Saliva in Diagnosis

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Abstract

Saliva is a complex fluid produced by the major and minor salivary glands and is a mixture of several constituents of non-salivary origin. Saliva as an aid in diagnosis is by no means limited to dental caries, periodontal diseases, HIV seropositivity or cancer. Tests using saliva have made substantial inroads into an array of clinical and research areas such as virology, immunology, microbiology, endocrinology, toxicology, psychiatry and forensics.
A pool of research articles is available on the potential use of saliva as a dignostic tool in medical sciences.
A comprehensive computer-based search using key words(in cochrane and pubmed databases) and hand searches were done for several journals were checked for relevant material related to the study. All articles were considered individually to eliminate non-peer- reviewed articles, those dealing with commercial laser technology, and those considered by the author to be purely opinion articles.
This review article aims at summarizing the variable use of saliva as a diagnostic aid on a daily basis when treating patients.

Key Words Sialometry , sialochemistry , HIV, Sjogren's syndrome

Introduction

Saliva is a complex fluid produced by the major and minor salivary glands [1] containing both organic and inorganic constituents. These constituents generally occur in small quantities varying with changes in flow, yet they continually provide an array of important functions. The multifactorial components within saliva not only protect the integrity of the oral tissues but also provide clues to various local and systemic conditions and disease. A growing number of researchers are finding saliva more useful as a diagnostic tool over blood due to easy and non-invasive collection methods [2]. The ability to measure a wide range of molecular components in saliva and compare them with serum has made it feasible to study microbes, chemicals and immunological markers [3]. The analysis of saliva has two purposes:To identify individuals with various diseases and to follow up the progress of affected individuals under treatment.

Autoimmune Diseases

Sjogren's Syndrome:

Sjogrens syndrome is a condition originally described by Henrik Sjogren in 1933 as a triad consisting of keratoconjunctivitis sicca, xerostomia and rheumatoid arthritis.The main etiologic factor for this disease is considered to be altered immunological response. Salivary gland dysfunction in one of the key manifestations of this disease and thus it seems logical to use the dysfunction of these glands for its diagnosis [4].

Methods for assessing salivary gland dysfunction include

Sialometry- Method for determining flow rate. Various studies have showed reduced SM/SL flow rate in SS. A possible explanation for this appreciably reduced flow rate is early involvement of SM/SL gland [4,5].

Sialochemistry-Chemical analysis of salivary composition.Sjogren's syndrome patients showed elevated levels of Na+ and C1-levels.Phosphate concentration was decreased in SS patients in unstimulated whole saliva and concentration was unaltered in stimulated saliva. The unaltered levels in stimulated saliva indicate a disease independent potassium secretion by duct cells or that potassium is secreted at other sites.Increased amount of anionic proteins, phospholipids and unsaturated fatty acids were observed in SS patients Increase in lactoferrin level and decrease in lysozyme level were seen in stimulated parotid saliva with primary SS. Amylase may be used as a marker/or acinar function. Salivary Kallikrein may serve as a marker for striated duct cell function and elevations found in SS may indicate damage to these cells. In SS albumin levels in saliva are elevated indicating a certain loss of parenchymal integrity [6,7,8].

Increased concentration of secretory IgA has been found in SS patients in view of the lymphocytic glandular infiltration. Marked elevation of IgG in SS patients due to activated local synthesis which is supported by reports detecting increased number or IgG producing plasma cells in the lymphocytic infiltrate of minor salivary gland biopsy specimens in SS patients.The presence of IgM

in saliva of SS patients was inconsistent [8].

Burning Mouth Syndrome(BMS)

Fernandes et al 2009 [9] found out that patients with BMS exhibit decreased morning salivary dehydroepian-drosterone(DHEA) which is a steroid produced in the adrenal cortex, in the gonads, CNS via spinal cord.

