

Clinical Reversal of
Primary Occlusal Fissure Carious Lesions
(POFCLs)
Using Ozone in General Dental Practice

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BACKGROUND

Occlusal and root surface caries forms the largest percentage, up to 70%, of new carious lesions seen in general dental practice, with inter-dental caries forming some 15% of all new cases. Traditional approaches to the diagnosis, management and treatment of occlusal caries have changed over the last 20 years, but at present it still consists of amputation therapy. That is where areas of diseased tissue are cut out, and replaced with some type of filling material or restoration. The dental profession has needed an alternative to this amputation therapy, and recent research has pointed the way towards a novel treatment with ozone.

There have been many advances in dental materials over the last 150 years, from gold foil, to packable amalgam and anaesthetics. Moreover, with the introduction of adhesive systems, a whole new range of restorative materials and techniques has been made available to the dental profession. Nevertheless, every advance in the material technology has started from the premise of prior caries removal, or tissue amputation. The dental profession has never had a system where caries can be left to remineralise predictably before some type of restorative system is placed, that would minimise tissue loss and patient trauma.

Current research with ozone has opened up a whole range of potential treatment protocols. The new treatment protocols do not require removal of the diseased or carious tissue. Published papers have shown that after ozone treatment for just 10 seconds, debris and bacteria impacted in a fissure can mineralise, 'sealing' a fissure against potential decay.

After the initial treatment with ozone and a period of remineralisation, a restoration may not be even required. However if a restoration is required, then minimal preparation to remove unsupported enamel is often all that is necessary without the need for anaesthesia, before an adhesive restoration is placed.

Ozone can now be considered as a clinical alternative management strategy for caries and this statement is well supported in the increasing volume of published research. Research has shown that ozone breaks up the acidic products of cariogenic bacteria, which may be more important in the aetiology of the developing carious lesion (Lynch, E *et al* (1997). Research by Baysan, A *et al.* (2000), reported that ozone application for either 10 or 20 seconds was effective to kill 99% or more (99.9% after 20 seconds) of micro-organisms in primary root carious lesions *in vitro* and *in vivo*, and an application for a period of 10 seconds was capable of reducing the numbers of *Streptococcus mutans* and *S. sobrinus in vitro*. Further research by OT.Abu-Salem, MM *et al* (2002) have shown that occlusal caries in deciduous teeth can be effectively controlled with ozone treatment. And recent research by H.Domingo *et al* (2001) has looked at the acceptability of this treatment modality by patients.

The Aims of this Study

The aim of this study was to assess the reliability of using the DIAGNOdent (KaVo, Germany) to assess the presence of decay, and the safety & efficacy of a novel ozone delivery system (HeaIOzone, CurOzone USA) with or without a final restoration for the management of primary occlusal fissure carious lesions (POFCLs) in general dental practice. The study data was collected over a 4-month period.

MATERIALS AND METHODS

Study population

Prior to the commencement of the study, ethical approval was obtained from the local Ethics Committee. The data sets were obtained from a total of 579 Primary Occlusal Fissure Carious Lesions (POFCLs) in 173 patients who presented to UKSmiles over a 4-month period from December 2001 through to the end of March 2002.



Figure 1 Appearance of an occlusal surface, with pit and fissure caries present

MATERIALS AND METHODS - Equipment used

Intra-Oral Camera & Image Management System

All patients were examined with an intra-oral camera (DPS, UK), with the video image displayed on a 40" Sony screen. This gave a final magnification of approximately x25 to x30. Where primary caries was detected, images of these teeth were captured into a video image storage system (Schick, USA). The Schick system allows annotation of images, and this was used to locate the points used for DIAGNOdent (KaVo) assessment.



Figure 2 DPS Intra-Oral Camera System (UK)



Figure 3 Image of a tooth with data annotation

The ProphyFlex

The original protocol developed by the Ozone Research group in Queens University Dental Department, Belfast, Northern Ireland was followed. They thoroughly cleaned each surface and lesion prior to assessment and measurement with the DIAGNOdent. The DIAGNOdent is very sensitive to caries, colours, stains, calculus and composite fillings. To avoid false readings, all stains and debris were removed with a sodium-bicarbonate slurry system under pressure (the ProphyFlex System, KaVo, Germany). This process only takes a few seconds for each tooth surface. The surface was then washed and dried. The surface was then photographed and assessed with the DIAGNOdent.



