May I spit on your photograph?

A preliminary investigation into the effectiveness of saliva and a synthetic alternative for surface cleaning silver gelatin photographs

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Abstract

There is a diverse range of views regarding body fluids (including saliva) among community groups. This range of opinion is reflected within the field of photographs conservation regarding the use of saliva to clean photographic prints. Attitudes to the use of saliva vary; some professionals use it openly and often, some are guarded about their use and others display a strong aversion to it. Given these widely ranging views there will be situations where the use of saliva will not be appropriate. Identifying a safe, effective alternative could be advantageous.

There is little published information available on the use of human saliva or synthetic alternatives in conservation treatments, and still less in the area of photographs conservation. Fundamental questions about the use of saliva require answers. For example, could the composition and variability of human saliva make this solution unsafe for use with photographic prints? Are human saliva and the synthetic alternative effective at removing surface grime and accretions from photographic prints? When used on photographic prints, how do the solutions compare to each other in preservation safety and cleaning efficacy?

This preliminary investigation compares the preservation safety and cleaning efficiency of human saliva and a synthetic alternative on silver gelatin photographic prints (referred to in this text as photographic prints). Information about the components and variability of saliva, collated from allied disciplines, is presented. The cleaning functions of the two solutions, Photographic Activity Test results and pre- and post-cleaning density readings are discussed. Observations are made relating to the clearing of enzymes from treatment sites.

Introduction

Most sources agree that water makes up approximately 99% of saliva. However, the materials making up the approximately 1% solids vary widely in quantity between individuals and within the individual (Edgar 1992: p308). These variations depend on many factors and it is therefore not surprising that sources listing the composition of human saliva tend to contradict each other regarding the presence and concentrations of the constituents. For a summary of human salivary components see Table 1.

Salivary components and surface cleaning

Solvents. The principal solvent in saliva is water and without its presence the other components would have little or no effect. Due to the surface tension and polarity of water it is not particularly effective at wetting surfaces and dirt. Water may dislodge only large, loosely bound particles of dirt that can be easily redeposited onto the treatment surface (Phenix and Burnstock 1992: p30, Wolbers 2000: p6).

Surfactants. Amino acids and proteins such as albumin act as weak surfactants during surface cleaning. Surfactants are crucial if the dirt contains oily or fatty substances as these substances repel water and

Table 1. Composition of saliva

Water	[makes up approximately 99%]
Buffers	both hydrogen phosphate and carbonate. pH is 6.8–7.4
Proteins	chiefly albumin
Amino acids	most of the 20 or so commonly found ones
Na^{+}	in substantial amounts, with traces of K^{+}, Ca^{+2}, Mg^{+2}, Cu^{+2}, Co^{+2}, SO_4, SCN^{-}, NH_4^{+} and Cl^{-}
Diammonium citrate	(0.01% w/v)
Enzymes	amylase, lipase, protease, lysozyme

Wolbers 2000: p6. Used with permission.

increase the adhesion between dirt particles (Phenix and Burnstock 1992: p30, Wolbers 2000: p6).

Buffers. The principal buffers in saliva are carbonate ions (CO_3^{2-}) and hydrogen phosphate ions ($H_2PO_4^-$). Maintaining a specific pH range ensures the stability and effectiveness of enzymes while decreasing the potential for damage to occur to the photographic print (DeSantis 1983: pp9–10, Phenix and Burnstock 1992: p31, Wolbers 2000: p6).

Ion or specific-ion effects. Using a slightly alkaline solution improves cleaning effectiveness by causing the dirt and the substrate surface to hold strong negative charges and therefore repel each other. In saliva citrate is thought to facilitate this effect (Phenix and Burnstock 1992: p35). This dislodges dirt particles and prevents them from being redeposited onto the item (Phenix and Burnstock 1992: pp30–31).

