Khoroushi and Kachuie, 2017], depending on the diagnostic techniques employed: their prevalence increases when quantitative light-induced fluorescence is used rather than visual inspection alone [Paris et al., 2013]. As concerns WS due to developmental defects of enamel, the prevalence ranges from 3.6% to 25%[Dogra et al., 2020]; in orthodontic patients, the prevalence is in the range of 30% to 70% while the treatment is underway[Julien et al., 2013]. Tüfekçi et al. [2011] found a higher incidence of WS during the first six months of treatment than during the six months thereafter. WS are typically located around orthodontic brackets or along the gum margin, usually on the vestibular surface, and adjacent to the areas most difficult to clean [Julien et al., 2013].WS occur mainly in the region of the upper front teeth, particularly on the lateral incisors, canines, premolars and central incisors. They are more common in males than in females [Mizrahi, 1982].

The main risk factor for the onset of WS is the presence of a fixed brace[Julien et al., 2013]: its irregular shape makes it difficult to ensure adequate oral hygiene, giving rise to areas where plaque can form. The uneven surfaces of the brackets, the bands and the wires all increase the area available for bacterial contamination and interfere with the natural self-cleansing effect of the saliva and oral muscles (tongue, lips, cheeks). This facilitates the brace's colonisation by the bacteria responsible for the formation of demineralised areas and non-cavitated caries, and even active, cavitated caries[Chang et al., 1997]. This situation is confirmed by the low incidence of WS in patients treated with lingual orthodontics. In the event of poor compliance with recommendations for oral hygiene, plaque stagnates and lowers the pH to a level with the enamel adjacent to the orthodontic brackets [Ludovichetti et al., 2022]. This prevents any remineralisation process, giving rise to decalcification [Gwinnett and Ceen, 1979] and the formation of WS already within four weeks of starting the orthodontic treatment [Øgaardet al., 1988; Chapman et al., 2010]. Other important risk factors are patient-related and include: a patient's medical history; dental history; diet; calcium, phosphate and bicarbonate levels in the saliva; fluoride levels; and genetic susceptibility. It is also important to pay attention to issues concerning any excess adhesive, the duration of the orthodontic treatment, excessive etching, and any existing tooth decay or previously treated caries [Heymann

and Grauer 2013]. In recent years, various preventive approaches have been introduced with the purpose of reducing the risk of WS developing. These include: special oral hygiene manoeuvres, possibly with the aid of an electric toothbrush, in patients with a high plaque index [Heintze et al.,1996]; paying greater attention to the spaces between the brackets, and between the bracket and the gum margin; rinsing with fluoridated mouthwash (containing 0.5% sodium fluoride); using toothpaste with a high fluoride content (1500-5000 ppm instead of 1000 ppm) and without any final rinsing of the mouth with water; applying fluoridated varnishes; more frequent sessions of iontophoresis (every three months instead of every six)[Al Mulla et al., 2010]; and applying fluoride-releasing orthodontic adhesives [Derks et al., 2004]. Various studies have cast doubts on the efficacy of such preventive measures, however, demonstrating that they have little efficacy in lowering the pH [Rølla et al., 1993], and the only noteworthy effect lies in an inhibition of the tendency for demineralisation [Benson et al., 2004; Shafi, 2008]. Like the mechanism of action responsible for the formation of caries, the onset of WS involves the colonisation of the enamel's surface by microflora (Streptococci non mutans) that are not initially cariogenic, but - in the case of a diet rich in fermentable carbohydrates - they tend to give rise to a prevalence of acidogenic and aciduric bacteria that alter the normal demineralisation and remineralisation cycles. The mineral substance is lost [Takahashi and Nyvad, 2011], the enamel crystals dissolve, and porosities develop between their prisms [Sudjalim et al., 2006]. These porosities then fill with air or water, which have a different refraction index from that of the healthy tooth (1.33 and 1 instead of 1.52)[Gorelick et al., 1982]. This changes how light is refracted on the surface of the enamel, giving rise to the characteristic appearance of WS. Treatment with infiltrating resins consists of applying a resin capable of penetrating inside the lesions in the enamel and modifying the refraction index of the WS to make it the same as that of the surrounding healthy enamel so that it is no longer so obvious. The only such infiltrating resin available on the market to date is called Icon (DMG, Hamburg, Germany). This is a hydrophilic resin based on unfilled triethylene glycol dimethacrylate. It features a high surface tension, a low viscosity, and a low contact angle, but a high coefficient of penetration. A fundamental characteristic of this resin is a refraction index similar to that of apatite crystals (1.52). Lowering the optical absorption reduces the difference in colour between the WS and the enamel, enabling the WS to be camouflaged [Paris et al., 2013]. The key element in this mechanism relates to the material's capacity for penetration, and the chances of it infiltrating the surface of the enamel [Manoharan et al., 2019]. Patients whose teeth have been treated with such infiltrating resins may ask their dentist for a dental whitening treatment. This is a process in which an oxidising agent (generally peroxide) is applied to the surface of the teeth. It attacks the organic compounds that cause discolouring of the teeth, forming epoxides first, and then alcohols that (being lighter) subsequently evaporate [Rodríguez-Martínez et al., 2019]. Clearly, an Icon resin treatment could be expected to interfere with the outcome of such dental whitening processes, but very few studies have investigated the efficacy of whitening agents on teeth previously treated with Icon. One such study[Santoset al., 2012], conducted on bovine teeth in vitro in 2012, concluded that the resin infiltration served as a barrier, preventing the whitening agent from completing its bleaching action. Another in vitro study on bovine teeth, published by Araùjo et. al [2015], involved teeth first treated with Icon, then discoloured with various types of stain, and then submitted to a whitening treatment. The Author concluded that the whitening process was effective in this case, probably thanks to a surface cleansing of the artificially induced stain. To our knowledge, no published studies specifically assessed the bleaching effect of a whitening treatment on human teeth previously infiltrated with Icon resin. The aim of the present study was thus to examine the effect - in terms of a chromatic variation - of a conventional whitening process on the vestibular surface of human teeth already treated with Icon infiltrating resin. Our study hypothesis was that the Icon resin would oppose the penetration of the bleaching agent in the teeth, consequently preventing their effective whitening.