Figure 4 The ProphyFlex

The DIAGNOdent

The DIAGNOdent (KaVo, Germany) was used to detect and quantify the severity of POFCs. The instant reading indicates the real time value that the probe tip is measuring, whilst the peak value refers to the highest level scanned on the tooth or surface. Variation in the tip angulation over the carious lesion can also be used to track the direction of caries penetration in a developing lesion. The peak values from several sites on an occlusal surface were recorded. Intra-oral photographs were used to record the position of the readings and the peak values for accurate re-assessment. The values obtained were subjected to statistical analyses.



Figure 5 DIAGNOdent (KaVo, Germany)

In addition to the DIAGNOdent values, each lesion measured was assessed as to its treatment needs. Various indices have been used in past studies, and a Clinical Severity Index (CSI), developed by Professor Edward Lynch (Queens University, Belfast) & Dr Julian Holmes (UKSmiles, Wokingham) was used to determine how long each lesion was to be treated with ozone. This varied from 10 seconds for the lowest (4) CSI score to 40 seconds with the highest (1) CSI score. The CSI was based on the Ekstrand index for clinical caries detection. The Ekstrand Index was modified to allow easy and fast indexing of lesions in a general dental practice environment, so that once this pilot study was completed, other general dental practices could send in data for an extended study. The Ekstrand Index has 7 groups as follows;

0	No or slight change in enamel translucency after prolonged air drying (>5s)
1	Opacity (white) hardly visible on the wet surface, but distinctly visible after air drying
1a	Opacity (brown) hardly visible on the wet surface, but distinctly visible after air drying
2	Opacity (white) distinctly visible with out air-drying.
2a	Opacity (brown) distinctly visible with out air-drying.
3	Localised enamel breakdown in opaque or discoloured enamel and or greyish discolouration from the underlying dentine.
4	Cavitation in opaque or discoloured enamel exposing the dentine beneath

Table 1 The Ekstrand Index

The modified index which was adopted, was called the Clinical Severity Index and now has 4 groups as follows;

Index	Assessed Treatment Needs	Tx (sec)
1	Lesion requiring drilling and filling (define this as deemed to have infected dentine where clinical infected demineralisation of the underlying dentine is deemed to be present)	40 seconds with O₃
2	Lesion possibly requiring drilling and filling (defined as possibly deemed to have infected dentine where clinical infected demineralisation of the underlying dentine is possibly considered to be present)	30 seconds with O₃
3	Lesion requiring a pharmaceutical approach but not drilling and filling (defined as deemed to have infected demineralised dentine which is reversing and getting smaller. This scenario is where clinical remineralisation of the underlying dentine is considered to be in the process of remineralising the demineralised dentine but is not yet complete)	20 seconds with O₃
4	Lesion arrested (defined as deemed to have had infected dentine which reversed and where clinical remineralisation of the underlying dentine is considered to be complete, with no infection remaining in the dentine)	10 seconds with O₃

Table 2 Clinical Severity Index

Data Recording & Retrieval

The data for each patient was collated into the Practice Clinical Management system (Software Of Excellence, UK). A custom record screen for each patient was programmed to allow easy data recording and retrieval.

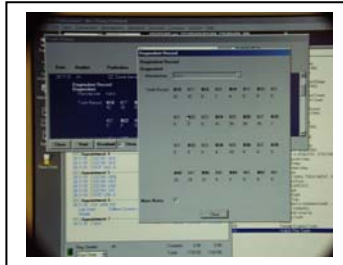


Figure 6 SOE DIAGNOdent data

The HealOzone

The HealOzone (CurOzone, USA) ozone delivery system is a portable apparatus. The unit comprises an ozone generator that delivers ozone (O_3) at a concentration of 2, 200 ppm (Figure 2). A vacuum pump pulls air through the generator at 615 cc/min to supply ozone to the lesion and purges the system of ozone after ozone treatment.



Figure 7 Ozone delivery system (HealOzone, Curozne, USA)

A disposable removable silicone cup (diameters ranging between 3 and 10 mm), attached to the handpiece, is provided for receiving and exposing a selected area of the tooth to ozone. The tightly fitting cup seals the selected area on the tooth to prevent escape of ozone (Figure 3). Unreacted ozone is sucked out of the sealing cup by the vacuum system, whilst the cup is still adapted to the POFCLs, through an ozone destructor (manganese (II) ions) that converts the ozone to oxygen. The system then delivers a liquid reductant to further neutralise any possible residual ozone and facilitate the remineralisation process.