Oncology

Because of the anatomic proximity of saliva to both the pre-malignant and malignant oral neoplasms salivary diagnosis could be part of a comprehensive diagnostic panel that will provide improved sensitivity and speficity in the detection of malignant diseases and will assist in monitoring the efficacy of trearment [1].

Molecular markers for head and neck cancers

p53: p53 gene is the most common target for genetic alteration in human tumors. [10] In a study conducted by **Liao et al 2000**[15] through PCR technique through p53 mutation observed mutation at exon 4 codon 63 in 63% of case and majority of base substitutions were C deletions suggesting a potential for clinical application of using gene mutation as markers of oral cancer detection [10,11,12,13,14].

Salivary soluble CD44

CD44 comprises a family of isoforms expressed in many cell types. These isoforms arise from alternative splicing of a variable exon present in CD44 , mRNA.[16] **Franzmann et al** reported detection of head and neck squamous cell carcinoma (HNSCC) when saliva specimen had soluble CD44 levels greater than 2.7ng/mL [15,16].

Hyaluronic acid and Hyaluronidase expression

Concentrations of HA are known to be elevated in several cancers including colon, breast, prostate, bladder and lung. They support metastasis by promoting tumor cell migration, offering protection against immune surveillance and causing a partial loss of contact mediated inhibition of cell growth and migration [17].

Hyaluronidase (HAase) is an endoglycosidase that degrades HA into small angiogenic HA fragments. HA and angiogenic HA fragments stimulate endothelial cell proliferation, adhesion and migration by activating the focal adhesion kinase and MAP kinase pathway. HAase has been shown to alter the expression of CD44 isoforrns, which may also be involved in tumor progression. It is also associated with increased tumor cell cycling. In their study, Elizabeth et al reported 4.9-fold elevated levels of HA in HNSCC patients.

Statherin: Contucci et al 2005 [18], showed decreased statherin levels in oral pre-cancerous and cancerous lesions in comparison to normal healthy controls by means of high performance liquid chromatography (HPLC). This protein has been proved to have a protective effect which can both delay and decrease the level of proliferation induced by any carcinogen [19].

Levels of statherin and truncated cystatin S may be potential risk indicators for development of caries and other oral diseases [20].

Adenosine de arninase [ADA] and 5'-nucleotidase activities

Several studies have found increased ADA activities in cancerous tissues and cells [19,20,21,22]. **Saracoglu et at** reported decreased ADA levels in oral cancer patients

Zhong L.P et al 2007 [23] observed that saliva Cyfra 21-1 concentration and CK19 protein expression increased significantly in OSCC tissues. Saliva testing may be an effective modality for diagnosis and for prognosis prediction of oral cancer, as well as monitoring post therapy status by measuring specific salivary macromolecules, examining proteomic or genomic targets such as enzymes,cytokines,growth factors, metalloproteinases, telomerase, cyto-keratins,and DNA transcripts [24].

Human Papillomavirus (HPV) 16 is present in upto 60% of patients with head and neck squamous cell carcinoma (HNSCC) which helps in favorable prognosis in terms of recurrence and mortality. HPV-16 DNA can be detected in initial salivary rinses of patients and also its presence in follow-up rinses preceded clinical detection of the disease [25].

Genetic heterogenecity

Loss of heterozygosity (LOH) is a term used to describe a condition where the cell becomes homozygous to a mutant allele. Studies using microsatellite markers from different chromosomal arms in HNSCC have shown that alterations at certain regions on chromosomes 3p, 9p, 17p and 18q to be associated with development of these tumors. Different studies have also shown a high incidence of loss of heterozygosity in non-invasive lesions indicating an early association with tumorigenesis [26].

In a study conducted by Naggar et al 2001, showed highest incidence of microsatellite LOH of chromosome 9p, 3p & 17p in 49% of the cases. Analysis of these markers in saliva may allow for rapid, inexpensive and objective assessment of genetic abnormalities at these sites for early detection, screening of individuals at high risk and follow of patients with cancer.