Chelating agents. Chelates (e.g. potassium) are capable of complexing metallic ions so that they are not free to react with other substances (Phenix and Burnstock 1992: p28). This prevents them forming insoluble precipitates on the treatment surface and facilitates the removal of a significant proportion of dirt by breaking the bonds between polyvalent metal ions (Hofenk de Graaff 1968: p125). Chelating agents may also damage historic deterioration products on photographic prints (e.g. silver mirroring).

Adjuvants. Mucin is a polymeric substance that increases the viscosity of saliva. It acts as a lubricant and suspension agent that helps prevent dirt particles being redeposited onto the treatment surface (Wolbers 2000: p6). Substances such metal ions, vitamins and various non-protein molecules may also be necessary to ensure the activities of enzymes.

Table 2 shows the effect of materials present in saliva in relation to conservation treatments.

Treatment concerns relating to saliva

The most commonly cited areas of concern when using saliva in conservation treatments are the complexity and variability of this secretion. However, the impact of compromised oral hygiene/health and effective clearing of the oral cavity after ingesting food and drink are also important issues. A further concern is the lack of conclusive research

Table 2. Effect of materials present in saliva

Water	solvent
pH/buffer(s)	hydrogen ion concentration or control
Surfactant(s)	wetting, emulsifying, dispersal, solubilization
Ion or specific-ionic effects	dissolution and formation of certain salts
Chelation	metal-complexing agents
Enzymes	materials that can effect a chemical change or modify a material to change solubility
Adjuvants	thickeners, suspension aids, solvents, corrosion controllers, preservatives, etc

Wolbers 2000: p7. Used with permission.

around the clearing of enzymes from the item post treatment. Coupled with this is the unknown period of time when enzymes could reactivate if suitable conditions reoccur.

As far as this author was able to establish, many of the variations in the composition or conditions of saliva are also associated with a decrease in pH arising from:

Dehydration. Commonly caused by inadequate water intake, dehydration is exacerbated by sustained exercise or by ingesting large amounts of coffee or alcohol.

Acidic substances. Consuming substances such as fruit drinks, fizzy drinks, wine, beer, citrus fruits and some medications (e.g. aspirin) significantly lowers the pH of saliva.

Food with a high carbohydrate content. Carbohydrates cause the bacteria in dental plaque to release acid by-products as they metabolise sugar. Carbohydrates also provide the nutrients necessary for oral bacteria to flourish in the oral cavity.

Nicotine. This inhibits salivary gland secretion and increases the risk of periodontal disease; it also makes the disease difficult to detect in smokers. Untreated gum disease causes more acid to be produced by bacteria and

this, combined with the reduced salivary flow, significantly lowers the pH of the oral cavity or mouth.

Eating foodstuffs. After eating, the pH of the oral cavity drops significantly before salivary secretions re-establish an equilibrium of approximately pH 6.5–7.5.

(Chin 2004: personal communication, Edgar 1992: p309)

The effects of compromised oral hygiene/health

Bacteria in the human oral cavity (numbering approximately 10 billion) produce enzymes during their metabolic functions. The saliva of people with halitosis (bad breath) or periodontal disease (gum disease) has elevated levels of enzymes associated with a proliferation of anaerobic bacteria in the oral cavity. These people also have a greater proportion of proteolytic organisms (including trypsome-like enzymes) in their saliva, which are primarily washed off the tongue or infected gums. Proteolytic enzymes break down proteins releasing hydrogen sulphide (H_2S) and methyl mercaptan (CH_3SH). These are VSCs (volatile sulphur-containing compounds) that give off the 'rotten egg' smell of bad breath when sulphur levels exceed 75ppb (Tagg 2004: personal communication).

Sulphur has been identified as one of the two main causes of degradation of photographic image silver (Reilly et al. 1989: p117). Hydrogen sulphide is also specifically mentioned as a factor in the deterioration of silver gelatin microforms (Zinn et al. 1994: pi).

