The use of QLF-D™ (Quantitative Light-induced Fluorescence-Digital™) as an oral hygiene evaluation tool to assess plaque accumulation and enamel demineralisation in pre orthodontic patients with suboptimal oral hygiene

Thesis submitted in accordance with the requirements of the University of Liverpool for the Degree of Doctor of Dental Science (Orthodontics)

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CONTENTS

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2.0 ABSTRACT

Aim: To assess the use of the Quantitative Light-induced Fluorescence-Digital Biluminator TM (QLF-D^{TM}) as an oral hygiene evaluation tool to assess plaque accumulation and demineralisation in patients with poor oral hygiene.

Design: Randomised clinical trial

Settings: Liverpool University Dental Hospital

Subjects: 60 patients (32 females, 28 males) with inadequate oral hygiene referred to dentists or hygienists for oral hygiene reinforcement before the start of orthodontic treatment were recruited for the trial. The median age of patients was 13.8 years with an IQR range from 11.1 to 26.7 years.

Methods: The patients were randomly allocated at baseline (T1) to receive oral hygiene reinforcement (OHR) at three consecutive appointments (T1-T3) using the White light (WL) or Quantitative Light-induced Fluorescence (QLF) images, taken with the QLF-D^{TM} device (Inspektor Research Systems BV, Amsterdam, The Netherlands), as visual aids. The standard of oral hygiene was assessed on the QLF images using customised software to provide quantitative scoring of fluorescence loss (ΔF) and plaque coverage (ΔR30) at each appointment. Inter-examiner reliability assessments were conducted by four examiners using QLF and WL images from 35 images of 7 patients. One examiner assessed the images on a second occasion two months later to ascertain the intra-examiner reliability. A debriefing questionnaire, distributed on completion of the study, was used to ascertain the patients' perspectives of the QLF-D^{TM} images.

Results: There were no significant differences in plaque accumulation (p=0.81) or demineralisation (P=0.69) between the WL and QLF groups. There was no significant change in demineralisation over the three visits in either group. However, there was a significant reduction in plaque in both groups (P<0.001) with a mean percentage change in ΔR30 of 51.8% and 95% CI of 40.36% to 63.26%.

All of the participants in the QLF group found being shown the images helpful and were able to see areas of demineralisation and plaque accumulation. 92.5% of the QLF group and 76.7% of the WL group expressed it would be useful to receive such OHR for the full duration of orthodontic treatment.

The inter-examiner reliability of QLF image assessment, using ICC, was 0.987 and 0.773 for ΔR30 and ΔF respectively. The inter-examiner reliability of WL image assessment, using kappa, ranged from -0.0932 to 0.447. The intra-examiner reliability scores were excellent with an ICC of 1.0 and 0.995 for

ΔR30 and ΔF respectively on the QLF images. The kappa score of demineralisation assessment on the WL images was 1.0.

Conclusion: QLF-D^{TM} can be used as an effective tool to assess plaque accumulation and detect and monitor demineralisation in patients with suboptimal oral hygiene to start orthodontic treatment. The image analysis demonstrated high levels of inter- and intra-examiner reliability. OHR using WL or QLF images as visual aids was effective in reducing plaque coverage in patients with suboptimal oral hygiene. There was no difference in the level of demineralisation or plaque coverage between the QLF and WL groups. More patients reported that the QLF images were useful than patients shown WL images.

Summary: OHR using WL or QLF images was an effective tool in reducing plaque in poor OH patients and reported QLF images were informative.

3.0 LITERATURE REVIEW

3.1 Dental enamel

Dental enamel consists of a highly crystalline structure arranged in rods. It is largely inorganic, with the main component being hydroxyapatite crystal of calcium phosphate, $Ca_{10}(PO_4)_6(OH)_2$. This inorganic component comprises 86-95% of the volume, which results in enamel being highly susceptible to demineralisation. The organic component, largely proteinaceous material, comprises 1-2% of the volume, with water constituting the remaining component (1). This results in spaces termed as pores, which allow the movement of ions within enamel and the surrounding oral environment.

3.2 Dental Plaque

Dental plaque can be defined as the diverse community of micro-organisms found on the tooth surface as a biofilm, embedded in an extracellular matrix of polymers of host and microbial origin (2). These communities are in a dynamic equilibrium with their environment, and there can be significant re- assortment and rearrangement of the composition and metabolic activity of these microbial consortia in response to changes in the biology of the mouth (e.g. eruption of teeth; flow of saliva; subversion of the host defences) and in the lifestyle of the individual (e.g. in response to smoking, dietary alterations, or to the side effects of medication, etc) (3). The composition of dental plaque also varies on distinct anatomical surfaces (e.g. fissures, approximal and smooth surfaces, gingival crevice, dentures) due to the prevailing physical and biological properties of each site(2).

Dental plaque accumulates preferentially at stagnant sites that afford protection from the vigorous removal forces that apply in the mouth. Distinct phases of development can be recognised (2), including:

(a) Adsorption of host and bacterial molecules to the tooth surface. The acquired pellicle forms immediately following eruption or cleaning and directly influences the pattern of initial microbial colonisation.

(b) Passive transport of oral bacteria to the tooth surface. Weak physicochemical interactions between the microbial cell surface and the pellicle-coated tooth create a weak area of net attraction that facilitates reversible adhesion. Subsequently, strong, short-range interactions between specific molecules on the bacterial cell surface (adhesins) and complementary receptors in the pellicle can result in the irreversible attachment. The pioneer species, frequently streptococci strains, adhere to the acquired pellicle and colonise (4)(5).

(c) Co-adhesion of later colonisers to already attached early colonisers. This stage involves specific interbacterial adhesin-receptor interactions (often involving lectins) and leads to an increase in the diversity of the biofilm and to the formation of unusual morphological structures, such as corn-cobs and rosettes. Co-adhesion may also facilitate the functional organisation of dental plaque. Bacteria engage in a range of antagonistic and synergistic biochemical interactions(4)(5).

(d) Multiplication of the attached micro-organisms. Cell division leads to confluent growth and, eventually, a three-dimensional spatially and functionally organised, mixed-culture biofilm. Polymer production results in the formation of a complex extracellular matrix made up of soluble and insoluble glucans, fructans and heteropolymers. Such a matrix is a common feature of biofilms and makes a significant contribution to the known structural integrity and general resistance of biofilms; the matrix can be biologically active and retain nutrients, water and key enzymes within the biofilm (5).

(e) Active detachment. Bacteria can respond to environmental cues and detach from surfaces, enabling cells to colonise elsewhere (5).

The plaque colony is produced in layers, beginning with pioneer species which adhere to pellicle followed by an increasingly complex collection of microflora ultimately producing a biofilm which includes filamentous and obligate anaerobic bacteria, many of which adhere to the tooth structure directly (5). When plaque calcifies it becomes calculus, and this can occur with either sub- or supra-gingival plaque deposits.

3.3 Role of plaque in enamel demineralisation

Dental caries development is considered to involve a triad of indispensable factors: bacteria (dental plaque), carbohydrates (the diet), and susceptible teeth (the host) (6). The aetiology of caries is based on W.D. Miller's 'chemico-parasitic' or 'acidogenic' theory 1982 that postulates that it is acid produced by plaque bacteria fermenting dietary carbohydrates that lead to decalcification of teeth and this can subsequently result in degeneration of the organic matrix by bacterial proteolytic action (7). Epidemiological studies have shown a strong association between the presence of mutans streptococci and the initiation of smooth surface and fissure caries, whilst lactobacilli are implicated as important contributory bacteria in tooth decay, but their role in the induction of lesions is not well supported (8). However, according to the extended caries ecological hypothesis, the caries process consists of 3 reversible stages (9). The microflora on clinically sound enamel surfaces contains mainly non-mutans streptococci and Actinomyces, in which acidification is mild and infrequent. The environment is compatible with the equilibrium of the demineralisation/

remineralisation balance or shifts the mineral balance toward net mineral gain (dynamic stability stage) (9). When sugar is frequently supplied, acidification becomes moderate and frequent and may enhance the acidogenicity and acidurance of the non-mutans bacteria adaptively. In addition, more aciduric strains, such as 'low-pH' non-mutans streptococci, may increase selectively. The microbial acid-induced adaptation and selection processes may, over time, shift the demineralisation/ remineralisation balance toward net mineral loss, leading to initiation/progression of dental caries (acidogenic stage) (9). Under severe and prolonged acidic conditions, more aciduric bacteria become dominant through acid-induced selection by temporary acid-impairment and acid-inhibition of growth (aciduric stage) (9). At this stage, mutans streptococci and lactobacilli as well as aciduric strains of non-mutans streptococci, Actinomyces, bifidobacteria, and yeasts may become dominant. Many acidogenic and aciduric bacteria are involved in caries. Environmental acidification is the main determinant of the phenotypic and genotypic changes that occur in the microflora during caries (9).

Red fluorescence in plaque has been observed in QLF images when the plaque was relatively old and has been assumed to be associated with caries risk (10). The Van der Veen et al. 2006 study showed that red-fluorescing plaque comprised only about 62% of total plaque and was mainly found at the gingival margins, which correspond to the location of initial plaque formation or a so-called plaque stagnation site (11). Acidogenic plaque bacteria particularly the mutans streptococcal strains generate acid when metabolising sugars and which then dissolve the mineral phase of enamel if the pH falls below a critical level of pH5.5 (12). Saliva has a protective effect due to its buffering and antimicrobial properties, however, as the plaque deposits increase, such factors have less of influence(5). The detection of plaque is crucial for both the patient and clinician. If a patient is to clean their mouth effectively, they must be able to identify areas that require more attention and those which are clean (4). Patients frequently have difficulty localising the deposits to enable optimal levels of oral hygiene to be achieved consistently. Clinicians making decisions about the prognosis of a patient's gingival condition, their commitment to an oral hygiene programme or their suitability for orthodontic treatments will need to be able to assess plaque levels.

White spot lesions remain a serious problem in orthodontics (13)(14)(15). White spot formation during orthodontic treatment has been attributed to the effect of prolonged accumulation and retention of bacterial plaque (16). In orthodontic treatment, the brackets and archwires are significant plaque stagnation sites. Conventional oral hygiene is more difficult due to the fixed appliances that increase plaque accumulation and retention as it is hard to visualise plaque stagnation sites. Clearance of plaque by saliva and cheeks is also further reduced. Progression rate between traditional caries formation and white spot lesions induced by deficient oral hygiene

combined with fixed orthodontic appliances seems to be different (17). The latter has a rather superficial and more rapid character and can become evident within one month after placement of fixed appliances (16). Gorelick and co-workers found that up to 50% of patients undergoing fixed appliance therapy developed white spot lesions during treatment (13). Mizrahi's similar cross-sectional survey found an incidence of white spot lesions of 12% following orthodontic fixed appliance treatment (18). The reported prevalence of white spots after fixed appliance treatment varies between 2 and 96 %(16).

3.4 Demineralisation

3.4.1 Description

Demineralisation of teeth occurs due to the bacterial fermentation of dietary sugars, which produces organic acids. Bacteria present in plaque, such as mutans streptococci and lactobacilli , lower the pH of the tooth surface to below the critical level, which causes dissolution of the mineral component (19). Decalcification is defined as loss of calcified tooth substance, and it occurs when the pH of the oral environment favours diffusion of calcium and phosphate ions out of the enamel. This dissolution follows the production of acid by bacterial plaque and results in an altered appearance of the tooth surface. The early lesion is an opaque white spot, which in active lesions appears chalky, and if mineral loss continues, frank cavitation may result (20). At this stage the lesion is past the point of spontaneous repair, and restorative intervention becomes necessary. The Stephan curve demonstrates the decrease in pH that occurs following consumption of sugary foods.

While the coexistence of four factors, namely, bacterial plaque, fermentable carbohydrates, a susceptible tooth surface and time, are necessary for demineralization to occur, salivary parameters such as pH, flow rate and buffer capacity can influence the degree of enamel mineral loss following an acid challenge, the rate of progression of demineralization, and the likelihood of repair (19). Following ingestion of a fermentable substrate, the pH of plaque fluid falls as acids are produced, and then recovers as salivary buffering occurs. As the frequency of consumption increases, the enamel surface may be exposed to overlapping episodes of acid challenge without significant intervening repair, resulting in a net loss of mineral over time (19).

The microradiography , polarised light experiments , microhardness data , electron microscopy, studies have identified the structure of early enamel caries into four zones (21). The zones are related to the degree of demineralisation and changes in mineral content that have occurred (13)(21)(22).

Surface zone

The zone is the outer surface layer of intact relatively unaffected enamel, displaying negative birefringence to polarised light, radio-opacity, with some focal holes and 10% mineral loss. This layer continuously changes mineral composition as a result of the changes in pH. Remineralisation may occur, resulting in the surface zone being porous although mineral rich. This allows the tooth surface to remain relatively intact despite substantial subsurface mineral having lost.

Body

The body of the lesion, with positive birefringence to polarised light, radiolucency and significant mineral loss of 24% is just below the surface and has a porosity that ranges from 5-25%. A greater amount of dissolution has occurred in this area, involving destruction largely of all the rods. These spaces are filled with water and bacteria. The area of demineralisation in the region can often be seen as a white spot; frequently it is termed as a 'white spot lesion'. However, extrinsic stains from tobacco, food and bacteria can accumulate in this area and cause the demineralisation to be brown in appearance.

Dark zone

The dark zone, with positive birefringence to polarised light, radio-opacity and 6% mineral loss. The mineral dissolution is mainly calcium and phosphate ions, with a greater number of rod structures and cross striations involved. The porosity is 2-4%.

Translucent zone

Finally, the translucent zone with negative birefringence to polarised light, radio-opacity and only 1% mineral loss and is the deepest part of the lesion. Dissolution of mainly magnesium and calcium ions occurs from the peripheral rod structures. The porosity is about 1%.

Demineralisation is the first stage of dental caries. Early demineralisation is reversible (Angmar-Mansson and ten Bosch, 1993), whereby when the pH in the oral cavity is restored to neutral, enamel remineralisation will occur. This process is aided by the protective components of saliva, including bicarbonate ions and proteins, which increases the pH. Remineralisation is also aided by fluoride ions supplied from external sources such as toothpaste and mouthwash. The enamel surface undergoes a regular dynamic process of ion exchange as the pH in the oral cavity varies. Caries may progress if the rate of ion loss from enamel, namely demineralisation, occurs at a greater rate than

remineralisation and would lead to dentinal involvement and ultimately the destruction of the tooth structure.

In studies conducted by Mizrahi et al. (20) to determine the prevalence of enamel opacities in school pupils who had not received any orthodontic treatment in a low fluoride area (23)(24), the results showed that 83 to 85 percent of the pupils had some evidence of enamel opacities (23). These results are in agreement with the findings of Hurme and Murray and Shaw (25)(26). A cross sectional study by Gorelick et al. carried out to determine the prevalence and severity of enamel opacities in a group of patients before starting and after receiving orthodontic treatment (18). The Mizrahi study with a sample consisting of 527 patients examined before and 269 patients examined after completion of multibanded orthodontic treatment showed that there was a significant increase in both the prevalence (before, 72.3 percent; after, 84.0 percent) and severity (Opacity Index: before, 0.125; after, 0.200) following completion of orthodontic treatment. Male patients experienced a significantly higher increase in the severity of enamel opacities following orthodontic treatment. There was no significant sex differential in the prevalence of enamel opacities either before or after orthodontic treatment. This study showed that orthodontic treatment with multibanded appliances contributed to the development of new areas of enamel demineralisation and to an increase in the severity of enamel opacities as measured by the Opacity Index (18). Another study of 173 patients undergoing orthodontic treatment with an edgewise-light wire technique investigated to determine the incidence of dental caries at the end of therapy at 19 months using caries index. The results indicated that, on an over-all basis, the number of new enamel alterations was small and the appliances did not markedly influence the total caries frequency. The correlations between caries incidence and age, duration of treatment, and initial caries experience were not significant. They found higher caries index recorded for male orthodontic patients and this was a result of a lower standard of oral hygiene based on high plaque index and gingival index scores recorded in male patients as compared to female patients (27). These two studies support the theory that female patients take more effort with their oral hygiene during orthodontic treatment than their male counterparts (18)(28).

3.4.2 Demineralisation and orthodontic treatment

The prevalence of demineralisation around fixed orthodontic appliances varies widely with documented rates ranging in the literature between 2-96% (13)(18)(20)(28).This large variation is due to the variety of methods used to assess and score the presence of decalcification, whether idiopathic enamel opacities were included or excluded, and the use or otherwise of a fluoride regime during treatment (20). Gorelick et al. (1982) conducted a retrospective cross-sectional study and

found that 3.6 % of the teeth had white spots in the control group and 10% of the teeth after treatment and that 50% of the patients experienced an increase in white spots. The study discussed the schematic diagram of how white spots at the gingival third were classified but did not explain how they distinguished this from developmental enamel opacity. They had not assessed the treatment group before the start of orthodontic treatment; therefore some of the lesions could have been present before orthodontic treatment. The study found no white spot lesions associated with the lingual canine-to-canine retainer which had been in position for an average of 24 months, despite having calculus around the wire of the canine-to-canine retainer. The authors suggested that accessibility to the free flow of saliva may be a significant factor in avoiding decalcification of enamel. Maxillary molars and second premolars had significantly less de- calcification than their mandibular opponents. Zachrisson and Zachrisson (1971), and Zachrisson (1977) employed a longitudinal design and recorded only new white spots developing, yet despite this, the prevalences they found were 89 and 15 %, respectively (27)(29). Mizrahi (1982), in a similar cross sectional study, found a significant 12% increase in the number of White Spot Lesions(WSL) in a group of patients who had undergone fixed orthodontic treatment compared to untreated controls (18). The severity of lesions, assessed using the opacity index, was also significantly greater. However, this study involved the use of multibanded appliances, which is no longer commonly conducted. An RCT by Stecksen-Blicks et al. (2007) on the use of a fluoridated varnish versus placebo varnish during fixed orthodontic treatment, found 25% of the participants in the control group developed demineralisation over the duration of their treatment (30). Ogaard (1989), compared 51 patients, aged 19 years, who had received orthodontic treatment an average of 5.7 years previously to a matched untreated control group of 47 patients. The prevalence of white spots was 96% and 85% in the orthodontic treatment and control groups respectively, representing an 11% increase. This study also highlights that demineralisation that develops during orthodontic treatment can be permanent, as white spots were still present and visible in a large number of the subjects (28). A retrospective study by Lovrov et al. (2012) (31) looked at 53 participants for WSL and plaque index before , during and post treatment showed 97.5% of teeth before and 73.6% after treatment were free of WSLs. Of all teeth, 24.9% developed new white spot lesions or a rise in their number. New or more numerous WSLs were more common in upper and lower premolars (34.4%) and front teeth (28.1%) than molars (11.8%). WSL incidence during therapy correlated with clinical attachment level, and the oral hygiene and fluoride-use scores. WSLs were graded from intraoral photographs taken before and after treatment. In cross sectional study designs (orthodontic patients after treatment compared with another group of patients who have not had orthodontics), it is difficult to distinguish between idiopathic white spots and demineralization, which artificially increases the prevalence quoted (32).

Developmental white areas, such as fluorosis and enamel hypoplasia, are often incorrectly diagnosed. Such pre-treatment white areas should be distinguished, noted at the outset of orthodontic treatment and be excluded from study values. This is to ensure there are minimal false positive results (33) which would lead to an overestimation of the prevalence. Demineralisation may also be present at baseline, due to suboptimal general mouth care (Figure 4.1).

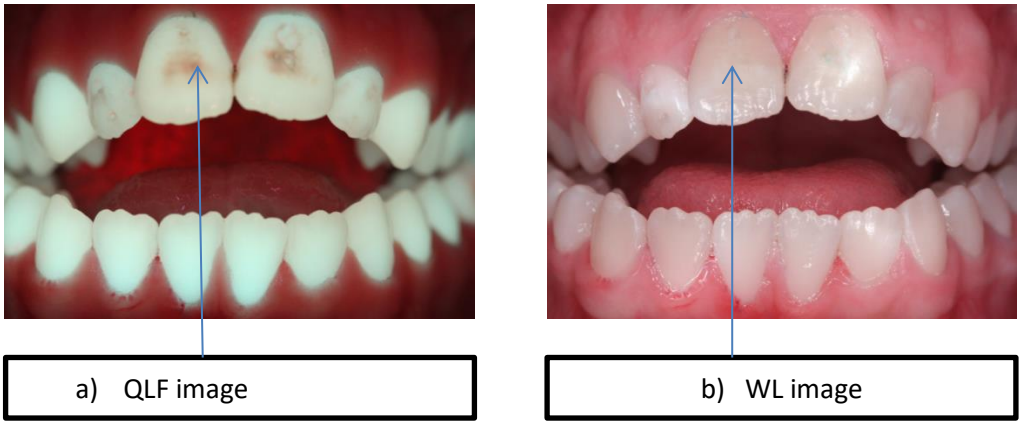

Figure 4.1: Clinical photographs illustrating demineralisation on a QLF image and a WL image.

The presence of fixed appliance components on tooth surfaces such as brackets and bands creates difficulties in keeping teeth clean, leading to the build-up of plaque and WSL formation (14). Compared to caries which takes 6 months to develop the demineralisation progresses quicker and have been demonstrated to be able to present within 4 weeks of appliance placement (17). It can be seen as white and brown areas, of varying size, around the appliance. The severity must be quantified in terms of the size of these areas but also the extent of mineral loss (34).

Additionally, differences in prevalence may be due to the use of various detection tools, which differ in their ability to diagnose demineralisation (15). Earlier studies frequently used direct visual examination as a method of assessment that does not allow reassessment and creates observer bias. More recent studies have reported on the newer equipment available. One such study by Al Maaitah et al. found the prevalence of white spot lesions was 71.7% in 230 subjects post orthodontic treatment assessed using Quantitative light-induced fluorescence (QLF) (35). The prevalence is much greater than the 25% reported by Stecksen-Blicks et al. and Lovrov et al. on assessing digital images(30) (31).

The anterior maxillary teeth were found to be the most affected, with studies reporting the highest incidence of WSLs on the maxillary lateral incisors of up to 23% (13), located adjacent to the gingival margin (36)(37). Frequently the maxillary lateral incisors are crowded palatally, and access to

brushing can be difficult. In a 5-year follow-up study by Ogaard, , WSLs were observed as a cosmetic problem for orthodontic patients (28). Similar findings were published in a 12-year follow-up study by Shungin et al. examining patients who had incidences of WSLs during orthodontic treatment (38). They found that although the size of the lesions reduced over time, they still presented a cosmetic problem for many orthodontic patients with scars on their teeth even 12 years after treatment. In severe cases, restorative treatment may be warranted (35). Van der Veen et al. found on debonding that the number of areas of demineralisation was significantly greater on posterior teeth than on anterior teeth. The canines and molars tend to be the most severely affected. The severity of the demineralisation was also significantly higher in the mandible than in the maxilla (39).

As orthodontics is elective treatment clinicians must adequately assess whether a patient has a satisfactory level of oral health suitable for treatment. Patients should exhibit sufficiently good levels of oral hygiene and have excellent dietary control. It is imperative that if demineralisation develops during treatment, it is detected as early as possible to prevent irreversible damage from occurring. Thus, methods for detecting early demineralisation and monitoring the lesions to ensure that they do not progress are of a significant benefit for both clinicians and indeed patients.

3.5 Methods of detecting plaque and demineralisation

3.5.1 Clinical Examination

Each patient assessed for orthodontic treatment undergoes plaque accumulation and demineralisation examination. Visual inspection is commonly undertaken as a first method of choice as it requires only clinical expertise of the clinician. No specific equipment is necessary and is a non-invasive method which makes it economical.

This direct visual assessment is an easy method of assessment of plaque, and the use of several plaque indices enables quantification of the amount observed. These indices use either selected teeth or the highest score for a group of teeth within a segment as the basis for their scores. When used for epidemiological studies or for evaluating the results of treatment in a study group the indices yield useful information. Some of the commonly used indices include the O'Leary index Modified Ramfjord index (Shick and Ash), the Quigley and Hein plaque index and the Silness and Loe index (40). The indices can be regarded as non-linear and therefore should be treated as scores assigned on an integer scale (41).

O’Leary Plaque Control Record

The Plaque Control Record is a simple method of recording the presence of plaque on individual tooth surfaces with the use of a disclosing agent. The record is a simplified process with a dichotomous scoring system for each tooth surface (simply marked for the presence or absence of plaque). Each tooth divided into four sections: buccal, lingual, mesial and distal. After the teeth disclosed, the number of surfaces with plaque present is completed on a full mouth charting. The total sum of surfaces positive for plaque presence is calculated and divided by the total number of surfaces present and multiplied by 100 to provide a percentage plaque score for the full mouth. The form also allows the patient to visualise his progress in learning plaque control. The process seems to have a motivating effect on patients and with a goal to reduce plaque accumulations until they reduce to 10% or less of the available tooth surfaces. The advantages of the index were the relatively simple criteria, which increased the repeatability; however, the lack of grading for the quantity of the deposits rendered the index relatively insensitive at differentiating between newly formed, thin plaque and older, thicker plaque (42).

Silness and Loe index.

