Review Article

Saliva as a Mirror of the Body Health

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Abstract

Saliva has an old history of study, It is of physiological importance. Saliva has hundreds of components which help detect systemic diseases and also provide biomarkers of health and disease status. Saliva has three major functions: digestion, protection and lubrication. Saliva also functions in maintenance of tooth integrity. Also, the carbohydrates of salivary glycoprotein carry the ABH blood group antigens, expressing in the salivary glands and secreted in the saliva. Saliva is a good indicator of the plasma levels of various substances such as hormones and drugs. The use of saliva as a diagnostic and monitoring method for periodontal diseases and many other infectious diseases has been studied. The relatively easy and non-invasive nature of sample collecting is considered as a simple low-cost stage but the problem is the low concentrations of the markers in comparsion to the plasma.

INTRODUCTION

Saliva has an old history of study but its physiological importance has only been recognized recently [(1)]. Saliva has hundreds of components that may serve to detect systemic diseases or as evidence of exposure to various harmful substances, as well as providing biomarkers of health and disease status. Nowadays, the saliva research field is rapidly advancing due to the use of novel approaches including metabolomics, genomics, proteomics and bioinformatics[(2)].

SALIVA PHYSIOLOGY

Saliva is a complex liquid consisting of secretions from the major and minor salivary glands. As estimated there are 450-750 minor accessory salivary glands, situated on tongue, buccal mucosa and palate except the anterior part of the hard palate and gums [(2-4)].The average daily volume of saliva production is 500-1000 ml. Submandibular glands produce 70% of the overall volume, the parotid glands 25%, and the sublingual glands about 5% [(5)]. The greatest volume of saliva is produced before, during and after meals, reaching its maximum peak at around 12.a.m. and falls considerably at night, while sleeping. Several physiological and pathological conditions can modify saliva production quantitatively, e.g. smell and taste stimulation, chewing, psychological and hormonal status, drugs, age, hereditary , oral hygiene and physical exercise [(3,6-9)] . Each salivary gland contains different regions, the acinar region, which is also referred to as the secretary end piece, and the ductal region. All the salivary fluid is produced from the local vascular bed in the acinar region, and is transported through the duct system, where excess sodium and chloride are reabsorbed and some additional proteins are secreted, and then empties into the oral cavity. A sodium gradient that is actively generated within the secretary end piece causes fluid to flow into the lumen through the tight junctions between the acinar cells [(10,11)]. Saliva is sterile when it leaves the salivary glands. The basis of

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saliva is interstitial fluid from blood capillaries which enters via the salivary gland ducts where it is modified from isotonic into hypotonic fluid [2,5,12]. Every type of salivary gland produces a typical secretion. The parotid glands produce serous fluids, the submandibular glands a sero-mucous secretion, and the sublingual glands secrete mucous saliva. The minor glands produce a viscous secrete [4]. About 99% of saliva is water and the other 1% is a complex of organic and inorganic molecules. Saliva has three major functions: digestion, protection and lubrication. Saliva also functions in maintenance of tooth integrity (Tables 1,2) [2,3,10,13].

Table 1: Important components of saliva with some examples within each component group

Electrolyte	Bicarbonate,calcium,fluoride, phosphate
Enzymes	α-amylase,I nvertase
Mucins	MUC5B(MG1) , MUC7(MG2)
Immunoglobulins	Ig A,IgG,IgM,IgAs
Lipids	Neutral lipids, glycolipids, phospholipids
Non-immunoglobulin Proteins	Histidin-rich proteins, lactoferrin , lysozyme, Peroxidase, proline-rich proteins, salivary agglutinins,statherin

