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TECHNICAL NOTE

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Applicability of Two Commercially Available Kits for Forensic Identification of Saliva Stains

ABSTRACT: The RSID-saliva test and the SALIgAE-saliva test are two recently developed forensic saliva detection kits. In this study, we compared the sensitivity and the specificity of the two test kits with the Phadebas® amylase test by analyzing amylases from various sources including human, animals, plants, and micro-organism. The data demonstrate that the RSID-saliva test and the SALIgAE-saliva test offer higher sensitivity and specificity for the detection of saliva than the Phadebas® amylase test. The detection limits of the RSID-saliva test, the SALIgAE-saliva test, and the Phadebas® amylase test equate to 10, 4, and 1000 nL, respectively for human saliva. The RSID-saliva test and the SALIgAE-saliva test were further evaluated by analyzing semen, vaginal secretion, breast milk, blood, urine, sweat, and feces. The results of the two tests are in good agreement. The two tests reacted with urine, breast milk, and feces, but not with semen, vaginal secretion, blood, and sweat.

KEYWORDS: forensic science, saliva, colorimetric test, immunochromatographic membrane test, sexual assault

α-amylases are ubiquitous enzymes existing in both plants and animals. These enzymes (E.C. 3.2.2.1) catalyze the hydrolysis of α-1,4-glucosidic linkages in large chain polymers such as starch and glycogen. In human, these enzymes are produced in human salivary glands and in the pancreas. Human salivary α-amylase is the major protein component in human saliva and it starts the digestion of starch. α-amylases are found in body fluids including serum, urine, semen, sweat, and lip mucus other than saliva (1,2). The human salivary α-amylase is also found in breast milk and cervical mucosa (3,4). Both salivary and pancreatic α-amylases were found in urine, serum, stool, and semen (3,5–8). Isoamylases distinguishable from the salivary and pancreatic isoamylases and specific for the genital tract were found in the Fallopian tube and male accessory genital glands (4). The salivary glands and pancreas have amylase concentrations that are several orders of magnitude greater than those of other tissues, and the amylases secreted by these two organs account for almost all of the serum amylase activity in a normal person (9).

Salivary and pancreatic α-amylases have been studied extensively because aberrant secretion and activity of these enzymes can be related to parotid or pancreatic diseases. Enzymatic and immunological detection are the two methodologies commonly used for identification of amylases clinically (8,10). The Phadebas® amylase test (Magle Life Sciences, Lund, Sweden), radial diffusion, and specific inhibitor are three examples of enzymatic detection of saliva (11–13). ELISA (14,15) and cross-over electrophoresis are examples of antigen-antibody immunological detection. All these methodologies are also used for forensic identification of saliva. Among these methodologies, the Phadebas® amylase test is the most common method of choice for forensic identification of saliva. The enzyme α-amylase is found in very high levels in saliva. Its activity in stains is used as an indicator for the presence of saliva. The Phadebas® amylase test consists of starch microspheres with a blue dye cross-linked to the starch. In the presence of amylase, the starch is digested, releasing the water soluble dye into solution. Furthermore, fluorescence spectroscopy has also been reported for detection of saliva stains (16).

The purpose of this study was to evaluate the two recent commercially available test devices for forensic identification of saliva stains, namely the SALIgAE-saliva test (Abacus Diagnostics, West Hills, CA) and the RSID-saliva test (Independent Forensics, Hillside, IL). The former is a colorimetric test while the latter is a membrane strip test based on immunochromatography principle. These two methods were evaluated for their sensitivity and specificity with reference to the Phadebas® amylase test. Casework samples were also employed to verify if the two methods could be adopted for identification of saliva for routine forensic samples.

Materials and Methods

Test Assays

Phadebas® amylase test was used in this study. The samples, fabric (5 × 5 mm) or swab (one-quarter) or liquid (100 μL), were placed in 15 mL sterile centrifuge tubes with 4 mL distilled water. One tablet was added to each tube, vortexed and placed in a 37°C water bath for 30 min. One milliliter of 0.5 M NaOH was added to each tube, which was then centrifuged for 5 min at 700×*g*. A blue coloration in the supernatant indicates a positive result, while a clear supernatant indicates a negative result.