The Plaque Index (PlI) is fundamentally based on the same principle as the Gingival Index, namely the desirability of distinguishing clearly between the severity and the location of the soft debris aggregates. Each of the four gingival areas of the tooth is given a score from 0-3; this is the PlI for the area. The scores from the four areas of the tooth may be added and divided by four to give the PlI for the tooth. The scores for individual teeth (incisors, premolars and molars) may be grouped to designate the PlI for the groups of teeth. Finally, by adding the indices for the teeth and dividing by the number of teeth examined, the Pll for the individual is obtained. The plaque index of Silness & Loe incorporated quantitative gradings of plaque thickness but was relatively more time consuming to perform (43). The surfaces of the teeth had first to be examined before disclosing for relatively thick plaque visible to the naked eye, and then again after disclosing to detect thinner deposits. An alternative technique described for the plaque index is to assess the tooth surface for plaque in situ; the surface was then wiped with a probe and the probe examined to enable thinner deposits not previously seen to be detected; both the disclosing and the probing methods required each surface examined in 2 separate phases. The Silness–Loe Index considers only the thickness of gingival plaque with no consideration of the coronal extension of the biofilm. The index has been criticised because, due to the examination technique, typically only one examiner can perform the assessment. If this index is to be used, it is recommended that a single, trained examiner be used throughout the trial (4).

The plaque index is described as below:

0 = Gingival area of tooth surface free of plaque tested by running probe across tooth.

1 = No plaque initially observed and only visualised after probe run across tooth.

2 = Gingival area is covered with a thin to a moderately thick layer of plaque visible to the naked eye.

3 = Heavy accumulation of soft matter, the thickness of which fills out the niche produced by the gingival margin and the tooth surface.

Modified Ramfjord index

Ramfjord developed one of the precursors to the contemporary plaque indices in 1956(44) as part of a larger periodontal disease index and focuses specifically on the gingival half of the interproximal tooth surfaces. This is because plaque here is more relevant to the development of periodontal diseases than coronal plaque. It was subsequently modified by Schick and Ash and has been employed in many clinical trials. The modification of the method involves the examination of facial and lingual surfaces of six selected teeth with the scoring of plaque restricted to the gingival half of the interproximal surfaces(4).

0 = Absence of dental plaque

1 = Plaque present on some, but not all, of the interproximal and gingival surfaces of the tooth.

2 = Plaque present on all interproximal and gingival surfaces but covering less than one-half of the entire clinical crown.

3 = Plaque extending over all interproximal and gingival surfaces, covering more than one-half of the entire clinical crown.

The total score is divided by the number of teeth examined to determine a mean score per tooth.

Oral Hygiene Index

Greene and Vermillion incorporated an index for calculus deposits as well as soft plaque deposits.

The plaque or ‘debris index’ is described below:

0 = No debris present.

1 = Soft plaque debris covering not more than one-third of the tooth surfaces being examined.

2 = Soft plaque debris is covering more than one-third but less than two-thirds of the exposed tooth surfaces.

3 = Soft plaque debris is covering more than two-thirds of the exposed tooth surfaces.

The total debris score for all teeth is divided by the number of surfaces scored to give an oral cleanliness score. The oral hygiene is termed 'good' if the score is 0.3-0.6, 'fair' when 0.7-1.8 and 'poor' if 1.9-3.

Quigley and Hein plaque index

Quigley & Hein modified the oral hygiene index in 1962 (45) to develop an index for the evaluation of different oral hygiene measures, and the extended score allows greater sensitivity in assessing therapeutic efficacy(46). The plaque index is described as below.

0 = No plaque

1 = Separate flecks of plaque at the cervical margin of the tooth.

2 = A thin continuous band of plaque (<1mm) at the cervical margin.

3 = A band of plaque wider than 1mm but covering less than 1/3 of the crown.

4 = Plaque is covering 1/3 – 2/3 of the crown.

5 = Plaque is covering 2/3 or more of the crown.

It is evident that any subjective evaluation could be expected to have a degree of variability both between and within the examiners as no strict measurements are employed (47). It could also be expected that the degree of this variability would be reduced if the criteria used in such evaluations were clear and unambiguous (47). The plaque index developed by Quigley and Hein and modified by Turesky (48) is one of the most frequently used indices in product-testing. The technique employs disclosed plaque that is counted on the facial and lingual surfaces and emphasises the difference in plaque accumulation in the gingival third. One difficulty with this index, and the reason it is used less frequently than the Turesky score is that it tends to over score the incisal half of crowns at the expense of the gingival margin.

The levels of plaque which are compatible with a healthy periodontium have yet to be defined . Some operators have suggested empirical levels; Greene & Vermillion and Grace & Smales considered that an oral hygiene index of 0.6 and 0.5, respectively, were appropriate objectives. Barrickman & Penhall and Garnick advocated the use of the plaque index as a clinical record and

advised that levels of 0.6 to 0.8 should be achieved by the patient (42). O'Leary et al. and Grant et al. suggested that values of 10% and 15%, respectively, should be attained using the plaque control record. Orthodontic treatment is conventionally provided if the O'Leary plaque score is less than 15% or less of the available tooth surfaces (40) (49).

Despite the common usage of these and other plaque indices, concerns over ambiguity in the interpretation of scores by different examiners have been expressed (47).

Silness and Loe and Turesky indices use has frequently been suggested (4) (41). The choice of an index system to be used in plaque trials should be made regarding the objective of the trial, the size of the population, the period of the study, and the type and extent of change anticipated. They have been demonstrated to be acceptable indices for the estimation of cleansing ability (41). Quigley Hein index emphasises differences in plaque accumulation in the gingival third of the tooth, the most important region in relation to periodontal infections (50).

Strong correlations are noted between two ordinal indices for assessing the plaque scoring, i.e., Turesky modification of QHI (TQHI) and Rustogi modified Navy Plaque Index (51). Marks et al. evaluated reliability and reproducibility of five of these indices and found an intra class correlation coefficient of 0.70 for TQHI (46). Traditional plaque indices are problematic due to their integral nature and their failure to detect small, but potentially clinically relevant changes in plaque area. Traditional plaque indices (QHP index and MNP index) are suggested to be not the most appropriate indices for orthodontic purposes as they reflect plaque at the gingival margins instead of focus on the surface along the gingival margin and areas around the bracket (52). Other difficulties lie with the subjective nature of the indices and the need for examiner training for calibration, and this can be time-consuming and expensive, often increasing the cost of clinical trials, as does the need for a clinician to conduct the examination. The ability to ensure that, within any one trial, all examiners are calibrated does not necessarily confer reliability to trials conducted within other centres or at differing times within the same centre. These methods lack precision, objectivity, sensitivity, specificity and reliability that the highest level of clinical trial design requires (4).

Disclosing agents

Staining of bacterial plaque is commonly used as a visual aid for patients in developing an efficient system of plaque removal and also in explaining and teaching the significance of plaque in dental disease (53). A two tone disclosing agent was designed to distinguish mature and immature plaque due to the result of diffusion phenomenon of active ingredient (53)(54). Plaque assessment and OHI using disclosing agents are frequently employed in dental settings. However, plaque disclosures have

some limitations of which they cannot selectively disclose only plaque, but dye soft debris and pellicle as well (55). A study looked at Plak-Lite system that consists of a fluorescent disclosing agent and a light source to make the agent visible (55). The study demonstrated that the Plak-Lite system revealed bacterial plaque on the teeth, tongue and gingiva and plaque free teeth or acquired pellicle did not fluoresce. Also, it needs time to remove the plaque at the chair side. Another method of measurement for the old plaque is using Silness & Löe plaque index and Quigley and Hein index with disclosing agents. These indices were developed for grading of plaque thickness. However, it is also relatively time consuming, and the result may be influenced by the examiner's subjective decision.

Planimetric measurements involve the use of photographic images to determine the Plaque Percentage Index (PPI) which relates to the percentage area of the tooth covered by plaque. The method requires disclosing teeth and then recording photographic images which can be scored either by hand tracing or by computer analysis using a count of pixels (4). The method allows the quantification of plaque on an interval scale as opposed to the discrete range of traditional indices. In this way, it may make it more sensitive for measuring small changes in plaque levels.

While studies have found planimetric methods to deliver excellent intra-examiner and inter-examiner reliability with the computer based methods proving particularly more precise, objective and sensitive than traditional indices, however it is time-consuming and can be expensive (56). As with any photographic analysis consistency with magnification, angulation and lighting can be difficult to achieve. Reproducibility is essential, and use of positioning jigs may restrict the analysis to buccal surfaces. It only measures the plaque area and not the depth.

Fluorescein is a UV fluorescent dye and is used for research purposes to penetrate and disclose plaque that is a significantly different colour (yellow green) to the surrounding oral hard and soft tissues which appear dark. In this way, images collected of the teeth can be analysed using digital image techniques and the amount of plaque accurately quantified. Fluorescein helps to increase the sensitivity of plaque detection, and thus reduce the number of participants and time required for clinical product trials(4). The system is, however, expensive and the plaque quantification is limited to the facial surfaces of the anterior teeth. The careful repositioning of subjects within the photographic 'jig' is essential for reliability and reproducibility is a disadvantage (57).

Assessment of demineralisation by clinical examination include the International Caries Detection and Assessment System (ICDAS) scale, which differentiates between cavitated and non-cavitated lesions (58). Additionally, the index of Gorelick et al. is commonly used for the labial surfaces, which allows classification of the severity of demineralisation into the following categories(13):

1. No white spot formation

2. Slight white spot formation

3. Severe white spot formation

4. White spot formation with cavitation

However, the disadvantage is that it is less sensitive and subclinical lesions will not be evident. Thus clinical examination alone may not be the most suitable for monitoring small changes in demineralisation. Furthermore, as demineralisation is only seen clinically when at the white spot lesion stage, the level of mineral loss that has occurred by that time point can be advanced. Ekstrand, Fyffe, Nyvad and Pitts and Fyffe are other visual criteria that are used in studies for clinical examination of non cavitated carious lesions (59).

Traditional diagnostic systems for detecting caries lesions, such as visual inspection and radiography, have limited accuracy and sensitivity when diagnosing occlusal caries at the pre-cavitated level (60). However, for both cost and practicality considerations, visual methods still remain the standard for clinical assessment in dental practice (59).

3.5.2 Clinical photography

Photographic images are routinely used in clinical practice, and these have the advantage of being readily available and relatively cheap to produce (61). Using black and white and colour photographic images from which to quantify the enamel changes by using various indices has been attempted in several studies, and colour photographs have been found to be valid and reproducible (62)(63)(64). Photographic techniques have been used in orthodontic patients to study enamel changes (13)(20)(65), however it has proved difficult to develop reliable, quantitative in vivo measurements of the extent of demineralisation (61).

Photographs are an easy and efficient method of recording the progress of treatment to obtain a permanent record (65). Many optical methods have been used to study the changes that occur during early demineralisation of the dental enamel (61). Studies have shown more false positives due to recording more details with photographs than with direct assessment (64) whilst other studies have shown it is more reproducible when controlled lighting and camera position is used (63)(65). Image storage, anonymising data, assessment of reliability, blinding of assessors help in reduces bias in producing high-quality research using photographs. However, the limitations with

photographs include the difficulties of standardising the angulation, magnification and lighting.it can be difficult for photographs to be (65).

Benson et al. assessed validity and reproducibility of enamel demineralisation with direct visual assessment and photographic assessment using a method of morphometry with 121 dot array. The photographic technique used was found to be a reproducible method of measuring artificial enamel demineralisation and more reproducible than direct assessment. However, subjectiveness of the index lead to most variation and the need for more objective means of assessing enamel demineralisation was discussed (65). Benson et al.assessed the reproducibility of measuring artificial enamel white spot lesions from captured photographic images using computerised image analysis. The photographs were converted into TIFF images and mean grey scale levels of the areas of etched enamel were measured using computerised image analysis. This study found good reproducibility, with no evidence of systematic error and a low random error, between repeat readings from the same slide taken below the occlusal plane. The reproducibility of readings from the slides taken above the occlusal plane was not as good, with evidence of some systematic error and a larger random error. The error was probably due to differences in light scatter from the camera flash (61). Benson et al. found that computerised analysis of digitally converted photographic slides was a reliable and valid method of analysing and quantifying levels of demineralisation. Benson et al. Investigated computerised image analysis from digitally converted photographic slides and quantitative light-induced fluorescence (QLF) for recording and quantifying demineralisation surrounding orthodontic brackets. The mean grey levels detected on human molars exposed to demineralising gel showed good repeatability with only small differences noted between the repeated readings and good validity. The study showed an identification of demineralisation was correct in 78 per cent of cases from photographs and 92 per cent of QLF images. A negative test result correctly predicted the absence of lesions in 94 per cent of cases when using the photographic technique, and 92 per cent of cases using QLF images (33). The limitations of photographic technique are that it produced a figure for the grey level, which in itself is relatively meaningless, as the absolute figure will be dependent upon lighting conditions, changes in processing, and even film type. Arbitarily, QLF estimates mineral loss by extrapolating the change in fluorescence of the lesion compared with the fluorescence of the surrounding sound enamel (34).

3.5.3 Quantitative light-induced fluorescence (QLF)

Quantitative light-induced fluorescence (QLF) has been described by a number of authors and is based on the auto fluorescing property of dental enamel under certain conditions. It is based on the fact that various (organic) substances in the mouth absorb light of a certain wavelength (colour) and

then re-emit the absorbed energy at a different wavelength. By filtering away the illuminating light the fluorescence or QLF™ the image is obtained (66). The optical phenomenon of tooth autofluorescing on illumination with ultraviolet light was first observed by Hans Stubel (67). Benedict was first to describe that fluorescence can be used for caries detection because of the difference in fluorescence observed between sound and demineralised enamel (68), which is greater when the enamel is illuminated by light in the blue-green range (488 nanometers)(69)(70)(71).

Laser light induced fluorescence as a diagnostic method for detection of enamel caries at an early stage was introduced in 1982(70). The tooth was illuminated with a broad beam of blue-green light of 488 nm wavelength from an argon ion laser source, demineralised areas appear dark. A quantitative version of the laser fluorescence method was compared with longitudinal microradiography (LMR) for assessment of mineral changes in enamel slices using an in vitro caries model (72).These fluorescent methods involved the use of ultraviolet or laser light, which are potentially dangerous forms of radiation, particularly to the eyes (34). The laser fluorescence method (LAF) was validated with longitudinal microradiography (LMR) for assessment of mineral loss in incipient caries lesions in human enamel. A high correlation (r=0.73) was noted between the amount of fluorescence loss and mineral loss (73).

Further changes by the development of a portable system were the use of a regular light source and filter system to replace the laser source. A colour CCD micro-video camera (Panasonic WV- KS 152) was used. The enamel surface was illuminated with white fluorescent image light emerging from an arc lamp on Xenon technology. A blue filter (peak intensity of X = 370 nm with a full- width half-measure of 80 nm) was placed in front of the lamp to produce blue light. To enable enamel autofluorescence to be detected, a yellow high-pass filter, with peak intensity > 520 nm was placed in front of the camera to exclude light with wavelengths less than 520 nm. The images were recorded with the video cameras in fluorescence set-up were stored, processed, and analysed by custom-made software (QLF 1.92, Inspektor Research Systems BV, Amsterdam, The Netherlands)(34) (74). A significant correlation was found between fluorescence changes with portable lamp and mineral loss with TMR of r = 0.84. The QLF system was validated and found to be a valid and reliable method for assessment of enamel during lesion formation and remineralisation in vitro (74).

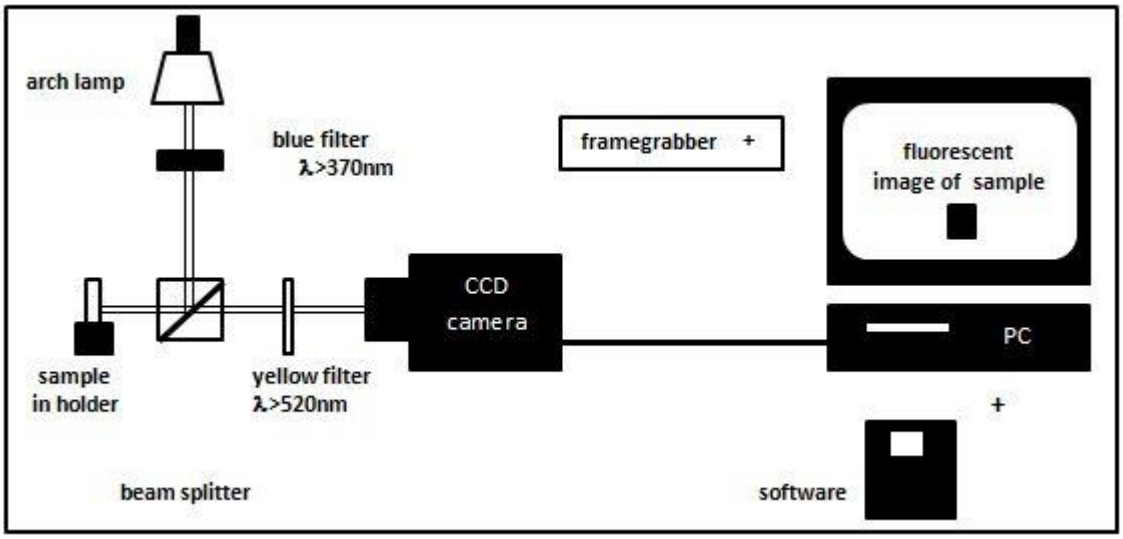

Figure 4.2: The portable QLF diagnostic system, reproduced from Al-Khateeb (74)

Areas, where porphyrins, generated by (anaerobic) bacterial activity have accumulated, show up brightly red/orange (Red Fluorescence or RF areas)(75)(76)(77). These effects can be observed visually, documented digitally and quantified. Red autofluorescence presumably originates from bacterial products (chromophores of porphyrins)(75). Red autofluorescence has been seen from *Actinomyces odontolyticus, Prevotella intermedia and Porphyromonas gingivalis and Peptostreptococcus micros* grown together (11). The invivo study has demonstrated QLF to be a reliable tool for assessing plaque accumulation present (4). It can be used longitudinally to assess levels present and hence monitor the success of any interventions aimed at reducing plaque accumulation.

Plaque accumulation on the teeth can be graded as the percentage tooth coverage using QLD software, which is based on the levels of red fluorescence evident at different cut off points. The value of ΔR30, which is a value related to how many pixels are covered with red fluorescence, must be greater than 30% for it be assessed as plaque.

QLF has been shown to be a valuable instrument for monitoring, detection and quantification of smooth surface caries (78). In these images, demineralised areas (e.g. white spots) show up as darkspots, where loss of fluorescence correlates with the mineral loss (72). The mechanism for this is that there is a greater degree of light scattering in demineralised enamel. The minerals have been replaced by water resulting in a decrease in light transmission and light absorption (79).

Demineralisation is assessed by comparing the fluorescence loss in the lesion to the fluorescence of the surrounding sound enamel to provide a quantitative assessment. False positives develop if the

outline of the analysis is not adjusted to be on sound enamel. Any areas with relative fluorescence loss greater than the 5% threshold are deemed part of the lesion (80).

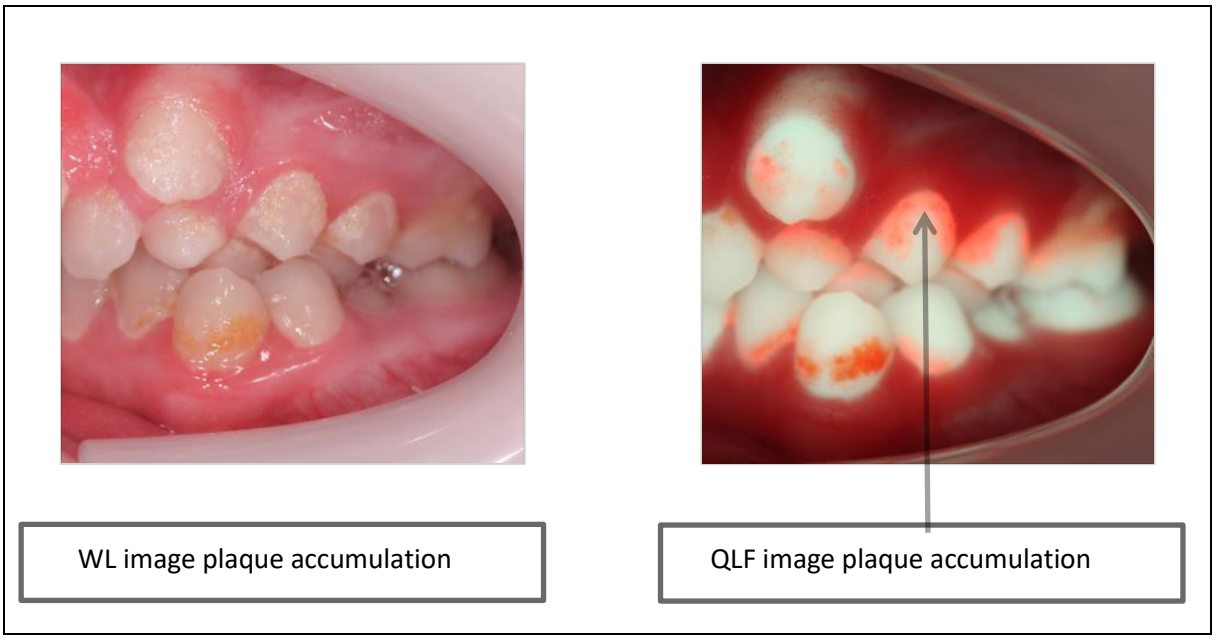

Figure 4.3: WL and QLF images demonstrating plaque accumulation

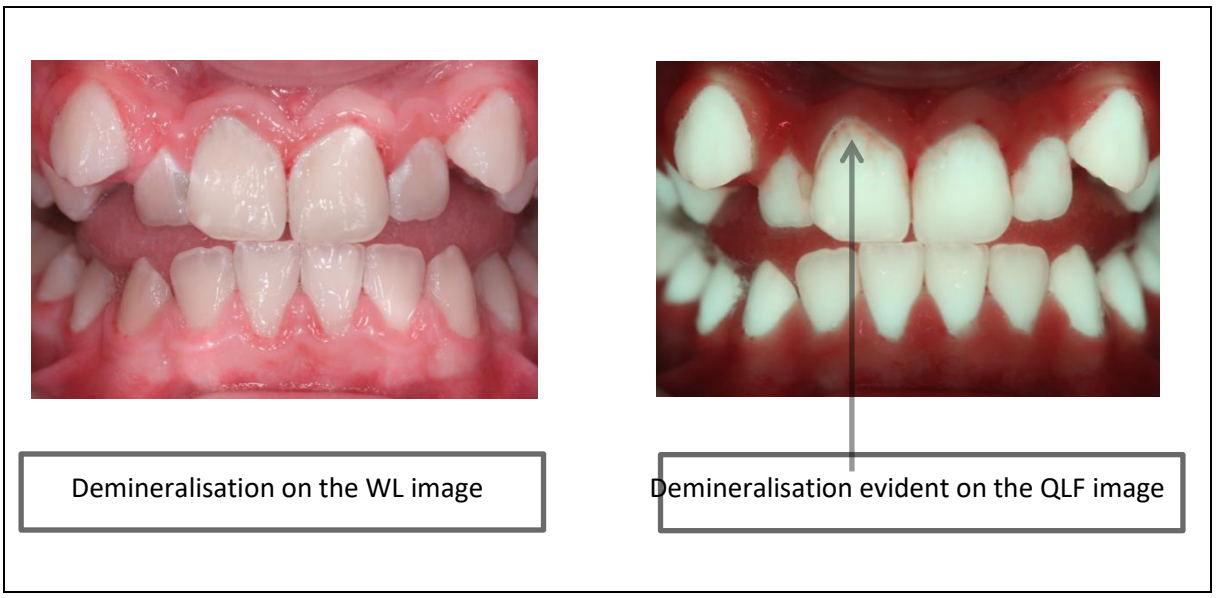

Figure 4.4: WL and QLF images demonstrating demineralisation

Besides the plaque, carious dental tissue also shows red autofluorescence(75) (81). A retrospective QLF study by Gomez et al. has shown a significant association between red fluorescence and progression to cavitation at thresholds ΔR0, ΔR10, ΔR20, ΔR60, ΔR70, ΔR80, ΔR90 and ΔRmax at baseline and for ΔR0 and ΔR10 at the final observation (82). Red fluorescence emission seen on QLF images is proposed to be the result of excitation of endogenous porphyrins by the violet-blue light at a wavelength range of 380–500 nm. Fluorescing porphyrins in caries detected to some extent are

protoporphyrin IX, coproporphyrin and uroporphyrin (83). It has been suggested that plaque associated with caries or gingivitis also shows red fluorescence (84). This red fluorescence corresponds with mature plaque (older than three to five days), calculus, or gingivitis, independent of gross unremoved food particles. The red fluorescence can be removed by professional cleaning in most instances. In some cases, no amount of non-destructive cleaning can remove the red fluorescence, and the activity may be contained within the tooth enamel and dentin. It also suggests that red fluorescence detected in a demineralised area or a fissure or a clean, sound surface signifies an active lesion (85).

QLF has been found to be a reproducible (34) and valid method (33) for assessing enamel demineralisation on a smooth surface. *In vitro*(33)(34)(80) and *in vivo* studies (39)(79)(86) have demonstrated that it is appropriate for identifying demineralisation and longitudinal monitoring of mineral changes for smooth surface lesions. A longitudinal study monitoring 406 carious lesions in 58 subjects for six months post debond showed an average ΔF at debond was 10.3% (39).