Whole saliva typically consists of stimulated and unstimulated secretions, gingival crevicular fluid, non-adhered oral bacteria, remains of food and minute amounts of ingested chemicals and medications (Edgar 1992: p305, Humphrey and Williamson 2001: p163). Stimulation of the salivary glands during and after ingestion of foodstuffs loosens and washes away food particles, dead cells and bacteria. If the oral cavity is too dry this debris remains in the mouth and becomes a food source for bacteria, eventually causing halitosis. Microbial populations also build up in the mouth during sleep due to reduced salivary flow.

Clearing enzymes post treatment

Due to their origin inside living cells, enzymatic reactions generally require an environment with moderate conditions of temperature and pH, and

the presence of moisture. Generally only a minute amount of an enzyme is required to facilitate reactions, particularly if optimum temperature and pH are maintained.

Testing for enzyme retention and future reactivation is beyond the scope of this research: however, reducing enzymes in the item post treatment is an important step. Proteins (e.g. enzymes) tend to darken as they age; discolouration of enzyme residue could therefore occur post treatment. Enzyme residues also have the potential to reactivate if suitable conditions reoccur. The length of time during which reactivation could occur is uncertain but it could be up to several years post treatment for paper items (Andrews et al. 1992: p319, DeSantis 1983: pp12–13).

Guidelines for the safer use of saliva during conservation treatments

- *Maintain adequate levels of personal hydration* before and during the treatment.
- *Allow a clearing period* after meals/drinks (at least one hour) to allow saliva to re-establish an equilibrium pH and clear food debris from the oral cavity (Edgar and Higham 2004: p86).
- *Test the pH of saliva* pre-treatment – aim for near-neutral or mildly alkaline pH and bear in mind that degraded gelatin will be particularly vulnerable to damage while gelatin in excellent condition is likely to be more stable (Nishimura 2006: personal communication).
- *Clean teeth and tongue* using a toothbrush and water (no toothpaste) prior to commencing treatment.
- *Exercise caution* using saliva from people with bad breath or gum disease; their oral cavity will contain increased levels of sulphur.

Methods for reducing enzyme residues in the item post treatment

- Several rinses of room-temperature distilled water. Note: this reduces residue/s but does not deactivate the enzyme/s.
- Several rinses of water (as above), followed by an ethanol rinse. Note: this deactivates the enzyme/s but they could re-form and then reactivate given suitable conditions.

(Andrews et al. 1992: pp318–9, DeSantis 1983: p11)

Testing for preservation safety and cleaning efficacy
Two solutions were tested and compared to each other: human saliva and Saliva-Like Unsoiling Gainful Solution.

Saliva-Like Unsoiling Gainful Solution (SLUGS)
SLUGS was chosen for testing as it was designed to duplicate a variety of the functions of saliva. Anne Rinuy (Conservation Scientist, Laboratoire des Musées d'art et d'histoire, Geneva) developed SLUGS as a synthetic alternative to saliva in paintings conservation treatments (2002: p959).

SLUGS formula
- 0.3% di-ammonium hydrogen citrate
- 1% sodium hydrogen carbonate
- 2% di-sodium hydrogen phosphate-2-hydrate
- Triacylglycerol lipase 350 mg/100ml distilled water
- Distilled water

See Table 3 for the functions of these materials.

Preparation of SLUGS
1. Dissolve the salts one by one in three-quarters of the final volume of warm water (about 37°C).
2. Maintain the temperature and add the enzyme.

Table 3. Functions of materials present in SLUGS

Water	principal solvent, facilitates the reactions of other components
Citrates	buffers and chelating agents
Hydrogen carbonates	buffers
Phosphates	surfactants (wetting, emulsifying agents) and calcium-chelating agents
Lipase enzyme	hydrolyses fatty substances (optimum activity pH 7.4–pH 7.6 @ 37°C)

Rinuy 2004: personal communication

3. Add the remaining water (heated to approximately 37°C) to reach the final volume.

Fresh solution should be frozen (-18°C or lower) to prevent the enzyme deteriorating. Ensure the containers are very well sealed; lipase is hygroscopic. Use a water bath to thaw and heat the solution to 37°C; maintain this temperature to facilitate the activity of the enzyme. Excess solution must be discarded post treatment (Rinuy 2003: personal communication). At 37°C the optimium pH for activity of lipase is pH 7.4–7.6. If necessary, adjust the pH of the SLUGS solution using citric acid (Nishimura 2003: personal communication).