Two tests, namely Rapid Stain Identification (RSID)-saliva test and SALIgAE-saliva test were employed in this study. The RSID-saliva test utilizes monoclonal anti-human salivary α-amylase antibodies in an immunochromatographic membrane assay technology. The samples, fabric (5 × 5 mm) or swab (one-quarter), were extracted in 200 μL TE buffer (10 mM Tris-HCl, 0.1 mM EDTA, pH 8.0) in a 1.5 mL microcentrifuge tube for 1 h at room temperature. After extraction, 20 μL was mixed with 80 μL TBS+ running buffer provided by the manufacturer and applied to the sample window of the device. The test results were read after 10 min. Two lines appear for a positive result whereas one control line

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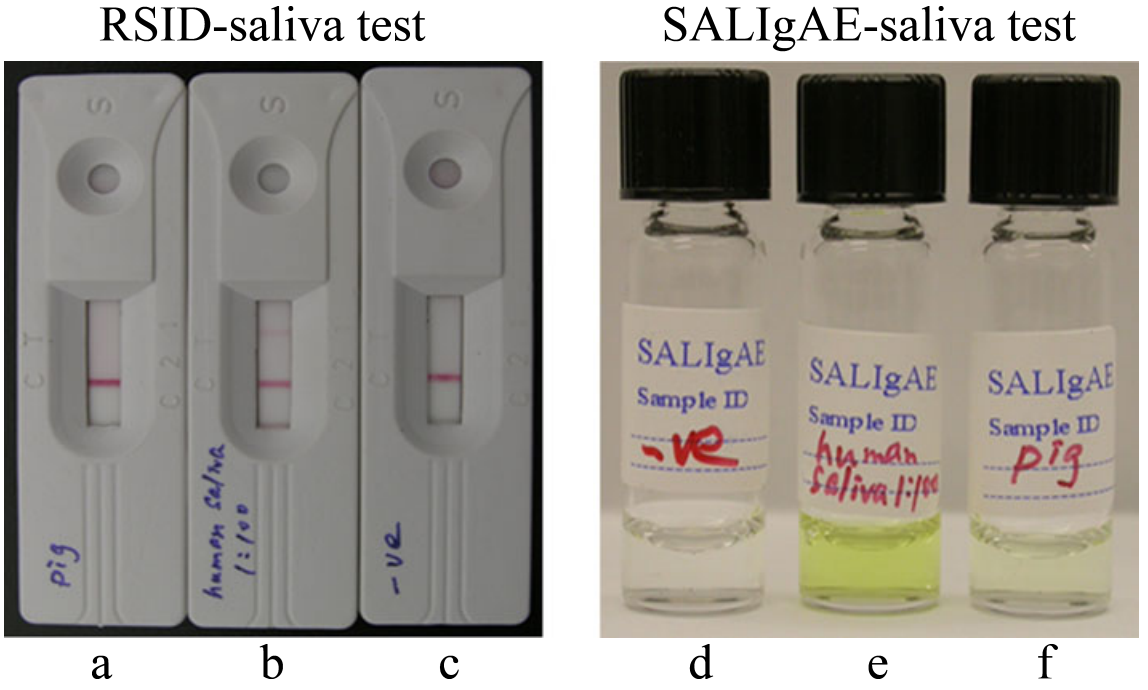

FIG. 1—*Positive and negative results obtained from the RSID-saliva test and the SALIgAE-saliva test. Samples applied are (a) 100 μL of a 100-fold diluted whole pig saliva; (b) 100 μL of a 100-fold diluted whole human saliva; (c) 100 μL of the supplied TBS+ buffer; (d) 8 μL of sterile water; (e) 8 μL of a 100-fold diluted whole human saliva; (f) 8 μL of a 100-fold diluted whole pig saliva. In the RSID-saliva test, a positive result is revealed by the presence of two lines, one in the test and the other in the control regions while a negative result is indicated by one line in the control area only. In the SALIgAE-saliva test, a positive result is indicated by a yellow color change from the colorless test solution whereas a negative result is indicated by the unchanged colorless test solution.*

