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Role of Saliva in Dental Practice – A Review

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ABSTRACT

As a diagnostic fluid, saliva offers distinctive advantages over serum because it can be collected non-invasively by individuals with modest training. Furthermore, saliva may provide a cost-effective approach for the screening of large populations. Gland-specific saliva can be used for diagnosis of pathology specific to one of the major salivary glands. Whole saliva, however, is most frequently used for diagnosis of systemic diseases, since it is readily collected and contains serum constituents. Analysis of saliva may be useful for the diagnosis of hereditary disorders, autoimmune diseases, malignant and infectious diseases, and endocrine disorders, as well as in the assessment of therapeutic levels of drugs and the monitoring of illicit drug use.

INTRODUCTION

Human saliva plays an important role in the health of the oral cavity and of the body as a whole. It is a complex fluid that is actively secreted by the major and minor salivary glands. It provides a window to the physiological and pathological state of the body as a whole. Salivary diagnostics is an emerging field that has tremendous potential in clinical applications because its collection is non-invasive and in contains a wide spectrum of analytes, which can serve as biomarkers for assessment of oral and systemic health. This review intends to give an overview of this exciting area of salivary research.

Saliva as Diagnostic Fluid

Salivary diagnosis is an increasingly important field in dentistry, physiology, internal medicine, endocrinology, pediatrics, immunology, clinical pathology, forensic medicine, psychology and sports medicine [1] A growing number of drugs, hormones and antibodies can be reliably monitored in saliva, which is an easily obtainable, non-invasive diagnostic medium [2,3,4]. Thus, salivary diagnosis is anticipated to be particularly useful in cases where repeated samples of body fluid are needed but where drawing blood is impractical, unethical, or both. Salivary concentrations of drugs and hormones also represent the free fractions of serum in many instances, with good correlations with the respective total concentrations in serum. Multiple specimens of saliva for steroid hormone analysis can be easily collected by the patient, at home, to monitor fertility cycles, menopausal fluctuations, stress and other diurnal variations [5].

Salivary antibody levels can be determined to screen for infectious diseases. Anti-HIV antibody immunocapture assays have also been developed and tested for saliva, which could be useful in high-risk groups under field conditions in developing countries [6]. Salivary assays have been used for monitoring of hepatitis A, B and C, measles, Epstain-Barr virus, rubella, parvovirus B 19, human herpesvirus 6, *Helicobacter pylori* and rotavirus infection [4]. In addition to measuring antibody, it is possible to identify a number of viral antigens in saliva, for example mumps and cytomegalo virus. Saliva has also proven to be a convenient source of host and microbial DNAs [7].

There has been growing interest in the use of saliva in pharmacokinetic studies of drugs and in therapeutic drug monitoring in a variety of clinical situations. It has been suggested that drug levels in saliva reflect the free, non-protein-bound portion in plasma and

hence may have a greater therapeutic implication than the total blood levels [4]. Lipid solubility is a determining factor in saliva excretion of drugs, and the degree of acidity and basicity of a drug will determine its salivary/plasma ratio. The salivary flow rate, pH, sampling conditions, contamination and many other pathophysiological factors may influence the concentrations of drugs in saliva [8]. Drugs currently monitored in saliva include anticonvulsants, theophylline, salicylate, digoxin, anti-arrhythmic drugs, lithium, benzodiazepines, amitriptyline, chlorpromazine, methadone, ethanol, marijuana, cocaine and caffeine[9]. It has become apparent that many systemic diseases affect salivary gland function and salivary composition. Studies of the effects of systemic diseases on salivary variables have been valuable in understanding the pathogenesis of the diseases, but their use as diagnostic markers has been limited.

Human saliva contains a large number of enzymes derived from the salivary glands, oral microorganisms, crevicular fluid, epithelial cells, and other sources. However, the use of whole saliva enzymes for diagnostic purposes has been more difficult than the use of serum enzymes. It has been difficult to standardize saliva collection methods and enzyme analytical procedures so that direct comparisons between different laboratories would be possible. Interpretation of results has also proved to be difficult. However, various studies have been made to find correlations between diseases or clinical situations and salivary enzyme levels [10].