The reported values for sensitivity and specificity for occlusal caries are 90.9% and 90.6%, respectively (87). A study looked at 5 different methods for detecting occlusal caries found no significant improvement in occlusal caries detection when compared to visual examination. The study also found that QLF had lower specificity, which could lead to more false-positive diagnosis and consequently over- treatments (60). Low specificity values for QLF have been reported previously (88)(89). Pretty et al. demonstrated that QLF image analysis was reliable and reproducible, based on the high intra- and inter-examiner agreements found in an *in vitro* study. The study showed using the QLF that an examiner novice to the technique demonstrated differences in intra-examiner reliability compared with other examiners with more training. They recommend that novices to the technique should be trained before they analyse experimental data. Pretty et al. reported that due to the subjectivity of the image analysis process, there is a risk of operator bias associated with the technique (87).

An advantage of QLF is that it provides a quantitative score of plaque accumulation and demineralisation. Quantification of lesion severity by determining fluorescence loss and lesion area is a great benefit compared to the qualitative and subjective data obtained by conventional visual examination (15). Quantification allows for monitoring of lesion progression or regression over time. Additionally, QLF demonstrates demineralisation before it is evident on white light digital images. Boersma et al. study of 64 posts debond patients found more demineralisation lesions with QLF than with visual examination and all lesions detected by visual assessment had a QLF- determined fluorescence loss in part of the lesion of >15%, and thus using QLF lesions can be detected earlier

(15). *In vivo* studies have also found that QLF demonstrated a greater amount of demineralisation and noted it an earlier stage than conventional photographic analysis(15)(90). An *in vitro* study of bonded orthodontic cleats on 13 human premolars placed in a demineralising solution. The level of fluorescence loss on QLF images, ΔQ, increased from 0.17 at baseline to 5.2, 29.7 and 68.2 at 24hours, 144 hours and 288 hours respectively. However, visual evidence of demineralisation was only evident on 5 of the 13 teeth at 144 hours and 8 teeth at 288 hours, indicating the greater sensitivity of QLF image assessment (80). Detecting subclinical lesions is beneficial to clinicians in reinforcing oral hygiene control and if appropriate, the knowledge that it may be appropriate to initiate remineralisation therapies.

3.5.4 Toothcare™

Toothcare TM is a hand-held device based on the same principles as QLF but without the ability to capture images and can be used to assess plaque accumulation and demineralisation. It also emits a blue light from a 450 nm LED to illuminate the tooth surface. The device is used by the operator in conjunction with goggles which filter the yellow and red light reflected (with transmission peaks around 500nm and 630nm) allowing the operator to visualise areas of fluorescent plaque and demineralisation at the chair side (91).The advantage is that the device is compact and inexpensive with no controlled unit or monitor. It is easily transportable and easy to use. However, it does not provide a direct quantitative measurement of the plaque accumulation and enamel demineralisation present.

An invivo study by Thomas et al. found Toothcare™ demonstrated plaque deposits more readily than QLF in 29 patients during fixed orthodontic treatment. It may be due to the device being more compact and easier to use. However, on assessing demineralisation, Toothcare™ had poorer sensitivity scores and 43% of lesions detected by QLF was not detected with the ToothcareTM device (90).

3.5.5 Diagnodent™

Hibst and Paulus developed Diagnodent using infrared light to detect caries based on the difference in fluorescence between sound and demineralised enamel. The instrument contains a laser diode (655 nm, modulated, 1 mW peak power) as the excitation light source, and a photo diode combined with a long pass filter (transmission > 680 nm) as the detector. The digital display shows quantitatively the detected fluorescence intensity (in units related to a calibration standard)(92)(93).In vitro studies have shown that the system has higher sensitivity and specificity for smooth surfaces (92)and occlusal surfaces (94) with the high inter-examiner agreement (94)(95)

(96). A systematic review showed reliability for Diagnodent, and Kappa values ranged from 0.54 to 0.94. The sensitivity scores ranged from 0.16 to 0.96, and the specificity ranged from 0.25 to 1.00 (59). *In vitro* studies have demonstrated the effective use for demineralisation adjacent to orthodontic brackets (93). An in vivo study showed that the system could be an adjunct to a visual examination for the detection of early enamel lesions on smooth surfaces (96), another found the system to have low detection ability for early lesions (89). The system may overscore lesions compared with a visual examination and can be a disadvantage resulting in over treatment (93).

3.4.6 Transverse microradiography

Transverse microradiography, originating from Thewlis (1940), was developed as a quantitative method by Angmar et al (97). Its principle is the measurement of the absorption of mono- chromatic x-rays by a tooth section, compared with absorption by a simultaneously exposed standard (97). The Transverse microradiography involves a tooth section with a thickness of the order of 0.1 mm, made by two cuts perpendicular to the surfaces. The section is then subjected to caries attack or de- and remineralisation treatments and radiographed with a calibrated wedge or step wedge, frequently made of aluminium. Transverse microradiography (TMR) is a valid and reliable technique for detecting demineralisation (4). It is considered to be the gold standard method for quantitative measurement of mineral content levels (85). Therefore, other techniques including QLF are often compared and validated against TMR results. Validation studies have compared QLF to total mineral loss or lesion depth. The choice of TMR as gold standard and use of correlation statistics when validating other techniques are criticised (85). This technique is employed in in vitro studies and therefore cannot be applicable when considering clinical trials. In 1999, ten Bosch reviewed the literature on calibration(validation) of QLF for smooth surface caries and determined the mean correlation coefficient as 0.75(±0.08) when compared to TMR/LMR for mineral loss measurements (98). When the relationship with lesion depth and QLF was considered the mean correlation was 0.73 (±0.10) (85). When considering the occlusal demineralisation the correlation coefficient of 0.82 (85) and were high.

3.5.7 Quantitative Light-induced Fluorescence-Digital (QLF-D)

Quantitative Light-induced Fluorescence-Digital (QLF-D BilluminatorTM, Inspektor Research Systems BV, Amsterdam, The Netherlands) is a novel dental diagnostic tool which is based on the autofluorescence of teeth. It is the updated version of the first product, the QLF device (InspektorTM Pro, Inspektor Research Systems BV, Amsterdam, The Netherlands), and it can get more clear plaque image in red using improved filter set (D007; Inspektor Research Systems BV, Amsterdam, The

Netherlands). The QLF-D Biluminator™ 2 consists of a Biluminator™ mounted on a Single Lens Reflex (SLR) camera fitted with a 60mm macro lens, based on QLF and Tooth care technology. The Biluminator™ provides the light sources and filters for making white-light which is a conventional digital photograph and QLF™-images and a connection to a computer that runs the necessary software for archiving and analysis. Archiving provides the ability to reduce and even eliminate observer recall bias and enables re randomisation of images, unlike the Toothcare device. The system enables documentation and monitoring over time of specific areas and supports information exchange. The photographs can be initiated either by hand or under computer control; the system takes two successive pictures: one standard white light image and one QLF image. The average duration of this procedure is less than 5 seconds. This method of taking two images almost simultaneously allows comparisons to be made as the magnification and angulation of images remain the same.

Figure 4.5 Image of QLF camera

The images are stored and analysed (semi-)automatically for demineralisation and red fluorescence or Two-Tone Plaque Score if applicable. Analysis for de- and remineralisation and red fluorescence is based on the same algorithms as used in the Inspektor™ Pro system and the correlation of the results is very good.

The WL images like conventional photographs require a direct visual assessment to assess the plaque accumulation and demineralisation present and can be conducted using qualitative or quantitative scoring criteria. The QLF images are analysed using the customised software to provide quantitative data. Plaque accumulation is graded as the percentage tooth covered by mature and immature plaque which is shown with two tone plaque score. Percentage of increase in the ratio of the red and the green component concerning that ratio of sound tissue is demonstrated as ΔR, at

different cut off points. This is related to the presence of porphyrins and indirectly related to the bacterial activity. The value ΔR30 is most commonly recorded, whereby the number of pixels covered with red fluorescence must be greater than 30%. Areas of demineralisation must be identified and assessed individually. Data is obtained regarding changes in the enamel fluorescence of the lesions, recorded as ΔF and ΔQ. All these parameters are computed by selecting, either automatically or manually, the area of interest in an image. Within this area, a reference is automatically generated or manually selected as seen below (Table 4.1).

Name	Symbol	Unit	Description
Delta F	ΔF	%	Percentage fluorescence loss with respect to the fluorescence of sound tooth tissue. Related to lesion depth.
Delta Q	ΔQ	% px^2	Percentage fluorescence loss with respect to the fluorescence of sound tissue times the area. Related to lesion volume.
Lesion Area	AΔF	px^2	The area with ΔF equal or smaller than a specific threshold value of ΔF (default -5%).
Delta R	ΔR	%	Percentage of increase in the ratio of the red and the green component with respect to that ratio of sound tissue. Related to the presence of porphyrins and indirectly related to the bacterial activity.
RF Area	AΔR	px^2	The area with ΔR equal or higher than a specific threshold value of ΔR.
Simple Plaque Score™	SPS™	-	A value from 0 (no mature plaque) to 5 (high amount of mature plaque).
Two-Tone Plaque Score™	TTPS™	%	Percentages of tooth area covered by mature (dark-blue) and immature plaque (pink-blue).

Table 4.1 lists the various parameters that can be obtained using QLF™.

An invivo study showed Δ*F* and Δ*Q* values may be useful in aiding clinical diagnosis and decision making in relation to the management of early mineral loss and restorative intervention of occlusal caries.(99)

4.0 AIMS AND OBJECTIVES

4.1 Aims

1. To assess the use of the QLF-D™ as an oral hygiene reinforcement tool to detect plaque accumulation before commencement of orthodontics.
2. To assess the use of the QLF-D™ as an oral hygiene evaluation tool to detect demineralisation before commencement of orthodontics.

4.2 Objectives

1. To quantify plaque accumulation on the surfaces of teeth using QLF-D™.
2. To quantify demineralisation on the surfaces of teeth present using QLF-D™.
3. To assess if OHR using the QLF-D™ device reduces plaque accumulation and the development of demineralisation before start of orthodontic treatment.
4. To assess the level of demineralisation that occurs and can be visualised on QLF images before being seen on WL images.
5. To evaluate the intra and inter-examiner reliability of QLF-D™ and WL image assessment.
6. To ascertain the patients' perspectives of QLF-D™ as oral hygiene evaluation tool.
7. To provide data on QLF-D™ as an oral hygiene evaluation tool to aid the design and methodology of future studies.

5.0 MATERIALS AND METHODS

5.1 Ethical approval

Ethical approval was gained from the North West Research Ethics Committee Liverpool Central (REC reference: 15/NW/0427 with an Integrated Research Application System (IRAS) project number 171796. The project was also registered with the Royal Liverpool and Broadgreen University Hospital Trust Research and Development Department.

5.2 Study design

Pilot Randomised clinical trial.

5.3 Sample

Sixty consecutive patients who met the inclusion and exclusion criteria attending Liverpool University Dental Hospital orthodontic department for new patient clinics, review clinics or pre-treatment oral hygiene visits with the hygienist were asked to participate.

5.3.1 Inclusion criteria

1. All participants consent.
2. All participants in good health.
3. At least 11 years of age.
4. Patients with poor level of oral hygiene undergoing pre-treatment visits with the hygienist.

5.3.2 Exclusion criteria

1. Patients with significant disabilities that may affect manual dexterity and oral hygiene practice.
2. Patients who have had antibiotics in the last two months.
3. Patients with full coronal coverage restorations.
4. Patients with visually cavitated lesions.

5.4 Setting

Liverpool University Dental Hospital. Patients with poor oral hygiene who were currently referred to hygienist before they were added on the waiting list to have fixed orthodontic appliance treatment were recruited.

5.5 Methods

5.5.1 Recruitment

Consecutive recruitment of 60 patients with high plaque score of 3 or more measured using modified Quigley Hein Index attending for new patient or review clinic orthodontic appointments were recruited. These were participants who required oral hygiene reinforcement by a hygienist or a dentist and were willing to take part in the trial. Patients were identified by consultants in the orthodontic department, and written patient information sheets were provided to the patients outlining involvement in the study (Appendix 14.1, Appendix 14.2). For individuals under 16 years of age, information sheets were additionally provided to a parent or guardian (Appendix 14.3). They were then contacted by the main examiner (PR). Patients who were interested in taking part in the trial were given appointments and consent was taken on the day of recruitment. Informed written consent was then obtained by PR from the patient (Appendix 14.4) or parent (Appendix 14.5). Assent forms were completed if the patient was less than 16 years of age (Appendix 14.6). Following recruitment, each participant was given a unique study number so that the data could be pseudo-anonymised. The personal details of each subject were not used in conjunction with the research project to ensure anonymity.

5.5.2 Randomisation

After gaining consent patients were recruited into two groups.The randomisation was conducted by the supervising statistician who was not invloved with the recruitment process. A random number sequence was produced by a computer generated programme. Allocation concealment was with consecutively numbered, sealed opaque envelopes. At the same appointment, the next envelope was opened and was allocated into one of the two parallel groups. Blinding of the patient or operator to the group allocation was not possible. All of the patients were treated by one operator (PR) and a baseline assessment (T1), was taken using the QLF-D^{TM} device (Inspektor Research Systems BV, Amsterdam, The Netherlands). QLF-D^{TM} device was used to photograph the maxillary and mandibular dentition when the patient was occluding edge to edge in frontal, buccal views. Occlusal views were taken with mouth wide open. O'Leary plaque record and Quigley Hein Plaque index were measured and the disclosing agent used. Prophylaxis was conducted to remove plaque deposits present. The photographs were then repeated to allow an assessment of any demineralisation present.

5.5.3 Data collection

The subjects were assessed at three consecutive hygiene appointments. These appointments were made approximately 3-4 week intervals. A QLF image was taken to identify the baseline plaque score at the first appointment. O'Leary plaque record and Quigley Hein Plaque index were measured and the disclosing agent used. Prophylaxis was conducted to remove plaque deposits present. The photographs were then repeated to allow an assessment of any demineralisation present. The standard of oral hygiene was re-assessed at three consecutive appointments (T1-T3), at approximately 3-4 week intervals.

At each visit, the subjects were given OHR using the WL or QLF images as visual aids depending on their group allocation. These images were at the same magnification, focus and direction. The OHI was a standardised reinforcement of the instructions usually given by the hygienist, although focused on the areas of poorer plaque control or where demineralisation was present. Patients were advised to brush their teeth twice a day, after breakfast and at bedtime. Demonstration on use of floss and interdental brushes and verbal information on the use of disclosing agents given. Patients were advised to use interdental brushes twice a day were applicable as shown to them and disclosing agents to be used twice per week 3 days apart. These instructions were given verbally.

On completion of the study, the participants were given a debriefing questionnaire (Appendix 14.7) to complete, which focused on their perception of being shown the images and their opinion of whether seeing them was a useful tool to aid their oral hygiene control.

As is normal clinical practice, if a subject's oral hygiene is continually poor and severe, or progressing demineralisation is noted, were discharged from the orthodontic department to the general dental practitioner for oral hygiene reinforcement and they are referred back to new patient's clinic when the oral hygiene is of an adequate standard.

5.5.4 Image analysis

The images taken were stored anonymously on a database based on the participant's study number. The images were recoded on a computerised programme by the statistician following data collection to avoid recall bias and observer bias. These images were then analysed by the main examiner PR 3-6 months after data collection.

The WL images were assessed for the number of areas of demineralisation present in addition to the number of teeth. The QLF images were evaluated using the customised computer software. A measurement of the plaque accumulation on each image as the percentage tooth coverage demonstrating red fluorescence at ΔR30 was graded as ΔR30.

For areas of demineralisation, an outline was drawn around each lesion with borders on sound enamel. The mean fluorescence loss (ΔF), maximum fluorescence loss (ΔF Max) was assessed in comparison to the fluorescence of the surrounding enamel. ΔQ was also ascertained, which enabled an assessment of the amount of fluorescence loss ΔF and the lesion area involved per pixel. If there was no sound enamel adjacent to the lesion on one side, such as when the lesion was adjacent to the gingival margin, the outline was adjusted for this. The maximum fluorescence loss (ΔF Max) was used for each tooth when more than 1 lesion was present.

5.5.5 Reliability assessments

The WL and QLF images from 7 patients were analysed by the main examiner and three additional examiners (Appendix 14.8) to assess inter-examiner reliability. Altogether 35 images were analysed for plaque coverage (ΔR30) and 35 images of 7 participants with varying size and severity of demineralisation to assess fluorescence loss (ΔF). Method of choosing 7 patient images allowed having 7 images of each view for the reliability analysis. These examiners all had previous experience with the software and analysing experimental data. The main examiner examined the images on a second occasion one month later to allow assessment of the intra-examiner reliability.

5.5.6 Sensitivity and specificity assessments

The sensitivity of a diagnostic test is its ability to correctly diagnose the presence of an outcome when the outcome is present or the true positive of an outcome, thereby whether the presence of demineralisation can be correctly diagnosed on the images. The specificity of a diagnostic test is the ability of a test to correctly confirm a negative outcome or true negative of an outcome, such as the absence of demineralisation.

Thirty-six WL images and their corresponding QLF images were assessed for the presence of demineralisation by three examiners to determine the ability of the examiners to identify the presence and absence of demineralisation correctly. The QLF and WL images were displayed in a

random order to ensure each image was assessed independently and to avoid recall bias. The results were compared to an additional main assessor's analysis, which was taken to be the gold standard.

6.0 STATISTICAL ANALYSIS

The statistical analysis was undertaken using IBM SPSS Statistics for Windows, version 24.0 (IBM Corp., Armonk, NY, USA) and SAS software version 9.3 (SAS Institute Inc., Cary, NC, USA).

6.1 SAMPLE SIZE CALCULATION

There were no previous studies using QLF-D^{TM} on a population with poor oral hygiene to base a sample size calculation and therefore the study was conducted as a pilot study.Thus a formal sample size calculation was not performed. The expert statistical advice was sought, and a sample size of 60 was deemed appropriate to allow estimation of parameters for a sample size calculation to be conducted for future definitive studies. Browne advocates assessing at least 30 participants when estimating the effect of specific factor during a pilot study (100)(101). Hence, this was deemed to be an appropriate number of patients to recruit.

6.2 NORMALITY TESTING AND HYPOTHESIS TESTING

The primary outcome variable was the percentage change in plaque accumulation from the first visit (T1) to the final visit of OHR (T3), measured from the QLF images as tooth coverage demonstrating red fluorescence at ΔR30, was analysed. Although the study was not powered to detect a difference between groups, a statistical comparison would be carried out to give initial estimates of effect size and variability for use in the design of future studies (101). The secondary outcome variable was the percentage change in demineralisation from the first visit (T1) to the final visit of OHR (T3), measured as ΔF from the QLF images. The measurement was taken at the tooth level, as separate areas of demineralisation.

As the outcome was measured at tooth level, but the randomisation was at a participant level, the analysis of the secondary outcome was controlled for the clustering of teeth within participants using multilevel modelling (102). This allowed estimation of the intra cluster correlation coefficient (ICC) for use in the design of future studies.

6.3 RECEIVER OPERATING CHARACTERISTIC CURVES

Receiving operator curves assess the relationship between the sensitivity of a test, which is the number of true positives and 1-specificity. As specificity is the correct diagnosis of true negatives, 1-specificity is the proportion of false negatives detected. A perfect test would have a sensitivity and specificity of 1. Graphically, when a diagnostic test is as likely to produce a true positive result as a false positive result, there would be a linear diagonal line from (0, 0) to (1, 1). The steeper the line,

the greater the sensitivity and specificity. Alongside this, the area under the curve (AUC) can be calculated to assess the performance of a diagnostic test with the best test having a value of 1. These factors were used to assess the level of demineralisation measured on QLF images, and that could be visualised on the WL images (103).

6.4 RELIABILITY DATA

For the QLF images, the plaque and demineralisation data collated was continuous, whereby a precise score was given using the QLF software. Thus intra and inter-examiner reliability were evaluated using intra-class correlation coefficient (ICC).For the WL images, the demineralisation data collated was categorical, whereby demineralisation was assessed as being present or absent. Thus intra and inter-examiner reliability were assessed using Cohen's kappa statistics.

6.5 SENSITIVITY AND SPECIFICITY OF DEMINERALISATION DATA

The sensitivity and specificity of demineralisation assessment on the QLF and WL images would be calculated by assessing the demineralisation data results obtained from the 3 examiners in comparison to the results of the gold standard, which was the main assessor's analysis of the QLF images. This would provide a measure of QLF and WL diagnostic accuracy of demineralisation.

7.0 RESULTS

7.1 Description of subjects

A total of 60 participants were recruited. The first patient enrolled in December 2015, and the last patient completed the study in October 2016. Baseline records were taken from the 60 participants at T1 at the start of intervention, and the images were analysed for the presence of plaque and demineralisation on the QLF images. All image assessments were taken after 6 months following the appointment. Both QLF and WL images were re randomised to avoid recall bias as the assessor and clinician providing the OHR was the same.

The baseline means ΔR30 was 2.3(SD 2.1) and 4.2 (SD 3.1) for the WL and QLF group participants respectively, which was not statistically significant (P=0.10, t-test). The baseline mean plaque percentage using the O'Leary index was 45.3 (SD 50.1) and 58.9 (SD 33.03) for the WL and QLF group participants respectively, which was statistically significant (P=0.03, t-test). The baseline mean plaque percentage using the Quigley Hein Index was and 1.37 (SD 0.58) and 1.68 (SD 0.59) for WL and QLF respectively, which was not statistically significant (P=0.81, t-test).

The 60 patients were randomly allocated to the WL or QLF groups at the first visit, T1. This resulted in 30 participants being allocated to the WL group and 30 to the QLF group. Figure 8.1 highlights the flow of patients through the trial.

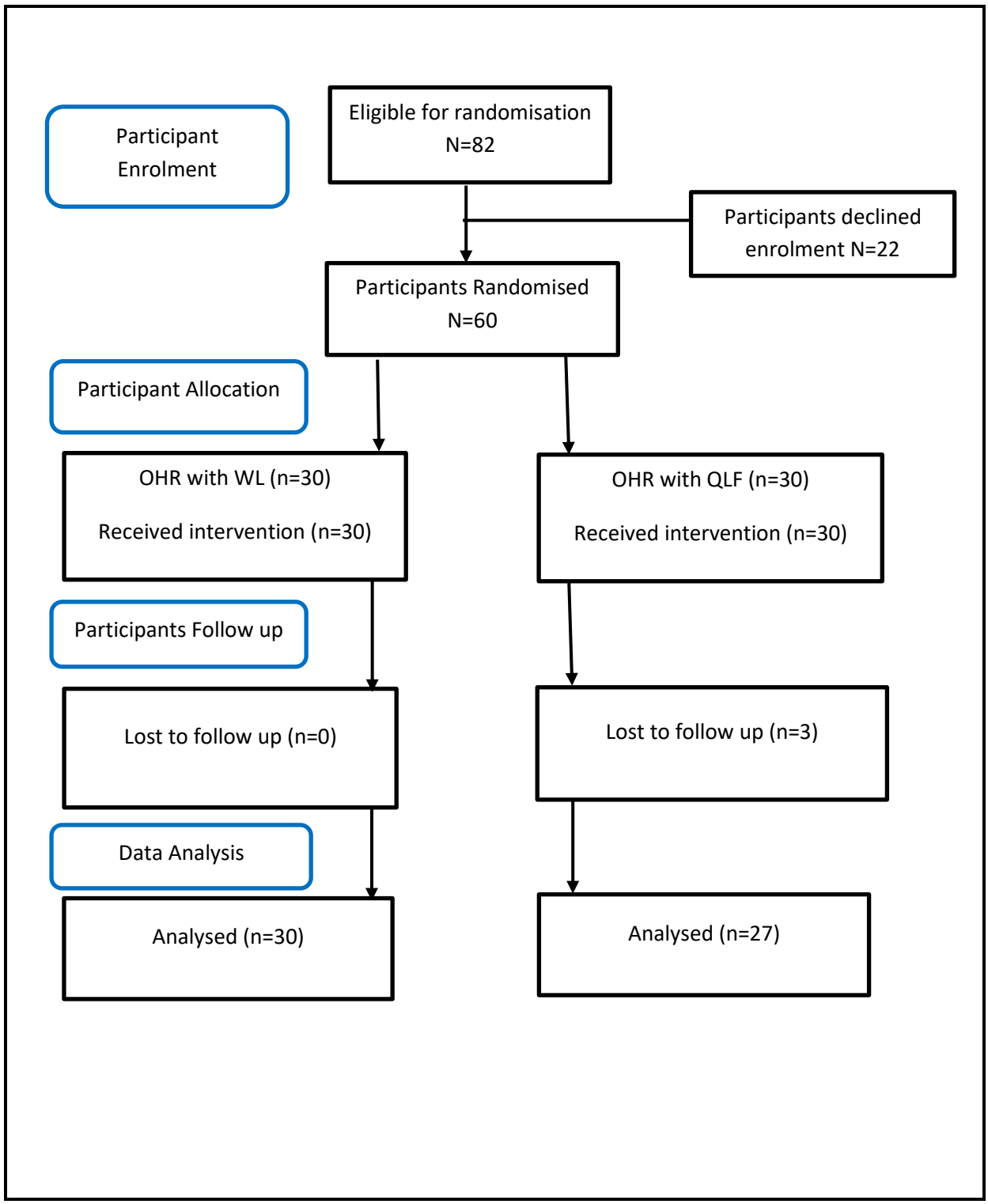

Figure 7.1: Flow diagram of the Randomised Clinical Trial

There were 3 drop-outs. One participant dropped out at the third visit as relocated to a different city and the second participant decided not to have orthodontic treatment and were both over 16yrs of age. These participants were given further 2 appointments which they failed to attend before they were considered drop outs. The third participant only attended the first visit and parents did not

bring the child for further two appointments despite given 4 different appointments. Rest of the patients completed the trial and had their data fully analysed. Intention to treat analysis was used as far as reasonably practicable; however, no replacements were made for missing data.