Figure 8 Handpiece with a cup

Data Recording & Retrieval

The data set values were transferred into an Excel (Microsoft UK) spreadsheet to allow statistical analysis. The pilot study data consisted of the patients ID, age, date of first assessment, tooth ID (FDI tooth notation) assessed, first DIAGNOdent value(s) DVs and the CSIs (CSI start value). The second data set recorded the date of re-assessment, the DIAGNOdent value (Dve) at re-assessment, and the CSIE (CSI end value). The spreadsheet then calculated the time elapsed between the two dates, the changes in the DIAGNOdent and CSI values. As patients were added into the study data, the average age, time between assessments and Index (DIAGNOdent & CSI) changes were all automatically updated.

The study involved 193 patients with 2 or more POFCLs which were randomly allocated to one of the 2 groups (Table 3).

Baseline DIAGNOdent measurements and CSI	
Group 1	Group 2
Application of O3	No O3 Application
2 month follow up & review DIAGNOdent measurements and CSI	
Group 1	Group 2
Re-application of O3	No O3 Application

Table 3 Schematic diagram of the study Patient recruitment

If patients presented between the start and re-assessment appointments with any form of discomfort, POFCLs for either group immediately treatment with conventional drilling and filling procedures was planned. Patients in all the above groups used a standard toothpaste and mouth rinse 1,100-ppm sodium fluoride (Curozone toothpaste and mouth rinse, Natural White, U.S.A) for at least 4 weeks after the first application of ozone to treated tooth surfaces.



Figure 9 Re-mineralising toothpaste & mouth rinse

MATERIALS AND METHODS *Materials Used*

Glass Ionomer / Composite Fissure Sealants

After a period of remineralisation, some patients requested the placement of a restoration for aesthetic reasons. A glass ionomer cement was used (Dentsply, UK) in this situation. For some other occlusal lesions, a fissure sealant was used (Ultradent, USA)



Figure 10 Some of the materials used to restore selected lesions after completion of this study

MATERIALS AND METHODS

Patient recruitment, assessment and data collection

Each patient that attended UKSmiles dental practice was asked if they would like to join this research project. No financial inducements were offered. After an initial examination to assess suitability for inclusion into this project, the project was explained to the patient. Appropriate paperwork was then completed and consent obtained. The tooth surfaces were cleaned with the ProphyFlex and then identified lesions were photographed, assessed with the DIAGNOdent and scored with the CSI. The data was collated into a custom screen within the practice management system, and each lesion was either treated with the HealOzone unit, or left as a control. The duration of treatment with the HealOzone unit for each lesion was determined by the CSI. At the end of treatment, each patient was given a standard Patient-Kit that complements the HealOzone treatment protocol and instructions as to its use. A date was set for recall and review, and booked into the practice management recall system.

On average, each patient attended a recall 2 months later. The previously treated teeth were identified, re-cleaned, and re-assessed with the DIAGNOdent and the CSI. The new values were recorded, and entered into the data collation system.

This data was then transferred to an Excel spread sheet, where statistical analysis was performed.

MATERIALS AND METHODS

Statistical analyses

DIAGNOdent Values

The values of the DIAGNOdent readings recorded at baseline and after 2 months were used for data analyses. In the statistical analyses of the data sets, the ozone-only group was compared to the control group with a paired statistical test.

Data sets were built up to show the mean values for the change in the DIAGNOdent values for each age group, and the differences between the ozone treated lesions compared to those left as controls. Data sets were built for the time elapsed between the initial measurement, and the re-assessment.

CSI Values

The differences in the number of lesions changing to a less severe index were tested. Data sets were built for each age group, and the differences between the ozone treated lesions compared to those left as controls.

It is important to realise that it is impossible to have a true “untreated” control surface lesion in this study, as the action of the patient kit which was dispensed to each patient seen will have an effect on the whole mouth. However, what is tested in this study is if ozone with or without improved oral care can reverse decay in an occlusal surface, and what, if any, difference there is in these two groups.