Saliva is essential for alimentation, remineralization of teeth, and the protection and lubrication of oral mucosal tissues.[4] Measurement of the patient's saliva flow is of primary importance in oral medicine and dentistry [11]. For many years dental investigators have been exploring changes in salivary flow rate and composition as a means of diagnosing and monitoring a number of oral diseases. It has even been suggested that analysis of saliva may also offer a cost-effective approach to the assessment of periodontal diseases in populations, even though no specific salivary marker of periodontal disease activity has been found so far [12].

A great number of studies with conflicting results have been published regarding various individual salivary agents and their possible association with oral health, particularly dental caries. However, it appears that no single chemical agent is much more important than others. Many of the various defense factors show additive or even synergistic interactions against oral pathogens [13].

Diagnostic Tests for Normal Dental Practice

Saliva is well adapted to protection against dental caries. The buffering capacity of saliva, the ability of saliva to wash the tooth surfaces and to control demineralization and mineralization, the antibacterial activity of saliva and perhaps other mechanisms all contribute to its essential role in the health of the teeth. Knowledge of the functional properties of saliva and of its separate components may permit a better assessment of dental caries susceptibility [14].

Measurement of salivary flow is an invaluable diagnostic tool in determining the prognosis of alternative treatment plans [15]. In modern dental practice, diagnostic salivary measurements, at least salivary secretion rate and buffering capacity, should be used to supplement the anamnestic information and clinical findings with regard to prevention of dental caries. In order to gain reliable standardized results from the diagnostic tests, detailed instructions should be provided and followed by the dentist and patient [16].

However, since caries lesions are the result of a multifactorial disease, assessment of a few salivary factors is not sufficient unless they are of overriding importance, which may occur in an individual patient [17]. Salivary bacterial counts, for example mutans streptococci and lactobacillus dip slide tests, are widely used in clinical practice in caries risk assessment. The current tests may be useful for estimating caries activity due to bad dietary habits, and establishing the presence of infection and salivary yeasts for the determination of the patient's medical condition [18]. However, these tests may be limited in their applicability in the assessment of caries activity and in caries prediction [19]. However, they can be effective in a group of persons with high or low caries experience [20].

Mutans streptococci are acidogenic and aciduric, and can produce extracellular glucans and adhere to tooth surfaces. Several methods are available to measure the levels of mutans streptococci in saliva and in plaque. The so-called ´Strip Mutans test´ is based on the ability of mutans streptococci to grow on hard surfaces, and it has been developed for chair-side use [21]. Other chairside tests have also been developed [22].

Lactobacilli are associated with caries. They are more dependent on retentive sites being available in high numbers, and hence lactobacillus counts have been used to predict the increment of new caries lesions [23]. The standard laboratory method of determining the number of lactobacilli includes the use of selective medium, Rogosa SL-agar. Chair-side methods for lactobacilli have also been developed, since the ´Dentocult LB´ method in 1975 [24].

It has recently been shown that low secretion rate of saliva and the high scores of lactobacilli and *Streptococcus mutans* have a significant influence on complications of fixed metal ceramic bridge prostheses and this should be taken into consideration in choosing patients for prosthetic treatment with fixed prosthodontics [25]. Since salivary flow and its composition is essential in the protection and lubrication of oral mucosal tissues, salivary tests have also significant predictive value in prosthodontic treatment planning. Successful

management of complete and removable partial dentures is complicated by a reduction in salivary flow [26]. It hasbeen suggested that salivary tests should be performed and analyzed before planning an extensive and expensive restorative therapy or orthodontic treatment and on aroutine basis with geriatric patients.

