

The cleaning of photographic retractors; a survey, clinical and laboratory study

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IN BRIEF

- This research is the first work to examine the effectiveness of various methods of cleaning photographic retractors.
- The technique described is very sensitive and can be applied to a number of different situations.
- It clearly demonstrates that washer-disinfectors are the first method of choice.
- No technique was 100% effective at removing all protein.

Objectives To determine the methods currently being used to decontaminate photographic retractors in specialist orthodontic practice and to investigate the effectiveness of the cleaning methods. **Design** The study was carried out in two parts: I – a postal self-report questionnaire, and II – a cross-sectional clinical and laboratory investigation. **Setting** The Orthodontic Department of the Charles Clifford Dental Hospital. **Subjects and materials** I – The questionnaire was sent to 278 specialist UK orthodontists. II – One hundred and twenty pairs of photographic retractors were collected following use. One retractor from each pair was randomly chosen to be the unwashed control and immediately placed in 20 ml of PBS-Tween for elution. The other was subjected to the one of four cleaning procedures: alcohol wipe, handwashing, ultrasonic bath or washer-disinfector, before being placed in PBS-Tween. Aliquots were taken for assay. **Main outcome measures** Antibody capture (ELISA) for amylase, to detect the presence of saliva, and for albumin, to detect the presence of serum. **Results** I – The questionnaire response rate was 65% and the majority of respondents (87.2%) were routinely taking clinical photographs. A wide variety of techniques were being used to decontaminate photographic retractors. II – All unwashed controls had detectable levels of amylase and albumin. All the retractors that were cleaned using an alcohol wipe had residual detectable levels of amylase and 80% had detectable levels of albumin. Only one retractor had detectable amylase and one had detectable albumin following cleaning using the washer-disinfector. There was a highly significant statistical difference between the techniques in the proportional reduction in both amylase and albumin detected from the unwashed control and cleaned experimental retractors ($p < 0.001$). The infective risk from inadequate cleaning of photographic retractors is discussed. **Conclusions** The washer-disinfector is the most effective method of cleaning photographic retractors, but no method was found to be 100% successful at removing amylase and albumin.

INTRODUCTION

Photography has become an important part of clinical practice.¹ In addition to being a valuable clinical record, photographic images have been used to communicate shade,² screen children for dental disease³ and identify appropriate orthodontic new patient referrals.⁴

Photographic retractors are used, when taking a clinical image, to keep cheeks and lips out of the way in order to obtain an adequate view of the teeth. Consequently,

these retractors will come into contact with saliva and possibly blood. It is widely recognised that bodily fluids and other contamination must be removed from dental instruments before sterilisation, otherwise blood borne pathogens might be protected from the sterilisation process.⁵ The UK Department of Health has recently provided guidance about cleaning in primary dental care.⁶ Recommendations for this initial cleaning include washing the instruments by hand, using an ultrasonic bath or an automated instrument washer. A recent observational study carried out in 179 general dental practices in Scotland found that the majority of practices were using a manual cleaning method for dental instruments with or without ultrasonic cleaning, but that this was poorly controlled and likely to increase the risk of cross-infection. None of the practices was using a washer-disinfector.

Various techniques have been developed to assess the effectiveness of cleaning procedures. These include visual inspection,⁷ culturing and examining the growth of bacteria^{5,8} and detecting the presence of blood contamination.^{9–11} Although dentistry is considered a low risk for transmission of the disease, the emergence of variant Creutzfeldt-Jakob disease (vCJD) has emphasised the need to find techniques to establish that adequate cleaning and sterilisation of all instruments that are used and reused on patients have been carried out.¹² The infective agent for vCJD is a prion protein that is resistant to normal sterilisation techniques.¹³ Methods have therefore been developed for detecting and determining the amount of protein contamination left on instruments after cleaning.^{14,15}

Few studies have been carried out to examine the extent of organic contamination of photographic retractors after use or

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of ways to effectively decontaminate them between patients. Some work performed on surgical retractors¹⁴ found that five out of thirteen retractors from five different UK hospitals, which were cleaned, autoclaved and ready to be returned to the operating theatre, showed significant levels of protein contamination.

The aim of this study was to determine the current methods used in specialist orthodontic practice to decontaminate photographic retractors and to assess the effectiveness of the cleaning methods using a quantitative antibody capture assay. The specific research questions were:

- What is the range of methods used to decontaminate photographic retractors in specialist orthodontic practice?
- What level of contamination with saliva and blood occurs when retractors are used?
- Are current methods of cleaning these retractors sufficient to remove the contamination?

SUBJECTS AND METHODS

The investigation was carried out in two parts:

1. A cross-sectional, postal, self-report questionnaire of specialist orthodontic practitioners in the UK to determine the methods used to decontaminate photographic retractors
2. A cross-sectional clinical and laboratory study to assess the effectiveness of the reported methods used to clean photographic retractors.

South Sheffield Research Ethics Committee was approached for advice regarding the project and confirmed that formal ethical approval was not required.