There were 32 females (53.3%) and 28 males (46.7%) recruited into the study (Table 7.1). In the WL group, there were 13 females (43.3%) and 17 males (56.7%). In the QLF group, there were 19 females (63.3%) and 11 males (36.7%).There were no significant differences between the groups at baseline for gender. (P=0.12, chi-square test).

The median age of the sample was 14.7 years (minimum 11.6yrs; maximum 26.7yrs). In the WL group, the median age was 14.5 years (SD 3.3), and QLF group was 15.0 years (SD 2.8). There was no significant difference between the groups for age (P=0.85, t-test).

The age of the participants showed evidence of non-normality when assessed using Kolmogorov Smirnov test and Shapiro Wilk tests. The median age of the sample was 13.8 years (IQR 4.4; minimum 11.1yrs; maximum 26.7yrs). In the WL group, the median age was 13.6 years (IQR 4.0; minimum 11.1yrs; maximum 26.7yrs), and QLF group was 14.7 years (IQR 4.2; minimum 11.1yrs; maximum 22.5yrs). There were no significant differences between the groups at baseline for age (P=0.79, Mann-Whitney U-test).

Baseline Characteristics	WL Group	QLF Group	All participants
Participants	30	30	60
Median Age in years (IQR)	13.6(4.0)	14.7(4.2)	13.8(4.4)
Gender -Female (Percentage)	13 (43.3%)	19 (63.3%)	32 (53.3%)
Gender -Male (Percentage)	17 (56.6%)	11 (36.6%)	28 (46.6%)

Table 7.1: Baseline characteristics of participants

The overall mean number of teeth assessed per participant was 23.1 (SD 1.3). In the WL group, the mean number of teeth assessed was 23.2 (SD 1.2) and in QLF group was 23.1 (SD 1.3). There was no statistical difference between the two groups (P=0.35, t-test).

7.2 PLAQUE DATA

The assessment of plaque accumulation was measured by ΔR30 on QLF images. The baseline means ΔR30 was 2.3 (SD 2.1) and 4.2 (SD 3.1) for the WL and QLF group participants respectively, which was not statistically significant (P=0.10, t-test).

Figure 7.2 demonstrates the mean ΔR30 scores for the participants at each of the visits in the two groups. As noted, there was a reduction in all the patients as the study progressed. The change in mean ΔR30 was greatest in the participants who were shown QLF images. These individuals had a reduction of 58% from T1 to T3. The participants who were shown the WL images had a 45% reduction (Table 7.2). This demonstrates a definite reduction in the ΔR30. However the standard deviation values were high. There was a significant reduction in plaque in both groups (P<0.001) with a mean of 51.8% and 95% CI of 40.36% to 63.26%. The confidence intervals indicate that the mean reductions noted were significantly lower in both groups at T3 than at T1 (P<0.05). Thus showing that the mean ΔR30 levels for the participants reduced over the course of the study as a result of the OHR being given from T1 to T3. This was the case for both groups, regardless of their group allocation. However, due to the nature of the study, these results were based on groups of low sample sizes, and thus the findings should be viewed with caution.

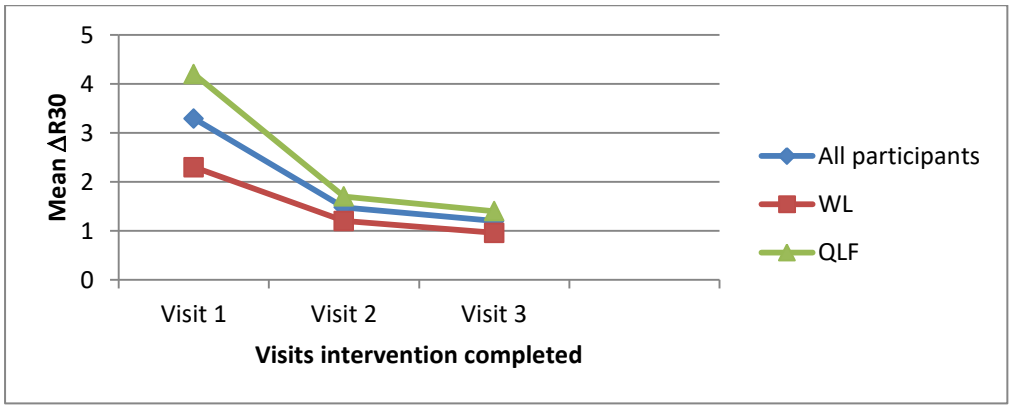

Figure 7.2: Mean ΔR30 level for the participants over the course of the study

	WL Group	QLF Group
Mean percentage change in ΔR 30	45.36%	58.93%
95% Confidence interval	-4.85% to 95.52%	25.92% to 91.96%
SE	9.1	6.3

Table 7.2: Adjusted mean percentage change in ΔR 30 from T1 to T3.

A repeated measure of analysis of variance was conducted for the overall mean ΔR30 for the participants at T1 to T3 using the first visit as a covariate. This assessment accounted for the high plaque score at the first visit and allowed the individuals to be analysed in the WL and QLF groups. The analysis found that there was a reduction in the mean ΔR30 values over the two visits as the study progressed, although this was not statistically significant (P=0.81). R squared of 0.29 shows that 29% of the variance in mean ΔR30 was explained by baseline mean ΔR30 and intervention group. The QLF participants appeared to have a greater reduction in mean ΔR30 from T1 to T3, yet again the difference between QLF and WL participants was not significant (P=0.148).

Figure 7.3 highlights an example of the QLF images of a participant at T1 and T3, clearly indicating the improvement in plaque control following 3 sessions of OHR.

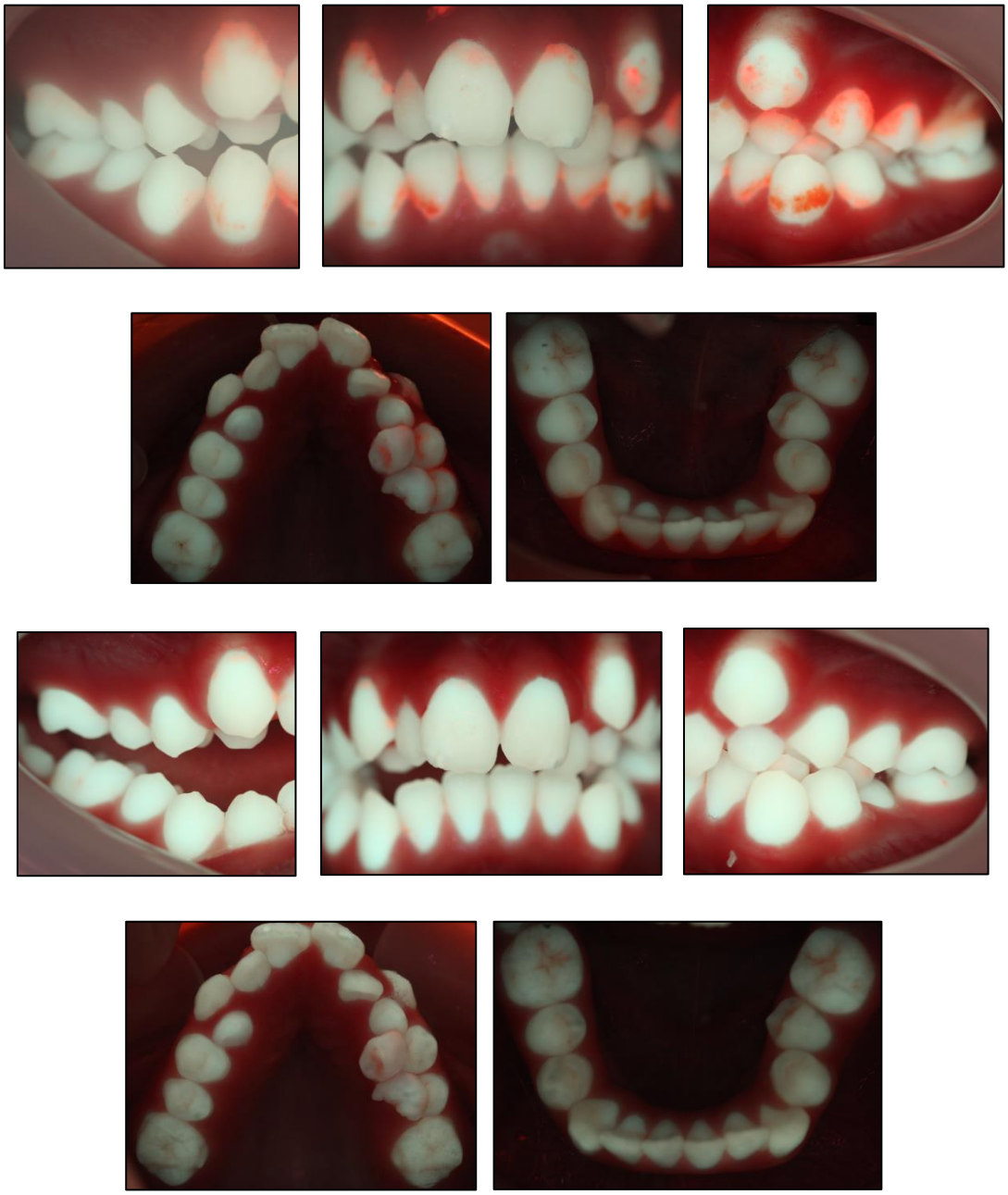

Figure 7.3: QLF images demonstrating the difference in plaque accumulation of a participant at T1 and T3. Upper 5 images taken at T1 and lower 5 images taken at T3.

7.3 DEMINERALISATION DATA

All the 60 participants had lesions at the start of the study. The total number of teeth with lesions present at the participant level at T1 was 800 and at T3 were 849. 649 lesions remained the same

from T1 to T3. 121 lesions were noted at T3, and 119 lesions at T1 were not present at T3. There was no change in lesion for 649. Comparing the total number of lesions at T1 and T3 showed a statistically significant change in the number of lesions per participant from T1 to T3 (P<0.001, paired t test). 121 lesions at T3 and 119 lesions at T1 were not analysed as the lesions showed red fluorescence from retained plaque and disclosing agent on occlusal lesions. However, following analysis, the teeth were re-assessed and analysed that found 29 subclinical lesions and there was demin present on 121 lesions at T3 and 119 lesions at T1.

At a tooth level, there were 649 teeth noted at T1 to have areas of demineralisation present on the QLF images of which 344 were in the QLF group and 305 in the WL group. The most commonly affected teeth were the mandibular premolars and maxillary premolars which accounted for 26.5 % and 25.7% of the lesions detected (Figure 7.4).

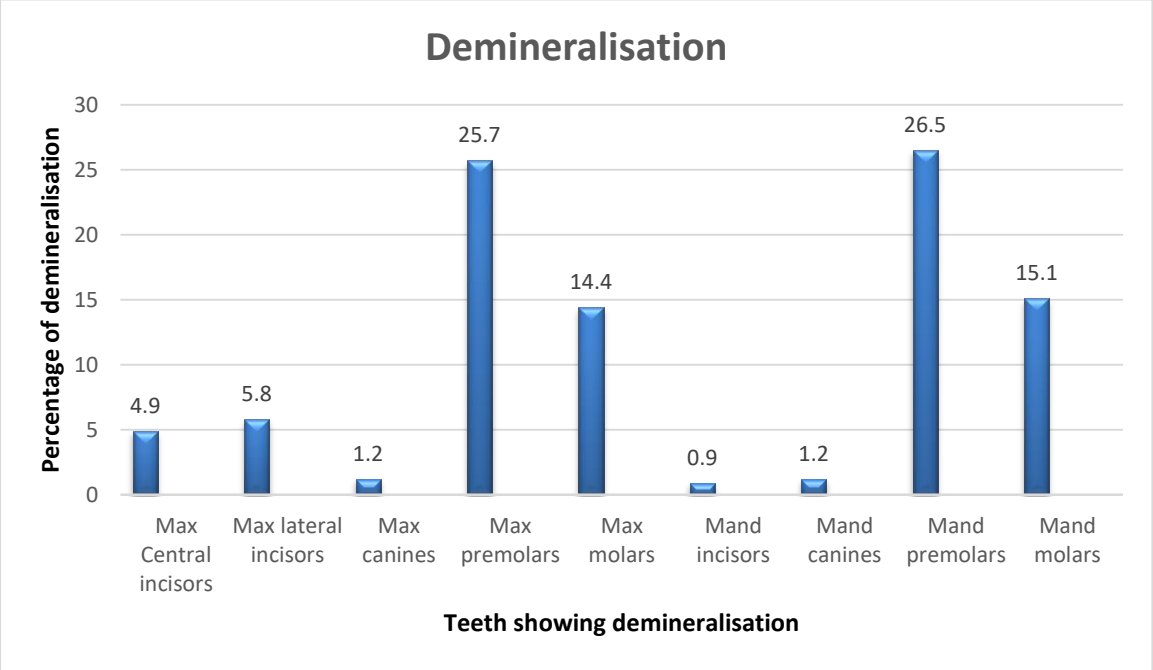

Figure 7.4: Percentage of teeth affected by demineralisation on all surfaces

Assessing the total number of lesions present at a patient level, all participants had demineralisation lesions present at T1 with a mean of 10 lesions in WL group (SD 3.6). The QLF group had a mean of 11 lesions (SD 3.1) at T1. At T3 the mean number of lesions in WL group was 9 (SD 3.7) and QLF group was 11 (SD 2.6) (Figure 7.5). There was a statistically significant change in the number of lesions from T1-T3 between WL and QLF group (P=0.03, ANOVA). The number of lesions at per participant level from T1-T3 was not statistically significant (P=0.21,

paired t test). 3 participants dropped out from QLF group and only 27 participants in QLF group were only included in the test.

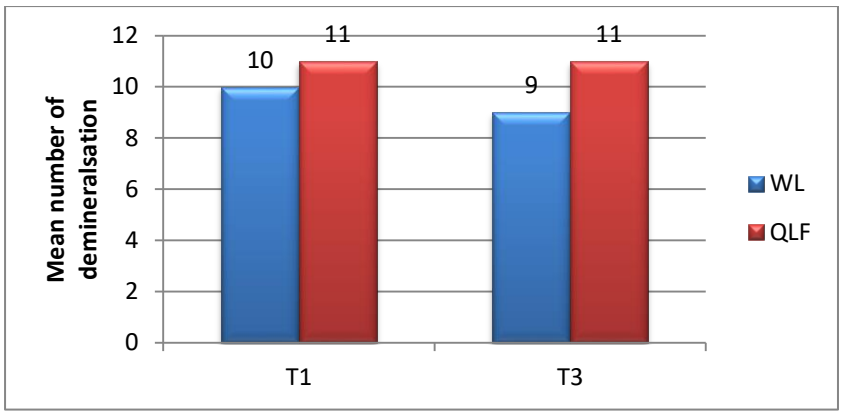

Fig 7.5: Per participant level mean demineralisation lesions observed at T1 and T3.

With regards to the outcome, the percentage change in ΔF at a tooth level, from T1-T3 in unadjusted means, not considering clustering within patients, the mean percentage change in ΔF from T1 to T3 was 0.04% (SD 0.22%). In the WL and QLF groups, the percentage change was 0.04% (SD 0.23%) and 0.04% (SD 0.21%) respectively. There was no difference between the two groups (P=0.69).

A repeated measure of analysis of variance was conducted for the mean ΔF for the participants at T1 to T3 using the first visit as a covariate. This assessment accounted for the mean ΔF at the first visit and allowed the individuals to be analysed in the WL and QLF groups. The analysis found that there was a reduction in the mean ΔF values over the third visit as the study completed, although this was not statistically significant (P=0.797). R squared of 0.43 shows that 43% of the variance in mean ΔF at T3 was explained by the mean ΔF at first visit. The QLF participants appeared to have a similar reduction in mean ΔF from T1 to T3, yet again the difference between WL and QLF participants was not significant (P=0.69).

The outcome measures in the study were measured at tooth level, but the randomisation was at participant level. Thus multilevel linear regression analysis was undertaken to control for the clustering of teeth within the participants. The adjusted mean ΔF was -14.0 (SE 0.50) and -14.19 (SE 0.53) in the WL and QLF groups respectively (Table 7.4). The 95% confidence intervals (CIs) demonstrate the narrow spread of the results and indicate no statistically significant differences in either the WL or QLF groups (P>0.05).

	WL Group	QLF Group
Mean ΔF	-14.0%	-14.19%
95% Confidence interval	-14.98% to -13.02%	-15.22% to -13.16%
SE	0.50	0.53

Table 7.4: Adjusted mean ΔF at T3.

Assessing the covariance parameters to determine the error variance of teeth being present within the same participant gave an intra cluster correlation coefficient (ICC) of 22.32%,. This indicated that 22.32% of the variance in the outcome was between patients and most the variation, 77.86%, was at a tooth level. This ICC estimation will be advantageous in planning the design and sample size required in future studies.

The maximum level of mineral loss noted within a lesion, ΔF Max, was additionally assessed to determine the extent of the lesion's severity. The overall unadjusted mean percentage change in ΔF Max from T1-T3 of 644 lesions was 0.29% (SD 0.61%). This was not statistically significant with 95% CI showing a narrow interval of 0.24% -0.34%. In the WL and QLF groups, the percentage change was 0.31% (SD 0.54%) and 0.27% (SD 0.67%) respectively. This was not statistically significant with P=0.29. Adjusting for clustering of teeth at a participant level (Table 7.5), the mean percentage change in ΔF Max was 0.04% (SE 0.04) and -0.10 % (SE 0.14) in the WL and QLF groups respectively. The CIs indicate the mean reductions noted were not significantly lower when comparing the two groups at T3 (P=0.48).

	WL Group	QLF Group
Mean percentage change in ΔF Max	0.04%	-0.10%
95% Confidence interval	-0.31% to 0.31%	-0.45% to 0.45%
SE	0.14	0.14

Table 7.5: Adjusted mean percentage change in ΔF Max from T1 to T3.

The difference in the adjusted means between the WL and QLF groups was 11.03 (SE 4.4) with a wide 95% CI of -20.0 to -2.03(P=0.017). Additionally, the test of the fixed effect of the intervention showed statistically significant difference between the WL and QLF groups (P=0.017).

ΔQ indicated the severity of the demineralisation with respect to the degree of mineral loss in conjunction with the lesion area involved, per pixel. The overall unadjusted mean percentage change in ΔQ from T1-T3 was -0.04% (SD 3.81%). In the WL and QLF groups, the percentage change was

0.04% (SD 1.81%) and -0.13% (SD 3.81%) respectively. Adjusting for clustering of teeth at a participant level (Table 7.6), the mean percentage change in ΔQ was -0.19 (SE 0.18) and -25.08 (SE 25.1) in the WL and QLF groups respectively. This suggests that there was a reduction in ΔQ in both groups. However the CIs indicate the wide variation within the groups from T1 to T3 and no statistically significant changes were noted (P>0.05).

	WL Group	QLF Group
Mean percentage change in ΔQ	-0.19%	-25.08%
95% Confidence interval	-0.54% to 0.16%	-20.67% to 20.52%
SE	0.18	25.1

Table 7.6: Adjusted mean percentage change in ΔQ from T1 to T3.

The difference in the adjusted ΔQ means between the WL and QLF groups was 40860.81 (SE 51848.88) with a wide 95% confidence interval of -63046.83 to 144768.30(P=0.43). Additionally, the test of the fixed effect of the intervention showed no statistically significant difference between the WL and QLF groups (P=0.96).

Participants were seen at 3-4 weekly intervals for the three appointments when OHR was provided. An assumption was made that, unlike ΔR30, changes in ΔF would occur linearly over time with the three visits of OHR. Thus, an assessment was required to ensure any variation in participants' duration in the study did not lead to differences in the potential development of demineralisation. The mean duration of the participants in the study from T1 to T3 was 61 days. There was no statistically significant difference between two groups with mean duration in WL group of 61 days (SD 20) and 60 days (SD 34) in QLF group.

Percentage change in ΔF at a tooth level of lesions on the 81 labial or buccal surfaces from the second premolar to the second premolar from T1-T3 in unadjusted means, not considering clustering within patients, the mean percentage change in ΔF from T1 to T3 was -0.74% (SD 2.98%). In the 50 lesions of QLF and 31 lesions of WL groups, the percentage change was 0.27% (SD 0.60%) and 0.47% (SD 0.69%) respectively. There was no difference between the two groups (P=0.16). This showed 81 lesions in 32 participants. 31 lesions were seen in 15 participants of WL group, and 50 lesions were seen in 17 participants in QLF.

A repeated measure of analysis of variance was conducted for the mean ΔF for the participants at T1 to T3 using the first visit as a covariate. This assessment accounted for the mean ΔF at the first visit and allowed the individuals to be analysed in the WL and QLF groups. The analysis found that there was a reduction in the mean ΔF values over the third visit as the study completed, although this was not statistically significant (P=0.797). R squared of 0.30 shows that 30% of the variance in mean ΔF at T3 was explained by the mean ΔF at first visit. The QLF participants appeared to have a lower reduction in mean ΔF from T1 to T3, and there was a statistically significant difference between WL and QLF participants (P=0.001).

The outcome measures in the study were measured at tooth level, but the randomisation was at participant level. Thus multilevel linear regression analysis was undertaken to control for the clustering of teeth within the participants. The adjusted mean ΔF was 0.08 (SE 0.11) and 0.68 (SE 0.11) in the QLF and WL groups respectively. The 95% confidence intervals (CIs) demonstrated the narrow spread of the results and indicated a statistically significant difference at the randomisation group level the QLF or WL groups (P=0.001). 15 of WL group had lesions on labial or buccal surfaces, and 17 of the QLF group participants had similar lesions. Table 7.7 shows adjusted mean ΔF from T1 to T3 for WL and QLF groups.

	WL Group	QLF Group
Mean ΔF 5 to 5	0.68%	0.08%
95% Confidence interval	0.22% to 1.14%	-0.38% to 0.54%
SE	0.11	0.11

Table 7.7: Adjusted mean ΔF from T1 to T3.

Percentage change fromT1-T3 in ΔF at tooth level of the occlusal surface lesions from the second molar to the second molar of 600 lesions showed an overall unadjusted mean percentage change of 0.18% (SD 0.40%). This was statistically significant with 95% CI showing a narrow interval of 0.14% to 0.21% with P=0.0001. In the 293 lesions of WL and 307 lesions of QLF groups, the percentage change was -0.19% (SD 0.56%) and 0.15% (SD 0.64%) respectively. This was not statistically significant with P=0.45. Adjusting for clustering of teeth at a participant level (Table 7.8) the mean percentage change in ΔF was 0.03 % (SE 0.04) and 0.02% (SE 0.04) in the WL and QLF groups respectively. The CIs indicate the mean reductions noted was significantly lower when comparing the two groups at T3 (P=0.84) with QLF showing more reduction. This showed 600 lesions in 57 participants. 293 lesions were seen in 30 participants of WL group, and 307 were seen in 27 participants in QLF.

	WL Group	QLF Group
Mean ΔF occlusal	0.03%	0.02%
95% Confidence interval	-0.04% to 0.1%	-0.05% to 0.09%
SE	0.04	0.04

Table 7.8: Adjusted mean ΔF Max of 5to 5 from T1 to T3.

The difference in the adjusted means between the WL and QLF groups was 1.21 (SE 1.03) with a wide 95% CI of -0.87 to 3.2(P=0.24). Additionally, the test of the fixed effect of the intervention showed no statistically significant difference between the QLF and WL groups (P=>0.999).

Percentage change in ΔF Max at tooth level of the 81 labial or buccal surface lesions from the second premolar to the second premolar from T1-T3 showed an overall unadjusted mean percentage change of 0.18% (SD 1.27%). This was not statistically significant with 95% CI showing a narrow interval of -0.10% -0.46% with P=0.20. In the 31 lesions of WL and 50 QLF groups, the percentage change was 0.30% (SD 1.47%) and 0.10% (SD 1.14%) respectively. This was not statistically significant with P=0.49. Adjusting for clustering of teeth at a participant level (Table 7.9), the mean percentage change in ΔF Max was and 0.61% (SE 0.16) and -0.06 % (SE 0.20) in the WL and QLF groups respectively.

	WL Group	QLF Group
Mean ΔF Max 5 to 5	0.61%	-0.06%
95% Confidence interval	0.3% to 0.92%	-0.45% to 0.33%
SE	0.16	0.20

Table 7.9: Adjusted mean ΔF Max of 5to 5 from T1 to T3.

The difference in the adjusted means ΔF max between the QLF and WL groups was 0.67 (SE 0.26) with a wide 95% CI of 0.13 to 1.21(P=0.016). The 95% CIs indicate the mean reductions noted was significantly lower when comparing the two groups at T3 (P=0.016) with QLF showing more reduction. Additionally, the test of the fixed effect of the intervention showed statistically significant difference between the WL and QLF groups (P=0.003).

Percentage change in ΔF Max at tooth level of the occlusal surface lesions from the second molar to the second molar from T1-T3 showed an overall unadjusted mean percentage change of 0.17% (SD 0.60%). This was statistically significant with 95% CI showing a narrow interval of 0.12% -0.22% with P=0.000. In the WL and QLF groups, the percentage change was 0.15% (SD 0.64%) and 0.19% (SD 0.56%) and respectively. This was not statistically significant with P=0.45. Adjusting for clustering of

teeth at a participant level (Table 7.10), the mean percentage change in ΔF Max was 0.02% (SE 0.07) and -0.13 % (SE 0.14) and in the WL and QLF groups respectively.

