



ANALYSIS OF SALIVARY PROTEINS AS THE BIOCHEMICAL INDICATORS OF NUTRITIONAL STATUS AND SALIVARY GLAND FUNCTION

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ABSTRACT

Saliva is increasingly used and well validated in diagnosing, monitoring health and disease status, mainly due to its origin, composition, functions, and interactions with other organ systems. The effect of nutrition status on saliva covers a broad spectrum of factors. This includes the degree of malnutrition, the period when malnutrition takes place, the consistency of the diet, the effect on salivary gland growth and function, and the effects on the different salivary glands. Protein-energy malnutrition (PEM) is a serious nutritional problem in slum children's and elder. Serum albumin, globulin, transferring, total protein and amylase activity have been used as biochemical indicators of protein nutritional status and salivary gland functions, but taking blood from the slum subjects are difficult and invasive. We therefore assessed the possibility of using saliva as noninvasive materials to estimate concentrations of albumin, transferrin, total protein and amylase activity. Saliva is collected from 155 slum and healthy subjects of different ages and control (aged 8-15, 24- 32, 58-77 yrs) and the concentrations of albumin, transferrin, total protein and amylase activity in saliva were evaluated. Altered levels of protein profile were observed in this study. This result suggested that nutritional deficiencies compromise salivary gland function, thus increasing susceptibility to dental caries and oral infections and reducing quality of life.

KEYWORDS: Malnutrition, Saliva, Protein, Amylase, Slum, Protein Energy Malnutrition.



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INTRODUCTION

Saliva is increasingly used and well validated in diagnosing, monitoring health and disease status, mainly due to its origin, composition, functions, and interactions with other organ systems. Saliva has hundreds of components which help detect systemic diseases and also provide biomarkers of health and disease status¹. Salivary assays present a lot of advantages when compared to blood assay: the sampling is very easy to do especially in non medical environment; multiple samples could be collected providing more information than that of single blood sample². Most researches have shown the advantage of using saliva in detecting physiologic or pathologic conditions because there is a close relationship between saliva and serum parameters^{3, 4}. The 'Nutrition' emerges as an important prerequisite for national development. Nutrition is one of the most important aspects of growth and development of human. It is well known that quantitative (energy and protein levels) and qualitative (nature of the diet, nutrient dynamic) changes in the nourishment of human affect their metabolism. Therefore, nutrition aims to optimise the metabolic activity of various organs⁵. In the present context malnutrition is synonymous with protein-energy malnutrition, which signifies an imbalance between the supply of protein and energy and the body's demand for them to ensure optimal growth and function⁶. Protein-energy malnutrition (PEM) is a serious nutritional problem in the slum and elderly subjects. The concentrations of albumin transferrin and total protein in serum have been used as the biochemical indicators of protein nutritional status. Recently, many studies revealed that the concentration of albumin in serum is associated with health and nutritional status among the elderly. In previous studies, a significant correlation between PEM and the concentration of total protein in saliva was demonstrated among children in India⁷. Saliva contains a large number of protein compounds, of which the structure and function have been studied with traditional biochemical techniques and lectin probe⁸. Salivary total protein is a vital component

of saliva, with salivary proteins, predominantly comprising proline rich proteins, mucin, amylase, immunoglobulins, statherin and antibacterial factors. Human saliva proteins can have a wide range of functional properties including the immune response, inhibition of calcium precipitation, taste perception, digestion, inhibition of proteinase, oral defence, transcription, cell proliferation; signal transduction, chemotaxis and cell motility⁴. Protein deficiency results in alterations to salivary gland structure and function. The effect of protein deficiency depends on the time period at which deficiency occurs⁹. One billion people or one third of the world's population are estimated to be living in either slum or squatter settlements. The largest proportion of the population living in slums in the world is in the Asian region due to low income cities¹⁰. Slum area means an area in which there is at least one-fourth (1/4) of all buildings or a predominance of improvements which by reason of dilapidation, deterioration, age or obsolescence, inadequate provision for ventilation, light, sanitation, or open spaces, high density of population and overcrowding, or any combination of such factors, are unsafe or unfit to occupy; are conducive to ill health, transmission of disease, infant mortality, juvenile delinquency, or crime; injuriously affect the entire area; or constitute a menace to the public health, safety and welfare¹¹. Health problems tend to be very high, as a result of improper sanitation and lack of access to basic health care. The ability to use saliva to monitor an individual's health and disease state is a highly desirable objective for healthcare research and promotion. Keeping in view, in the present study was to examine the correlation between the concentrations of total protein, albumin transferrin and α -amylase in saliva, and to examine the usefulness of the concentrations of the proteins for evaluating protein nutritional status and salivary gland function in slum and healthy subjects of different ages.