Materials and Methods

Study design

This was an experimental study conducted in vitro on 12 upper and lower incisors selected from a total sample of 30 teeth. Teeth were excluded if they showed signs of decay, demineralisation, reconstructions, WS or brown spots. All the teeth included in the study had been preserved correctly in a physiological solution at room temperature. The crown of each tooth was divided into three areas for the purposes of our analysis.

Sample preparation

The samples were prepared by removing any plaque with a sickle scaler (LM Ergonorm 312) and a low-power piezoelectric ultrasonic scaler (Satelec). Any external pigmentation was removed with carborundum burs and rubbers, and the teeth were polished with a brush and pumice (Fig. 1/A). Each tooth was attached with resin on a plastic stand, with the apex of the tooth facing downwards, and identified with a number. A longitudinal line was marked along the greater axis of the crown to separate it into two equal halves (Fig. 1/B).

At T0, after being stored for 24 hours in a physiological solution at 37°C, the teeth were analysed with a Vita Easyshade Advance spectrophotometer (VITA Zahnfabrik, Bad Sackingen, Germany) to record their initial colour. The spectrophotometer recordings were obtained on three portions of each tooth (apical, medial and coronal), distinguishing the right-hand side from the left, for a total of 72 recording sites.

Creation of white spots

At T1 (Fig. 1), WS were created on each sample following the protocol used in the study by Araùjo et al. [2015]. They were created only on the left-hand side of each tooth (the "ICON side", Fig. 2). The right-hand side (the "no-ICON side") was protected by means of an etching treatment with orthophosphoric acid (Scotchbond™ Universal Etchant 37%, Fig. 1/C), and impregnated with adhesive (3M™ Scotchbond™ Universal Adhesive, Fig. 1/D). After photopolymerisation for 40 s (3M™ Elipar™ DeepCure-S), the samples were immersed in an acid solution to induce the formation of WS. This was a buffer solution containing 0.05 M acetic acid (CH_3COOH)[Meyer-Lueckel and Paris, 2008]. The teeth were kept in the solution for 10 hours at 37°C, then rinsed and dried. There was evidence of chalky WS that developed only on the ICON side of each tooth (Fig. 1/E), the no-ICON side having been protected in order to serve as a control vis-à-vis the same tooth.

Resin infiltration procedure

At T2 (Fig. 2), infiltration of the ICON side was done by means of a process involving the following steps:

1. abundant rinsing with air-water spray;
2. drying, again with air-water spray;
3. etching with 15% Icon Etch HCl (Fig. 2/F);
4. rinsing;
5. drying;
6. applying 99% ethanol on the WS (Fig. 2/G). Ideally, the