Salivary Flow

Diminished salivary output can have deleterious effects on oral and systemic health [27]. Unstimulated whole saliva is the mixture of secretions which enter the mouth in the absence of exogenous stimuli such as tastants or chewing. Several studies of unstimulated saliva flow rates in healthy individuals have found the average value for whole saliva to be about 0.3 ml/min. Values below 0.1 ml/min are considered as hyposalivation, and values between 0.1-0.25 ml/min low[16]. The normal range is very large and includes individuals with very low flow rates who do not complain of a dry mouth [28]. There is significant difference between genders in unstimulated flow rate. Xerostomia (dry mouth) is the subjective feeling of oral dryness. It is generally accompanied by salivary gland hypofunction and a severe reduction in the secretion of unstimulated whole saliva, but xerostomia is not necessarily reflected in the actually measured flow rates [29].

Unstimulated saliva is usually collected with the patient sitting quietly, with the head bent down and mouth open to allow the saliva to drip from the lower lip into a sampling tube (the so-called draining method). The other most commonly used techniques for measuring unstimulated saliva are the spitting method, suction method and swab method [17]. The factors affecting unstimulated saliva flow rate are degree of hydration, body position, exposure to light, previous stimulation, circadian rhythms, circannual rhythms, and drugs. Less important factors are age, body weight, psychic effects, and functional stimulation [30].

Stimulated saliva is secreted in response to either masticatory or gustatory stimulation, or to other less common stimuli such as activation of the vomiting centre. A wide variation among individuals has been found. Men have higher flow rates than women. The factors affecting the flow of stimulated saliva are nature of stimulus, vomiting, smoking, gland size, gag reflex, olfaction, unilateral stimulation, and food intake [11]. Reduced salivary flow may cause a variety of mostly unspecific symptoms to the patient, so the establishment of salivary flow rates is of primary importance in oral medicine and dentistry. Saliva influences caries attacks mainly by its rate of flow and its fluoride content. The salivary flow rate influences to a high degree the rate of oral and salivary clearance of bacterial substrates [31].

Buffering Capacity of Saliva

Salivary buffering capacity is important in maintaining a pH level in saliva and plaque. The buffer capacity of unstimulated and stimulated whole saliva involves three major buffer systems. The most important buffering system in saliva is the carbonic acid / bicarbonate system. The dynamics of this system is complicated by the fact that it involves the gas carbon dioxide dissolved in the saliva. The complete simplified equilibrium is as follows:

$$CO2 + H2 O \square H2 CO3 \square HCO3^{-} + H+$$

The increased carbonic acid concentration will cause more carbon dioxide to escape from the saliva. The saliva bicarbonate increases the pH and buffer capacity of saliva, especially during stimulation [32].

The second buffering system is the phosphate system, which contributes to some extent to the buffer capacity at low flow rate. The mechanism for the buffering action of inorganic phosphate is due to the ability of the secondary phosphate ion, $HPO4^{2-}$, to bind a hydrogen ion and form an $H2PO4^{-}$ -ion. The third buffering system is the protein system. In the low range of pH the buffering capacity of saliva is due to the macromolecules (proteins) containing H-binding sites.

The bicarbonate concentration is strongly dependent on secretion rate. Since bicarbonate is the chief determinant of the buffer capacity, there is an interrelationship between pH, secretion rate and salivary buffering capacity [16].

Various methods have been used to measure the salivary buffer capacity, including titration under oil, titration while open to air and titration with CO_2. Values obtained for buffer capacity in different studies are not comparable. However, final pHs under 3.5 for unstimulated saliva and 4.0 for stimulated saliva are considered low. From a practical point of view, the Dentobuff method has been developed to assess the buffering capacity in dental practice. Based on the color change of the indicator paper, the buffering capacity is assessed in comparison with a color chart. The Dentobuff method to assess the salivary buffering capacities has been shown to be valid [22].

Salivary Viscosity

The lubricating action of saliva is important for oral health. It facilitates the movements of the tongue and the lips during swallowing and eating and is important for clearly articulated speech. The efficacy of saliva as a lubricant depends on its viscosity and how it changes with shear rate. The shear rate can obtain high values, e.g. 160 and 60 1/s during speaking and swallowing, respectively [33].