MATERIALS AND METHODS

Subjects were referred from the major two groups were established as group I and group II. The group I was sub-divided into three different age group of health subjects (5-15, 24-32, and 58-78). Group II sub-divided into three different age group slum subjects (5-15, 24-32, and 58-78). The Candidate were analysed in the protein, albumin, amylase and transferrin. Total number of subjects is 60. The study was conducted in children, young and elder from Kilavasal Thanjavur where healthy and slum age-matched subjects were included in the study. The purpose of the study was explained to the parents/guardians and an informed consent for participation was obtained from them. Group I includes 25 male and 22 female in the group mean age of 10, age range 5-15 years. Group II includes 35 male and 32 female in the group mean age of 28, age range 24-32 years. Group III includes 20 male and 21 female in the group mean age of 68, age range 58-77 years. The control group consisted of healthy subjects without a past history of the systemic or cardiovascular problem. Smokers were excluded from both groups. For saliva collection, the subjects' oral cavities were rinsed with distilled

water was chewed for several minutes. Through this method, 10ml of the saliva samples was collected from each subject and immediately kept in a refrigerator. It was centrifuged at 3000 rpm for 15min and the supernatant was delivered into tubes (1mL/tube). The tubes were stored frozen at -80°C, thawed on measurement, and analyzed.

(i) Sample collections

We collected unstimulated whole expectorated saliva from each subject, into sterile centrifuge tubes, between 9 and 10 a.m. after a single mouth rinse with 15 ml of distilled water to wash out exfoliated cells¹². We placed collected samples on ice immediately. We then centrifuged them at 3000 rpm and the supernatant was used for analysis of various biochemical parameters.

(i) Biochemical estimations

Protein was estimated by the method of ¹³. The results were expressed as g/l. Albumin was estimated by the method of ¹⁴. The results were expressed as mg/dl. Transferrin was estimated by the method of ¹⁵. The results were expressed as mg/dl. Amylase was estimated by the method of ¹⁶. The results were expressed as U/ml.

Table 1
Analysis of protein, albumin, transferrin content and amylase activity in saliva of healthy and slum subjects.

Parameters	Age group (5-15)		Age group (24- 32)		Age group (58-77)	
	Healthy	Slum	Healthy	Slum	Healthy	Slum
Protein (g/l)	0.88 ± 0.003	0.63 ± 0.02*	1.5 ± 0.06#	1.01 ± 0.04*#	0.96 ± 0.03	0.91 ± 0.03*
Albumin (mg/dl)	3.54 ± 0.14	2.31 ± 0.09*	5.34 ± 0.21#	4.06 ± 0.16*#	3.81 ± 0.15	3.53 ± 0.14*
Transferrin (mg/dl)	0.5 ± 0.02	0.4 ± 0.01*	0.8 ± 0.03 #	0.7 ± 0.02*#	0.6 ± 0.02	0.6 ± 0.02*
Amylase (U/ml)	52.3 ± 2.09	48.7 ± 1.94*	84.6 ± 3.38 #	62.3 ± 2.49*#	60.2 ± 2.40	56.4 ± 2.25*

Values were expressed as mean ± SD

*Significantly different from respective healthy age group

#Significantly different from age group of 5-15 and 58-77.

RESULTS

In the age group 5-15 of slum subjects of salivary protein, albumin, transferrin and amylase activity was significantly decreased as

compared to healthy subjects. Among the various age groups, the protein content and amylase activity of healthy and slum subjects

were significantly decreased in the age group 5-15 as compared to the age group of 24-32 and 58-77. In the age group 24-32 of slum subjects of salivary protein, albumin, transferrin and amylase activity was significantly decreased as compared to healthy subjects. Among the various age groups, the protein content and amylase activity of healthy and slum subjects were significantly increased in the age group 24-32 as compared to the age group of 5-15 and 58-77. In the age group 58-77 of slum subjects of salivary protein, albumin, transferrin and amylase activity were significantly decreased as compared to healthy subjects. Among the various age groups, the protein content and amylase activity of healthy and slum subjects were significantly decreased in the age group 58-77 as compared to the age group of 24-32 and 5-15.