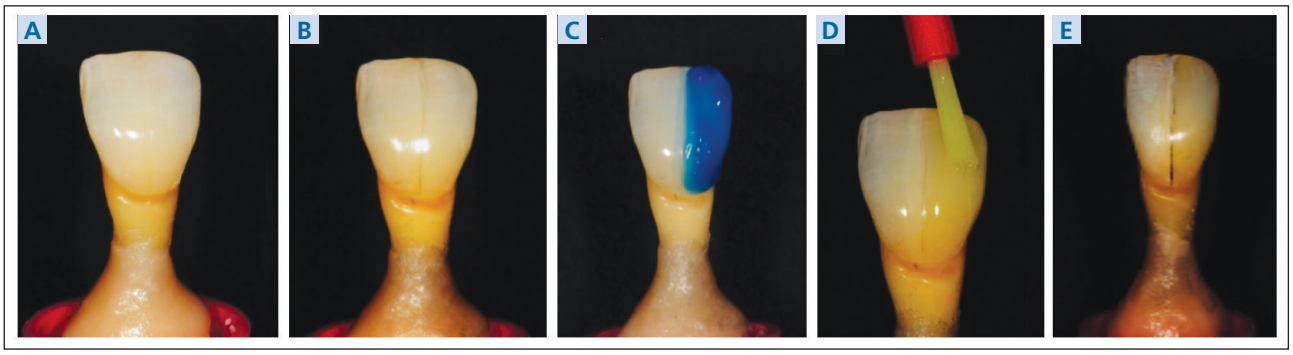


FIG. 1 Applying adhesive to one side of each tooth, and creating white spots.

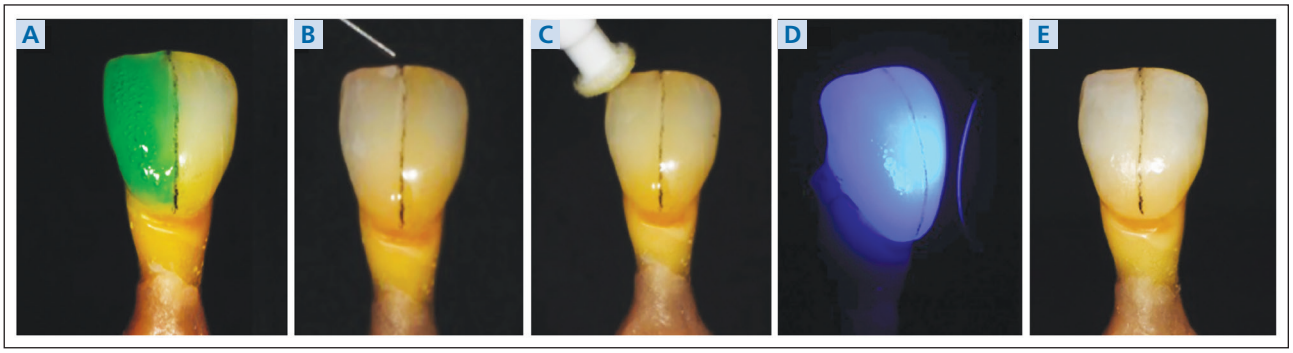


FIG. 2 Infiltrating half of the crown with Icon.



FIG 3 Applying the whitening agent all over the vestibular surface of the tooth.

purpose of this step is to obtain a first impression of the results achievable with the infiltrating resin, and the etching step has to be repeated if the WS is not camouflaged;

7. drying the sample again;
8. applying Icon resin (DMG Dental Company, Fig. 2/H) for two minutes, using the applicator tip provided;
9. blowing the resin coating gently for two minutes to ensure its adequate penetration inside the WS;
10. photopolymerisation of the resin for approximately 40 seconds (Fig. 2/I);
11. polishing of the resin coating with a toothbrush and pumice (Fig. 2/L).

The resin-infiltrated teeth were then stored in physiological solution. At T3, four weeks after the infiltration procedure (the time specified by the manufacturer to obtain a definitive outcome of the infiltration treatment), a second spectrophotometer recording was obtained on three levels, on the infiltrated right-hand side of each tooth. A different operator from the one taking the first recordings completed this step in order to introduce a randomisation effect.

Whitening process

At T4, after removing the adhesive from the N-IC SIDE with the aid of a carborundum bur and rubber, the whitening process was performed (Fig. 3) on both sides of the crown of each tooth. The whitening agent was M+W Select Power Whitening Xtra 38% (38%hydrogen peroxide gel). It was applied to the vestibular surface and left in place for 20 minutes, then rinsed off. This procedure was then repeated. At T5, after 24 hours of storage in a physiological solution at 37°C, a third and final spectrophotometric reading was obtained by a third, different operator, on both the IC side and the N-IC SIDE of each tooth. Apart from the spectrophotometer recordings, the whole procedure was completed by the same operator, who has considerable experience in the management of both Icon and bleaching treatments.

