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Comparative Evaluation of Salivary Total Proteins in Mixed and Permanent Dentition.

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ABSTRACT

Human saliva plays a vital role in maintaining the integrity of oral tissues and its composition changes during childhood due to maturation of the salivary glands thus indicating the need of age-matched controls for the clinical use of saliva as a diagnostic tool for diseases. This study aims at analyzing physiologic variability of naturally occurring total protein concentration in unstimulated whole saliva of children as a function of age. For this study unstimulated whole saliva specimens were collected from 20 healthy children equally divided into: Mixed (6 - 11 years); Permanent (12 – 18years) dentition age groups. The samples were studied for total protein content by light chromatography coupled with mass spectrometry. The total protein content showed a linear increase with age. Thus this study establishes a correlation between age and salivary composition hence constructing a comprehensive catalogue which forms the basis for salivary total proteins with newer biochemical aids necessary for saliva to serve as a diagnostic aid.

Keywords: Saliva, total proteins, mixed dentition, permanent dentition.

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INTRODUCTION

Mouth is a unique, highly complex multifactorial interface between the body and its external environment. It is very complex structurally and biologically. It contains mineralized tissues (teeth) continuously exposed to environmental changes. Thus saliva, an oral bio-fluid is important in maintaining homeostasis in the oral cavity and its presence is vital to the maintenance of healthy oral tissues [1].

Saliva is composed of organic, inorganic contents and macromolecules. Its composition changes from childhood to adulthood due to maturation of salivary glands. Thus for saliva to serve as a diagnostic aid there is a need for age-matched controls with physiologic levels of salivary proteins established for particular age groups[2].

The role of saliva is much broader and it can also serve as a diagnostic tool for monitoring health and disease status of an individual [3]. This aspect of saliva is a late bloomer and is now coming to the forefront. But for this approach to succeed we must understand the basic concept of salivary composition and the role of its constituents [1].

There are only few studies on salivary composition of healthy children which are available [4]. Thus this study aims at analyzing physiologic variability of naturally occurring total protein concentration in unstimulated whole saliva of children as a function of age.

MATERIAL AND METHODS

Criteria for patient selection

In the present study, 20 normal healthy children ranging from 6 to 18 years were selected from housing societies in and around Pimpri- Chinchwad area of Pune district who were free from any systemic or local diseases which affect salivary secretions and totally caries free with dmft/DMFT score of 0 [5] in 2015. After assessing and confirming their caries status these children were stratified equally into two dentition groups: Mixed (10 children ranging from 6 -11 years), Permanent (10 children, ranging from 12- 18 years). Exclusion criteria included patients who were physical or mentally compromised, having developmental delay, auditory or visual dysfunction, known neurological diseases, history of drug intake and patients with arrested carious lesions [6]. Informed consent forms were obtained from the custodial parent or guardian of the subject after explaining the procedure to the parent or guardian.

Method of saliva collection

To minimize the effect of circadian rhythms, all whole saliva samples were collected one hour after lunch for the unstimulated condition [4]. The child was seated in a well-ventilated and well-lit room. The head was kept at 45 degrees flexion with one hand holding onto a 4ml cryoprecipitation vial with a funnel inserted into it, in a calm atmosphere to simulate unstimulated conditions. The saliva was allowed to drip into the funnel held to the lower lip. For each trial, the collection continued for 2 minutes but if the saliva sample was insufficient within 2 minutes, the collection was continued until 2 ml of saliva per subject was obtained [6].

Methods of laboratory analysis

For detection of total proteins in saliva, the saliva samples obtained from each subject were diluted with distilled water in a proportion of 1:4. This diluted saliva sample was then subjected to inductively coupled plasma emission spectroscopy for detection of total proteins, light chromatography coupled with mass spectrometry (LCMS) was used. Mass spectrometry (MS) is an analytical technique used for determining masses of particles, for determining the elemental composition of a sample or molecule and for elucidating the chemical structures of molecules, such as peptides and other chemical compounds.