DISCUSSION

Most of the people who live in slums are extremely poor, and many are treated as second class citizens by their society. Malnutrition is a serious problem in many slums. Slums are further characterized by the following attributes: (a) lack of basic services, (b) substandard housing or illegal and inadequate building structures, (c) overcrowding and high density, (d) unhealthy living conditions and hazardous locations, (e) insecure tenure and irregular or informal settlements, (f) poverty and social exclusion, and (g) minimum settlement size¹⁷. Another determinant of health is access to insufficient quantities of safe and quality food¹⁰. The health problems experienced by slum residents included worms in children. Food and Water-borne illnesses, cholera and dysentery, were a perennial problem, largely because of the lack of adequate food and potable water supply. Other common illnesses because of congested living conditions and low standards of hygiene included tuberculosis¹⁰. The proteins in saliva have been found in concentrations of approximately 3% of plasma, and most have antibacterial properties, as they include both non immunoglobulin and secretory antibodies,

especially secretory IgA, lysozyme, lactoferrin and peroxidase. Salivary components, particularly proteins, are multifunctional (performing more than 1 function), redundant (performing similar functions but to different extents), and amphifunctional (acting both for and against the host)¹⁸. Research on the effect of nutritional status on saliva covers a broad spectrum of factors. This includes the degree of malnutrition, the period when malnutrition takes place, the consistency of the diet, the effect on salivary gland growth and function, and the effects on the different salivary glands¹⁹. Nutritional changes and deficiencies can influence salivary function as well. A modest reduction in daily food intake may result in decreased salivary protein, whereas severe caloric restrictions tend to reduce salivary flow, cell numbers, and salivary composition²⁰. The salivary levels of total protein increase also through β -sympathetic activity in the salivary glands, since saliva secretion is mainly evoked by the action of adrenergic mediators²¹. Protein energy malnutrition (PEM) interferes with the action of β adrenergic mediators²² and decrease protein content in saliva were observed in this study. Our study also supported with²³. A study was done to investigate the relation between protein deficiency and dietary texture (which affects the requirement for mastication) and comparing low- with normal-protein diets fed in a powdered or pellet form. In addition to a marked effect of protein deficiency on salivary gland growth and function, the powdered diet reduced gland function. This is supported by others where a change in saliva volume or protein content has been found in relation to protein malnutrition²². Previously reported in children have shown that the severity of protein energy malnutrition (PEM) is related to the extent of reduction of stimulated salivary secretion rate²⁴. PEM also results in a decrease in calcium, chloride and secretion of protein of stimulated saliva and a reduction in immunologic and agglutinating defense factors of unstimulated saliva. Because an increased severity of PEM is accompanied by a decrease in salivary protein concentration and arginase activity, protein content of saliva and salivary arginase activity in

the early stages of the disease might be used as an index of PEM^{25, 22}. There was a decrease in stimulated salivary secretion and reduced protein concentrations in human adults fed a low-energy liquid diet for 7 days²⁶. Few studies have evaluated the effects of protein deficiency on salivary gland function in human adults^{27, 28}. Significant decreases in the secretion rate of protein concentration in stimulated whole saliva also have been observed in adults²⁹. In conclusion, saliva has hundreds of components which provide biomarkers of health and disease status. Saliva has an important role in ensuring adequate nutrition, salivary gland function and in

providing protection from oral diseases. Our data suggest that the possibility of usefulness of the concentrations of the proteins for evaluating protein nutritional status and salivary function in the healthy and slum subjects. The results of the present study indicate that the decrease in protein content in slums as compared to healthy subjects may be due to the poor nutrition. The poor nutrition may lead to decrease in salivary gland activity and a further decrease in salivary flow. At the same time, nutritional deficiencies compromise salivary gland function, thus increasing susceptibility to dental caries and oral infections and reducing quality of life.

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