Postal questionnaire

The methodology followed a recent survey into the reuse, cleaning and sterilisation of orthodontic bands.¹⁶ A pilot study was carried out amongst ten specialist orthodontists at the Charles Clifford Dental Hospital, Sheffield to show the level of response and acceptability of the questionnaire. Constructive suggestions led to the questionnaire being modified.

The questionnaire was sent by post to 278 orthodontists on the Specialist Orthodontist list held by the General Dental Council in the UK. Participants were chosen using a



Fig. 1 Plastic photographic retractors in use

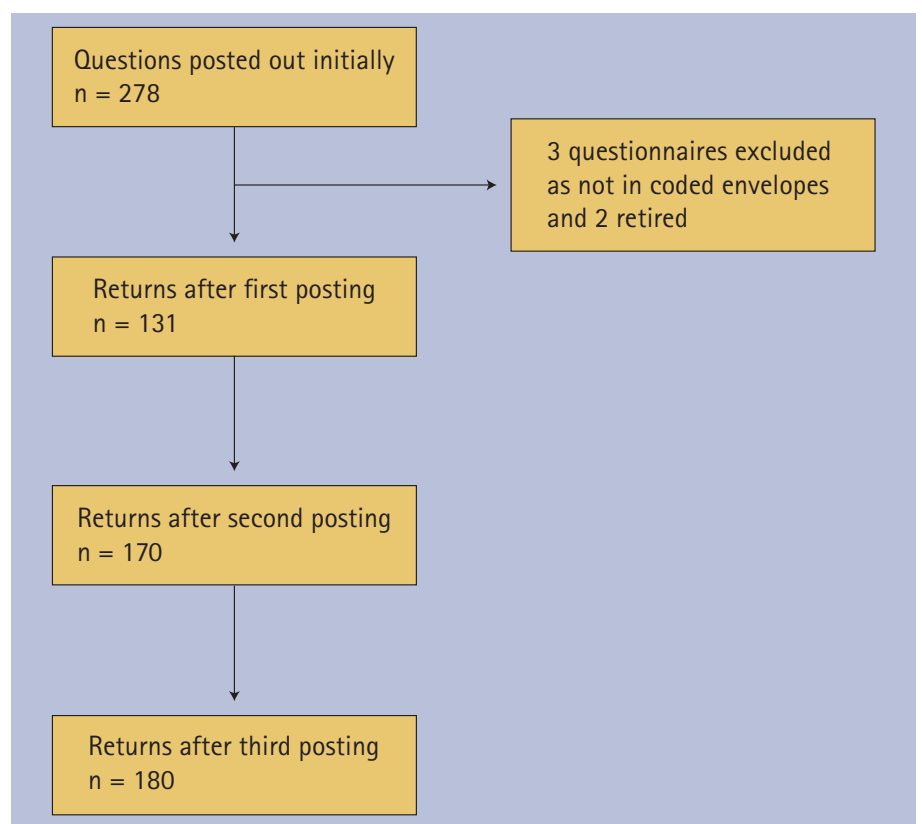


Fig. 2 The number of responses at the different stages of the survey

random method based on their position in the specialist list on the GDC website. The list was divided into groups of five names and the second and fifth of each group were chosen. These included all sexes, age groups and practice locations.

Each specialist was allocated a code that enabled responses to be monitored. The questionnaires were sent out in June 2007, with a covering letter and return self addressed envelope. Replies were collated after approximately six weeks and those that had not responded to the initial questionnaire were sent a reminder questionnaire in August 2007. The second questionnaire

was accompanied by a covering letter emphasising the importance of the survey and requesting the practitioners to respond. Each response envelope was again coded to monitor the response rate. Sending out the second questionnaires and the receipt of the responses took about four weeks.

Those who did not respond to the first two mailings of the questionnaire were sent a final reminder questionnaire with covering letter in September 2007. This time the return envelopes were not coded to encourage those who might have been concerned about anonymity. No further reminders were posted.

Data collected were entered into an Excel spreadsheet (Microsoft Corp, USA) and a descriptive analysis was undertaken.

Clinical and laboratory study

The results of the survey were used to inform the second part of the study, the objective of which was to examine the effectiveness of the common methods used to clean photographic retractors. The investigation was carried out in the Orthodontic Department of the Charles Clifford Dental Hospital and the Microbiology Laboratory of the School of Clinical Dentistry, Sheffield. Four of the most commonly reported methods of cleaning from the first part of the study were investigated:

- Alcohol wipe
- Handwashing
- Ultrasonic cleaning
- Washer-disinfector.

Thirty pairs of photographic retractors (American Orthodontics UK, Bucks, UK) which had been used on orthodontic patients requiring photographs (Fig. 1) were collected for each of the cleaning methods. One retractor from each pair was randomly chosen to be the unwashed control and immediately placed in 20 ml of PBS-Tween for elution. The other was subjected to the cleaning procedure outlined below and was deemed to be the experimental retractor. After each treatment the experimental retractors were also placed in 20 ml PBS-Tween for elution of residual protein.

Alcohol wipe

The retractors were thoroughly cleaned with one alcohol impregnated wipe (Azo Active, Synergy Health PLC, UK) and allowed to dry for five minutes.