Tribology is the science and practice of friction, lubrication, and wear applied to surfaces in relative motion [34]. Rheology is the science associated with the deformation of materials subjected to stresses and forces. The rheological aspect includes viscosity and viscoelasticity. Saliva possesses specific rheological properties as a result of its chemical,physical and biological characteristics, these properties being essential for maintaining balanced conditions within the oral cavity [35].

It has been found that salivary viscosity is greatly influenced by pH and calcium [36]. Increased salivary viscosity may also be associated with an increase in dental caries, although it is difficult to examine flow rate and viscosity independently from each other [37]. The apparent viscosity contributes to the rheological properties of saliva, and the elastic properties could be important as well [38]. Salivary viscosity is also suggested to contribute to denture retention. Retention of dentures is a dynamic issue dependent on the control of the flow of the interposed fluid and thus its viscosity and film thickness. In this, the most important concerns are good base adaptation and border seal of the prosthesis, so that full advantage is taken of the saliva flow-related effects. Alterations in salivary composition appear to be reflected in its viscosity and in oral complaints [39].

Salivary Immunoglobulins

Salivary secretory immunoglobulins (sIgA and sIgM) originate from immune cells which home to the salivary glands, and are produced as a host response to an antigenic stimulus [40]. The immunoglobulins may be directed at specific bacterial molecules, including cell surface molecules such as adhesins, or against enzymes. By binding to such molecules, adhesion of specific bacteria to oral surfaces may be blocked, so preventing colonisation by the affected species [41]. Several studies have confirmed that sIgA is mainly dimeric rather than monomeric, and it is associated with an epithelial glycoprotein called SC (secretory component) [42]. At least 95% of the IgA normally appearing in saliva is produced by the local gland-associated immunocytes rather than being derived from the serum.

Salivary Non-immunoglobulin Proteins

Salivary lysozyme hydrolyses specific bonds in exposed bacterial cell walls, causing cell lysis and death. It is also known that lysozyme contributes to mucosal protection and modulates Candida populations in the oral cavity [43]. Peroxidases, salivary peroxidase and myeloperoxidase, catalyze a reaction involved in the inhibition of bacterial growth and metabolism, and the prevention of hydrogen peroxide accumulation, thus protecting proteins from the action of oxygen and reactive oxygen species [44]. Salivary lactoferrin has antibacterial activity. Lactoferrin binds iron, making it unavailable for microbial use. Lactoferrin, in its unbound state, also has a direct bactericidal effect on some microorganisms including *Streptococcus mutans* strains [45]. Histatins shows anti fungal activity against Candida Albicans. Histatins have been shown to be tannin-binding proteins in human saliva. Histatins also bind to enamel surfaces and hydroxyapatite in a complex manner [46]. Salivary agglutinins are glycoproteins which have the capacity to interact with unattached bacteria, resulting in clumping of bacteria into large aggregates which are more easily flushed away by saliva and swallowed. Bacterial binding to salivary proteins may in part account for individual differences in the colonization of tooth surfaces. Agglutinins induce the aggregation and clearance of streptococci from the oral cavity and are also important modulators of initial plaque formation [47].

CONCLUSION

The ongoing development of salivary diagnostics and the ease of collection of saliva are resulting in a shifting paradigm in diagnostic and treatment planning approaches in many areas of medicine and dentistry.

REFERENCES

1. Mandel ID. Salivary diagnosis: more than a lick and a promise. J Am Dent Assoc. 1993;124:5-7.
2. Mandel ID. Sialochemistry in diseases and clinical situations affecting salivary glands. Crit Rev Clin Lab Sci. 1980;12:321-66.
3. Mandel ID. The role of saliva in maintaining oral homeostasis. J Am Dent Assoc. 1989;119: 298-304.
4. Mandel ID. The diagnostic use of saliva. J Oral Pathol Med.1990; 19:119-25.
5. Hofman LF. Human saliva as a diagnostic specimen. J Nutr. 2001;131:1621S-5S.
6. Pasquier C, Bello PY, Gourney P, Puel J, Izopet J. A new generation of serum anti-HIV antibody immunocapture assay for saliva testing. Clin Diagn Virol. 1997; 8:195-7 .
7. Tabak LA. A revolution in biomedical assessment: the development of salivary diagnostics. J Dent Educ. 2001;65:1335-9.