Table 2: The Major Functions of Saliva

Function	
(1) Protective Functions	
Lubrication	Mucins , proline-rich glycoproteins , water
Antimicrobial	Amylase, complement, defensins, lysozyme, lactoferrin, lactoperoxidase, mucins, cystatins, proline-rich glycoproteins, secretory IgA, secretory leukocyte protease inhibitor , statherin , thrombospondin , sialoperoxidase
Growth factor	Epidermal growth factor(EGF), transforming growth factor- alpha(TGF-α) , transforming growth factor-beta(TGF-β) , fibroblast growth factor(FGF) , insulin-like growth factor(IGF-I & IGF-II), nerve growth factor(NGF).
Mucosal integrity	Mucins , electrolytes , water
Lavage/cleansing	Water
Buffering	Bicarbonate , phosphate ione , proteins
Remineralization	Calcium , phosphate , statherin , anioic proline-rich proteins
(2) Food-and speech-related functions	
Food preparation	Water , mucins
Digestion	Amylases , Lipase , ribonuclease , protease , water , mucins
Taste	Water , gustins
Speech	Water, mucins
Prevention of caries	Ca-alkaline , Buffer acids , delivered Fluoride

Saliva is involved in the perception of taste, flavor and texture of foods. The mixing of saliva with food can influence flavor release. The α-amylase enzyme presented in saliva, initiating the digestion of starch. Furthermore, saliva acts as a buffering system. In addition, the large proteins may influence the lubrication [14-18].

The amount and composition of secreted human saliva depends on many factors, i.e. the flow rate, circadian rhythm, type and size of the salivary glands, duration and type of the stimuli, diet, drugs, age, gender, blood type and physiological status. Stimuli for increased salivation include the presence of food or irritating substances in the mouth, and the sight or the smell of food [2,15,16,19-22]. Saliva can be considered as gland specific and whole saliva. Gland specific can be collected directly from

individual salivary glands: parotid, submandibular, sublingual and minor salivary glands [23] .The collection and evaluation of the secretions from the individual salivary glands are primarily useful for the detection of gland-specific pathology, i.e. infection and obstruction. However, whole saliva is most frequently studied when salivary analysis is used for the evaluation of systemic disorders and hormones [24]. Whole saliva (mixed saliva or oral fluid) is a mixture of oral fluids and includes secretions from both the major and minor salivary glands. In addition to several constituents of non-salivary origin, such as gingival crevicular fluid GSF , the area between tooth and marginal free gingiva,oro-naso-pharyngeal secretions, serum and blood derivatives from oral wounds, bacteria and their metabolites, viruses and fungi, desquamated epithelial cells, electrolytes, leukocytes, other cellular components and food debris [4,6,10] .

Subjective and objective functional losses occur in people with hyposalivation. These include dry mouth feeling (xerostomia), difficulty with swallowing food, and an increased susceptibility for opportunistic infections. The last issue points to an active protective role of saliva in maintaining of oral health under normal conditions [25, 26]. Cut off values of 0.1 ml/min for resting whole saliva and 0.5 ml/min for stimulated saliva may be considered as indicative of salivary gland hypofunction [5]. Studies showed that females had higher protein concentrations and lower salivary flow rates than males, and subjects with highest DMFT also have the highest unstimulated flow rates [27]. Mucin, the major organic component of submandibular/ sublingual saliva, provide lubrication. They may also bond to toxins, agglutinate bacteria, interact with host cells, and are important components of the acquired pellicle and plaque matrix [3,22,28].

BUFFERING CAPACITY OF SALIVA

Saliva acts as a buffering system. The buffering effect of saliva is largely due to bicarbonate/carbonate ions, and to a lesser extent to phosphate-ions and proteins present in saliva, neutralizing acids ingested or produced by microorganisms in the mouth [29,30]. The buffer activity is usually assessed immediately after collection of saliva sample. Several studies indicate that buffer capacity may increase after storage at room temperature. Generally, the accuracy of pH measurements depends on the method of saliva collection and on the time interval between collection and analysis [1].