Instrumental analyses

The spectrophotometric analysis was performed by another operator, who completed all the measurements. The Vita scale was used to measure changes in colour, from the palest (A1) to the darkest (C4). Given the low numerosity of the sample, the Classical Vita scale was adopted, which contains six values in all (Table 1). The colour differences obtained were calculated as: ΔE_1= ET3-ET0, ΔE_2= ET6 – ET3, ΔE_3= ET6 – ET0.

Intraclass correlation coefficients (ICCs) were used for reliability testing, with a target value of 0.8, and a 95% confidence interval (CI) of 0.2.

Statistical analysis

After recording the colour of all the teeth, the data were analysed using R software (version 3.6.1 [2019-07-05] – "Action of the Toes", Copyright © 2019; The R Foundation for Statistical Computing. The tables show the medians and the first and third quartiles for the continuous variables. For the purposes of our analyses, the data were grouped from the 1st to the 16th value using the Classical Vita scale (Table 1). In the statistical analysis, the Wilcoxon test for paired data was used for continuous

N	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
	B1	A1	B2	D2	A2	C1	C2	D4	A3	D3	B3	A3,5	B4	C3	A4	C4

TABLE 1 Values from the 1st to the 16th on the Classical Vita scale.

	T0 (N=12)	T3 (N=12)	ΔE_1	p-value
Incisal third	2.8/8.0/12.5	4.5/7.5/ 9.5	-0.75	0.47
Medial third	5/12/14	3/11/12	-1.66	0.017*
Apical third	10/12/15	9/11/12	-1	0.31

TABLE 2 Wilcoxon test T0-T3 (ICON side).

	T3 (N=12)	T6 (N=12)	ΔE_2	p-value
Incisal third	4.5/7.5/195	4.2/7.0/ 11.0	-0.11	1
Medial third	3/11/12	3/11/12	-0.19	0.59
Apical third	9/11/12	9/11/12	-0.12	0.59

TABLE 3 Wilcoxon test T3-T6 (ICON side).

	T0 (N=12)	T6 (N=12)	ΔE_3	p-value
Incisal third	2.8/8.0/12.5	4.5/6.5/11.0	-0.9	0.45
Medial third	5.0/11.5/14.2	2.8/10.0/12.2	-1.91	0.013*
Apical third	10.5/12.0/15.0	7.5/11.0/14.2	-1.16	0.094

TABLE 4 Wilcoxon test T0-T2 (no-ICON side).

variables. A p-value < 0.05 was considered statistically significant.

Results

Statistical analyses were conducted on the data obtained with the three spectrophotometer recordings. We analysed the colour differences from T0 to T3 (ΔE_1), and from T3 to T6 (ΔE_2), always on the resin-infiltrated side (ICON side) of the teeth. Then we considered the differences between T0 and T6 (ΔE_3) on the un-infiltrated side (no-ICON side) of the teeth, distinguishing between the incisal, medial and apical thirds (for a total of 36 recordings on each side).

T0-T3: From T0 to T3, there was evidence of some changes in colour on the ICON side after it was infiltrated with resin: in 72% of cases (26 recordings) the resin application led to a change in colour, while in the remaining 28% (10 recordings) the colour of the tooth remained the same. In 6 cases, the colour became darker, while in 20 it was paler than at the baseline. It was only for the medial third that the result was statistically significant (p-value=0.017*, Table 2), with a mean colour change of -1.66, and a range of 8 values, from -6 a +1. There was no change in colour in medial third in only 3/12 cases.

T3-T6: The whitening treatment was completed between T3 and T6. There was evidence of the portion of tooth on the IC side changing in colour in 28% of cases, while there was no change in 72% of cases. The findings were the same all over the half of each tooth. The mean change in colour ΔE_2 was -0.14, with a range of six values, from -3 to +2. The results were not statistically significant for any of the three areas of tooth considered (Table 3).

T0-T6: There was evidence of changes in colour occurring on the no-ICON side between T0 and T6. The spectrophotometric results showed changes in colour that were due to the whitening treatment. In 22% of cases (8 recordings), the whitening process made no difference, while in 64% of cases (23 recordings) it led to a gain in colour - even of as much as 7 values - with a mean change of -1.3. In 14% of cases (5 recordings), the whitening treatment led to a loss of colour; 3 of these recordings were obtained from the incisal third and 2 from the apical third. The results obtained on a level with the medial third were statistically significant (p-value=0.013*), where the mean change in colour was -1.91 (Table 4). For the apical third, the results were weakly significant (p-value=0.094), with a mean of -1.16, and a range of values from -6 to +2.