Thus saliva is a readily available source of DNA for genetic analysis and can be frequently used for molecular screening and early detection of these diseases.

Salivary transcriptome diagnostics

Salivary transcriptome diagnostics constitutes a novel clinical approach where a large panel of human RNA's can be reliably detected in saliva by use of microarray technology **(Yang et al 2004 [27])** Seven salivary mRNA biomarkers showed significant elevation in saliva of OSCC patients.

- High up-regulated mRNA - ILS
- Moderate up-regulated mRNA - H3F3A, IL 1 β, S100P
- Low up-regulated mRNA - DUSP 1, OAZ 1, and SAT.

Interleukin-6 levels were found to be up-regulated in saliva in the OSCC patients. The IL-6 level tended to increase before treatment and returned to baseline levels after treatment [28].

Salivary epidermal growth factor (EGF)

The EGF concentration in resting saliva may be a more important determinant because the resting saliva and the substance contained in it create the oral cavity environment for most of the day [29]. The reason of lowered EGF concentration in saliva of oral cancer patients could be attributed to the interrelations between EGF-receptor - (EGFR) and ligands competing for EGFR.In another study by Ino et al, they found a major drop in EGF concentration of resting saliva in oral cancer patients before surgery in comparison to healthy controls. Epstein et al have also found decreased EGF concentration in saliva of oral cancer patients.Thus it is suggested that EGF may play an important role in oral cancer development and determination of their values in saliva could be useful as an important diagnostic aid. Elevated salivary defensin-1 in comparison to healthy controls was also observed [1].

Oxidant and antioxidants: Oral squamous cell carcinoma causes alterations in free radicals and antioxidant levels in plasma and erythrocytes. They cause a decline in activities of antioxidant enzymes which causes greater accumulation of free radicals that further causes structural and functional damages to antioxidant enzymes and impairing their efficiency [30]. It has been found that salivary composition of OSCC is substantially altered with respect to free radical involved mechanisms [31]. This non-invasive, efficient and low-cost approach can be developed as a promising method for population-based screening of cancers and pre-cancers in the oral cavity [32].

Proteomic Analyses Of Saliva: Cancer proteomics encompasses the identification and quantitative analysis of biomarkers, which are subsequently objectively measured indicators that characterize the state of the health of the biologic system being analyzed [33,34,35] . Transcriptome IL8,IL1B,SAT1 and S100P is significantly elevated in OSCC patients [12].

The discovery of breast cancer markers in saliva has offered renewed interest in the potential use of saliva as a diagnostic fluid

Salivary Markers For Diagnosis of Cancers of Other Sites: The use of saliva for detection of other malignancies deals primarily with the identification and quantification of cancer related proteins that were previously discovered to be present in cancer tissue supernatants or elevated in serum of diagnosed cancer patients.[36]

The various commonly used markers are: C-erbB-2,CA-I25,CA-15-3,Kallikrein, EGF,

p-53,Cathepsin-D

C-erb-B2 is the salivary biomarker of choice in detecting breast cancer because they have been shown to be present on the membrane of the ductal epithelium of salivary gland tissues. In a study performed by **Sterkfus et al** (2005) [36] reported that salivary levels of C-erB-2 were significantly higher in benign tumor patients.

CA-125 (Cancer Antigen-125) is a glycoprotein complex that is often used as a marker for ovarian cancer as reported by **Chen et al (1990).**

Various studies have reported the use or **kallikrein** as a diagnostic marker among patients with malignant tumors **(Streckfus et al 2005)** [36]. Navarro et al 1997 demonstrated that EGF concentration were higher in saliva of women with primary or recurrent breast cancer.

Viral Diseases

The antibody response to infection is the basis for many diagnostic tests in virology.Antibodies against viruses and viral components can be detected in saliva and can aid in diagnosis of acute viral infection, congenital infections and reactivation of infection. (Kaufman et al 2002) [1].