8. Liu H, Delgado MR. Therapeutic drug concentration monitoring using saliva samples. Focus on anticonvulsants. Clin Pharmacokinet. 1999;36:453-70.
9. Knott C. Excretion of drugs into saliva. In: Human saliva: clinical chemistry and microbiology.Volume I. Editor Tenovuo JO. CRC Press, Boca Raton, Florida, USA, 1989 Pp. 177-201.
10. Mäkinen KK. Salivary enzymes. In: Human saliva: clinical chemistry and microbiology. Volume II. Editor Tenovuo JO. CRC Press, Boca Raton, Florida, USA, 1989. Pp. 93-120.
11. Ghezzi EM, Lange LA, Ship JA. Determination of variation of stimulated salivary flow rates. J Dent Res. 2000; 79:1874-8.
12. Kaufman E, Lamster IB. Analysis of saliva for periodontal diagnosis-a review. J Clin Periodontol. 2000;27:453-65.
13. Tenovuo J. Antimicrobial function of human saliva-how important is it for oral health? Acta Odontol Scand. 1998;56:250-6.
14. Dowd FJ. Saliva and dental caries. Dent Clin North Am. 1999;43:579-97.
15. Strahl RC, Welsh S, Strechfus CF. Salivary flow rates: a diagnostic aid in treatment planning of geriatric patients. Clinical Preventive Dentistry. 1990;12:10-2.
16. Tenovuo J, Lagerlöf F. Saliva. In: Textbook of clinical cardiology. Second edition. Editors Thylstrup A and Fejerskov O. chapter 2. Munksgaard, Copenhagen, Denmark, 1994. Pp. 17-43.
17. Birkhed D, Heintze U. Salivary secretion rate, buffer capacity, and pH. In: Human saliva: clinical chemistry and microbiology. Volume I. Editor Tenovuo JO. CRC Press, Boca Raton, Florida, USA, 1989 Pp. 25-74.
18. Larmas M. Saliva and dental caries: diagnostic tests for normal dental practice. Int Dent J. 1992; 42:199-208.
19. Pinelli C, Serra MC, Loffredo LC. Efficacy of a dip slide test for mutans streptococci in caries risk assessment. Community Dent Oral Epidemiol. 2001;29:443-8.
20. Bowden GH. Does assessment of microbial composition of plaque/saliva allow for diagnosis of disease activity of individuals? Community Dent Oral Epidemiol. 1997;25:76-8.
21. Jensen B, Bratthall D. A new method for the estimation of mutans streptococci in human saliva. J Dent Res.1989; 68:468-71.
22. Bratthall D, Ericsson D. Tests for assessment of caries risk. In: Textbook of clinical cariology.Second edition. Editors Thylstrup A and Fejerskov O. Munksgaard, Copenhagen, Denmark, 1994,Pp. 333-53.
23. Smith SI, Aweh AJ, Coker AO, Savage KO, Abosede DA, Oyedeji KS. Lactobacilli in human dental caries and saliva. Microbios. 2001;105:77-85.
24. Larmas M. A new dip-slide method for the counting of salivary lactobacilli. Proc Finn Dent Soc. 1975;71:31-5.
25. Näpänkangas R, Salonen-Kemppi MA, Raustia AM. Longevity of fixed metal ceramic bridge prostheses: a clinical follow-up study. J Oral Rehabil. 2002;29: 140-5.
26. Massad JJ, Cagna DR. Removable prosthodontic therapy and xerostomia. Treatment considerations. Dent Today. 2002; 21:80-2, 84, 86-7.
27. Navazesh M, Christensen C, Brightman V. Clinical criteria for the diagnosis of salivary gland hypofunction. J Dent Res.1992;71:1363-9.