Safety precautions for handling enzymes
Lipase and alpha-amylase can cause allergic reactions, and saliva is a biohazard, so appropriate safety measures must be taken. Some enzymes can cause allergic reactions and problems such as dermatitis; avoid direct contact with skin and eyes and avoid inhaling dried enzymes (Nishimura 2003: personal communication, Hay 1980–81: p7, Shibayama and Estop 1996: p62).

Photographic Activity Test for preservation safety – saliva vs SLUGS

Methodology
Three separate Photographic Activity Tests (PATS) were carried out to indicate if either saliva or SLUGS cause damaging chemical reactions when in contact with photographs, and whether clearing has an effect. The PAT will not test for the viability of enzymes over time; amylase and lipase are likely to be destroyed during the test by the elevated temperature.

The PAT is a predictive test that establishes whether enclosures or components of enclosures (e.g. papers or paper-based products, inks, adhesives, etc) will chemically react with photographic materials. Layers of samples and detectors are sandwiched together and subjected to conditions of elevated temperature and humidity (70°C and 86% RH for 15 days). Comparison of pre- and post-test density readings establish if unacceptable levels of fading or staining have occurred on the detectors. Visual examination establishes whether unacceptable levels of mottling have occurred.

In these tests both cleared and uncleared samples were used. The arrangement of cleared samples (Test 2 and Test 3) differed from the PAT standard as the solutions were applied to the sample detector strips and distilled water was applied to the control strips to mimic the wetting of the sample detectors (Nishimura 2004: personal communication). Applying the solutions to the detectors was an attempt to mimic the use of cleaning solutions on photographic prints during treatment. To minimise false fail results all samples and detectors were allowed to dry before testing commenced (Nishimura 2004: personal communication).

Samples not cleared of enzymes
Test 1. Strips of fibre-based developing-out paper (DOP) (fixed and washed) coated in each solution. Detectors in this test were not coated directly with the solutions because the uncleared solutions had created striations/application marks on the detectors in an earlier test. These striations could have contributed to unacceptable levels of density change on the detectors and therefore caused a false fail result. To avoid this problem the solutions were applied to developing-out paper and interleaved with Whatman filter paper as per the usual PAT method. The interleaving also served to prevent the DOP strips from blocking to the detectors during the test (Nishimura 2005: personal communication).

Samples cleared of enzymes
Test 2. Detectors coated with solution. Solution cleared by wiping with a fresh cotton wad dipped in distilled water. Repeated.

Test 3. Detectors coated with solution. Solution cleared by wiping with a fresh cotton wad dipped in distilled water. Repeated. Then wiped with a cotton wad dipped in ethanol.

In order to see the effect of a mildly acidic solution, pH 6 saliva was used for Test 3 while Test 1 and Test 2 used saliva of pH 7. SLUGS was tested at pH 8 in all three tests.

Photographic Activity Test results
The test results are shown in Table 4.

Table 4. PAT test results.

PAT	*Description*	*Results*
Test 1	• Solution applied to developing-out paper samples No enzyme clearing steps	Saliva FAILED SLUGS FAILED
Test 2	• Solution applied to Fade and Stain detectors Two enzyme clearing steps: • Two rinses of distilled water.	Saliva PASSED SLUGS PASSED
Test 3	• Solution applied to Fade and Stain detectors. Three enzyme clearing steps: • Two rinses of distilled water. • Final rinse of ethanol.	Saliva PASSED SLUGS PASSED

Testing for cleaning efficacy – saliva vs SLUGS

To determine cleaning efficacy, photographic prints were spot-cleaned using each of the solutions. The prints used were sourced from local second-hand dealers. They were of unknown origin and storage history. The main selection criteria was the presence of significant surface soiling. Prior to the cleaning treatment the prints were spot-tested with distilled water to check the extent of swelling of the gelatin and potential damage caused by water.