RESULTS

Sr No.	Mixed dentition	Permanent dentition
1.	50	21.4
2.	21.6	20.2
3.	33	21.4
4.	22.7	26.3
5.	20.05	22.1
6.	20.5	50
7.	42	33.5
8.	35	51
9.	50.5	78.06
10.	48	44

Table 1: Salivary total protein in mixed and permanent dentition

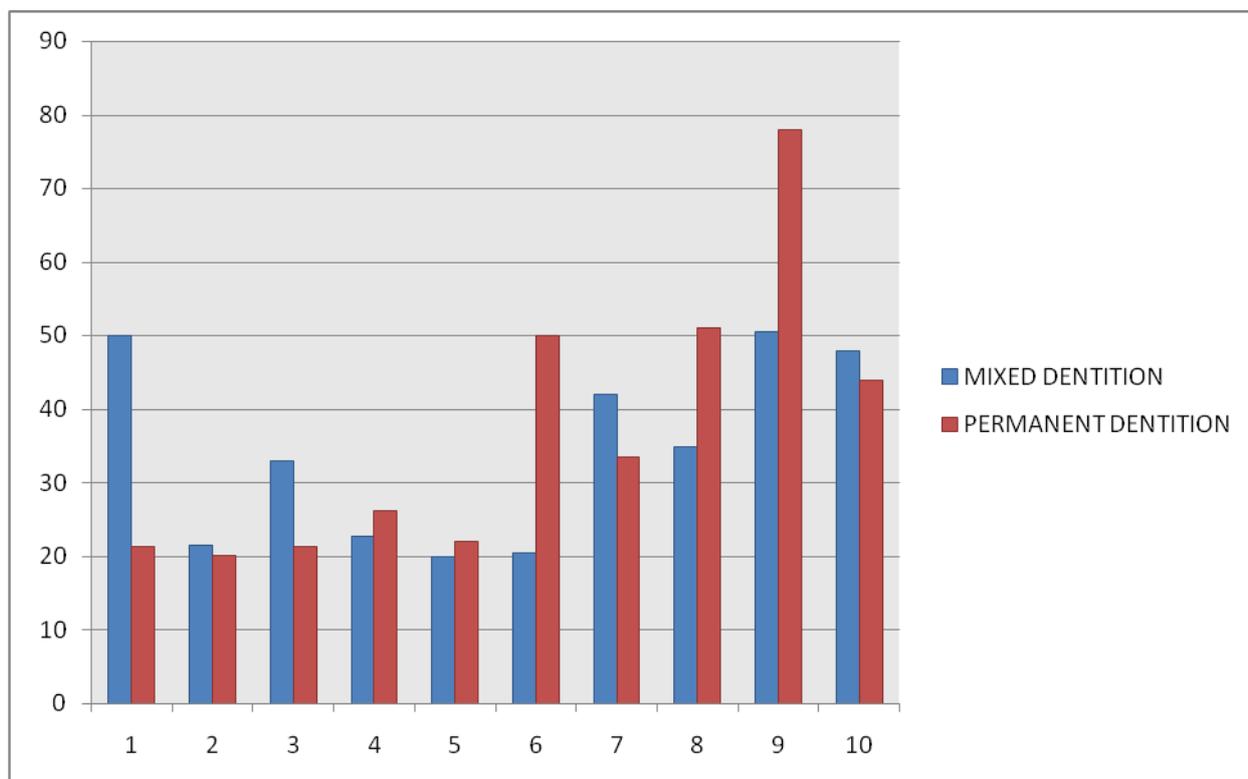


Figure 1: Bar graph showing Salivary total proteins values in mixed and permanent dentition.

The above mentioned results have been statistically analyzed by Mann Whitney test (not significant).

DISCUSSION

This study involved the collection of whole saliva to analyse its organic and inorganic contents. The compositions of the contents of saliva are never constant. A number of physiological factors influence them. The factors being the source of saliva, the method in which it is collected and the degree of stimulation. It is difficult to use any collecting device in children. Hence for the study, unstimulated whole saliva was collected. Apart from the factors mentioned above the time of saliva collection is also important. In this study saliva was collected in the acrophase salivary flow rate peaks during afternoon time [8].

Total proteins in saliva may have both protective and detrimental properties [9]. The function of the proteins is largely dependent on the molecule’s location as well as the site of action. Some proteins such as antimicrobial and pH modulating proteins play a protective role in the oral cavity, while adhesions

and agglutinins play a detrimental role by increasing the colonization of micro-organisms. Thus quantitative and qualitative identification of salivary proteins is a necessary first step in identifying potential protein biomarkers of disease[10]. In this study the total protein concentration showed a linear increase with age, but qualitative analysis of these proteins is essential to prove its role in health and disease.

CONCLUSION

From this study we can conclude that saliva has a great potential for clinical disease diagnostics. It has long been recognized that saliva serves as a mirror of body's health as it contains proteins that are frequently measured in standard blood tests to monitor health and disease[11]. Thus there is a necessity for constructing a comprehensive catalogue which is physiologic for salivary total proteins along with the qualitative analysis of individual amino acids, their linkages and formations with newer biochemical approaches. Thus this study lays a foothold and may serve as a reference value for growing interest in saliva as a diagnostic tool.

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