RESULTS

DIAGNOdent scores (Table 4)

The mean DIAGNOdent scores for each age group are shown in Table 4. At baseline, the DIAGNOdent readings were greater for the younger age groups and oldest, which could be related to be tooth development stage and structure. The mean DIAGNOdent readings for the control group tended to increase after 2 months when compared to baseline ($p < 0.01$). In contrast, the mean DIAGNOdent readings in the ozone-only group tended to decrease during the study when compared to baseline measurements ($p < 0.01$). There were statistically significant differences in the changes in DIAGNOdent readings between the two groups at 2-month reviews ($p < 0.001$).

Age Group	0-6	7-14	15-25	26-35	36-45	46-55	55-85		
Number of Patients	64	39	19	15	18	15	3		n = 173
Number of Lesions	152	127	153	37	59	42	9		n = 579
Average Lesions/Pt	2.4	3.3	7.1	2.5	3.3	2.8	3	3.4	
Average DV at start	84	76	43	41	36	43	73	56	
Average DV O₃at finish	18	22	14	15	18	22	47	22	Treated c O₃
Average DV O₃ Change	66	54	29	26	18	21	26	34	Treated c O₃
Average DV O₃at finish	86	79	44	39	41	44	77	58	Control Grp
Average DV O₃Change	2	3	1	-2	5	1	4	2	Control Grp

Table 4 DIAGNOdent data

CSI scores ~ at 2 months recall (Table 5)

The CSI scores are shown in Table 5.

The majority of lesions reversed from CSI 2 to 3 in the ozone group, when compared to the control group ($p < 0.001$). The control group over all showed signs of continued caries development. The effects of the use of a remineralising tooth paste and mouth rinse without eliminating the micro-organism niches is seen to be limited.

Age Group	0-6	7-14	15-25	26-35	36-45	46-55	55-85	Av	
Number of Patients	64	39	19	15	18	15	3		n = 173
Number of Lesions	152	127	153	37	59	42	9		n = 579
Average Lesions/Pt	2.4	3.3	7.1	2.5	3.3	2.8	3	3.4	
Average CSI at start	1.9	1.8	1.9	1.8	1.5	1.6	1	1.6	
Average CSI O₃at finish	3.5	3.3	3.9	3.4	3.3	3.4	3.5	3.5	Treated c O₃
Average CSI O₃ Change	-1.6	-1.5	-2.0	-1.6	-1.8	-1.8	-2.5	-1.8	Treated c O₃
Control Av CSI at finish	1.3	1.7	1.8	1.8	1.4	1.4	1	4	Control Grp
Control Change CSI at finish	+0.6	+0.1	+0.1	0	+0.1	+0.2	0	+0.16	Control Grp

Table 5 CSI data

DISCUSSION

Previous research has shown that remineralisation *in-vivo* can be achieved when the correct oral conditions exist, but it can be unpredictable. The main clinical problem with non-invasive or pharmaceutical approaches to the management of caries seems to be the difficulty in suppressing or eliminating micro-organisms for a long enough period of time to allow caries reversal and remineralisation of the lesion. After treatment with pharmaceutical agents, organisms may re-colonise in POFCLs.

Caries reversal is associated with several factors including the level of microbial reduction and the oxidant effects of ozone on POFCLs. The dramatic reduction in microbial flora will have eradicated the ecological niche of the acidogenic and aciduric micro-organisms. This shifting of microbial flora to normal oral commensals would predominantly allow remineralisation to occur within the carious process. Previous studies have also shown that ozone reduces bacterial by-products and metabolites. This has two effects. First, the by-products cannot cause further demineralisation of the lesion. Second, the removal of important bacterial metabolites denies other bacterial types in the lesion their important nutrients. In this way, ozone has a dual effect when used to control caries.

In a recent paper on the clinical management of primary root carious lesions (PRCLs) using ozone *in-vivo*, Baysan, A *et al* (2001) the results were staggering in comparison to other agents and studies. Just 10 seconds of exposure to ozone reduced the number of colony forming units (cfu) from 7,000,000 to 4,000 after 10 seconds, and less than 1 after 20 seconds! Since less than 1 cfu cannot exist, it is safe to postulate that 20 seconds or more treatment with ozone is effective in sterilising the lesions examined in this study.