	WL Group	QLF Group
Mean ΔF Max occlusal	0.02%	-0.13%
95% Confidence interval	-0.11% to 0.15%	-0.27% to 0.1%
SE	0.07	0.14

Table 7.10: Adjusted mean percentage change in ΔF Max of occlusal lesions from T1 to T3.

The difference in the adjusted means between the QLF and WL groups was 0.15 (SE 0.15) with a wide 95% CI of -0.15 to 0.46(P=0.31). The 95% CIs indicate the mean reductions noted was significantly lower when comparing the two groups at T3 (P=0.31) with QLF showing more reduction. Additionally, the test of the fixed effect of the intervention showed statistically significant difference between the QLF and WL groups (P=0.003).

The overall unadjusted mean percentage change from T1 to T3 in ΔQ of labial and buccal surface lesions from the second premolar to second premolar was -112.42% (SD 995.51%). In the WL and QLF groups, the percentage change was -1.89% (SD 11.89%) and -180.95% (SD 1267.05%) and respectively. Adjusting for clustering of teeth at a participant level (Table 7.11), the mean percentage change in ΔQ was 0.56 (SE 0.85) and-15.34 (SE 14.82) and in the WL and QLF groups respectively.

	WL Group	QLF Group
Mean ΔQ 5 to 5	0.56	-15.34
95% Confidence interval	-1.1 to 2.22	-44.38 to 13.7
SE	0.85	14.82

Table 7.11: Adjusted mean percentage change in ΔQ of 5 to 5 from T1 to T3.

The difference in the adjusted means of ∆Q of 5 to 5 between the QLF and WL groups was 15.86. (SE 16.28) With a wide 95% confidence interval of -17.35 to 49.07(P=0.33). This suggests that there was a reduction in ΔQ in both groups. However the CIs indicate the wide variation within the groups from T1 to T3 and no statistically significant changes were noted (P>0.05). Additionally, the test of the fixed effect of the intervention showed no statistically significant difference between the QLF and WL groups (P=0.22).

The overall unadjusted mean percentage change from T1 to T3 in ΔQ of occlusal surface lesions at tooth level from the second molar to the second molar was -1.1315 (SD 17.93%). In the WL and QLF groups, the percentage change was -2.17% (SD 25.37%) and -0.17% (SD 5%) and respectively. Adjusting for clustering of teeth at a participant level (Table 7.12), the mean percentage change in ΔQ was 0.51 (SE 0.22) and -15.34 (SE 14.82) in the WL and QLF groups respectively.

	WL Group	QLF Group
Mean ΔQ occlusal	0.51%	-15.34%
95% Confidence interval	0.08% to 0.94%	-44.38% to 13.7%
SE	0.22	14.82

Table 7.12: Adjusted mean percentage change in ΔQ of occlusal lesions from T1 to T3.

The difference in the adjusted means of ΔQ of occlusal surfaces between the QLF and WL groups was -1.99 (SE 1.4) with a wide 95% confidence interval of -4.84 to 0.85(P=0.16). This suggests that there was a reduction in ΔQ in both groups. However the CIs indicate the wide variation within the groups from T1 to T3 and no statistically significant changes were noted (P>0.05). Additionally, the test of the fixed effect of the intervention showed no statistically significant difference between the WL and QLF groups (P=0.26).

7.4 DEMINERALISATION WITH VISUAL ASSESSMENT

Except for 17 participants, all the other participants had demineralisation on the labial or buccal surfaces at the start of the study under direct visual assessment. Of these, 11 participants had white spots on teeth that were clinically diagnosed as hypoplasia or molar incisor hypomineralisation. The total number of teeth with demineralisation lesions present at the participant level at T1 was 189 and at T3 were 189. On average each participant had 3 lesions on the labial surface with one participant having 18 lesions. The hypoplastic lesions were high as well in these participants, and there were 127 lesions altogether in 30 participants. 35 lesions were difficult to classify as demineralisation or hypoplasia and therefore categorised as unsure (Figure 7.6). These were included among the demineralisation lesions.

The most commonly affected teeth were the maxillary incisors followed by mandibular molars which accounted for 34.9 % and 24.8% of the lesions detected. The least affected were lower incisors followed by lower canines.

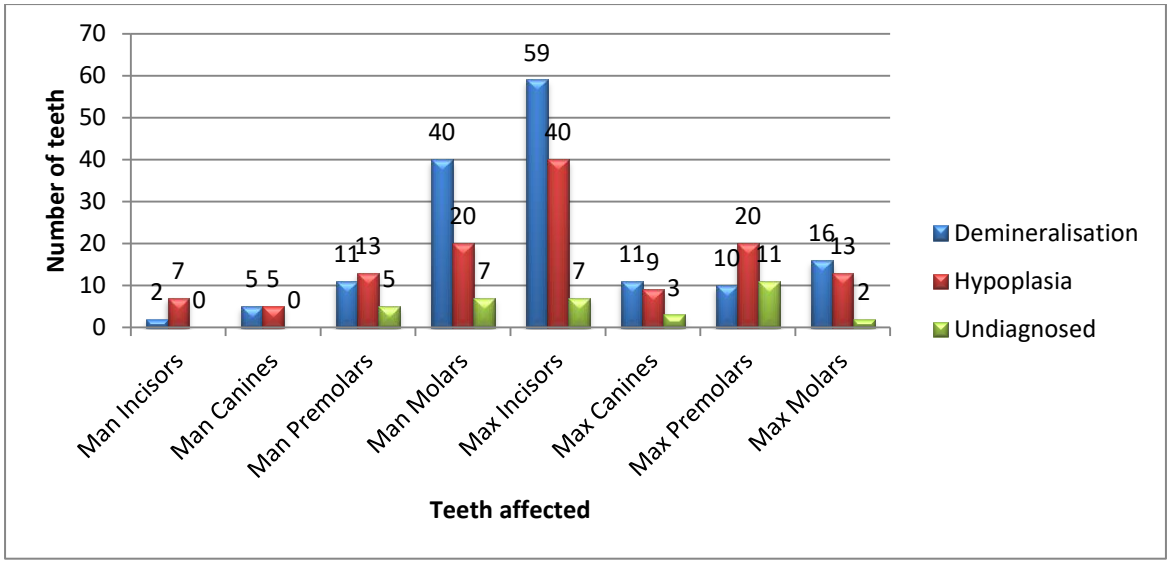

Figure 7.6: Number of teeth affected clinically by demineralisation, hypoplasia and undiagnosed white spots on the labial or buccal surface.

7.5 RELIABILITY ASSESSMENTS

7.5.1 QLF IMAGES

The QLF images were analysed by the main assessor and 3 additional examiners who had previous experience in the software and in analysing experimental data. Examiner 2 had the most experience using the software and therefore was considered as the gold standard for reliability. QLF images from 7 participants were used, and the assessors were given a questionnaire (Appendix 14.8) outlining which specific teeth should be assessed. There were 97 teeth with demineralisation and 35 images assessed for demineralisation and plaque data separately. The data obtained was continuous. Thus an intra class correlation coefficient (ICC) was used as a measure of inter-examiner reliability. The inter-examiner agreement for the assessment of plaque accumulation, measured by ΔR30 indicated a strong level of agreement with an ICC 0.987 when assessed by 4 examiners (Table 7.14). The results (Table 7.15) indicate moderate levels of agreement for assessing demineralisation, with ICC values of 0.773 for ΔF when assessed by 4 examiners. However, the inter-examiner reliability between the gold standard and the main examiner was 0.934 which showed strong levels of agreement for assessing demineralisation. The examiner who showed low reliability had the least experience in using the software and might have reflected in the ICC values with the most experienced examiner.

To measure the intra-examiner reliability of the demineralisation and plaque assessments, the main assessor examined the same images, in an alternative random order, on a second occasion one month later. All the data were noted to have strong levels of agreement (Table 7.14 and 7.15). The outcome measures, ΔF and ΔR30, had ICC scores of 0.995 and 1.0 respectively.

ΔR30	ICC	95% Confidence Interval
All examiners	0.987	0.978-0.993
Gold standard -Main examiner	0.997	0.995-0.999
Gold standard-Examiner 1	0.978	0.957-0.989
Gold standard-Examiner 2	0.997	0.994-0.998
Intra examiner reliability	1	1

Table 7.14: Inter- and Intra-examiner reliability assessment of the QLF images for plaque

ΔF	ICC	95% Confidence Interval
All examiners	0.773	0.704-0.831
Gold standard - Main examiner	0.934	0.902-0.955
Gold standard - Examiner 1	0.812	0.706-0.878
Gold standard - Examiner 2	0.665	0.530-0.765
Intra examiner reliability	0.995	0.993-0.997

Table 7.15: Inter- and Intra-examiner reliability assessment of the QLF images for demineralisation.

7.5.2 WL IMAGES

Similarly, using a WL images of 7 participants with demineralisation to record the data (Appendix 14.8) were assessed by the main assessor and 3 additional examiners to determine the inter- and intra-reliability of assessing demineralisation on WL images. The categorical data, analysed using kappa statistic, demonstrated that the inter-examiner agreement ranged from -0.0932 to 0.447 (Table 7.16). The intra-examiner assessment of examiner 1, which was conducted similarly as the QLF assessment after a month interval, demonstrated a kappa score of 1.0.

Examiner	1	2	3	4
1	1	0.447	0.239	0.051
2	-	-	-0.093	0.121
3	-	-	-	-0.147
4	-	-	-	-

Table 7.16: Inter-examiner reliability assessment on the WL images

7.6 SENSITIVITY AND SPECIFICITY ASSESSMENTS

The WL and QLF images were assessed to determine the ability of the examiners to identify the presence and absence of demineralisation correctly. This was undertaken in addition to the ROC analysis primarily to ascertain the sensitivity of the QLF image assessment. 36 WL images and their corresponding QLF images were assessed for the presence of demineralisation by three examiners. The images (Appendix 14.9) were shown in a random order, and the examiners were given proforma to complete if they could observe areas of demineralisation and mark the tooth. Their results were compared to an additional main assessor's analysis, which was assumed to be the gold standard. The main assessor noted 145 lesions on the QLF images, of which 102 could be identified on WL, giving the WL images a sensitivity of 0.70. The specificity of demineralisation detection on the WL images was 1.0.

The results (Table 7.17), demonstrate that the sensitivity of the WL images was moderately good. Examiner B and C correctly identified 97 and 95 lesions with a sensitivity of 0.93 and 0.91 missing 5 and 7 lesions respectively. Examiner A had much lower sensitivity scores of 0.46 missing 36 lesions. The specificity of WL image assessment was good, ranging from 0.81 -0.97 with the majority of sound images being correctly identified by Examiner A and C. Examiner A, B and C incorrectly diagnosed 2, 18 and 23 additional areas of demineralisation lesions respectively. The sources of error were incorrectly noting the presence of hypoplasia (46.5%), light reflection (46.5%) and staining (7%).

The sensitivity and specificity of the QLF images were found to be higher than WL images. The sensitivity scores were 0.96 and 0.97 with 9 and 11 lesions being missed respectively by Examiner A and B. Examiner C had lower sensitivity scores 0.55 with missing 65 lesions. Additionally, the specificity of QLF images was higher at 0.88-0.97. Examiner A and C incorrectly diagnosed 6 and 11 additional lesions, noting the appearance of staining to be demineralisation. Examiner B showed high specificity and identified only 3 lesions, noting staining to be demineralisation.

	A	B	C
WL Image sensitivity	0.46	0.93	0.91
QLF Image sensitivity	0.92	0.72	0.55
WL Image specificity	0.97	0.84	0.81
QLF Image specificity	0.96	0.97	0.88

Table 7.17: The sensitivity and specificity of demineralisation assessment.

ROC curves were used to assess the level of demineralisation measured on QLF images and that could be visualised on WL images was assessed using ROC curves. There were 143 areas of demineralisation noted on the QLF images. All of these areas of demineralisation were included in the assessment (Figure 7.7). 91 lesions noted on WL images did not show a ΔF value. Whilst 29 lesions without a white spot had a ΔF valuesuggesting presence of subclinical lesion. The area under the curve was 0.667 (95% confidence interval 0.611 to 0.722).

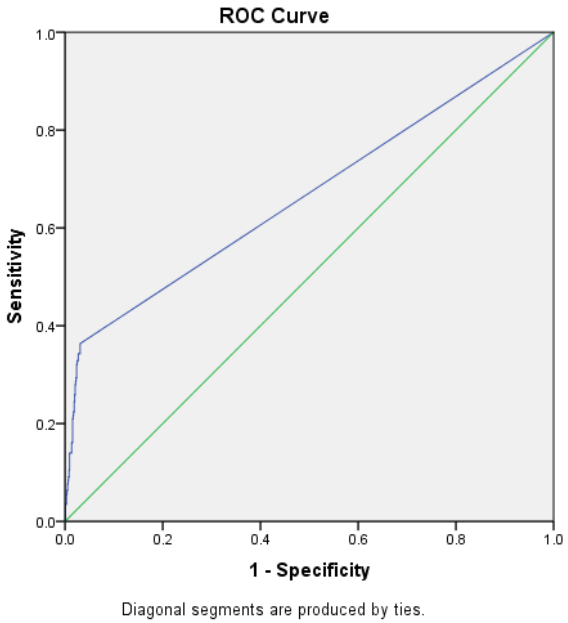

Figure 7.7: ROC of demineralisation assessed on WL and QLF images of demineralisation lesion on the labial surface of the second premolar to the second premolar in the maxillary and mandibular arch.

7.7 PATIENT PERSPECTIVE

A debriefing questionnaire (Appendix 14.7) was provided to all participants on completion of the study. The patients were asked to identify whether they were shown WL or QLF images. This question was added to identify their allocation which would determine the validity of their answers.

The results (Table 7.18), demonstrate that the patients were very positive about being shown the images. 100% of the participants found being shown the images helpful. 91.22% of the participants had no problems with taking photographs. 5 participants in WL group commented they had problems having the images taken. They commented they didn't like the stretching the lips and cheeks with the retractors. 89.24% could see areas of food accumulation. All the QLF participants could see the food accumulation, however, 6 of the WL participants was unsure if they saw the food accumulation. 89.4% of the participants could see the tooth damage. Interestingly, 2 participants from the QLF group were unsure if it would be useful to have the photographs taken whole way through treatment. Likewise, 7 of the WL group were unsure if it would be helpful to have photographs taken whole way through treatment. This was due to the discomfort from stretching of lips while taking photographs. Except for 1 participant in WL group, all other participants allocated to the QLF and WL group thought their tooth brushing improved over the appointments.

Questions	All	WL	QLF
Number of participants	60	30	27
Reported not having problems with the photographs	91.22%	83.4%	100%
Reported the photographs were helpful	100%	100%	100%
Reported tooth-brushing improved	98.25%	96.67%	100%
Reported able to see food accumulation	89.4%	80%	100%
Reported able to see tooth damage	89.4%	80%	100%
Reported it would be useful to be shown images for the whole duration of treatment	84.3%	76.67%	92.6%

Table 7.18: Patient perspectives of OHR with QLF-D^{TM} images

8.0 DISCUSSION

8.1 Summary of the main findings

1. OHR resulted in a reduction in plaque accumulation, assessed on the QLF images, from T1 to T3. This reduction was noted in all study participants, regardless of their allocation to the WL or QLF groups.

2. OHR provided at three consecutive hygiene appointments using WL or QLF images as visual aids does not significantly reduce the development of demineralisation.

3. There was no advantage in terms of plaque accumulation or demineralisation being given OHR using the QLF images as visual aids rather than the WL images.

4. The QLF image assessment displayed high levels of inter- and intra-examiner reliability.

5. The QLF images have a greater sensitivity and specificity, allowing lesions of demineralisation to be detected.

6. The patients' perspectives of the use of both the WL and QLF images, as visual aids for OHR, were positive. This suggests that the QLF images may be a suitable tool that could be used to supplement routine oral hygiene control measures in the orthodontic setting.

8.1.1 The relationship of OHR and plaque accumulation

Studies on OHR during orthodontics tend to focus on measures of periodontal health, such as bleeding on probing, gingival indices and plaque indices. Three systematic reviews found oral health promotion during fixed orthodontic treatment and oral health education programmes to show short term improvements in plaque levels and gingival health (104) (105)(106). These studies have largely used ordinal indices which may lack precision. In this study, plaque accumulation was assessed on the QLF images as ΔR30. This provides quantitative score that may increase the validity of the true effect of the intervention and is not a subjective score of plaque. A significant reduction in the adjusted mean percentage change ΔR30 at participant level was found in both groups from T1 to T3, of 45.36%(95 % CI of -4.8 to 95.52) and 58.93% (95% CI of 25.92 to 91.96) in the WL and QLF groups respectively. However, this was not statistically significant between the groups. Irrespective of the allocation group the participants were recruited, the study showed a statistically significant plaque reduction of 51.8% (95% CI of 40.36% to 63.26%) with a ($P<0.001$). The improvements noted in ΔR30 from T1 to T3 may be partly due to the patients being involved in a clinical trial. However, the trend continued despite having longer gaps than 3 to 4 weeks between appointments in patients who missed or cancelled appointments and from second visit to third visit. This highlights that the overall result is unlikely to be due to Hawthorne effect.

It is paramount that patients have adequate levels of oral hygiene to prevent caries and periodontal disease during orthodontic treatment. Hobson and Clark (1998), Eppright (2014) express it is an obligation of the treating clinician to ensure patients are advised about the importance of adequate plaque control and a method to ensure this is achieved. The clinician has the duty of care to monitor the effectiveness of patient's oral hygiene throughout treatment. If sufficient levels of oral hygiene are not being maintained to support treatment, the appliances should be removed to prevent progressing demineralisation and periodontal disease as this would be in the best interest of the patient. A survey was distributed to 1038 UK orthodontists to determine the oral hygiene advice routinely given to patients (107). There was a 46% response rate, with the results indicating that the majority of orthodontists routinely provide instruction on tooth-brushing, disclosing tablets and floss in addition to dietary advice. However, as the response rate was low, this might not depict the true picture. Many oral health promotion techniques have been proposed in dental health education programmes and orthodontics, including the use of a specially made videotape (Lees and Rock, 2000), disclosing agents for patients' to self-assess the effectiveness of their plaque control (Boyd, 1983), the provision of regular report cards with written feedback (Richter and Nanda, 1998), a personalised 40-minute oral health counselling session (Lalic et al. 2012), rewards such as coupons for ice cream sundaes for clinical compliance (Richter and Nanda, 1998) and weekly text message

oral hygiene reminders (Eppright et al. 2014), what's app chat room discussion (Francesca Zotti 2016), illustration catalogue (Ay 2007), Scaling and TV (Glavend and Zeunur 1985) and cognitive behaviour programme (Stewart 1991). Additionally, written instructions are often given alongside other techniques; although the former has been shown to be the least effective method of improving plaque scores (Lees and Rock, 2000).

This study demonstrated that regular verbal OHR with WL and QLF images as visual aids could be a useful technique in improving plaque control. Marini et al. found similar findings(108). In their study, 60 patients were recruited one month after bond up to receiving repeated OHI and motivational reinforcement by a registered hygienist at six 4-weekly visits or just at the baseline visit. The participants were randomly allocated to using an electric or manual toothbrush. The plaque coverage scores, graded using the modified Quigley-Hein index, were assessed at each visit by a blinded examiner, and demonstrated a statistically significant reduction with repeated OHI and motivation, regardless of the type of toothbrush allocation. This highlights that active reminder systems should be in place to reinforce the importance of adequate standards of oral hygiene throughout treatment. The present study showed that plaque levels consistently reduced, highlighting the benefit of the OHR intervention that was being given.

8.1.2 The relationship of OHR and demineralisation

Over the course of the study, there was no significant improvement in demineralisation in both the WL and QLF groups with an adjusted mean change in ΔF of -14.19 (95% CI; -14.98 to -13.02) and -14.9 (95% CI; -15.22 to -13.16) respectively. This was a short term study of mean 61 days and may have led to any intervention, using the OHR, having minimal effect on improving the ΔF values. A prospective longitudinal study (39) using QLF-D^{TM} on 51 patients who had completed fixed orthodontic treatment showed the median ΔF of lesions at debonding as 8.5 (Quartiles 6.6%; 11.9%). In the two year post-treatment assessment, 39% of lesions showed an improvement. There was a statistically significant improvement in ΔF within the first 6 months. However no further improvement was achieved following this. This suggests that lesions with a median ΔF 8.5 do have a potential for improvement, particularly immediately following debond. In this study, the mean ΔF at baseline was -14.19, indicating similar findings could not have been accomplished.

The most commonly affected teeth with demineralisation assessed using QLF were the mandibular premolars (26.5%) and maxillary premolars (25.7%).This included the occlusal demineralisation lesions as well as labial and buccal lesions. When comparing the labial lesions, mandibular molars had more lesions followed by maxillary molars and the maxillary central and lateral incisors. On the

WL assessment of labial and buccal lesions, the most commonly affected teeth were the maxillary central incisors (34.9 %) and mandibular molars (24.8%) and maxillary premolars (16.8%). It is slightly unusual for the maxillary central incisor to be affected to such a degree and the overall results were different from other studies. Stecksen-Blicks found the maxillary lateral incisors were the most commonly affected tooth in their RCT comparing the application of a fluoridated varnish against a placebo varnish in the prevention of demineralisation using digital image assessment (30). The authors suggest that the maxillary lateral incisor may be more affected due to its frequently crowded position palatally at baseline which would lead to greater plaque levels and thus be at a higher risk of demineralisation. The second most commonly affected tooth in their study was the maxillary cuspid and the maxillary central incisor in the fluoride and placebo varnish group respectively. Our study had participants who had poor oral hygiene and this cohort is different to participants in the demineralisation studies and therefore the data shows difference in the teeth showing demineralisation. Khalef systematic review found poor oral hygiene was the highest risk factor for developing demineralisation with an relative risk of 8.55 (109).

8.1.3 OHR using the QLF images compared to the WL images

The study did not show a statistically significant difference between QLF and WL images used as visual aids for OHR. QLF images are more effective with regards to plaque accumulation, as deposits can be visualised more easily than on WL images. However, the only mature plaque is seen as red areas and therefore might not show the true picture of patients who are good at brushing on the day of the appointment and not being consistent in their approach and technique. The gingival status is not accounted for using QLF images, but this can be clinically assessed. Patients are more receptive to advice as mature plaque is evident on the QLF images. By making them aware that the plaque is mature and has not been brushed for 3 days reduces the resistance from patients in giving lame excuses of why they could not complete a thorough brushing on the day of the appointment. Disclosing agents overestimate the plaque and this can be more advantageous at times for those patients who are not consistent in their OH management. Visualising mature and immature plaque by using disclosing tablets is beneficial to use at home provided patients are fully aware of the color differences of mature and immature plaque. WL images were more effective with white spots than QLF images again as they can be visualised better than faint darker areas on QLF images. This could also be due the variation in the color of the tooth, giving more contrast and therefore white spot lesions are easily noticeable.

A possible factor which may have contributed to plaque reduction in our study group could relate to patient motivation as they were waiting to be added to waiting list for orthodontic treatment. A

cross-sectional study by Ericson et al. found that adolescents with more negative perceptions, attitudes and behaviours to oral health had poorer oral hygiene with higher plaque and gingivitis scores. Although it is important that patients are adequately informed, unless they display sufficient levels of motivation, the specific method used during the OHR session will be largely ineffective and will not necessarily result in reduced demineralisation and periodontal disease. Oral hygiene compliance is one of the most important factors controlled by the patient during orthodontic treatment. Hadler-Olsen et al. assessed patient compliance following oral hygiene instructions using a questionnaire and found a relationship between the level of reported compliance and the number of areas of demineralisation that had developed during fixed orthodontic treatment, although this relationship failed to reach statistical significance. Umaki et al. found that non-compliant patients had a higher frequency of stressful life events, and suggested that the response of patients to stress will affect their ability to follow a maintenance programme.

Systematic reviews that analysed if knowledge and attitudes could be improved through dental health education, only one study showed positive effects among multiple studies. Tolvanen et al, reported that children in the experimental group of the randomised controlled trial tended to improve their behaviour more than did those in the control group (110). The authors concluded that the oral health-promotion programme could improve oral health-related behaviour but has less effect on improvement of knowledge and attitudes. de Farias et al. observed the outcomes knowledge, attitude and practices regarding oral health and plaque and gingival scores(111). Final plaque scores ($P = 0.014$, OR = 0.46, CI = 0.24–0.86) and gingival bleeding ($P = 0.013$, OR = 0.49, CI = 0.28–0.90) indices decreased more in the experimental group. The experimental group also showed a statistically significant difference ($P < 0.001$) between the numbers of correct answers in the questionnaire after the education intervention. The authors concluded that contextualised educational activities in the school routine had positive effects on oral hygiene and the level of information about oral health, although the more informed individuals did not always practice better oral hygiene. It may be that patients do not fully comprehend the potential risks that may result from poorer oral hygiene and dietary control combined with poor motivation makes it harder for clinicians to improve oral health related beahaviour. Peng et al. demonstrated a significantly greater improvement in plaque and gingival indices in patients who were shown images of the severe potential consequences of plaque deposits compared to a group who were only provided with routine OHI (112). This suggests that greater patient awareness of the risks and hence the importance of plaque control improved their motivation, which led to better oral hygiene. In addition to motivation, self-application of the technique is important to ensure patients are aware of the level that they should aim to achieve. A short duration study by Ay et al. assessed different oral

hygiene motivation methods including verbal information with the model demonstration, verbal communication with catalogue illustration and the former methods with patient self-application under the supervision of the treating clinician. The group who had verbal OHI with catalogue illustration alongside self-application had significantly lower plaque scores at 4 weeks (113).