SALIVA SPECIMEN

The idea of using saliva in diagnoses was made in the second half of the 20th century [5]. Saliva is an accessible fluid that can easily be collected by the patient. Advantages of saliva testing sample are easy and non invasive collection procedure that is neither painful nor traumatic. Saliva is reliable for early detection of certain diseases and monitoring the disease course in conjunction with treatment and detection of addictive drugs [5,10]. These characteristics make it possible to monitor several biomarkers in infants, children, elderly and non- collaborative subjects and in many circumstances in which blood and urine sampling is not available [7] . The most commonly used laboratory diagnostic procedures involve the analyses of cellular and chemical constituents of blood. No special equipment is needed for collection of saliva. Diagnosis of disease via the analysis of saliva is potentially valuable for children and older adults. Further, analysis of saliva may provide a cost-effective approach for the screening of large populations[6].Blood collection is more expensive because it requires the help of a trained technician,it may evoke ethical issue in special populations (infants or elderly persons), it is inconvenient for the patient because samples must be collected in the clinic, and it can be traumatic and stress inducing, especially when repeated samples must be taken[10, 25].

SALIVA COLLECTION

Saliva can be easily collected from human. The patient must be aware of the collection protocol: importance of the exact timing of the samples, excluding brushing teeth before the collection, avoiding food and

fluid (apart from water) ingestion or chewing gum for at least 30 min before collection, and rinsing the mouth with water (preferably distilled). They should also be informed on the procedures for storing samples. If necessary, inhibitors or specific additives should be added to the specimen before the samples are collected /analyzed, otherwise it can be frozen until the laboratory analyses [7]. Although a variety of devices have been developed to collect saliva from individual glands, according to the previous studies mixed (whole) saliva is more preferred [10,31]. The components of saliva play a considerable role in mastication and perception. Different types of stimulation produce significantly different salivary flow rates: citric acid elicits the largest volumes of saliva, followed by mechanical stimulation (paraffin), odor stimulation and unstimulated saliva. The composition of saliva as a response to the four types of stimulation varies significantly in protein concentration which is highest in unstimulated saliva, followed by saliva stimulated by odor, chewing or citric acid. The highest buffer capacity is found in the mechanically stimulated saliva, followed by resting and odor stimulated saliva. Mucin concentration is the highest in resting time [29].

SALIVA AND DENTAL CARIES

In the last few decades, there has been a focus on the utilization of saliva for bacteriological tests that give an indication of dental caries risk[32,33,34]. Salivary proteins interfere with bacterial colonization and also promote colonization [35]. These proteins influence the enamel demineralization-remineralization process and dental caries formation. Furthermore, it has been demonstrated that functional formation of heterotypic complexes between salivary molecules such as MG-1 (high-molecular-weight mucin glycoprotein-1), Amylase, PRPs (acidic prolin-rich protein-1), and Statherin are determination for plaque formation and dental caries. In addition, genetic factors should be included in associated with phenotypic expression of these proteins in mixed saliva, which may contribute to oral disease etiology [36]. There appears to be a correlation among MG-1, MG2 (low-molecular-weight mucin glycoprotein-2), PRP-1and DMFT. Salivary mucin plays an important role in protection of oral surfaces. Also these salivary proteins participate in acquired enamel pellicle formation, thus increase protective features of saliva [27]. Absence of these proteins is associated with increased prevalence of dental caries [7,37]. It has been suggested that adsorption of PRPs to hydroxyapatite enamel causes a structural change in protein, to expose bacterial binding sites which are hidden in the tertiary structure. Statherin allows saliva to maintain its state of supersaturation because of calcium and phosphate salts [13, 38].

SALIVA AND ABH BLOOD GROUP

The carbohydrates of salivary glycoprotein carry the ABH blood group antigens, expressing in the salivary glands and secreting in saliva. The expression of the blood group antigens in saliva may change the specific interaction between microorganisms and their salivary glycoprotein receptors, which might interfere in development and prevention of oral infectious diseases. The performed investigations indicated that high-molecular-weight salivary mucin is the primary carrier of ABH blood group antigens. The ABH blood group antigens were better detected in centrifuged samples stored at lower temperature. These studies also indicated a distinct difference in the structure and function of high-molecular-weight salivary mucin between secretors and non-secretors people. This finding could be used for further researches on the relationship between the blood group antigens and oral diseases as well as on its forensic applications [39, 40].