Mean \pm SE log₁₀ cfu + 1 before and after ozone application for a period of either 10 or 20 s

Groups	10 Seconds	20 Seconds
Control Samples	7.00 \pm 0.24	6.00 \pm 0.20
Test Samples	4.35 \pm 0.49	0.46 \pm 0.25

Ozone application either for a period of 10 (99%) or 20 s (99.9%) substantially killed micro-organisms in PRCLs

Table 6; Baysan, A *et al* (2001)

Other studies have shown an oxidant (sodium hypochlorite) can improve the remineralisation potential of demineralised dentine. Inaba *et al*, (1995) found that the use of an oxidant (10% sodium hypochlorite) on demineralised root dentine lesions improved their potential to remineralise since sodium hypochlorite is a non-specific proteolytic agent and was effective in removing organic components in the lesions. Further published research in this field by Inaba *et al*, (1996) showed that when root dentine samples were treated with this oxidant for 2 minutes, the permeability of the lesion to fluoride ions increased. The conclusion of this study was that removal of organic materials from dentine lesions was an acceptable approach to enhance remineralisation.

In this light, part of the dramatic remineralisation results shown after ozone application in this and other studies can be accounted for, as it is known that ozone is one of the most powerful oxidants available. It may also indicate that ozone has the ability to remove proteins in carious lesions, and to enable calcium and phosphate ions to diffuse through the lesions, a phenomenon resulting in remineralisation of the majority of POFCLs after ozone application in this study.

After the initial elimination of the numbers of total micro-organisms, decolonisation of the micro-organisms may be retarded by the lack of available organic substrates essential for the metabolism of cariogenic bacteria. As the ecological niche of these acidogenic and aciduric micro-organisms would be severely disrupted, this will in turn interfere with recolonisation and re-growth by this specific micro flora. This may result in long-term suppression of acidogenic and aciduric micro-organisms in POFCLs. Previous workers, for example Emilson (1981), have also reported that after a short-term intensive pharmacological treatment, there is significant *in-vivo* suppression of bacterial growth.

One area that further research is needed, is to what depth the remineralisation process penetrates. Is it just within the surface layers, or does remineralisation penetrate to the full depth of the treated lesion? This is obviously very important factor in the clinical management of caries in a general dental practice setting, and one that patients need reassurance about. Baysan, A (2002) postulated that surface hypermineralisation is less likely to occur following the application of ozone. She argued that since ozone is a strong oxidant, it would undoubtedly oxidise PRCL biomolecules and hence open dentine channels in the lesions, helping the diffusion of calcium and phosphate ions throughout the depth of the lesion. Previous research by Martens and Verbeeck (1998) reported that low concentrations of fluoride have the capacity to remineralise carious lesions to their full depth. It is possible that ozone treatment, followed by low mineral concentration rinses is the only treatment required. This argument is supported by the results of this study. The requirement for a restoration is only cosmetic, as the "healing" ability of the treatment in carious lesions is well demonstrated. Previous studies (Lynch, E *et al* 1999) have also shown that a patient's saliva contains all the bio-available mineral components required for the remineralisation process, so possibly ozone treatment alone, without the adjunct specialised toothpastes and mouth rinses may be all that is required.

The Clinical Severity Index developed by Professor Edward Lynch and Dr Julian Holmes was necessary to avoid errors from the DIAGNOdent values. I have mentioned that three lesions showed increased DIAGNOdent values after treatment with ozone. These lesions consisted of exposed dentine. In these lesions, it is postulated that the remineralisation process has led to an increase in the stain or colouration of the lesions. The DIAGNOdent is very sensitive to stains, which is why the cleaning protocol prior to DIAGNOdent assessment is so important. But in cases where remineralisation leads to an increase in the 'staining', it is possible to assume the caries process has continued in the lesion. By measuring a CSI for each lesion and recording this data, this potential error is reduced.

The two data sets presented show how caries develops in different age groups. The age groups were chosen with care to minimise the number of data sets, and to show the average age-related dental development. The 0-6 age group represent the majority of child patients with only primary dentitions. Previous studies have shown that as the teeth are smaller, and the nerve tissue cavity large, decay quickly spreads to cause nerve tissue death and necrosis. Ozone has been shown to be very effective in this age group. The 7-14 age group represents the mixed dentition stage, and the 15-25 age group will have developing pre-molar, second and third molar surfaces erupting. All these developing occlusal fissure surfaces are prone to potential decay but improved oral hygiene, coupled with ozone treatment and specialised oral mouth rinses would seem to provide the greatest possible protection in this study. The last age group, the 56-85 age group, were bundled due to few numbers. Decreased manual dexterity, as well as dry mouth could account for most of the developing lesion seen in this group. However, ozone treatment provided a great improvement for the treated lesions.