Systematic reviews shows that repetition and reinforcement of dental health education instructions are key factors to improving oral hygiene performance long-term (105)(106). The results of these reviews showed plaque removal programmes effective in the short term but no long term benefits and cannot be supported if costs were considered. Boyd et al. (1983) conducted a study to evaluate the effect of plaque control measures on gingivitis and found that a structured plaque control program only was effective in reducing dental plaque and gingivitis, provided there was periodic reinforcement at 4 to 7 week intervals; otherwise, the gingivitis scores tend to increase to pre-orthodontic treatment level, on cessation of reinforcement (114). A study involved taking plaque samples from participants and showing the patients the live, motile bacteria present in their plaque using a phase contrast microscope (115). This study included two other interventions which conventional plaque control measures and chair side motivational tests with conventional plaque control measures. The authors found this, alongside conventional plaque disclosure and OHI was more effective than conventional plaque disclosure alone. They advocated this had a long-lasting effect on plaque levels, which would reduce the need for conducting regular OHR. However, only a 6 month period was assessed. The majority of these studies are short term studies. Several studies have demonstrated that the first months after placement of braces are the most challenging for patients who have to familiarise themselves with and acquire new manual skills to maintain the orthodontic appliance in an adequate hygienic condition. During the middle part of orthodontic treatment, patient enthusiasm and motivation tend to decrease progressively, often leading to worsening oral hygiene. At this time, motivational strategies play a crucial role in maintaining adequate compliance until the final phase of treatment, when patients' motivation usually increases due to the approach of braces debonding. It would be beneficial to follow patient during a full course of orthodontic treatment and use QLF-D^{TM} as a visual aid for OHR. This could potentially show the benefit of plaque reduction and as a result a possiilty of low demineralisation post orthodontic treatment.

8.1.4 The sensitivity and specificity of the QLF images

QLF-D^{TM} device provides quantitative scores to allow monitoring of plaque coverage and demineralisation. QLF has been shown to detect subclinical lesions (15)(80)(90). ROC curves have been used to determine that a maximum combined sensitivity and specificity of 1.65 was noted at a

level of demineralisation of ΔF 7.25, indicating that this is the level demineralisation will become apparent using direct visual assessment (116). Boersma et al. (2005) found the mean fluorescence loss of lesions noted with direct vision was greater than 15% and those with QLF was 12.6%, indicating the results of this study are more sensitive (15). ROC curve did not show a good discrimination using QLF images in this study. This is partly due to having similar hypoplastic and developmental opacities in participants along with demineralisation lesions. 91 lesions were seen on WL images which had no ΔF scores using the QLF Images , and there were 29 lesions those were subclinical lesions with no white spot.

The ability of QLF to detect more demineralisation was supported by the sensitivity assessments which demonstrated moderate scores of 0.55-0.92 and 0.46-0.91 for the QLF and WL groups respectively. In contrast, the QLF and WL images had higher and good specificity scores of 0.88-0.96 and 0.81-0.97. There was significant variation in the results with examiner A showing low sensitivity with WL images and Examiner C showed low sensitivity for QLF images. Altogether 72 images were used for assessing sensitivity and specificity of three examiners. On the WL images multiple areas were incorrectly diagnosed. 46.5% were a light reflection and 46.5% were hypoplasia and other enamel opacities were incorrectly diagnosed as demineralisation. It is also possible that some of the demineralisation lesions were shallow and therefore did not give a ΔF score. It could also be possible that the image was further away or at an angle and did not provide the contrast needed. The incorrect diagnosis of light reflection as demineralisation is of particular concern as a large number of studies in the literature use digital images for demineralisation assessment and may suffer from this source of error. Diagnosing hypoplasia or development enamel opacities can be very difficult to ascertain using WL images. Clinical assessment has to be undertaken thoroughly in addition to using visual aids such as WL or QLF images to allow a fully accurate diagnosis.

8.1.5. The inter- and intra-examiner reliability of the QLF and WL image assessment

The good inter- and intra-examiner ICC scores for ΔR30 and ΔF on the QLF images demonstrate that QLF image assessment is reliable amongst different examiners. The inter-examiner ICC for ΔR30 was 0.987 which was excellent. The inter-examiner ICC for ΔF was 0.773 which was good. The inter examiner reliability was assessed separately for each examiner to the gold standard examiner who had the maximum research experience. This found one examiner to have moderate reliability compared to the gold standard. The main examiner had excellent reliability to the gold standard with ICC of 0.997 and 0.934 for ΔR30 and ΔF. This variation among examiners is likely to be attributable to difficulties in outlining the extent of some of the demineralised lesions. The outline

has to be in the sound enamel and this can be difficult when the lesions are in the cervical area are closer to gingiva. This study involved examiners with prior research experience. This is important as the reliability study by Pretty et al. (2002) found that a novice who had few hours of previous experience with the QLF analysis software had higher levels of disagreement than more experienced assessors.

The outcomes for the study on which the conclusions were based were ΔF and ΔR30. In contrast, the ΔF, ΔF Max ΔQ values of the individual areas of demineralisation in the participants from T1-T3, had very large variations. This was due to having multiple lesions not being analysed from having disclosing agent covering the occlusal surfaces following prophylaxis. Also, minor residual plaque deposits were occasionally present despite a prophylaxis having been performed which contributed to difficulties in ascertaining the outline of the lesions. This led to assessing the mean percentage change with some of lesion s having a baseline 0 value and some lesions having 0 for T3. This variation is also likely attributable to difficulties in outlining the extent of some of the demineralisation lesions. Pretty et al. (2002) discuss similar difficulties in outlining lesions lying adjacent to confounding factors such as enamel defects and stain. They report these lesions raise the complexity of image analysis, although suggest that with rigid instructions in place to ensure appropriate management of these areas, high levels of reliability can be achieved.

The inter-examiner reliability of demineralisation assessment on WL images using kappa ranged from -0.0932 to 0.447, indicating a poor reliability in comparison to the QLF image assessment. This confirms that often demineralisation can be difficult to detect in from photographs accurately. This significant variation could have been due to several reasons. The images used had 14 that were of occlusal surfaces which are difficult to assess for occlusal demineralisation. Patients having enamel opacities and hypoplasia, optical light reflection and staining could have resulted in misinterpreting the white spots. This is quite different to previous studies which have shown good kappa scores (116). Stecksen-Blicks et al. study found similar scores in their WL reliability measurements of demineralisation. Their intra- and inter-examiner assessment results, using kappa, were 0.77 and 0.69 respectively (30).

8.1.6. The positive patient perspective of the use of the QLF-D™ images for OHR

The participants' perspectives of having regular OHR with the QLF and WL images as visual aids demonstrated a positive response. Except for two participants in the QLF group, the rest were very positive on use QLF images. 92.6% of the participants allocated to the QLF group felt it would be

useful to have the OHR for the complete duration of treatment, compared to 76.67% of the WL group, with an odds ratio of 0.26 (95% CI: 0.04-1.3). This did not show a statistically significant difference in the participants' opinions between the groups (P> 0.05).However, more participants in QLF image found it more useful as an oral hygiene aid. This was different to the results shown by Miller et al. who found an odds ratio of 2.3 (95% CI: 1.5-3.5) with all QLF participants giving a positive response to the questionnaire and only 81% from WL images giving a positive response. This difference in our study could be due to few reasons. Our participants were not exposed to routine photographs being taken and this intervention was their first experience with a clinician in the orthodontic department. Majority of the participants found stretching with retractors uncomfortable during exposure with photographs. Whereas in the study group of Miller et al., the participants were in treatment for a while with repeated photographs taken each visit and comfortable using retractors. They could see the areas with plaque deposits which was difficult to visualise and difficult to access. They would have seen the benefit of maintaining good oral hygiene and the benefit of not developing demineralisation, making them more receptive to the idea of the usefulness of the images taken throughout the orthodontic treatment.

Patients often have difficulty localising plaque deposits due to the nature of the plaque deposits. WL images are similar to conventional photograph and except for the fact that participants could see them close enough to identify food deposits and plaque. Demineralised lesions were more obvious, and participants found it easier to locate them on WL images. QLF images showed plaque as red fluorescence, and this was very obvious. Participants were more willing to locate these areas and took the advice provided with less reluctance. Also the fact that mature plaque is seen only after 3 days, suggested their oral hygiene maintenance technique was inadequate if the patients could see any red areas. The clinician found less resistance to listen to the advice provided to participants shown QLF images compared to WL images. It was clear that this could be due to better visualisation of plaque. Disclosing agents showed the plaque and pellicle whereas QLF images showed red areas that were more than 3 days old. Participants who did not improve significantly even after showing QLF images were possibly the ones who were not motivated enough to have orthodontic treatment. The questionnaire did not have questions to identify their attitude on orthodontic treatment, motivation or knowledge of maintenance of good oral hygiene. Some participants felt they still had time to improve before the consultants review OH and there is still time and scope for improvement. One participant decided not to continue with orthodontic treatment as they found they could not commit to regular monitoring of oral hygiene and dropped out. One participant completed the trial, however, mentioned at the final appointment that they might not go ahead for orthodontic

treatment. These two participants were over 16 years of age. Nevertheless, this did not result in any statistically significant differences being detected between the QLF or WL groups.

A structured questionnaire was distributed to 122 patients undergoing active orthodontic treatment with fixed appliances in a study by Berlin-Broner et al. (117). Patients were treated by 38 different orthodontists. The questionnaire accessed information regarding instructions patients received from their orthodontist concerning maintenance of their oral hygiene during orthodontic treatment. A significant positive correlation was found between explaining the patients the importance of tooth brushing and instructing them on how to brush their teeth correctly. Most of the patients (94%) reported that their orthodontists informed them at least once about the importance of tooth-brushing, and 74.5% received instructions for correct performance of tooth brushing or were referred to the dental hygienist. It concluded emphasising the necessity of orthodontists to increase their commitment to providing thorough and comprehensive oral hygiene advice to patients to reduce the risk of developing caries and periodontal disease. This study suggested that an increasing interest during the last 5 years in the use of new technologies in orthodontic patient motivation protocols, with several studies demonstrating the efficacy of short message service (SMS) and e-mail reminders in improving patient compliance with and acceptance of orthodontic treatment (118)(119).

Questionnaires with quantitative investigations focussing on patient motivation levels are infrequently conducted in studies to knowing attitude, behaviour and motivating factors in orthodontic patients. Assessing the patient's perspectives' of the methods of OHR used in this study is advantageous to gain their opinion.

8.2 Study Limitations

8.2.1 Sample size

The sample size was limited to 60 participants due to the constraints of recruiting according to the inclusions criteria and providing detailed OHR at three consecutive appointments. There was no data in previous studies on the similar population to base a sample size calculation. Thus it was deemed acceptable to recruit at least 60 individuals to ensure adequate clinical time was available for standardised appointment intervals for all participants. If we recruited a sample with an excessive number of subjects it would have taken two to three years to complete data collection. The study was subsequently conducted as a pilot study, and a formal sample size calculation was not carried out. The results of this study would allow estimation of parameters for a sample size calculation to

be conducted in future definitive studies. However, in this study there is a greater risk of type 2 statistical error, due to the sample size lacking statistical power to detect a difference.

8.2.2 Sample

The baseline mean plaque percentage using the Quigley Hein Index was and 3.37 (SD 0.58) and 3.68 (SD 0.59) for WL and QLF respectively, which was not statistically significant (P=0.81, t-test). The baseline means ΔR30 was 2.3(SD 2.1) and 4.2 (SD 3.1) for the WL and QLF group participants respectively. However, the baseline means plaque percentage using the O'Leary index was 45.3 (SD 50.1) and 58.9 (SD 33.03) for the WL and QLF group participants respectively which were statistically significant. This could be a source of bias as the two groups were different when considering O'Leary plaque record and therefore the difference at the end of the trial could be due to the existing difference. However, it might also be due to the insensitivity of the plaque score as it is represented as a percentage compared to the QHI Index or ΔR30.

Of the 60 patients, 53.3% were female, and 46.6% were male. In the WL group, there were 13 females (43.3%) and 17 males (56.7%). In the QLF group, there were 19 females (63.3%) and 11 males (36.7%). This could be a source of bias in QLF group. The mean plaque reduction for females was 52.57(SD 49.12) and males 51 (SD 36.69). Males have been shown to develop more demineralisation during fixed orthodontic treatment and of a greater severity as male patients take less effort to maintaining good oral hygiene (13)(15)(27)(86) . The girls are shown to take more effort in maintaining good oral hygiene. However, our data did not show a difference. There seem to be more female patients taking orthodontic treatment, and sometimes the difference could be solely based on this difference in the gender differences of individuals seeking orthodontic treatment. Al Maaitah et al. assessed 230 patients on completion of fixed orthodontic treatment, 65% of which were female (86). This study recruited 32 females and 28 males, and this was not found to be different than reported studies.

Of the recruited patients, all participants had demineralisation at baseline. Labial or buccal surface demineralisation lesions were present in 32 participants, and all participants had occlusal demineralisation. The changes in demineralisation at a participant level were conducted on all of the individuals that were recruited. The primary tooth level change assessed was the percentage change in ΔF for labial and occlusal lesions. Mathematically a percentage change cannot be undertaken when the baseline value (T1) is 0. Thus this assessment was conducted on the participants who had demineralisation at baseline. Additionally, having no value at the T3 will give a score of 1 when calculating percentage changes and this can artificially create a positive result. Therefore 3

participants who were dropped out were removed when this calculation was undertaken. This is unlikely to be a significant source of bias as the number of individuals who were excluded from the assessment who did not have lesions was 15 in the QLF group and 13 in the WL group, which is relatively similar. Additional analysis on demineralisation at tooth level, regarding the mean total ΔF per tooth, was undertaken to include the results of all of the participants to account for this. Furthermore, as mentioned, except for 3 participants who dropped out the rest of 57participants of the sample were included in the participant level analysis.

8.2.3 Study duration

The study was a short term study with a mean duration of 61 days. Lack of any significant changes being noted in demineralisation may be that the study was relatively short. We know from previous studies that demineralisation with orthodontic treatment is a slower process compared to when in treatment. Longitudinal monitoring of demineralisation lesions is required over 6 months. An RCT involving the use of a weekly text message being sent to the parents of patients undergoing orthodontic treatment and a control group who did not receive such a text reminder found no difference in the prevalence of demineralisation, measured by direct visual examination, between the two groups (119). However, there was a trend in the control group for increasing levels of demineralisation to occur between two and four appointments after baseline. Bleeding Index, Marginal Gingival Index, and Plaque Index scores were significantly lower in the text message group than in the control group at T2.The authors advised that to assess the development of demineralisation with an intervention accurately, longitudinal monitoring should be undertaken for greater than six months. Orthodontists should add an active reminder system of the importance of oral hygiene compliance to their typical protocol during treatment. They also found that the significant improvements that were noted in the gingival indices did not appear to influence the development of demineralisation. Although, again this could be due to the short duration of the study as it was 5.44 months.

Another RCT by Miller et al. the participants were assessed over five visits (T0-T4), held at approximately six weekly intervals (116). The overall length of involvement in the study was on average 30 weeks, slightly over six months found no significant difference in the development of new additional lesions.

Khalef found that increased treatment duration was correlated with significantly more areas of demineralisation with an RR of 3.65 when treatment length was >36 months compared with <24 months (109). It would have been advantageous to assess the impact of OHR when these

participants start their orthodontic treatment. How much of the improvement is maintained would decide the actual benefit of this trial being undertaken. These participants, especially with multiple demineralisation lesions, would certainly benefit with having QLF images taken throughout the whole active orthodontic treatment. These participants would benefit from reminders on OH and post debond assessment of the demineralisation lesions would be beneficial. A recent RCT used an app-based approach in a protocol for domestic oral hygiene maintenance in a group of adolescent patients wearing fixed multibracket appliances (118). Study group (SG) patients were enrolled in a WhatsApp chat room–based competition and instructed to share monthly with the other participants two self-photographs (selfies) showing their oral hygiene status.SG patient participation in the chat room was regular and active throughout the observation period. At 3, 6 and 9 and at the end of the first year, SG patients had significantly lower values of both Plaque Index and Gingival Index and a lower incidence of new white spot and caries, compared with the control group. Also, following the participants after the appliances were removed would have allowed assessment of post-debond changes that may occur. This is a limitation that has been recognised by other similar short-duration studies. However, long term full treatment studies become cumbersome to complete, expensive and time-consuming.

8.2.4 Blinding

The allocation was concealed by the use of consecutively numbered, sealed opaque envelopes to reduce selection bias. The participants were identified as eligible for the trial by the Orthodontic consultants who provided the information leaflets and consent forms to the participants. The participants were contacted by the main examiner and recruited if the participants were interested in taking part in the trial. The OHR was given to all participants by the same clinician who was the main examiner. It was not possible to blind the clinician to the treatment allocation, which has the potential to lead to bias. However, the OHR advice was standardised instructions, with the only difference being the areas of focus, which were the plaque and demineralisation areas that could be visualised on either the QLF or WL images. Thus, there should be minimal bias as a result. Similar studies assessing OHR (113)(116) have used the same clinician for standardisation purposes.

8.2.5 Time point of data collection

Participants were seen for OHR every 3 to 4 weeks as was routine at the dental hospital hygiene appointments. Before the start of the trial, an audit was undertaken by the main examiner to check the failed to attend rates for the hygienist and was found to be 40%. During the trial, each participant were contacted by the main examiner on Friday for their appointments booked for the

next Tuesday. Occasionally, patients cancelled or failed to attend their appointment which was not within the control of the main examiner and demonstrates the difficulties with conducting clinical trials. However, in real-life participants receives text reminders the day before their appointments at the dental hospital which was not provided as a part of this trial. These participants were rebooked for the next available session, however assuming that changes in ΔR 30 and ΔF occur linearly over time, a short delay could have potentially lead to performance bias in that there was a greater period for demineralisation to develop or improve. The mean duration in WL group of 61 days (SD 20) and 60 days (SD 34) in QLF group indicating the limited variation of the participants' study duration. This is likely due to the imminent rescheduling of patients who cancelled or failed to attend their appointments. Despite providing 4 appointments to one participant, they did not attend further appointments and 2 of the participants failed two appointments and dropped out from the trial.

8.2.6 Data analysis

The mean number of teeth assessed per patient was 23.1 (SD 1.3) with no statistical difference noted between the WL or QLF groups. The variation was related to the number of teeth present in the arches, in that second molars might have been erupting or premolars have been erupting as the majority of the participants were 13 years of age. There have been some discrepancies with including all the teeth that were visible in the image and not excluding these teeth if they were not present in the next visit. As images were recoded and anonymised the discrepancies were checked following analysis, and we found 22 teeth that were not visible. It was necessary to exclude teeth that could not be fully assessed. It may have been more appropriate to have completely standardised the process. However, this would have been difficult as there were 600 images those were analysed for demineralisation and excluding posterior teeth would not have shown the true outcome. In this study, all teeth visible in the image was analysed and no limitation was placed on which teeth to assess for plaque coverage and demineralisation. It was possible to assess the first molars and even second molars on the QLF and WL images of some participants, allowing their inclusion. Should these teeth have been excluded, it would have allowed the assessment to be more standardised, although it would have reduced the amount of data obtained. One of the limitations we found with the study was there were multiple demineralisations or enamel opacities visible on WL images that were not showing demineralisation as a darker area on QLF images. These lesions could have been hypoplastic lesions or a tooth that was comparatively discoloured to the rest of the teeth, and therefore white spot was more obvious on the WL image whereas QLF did not show any darker area or ΔF value. It could also be postulated that these teeth being further away from the

Biluminator and might not be in focus which reduced the sharpness and does not show darker area. ROC showed AUC of 0.667 of buccal or labial lesions from the second premolar to the second premolar with a 95% confidence interval 0.611 to 0.722. This showed a discrimination that was slightly better than by chance. It would have been ideal to take multiple images of any tooth that showed white spot lesion to confirm the area and this would help with monitoring the lesion longitudinally. However this would have made each appointment longer, and participants might have found this difficult to cope. Participants commented the photographs to be the difficult part as they found stretching with retractors uncomfortable.

Majority of the studies have assessed anterior maxillary teeth or maxillary incisors, canines and premolars (30)(120) (121). Bailey et al. assessed upper and lower incisors, canines and first premolars in their RCT(120)(122). Some studies looked at maxillary and mandibular anterior teeth from canine to canine and Miller et al. assessed as many teeth that were visible including first molars, as these participants had extractions of premolars (116)(123). Our study had participants with different malocclusions and therefore to get all the teeth from the first molar to the first molar was difficult especially when they were severe class II or class III incisor and skeletal relationship. The participants were given a unique identification number on enrollment which was used throughout. Following data collection, these images were recoded and anonymised as the main examiner who provided the OHR completed the data analysis. To avoid recall bias, the analysis was completed 3 to 6 months after data collection, and the WL and QLF images were recoded to analyse. However, a risk of recall bias still exists especially with certain type of malocclusion that can be characteristic for a participant.

Despite the use of customised software for image analysis, measurement bias has been reported (85).This is due to the subjectiveness of marking the outline of the lesion for QLF assessments. This is more so for WL image analysis and clinically when assessing demineralisation even though strict guideline was used is unlikely to be significant in this study as strict guidelines were in place regarding the assessment of lesions. The RCT by Eppright et al. used the following scale for assessing demineralisation clinically (119):

1. No visible white spots or surface disruption

2. Visible white spot without surface disruption

3. Visible white spot lesion with roughened surface

4. Visible white spot requiring restoration

Gorelick's scale is commonly used for assessment of white spots in most studies. Clearly, not only is there similar risks of subjectivity associated with such a scoring system, but there is poorer accuracy with regards to the classification of the severity of the demineralisation present. Photographs tend to overestimate the incidence of opacities, partly due to the reflection of the flash from the tooth surface which is similar to WL images even though this is much less severe. Benson et al. study found difficulties with standardisation of the procedures , particularly concerning the wetness of the tooth (124). However, QLF has been validated in *in vitro* and *in vivo* studies, suggesting it is a valid assessment tool. Al-Khateeb et al. demonstrated a strong correlation between QLF fluorescence changes and TMR observed mineral loss (74).

The data analysis was standardised by the same examiner conducting all of the image analysis. The images were recoded to prevent observer bias and analysed 3 to 6 months after data collection to reduce recall bias. Intra-examiner reliability assessment of this examiner demonstrated high levels of consistency, suggesting good reliability for the main examiner. Also, the inter-examiner reliability assessments conducted demonstrated high levels of agreement, as has been reported in previous studies (87)(116). One of the examiners showed a moderate level of agreement and this could be due to the reduced experience as previously reported. Pretty et al. selected 16 demineralisation lesions of varying size and severity to be assessed by 10 examiners using QLF software of which majority of the significant differences in the agreement was from a single novice (87). This method of using different types of lesions provided a sample with a range of difficulty. Similarly, in this study, the images that were chosen to be used were of different sizes and severity. However, randomly selecting the images would have been ideal to reduce any associated bias. As not all patients had demineralisation lesions, these lesions had to be deliberately chosen. Miller et al. and other studies have randomly selected patients or images(120) for WL image assessment (30) (116) (120).

8.2.7 Bias associated with the method

Participants were seen for 30 minutes at each visit by the main examiner. This time was allocated for providing OHR using QLF, or WL images along with feedback on how they followed the instructions and data were collected on how often they brushed their teeth every day, use of the interdental brush, floss, and disclosing tablets per week. Participants had their teeth disclosed and plaque score was recorded using O'Leary plaque record and Quigley Hein Index at each visit. Following this, they had prophylaxis with full mouth scaling and polishing with prophy paste. QLF images were taken post prophylaxis at T1 and T3 visits. Demineralisation lesions were identified, and patients were informed after prophylaxis was completed. There were time restrictions to provide all the above and at times removal of plaque from occlusal surfaces proved difficult. As this was the case for all of the

individuals regardless of their treatment allocation, it is unlikely this would have significantly affected the between- group results. However, this impacted when data analysis was undertaken. Occlusal lesions with obvious disclosing agent ΔF assessment was not completed on these teeth which resulted in showing a statistically different number of lesions at T1 and T3 visit. After data analysis, a further analysis was undertaken on these teeth to check if the lesions had fully subsided or were missed and the analysis showed no statistically significant difference between the numbers of lesions or the ΔF value. It would have been interesting to ascertain if allocating more time to give further detailed OHR instructions affected the results and led to reduction in the lesions.

Hobson and Clark have advocated that it may be more cost-effective for oral hygiene measures to be provided by trained auxiliaries (107). Before start of the trial, the main examiner observed the hygienist at the dental hospital and developed the standardised protocol on the OHR based on the clinical practice that was routine at the hospital. These appointments are routinely 30 minutes long, and patients were seen fortnightly for three visits. Having to take QLF images at each visit and providing OHR based on this along with what is routinely practised meant more time allocation for the trial. However, this would have reduced the number of participants that could have been seen every session and the trial would have taken longer to finish. Prophylaxis was conducted if plaque deposits were present before the QLF-D^{TM} images were taken for demineralisation assessment. Every patient required a scaling and polishing all three visits. Despite this, occasionally, residual plaque deposits remained, or calculus was present that was not removed during the prophylaxis. This contributed to difficulties visualising demineralisation during QLF image analysis. It may have been advantageous for the prophylaxis to have included ultrasonic scaling of occlusal surfaces, although there would have been time implications associated with including this and still it can be difficult to remove stain.