SALIVA AND DIAGNOSIS OF SYSTEMIC DISEASES

With advances in microbiology, immunology and biochemistry, salivary testing in clinical and research settings is rapidly proving to be a practical and

reliable means of recognizing oral signs of systemic illness and exposure to risk factors. The components of saliva act as a "mirror of the body 's health", and the widespread use and growing acceptability of saliva as a diagnostic tool is helping individuals, researchers, health care professionals and community health programs to better detect and to monitor diseases and to improve the general health of the public. Many studies show that analysis of saliva sample is a convenient means for assessment of physiological conditions, evaluation the serum concentration of medicine and assessment of the severity of an illness [18,41]. Saliva is a good indicator of the plasma levels of various substances such as hormones and drugs and can therefore be used as a non-invasive method for monitoring plasma concentrations of medicines or other substances (Tables 3,4) [13].

Table 3: Systemic Diseases Affecting Salivary Glands and Saliva

Affective disorder	
Autoimmune disease	Sjögren's syndrome, Rheumatoid Diseases, Myasthenia Graft-vs.-host disease
Cancer	
Cirrhosis	
Cystic fibrosis	
HIV infection	
Hormonal disorders	Adrenal-cortical disease, Diabetes mellitus, Thyroiditis, Acromegaly
Hypertention	
Metabolic disturbances	Malnutrition, Dehydration, Vitamin deficiency
Neurological disease	Parkinsonism, Bell's palsy, Cerebral palsy, Alzheimer's disease
Renal disease	
Sarcoidosis	

Table 4: Salivary/Oral Fluid Biomarkers Possibilities of Use

Salivary/Oral Fluid Biomarkers	
DNA	Standard genotyping
	Bacterial infection
	Diagnosing carcinoma of the head and neck
	Forensics
RNA	Viral/bacterial identification
	Carcinomas of the head and neck
Proteins	Diagnosing periodontitis
	Diagnosing carcinomas of the head and neck
	Detecting dental cavities
Mucins/glycoproteins	Diagnosing carcinomas of head and neck
	Detecting dental cavities
Immunoglobulins	Diagnosing viruses (HIV, Hepatitis B and C)
Metabolites	Diagnosing periodontitis
Drug and their metabolites	Detection of drugs in the body
Viruses,bacteria	Epstein-Barr virus reactivation(mononucleosis)
Cellular material	Diagnosis of carcinomas of the head and neck

INFECTIOUS DISEASE

The use of saliva as a diagnostic and monitoring method for periodontal diseases has been increasing studied. Todorovic et al. analyzed the saliva of patients with periodontitis and demonstrated significant increases in enzyme activity in association with cell injury and tissue cell death , salivary enzyme activity , as biochemical markers , may be useful in diagnosis , prognosis , and monitoring of periodontal diseases[33, 42].The levels of amylase and secretary IgA were significantly higher in whole saliva of subjects with periodontitis than in healthy controls[37].Mycobacterium tuberculosis is detected in oral fluid by PCR only when this infectious disease is in an acute phase and the level of bacteria is very high. Mycobacteria appear in the oral cavity in a great number. The diagnosis of streptococcal pneumonia and detection of pneumococcal C-polysaccharides are possible in saliva without detection of nucleic acids [6].The use of saliva as a diagnostic tool for Helicobacter pylori infection (a species of bacteria that infects the mucosal surface of stomach which causes peptic ulcers, gastritis, and cancrum) is an attractive option for epidemiologic studies in children when the non-invasive nature of the test is considered. PCR technology has been proved to be highly sensitive and specific for detecting of H.pylori DNA in the mouth using biological markers found in saliva [5,33,43].Studies successfully used saliva for the diagnosis of HIV by using specific antibodies as biological markers .This method made the first quick test for detection of HIV-1 infection possibility, a test that is presently used in investigations and has a high sensitivity and specificity, at 99.4%. For detecting HIV infection using the ELISA method in combination with the Western blot test, saliva, comparing to blood and urine has greater advantage owing to its higher specificity and sensitivity. Saliva can also be used to