Hepatitis A and B: (Kaufman et al 2002) : Acute hepatitis A&B can be diagnosed based on the presence of IgM antibodies in saliva. The ratio of IgM to IgG anti HAV antibody correlated with time interval from the onset of infection. Saliva has also been utilized for screening for HbsAg in epidemiologic studies [1].

Hepatitis C: Hepatitis C virus mainly spreads through sharing needles or 'works' when 'shooting' drugs through needlesticks or sharp exposures or from an infected mother to her baby during birth. Investigation of body fluids by PCR demonstrates HCV-RNA in semen, urine and saliva [37].

Human Herpes Virus: Salivary glands have been proposed as sites for persistent viral infection, because of the high frequency of virus isolation from saliva samples. Thus it has been speculated that HHV-7 shedding in saliva might play a role in virus transmission [38].The frequency of HSV-1 positive saliva samples was higher in patients with severe cytopenia. This finding could be an important susceptibility factor for increased HSV-1 salivary shedding [39].

Cytomegalovirus (CMV): another β-herpesvirinae subfamily member is frequently recovered from saliva of seropositive adults CMV, HHV-6,7& 8 are all detected in saliva of HIV infected patients by PCR technique.

Epstein Barr Virus (EBV): is an orally transmitted human herpes virus that is usually asymptomatic but can cause infectious mononucleosis and certain epithelial cell malignancies. EBV has been isolated from salivary secretion using PCR technique.

Mumps, Measles and Rubella: IgM specific antibody for these viruses can be detected in saliva using radioimmunoassay within six weeks from the onset of infection. Saliva can also be tested for total IgG content by radial immunodiffusion. (Brown et al 1994) [40].

Dengue Virus: It is a mosquito transmitted viral disease. Salivary levels of anti-dengue IgM& IgG demonstrated very high specificity and sensitivity in diagnosis of primary and secondary infection. (Kaufman et al 2002) [1].

Rotavirus Infection: Salivary IgA response was found to be a better marker than the serum response in new born infants. Saliva can also be used to monitor the immune response to vaccination. (Kaufman et a12002 [1]).

Parvovirus B-19: Saliva was found to be a reliable alternative to serum for identification of antibody to parvovirus B-19. (Kaufman et al 2002 [1])

HIV-It is well known that HI V is poorly transmitted through saliva. The reason for this is believed to be the presence of numerous salivary proteins that could hinder viral infection through the oral route [41]. Various observations suggest that anti HIV-1 effects of saliva involve a specific mode of action such as physical blocking via high viscosity or virai lysis aue to low hypotonicity. Salivary antibodies against HIV were identified since alongtime. HIV infection is associated with decreased salivary IgA levels [42]. Presence of specific anti-HIV antibodies (lgA, IgG, IgM) can be readily detected in saliva but at lower concentration than serum, and hence require sensitive methods for their detection.

The first testing system to be used is the OraSure HIV-l oral specimen collecting device which was approved in 1996 [43]. This testing system utilizes methods designed to isolate transudates rich.in IgG. The levels of IgG present in GCF which enters the oral cavity through passive diffusion of serum transudates are considered comparable to serum as an HIV testing medium. IgG antibody to the virus is predominant type of anti-HIV immunolog-lobulin [1].

Promising salivary markers that are identified are: Viral RNA,IgA, T-helper cytokine profiles,Calprotectin,Secretory leukocyte protease inhibitors,Neopterin,B-2 microglobulin

Viral RNA -- Given the value of plasma viral RNA levels in predicting disease course, HIV - t RNA quantitation in saliva would seem a logical alternative to plasma. **Shugars et al 2002,** in his study found that viral RNA levels in whole saliva exceeded plasma levels by at least five fold. The presence of viral RNA at such high concentration in saliva could be because the virus may be actively replicating in oropharyngeal tissues such as salivary glands and tonsils. Further research is necessary to determine the biological implications of high levels of HIV-l shedding in oral cavity to document this finding.