appears for a negative result. The control line must appear for a valid test (Fig. 1). On the other hand, the mechanism of the SALIgAE-saliva test has not been disclosed. It is a colorimetric test in that the colorless solution changes to yellow in the presence of saliva. After extraction, 8 μL of an extract was aliquoted into the colorless test solution. The test results were read after 10 min. A yellow color in the test solution is interpreted as a positive result while no change in color (colorless) is considered to be a negative test result (Fig. 1). Liquid samples such as saliva, semen, and urine were mixed with TBS+ for the RSID-saliva test or sterile ddH_2O for the SALIgAE-saliva test. One hundred microliters (20 μL extract + 80 μL TBS+) and 8 μL were used for the RSID-saliva test and the SALIgAE-saliva test, respectively.

Human saliva and distilled water were used as positive and negative controls respectively.

Samples

Human saliva, blood, urine, semen, vaginal secretion, breast milk, sweat, and feces were obtained for this study. Human saliva, semen, and urine samples were used either in liquid form directly or as dried stains (fabric or swab) after being placed onto sterile fabric or cotton swab, and air-dried. The other samples were prepared as stains. All these samples were frozen at −20°C before use.

Saliva samples from pig, rat, rabbit, guinea pig, mouse, hamster, dog, and cat were used in this study. One hundred microliters of each animal saliva sample was pipetted to a sterile cotton swab, which was then dried overnight at room temperature. One-quarter of the swab was used as a dried stain.

Extracts of fruit and vegetable including apple, sweet corn, red carrot, green radish, and cabbage were used in this study. Plant extracts were prepared by manual homogenization with sterile ddH_2O for 10 min at room temperature. The supernatants were used as extracts.

Lyophilized samples of human salivary α-amylase (Sigma, St. Louis, MO, A1031) and human pancreatic α-amylase (Sigma, A9972), liquid samples of bacillus α-amylase (21 mg/mL, Sigma, A3403) and sweet potato β-amylase (43 mg/mL, Sigma, A7005), powdered samples of porcine pancreatic α-amylase (Sigma, A3176) and barley β-amylase (Sigma, A7130) were purchased for this study. Lyophilized human salivary α-amylase and pancreatic α-amylase were reconstituted with sterile water to a concentration of 10 mg/mL and 1 μg/μL respectively according to the manufacturer's instructions. Liquid samples were used directly. Porcine and barley samples were dissolved in sterile water at a concentration of 100 mg/mL.

A total of 35 samples were obtained from actual sexual assault cases where no oral intercourse was involved. Included in these samples were buccal swabs, vaginal swabs, penile swabs, liquid semen inside condoms and on clothing, which were stored at −20°C before use.

A total of 15 non-casework samples were employed and included (i) beverage bottles and cans (samples were taken by sterile moistened cotton bud swabs from the openings of the containers after the beverages were consumed); (ii) cigarette butts (5 × 5 mm filter tips were cut out after the cigarettes were smoked); and (iii) skin swabs (saliva was deposited on human skin by licking and allowed to dry in air. The saliva was collected with sterile moistened cotton bud swabs, which were then air-dried before used).

Determination of Sensitivity

Whole human saliva and human salivary α-amylase (Sigma, A1031) were used to determine the sensitivity of the three saliva

detection tests. The whole human saliva and the human salivary α-amylase (10 mg/mL) were prepared at the following dilutions: 1/100, 1/1000, 1/2000, 1/5000, 1/10,000, 1/20,000, and 1/50,000. The equivalent amounts of human saliva for these dilutions are 10, 1, 0.5, 0.2, 0.1, 0.05, 0.02 nL per μL whereas those of the human salivary α-amylase for these dilutions are 100, 10, 5, 2, 1, 0.5, 0.2 ng per μL. The normal range of alpha amylase found in human saliva is 0.2–6.4 mg/mL (11,12,17).

Body Fluid Interference

Mixed body fluid stains were prepared by adding human saliva to semen, vaginal secretion, blood, and sweat stains. The mixed stains were air-dried and tested for saliva using the two tests in order to study the possible interference of these body fluids with human saliva detection.