measure the level of beta -2 microglobulins and/or the level of tumor necrosis factor – alpha receptors and confirm the possibility of monitoring the activity of HIV virus or AIDS related inflammatory diseases [32,33,44-46]. Regarding the certain viral diseases, detection of hepatitis A antigen and hepatitis B surface antigen in the saliva has been used in epidemiological studies of to both types of hepatitis. Analysis of saliva provided a highly sensitive and specific method for the diagnosis of viral hepatitis B and hepatitis C. There are commercial kits for determining antibodies of hepatitis B and C viruses that are 100% sensitive and specific [5,6,32,33,47]. PCR is also used for diagnosing CMV , HPV 6,7,8 and human forms of rabies [5,48] . Shedding of herpes viruses (HHV-8, CMV and EBV) in saliva of affected patients has been reported. Saliva was also found to be a reliable alternative to serum for identification of the antibody of parvovirus B19 [6,49]. Saliva samples can also be used for the laboratory diagnosis of rubeola through the detection of IgM. Studies showed that IgM antibodies against rubeola were found in 84.4% of salivary samples with a test specificity of 96%.Saliva has been used to detect antibodies against rubella, parotiditis and rubeola viruses [30,31,46]. Saliva may also been used for determining immunization and detecting infection with measles, mumps, and rubella [6,50].

For newborn infants, the salivary IgA response was found to be better marker of rotavirus (RV) infection. Saliva can be used for monitoring of immune response to vaccination and infection with RV [51]. Some studies have suggested that reactivation of herpes virus type 1 infections is related to the pathogenesis of and that PCR detection of the virus in the saliva would be a suitable method for early detection of reactivation of this illness. The shed of HSV-1 reactivation in patients with Bell's palsy was 50% in comparison with 19% in healthy controls [6,32,52]. Another infectious disease of the oral cavity that can be diagnosed by saliva is candidiasis through the presence of Candida spp in saliva [32]. Histatins are another group of proteins that have been received much interest. Of particular interest is the fact that the histatins have potent anti-candida effects, and it seems that these activities could be exploited as a natural defense against candida [13]. Evaluation of the secretory immune response in the saliva of children infected with Shigella revealed higher titers of anti-lipopolysaccharide and anti-Shiga toxin antibody in comparison with healthy controls. It was suggested that salivary levels of these immunoglobins could be used for monitoring of the immune response in shigellosis [53]. The presence of antibodies to other infectious organism such as Borrelia burdogferi can also be detected through the saliva [32]. Lyme disease is caused by the spirochete Borrelia burgdorferi and is transmitted to humans by blood-feeding ticks. The detection of anti-tick antibody in saliva has potential as a biologic marker of exposure to tick bites, which in turn may serve as a screening mechanism for individuals at risk for Lyme disease [54]. Dengue is a viral disease transmitted by the mosquito Aedes aegypti. Primary infection can lead to a self limiting febrile disease, while a secondary infection can cause serious complications such as hemorrhagic fever or dengue shock syndrome. Additionally, salivary IgG level is useful for the differentiation between primary and secondary infections [6,32,55]. Pigeon breeder's disease (PBD) is an interstitial lung disease induced by exposure to antigens derived from pigeons. Measurement of salivary IgG against these antigens may assist in the evaluation of patients with this disease [56]. The detection of pneumococcal C polysaccharide in saliva by ELISA may offer a valuable complement to conventional diagnostic methods for pneumococcal pneumonia [57]. Specific antibody to Taenia solium larvae has been demonstrated in saliva for detection of neurocysticercosis. It was suggested that saliva could be used in epidemiologic studies of this disease [58].