T-helper 1 (Th-l) and T-helper 2 (Th-2) represents 2 distinct immune conditions.**Th 1** - induce cell mediated immune response. **Th 2** - drive antibody production.Studies have suggested that progression to AIDS and susceptibility to mucosal candidiasis are reflected by enhanced Th-2 cytokine receptors. A constitutive mixed Th l/Th2 cytokine expression in whole saliva from healthy uninfected patients contrasts with a dominant Th2 type salivary cytokine profile in infected individuals. However, technical difficulties in measuring cytokine levels in saliva may restrict its value for screening immunological status in HIV / AIDS patients.

Calprotectin is a calcium binding protein. Its levels in parotid saliva are low in HIV infected patients suffering from oral candidiasis. In contrast, calprotectin levels in whole saliva increases with candidal infection in HIV-l infected patients.

Neopterin a pteridine derivative present in body fluids in typically elevated in serum due to immune activation, malignancy, allograft rejection and viral infections. Neopterin levels in oral mucosal transudate (OMT, serum transudate collected from buccal sulcus) but not whole saliva appear to mirror levels in serum, suggesting than OMT-based test for salivary neopterin may show promise as surrogate marker assay.

β-2 microglobulin concentration in OMT show good correlation with serum levels and may be more sensitive in detecting clinical or immunological deteoration. Based on in-vitro studies, B-2 microglobulin and neopterin to some extent are best options for oral-fluid surrogate markers for predicting disease progression.

In conclusion, HIV transmission through saliva is extremely rare and oral HIV testing being simple, safe and well tolerated can be used as an alternative to serum testing for diagnosis of HIV infection.

Bacterial Infections

Saliva samples were tested by PCR assay for the presence of H.pylori DNA and were comparable with the results obtained from biopsy DNA and histopathological analysis of gastric tissues of the infected subjects (Tiwary B et al 2005) [42]. A novel PCR method analyzing the salivary bacterial flora showed that the dominant genera differed among the patients with odontogenic infections when compared from those of the healthy subjects [44,45].

Dental caries and periodontal disease

Saliva can be used in diagnosing caries risk owing to the possibility of detecting the presence of S.mutans & lactobacillus species as well as lactic acid which causes the subsurface demineralization that causes the onset of carious lesion (Puy 2006) [46] Also tests have been developed to conveniently measure P.gingivalis which is associated with periodontal disease [47], saliva based diagnostic tests are now utilized as indicators of patient's risk potential for either disease and the consequent need for aggressive preventive measures (Streckfus et al 2002) [36]. Gopinath v.k 2006 [48] showed that the flow rate,

viscosity, pH, and buffering capacity of saliva in subjects with hingh caries index was significantly lower.

Shigellosis: Evaluation of the secretory immune response in the saliva of children infected with shigella revealed -higher titers o anti-lipopolysaccharide and anti-shiga toxin antibody [1]. **Pneumococcal pneumonia:** can be diagnosed by the detection of pneumococcal C polysaccharide in saliva by ELISA technique.

Lyme disease: is caused by the spirochete Borrelia Burgdorteri and is transmitted to humans by blood feeding ticks. The detection of anti-tick antibody in saliva is a potential biomarker for exposure to tick bites which in turn may serve as screening mechanism for individuals at risk for Lyme disease.

Fungal Infections :

Saliva has been a useful tool in diagnosis of oral candidiasis and salivary fungal counts may reflect mucosal colonization. Saliva from infected individuals especially oral cancer and immunocompromised patients can be collected and the colony count can be evaluated after incubation to detect the presence of infection.Salivary glucose levels are significantly correlated with blood glucose levels in people with diabetes and could therefore be a potentially useful, non-invasive tool to monitor glycemic control [49].

Use Of Saliva In Drug Monitoring:

Although many assumptions for drug level monitoring in saliva are made, the primary requisite for salivary monitoring to be useful in a constant or predictable relationship between drug concentration in saliva and drug concentration in plasma [50].