Results and Discussions

Sensitivity

The manufacturer of the RSID-saliva test states that 1 μL human saliva can be detected while that of the SALIgAE-saliva test claims that the test device is able to detect trace levels of saliva. Human saliva and lyophilized human salivary amylase, which was reconstituted with sterile water to a concentration of 10 mg/mL, were diluted to determine the sensitivity of the two tests. While the RSID-saliva test could detect human saliva up to a 10,000-fold dilution and human salivary amylase up to a 20,000-fold dilution, the SALIgAE-saliva test could detect both human saliva and human salivary amylase up to a 2000-fold dilution. These two tests were more sensitive in detecting saliva than the Phadebas® amylase test, which could only detect both human saliva and human salivary amylase up to a 100-fold dilution (Table 1).

The detection limits of the RSID-saliva test, the SALIgAE-saliva test, and the Phadebas® amylase test equate to 10 nL (20 μL × 1/10,000), 4 nL (8 μL × 1/2000), and 1000 nL (100 μL × 1/100), respectively for human saliva and 10, 40, and 10,000 ng, respectively for human salivary amylase. The RSID-saliva test and the SALIgAE-saliva test are regarded as highly sensitive tests for detection of saliva since they can detect as little as 10 nL saliva.

TABLE 1—*Sensitivity of the RSID-saliva, the SALIgAE-saliva, and the Phadebas® amylase tests in saliva detection with human saliva and human salivary α-amylase.*

	Dilutions						
	1:100	1:1000	1:2000	1:5000	1:10,000	1:20,000	1:50,000
RSID-saliva test							
Human saliva	+	+	+	+	+	−	/
Human salivary α-amylase	+	+	+	+	+	+	−
SALIgAE-saliva test							
Human saliva	+	+	+	−	/	/	/
Human salivary α-amylase	+	+	+	−	/	/	/
Phadebas® amylase test							
Human saliva	+	−	/	/	/	/	/
Human salivary α-amylase	+	−	/	/	/	/	/

+/−, stands for positive and negative results; /, stands for not determined.

Species and Amylases Specificity

Liquid whole saliva samples were obtained from animals including pig, rat, mouse, rabbit, guinea pig, hamster, dog, and cat. A 100-fold diluted liquid saliva from rat was tested positive and all other 100-fold diluted liquid animal salivas were tested negative in the RSID-saliva test. On the other hand, all 100-fold diluted liquid animal salivas were tested positive in the SALIgAE-saliva test and the Phadebas® amylase test except those from dog and cat. Saliva stains from all these animals were prepared with sterile cotton bud swabs. The saliva stains were extracted and analyzed with the two test devices. The results for all saliva stains were the same as those for liquid saliva (Table 2). Among the test animal salivas, dog and cat salivas are reported to have no salivary amylases activity (14). Amylase transcripts were not detectable in the dog parotid gland (18).

Purified amylases including human pancreatic, porcine pancreatic, and bacillus α-amylases, as well as barley and sweet potato β-amylases were purchased for analyzing the specificity of the three saliva tests in addition to human salivary α-amylase. The RSID-saliva test is also reactive to human pancreatic α-amylase among these purified α-amylases. The SALIgAE-saliva test is also reactive to human pancreatic α-amylase and porcine pancreatic α-amylase, but not bacillus α-amylases. No reaction between these two devices with the barley and sweet potato β-amylases was observed. The Phadebas® amylase test gave positive results for all of these α- and β-amylases (Table 3). β-amylases are normally present in germinating seeds prior to germination.

In order to determine if the RSID-saliva test and the SALIgAE-saliva test are more specific to the salivary or pancreatic amylases, the lyophilized pancreatic α-amylases were diluted serially and analyzed with the two devices. It was found that the RSID-saliva test and the SALIgAE-saliva test could detect human pancreatic amylase up to a 10-fold and a 100-fold dilutions, respectively. In other

TABLE 2—*Species specificity of the RSID-saliva, the SALIgAE-saliva, and the Phadebas® amylase tests with whole saliva from various animals.*

Sample	Whole Saliva	Saliva Stain
RSID-saliva test		
Pig	−	−
Rat	+	+
Mouse	−	−
Rabbit	−	−
Guinea pig	−	−
Hamster	−	−
Dog	−	−
Cat	−	−
SALIgAE-saliva test		
Pig	+	+
Rat	+	+
Mouse	+	+
Rabbit	+	+
Guinea pig	+	+
Hamster	+	+
Dog	−	−
Cat	−	−
Phadebas® amylase test		
Pig	+	+
Rat	+	+
Mouse	+	+
Rabbit	+	+
Guinea pig	+	+
Hamster	+	+
Dog	−	−
Cat	−	−

+/−, stands for positive and negative results.