Hand washing

The retractors were hand washed according to normal practise within the department, which involved placing them in plain tap water for 15 minutes and applying slight agitation for a few seconds by hand.

Ultrasonic cleaning

Without prior rinsing, the retractors were placed in an ultrasonic bath (Prosonic 1000, Sultan Healthcare, NJ, USA) containing Opti-Prep Ultra (Optident, UK) for fifteen minutes and then transferred to elution buffer. Routine practise in the Orthodontic

Department of the Charles Clifford Dental Hospital is to change the solution in the ultrasonic bath twice daily and once the retractors were in the ultrasonic bath, no other instruments were added until after the retractors had been removed.

Washer-disinfector

The retractors were placed in sterilisation pouches and then on trays in a Deko D32 washer disinfector (Dekomed, UK), sited in the Royal Hallamshire Hospital, Sheffield, within 10 to 20 minutes after removal from the patient. The machines were fully commissioned and regularly validated. The full cycle lasted 90 minutes and the retractors were then placed in elution buffer.

Elution was performed with gentle agitation for 20 minutes at room temperature and aliquots (100 µl) were subjected to an antibody capture Enzyme-Linked Immunosorbent Assay (ELISA) for amylase as a representative protein of saliva, and for albumin as a representative protein of blood.

ELISA

Anti-human albumin and anti-human amylase (diluted 1:10,000 in bicarbonate buffer pH 9.6; Sigma) were coated onto ELISA wells (Corning Costar, High Wycombe, UK) overnight at 4°C. After washing and blocking with 1% (w/v) skimmed milk, suitably diluted samples were placed in wells for 1 hour at 37°C, washed again and probed with biotin-labelled anti-albumin or anti-amylase antibodies (1:10,000). Antibodies were biotin-labelled by reaction with biotin-N-hydroxysuccinimide ester (Sigma; 44 µg/ml in PBS pH 7.5) as described previously. Fourteen wells were developed with avidin-conjugated horseradish peroxidase (1:10,000; Dako, Ely, UK) and o-phenylenediamine (1 mg/ml). The colour generated was measured in a plate reader (FLUOStar Galaxy, BMG Lab technologies, Offenburg, Germany). Quantitative data of the level of contamination on each retractor were obtained by comparison with standard curves generated using pooled, clarified stimulated human whole saliva (freshly collected from four volunteer laboratory personnel) and pooled human serum (Sigma) and purified albumin (Sigma) as appropriate. The assay was sensitive to approximately 0.0005 µg of albumin and could detect 10⁻⁹ ml of saliva.

To determine the recovery of contaminating material from the retractor after elution in PBS, each retractor was probed with the two antibodies to detect retained proteins. The resultant colour generated in solution was measured as above.

Statistical methods

A sample size calculation was performed using data from a previous investigation.¹⁵ This study found that 50% of orthodontic bands that had been tried in the mouth and cleaned using ultrasonic cleaning had detectable levels of amylase, albumin or both. Based on the assumption that this proportion could be reduced to 20% after cleaning with a washer-disinfector, the sample size calculation for categorical data with a binary outcome¹⁷ was used to determine that a sample size of 30 would be sufficient to determine a significant difference to a power of 0.85 and significance level of 0.05. The data were examined using descriptive statistics. The proportions of samples with detectable volumes of amylase, albumin or both were calculated. The amount of protein detected on the control samples varied slightly between the different cleaning techniques, therefore the proportional reduction in protein was determined to overcome this. The amount of detected protein (amylase or albumin) on the experimental retractors was subtracted from the amount of detected protein on the unwashed control retractor from the same individual and the percentage reduction in the volume of detected protein was calculated. These data were not normally distributed, therefore the differences in the percentage reductions in detected proteins between the different cleaning techniques was tested using the non-parametric one way analysis of variance (Kruskal-Wallis).

RESULTS

Postal questionnaire

Figure 2 shows a flow chart of the responses at each stage of the survey. A total of 278 questionnaires were sent out. The number of questionnaires returned after the first posting was 131, which was a response rate of 47.1%. After the first reminder a further 39 were returned and a further 10 were returned after the second and final reminder. This gave a total of 180

questionnaires returned after three postings, an overall response rate of 65%.

In the first return three questionnaires were without the coded envelopes and were excluded since it was not possible to show who had sent them. Two of the responders said they were no longer practising because of retirement and were excluded from the data analysis.

The majority of responders were male ($n = 121$; 66.9%) and had obtained their first dental degree between 1954 and 2000 (median 1987). Over three quarters had the Membership in Orthodontics diploma from one of the Royal Colleges or the MOrth and the DOrth ($n = 140$; 77.3%) and nearly one half ($n = 89$; 49.1%) had a masters degree. Most responders worked only in specialist practise ($n = 95$; 52.8%) or only in hospital practise ($n = 39$; 21.7%).