The predominant immunoglobulin in saliva is secretary IgA (sIgA), which is derived from plasma cells in the salivary glands, and constitutes the main specific immune defense mechanism in saliva. In contrast, salivary IgM and IgG are primarily derived

from serum via GCF, and present in lower concentrations in saliva than IgA. Antibodies against viruses and viral components can be detected in saliva and can aid in the diagnosis of acute viral infections, congenital infections and reactivation of infection [59].

MALIGNANT TUMORS OF THE ORAL CAVITY

In case of primary oral tumors and recurrence measuring the level of selected biomarkers is adequately sensitive and specific monitor. Saliva much has also been considered in the detection of DNA biomarkers to diagnose spinocellular carcinoma of the oral cavity. Mutation of the tumor suppressor gene p53 is common in many malignancies. For malignant tumors of the head and neck, the mutation can be up to 50%. This is confirmed in a number of studies analyzing mutation of the p53 oncogene which can be done by the PCR method. Using the ELISA method, the p53 antibody was detected in 13% of patients treated for spinocellular carcinoma of the oral cavity Defensine are peptides which possess antimicrobial and cytotoxic properties. Elevated levels of salivary defencine-1 were found to be indicative of the presence of oral squamous cell carcinoma [5,13,28,32,60].Oral fluid is also used in diagnosing of other malignancies. Breast cancer is one of the first malignant tumors detected with the help of genetic protein biomarkers. Streckfus et al. drew attention to raised levels of CA15-3, epidermal growth factor (EGF) receptor and c-erb B-2 in patients with breast cancer while Di-Xia, Schwartz, and Fan-Qin described elevated [5,63,64].

HORMONAL ANALYSIS

Saliva contains some free hormones that can be easily measured to show their availability in human tissue and to evaluate endocrine function. The liposoluble hormones with lower molecular weights can be detected most reliably in saliva, but protein- bound hormones (such as gonadotropins, prolactin and thyrotropin) cannot be accurately monitored by means of salivary analysis. Hormones variation in saliva can be an indicative of cancer progress or the possibility of a disease like Cushing syndrome and Addison's , Physical exercise and Stress (cortisol), Primary aldestronism or Conn's syndrome (aldosterone), Testicular function and behavioral studies of aggression, Depression, abuse , Violent and antisocial behavior (testosterone and dehydroepiandrosterone) feto-placental function (estriol), Prediction of ovulation and ovarian function(progesterone). Other

levels of CA125 and the glycoprotein complex in saliva of patients with ovarian cancer [5,61]. A recent report suggests that head and neck carcinomas can be detected by utilizing DNA derived from the exfoliated oral mucosa cells collected in saliva. Franzmann et al. used CD44 protein in saliva as a potential molecular marker in diagnosing of head and neck cancer in all stages [6,33,62].

In the presence of oral cavity malignancies the level of carcino embryonic antigen (CEA) in saliva increases, while the level of gastrointestinal cancer antigen decreases.IL-8 and thioredoxin can be used in the diagnosis of spinocellular carcinoma [33].

HEMATOLOGICAL ONCOLOGY

In recent years, publications have been appeared on the detection of neutrophils in oral fluid of hematological oncology patients after bone marrow transplants. Wright et al. sought to establish high level of neutrophils in oral fluid that potentially indicated the success of bone marrow transplant. They detected neutrophils in saliva 2-3 days earlier than in the peripheral blood. In healthy people, they found diurnal fluctuating levels of neutrophils in saliva. Researchers also examined the importance of oral mucositis after myoablative chemotherapy and found that improvement was related to the appearance of neutrophils in the saliva. Lieschke et al. pointed to a steep rise in neutrophils after administering growth factors (G-CSF) in both blood and saliva which again proves their interdependence.

hormones in saliva are Androstenedione, Dihydrotestesterone, estradiol and insulin [6,32,33,65]. Saliva is being used to detect a specific estrogenic hormone, estradiol, which has been found to predict preterm labour. The FDA-approved the test for detecting of estradiol at home by women at risk for premature,low-birth-weight babies [10,66,67].