8.2.8 Performance bias

Risk of performance bias was high in this study as these were participants who had poor oral hygiene and could not have been added on to the waiting list for orthodontic treatment without showing an improvement in oral hygiene. The patients are usually added to the waiting list when the O'Leary plaque record is below 15%. As in any clinical research study, there is a risk of performance bias when the participants are aware of being assessed. However, this could have been higher in this study group as the majority wanted orthodontic treatment. Data was not collected on the day of recruitment and was collected only when intervention was provided. It would be hard to say if the oral hygiene improved and whether this impacted on the plaque score during this time period.

8.2.9 Confounding factors

The study showed no statistical differences between the WL and QLF groups in terms of age or gender, which demonstrates the groups, were well balanced in terms of these potentially confounding variables. The baseline QHI plaque scores and ∆R 30 were similar in WL and QLF group wheras O'Leary plaque record showed statistically significant difference between the groups. However, when the repeated measure of analysis of variance was conducted for the overall mean ΔR30 for the participants at T1 to T3 using the first visit as a covariate, this was not found to be statistically significant. Only 29% of the variance was attributed to the ΔR30 at first visit. The participants were randomly allocated to the QLF and WL groups at baseline, without stratification for the presence of demineralisation. It can be assumed that patients with baseline demineralisation present a higher risk of developing additional lesions, which would have the potential to confound the results if not controlled for. However, our group had almost equal participants with labial demineralisation lesions with 15 from QLF and 17 from WL group. All the participants had occlusal demineralisation lesions, and therefore the risk of this factor being a confounder is low. Also, the fact that this was a short term study, any difference shown would have been a clinically meaningful difference between visits or between groups. Al Maaitah et al. found the pre-treatment oral hygiene status and the presence of diseased first molars were related to the number of areas of demineralisation and the severity of demineralisation respectively. In our study, all participants had occlusal demineralisation, and therefore this might not have been a confounding factor (35).

A potential confounding factor was the variation in oral hygiene practice between the individuals. Some of the individuals followed the instructions given to them, and the rest might not have. All participants brushed their teeth twice as advised. Some would have spent more time brushing on the day of appointment as they were getting assessed. Some of them could have brushed their teeth in the waiting room before the appointment, which would have led to differences in the plaque accumulation. The time of the day that the appointment was held may also have been a potential factor, in that if the appointment was after breakfast, residual food deposits may have been present. The patients were seen from 9 am to 12.30pm and therefore the time between brushing and appointment could have acted as a confounding factor. In real life scenario, patients maintain oral hygiene levels in a similar way and therefore standardising this would have created an unrealistic situation and seeing participants at the same time each visit or seeing all participants same time was not considered reasonable. By assessing the real-life situation, the effectiveness of our intervention was assessed compared to efficacy. This would have added to the external validity of the trial. Additionally, the oral hygiene products that were used at home were not controlled. All of the participants were given standardised OHI on tooth brushing techniques and the daily use of

mouthwash, alongside the frequency and duration of the use of the above. They were shown to use floss, interdental brushes and disclosing tablets were provided to be used twice per week. The toothpaste and mouthwash studies supplied participants with toothbrushes and toothpaste to standardise this. As ΔR30 showed mature plaque, the time of tooth brushing might not have made a significant difference for this study.

9.0 CONCLUSION

This study assessed the use of QLF-D^{TM} images as visual aids for OHR in patients with poor oral hygiene before starting orthodontic treatment. Data were collected at three consecutive hygiene appointments, with OHR being provided at three-time points. The following conclusions were drawn:

1. The QLF-D^{TM} device was found to be a good evaluation tool to assess plaque accumulation and demineralisation. The QLF images allow quantitative data to be obtained on demineralisation and plaque coverage. Analysis of the QLF images has high levels of inter- and intra-examiner reliability.

2. While OHR using the QLF images as visual aids did not reduce the demineralisation over the mean 4 month period of assessment, there was a significant reduction in plaque noted both clinically and statistically. This was significant in both the QLF and WL groups. However, there was no significant difference between the two groups.

3. There was no clinical benefit of having OHR with the QLF images than with the WL images, although the patient perspective questionnaires indicated that the QLF images might be more useful. For both groups, the response of being shown the images was positive, and problem reported were from having photographs taken.

4. It is beneficial to use both WL and QLF images for OHR by hygienists and during routine orthodontic new patient and treatment clinics.

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10.0 CLINICAL RECOMMENDATIONS

This study demonstrates clinician led OHR using QLF-D ™ Biluminator as a visual aid is effective in patients with suboptimal oral hygiene. It may be worthwhile for these to be used as direct visual aids to provide regular personalised focused OHR in hygiene clinics. Clinician led OHR clinics could be used to educate patients on the commitment, motivation and compliance that is required during orthodontic treatment with regards to OH and diet to prevent demineralisation during orthodontic treatment. This would help in reducing patients who are not motivated to commit to undergo orthodontic treatment. Photographic records can regularly be taken in new patient clinics and hygienist clinics for OHR. Taking QLF images throughout treatment to monitor demineralisation lesions if present or to provide OHR to patients on a regular basis would provide information to clinician along with the changes in the occlusion, thus they should be readily available.

While there was no apparent clinical benefit of using QLF images over WL images as oral hygiene aids, the patient perspective suggested that the QLF images may be more useful. Our suggestion would be useful to use both QLF and WL images for OHR using QLF-D™ Biluminator. Using WL images for demineralisation as they are easily visible and QLF images for plaque makes QLF-D ™ device more advantageous to use instead of a conventional camera during orthodontics.

11.0 RESEARCH RECOMMENDATIONS

As a result of this study, the recommendations for future studies are:

1. It would be beneficial to conduct a similar study using the QLF and WL images taken with the QLF-D^{TM} Biluminator for OHR with minor modifications being made to the protocol . To have a patient group with suboptimal OH and routine orthodontic patients with optimal standard of OH and no demineralisation lesions at baseline. The suboptimal group to receive OHI with QLF images and optimal OH group to receive routine OHI . Then to assess the demineralisation post debond in the two groups.

- Conduct a sample size calculation based on the results of this study and thereby recruit a sufficient number of participants to ensure the study is adequately powered to detect a difference.
- Applying stratification of the sample for age and gender.
- Have the optimal OH group who would receive OHI alone, to allow comparisons to be made against the standard of oral hygiene care which is routinely provided.
- Extend the length of the study's duration to incorporate the full course of fixed orthodontic treatment, with the participants being assessed before commencing treatment and at debond.
- Assess pre-treatment demineralisation and enamel opacities and monitor these lesions longitudinally.

2. QLF-D^{TM} Biluminator could be used to assess improvements in demineralisation and plaque during RCTs with one group to receive OHI alone and intervention group to receive regular fluoride application along with OHR using QLF as visual aids. QLF images have a greater sensitivity in detecting demineralisation, which would allow a more detailed and precise analysis process and assess the difference in demineralisation in each group.

3. QLF-D^{TM} Biluminator could be used to assess improvements in demineralisation and plaque during RCTs with one group to receive OHI alone and intervention group to receive OHR using QLF as visual aids. QLF image taken to be printed out for patients to take home and maintain better OH in areas that require specific attention.

4. Further investigation, to understand the opinions of patients and parents regarding their motivation about orthodontic treatment and how much this impacts the oral hygiene care and

dietary habits and compliance with different appliances using expanded questionnaires or a framework approach. Stratifying participants based on their age and gender along with having to assess their motivation would show why some patients respond well to OH interventions. This would help in developing the best practice for OHR techniques.

12.0 REFERENCES

1. Weatherell JA. Composition of dental enamel. Br Med Bull. 1975;31(2):115–9.

2. Marsh PD. Dental plaque as a microbial biofilm. Caries Res. 2004;38(3):204–11.

3. Marsh PD, Head DA, Devine DA. Dental plaque as a biofilm and a microbial community - Implications for treatment. J Oral Biosci. 2015;57(4):185–91.

4. Pretty IA, Edgar WM, Smith PW, Higham SM. Quantification of dental plaque in the research environment. J Dent. 2005;33(3):193–207.

5. Donlan RM, Costerton JW. Biofilms: survival mechanisms of clinically relevant microorganisms. Clin Microbiol Rev. 2002;15(2):167–19.

6. van Houte J. Role of micro-organisms in caries etiology. J Dent Res. 1994;73(3):672–81.

7. Herschfeld J. W.D. Miller and the “chemico-parasitic” theory of dental caries. Bull Hist Den. 26(1):11–20.

8. Tanzer JM, Livingston J, Thompson AM. The microbiology of primary dental caries in humans. J Dent Educ. 2001;65(10):1028–37.

9. Takahashi N, Nyvad B. The role of bacteria in the caries process: ecological perspectives. J Dent Res. 2011;90(3):294–303.

10. Heinrich-Weltzien R, Kühnisch J, van der Veen M, de Josselin de Jong E. Quantitative light-induced fluorescence (QLF)— A potential method for the dental practitioner. 2003;34:181–8.

11. Van Der Veen MH, Thomas RZ, Huysmans MCDNJM, De Soet JJ. Red autofluorescence of dental plaque bacteria. Caries Res. 2006;40(6):542–5.

12. Marsh P, Martin M. Oral Microbiology fifth edition. Churhill and Livingstone; 2009.

13. Gorelick L, Geiger AM, Gwinnett A. Incidence of white spot formation after bonding and banding. Am J Orthod. 1982 Feb;81(2):93–8.

14. Øgaard B, Rølla G, Arends J. Orthodontic appliances and enamel demineralization. Part 1. Lesion development. Am J Orthod Dentofac Orthop. 1988;94(1):68–73.

15. Boersma JG, Van Der Veen MH, Lagerweij MD, Bokhout B, Prahl-Andersen B. Caries prevalence measured with QLF after treatment with fixed orthodontic appliances: Influencing factors. Caries Res. 2005;39(1):41–7.

16. Mattousch TJH, Van Der Veen MH, Zentner A. Caries lesions after orthodontic treatment followed by quantitative light-induced fluorescence: A 2-year follow-up. Eur J Orthod. 2007;29(3):294–8.

17. Ogaard B, Ten Bosch JJ. Regression of white spot enamel lesions. A new optical method for quantitative longitudinal evaluation in vivo. Am J Orthod Dentofacial Orthop. 1994;106(3):238–42.

18. Mizrahi E. Enamel demineralisation following orthodontic treatment. Am J Orthod. 1982;82(1):62–7.

19. Chang HS, Walsh LJ, Freer TJ. Enamel demineralization during orthodontic treatment. Aetiology and prevention. Aust Dent J. 1997;42(5):322–7.

20. Mitchell L. Decalcification during orthodontic treatment with fixed appliances-an overview. Br J Orthod. 1992;19(3):199–205.

21. Arends J, Christoffersen J. The nature of early caries lesions in enamel. J Dent Res. 1986;65(1):2–11.

22. Deyhle H, White S, Bunk O, Beckmann F, Muller B. Nanostructure of carious tooth enamel lesion. Acta Biomater. 2014;10(1):355–64.

23. Mizrahi E. Enamel opacities in primary and high school pupils. J Dent. 1982;10(1):28–38.

24. Mizrahi E, Cleaton-Jones PE. Enamel decalcification in primary school children. J Dent Assoc S Afr. 979;34:495–501.

25. Hurme VO. Developmental opacities of teeth in a New England community. Their relation to fluorine toxicosis. Am J Dis Child. 1949;77(1):61–75.

26. Murray J, Shaw L. Classification and prevalence of enamel opacities in the human deciduous and permanent dentitions. Arch Oral Biol. 1979;24(1):7–13.

27. Zachrisson S, Zachrisson B. Caries incidence and orthodontic treatment with fixed appliances. Scand J Dent Res J Dent Res. 1971;79:183–92.

28. Øgaard B. Prevalence of white spot lesions in 19-year-olds: A study on untreated and orthodontically treated persons 5 years after treatment. Am J Orthod Dentofac Orthop. 1989;96(5):423–7.

29. Zachrisson B. Direct bonding in orthodontics. Am J Orthod. 1977;71:173–89.

30. Stecksén-Blicks C, Renfors G, Oscarson ND, Bergstrand F, Twetman S. Caries-preventive effectiveness of a fluoride varnish: A randomized controlled trial in adolescents with fixed orthodontic appliances. Caries Res. 2007;41(6):455–9.

31. Lovrov S, Hertrich K, Hirschfelder U. Enamel demineralization during fixed orthodontic treatment - Incidence and correlation to various oral-hygiene parameters. J Orofac Orthop. 2007;68(5):353–63.

32. Willmot D. White Spot Lesions After Orthodontic Treatment. Semin Orthod. 2008;14(3):209–19.

33. Benson PE, Pender N, Higham SM. Quantifying enamel demineralization from teeth with orthodontic brackets — a comparison of two methods . Part 2 : validity. Eur J Orthod. 25:159–65.

34. Benson PE, Pender N, Higham SM. Quantifying enamel demineralization from teeth with orthodontic brackets — a comparison of two methods . Part 1 : repeatability and agreement. Eur J Orthod. 25:149–58.

35. Al Maaitah EF, Adeyemi AA, Higham SM, Pender N, Harrison JE. Factors affecting demineralization during orthodontic treatment: a post-hoc analysis of RCT recruits. Am J Orthod Dentofac Orthop. 2011;139(2):181–91.

36. Artun J, Brobakken B. Prevalence of carious white spots after orthodontic treatment with multibonded appliances. Eur J Orthod. 1986;8(4):229–34.

37. Lucchese A, Gherlone E. Prevalence of white-spot lesions before and during orthodontic treatment with fixed appliances. Eur J Orthod. 2013;35(5):664–8.

38. Shungin D, Olsson A, Persson M. Orthodontic treatment-related white spot lesions: a 14-year prospective quantitative follow-up, including bonding material assessment. Am J Orthod Dentofac Orthop. 2010;138(2):136.e1-8; discussion 136-7.

39. van der Veen MH, Mattousch T, Boersma JG. Longitudinal development of caries lesions after orthodontic treatment evaluated by quantitative light-induced fluorescence. Am J Orthod Dentofac Orthop. 2007;131(2):223–8.

40. O ’leary T, Drake R, Naylor J. The Plaque Control Record. J Periodontol. 1972;43(1):38.

41. Fischman S. Current status of indices of piaque. J Clin Periodontol. 1986;13:371–4.

42. Lim LP, Tay FBK, Waite IM, Cornick DER. A comparison of 4 techniques for clinical detection of early plaque formed during different dietary regimes. J Clin Periodontol. 1986;13(7):658–65.

43. Löe H. The Gingival Index, the Plaque Index and the Retention Index Systems. J Periodontol. 1967;38(6):610–6.

44. Ramfjord. S. P. Indices for prevalence and incidence of periodontal disease. J Periodontotogy. 1959;30:51–9.

45. Quigley, G A, Hein, J W. Comparative cleansing efficiency of manual and power brushing. J Am Dent Assoc. 1962;65:26–9.

46. Marks RG, Magnusson I, Taylor M, Clouser B, Maruniak J, Clark WB. Evaluation of reliability and reproducibility of dental indices. J Clin Periodontol. 1993;20(1):54–8.

47. Dababneh RH, Khouri AT, Smith RG, Addy M. Correlation and examiner agreement between a new method of plaque scoring and a popular established plaque index, modelled in vitro. J Clin Periodontol. 2002;29(12):1107–11.

48. Tureski S, Gilmore N, Glickman I. Reduced plaque formation by the chloromethyl analogue of victamine C. J Periodontol. 1970;41:41–3.

49. Grant D. A, Stern B, Everett FG. Periodontics. 5th editio. Mosby. SLC V., editor. 1979. 529 p.

50. Quirynen M, Dekeyser C, van Steenberghe D. Discriminating power of five plaque indices. J Periodontol. 1991;62(2):100–5.

51. Cugini M, Thompson M, PR W. Correlations Between Two Plaque Indices in assessment of toothbrush effectiveness. J Contemp Dent Pract. 2006;7(5):1–9.

52. Paschos E, Bücher K, Huth KC, Crispin A, Wichelhaus A, Dietel T. Is there a need for orthodontic plaque indices?-diagnostic accuracy of four plaque indices. Clin Oral Investig. 2014;18(4):1351–8.

53. Gallagher IHC, Fussell SJ, Cutress TW. Mechanism of Action of a Two-Tone Plaque Disclosing Agent. J Periodontol. 1977;July:395–6.

54. Block PL, Lobene RR, Derdivanis JP. A two-tone dye test for dental plaque. J Periodontol. 1972;43(7):423–6.

55. Lang N, ØStergaard E, Löe H. A Fluorescent Plaque Disclosing Agent. J Periodontal Res. 1972;59–67.

56. Söder P, Jin L, Söder B. Computerised planimetric method for clinical plaque measurement. Scand J Dent Res. 1993;(101):21–5.

57. Sagel P, Lapujade P, Miller J, Sunberg R. Objective quantification of plaque using digital image analysis. Monogr Oral Sci. 2000;17:130–43.

58. Pitts NB. Modern concepts of caries measurement. J Dent Res. 2004;83 Spec No:C43-47.

59. Gomez J, Tellez M, Pretty IA, Ellwood RP, Ismail AI. Non-cavitated carious lesions detection methods: A systematic review. Community Dent Oral Epidemiol. 2013;41(1):55–66.

60. Pereira AC, Eggertsson H, González-Cabezas C, Zero DT, Eckert GJ, Mialhe FL. Quantitative light-induced fluorescence (QLF) in relation to other technologies and conventional methods for detecting occlusal caries in permanent teeth. Brazilian J Oral Sci. 2011;10(1):27–32.

61. Willmot DR, Benson PE, Pender N, Brook AH. Reproducibility of quantitative measurement of white enamel demineralisation by image analysis. Caries Res. 2000;34(2):175–81.

62. Hill B, Geddes D. A photographic approach to recording pre-carious changes on teeth in situ. Med Biol Illus. 1975;25:79–88.

63. Edgar WM, Rugg-Gunn AJ, Jenkins GN GD. Photographic and direct visual recording of experimental caries-like changes in human dental enamel. Arch Oral Biol. 1978;23:667–73.

64. Ellwood R. Enamel Opacities and Dental Esthetics. J Public Health Dent. 1995;55(3):171–6.

65. Benson PE, Pender N, Higham SM, Edgar WM. Morphometric assessment of enamel demineralisation from photographs. J Dent. 1998 Nov;26(8):669–77.

66. Waller E, Daelen CJ van, Veen MH van der. Application of QLF™ for Diagnosis and Quality Assessment in Clinical Practice. 2012. p. 1–27.

67. Buchalla W. Comparative fluorescence spectros- copy shows differences in noncavitated enamel lesions. Caries Res. 2005;39:150–6.

68. Benedict HC. Notes on the fluorescence of teeth in ultra-violet rays. Science (80-). 1928;67(1739):442.

69. Alfano R, Yao S. Human teeth with and without dental caries studied by visible luminescent

spectroscopy. J Dent Res. 1981;60(2):120–2.

70. Bjelkhagen H, Sundström F, Ångmar-Månsson B, Rydén M. Early detection of enamel caries by the luminescence excited by visible laser light. Swed Dent J. 1982;6(1):1–7.

71. Sundström F, Fredriksson K, Montan S, Hafström-Björkman U, Strom J. Laser-induced fluorescence from sound and carious tooth sub- stance: spectroscopic studies. Swed Dent J. 1985;9(2):71–80.

72. Hafström-Björkman, U Sundström F, de Josselin de Jong E, Angmar-Månsson B. Comparison of laser fluorescence and longitudinal microradiography for quantitative assessment of in vitro enamel caries. Caries Res. 1992;26(4):241–7.

73. Emani Z, Al-Khateeb S, de Josselin de Jong, E Sundstrom F, Trollsas K, Angmar-Mansson B. Mineral loss in incipient caries lesions quantified with laser fluorescence and longitudinal microradiography. A methodologic study. Acta Odontol Scand. 1996;54(1):8–13.

74. Al- Khateeb S, Ten Cate JM, Angmar-Månsson B, de Josselin de Jong E, Sundström G, Exterkate RAM, et al. Quantification of formation and remineralisation of artificial enamel lesions with a new portable flourescence device. Adv Dent Res. 1997;11(4):502–6.

75. Konig K, Hibst R, Meyer H, Flemming G, Schneckenburger H. Laser-induced autofluorescence of carious regions of human teeth and caries-involved bacteria. Spie. 1993;2080:170–80.

76. Lennon Á M, Buchalla W, Brune L, Zimmermann O, Gross U, Attin T. The ability of selected oral microorganisms to emit red fluorescence. Caries Res. 2005;40(1):2–5.

77. Coulthwaite L, Pretty IA, Smith PW, Higham SM, Verran J. QLF is not readily suitable for in vivo denture plaque assessment. J Dent. 2009;37(11):898–901.

78. Angmar-Månsson B, Ten Bosch JJ. Quantitative light-induced fluorescence (QLF): A method for assessment of incipient caries lesions. Dentomaxillofacial Radiol. 2001;30(6):298–307.

79. AI-Khateeb S, Forsberg C-M, de Josselin de Jong E, Angmar-Månsson B. A longitudinal laser fluorescence study of white spot lesions in orthodontic patients. Am J Orthod Dentofac Orthop. 1998;113(6):595–602.

80. Pretty IA, Smith PW, Edgar WM, Higham SM. Detection of in vitro demineralization adjacent to restorations using quantitative light induced fluorescence (QLF). Dent Mater. 2003;19(5):368–74.

81. Lennon A, Buchalla W, Switalski L, Stookey GK. Residual caries detection using visible fluorescence. Caries Res. 2002;36(5):315–9.

82. Gomez GF, Eckert GJ, Zandona AF. Orange/Red fluorescence of active caries by retrospective quantitative light-induced fluorescence image analysis. Caries Res. 2016;50(3):295–302.

83. Buchalla W, T A, Y N, PD N, AM L. Porphyrins are the cause of red fluorescence of carious dentine: verified by gradient reversed-phase HPLC. Caries Res. 2008;42:223.

84. Izu M, Miyake T, Uene M, Tanaka H, Takashima R, Nishijima N, et al. New analysis method of gingiva with quantitative light-induced fluorescence. J Dent Res. 2005;84:1069.

85. Pretty IA, Edgar WM, Higham SM. A review of the effectiveness of quantitative light induced fluorescence (QLF) to detect early caries. Proceedings of the 6th Indiana Conference. 2005. p. 253–91.

86. Al Maaitah E, Adeyemi A, Higham SM, Pender N, Harrison JE. Factors affecting demineralization during orthodontic treatment: A post-hoc analysis of RCT recruits. Am J Orthod Dentofac Orthop. 2011;139(2):181–91.

87. Pretty IA, Hall AF, Smith PW, Edgar WM, Higham SM. The intra- and inter-examiner reliability of quantitative light-induced fluorescence (QLF) analyses. Br Dent J. 2002;193:105–9.

88. Veen van der MH, e Josselin de Jong E, Faller F V. Application of quantitative light-induced fluorescence for assessing early caries lesions. Monogr Oral Sci. Karger; 2000;7:144–62.

89. Ferreira Zandoná A, Ando M, Eggertsson H. Clinical validation of caries detection methodologies: preliminary results. J Den Res. 2004;83(special issue A).

90. Thomas E. Assessment of three methods to detect plaque and white spot lesions [M Dent Sci Thesis]. University of Liverpool; 2010.

91. van Daelen C, Smith P, de Josselin de Jong E, Higham SM, MH van der V. A simple blue light to visualise caries by means of fluorescence. Caries Res. 2008;42:223.

92. Lussi a, Hibst R, Paulus R. DIAGNOdent: an optical method for caries detection. J Dent Res. 2004;83 Spec No:C80–3.

93. Sheehy E, Brailsford S, Kidd E, Beighton D, Zoitopoulos L. Comparison between visual examination and a laser fluorescence system for in vivo diagnosis of occlusal caries. Caries Res. 2001;35(6):421–6.

94. Bamzahim M, Shi X, Ångmar-Månsson B. Occlusal caries detection and quantification by DIAGNOdent and electronic caries monitor: in vitro comparison. Acta Odontol Scand. 2002;60(6):360–4.

95. Lussi A, Imwinkelried S, Pitts N, Longbottom C, Reich E. Performance and reproducibility of a laser fluorescence system for detection of occlusal caries in vitro. Caries Res. 1999;33(4):261–6.

96. Pinelli C, Campos SM, de Castro Monteiro Loffredo L. Validity and reproducibility of a laser fluorescence system for detecting the activity of white-spot lesions on free smooth surfaces in vivo. Caries Res. 2002;36(1):19–24.

97. Ten Bosch JJ, Angmar-Månsson B. A review of quantitative methods for studies of mineral content of intra-oral caries lesions. J Dent Res. 1991;70(1):2–14.

98. Ten Bosch JJ. Summary of research of Quantitative light induced fluorescence. Early detection of dental caries II. Proceedings of the 4th Annual Conference, Indianapolis, Indiana University. 2000.

99. Alammari MR, Smith PW, Jong E de J de, Higham SM. Quantitative light-induced fluorescence (QLF): A tool for early occlusal dental caries detection and supporting decision making in vivo. J Dent. 2012;41(2):127–32.