Photographic retractors

Most of the respondents ($n = 157$; 87.2%) were taking photographs as part of their orthodontic practise and nearly all of these (98.7%) were routinely using photographic cheek retractors. The 22 individuals who were not taking photographs and the 1 respondent that did not respond were excluded from the rest of the analysis. Plastic retractors were most commonly used ($n = 145$; 92.4%). Eight individuals were using metal retractors and four were using both plastic and metal retractors. The majority of respondents ($n = 134$; 85.4%) stated that they were disinfecting/cleaning the retractors between patients.

Many different methods were used for cleaning photographic retractors (Table 1). The commonest method was hand washing either on its own ($n = 36$; 22.9%) or in combination with another method ($n = 35$). The next commonest method was the washer-disinfector, which was used on its own by 19 respondents (12.1%) or in combination with another method by 14 people. An alcohol wipe was used on its own by 8 people (5.1%) or in combination with other methods by 28 individuals. The ultrasonic was used exclusively by 8 individuals (5.1%) or in combination with other methods by 10 individuals. Four responders stated that they autoclaved the retractors without cleaning them first and 36 did not state the method of cleaning.

Table 1 The different methods of cleaning photographic retractors

Method	n	%
Handwash only	36	22.9%
Handwash & alcohol wipe	17	10.8%
Handwash & ultrasonic	2	1.3%
Handwash & washer-disinfector	6	3.8%
Handwash, alcohol wipe & ultrasonic	4	2.5%
Handwash, alcohol wipe & washer-disinfector	4	2.5%
Handwash, ultrasonic & washer-disinfector	1	0.6%
Handwash, alcohol wipe, ultrasonic & washer-disinfector	1	0.6%
Alcohol wipe only	8	5.1%
Alcohol wipe and ultrasonic	1	0.6%
Alcohol wipe & washer-disinfector	1	0.6%
Ultrasonic cleaner only	8	5.1%
Ultrasonic & washer-disinfector	1	0.6%
Washer-disinfector only	19	12.1%
CSSD	8	5.1%
No specified decontamination	40	25.4%
Total	157	100

Table 2 The numbers and proportions of samples with detectable amylase from the unwashed control and cleaned experimental retractors

	Control		Experimental	
	n	%	n	%
Alcohol wipe	30	100%	30	100%
Handwash	30	100%	12	40%
Ultrasonic	30	100%	5	17%
Washer-disinfector	30	100%	1	3%

Table 3 The numbers and proportions of samples with detectable albumin from unwashed control and cleaned experimental retractors

	Control		Experimental	
	n	%	n	%
Alcohol wipe	30	100%	24	80%
Handwash	30	100%	13	43%
Ultrasonic	30	100%	16	53%
Washer-disinfector	30	100%	1	3%

The majority of responders claimed to sterilise retractors following cleaning ($n = 115$; 73.2%). Amongst those that sterilised the retractors the commonest reported method was an autoclave either alone ($n = 62$; 53.9%) or in combination with cold sterilisation ($n = 3$; 1.9%). One reported using a hot air oven, as well as an autoclave; however a large number of respondents were using cold sterilisation methods ($n = 50$; 43.5%).

Clinical and laboratory study

A total of 240 photographic retractors (120 unwashed control samples and 120 cleaned experimental samples) were collected during the study from 120 patients.

Table 2 shows the numbers and proportions of unwashed control and cleaned experimental samples that had detectable levels of amylase as determined by ELISA. The results from the control samples show that all the retractors were contaminated

with amylase after being used in the mouth and prior to cleaning; however there were marked differences in the effectiveness of the different methods of cleaning the retractors. Whereas all the retractors cleaned using an alcohol wipe had residual detectable levels of amylase, only one retractor had any detectable amylase following cleaning using the washer-disinfectant. Five out of 30 (17%) experimental retractors subjected to ultrasonic cleaning and 12 out of 30 (40%) cleaned by hand had residual amylase.

Table 3 shows the numbers and proportions of unwashed control and cleaned experimental samples that had detectable levels of albumin as determined by the ELISA for albumin. Again this shows that all the retractors were contaminated with albumin after being used in the mouth and prior to cleaning; however the results from the experimental retractors subjected to the four cleaning procedures demonstrated an even proportion of retractors with residual detectable levels of albumin compared with amylase. The retractors cleaned using the alcohol wipe method demonstrated a high proportion with residual albumin compared with the other techniques (80%). The proportion of retractors with residual albumin following hand washing (43%) was similar to the proportion with residual amylase, whereas the proportion of retractors subjected to ultrasonic cleaning that had remaining albumin (53%) was much higher compared to those with the residual amylase. The washer-disinfectant again proved to be the most efficient method of cleaning photographic retractors of albumin.

Figure 3 shows the proportions of the 30 experimental retractors cleaned using the four techniques with detectable amylase only, the proportions with detectable albumin only and the proportions with both proteins detected.