TABLE 3—*Reactivity of RSID-saliva, SALIgAE-saliva, and the Phadebas® amylase tests with various purified amylases and plant extracts.*

Samples	RSID-Saliva Test	SALIgAE-Saliva Test	Phadebas® Amylase Test
Human salivary α-amylase	+	+	+
Human pancreatic α-amylase	+	+	+
Porcine pancreatic α-amylase	−	+	+
Bacillus α-amylase	−	−	+
Barley β-amylase	−	−	+
Sweet potato β-amylase	−	−	+
Apple extract	−	−	+
Sweet corn extract	−	−	+
Red carrot extract	−	−	+
Green radish extract	−	−	+
Cabbage extract	−	−	+

+/−, stands for positive and negative results.

TABLE 4—*Results of the RSID-saliva and the SALIgAE-saliva tests for body fluids/materials.*

Category	Bodily Fluids/ Materials	No. of Samples	RSID-Saliva Test	SALIgAE-Saliva Test
1	Semen (neat)	7	−	−
2	Vaginal swab without semen	8	−	−
3	Breast milk stain	2	−	−
		1	+	+
4	Blood (neat)—male	3	−	−
	Blood (neat)—female	3	−	−
5	Urine (neat)—male	4	+	+
	Urine (neat)—female	2	+	+
		1	−	−
	Urine stain—male	4	−	−
	Urine stain—female	3	−	−
6	Sweat swab—male	4	−	−
	Sweat swab—female	3	−	−
7	Fecal swab	3	+	+
Total		43		

+/−, stands for positive and negative results.

words, the detection limits of the RSID-saliva test and the SALIgAE for human pancreatic amylases were about 2000 ng (20 μL × 1/10) and 80 ng (8 μL × 1/100) respectively. It appears that RSID-saliva test is more specific to human salivary α-amylase. The RSID-saliva test is about 200 times more sensitive to salivary α-amylase than the pancreatic α-amylase while the SALIgAE-saliva test shows similar sensitivity to the two α-amylases in the present study.

Considerable amino acid sequence similarity exists amongst α-amylases from different sources. The amino acid sequences of the human salivary α-amylase and human pancreatic α-amylase are 94% homologous as predicted from the cDNA sequences of the two proteins, which are 96% homologous themselves (19). Both human salivary α-amylase and human pancreatic α-amylase are very similar to pig pancreatic α-amylases, rat pancreatic α-amylases, mouse salivary, and pancreatic α-amylases (20). The α-amylases between human and porcine pancreas are especially similar (21).

Extracts of fruit and vegetable including apple, sweet corn, red carrot, green radish, and cabbage were all tested negative in the RSID-saliva test and the SALIgAE-saliva test, but positive in the Phadebas® amylase test (Table 3).

The RSID-saliva test appears to be more specific than the SALIgAE-saliva test and the Phadebas® amylase test for human saliva detection in the present study. The RSID-saliva test is only reactive to the purified human pancreatic α-amylases and whole saliva of rat among the tested purified amylases, plant extracts, and animal salivas in this study in addition to human salivary α-amylases in saliva and in purified form. On the other hand, the SALIgAE-saliva test is reactive to a number of amylases other than the human salivary α-amylase. Positive Phadebas® amylase tests were obtained for all tested purified amylases, plant extracts, and animal salivas except those from dog and cat.