CARDIOVASCULAR DISEASES

Cardiovascular diseases are a leading cause of death all over the world. Enzymes found in saliva, such as amylase, have been used for post-operative control of patients who had cardiovascular surgery. A study of Adam et al. showed that low levels of salivary amylase in pre-operative stage of patients with aorta aneurism is associated with an increase in mortality. Chatteron et al found the direct relationship between raised levels of alpha amylase and heart rate which increases under stress [5,32,68].

AUTOIMMUNE DISEASES

Sjögren's syndrome is a chronic disease affecting the lachrymal, salivary and other exocrine glands. Some procedures for the diagnosis of Sjögren's syndrome include sialography, salivary scintiography, biopsies, and serological tests. These tests are invasive, expensive, and sometimes conclusive. Researchers have measured specific concentrations of cytokines in saliva of patients with Sjögren's syndrome.Lactoferin, Beta 2 Microglobulin, Lysozyme C, Cyctatin C, Cyctatin S, Sodium, chloride, Albumin, Alpha 2 microglobulin, Lipid and inflammation mediators such as Eicosanoids, Prostegelandine E2, Thromboxane B2 and Interlukins 2 and 6, IgA,IgG and IgM autoantibodies arised in individuals that suffer from autoimmune diseases . Levels of amylase, carbonic anhydrase and phosphate decrease in saliva but levels of calcium and potassium are usually normal. Salivary kallikerin is in association with Sjögren' syndrome. Ss-anti La antibodies were primarily found in saliva .Any changes of these antibodies in saliva can be useful in diagnosis of Sjögren's syndrome as well as in control of its progression. Furthermore, analysis of unstimulated whole saliva was more sensitive than analysis of stimulated whole saliva for detection of these changes. Parotid saliva from patients with rheumatoid arthritis and Sjögren's syndrome contains higher level of multiple forms of tissue kallikrein [4,5,32,33,69,70].

CELIAC DISEASE

Celiac disease is a congenital disorder of the small intestine that involves malabsorption of gluten. To diagnose Celiac disease, detection of IgA and antigliadin antibody in saliva shows high specificity and low sensitivity, whereas their determination in serum is highly sensitive and less specific [30,71].

CYSTIC FIBROSIS (CF)

Besides the increased visco-elasticity of saliva in cystic fibrosis patients, there are several electrolyte and protein concentration differences compared with healthy individuals (not CF-heterozygote). The RNase-activity is four times higher in CF homozygotes than in control subjects. Due to the increased calcium and total protein in saliva, insoluble calcium-protein complexes are formed. The salivary concentrations of sodium, phosphate, chloride, lipid, epidermal growth factor and prostaglandin E2 also increase which are believed to play an important role in protection against dental decay [6,32,72].

MULTIPLE SCLEROSIS

Very little changes are observed in whole saliva of multiple sclerosis patients. However, there is a significant reduction in IgA production during rest, but the absence of protein band of 140 kDa is also found. Although, these findings can not be used in diagnoses of the disease [4].

GRAFT-VERSUS-HOST DISEASE

Graft-versus-host disease presents as destruction of the salivary gland tissues, resulting in decreasing of salivary flow rate.

This can either be acute, shortly after transplantation, or chronic, about 100 days after the surgical procedure. It should be mentioned that decreasing in salivation can be induced, in the first place, by the accompanying irradiation or chemotherapy. However, when the hyposalivation increases 100 days after the transplantation, it may be indicative for a graft-versus-host disease. In these situations sodium and lysozyme concentrations are increased, while phosphate and s-IgA are decreased in saliva [4,73].

DIABETES MELLITUS

Insulin is able to stimulate salivation so in diabetes mellitus patients the salivary flow rate decreases. In this situation salivation is easy to stimulate. It should be noted that medications used in these patients can also be responsible for the decreased salivary flow rate .Albumin and IgG concentrations of non stimulated saliva are lower than healthy individuals [74]. Patients with diabetes mellitus express higher levels of amylase and secretory IgA in whole saliva constituents [4, 44].