The washer-disinfectant was clearly the most effective method of cleaning photographic retractors. Following cleaning in the washer-disinfectant only one retractor out of the 30 collected showed detectable levels of amylase; one retractor showed detectable levels of albumin and no retractors showed detectable levels of both proteins. This contrasts with the retractors cleaned using an alcohol wipe, in which 80% showed residual contamination with both amylase and albumin and the remaining 20% were

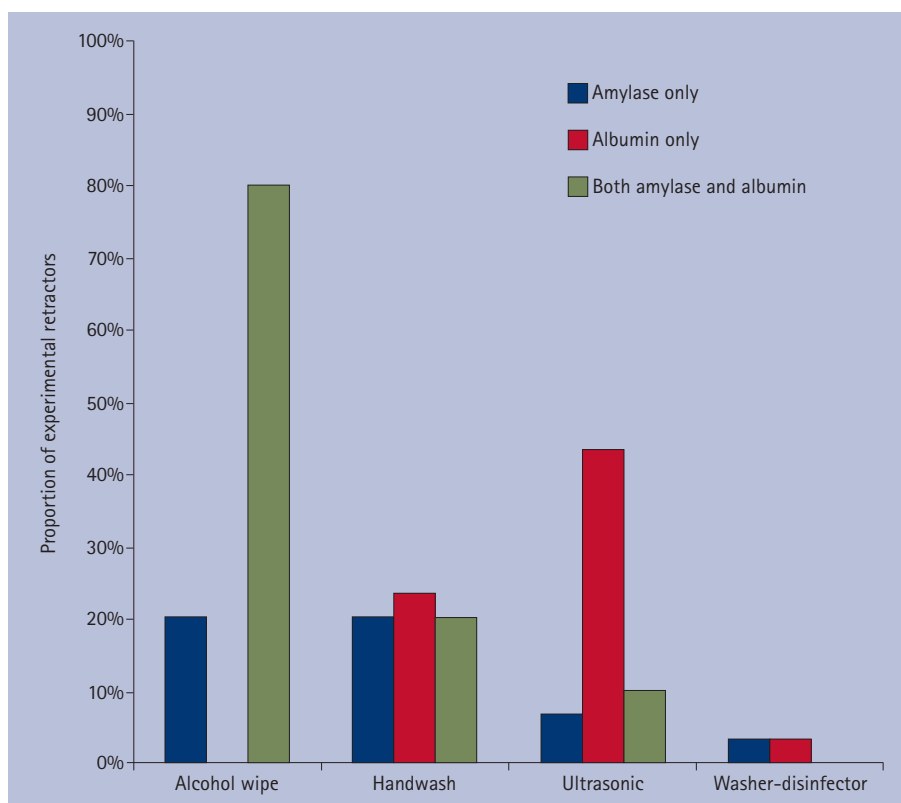


Fig. 3 The proportion of experimental retractors with detectable amylase only, detectable albumin only and when both proteins were detected

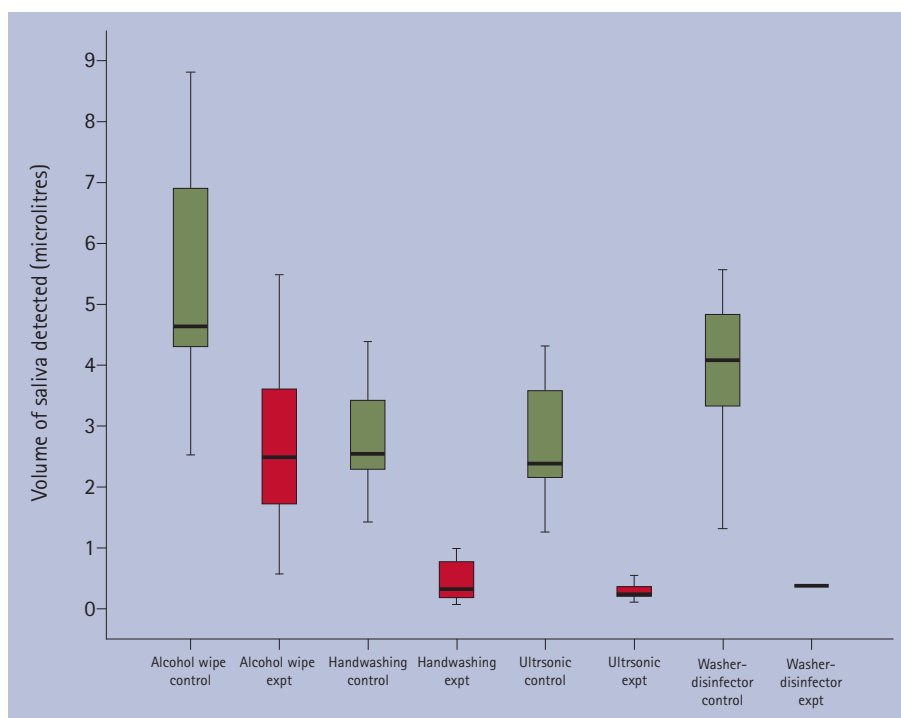


Fig. 4 Box plots showing the medians, interquartiles and range for the amylase detected (μ l) on the unwashed controls and cleaned experimental retractors on which amylase was detected for the four cleaning techniques

still contaminated with residual amylase. Following hand washing the proportions of retractors with residual contamination with amylase, albumin and both were approximately equal, whereas ultrasonic cleaning

was much less effective at removing albumin than amylase.