Cleaning sites

Eight prints were used, described here as Sample 1, Sample 2 etc. There were four discrete treatment sites (a–d) on each print, located using templates:

Sites cleaned with human saliva:	*Sites cleaned with SLUGS:*
(a) Minimum density area (Saliva D_{min})	(c) Minimum density area (SLUGS D_{min})
(b) Maximum density area (Saliva D_{max})	(d) Maximum density area (SLUGS D_{max})

Each site measured approximately 15 × 15 mm.

Cleaning sequence and method for each test site

1. A swab dampened in the cleaning solution was rolled across the treatment site.
2. A second dampened swab was briefly rolled on the site.
3. Three gentle wipes using the second swab completed the process.

The rolling action of the swab helped to thoroughly wet the site and lift dirt and grime from the surface of the print (Arnold et al. 1992: 14.4.3a, p33). Wiping the site at the end of the cleaning sequence helped remove dirt redeposited by the rolling action and ensured a minimum amount of dirt was pulled over the emulsion.

Assessment

- Changes were visually assessed (using the unaided eye, and optical microscope at 25x magnification).
- To quantify the changes noted during the visual assessments, density readings were taken on a MacBeth TR924 densitometer using Status A filters. The density changes were calculated by subtracting the post-treatment readings from the pre-treatment readings for the red, green and blue channels. These three density readings were then added together to calculate the final density change for each treatment site.

Each site was allowed to dry for 30 minutes post cleaning to ensure accurate density readings. No enzyme clearing was undertaken until after these readings were finished to avoid affecting the results.

Results

Cleaning of Sample 7 resulted in the sudden and complete removal of silver mirroring by both solutions. There was also possible removal of some of the underlying image material (see Figure 1 and Figure 2). This may have been due to the presence of chelating agents in both saliva and SLUGS. Removing silver mirroring was not the aim of this comparison but it is important to know that this deterioration product (which is essentially re-deposited image silver) was affected by the two cleaning solutions.