ALCOHOLIC LIVER CIRRHOSIS

Parotid enlarges in 50% of the patients with alcoholic liver cirrhosis, which results a 50% reduction of the salivary flow rate and a reduction of salivary sodium, bicarbonate and chlorine concentrations. The total salivary protein concentration decreases as well [4].

EPILEPSY

Gingival hypertrophy can be observed in patients with epilepsy who take phenytoin due to increasing of collagen synthesis and accumulation of proteoglycans. These patients should have a high quality oral hygiene. IgA deficiency is another side effect of phenytoin, resulting in a decreased immunological defense. Cyclosporine A and nifedipine are not associated with similar effects [4,75].

BURNING MOUTH SYNDROME

This syndrome is relatively most common in post-menopausal women with male: female ratio 1:7. Patients complain of oral pain and dry mouth. Dry mouth can be developed after taking some medications like antidepressants. However, their salivation can be easily stimulated mechanically and chemically. The total salivary protein concentration of stimulated saliva is lower than in control subjects, but the total mucin concentration is higher. Salivary potassium, chloride and phosphate concentrations are also increased in the patients [76,77].

KIDNEY DYSFUNCTION

Half of all hemodialysis patients complain of hyposalivation, changes in taste, ammonium smelling breath and oral mucosal pain. The total salivary protein, sodium and potassium concentrations are similar to the plasma. Salivary pH of these patients is significantly higher than the healthy controls, due to the significantly increased salivary urea concentration [4,78]. Gonzalez et al. found that saliva can be a good tool for early detection of exposure to lead and cadmium since salivary levels of these elements arise from the diffusible fraction of plasma [79].

SALIVA AS A DIAGNOSTIC TEST FOR DRUGS

Diagnostic testing of drugs using saliva/oral fluid is now widespread and replacing the previously used urine. Saliva is used to measure the level of Lithium, Carbamazepine, Barbiturates, Benzodiazepines, Phenytoin, Theophyline and Cyclosporine, Antipyrine, Caffeine, Cisplatin, Diazepam, Digoxin, Ethosuximide, Irinotacan, Methadone, Methoprolol, Oxpernolol, Parecetamol, Primidone, Procainamide, Quinine, Sulfanilamide, Tolbutamide and for drug abuse such as Amphetamine, Benzodiazepines, Cocaine, Ethanol, Marijuana, Nicotine, Opioids and Phencyclidine [5,6,32].

21-HYDROXYLASE DEFICIENCY

21-hydroxylase deficiency is an inherited disorder of steroid genesis which causes congenital adrenal hyperplasia. Early morning salivary level of 17-hydroxyprogestrone(17-OHP) is an excellent screening test for the diagnosis of 21-hydroxylase deficiency. There is a correlation between 17-hydroxyprogestrone levels in saliva and serum in both affected and healthy individuals [(6)].

SIALOCHEMISTRY ANALYSIS

Researchers found that saliva can be a good tool for early monitoring of an exposure to lead and cadmium, because of higher salivary levels of these elements[(7)].

Sialochemistry of environmental heavy metals (cadmium, lead, mercury) may be useful in monitoring environmental, atmospheric and occupational pollutants.

CONCLUSION

Saliva is an important body fluid for detecting the physiological and pathological situations of the human body. Saliva is a complex and dynamic biological fluid containing wide range of compounds. The biochemical and physical chemical properties of these salivary components and their interaction function in the oral cavity. In the last few years scientific interest has been raised to saliva not only for the various compounds, e.g., drugs, pollutants, hormones into saliva, but also the well-documented relation of saliva with bacterial, viral and systemic diseases. The relatively easy and non-invasive nature of sample collecting is considered as a simple low-cost stage but the problem is the concentrations of its biological markers are in lower levels in compariing with plasma, and there still are not reference values. The development of better technology (nanotechnology) is improving this situation.

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