Drugs monitored in saliva: [50,1]

Therapeutic Drugs:

a) Anticonvulsants:

Carbamazepine concentration in oral fluid ranged from 0.1-5.5mg/L while free CBZ concentration in plasma ranged from 07-4 2mg/L S/P ratios were highly correlated and thus signifies the use of carbamazepine monitoring in oral fluids.

Primidone has an S/P ratio of approx. in part due to its limited protein binding, making oral fluid a god matrix for its therapeutic drug monitoring.

Ethosuximide is not highly protein bound, has a good oral fluid to serum correlation and thus oral fluid monitoring is considered potentially useful.

b) Bronchodilators:

Theophylline is methylxanthine derivative used in treatment of asthma, bronchitis and COPD. In general, correlation coefficients between saliva and plasma theophylline levels were high SIP ratio of about 0.5 was found. Some studies found stimulated saliva samples improved the correlation sufficiently between saliva and free fraction in serum.

c) Anti-microbials

Ofloxacin, an antibacterial used to treat gram positive and negative bacteria was found to have S/P ratio of 0.51. The half lives of otloxacin in oral fluid and plasma were similar, making it suitable for therapeutic monitoring.

Gentamycin is a good candidate for oral monitoring when administered once a day. However, when administered thrice daily no correlation was found between oral fluid and plasma.

Metronidazole concentration in saliva correlated well with levels in serum and half lives of drug samples in saliva were similar to that in serum. Hence measurement of saliva metronidazole could potentially be used to estimate pharrnacokinetics.

Fluconazole, an anti-fungal drug showed saliva concentration that were sufficient to test for compliance and even semi-quantitative predictors of plasma concentration.

Quinine, an anti-malarial drug has similar elimination half lives in saliva and plasma making it a good candidate for non-invasive means monitoring active drug concentration.

Zidovudine, a reverse transcriptase inhibitor used in treatment of HIV shows salivary concentration similar to plasma levels with average S/P ratio of 0.68.

Nevirapine, a-non-nucleoside reverse transcriptase inhibitors also showed strong relation and could be monitored in saliva

d. Anti-pyreticsand analgesics:

Acetaminophen concentrations are highly correlated in plasma and oral fluid specimens.

Paracetamol showed an adequate correlation between oral fluid and a plasma levels

e. Anti-neoplastic:

Doxyrubicin is concentrated in oral fluid with SIP ratio of 2.8 and is considered to be good predictor or free plasma fraction.

Taxol, a highly protein bound chemo-therapeutic agent, has an oral fluid concentration that parallels that found in plasma.

f) Anti-arrythmias

Digoxin levels in saliva could be correlated with plasma levels with free fraction in plasma S/P ratio for digoxin was below 1.0 alter a single dose and above 1 in steady state. Digoxin concentration was relatively high in unstimulated salvia and decreased with stimulation of salivary flow rate. It is suggested that unstimulated saliva reflects the intracellular digoxin concentration stimulated saliva reflects free digoxin concentration of serum.

g) β-blockers: Metaprolol,Oxprenolol

h) Psychoactive drugs:

Lithium is considered to be one of the drugs which is transported into saliva through the mechanisms of active transport. Lithium appears to have an inverse relationship to the rate of flow of salvia and was found to be actively secreted into saliva. This results in concentrations above serum levels

Chlorpromazine. . It was found that chlorpromazine concentration in saliva were 4-50 times higher when compared to plasma

i) Immunosuppresants:

Cyclosporine. Mycophenolic acid, (drug used in organ transplant recipients) concentrations in saliva correlated well with plasma levels. Tacrolimus. Saliva may also provide a practical approach for measuring the unbound concentration of cyclosporine, sirolimus and tacrolimus.

j) Drug abuse Recreational drugs

Ethanol was the first drug to be investigated in oral fluid 'and it appears to reach a higher peak concentration in saliva than in peripheral blood. Ethanol is not ionized in serum, not protein bound and due to its low molecular, weight and lipid solubility diffuse rapidly into saliva. Consequently S/P ratio is generally about 1. (Kaufman 2002) [1].