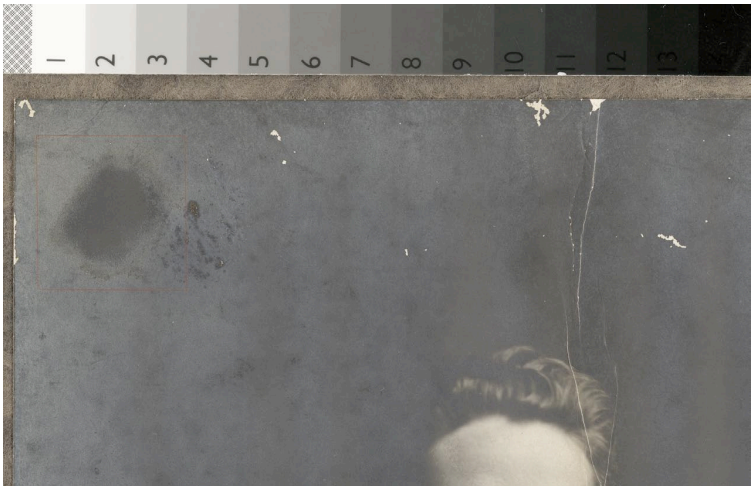


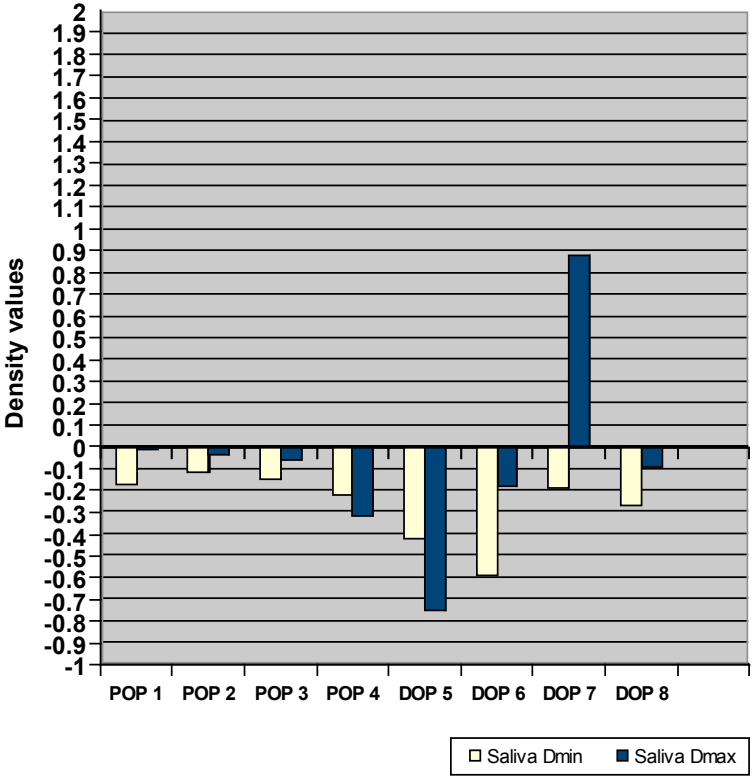
Figure 1. Removal of silver mirroring by saliva – Sample 7 treatment site (b).

Figure 2. Removal of silver mirroring by SLUGS – Sample 7 treatment site (d).

Comparison of density changes
Sample 7 is excluded from all cleaning efficacy comparisons (including all calculations).

Density changes D_{min} and D_{max}
The first comparison of results was between treatment sites where minimal amounts of silver particles were present (i.e. D_{min} sites) and sites where the greatest amounts of silver particles were present (i.e. D_{max} sites) (see Figure 3 and Figure 4). The aim of this comparison was to establish whether image material (i.e. silver particles) was being removed at the

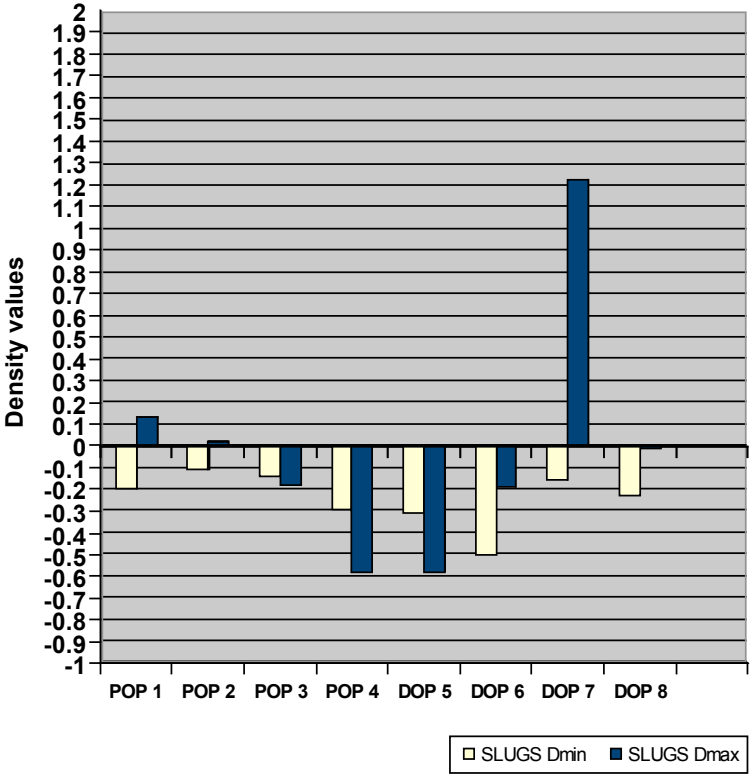
Figure 3. Saliva density changes – D_{min} and D_{max}.



treatment sites. If, for example, there was consistently more density loss at the D_{max} sites than at the D_{min} sites this could indicate that image silver was being removed by the solutions.