Figure 4 shows box plots of the descriptive data for the volumes of saliva (as represented by amylase) measured using the

ELISA on the unwashed controls and those cleaned experimental retractors that had detectable levels of amylase. The alcohol wipe was the least effective method of reducing contamination with saliva, as the median volume of detected saliva reduced from 4.65 μl on the unwashed control retractors to 2.49 μl on the cleaned retractors. The median proportional reduction in volume of saliva was 50% (range 0–83%). The equivalent values for the hand washed retractors was 2.55 μl for the unwashed controls to 0.32 μl for the cleaned hand washed retractors (median proportional reduction 87%; range 56–98%); 2.30 μl for the unwashed ultrasonic controls to 0.22 μl for the cleaned ultrasonic experimental retractors (median proportional reduction in volume 90%; range 77–95%); and only one retractor in the sample cleaned using the washer-disinfector showing a detectable volume of saliva (2.61 μl control to 0.37 μl experimental; proportional reduction in volume 86%). The Kruskal-Wallis test showed a highly significant statistical difference in the reduction between the various cleaning techniques in saliva detected on the unwashed control to that of the cleaned experimental retractors ($p < 0.001$).

Figure 5 shows box plots of the descriptive data for the amounts of albumin measured using the ELISA on the unwashed controls and those cleaned experimental retractors which had detectable levels of albumin. This shows that again the alcohol wipe was the least effective method for reducing contamination with albumin, as the median amount of albumin reduced from 7.17 μg from the unwashed control retractors to 1.22 μg from the cleaned retractors. The median proportional reduction in the amount of albumin was 80% (range 0–97%). The equivalent values for the hand washed retractors was 3.34 μg for the unwashed controls to 0.26 μg for the cleaned hand washed retractors (median proportional reduction in amount 86%; range 36–98%); 3.72 μg for the unwashed ultrasonic controls to 0.54 μg for the cleaned ultrasonic experimental retractors (median proportional reduction in amount 85%; range 54–98%); and only one retractor in the sample cleaned using the washer-disinfector showing a detectable volume of albumin (2.5 μg control to 0.45 μg experimental; proportional reduction in volume 90%). The Kruskal-Wallis

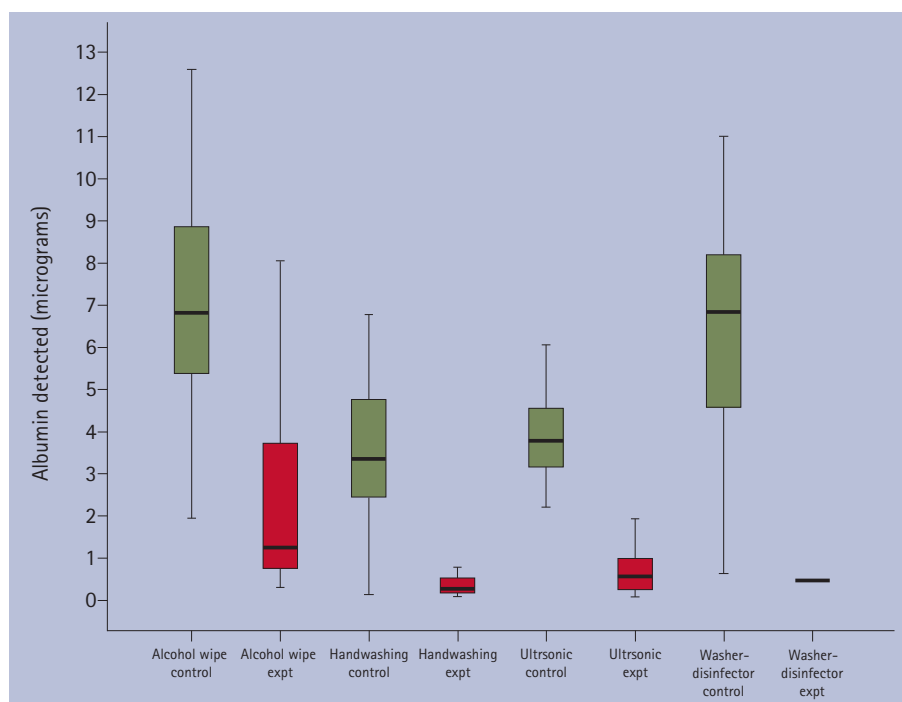


Fig. 5 Box plot showing the medians, interquartiles and range for the albumin detected (μg) measured on the unwashed controls and cleaned experimental retractors on which albumin was detected for the four cleaning techniques

test showed a highly significant statistical difference between the techniques in the proportional reduction in albumin detected from the unwashed control and cleaned experimental retractors ($p < 0.001$).