28. Ship JA, Fox PC, Baum BJ. How much saliva is enough? "Normal" function defined. J Am Dent Assoc. 1991;122:63-9.
29. Nederfors T, Isaksson R, Mornstad H, Dahllöf C. Prevalence of perceived symptoms of dry mouthin an adult Swedish population - relation to age, sex and pharmacotherapy. Community Dent Oral Epidemiol. 1997; 25:211-6.
30. Dawes C. Physiological factors affecting salivary flow rate, oral sugar clearance, and the sensation of dry mouth in man. J Dent Res.1987;66:648-53.
31. Lagerlof F, Oliveby A. Caries-protective factors in saliva. Adv Dent Res1994;8:229-38.
32. Bardow A, Moe D, Nyvad B, Nauntofte B. The buffer capacity and buffer systems of human whole saliva measured without loss of CO_2. Arch Oral Biol 2000;45:1-12.
33. Waterman HA, Blom C,Holterman HJ,s-Gravenmade EJ, Mellema J. Rheological properties of human saliva. Arch Oral Biol. 1988; 33:589-96.
34. Schwarz WH: The rheology of saliva. J Dent Res. 1987; 66:660-4.
35. Aguirre A, Mendoza B, Levine MJ, Hatton MN, Douglas WH. In vitro characterization of human salivary lubrication. Arch Oral Biol. 1989;34:675-7.
36. Nordbo H, Darwish S, Bhatnagar RS. Salivary viscosity and lubrication: influence of pH and calcium. Scand J Dent Res. 1984; 92:306-14.
37. Biesbrock AR, Dirksen T, Schuster G. Effects of tung oil on salivary viscosity and extent and incidence of dental caries in rats. Caries Res.1992; 26:117-23.
38. Van der Reijden WA, Veerman ECI, Nieuw Amerongen AV. Shear rate dependent viscoelastic behavior of human glandular salivas. Biorheology.1993; 30:141-52.
39. Chimenos-Kustner E, Marques-Soares MS. Burning mouth and saliva. Med Oral. 2002;7:244-53.
40. Brandtzaeg P, Berstad AE, Farstad IN, Haraldsen G, Helgeland L, Jahnsen FL, et al. Mucosal immunity - a major adaptive defense mechanism. Behring Inst Mitt. 1997;98:1-23.
41. Zee KY, Samaranayake LP, Attstrom R. Salivary immunoglobulin A levels in rapid and slow plaque formers: a pilot study. Microbios. 2001;106; suppl 2:81-7.

42. Seidel BM, Schubert S, Schulze B, Borte M. Secretory IgA, free secretory component and IgD in saliva of newborn infants. Early Hum Dev. 2001; 62:159-64.
43. Samaranayake LP. Nutritional factors and oral candidosis. J Oral Pathol. 1986;15: 61-5.
44. Salvolini E, Martarelli D, Di Giorgio R, Mazzanti L, Procaccini M, Curatola G. Age related modifications in human unstimulated whole saliva: a biochemical study. Aging. 2000;12:445-8.
45. Van de Strate, BW, Harmsen MC, The TH, Sprenger HG, de Vries H, Eikelboom MC, Kuipers ME, Meijer DK, Swart PJ. Plasma lactoferrin levels are decreased in end-stage AIDS patients. Viral Immunol. 1999;12:197-203.
46. Tsai H, Bobek LA. Human salivary histatins: promising anti-fungal therapeutic agents. Crit Rev Oral Biol Med.1998; 9:480-97.
47. Carlen A, Bratt P, Stenudd C, Olsson C, Strömberg N. Agglutinin and acidic proline-rich protein receptor patterns may modulate bacterial adherence and colonization on tooth surfaces. J Dent Res.1998;77:81-90.