Both solutions produced a greater average density change at the D_{min} treatment sites compared to the D_{max} sites. Samples 5 Saliva and 4 and 5 SLUGS showed significantly greater density changes at D_{max} treatment sites. The remaining samples showed that there was minimal difference between the two solutions in terms of the average amount of density change from D_{min} and D_{max} sites: 0.08 for saliva and 0.06 for SLUGS.

Figure 4. SLUGS density changes – D_{min} and D_{max}.

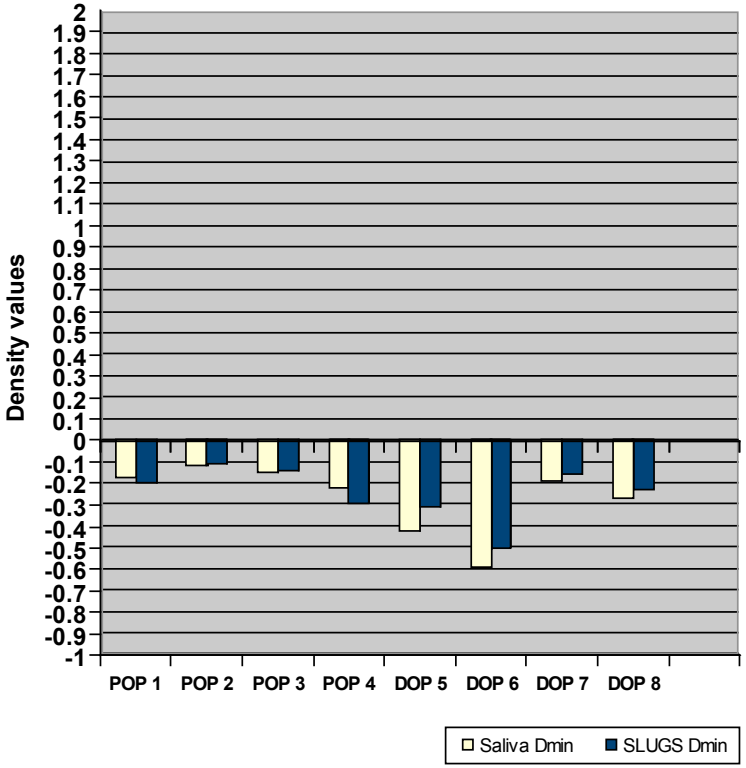


The decrease in density on the treatment sites post cleaning suggests image silver was not being removed from the photographic prints. Due to the small numbers of samples tested no conclusive statement can be made from these results.

Comparison of D_{min} changes

Overall very similar amounts of material were removed from the D_{min} treatment sites by the two solutions at an average difference of -0.02 density units per sample (see Figure 5). Some individual samples (numbers 4–6) showed significantly different density changes post treatment.

Figure 5. Saliva and SLUGS – comparison of D_{min} changes.