Marijuana can be detected in saliva by means of radioimiiiunoassay

Hormone Monitoring In Saliva:

Saliva can be analyzed as part of the evaluation of endocrine function. The majority of hormones enter saliva by passive diffusion across the acinar cells [1].

Hormones whose salivary levels reflect serum levels include:

Cortisol. Salivary cortisol is a better measure of adrenal cortical function that serum cortisol and is particularly useful in children(Mandel 1990)2.Considered to be reliable and practical index of hypothalamic pituitary adrenal axis activity in depression, especially in out patients. (Mandel 1990) [2].

Aldosterone: Increased salivary levels were found in patients with primarily aldo-steronism in correlation with serum levels.

Testosterone: Salivary levels were used for the assessment of testicular function.Signi-ficant correlation between salivary and unbound serum levels were observed in normal and hyperandrogenic women. May be useful in behavioural studies of aggression, depression abuse, violent and antisocial behaviour

Estriol Salivary estradiol follows the same trend as serum levels during menstrual cycle. Salivary levels show a very high correlation with serum levels in pregnant women. Suggested as a means for assessment of Feto-placental function.Decreased levels are an excellent means of detecting fetal growth retardation. Progesterone Salivary proge-sterone helps in asse-ssing the func-tional capacity of corpus luteum in normal women and those with defects in the hypothalamic pituitary ovarian axis. Helps in studies of subfertile women. Assessing hormonal changes during adolescence. Useful for prediction of ovulation. Increased estriol-progesterone ratio may be a predictor of pre-term delivery

Insulin: Positive correlation between salivary and serum levels were observed following glucose tolerance test in healthy subjects, NIDDM and obese non-diabetic patient. Such a correlation indicated the potential use of salivary insulin measurement in clinical

practice. (Kaufman et al 2002) [1].

Melanin: Salivary melatonin levels can be used purely as a marker of the free hormone fraction or as a tool in approximation of the concentration in plasma. It is of novel clinical utility in the prognostic monitoring of patients treated fell' melatonin-secreting pineal tumors. The appearance of melatonin in saliva of patients treated for pineal tumors may serve as an early warning of tumor regeneration or metastatic complication and could form a part of long term follow-up in these individuals.

Salivary cortisone levels do not bear any diagnostic significance as their levels are higher than in serum due to conversion of cortisol into cortisone by the enzyme 11β-hydroxysteoid dehydrogenase in the salivary glands. ACTH stimulated basal salivary cortisol levels is an accurate biomarker for the diagnosis of adrenal insuffiency in hypotensive end stage renal disease patients [46].

Expression of Ghrelin in human salivary glands:

Overweight or obesity is a serious health issue,especially for children and adolescents. Ghrelin is a human-growth-hormone releasing peptide with a stimulatory effect on food intake,energy expenditure and fat accumulation and thereby contributing to the control of body weight gain. Parotid and submandibular glands were primary sources of ghrelin produced and released in saliva. Although whether salivary ghrelin could be useful in the diagnosis of obesity remains to be determined,salivary ghrelin might be a possible alternative to serum ghrelin for predicting obesity [51].

Salivary Markers Of Systemic Diseases:

Hereditary Diseases

Cystic fibrosis (CF) is a genetically transmitted disease of children and -young adults which is considered a generalized exocrinopathy. The gene deficit causing cysts fibrosis is present on chromosome 7 and codes for a transmembrane regulating protein called cystic fibrosis transmembrane conductance regulator. The abnormal secretion present in cystic fibrosis caused clinicians to explore the usefulness of saliva for diagnosis of disease.