Body Fluids/Materials

Having determined the sensitivity and the specificity of the RSID-saliva test and the SALIgAE-saliva test, they were further evaluated by analyzing various types of body fluid. The results are given in Table 4. All seven semen samples, eight samples of vaginal swabs without semen, six blood samples, and seven sweat swabs gave negative results for both the RSID-saliva test and the SALIgAE-saliva test. Six neat urine samples gave positive results for both tests; one of the tested female urine produced negative results for both tests. When urine stains were prepared from the neat urine samples and tested with the two assays, none gave positive results. The levels of α-amylase in dried stains were lower. α-amylase proteins might have been denatured during the stain preparation. Positive results were also obtained from all the three tested fecal swabs for both assays. One of the three breast milk stains yielded positive results for both tests. The results of the tested body fluids/materials of the two assays were in good agreement. Stains of mixed body fluids including semen/saliva, vaginal swab/saliva, blood/saliva, and sweat/saliva were prepared and tested by the two devices. All mixed stains with saliva were tested positive in the two devices.

No detectable amount of α-amylase/saliva was found in semen by the two test devices. These findings were consistent with the previous reports that only very low levels of α-amylase were reported in semen (1,2,5). Amylase level for saliva was reported to be approximately 1000 times greater than that found in semen (1). Salivary α-amylase should account for all amylase level in saliva because it was found that AMY1 gene was exclusively expressed in the salivary glands (22). The negative findings obtained from semen samples suggested that the two test devices could be used for saliva detection in forensic samples with semen. Stains made of semen and saliva were prepared for saliva detection. Semen/saliva stains were all tested positive and responded as well as equivalent saliva-only stains, indicating that semen did not interfere with the saliva detection in the two tests.

Negative results obtained from the eight vaginal swabs suggested that the two tests could be used to identify saliva stains in this type of sample. Positive saliva detection from vaginal swabs probably indicates the involvement of saliva in the sexual activity, for example, oral intercourse. However, high amylase activity was detected in human serous ovarian tumors. Samples taken from the vagina of a patient with human serous-type ovarian tumors might contain a high level of amylase (23). The clinical information of the victim might be required for accurate determination of the presence of saliva in the vaginal samples.

All the blood samples tested in this study were negative in both the RSID-saliva test and the SALIgAE-saliva test. Stains of blood and saliva were prepared and these stains were tested positive in both tests. The sensitivity of the blood/saliva and the saliva-only stains was similar, indicating that the blood did not interfere with

the positive identification of saliva in the two test devices. These results suggested that the two tests could be used for determination of trace amounts of saliva in blood. The two assays might be used to analyze the suspected expirated bloodstains in bloodstain pattern analysis. However, further work in this area is required. It should also be noted that an elevated salivary amylase level was detected in the serum of a patient with hyperamylasemia in lung cancer. Amylase contained in lung tissues was released into the blood stream by some inflammatory process (24). Furthermore, discoloration of test solution by blood in the SALIgAE-saliva test was observed. Careful interpretation of the test result with colored samples should be required. No special precautions on saliva detection with the RSID-saliva test are required since blood does not mask the lines in this device.

Casework Samples

Table 5 summarizes the results of the RSID-saliva test and the SALIgAE-saliva test for 35 real forensic casework samples. The five buccal swabs gave positive results for saliva detection in both assays and none of vaginal swabs, penile swabs, semen samples from condom, and fabric samples with semen gave positive results. The results were consistent with the findings for the body fluid/materials determined in the last section. These samples were taken from forensic cases where no oral intercourse was involved. Vaginal secretions and semen appeared not to contain sufficient amount of α-amylase to give rise to a positive result in the two assays. The negative results obtained in all the tested vaginal swabs with/without semen could probably indicate the feasibility of using the test kits for the forensic identification of saliva in vaginal swabs. Positive saliva detection from vaginal swabs could indicate the presence of saliva, and oral intercourse might be involved.

Unusually high levels of α-amylase in seminal fluid have been reported (1,2). However, it was argued that the high levels of α-amylase in seminal fluid as reported in the literature might simply represent an artifact in the collection process (25). None of the semen samples used in this study gave positive result in the two saliva assays. Positive results obtained for saliva detection can be regarded as a strong indication of the presence of saliva. Since saliva and saliva-stained materials are good sources of DNA for analysis and for DNA typing (26), the potential source of the saliva could probably be identified with positive DNA profiling results.