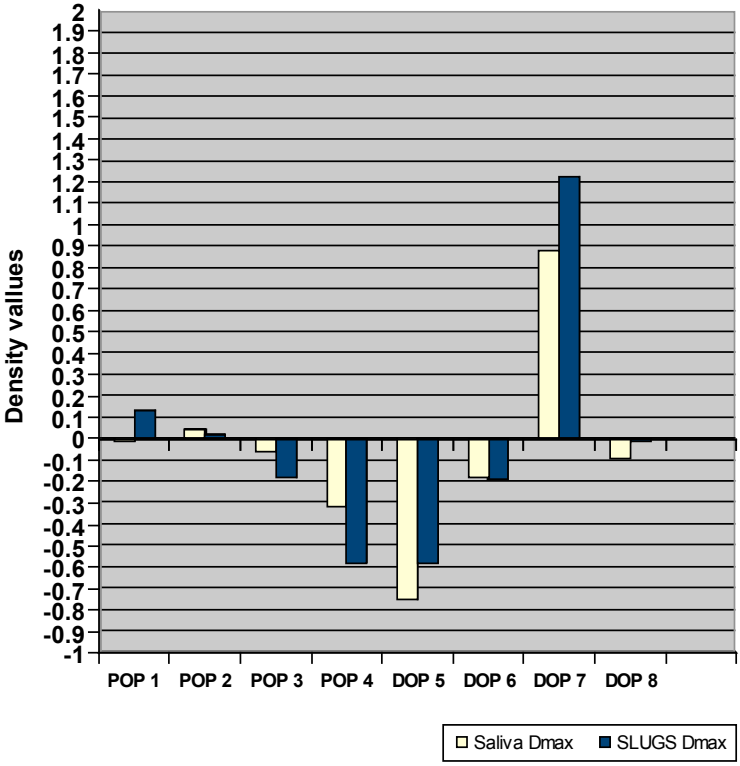


Comparison of D_{max} changes

Overall very similar amounts of material were removed from the D_{max} treatment sites by the two solutions, with an average difference of 0.002 density units (see Figure 6), although most individual samples showed substantially different density changes caused by the two solutions.

The indication from this data is that the two solutions may have similar cleaning capabilities when tested on comparable densities of the print. Due to the small numbers of samples tested no conclusive statement can be made from these results.

Figure 6. Saliva and SLUGS – comparison of D_{max} changes.



Summary and discussion of results

Preservation safety of saliva and SLUGS

- Standard cautions and controls apply when using aqueous-based solutions to clean photographic prints, e.g. accurate process identification, comprehensive condition assessment, etc.
- Comprehensive spot testing is crucial – and a more accurate indication of potential damage may be gained by including the mechanical action of the cotton swab.
- Saliva and SLUGS should not be used on photographic prints exhibiting silver mirroring unless the intention of the conservation treatment is to remove this form of deterioration. Exercise extreme caution; the action of both solutions was immediate with possible removal of the underlying image material.
- Samples failed the PAT when saliva or SLUGS residues were not cleared from the detectors. However both solutions passed when residues were cleared from the detectors using cotton wads dampened with two rinses of distilled water, whether or not this was followed by a third rinse using ethanol.
- The potential for the enzymes in the solutions to reactivate over time was not explored in this research and remains unknown.

Cleaning efficacy of saliva and SLUGS

Due to the small number of samples tested no statistically significant conclusions can be drawn from the results, although general comments can be made:

- Saliva and SLUGS both appear effective at removing soiling from photographic prints.
- Both solutions appear to remove soiling from photographic prints without removing image material.
- Results suggest the solutions have similar cleaning capabilities on photographic prints.

Conclusion

Conservators continue to use saliva to surface clean photographic prints and many consider this acceptable if measures such as comprehensive spot testing of the item and clearing of enzymes post treatment are carried out. It is significant that samples cleared of the solution residues passed the Photographic Activity Test. However, in the absence of conclusive research targeting the long-term potential for enzyme reactivation on photographic prints, preservation concerns remain. Saliva and SLUGS are both efficient aqueous-based surface cleaners when used on photographic prints.

Further research

It would be useful to carry out further testing on a statistically significant number of samples to establish and compare the cleaning efficiency of saliva and SLUGS.

There are also many other areas of this topic that require further investigation:

- the long-term effects of amylase and lipase on photographic prints, and the possible effects of protease in saliva on gelatin emulsions
- halitosis and gum disease; quantifying the risk of sulphur contained in the oral cavity
- alternative synthetic salivas – particularly non-enzyme solutions
- foods that safely increase the cleaning activity of saliva, and foods to avoid when cleaning with saliva.

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