Changes observed in saliva of cystic fibrosis patients are:

Elevated levels of Ca++ and proteins were found submandibular saliva and resulted in a calcium protein aggregation which caused turbidity of saliva. Elevations in electrolytes, urea and uric acid were observed in saliva of these patients. Elevated levels of Na+ and decreased flow rate were observed in minor salivary glands of these patients.

Decrease protease activities in saliva were observed in this disease. Saliva from these patients was found to contain an unusual form of EGF. The EGF from these patients demonstrated poor biological activity compared to healthy controls.

Coeliac disease is a congenital disorder of the small intestine that involves malabsorption of gluten. Gliadin is a major component of gluten. Serum IgA antigliadin antibodies (AGA) are increased in patients with coeliac disease.Measurement of salivary IgA-AGA has been reported to be a sensitive and specific method for the screening of coeliac disease and for monitoring compliance with the required gluten-free diet.

Cardiovascular Diseases:

Markers in saliva may be useful in postoperative follow up among patients undergoing cardiovascular surgery.Total serum amylase and salivary amylase activity have been determined before and 6 hrs after cardiovascular surgery. The results indicated that if serum amylase levels were low in pre-operative patients with ruptured aortic aneurysms, there was an associated increase in mortality.. 3 Salivary analysis has also provided valuable information to clinical problems involving digitalis toxicity.

Application of classification and regression tree (CART) analysis demonstrates that salivary biomarker measurements used in the absence of electrocardiograph findings confer a high degree of sensitivity and specificity for the identification of acute myocardial infarction [2,3,52].

Psychiatry: The studies have reported that the patients with effective disorders secrete significantly less saliva than normal. (Mandel 1990) [2]. Apparently wide scale use of psychoactive drugs causes xerostomia-as side effect that contributes to reduced flow rate. Measurement of salivary prostaglandins may be a good indicator of major depressive disorders. In the saliva of patients with major depressive disorders the concentrations of immunoreactive prostaglandins were significantly higher than· those of healthy controls. Saliva has been used to monitor therapeutic responses in the treatment of anxiety by measuring salivary levels of 3-methoxy 4-hydroxy phenylglycol (MHPG) [3].

Nephrology: Few studies have employed saliva to screen for renal diseases. They have suggested that salivary creatinine concentration shows a high sensitivity and specificity for determining the presence of renal disease. Further studies are required to confirm such findings and to use for diagnosing renal diseases. Salivary cytokines and secretion rates are significantly decreased in chronic kidney disease(CKD) patients [54].

Respiratory Diseases: Chronic respiratory infection especially in children is often associated with specific secretory IgA deficiency. Determination of complete or near-complete IgA deficiency can readily be made with whole saliva sample aspirated from flood of the mouth or expectorated in older children. Flow rate should also be determined for the most precise measurement of IgA level since salivary IgA concentration varies inversely with flow rate.

Time Of Ovulation: Measurement of electrical resistance in saliva could prove to be a very useful method for determination of ovulation time. A special device which provides a digital measurement of the electrical resistance of saliva has been shown to predict ovulation on average of 5.3 days in advance.

Diagnostic Significance Of Salivary Testosterone Measurement: Saliva as a material for screening biomarkers has several advantages in the study of large scale populations. Since testosterone is not bound to protein in saliva, salivary testosterone determination provides an excellent approach for the evaluation of serum bioavailable or free testosterone. It is measured by liquid chromatography/mass spectrometry (LC-MS) is a non-invasive, reliable substitute for serum calculated free or bioavailable testosterone. For its cost advantage and technical convenience, ELISA for salivary testosterone is now recom-mended for the purpose of screening the androgen bioavailability level [55].

Conclusion

Saliva has been considered as the mirror image of blood since a long time and its various components acts as a mirror of the body's health. Thus saliva could be considered as an attractive diagnostic tool.

The use of saliva as a diagnostic aid is progressing at a rapid pace and will be considered as a popular bodily fluid in the near future.

References

Reference are available on request at editor@healtalkht.com