The data sets and the statistical analysis has lead to the hypothesis that the change in the DIAGNOdent readings from the initial lesion assessment, to the 2 month re-assessment could be regarded as a “healing index” or “remineralisation index”. Until a technique or a technology is developed that allows us to ‘look’ inside a treated tooth to show us how far the remineralisation process has penetrated, this sort of index may not be reliable. However, Baysan, A *et al* (2002) has already shown that the DIAGNOdent readings and the ECMIII values have a very close correlation. That is, any changes in the ECMIII values that indicate caries reversal and re-mineralisation, are mirrored in the changes in the DIAGNOdent values. Three lesions in this study showed increased DIAGNOdent values, and it was expected that we would find these lesions with advancing caries. In fact, this was not the case. The CSI showed that these three areas were hard, re-mineralised lesions, and since they consisted of exposed dentine, the remineralisation process had lead to increased staining of these lesions. These three lesions illustrate the value of the CSI used along side the DIAGNOdent to score the lesions.

The results acquired from this initial ozone study mirror those from studies in research centres. It is known that the delivery system used is safe, and cannot expose the patient, the operator, or any surrounding team members to ozone. The use of ozone is very cost-effective, as ozone is only made by the unit when required. The initial investment required is high, but an unpublished study by Holmes, J & Lynch, E (2001) has shown that patients are prepared to pay for this treatment. Published research into patient acceptability by H.Domingo *et al* (2001) show that this treatment modality is well accepted, as there are none of the usual problems associated with conventional treatment (injections, pain, drilling, noise, smells, etc) and that patients are willing to pay an increased fee as an alternative to traditional treatment. Treatment with ozone is also cost-efficient, and very time-efficient.

Current research with ozone has opened up a whole range of potential treatment protocols. The new treatment protocols do not require removal of the diseased or carious tissue. Published papers have shown that after ozone treatment for just 10 seconds, debris and bacteria impacted in a fissure can mineralise, ‘sealing’ to fissure against decay. This morphological change in the surface of a tooth can be measured by a technique developed by Jovanovski V. and Lynch E. (2000) at Queen’s University, Belfast, to an accuracy of 1 micron.

Previous studies and literature reviews have shown that a surface that re-mineralises is never seen to decay. So it follows that if this morphological change and re-mineralisation is predictable, the current protocol could be modified, by modifying the cleaning stage of the treatment protocol to the use of bristle brushes only. Whilst this may appear on the face of it to be a retrograde step, the use of a brush to remove the bulk of the impacted debris will allow ozone induced remineralisation of the remaining debris to begin. By leaving the debris impacted into the fissures and sterilising it with a 20+ second exposure to ozone with the HealOzone unit, then there would be no need to place fissure sealant materials which are prone to bonding failure due to poor preparation, compromised chemical bonding at the margins, and where debris has been incompletely removed before enamel etching. In addition, the problem of inadequate drying or isolation prior to placement of a resin-sealed type restoration or preventive restoration technique would be eliminated; and with it, the preventive care of patients could be improved.

At UKSmiles, we have treated successfully patients from a wide age group. All our patients want the benefits of modern dental care that is non-invasive, painless, and does not require the use of a drill. The young patient’s attitude to future dental care is often highly influenced by traumatic dental care in the early years. And with increasing age and anxiety about dental treatment, dental care is often postponed until more radical treatment is required due to past experiences. These situations can be overcome using early intervention strategies. In this respect, the use of ozone should be considered especially for the young, medically compromised patients, those who are in long-term care, domiciliary care patients and homebound people (Baysan *et al.*, 2001). There is no injection or tissue amputation involved in ozone treatment and the ozone delivery system is portable. Research by H.Domingo *et al* (2001) has already shown that this treatment modality is acceptable to the public.

To conclude, this pilot study has shown that this novel treatment regime using ozone is capable of clinically reversing POFCLs and can be considered a revolutionary alternative to conventional "drilling and filling" or "amputation" therapy for caries treatment.

(All lesions were treated with ozone at the end of this study, and not left to further decay.)

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