100. Browne RH. On the use of a pilot sample for sample size determination. Stat Med. 1995;14(17):1933–40.

101. Lancaster GA, Dodd S, Williamson PR. Design and analysis of pilot studies:recommendations for good practice. J Eval Clin Pract. 2004;307–12.

102. Harrison JE, Burnside G. Why does clustering matter in orthodontic trials? Eur J Orthod. 2012;34(3):293–5.

103. Bewick V, Cheek L, Ball J. Statistics review 13: Receiver operating characteristic curves. Crit Care. 2004;8(6):508–12.

104. Gray D, McIntyre G. Does oral health promotion influence the oral hygiene and gingival health of patients undergoing fixed appliance orthodontic treatment? A systematic literature review. J Orthod [Internet]. 2008;35(4):262–9. Available from: http://www.ncbi.nlm.nih.gov/pubmed/19074364

105. Habbu SG, Krishnappa P. Effectiveness of oral health education in children - A systematic review of current evidence (2005-2011). Int Dent J. 2015;65(2):57–64.

106. Kay E, Locker D. Is dental health education effective ? A systematic review of current evidence. Community Dent Oral Epidemiol. 1996;Aug 24(4):231–5.

107. Hobson RS, Clark JD. How UK orthodontists advise patients on oral hygiene. Br J Orthod. 1998;25(1):64–6.

108. Marini I, Bortolotti F, Parenti SI, Gatto MR, Bonetti GA. Combined effects of repeated oral hygiene motivation and type of toothbrush on orthodontic patients: a blind randomized clinical trial. Angle Orthod. 2014;84(5):896–901.

109. Khalaf K. Factors affecting the formation, severity and location of white spot lesions during orthodontic treatment with fixed appliances. J oral Maxillofac Res. 2014;5(1):e4.

110. Tolvanen M, Lahti S PR. Changes in children's oral health-related behavior, kowledge and attitudes during a 3.4-yr randomized clinical trial and oral health-promotion program. Eur J Oral Sci. 2009;117:390–2.

111. de Farias IA, Souza CG FM. A health education program for Brazilian public schoolchildren: the effects on dental health practice and oral health awareness. J Public Heal Dent. 2009;225–230.

112. Peng Y, Wu R, Qu W, Wu W, Chen J, Fang J, et al. Effect of visual method vs plaque disclosure in enhancing oral hygiene in adolescents and young adults: A single-blind randomized controlled trial. Am J Orthod Dentofac Orthop. American Association of Orthodontists; 2014;145(3):280–6.

113. Ay ZY, Sayin MÖ, Özat Y, Goster T, Atilla AO, Bozkurt FY. Appropriate oral hygiene motivation method for patients with fixed appliances. Angle Orthod. 2007;77(6):1085–9.

114. Boyd RL. Longitudinal evaluation of a system for self-monitoring plaque control effectiveness in orthodontic patients. J Clin Periodontol. 1983;10:380–8.

115. Acharya S, Goyal A, Utreja AK, Mohanty U. Effect of three different motivational techniques on oral hygiene and gingival health of patients undergoing multibracketed orthodontics. Angle Orthod. 2011;81(5):884–8.

116. Miller CC, Burnside G, Higham SM, Flannigan NL. Quantitative Light-induced Fluorescence-

Digital as an oral hygiene evaluation tool to assess plaque accumulation and enamel demineralization in orthodontics. Angle Orthod. 2016;86(6):991–7.

117. Berlin-Broner Y. Awareness of orthodontists regarding oral hygiene performance during active orthodontic treatment. Eur J Paediatr Dent. 2012;13(3):187–91.

118. Zotti F, Dalessandri D, Salgarello S, Piancino M, Bonetti S, Visconti L, et al. Usefulness of an app in improving oral hygiene compliance in adolescent orthodontic patients. Angle Orthod. 2016;86(1):101–7.

119. Eppright M, Shroff B, Best AM, Barcoma E, Lindauer SJ. Influence of active reminders on oral hygiene compliance in orthodontic patients. Angle Orthod. 2014;84(2):208–13.

120. Huang G, Roloff-Chiang B, Mills B, Shalchi S, Spiekerman C, Korpak AM, et al. Effectiveness of MI Paste Plus and PreviDent fluoride varnish for treatment of white spot lesions: A randomized controlled trial. Am J Orthod Dentofac Orthop. 2013;143(1):31–41.

121. Sonesson M, Twetman S, Bondemark L. Effectiveness of high-fluoride toothpaste on enamel demineralization during orthodontic treatment - A multicenter randomized controlled trial. Eur J Orthod. 2014;36(6):678–82.

122. Bailey DL, Adams GG, Tsao CE, Hyslop a, Escobar K, Manton DJ, et al. Regression of post-orthodontic lesions by a remineralizing cream. J Dent Res. 2009;88(12):1148–53.

123. Andersson A, Anders S, Petersson LG. Calcium phosphate complexes on white spot lesion regression assessed by laser fluorescence. Oral Heal Prev Dent. 2007;5(3):229–34.

124. Benson P. Evaluation of white spot lesions on teeth with orthodontic brackets. Semin Orthod. 2008;14(3):200–8.

3.0 APPENDICES

13.1 Information sheet for under 16s

INFORMATION SHEET FOR UNDER 16s

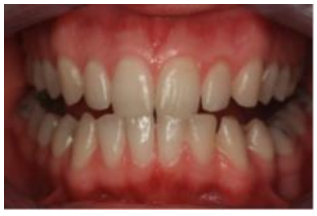
The use of QLF-D in orthodontics

We want to tell you about a research study we are doing. A research study is a special way to find out about something. We would like you to join this study that looks at a new way which shows if food is stuck on your teeth or your teeth have any damage. Before you decide, please read this information sheet. Please ask us if there is anything that is not clear or you have any questions.

Quantitative Light Induced Fluorescence digital (QLFD™) is a camera which records a picture of your teeth. This camera takes a normal photograph and a blue light photograph of the teeth. It will help us monitor your the teeth and see how clean they are or if there is any damage. You will be given tooth brushing advice and shown either the normal or blue light photographs. These photographs will show the areas of food stuck on your teeth and help you know where to brush.

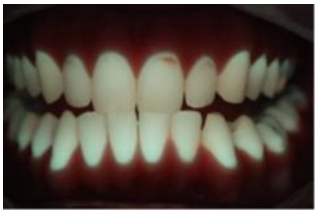
The study will not change your treatment. It will only make your appointments a few minutes longer.

What is the purpose of the study?



We are using a Quantitative Light Induced Fluorescence digital (QLFD™) camera to take normal and blue light photographs of your teeth. We are trying to find the areas where food has not been cleaned away, or there is damage to your teeth.

We are also trying to find out if showing you the camera photographs is useful for your tooth brushing.



Why have I been asked to take part?

You are the right age and are having hygienist visits to improve your brushing.

What will happen if I say yes?

At every visit, we will take photographs of your teeth and will clean them if required. We will show you either the normal or blue light photographs and give you tooth brushing advice on the areas that need better cleaning.

How long is the project?

It will last for three of your normal hygienist appointments.

What if I am not happy or have a problem?
You can stop taking part in this project at any time. Your hygienist visits will continue as normal.

What if I have a question?
If you have any questions, feel free to ask, and I will be happy to answer them.
Thank you for taking the time to read this.

13.2 Information sheet for adults

INFORMATION SHEET FOR THE PARTICIPANT

The use of QLFD in orthodontics

You are being asked to participate in a research study which is looking at a new way to assess the level of cleanliness of your teeth and any damage present. Before you decide to take part in the study, please take time to read this information sheet. Please ask us if there is anything that is unclear if you have any questions or would like further information.

Quantitative Light Induced Fluorescence digital (QLFD™) is a digital camera which can record images of teeth. It takes a normal photograph and a blue light photograph of the teeth. The blue light enables plaque debris to be seen as fluorescent areas on teeth. It is also able to show enamel damage, which can leave permanent marks on teeth, at an earlier stage than eye sight alone. This camera will help us monitor the health of the teeth and assess the cleanliness and damage to the teeth more accurately.

The investigation will not involve any alteration to the hygienist appointments apart from a slight extension of the appointment times.

What is the purpose of the study?
We are investigating a method used to assess the level of cleanliness of your teeth and any damage present. We will study the cleanliness of teeth using a digital camera under blue light conditions, which is a recognised technology, called Quantitative Light Induced Fluorescence (QLF), and has been used in many previous clinical trials as well as in dental practice. We will either show you the white light or blue light images taken with the QLFD camera and give tooth brushing advice to determine if seeing these images helps reduce plaque debris build up and reduce any enamel damage.

Has the study been approved?
The project was reviewed by NRES Committee North West- Liverpool Central.

Who is paying for the study?
The University of Liverpool is providing funds for this study.

Who will be conducting the study?
The study is being run by Prof. Susan Higham (Professor of Oral Biology), Dr Norah Flannigan (Senior Clinical Lecturer in Orthodontics) and Puthri Raphy (Specialist Registrar in Orthodontics).

Why have I been asked to take part?
You have been asked because we are looking for healthy volunteers who are currently having hygienist appointments before they are added on the waiting list to start orthodontic treatment.

What will happen if I take part?
The QLFD camera will be used to take normal photographs and blue light photographs of the teeth. This will be repeated at three consecutive appointments. Your teeth will also be given a polish to remove any plaque deposits present if required. This will lengthen the appointment time by no more than 15 minutes. You will then be shown either the normal or blue light images of your teeth on a screen and given tooth brushing instruction.

How long will the study last?
You will be monitored for three consecutive appointments with the hygienist

What if I do not want to take part?
Your treatment will continue as normal. You should not feel obliged to take part, and you do not have to give a reason if you do not want to. If you do take part in the study, but later decide that you do not want to continue, you can also withdraw at any time without giving a reason.

What if I have a question of there is a problem on the trial?
You may ask questions at any time, before and during the study. If you wish to make any enquiry subsequently, you may contact, Puthri Raphy, Orthodontic Department, Liverpool University Dental Hospital, Pembroke Place, Liverpool, L3 5PS. Email: puthri.raphy@liverpool.ac.uk

How will the data collected be managed?
All information about you will be processed and analysed by the research staff involved in the study. Data will be stored for ten years. As soon as we have collected the necessary data all information, which identifies you, will be removed and replaced by a code. The person responsible for security and access to your data is Dr Flannigan, the Chief investigator of the Study.

What do I do if I want to take part?
If you would like to take part, please sign all the relevant sections of the consent form that you will have been provided with.

Thank you for taking the time to read this.

13.3 Information for parents

INFORMATION SHEET FOR THE PARENT

The use of QLFD in orthodontics

Children are being asked to participate in a research study which is looking at a new way to assess the level of cleanliness of teeth. Before deciding to take part in the study, please take time to read this information sheet. Please ask us if there is anything that is unclear if you have any questions or would like further information.

Quantitative Light Induced Fluorescence digital (QLFD™) is a digital camera which can record images of teeth. It takes a normal photograph and a blue light photograph. The blue light enables plaque debris to be seen as fluorescent areas on teeth. It is also able to show enamel damage, which can leave permanent marks on teeth, at an earlier stage than eye sight alone. The child will either be shown the normal or blue light photographs and be given tooth brushing advice based on these images. This will help us monitor the health of the teeth and assess the cleanliness and damage to the teeth more accurately.

The investigation will not involve any alteration to the hygienist appointments apart from a slight extension of the appointment times.

What is the purpose of the study?
We are investigating a method used to assess the level of cleanliness of teeth and any damage present. We will study the cleanliness of teeth using a digital camera under blue light conditions, which is a recognised technology, called Quantitative Light Induced Fluorescence (QLF), and has been used in many previous clinical trials as well as in dental practice. We will show the child either the white light or blue light images were taken with the QLFD camera and give tooth brushing advice to determine if seeing these images helps reduce plaque debris build up and reduce any enamel damage.

Has the study been approved?
The project was reviewed by NRES Committee North West- Liverpool Central.

Who is paying for the study?
The University of Liverpool is providing funds for this study.

Who will be conducting the study?
The study is being run by Prof Susan Higham (Professor of Oral Biology), Dr Norah Flannigan (Senior Clinical Lecturer in Orthodontics) and Puthri Raphy (Specialist Registrar in Orthodontics).

Why has my child been asked to take part?
We are looking for healthy volunteers who are currently having hygienist appointments before they are added on the waiting list to start orthodontic treatment.

What will happen if my child takes part?
The QLFD camera will be used to take normal photographs and blue light photographs of the teeth. This will be repeated at three consecutive appointments. The child's teeth will also be given a clean if required. This will lengthen the appointment time by no more than 15 minutes. The child will be shown either the normal or blue light images of your teeth on a screen and given tooth brushing instruction.

How long will the study last?
Your child will be monitored for three consecutive appointments with the hygienist.

What if I do not want my child to take part?
Your child's treatment will continue as normal. You should not feel obliged to consent to take part in this study and do not have to give a reason if you do not want to. If you do consent but later decide that you do not want to continue, you can also withdraw at any time without giving a reason.

What if I have a question of there is a problem on the trial?
You may ask questions at any time, before and during the study. If you wish to make any enquiry subsequently, you may contact, Puthri Raphy, Orthodontic Department, Liverpool University Dental Hospital, Pembroke Place, Liverpool, L3 5PS. Email: puthri.raphy@liverpool.ac.uk

How will the data collected be managed?
All information will be processed and analysed by the research staff involved in the study. Data will be stored for ten years. As soon as we have collected the necessary data, all information, which identifies you, will be removed and replaced by a code. The person responsible for security and access to your data is Dr Flannigan, the Chief investigator of the Study.

What do I do if I am happy for my child to take part?
If you are happy for your child to take part, please sign all the relevant sections of the consent form that you will have been provided with.

Thank you for taking the time to read this.

1 3.4 Consent form for under 16s

Patient Identification Number for this trial:

ASSENT FORM FOR UNDER 16s

Research project: The use of QLFD as an oral hygiene evaluation tool in orthodontics

Researcher: Puthri Raphy

Please circle YES or NO

1. I have read the information sheet dated 06/04/2015(V1.2). YES NO
2. The project has been explained to me. YES NO
3. I have been able to ask questions and have had these answered. YES NO
4. I understand what the project is about and what I need to do. YES NO
5. I understand that I can stop taking part at any time. YES NO
6. I am happy to take part in the project. YES NO

_____	_____	_____
Name of patient	Date	Signature

_____	_____	_____
Name of Person taking assent	Date	Signature

13.5 Consent form the adults

Patient Identification Number for this trial:

CONSENT FORM 1
Patient's agreement for participation in research

Research project: The use of QLFD as an oral hygiene evaluation tool in orthodontics.

Researcher: Puthri Raphy

Please initial box

1. I confirm that I have read and understand the information sheet dated 06/04/2015 (Version 1.2) for the above study. I have had the opportunity to consider the information, ask questions and have had these answered satisfactorily. ☐

2. I understand that my participation is voluntary and that I am free to withdraw at any time without giving any reason, without my medical care or legal rights being affected. ☐

3. I understand that the data collected during the study will be analysed by the study investigators and that relevant sections of data may be looked at by individuals from regulatory authorities, where it is relevant to my taking part in this research. I give permission for these individuals to have access to my records. ☐

4. I agree to take part in the above study. ☐

_____	_____	_____
Name of Volunteer	Date	Signature

_____	_____	_____
Name of Person taking consent	Date	Signature

13.6 Consent form for parents

Patient Identification Number for this trial:

CONSENT FORM 2
Parental agreement for participation in research

Research project: The use of QLFD as an oral hygiene evaluation tool in orthodontics.

Researcher: Puthri Raphy

Please initial box

1. I confirm that I have read and understand the information sheet dated 06/04/2015 (Version 1.2) for the above study. I have had the opportunity to consider the information, ask questions and have had these answered satisfactorily. ☐

2. I understand that my child's participation is voluntary and that I am free to withdraw them at any time without giving any reason, without my medical care or legal rights being affected. ☐

3. I understand that the data collected during the study will be analysed by the study investigators and that relevant sections of data may be looked at by individuals from regulatory authorities, where it is relevant to my taking part in this research. I give permission for these individuals to have access to my child's records. ☐

4. I agree to my child taking part in the above study. ☐

_____	_____	_____
Parent / Guardian	Date	Signature
_____	_____	_____
Name of Person taking consent	Date	Signature

13.7 Debriefing form

Patient Identification Number for this trial:

DEBRIEFING FORM

Research project: The use of QLFD as an oral hygiene evaluation tool in orthodontics

Researcher: Puthri Raphy

Please circle answer

1. Which photographs were you shown?

NORMAL PHOTO BLUE LIGHT PHOTO NOT SURE

2. Did you have any problems with the additional photographs that were taken?

YES NO NOT SURE

3. Were you able to see the areas of food accumulation on the photographs?

YES NO NOT SURE

4. Were you able to see the areas of tooth damage on the photographs?

YES NO NOT SURE

5. Did you find it helpful to be shown the photographs?

YES NO NOT SURE

6. Do you think it would be useful to have the photographs taken the whole way through treatment?

YES NO NOT SURE

7. Do you think your tooth brushing improved over the appointments?

YES NO NOT SURE

_____ _____ _____

Name of patient Date Signature

13.8 Reliability test for plaque accumulation

Image Number	∆R30
QLF Image 1	
QLF Image 2	
QLF Image 3	
QLF Image 4	
QLF Image 5	
QLF Image 6	
QLF Image 7	
QLF Image 8	
QLF Image 9	
QLF Image 10	
QLF Image 11	
QLF Image 12	
QLF Image 13	
QLF Image 14	
QLF Image 15	
QLF Image 16	
QLF Image 17	
QLF Image 18	

QLF Image 19	
QLF Image 20	
QLF Image 21	
QLF Image 22	
QLF Image 23	
QLF Image 24	
QLF Image 25	
QLF Image 26	
QLF Image 27	
QLF Image 28	
QLF Image 29	
QLF Image 30	
QLF Image 31	
QLF Image 32	
QLF Image 33	
QLF Image 34	
QLF Image 35	

13.9 Reliability test for demineralisation

Image Number	ΔF
QLF Image 2	UR2 UR1 UL1 UL2
QLF Image 4	UL2 UL6
QLF Image 6	LR6 LR5 LL5 LL6
QLF Image 8	UR2 UR6
QLF Image 10	UR4 UR2 UR1 UL1 UL2 UL4
QLF Image 12	UR2 UR1 UL1 UL2
QLF Image 14	UL3 LL6
QLF Image 16	UR5 UR4 UL4 UL5
QLF Image 18	UR2 UR1 UL1 UL2
QLF Image 20	UL4 UL6
QLF Image 22	UR3 UR4 UR6 UR7
QLF Image 24	UR5 UR1
QLF Image 26	LR6
QLF Image 28	UR1 UL1

	UL2
QLF Image 30	UR6 LR6
QLF Image 32	LR6 LR5 LL5 LL6
QLF Image 34	UR4 UR6 UL4 UL6
QLF Image 36	UL6 LL6
QLF Image 38	LL6
QLF Image 40	UR2 UR1 UL1 UL2
QLF Image 42	LR6
QLF Image 44	LR6 LL6
QLF Image 46	LR6 LR5 LL6
QLF Image 48	UR2 UR1 UL1 UL2
QLF Image 50	UR6 LR6
QLF Image 52	UL4 LL6
QLF Image 54	UR6 UL6
QLF Image 56	LR6 LL6
QLF Image 58	UR6 LR6
QLF Image 60	UR1 UL1

QLF Image 62	LR6 LL6
QLF Image 64	UR2 UR1 UL1 UL2
QLF Image 66	UL3
QLF Image 68	UR6 UL6
QLF Image 70	UR3 UR1 UL1 UL3
QLF Image 72	LR6

13.10 Organisation of Caries Research Conference- poster presentation

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QLF-D as oral hygiene evaluation tool to assess plaque and demineralisation in Orthodontics

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Background

- Good oral hygiene is essential in minimising plaque during orthodontic treatment to prevent periodontal diseases and caries.[1]
- Demineralisation during orthodontic treatment affects between 2-96% of patients.[2]
- Plaque control is made more difficult by the presence of orthodontic appliances and demineralisation may develop within 4 weeks of appliance placement.[3]
- The Quantitative Light- Induced Fluorescence-Digital (QLF-D) device, allows White Light (WL) and Quantitative Light-Induced Fluorescence (QLF) images to be taken.
- On the QLF images, plaque is seen as red due to auto fluorescence of bacterial porphyrins (Figure 1) and demineralisation is seen as darker area due to the reduced fluorescence (Figure 2).

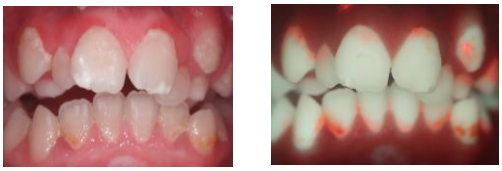

Figure 1:WL and QLF images taken for plaque assessment

Figure 2 :WL and QLF images taken for demineralisation assessment

Aim

- To assess the use of Quantitative Light-induced Fluorescence-Digital Biluminator TM (QLF-D™) to detect plaque coverage and demineralisation in patients with poor oral hygiene prior to start of orthodontics.

Material and Methods

- 1:1 parallel arm randomised clinical trial was conducted at Liverpool University Dental Hospital.
- 60 patients with poor oral hygiene who required oral hygiene reinforcement and prophylaxis before they are added on the waiting list to have fixed orthodontic appliance treatment were randomly allocated to receive oral hygiene reinforcement (OHR) at three consecutive appointments using white light (WL) or QLF-D images taken with the QLF-D device as visual aids.
- For both groups, plaque coverage, ΔR30 and change in demineralisation, measured by the degree of fluorescence loss, ΔF, were assessed on QLF-D images from the baseline to the final appointment.
- A questionnaire was used to ascertain the patients' perspectives of the images being used as oral hygiene aids.

Results

- There were no significant differences in plaque accumulation (P=0.81) or demineralisation (P=0.69) between the WL and QLF-D groups.
- There was no significant change in demineralisation over the three visits in either group, however there was a significant reduction in plaque in both groups (P<0.001) with a mean percentage change in ΔR30 of 51.8% and 95% CI of 40.36% to 63.26%.
- 92.5% of the QLF-D group and 76.7% of the WL group expressed it would be useful to be given such OHR for the full duration of orthodontic treatment.

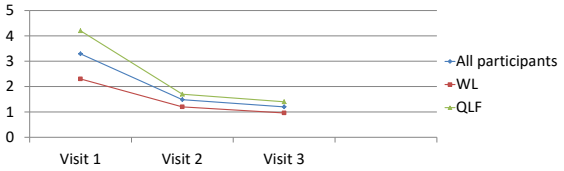

Figure 3: Reduction in ΔR30 level for the participants over the course of the study

Discussion

- There was no statistical significance in terms of reducing levels of plaque or demineralisation using QLF images over WL images, a greater number of patients allocated to the QLF group felt it would be useful to have this OHR for the whole duration of treatment, suggesting that QLF images may be more useful.
- Main limitation of the study was that it was not possible to blind the treating clinician, who provided the OHR and undertook the image analysis. The images were randomised to reduce bias.

Conclusions

- OHR using WL or QLF-D images as visual aids was effective in reducing plaque coverage.
- There was no difference in the level of demineralisation or plaque coverage between the QLF-D and WL groups.
- More patients reported that the QLF-D images were useful than patients shown WL images.

References

1. *Gray D and McIntyre G 2008. Does oral health promotion influence the oral hygiene and gingival health of patients undergoing fixed appliance orthodontic treatment? A systemic literature review. Journal of Orthodontics 35(4): 262-269.*
2. *Gorelick L, Geiger AM, Gwinnett AJ 1982. Incidence of white spot formation after bonding and banding. American Journal of Orthodontics 81(2): 93-98.*
3. *Ogaard B, ten Bosch JJ 1994. Regression of white spot enamel lesions. A new optical method for quantitative longitudinal evaluation in vivo. American Journal of Orthodontics and Dentofacial Orthopedics 106(3): 238-242.*

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13.11 British Orthodontic Conference- oral presentation abstract

Use of QLF-D as oral hygiene evaluation tool in Orthodontics.

Raphy P*, Burnside G, Higham.S.M, Flannigan N.L.
Liverpool University Dental Hospital.

Objective: To assess the use of Quantitative Light-induced Fluorescence (QLF-D) to detect plaque coverage and demineralisation.

Design and Setting: A prospective RCT was conducted at Liverpool University Dental Hospital.

Materials and Methods: 60 patients with poor oral hygiene prior to start of orthodontic treatment were randomly allocated to receiving oral hygiene reinforcement (OHR) at three consecutive appointments using white light (WL) or QLF-D images as visual aids. For both groups, plaque coverage, ΔR30 and change in demineralisation, measured by the degree of fluorescence loss, ΔF, were assessed on QLF-D images from baseline to the final appointment. A questionnaire was used to ascertain the patients' perspectives of the images being used as oral hygiene aids.

Results: There were no significant differences in plaque accumulation (P=0.81) or demineralisation (P=0.69) between the WL and QLF-D groups. There was a significant reduction in plaque in both groups (P<0.001) with a mean percentage change in □R30 of 51.8%. 92.5% of the QLF-D group and 76.7% of the WL group expressed it would be useful to be given such OHR for the full duration of orthodontic treatment.

Conclusions: OHR using WL or QLF-D images as visual aids was effective in reducing plaque coverage. There was no difference in the level of demineralisation or plaque coverage between the QLF-D and WL groups. More patients in QLF-D group found images useful than WL group.