Discussion

Our research hypothesis was confirmed regarding the efficacy of a tooth whitening treatment in changing the colour of teeth treated with Icon resin. Our first analysis concerned ΔE_1, i.e. a comparison between the ICON side before and after infiltration with Icon resin. The statistically significant findings mainly concern the medial third of the tooth, where the colour became paler. This new colouring of the ICON side was taken for reference in the subsequent comparisons after the whitening treatment.

It is worth emphasising the ΔE_2 relating to the ICON side (from T3 to T6): in the majority of cases (72%) the whitening agent had no effect. These results suggest that Icon serves as a barrier to the whitening agent, which consequently fails to penetrate inside the organic matrix of the tooth. This finding is consistent with the suggestion advanced by Santos et al.[2012] that, after polymerisation, Icon would alter the enamel's permeability to the whitening agent. Some bleaching effect was nonetheless apparent in a minority of cases: this finding could corroborate previous reports [Meyer-Lueckel and Paris, 2008; Paris et al., 2007], of the barrier effect of the resin depending on the 'pore volume of the lesion'. The 'resin shrinkage' phenomenon occurring during polymerisation could also give rise to an incomplete barrier [Paris et al., 2007], enabling part of the whitening agent to penetrate underneath and take effect on the underlying structures. Our finding is in marked contrast with the conclusions drawn in the study conducted in 2015 Araùjo [Araùjo et al.], who found a whitening effect irrespective of any infiltration with Icon. The Author also mentioned, however, that the bleaching effect was assessed in relation to artificially-created surface spots, and might have been due more to the tooth's surface cleaning than to the action of the hydrogen peroxide. As for the no-ICON side of the teeth, which served as a control group, the whitening agent had an effect on most of the sample teeth. This effect mainly concerned the medial third of the tooth, which is the area probably most heavily impregnated with the pigments on which the bleaching agent can take effect [Paris et al., 2013]. One aspect to consider concerns the nature of the artificially-created white spots, which can vary in depth and surface porosity from one tooth to another. This could interfere with both the infiltration of the Icon and the whitening effect obtained on the ICON-treated side, and consequently with the final results. The whitening is achieved by diffusion through the tissues from the outer surface, so the effect will be more or less evident depending on the extent and depth of the porosity of the defect created.

Another factor that may have interfered with the results concerns the procedure used to protect part of the tooth while

creating the white spot. Using orthophosphoric acid, applying an adhesive and then removing it with a carborundum bur, and polishing with a rubber can partly alter the surface porosity, making the tooth more permeable to the whitening agent. In fact, there was a statistically significant change of colour between before and after the whitening treatment on the no-ICON side. The treatment was expected to have some effect, however, because a professional whitening agent was used. The lack of any statistically significant difference on the ICON-treated side is more intriguing. In short, whitening procedures would be scarcely effective on teeth previously infiltrated with Icon resin. In cases where the use of Icon is indicated, it would therefore be important to inform patients that: should they wish to have their teeth whitened, it would be more effective if it were done before the Icon infiltration; and patients already treated with Icon who request a whitening treatment should be warned that it might have little effect on their resin-infiltrated teeth. Using a self-applied whitening agent (containing 10-16% carbamide peroxide) at home instead of a professional one at the dentist's could have a stronger whitening effect on the teeth not treated with ICON because of the longer treatment times involved; and a greater efficacy of the former has already been reported [Mounikat al., 2018]. Further studies could test this hypothesis.

Conclusions

Within the limitations of this in-vitro work, the following conclusions can be drawn:

- there was a statistically significant (p-value<0.05) change in colour between before and after infiltration with Icon on the side of the tooth treated with the resin;
- there was a statistically significant (p-value<0.05) change in colour between before and after the whitening treatment on the side of the tooth not infiltrated with Icon;
- there was no statistically significant evidence of any efficacy of the whitening procedure on the side of the tooth previously infiltrated with Icon (p-value>0.05).

Contributions

RF: Conceptualisation
PM: Investigastion
BD: Writing
DFA: Methodology
AV: Conceptualisation, Writing
LF: Supervision, Validation
All authors read and approved the final manuscript.

Clinical Relevance

Treating WS with ICON resin represents today the gold standard in the world. It is possible that patients treated with ICON require a bleaching treatment in subsequent years; studies in the literature assessing the effectiveness of the whitening agent on a tooth already treated with ICON are few, conflicting and not performed on human teeth. This in vitro study on human teeth found a clear decrease in the effectiveness of the whitening action on teeth already infiltrated. From a clinical point of view, it is advisable to warn a patient already treated with ICON of the reduced efficacy of whitening, or to perform a possible whitening treatment before infiltrating with ICON.

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