TABLE 5—*Results of the RSID-saliva and the SALIgAE-saliva tests for 35 forensic casework samples and 15 non-casework samples.*

Category	Descriptions	No. of Samples	RSID-Saliva Test	SALIgAE-Saliva Test
Casework				
1	Buccal swab	5	+	+
2	Vaginal swab without semen	12	−	−
3	Vaginal swab with semen	3	−	−
4	Penile swab without semen	6	−	−
5	Penile swab with semen	2	−	−
6	Semen from condom—retrieved by swab	3	−	−
7	Fabrics with semen	4	−	−
Total		35		
Non-casework				
8	Cigarette butt	5	+	+
9	Saliva on skin	5	+	+
10	Beverage container (can/bottle)	5	+	+
Total		15		

+/−, stands for positive and negative results.

Non-Casework Samples

A total of 15 samples were prepared to simulate casework samples to examine the efficiency in the use of the two assays to detect human saliva. All filters of cigarette butts, all swabs on dried human saliva on skin and the opening of the beverage containers were tested positive for saliva in both assays (Table 5).

Comparison of the Membrane-Strip and Colorimetric Tests for Saliva Identification with Reference to the Phadebas® Amylase Tests

The RSID-saliva test and the SALIgAE-saliva test gave higher sensitivity than the Phadebas® amylase tests. The sensitivity of the Phadebas® amylase tests was determined to be about 1 μL of saliva, which should be good enough to detect saliva stains in forensic casework. However, the Phadebas® amylase test gives a positive result when the product of amylase activity is released (13). In other words, any amylase that is present in plants or micro-organisms capable of catalyzing the hydrolysis of α-1,4-glucosidic linkages, will give a positive result. While the Phadebas® amylase test may not be specific enough for detecting human amylases, both RSID-saliva test and the SALIgAE-saliva test were found in the present study to have a higher specificity for amylases. In particular, the RSID-saliva test reacted only with human amylases and amylases from rat saliva, but not with any other sources of amylases tested. The two tests are also not reactive to all plant extracts tested in this study compared with the positive results obtained for these plant extracts with the Phadebas® amylase test (13,14). The specificity of the two devices is therefore considered much better than the Phadebas® amylase tests.

Although the RSID-saliva test and the SALIgAE-saliva test offer alternative methods of saliva identification with higher sensitivity and specificity, they both cannot be used as a searching device like Phadebas® amylase test. The Phadebas® amylase test remains as the primary presumptive screening test in searching for potential saliva stains on articles of evidence. The quick, qualitative Phadebas® amylase test is easy to use. The Phadebas® impregnated paper can be placed into contact with an article of evidence, and the positive Phadebas® tested areas will leave a kind of "image" on the spotty paper. These areas can then be subjected to more rigorous tests. These areas can further be tested with the two assays during the DNA extraction process. An aliquot can be taken from the extraction supernatant for the two assays to obtain a more accurate result on saliva detection. The Phadebas® spotty paper method is an indispensable tool for localizing saliva stains on a large surface such as a piece of clothing (27).

Conclusion

This study demonstrates that the RSID-saliva test and the SALIgAE-saliva test are effective in forensic detection and identification of saliva. The membrane test strip and the colorimetric tube test are easy to perform and the result can be obtained in 10 min. While the RSID-saliva test and the SALIgAE-saliva test show a higher sensitivity and specificity than the Phadebas® amylase test for the detection of amylase, they are not without their limitations. In addition to human salivary amylases, the RSID-saliva test is reactive to the purified human pancreatic amylases and whole

saliva of rat. The SALIgAE-saliva test is reactive to a number of amylases other than the human salivary-amylase. The present study further substantiates the need of conducting an internal validation to define the limitations of a procedure before a forensic laboratory adopting a method for casework analysis. The RSID-saliva test and the SALIgAE-saliva test allow rapid detection of saliva, offering high sensitivity and specificity. With a clear understanding of the limitations of these procedures, the RSID-saliva test and the SALIgAE-saliva test could each be an effective tool in forensic saliva detection, supplementing the indispensable screening procedure offered by the Phadebas® amylase test.

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