To estimate the concentration of salivary albumin, saliva was collected from four volunteer laboratory personnel who had had no dental procedures carried out prior to saliva collection and who had no obvious gingivitis. A standard curve was constructed and the mean value of albumin in saliva was estimated to be 38.3 $\mu\text{g}/\text{ml}$, which was within the values reported by others.¹⁸ Hoek *et al.* also found that the concentration of albumin in blood (34–54 mg/ml) is approximately 1,000 times the amount in saliva. For the purposes of this study samples that had values close to or below 38.3 $\mu\text{g}/\text{ml}$ albumin were taken to be due to saliva contamination, whereas samples with an albumin concentration greater than this were considered to be contaminated with blood. The potential amount of blood contamination detected on the retractors was estimated in the following way. The mean amount of albumin detected on all the uncleaned control retractors was 5.2 μg (SD 2.7 μg). If this entire albumin was derived from saliva, it would represent the equivalent of 132 μl of saliva. However, the largest

volume of saliva detected on a retractor was 8.83 μl . The average volume of saliva detected on the uncleaned retractors was 3.76 μl (SD 1.69 μl), which, based on our assay of saliva from four volunteer laboratory personnel, showed this will contain approximately 0.14 μg of albumin. Subtracting this value (0.14 μg) from the average amount of albumin detected on the retractors, (5.2 μg) indicates an average blood/serum contamination of approximately 0.1 μl . Applying the same calculations to cleaned retractors it was found that after hand washing the amount of blood/serum remaining was 0.003 μl , after ultrasonic cleaning it was 0.005 μl , after alcohol wiping it was 0.05 μl and after going through the washer-disinfection it was 0.0004 μl . However, using the highest amount of albumin left on the retractors after cleaning then the highest level of blood/serum that might be present after cleaning was: hand wash 0.06 μl ; ultrasonic 0.05 μl ; alcohol wipe 0.17 μl ; and washer-disinfector 0.01 μl .

DISCUSSION

This investigation was undertaken in two parts. The first stage involved a postal questionnaire to determine the proportion of specialist orthodontic practitioners who perform clinical photography and the

methods they used to decontaminate photographic retractors. Although many consider a self-report questionnaire to be an unreliable method of determining clinical practise, particularly with regard to cross-infection control,^{19,20} the results were used to inform the second part, which investigated the effectiveness of commonly reported methods of cleaning.

The survey found that the majority of orthodontic practitioners were routinely taking photographs, underlining the importance of this as a clinical record for monitoring progress and treatment outcome. The majority of clinicians who did carry out clinical photography were using plastic retractors and reported that they were decontaminating and sterilising the retractors between patients using recognised and effective procedures, but the number and combinations of techniques demonstrates the lack of clear guidelines for dealing with the potential risk of cross-infection from photographic retractors.

The results of the second part of the study clearly show that the most effective method of cleaning photographic retractors prior to sterilisation is with a washer-disinfector. Out of the sample of 30 contaminated retractors only two had detectable levels of protein following cleaning in a washer-disinfector; however it should be noted that none of the methods was 100% effective at removing all protein contamination from photographic retractors.

Several studies have been carried out examining the effectiveness of cleaning procedures in dentistry. The inadequacy of manual methods compared with the ultrasonic washer has been shown several times.^{5,21,22} Cafruny *et al.*²¹ used an IgG marker to test for blood contamination on dental instruments following a routine prophylaxis and found that ultrasonic cleaning was both more effective and more consistent at reduced blood contamination than hand washing. Lowe *et al.*,¹⁰ using the Kastle-Meyer test for blood, found that a far smaller proportion of matrix bands and their retainers had residual contamination following ultrasonic cleaning compared with those washed by hand.

Several studies have shown that automated cleaning is more effective than manual cleaning, particularly with instruments and equipment that have a lumen.^{23,24} Apart from the effectiveness, other advantages of automated

cleaning techniques such as washer-disinfectors include minimal handling of instruments reducing the risk of accidental injury, a reproducible closed cycle and production of a printed record verifying that the correct cycle has been achieved.⁵

Smith *et al.*²⁵ have also shown that the instrument shape and material might affect how well it is cleaned prior to sterilisation. They found that 98% of their sample of endodontic files used in general practice retained visible debris after washing, with a median amount of 5.4 µg residual protein. This is a much higher level of detectable residual protein compared with the worst median value of 0.99 µg found on the retractors cleaned with the alcohol wipes. This is probably again due to the smoother surface features of the photographic retractors, but also the greater amount of contamination of endodontic files following their use within the root canal. The design of the cleaner may also affect the outcome of cleaning. Perakaki⁷ found higher mean debris scores on endodontic files cleaned in a washer disinfector compared to an ultrasonic cleaner, but concluded that this was probably due to the design of the file holder, which prevented access to the water jets of the washer disinfector. Following this research endodontic files are now all considered single use only in the UK.²⁶

The finding that no method of cleaning is 100% effective has been shown in several studies.^{11,27} Both Sanchez and Macdonald²⁷ studying dental instruments and Whitworth *et al.*¹¹ with matrix bands found residual contamination even following cleaning in washer-disinfectors. Whitworth *et al.* found a 99.7% reduction in median blood volume following cleaning in a washer-disinfector, however this reduced to 91.4% when they examined 20 clinically contaminated bands collected by 10 general practitioners. These results are similar to those found in this study.

We found that methods involving a denaturing agent, such as alcohol, were not effective at cleaning photographic retractors. It is possible that albumin is denatured by the alcohol treatment and in that form is more persistent and so not removed satisfactorily. This is quite worrying since in 5% of responses to the survey this was the only means of cleaning photographic retractors.

The ELISA technique used in this study

involves a specific test for amylase and albumin and although it does not measure the exact volumes of saliva and blood, it does allow an estimation of the volumes of saliva and blood present. The disadvantage of the method is that it is time consuming and requires considerable expertise. Other studies have showed the presence of residual proteins and organic debris on cleaned instruments using different techniques.^{14,28} These methods do not directly measure viral or bacterial loads or attempt to determine the viability/infectivity of any bacteria or viruses that might have been present. The presence of residual protein does not indicate definite infection risk; however, the fact that protein still remained after some cleaning procedures suggests a potential for cross-infection from prions or viruses. Prions are resistant to chemical and thermal decontamination and the potential risk of cross-infection is worsened when the instruments are not cleaned properly, then subjected to heat, as prions are known to undergo fixation when subjected to heat.²⁹

Infective risk

The survey invited free comments from responders and some of these showed potential confusion over the risk of cross-infection from photographic retractors. For example one person wrote:

'Retractors are like cutlery used in restaurants and since these are not sterilised there is no need to sterilise retractors.'

Another wrote:

'No blood contact when retractors are used for photography therefore no need for sterilisation, decontamination is okay.'

There was also a clear frustration that the requirements of cross-infection control were considered to be onerous and unnecessary:

'No cross-infection has been traced back to dentistry, microbiology is wagging the dog.'

There is no doubt that the presence of small amounts of infected blood or serum on used retractors could pose a cross-infection risk from hepatitis B virus and other viruses found in blood or saliva. The potential risk from prions would seem to be extremely low, but this is very difficult to interpret both from these data and

from the clinical procedure being studied.

It has been shown that the presence of a viral hepatitis B DNA level of 10^5 genome equivalents per ml (geq/ml) in serum is sufficient for the transmission of viral hepatitis during surgery.³⁰ The median HBV in serum in that study was 2.10×10^5 geq/ml (range 373– 4.13×10^9 geq/ml) while in saliva the median level of HBV DNA was 2.27×10^4 geq/ml (range 0– 9.25×10^6 geq/ml). The infective dose of hepatitis B virus is estimated to be 20–1,000 geq. These estimates generally apply to an inoculation route of delivery, but it is uncertain what the level would be for mucosal contact with minor breaks at the angles of the mouth, as might occur during photography.

In the present study it has been shown that the median volumes of saliva (using the amylase estimates) before cleaning ranged between 2.4 μ l and 4.6 μ l with a mean of 3.76 μ l. These volumes equate to a mean of <1–3 infective doses, using the median estimate of HBV load in saliva, but there could be up to 1,500 infective doses if the subject's saliva contains a high viral load (9.25×10^6 geq/ml). After cleaning, the only procedure that regularly left detectable amounts of saliva was the alcohol wipe. The median volume remaining was 2.5 μ l, which equates to a median number of 790 viruses, equating to <1–40 infective doses. If the subject was a high HBV load carrier, this could increase to approximately 22–1,000 infective doses.

Each of the cleaning methods employed reduced the amount of albumin present on the photographic retractors by >90%, with the exception of the alcohol wipe, which removed approximately 73% of it. The quantity of albumin remaining after cleaning ranged between 0.015–1.9 μ g depending upon the method. This is equivalent to 0.0004–0.05 μ l of blood and, therefore, less than one infective dose using the median level of HBV in blood or 20–1,000 infective doses for a high HBV load carrier. Even after cleaning with the washer-disinfector, there might be as many as 1–80 infective doses remaining on those rare retractors that retained detectable albumin.

Of course such theoretical estimates do not take into consideration the effect of viral inactivation on these loads, which would greatly reduce the amount of infective HBV present on the retractors. Also, as mentioned above, it seems likely that

the dose of HBV that would be required to establish an infection following contact with a contaminated retractor would be much higher than the doses quoted for parenteral routes of delivery. However, the fact that we have demonstrated a significant amount of protein on uncleaned retractors and even following certain cleaning procedures makes the efficiency of a sterilisation procedure possibly uncertain.

Although the level of blood contact in the orthodontic clinic seems to be minimal, this study showed that there was albumin easily detectable on most retractors at a level that is likely to represent some serum/blood contamination and thus could pose a cross-infection hazard in the clinic. By extrapolation, in other clinical areas such as oral surgery and periodontics, that perform procedures more prone to bleeding, these retractors would be potential vehicles for transmitting infection unless cleaned carefully. Thus, there does seem to be a tangible cross-infection risk associated with inadequately cleaned retractors and so clear guidelines on cleaning and sterilisation of these items should be introduced.

CONCLUSIONS

The most effective method of cleaning photographic retractors prior to sterilisation is with a washer-disinfector. No method of cleaning photographic retractors was 100% successful at removing salivary proteins and it is essential to use an effective method of sterilising retractors prior to reuse. Use of the alcohol wipe left extensive residual protein on the retractor and therefore cannot be recommended. Further studies need to be carried out in this field to show the effect these different methods of cleaning would have on viral and bacterial loads from used photographic retractors.

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