Salivary lysozyme levels in patients with primary immunodeficiencies

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ABSTRACT

Background: Lysozyme is a muramidase that acts on the peptideoglycan wall of Gram positive bacteria, causing cell death. It plays part in innate immunity and is present in blood, external fluid, as well in lysossomal granules of the phagocytes. Primary Immunodeficiencies are a diverse group of illnesses that, as a result of abnormalities of the immune system, increase susceptibility to infection. Among the examples of impaired natural immunity are defects in phagocytes and in the complement system. Innate immunity could be important in protecting mucosas against infections in patients with different forms of primary immunodeficiencies. The aim of this study was to investigate lysozyme concentrations in saliva from patients with primary immunodeficiencies.

Methods: Lysozyme levels in saliva samples from 34 patients with primary immunodeficiency (30 children and adolescents between the age of 3-13 years and 4 adults between the age of 20-33) and 60 age-matched healthy controls (49 children and adolescents between the ages of 3-15 and 11 adults between the ages of 22-42) were determined by the lysoplate method.

Results: There was no statistically significant difference between the lysozyme concentrations in the saliva of the immunodeficient subjects and those of the healthy controls.

Conclusion: The results in the present work clearly show that salivary lysozyme levels in primary immunodeficient patients are equivalent to those found in healthy controls, suggesting that this enzyme still represents a remaining (but not a compensatory mechanism), contributing to the protection of there patients against infections.

Key words: Primary immunodeficiency. Lysozyme. Saliva. Compensatory mechanism. Mucosal immunity.

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RESUMEN

Introducción: La lisozima es una muramidasa que actua sobre la pared de peptidoglicano de las bacterias Gram-positivas, provocando la muerte celular. Desempeña un papel en la inmunidad innata y está presente en la sangre, secreciones externas y en los gránulos de los fagocitos. Las inmunodeficiencias primarias constituyen un grupo diverso de enfermedades que, como consecuencia de las anormalidades en el sistema inmune, presentan un aumento de susceptibilidad a las infecciones. Ejemplos de deficiencias de la inmunidad natural son los defectos de los fagocitos y del sistema de complemento. La inmunidad innata puede ser importante para la protección

contra las infecciones de las mucosas en los pacientes con diferentes tipos de inmunodeficiencia primaria. El objetivo de este estudio es investigar la concentración de lisozima en la saliva en pacientes con inmunodeficiencia primaria.

Métodos: Los niveles de lisozima en las muestras de saliva de 34 pacientes con inmunodeficiencia primaria (30 niños y adolecentes con edad entre 3-13 años y 4 adultos con edad entre 20-33 años) y 60 controles sanos de la misma edad (49 niños y adolescentes con edad entre 3-15 años y 11 adultos con edad entre 22-42 años) fueron determinados por el método *lysoplate*.

Resultados: No se observó una diferencia estadística significativa entre las concentraciones de lisozima en la saliva de los individuos inmunodeficientes y de los controles sanos.

Conclusión: Los resultados del presente trabajo claramente indican que los niveles de lisozima en la saliva de los pacientes con inmunodeficiencias primarias son equivalentes a los de los controles sanos, sugiriendo que este enzima representa un mecanismo remanescente, pero no compensador, que contribuye a la protección contra las infecciones en estos pacientes.

Palabras clave: Inmunodeficiencias primarias. Lisozima. Saliva. Mecanismo compensador. Inmunidad de las mucosas.

INTRODUCTION

Defense against microbes is mediated by the early reactions of innate immunity and the later response of adaptative immunity. Adaptative immunity is stimulated by exposure to infectious agents, which make the body capable of recognizing and reacting to a larger number of microbial and nonmicrobial substances. Innate or natural immunity consists of cellular and biochemical defense mechanisms that are in place even before infection and poised to respond rapidly to infection. These mechanisms react only to microbes and not to noninfectious substances, and they respond in essentially the same way to repeated infections. The principal components of innate immunity are physical and chemical barriers, phagocytic cells and NK cells, blood proteins - including members of the complement system and other mediators of inflammation, like lactoferrin and lysozyme.

Lysozyme is a 1-4-β-N acetylmuramidase, which acts on the peptideoglycan layer of Gram positive bacterial wall, causing cell lysis. It's a soluble endoglycosidase present in blood, in external fluids as well in the lysossomal granules of the phagocytes[1,2]. Lysozyme is also present in human milk throughout lactation in a concentration of 100 μg/ml, and unlike other factors, its concentration increases towards the end of lactation. Lysozyme is produced by monocytes, neutrophils, Paneth cells (in the intestinal tract)[3] and salivary glands[4]. In phylogeny, lysozyme is highly conserved, being present right through from plants to mammals[5].

Primary immunodeficiency disorders are a diverse group of illnesses that, as a result of abnormalities in the immune system, increases susceptibility to infection[6]. However, little is known about mucosal innate soluble factors that contribut to the host defense of these patients. Synergism between lysozyme, IgA and the complement system has been reported to occur in microorganism lysis, but the biological role of lysozyme is not yet fully known[7]. In the present work, we investigated the lysozyme concentrations in saliva from patients with primary immunodeficiencies to verify if lysozyme might act as a compensatory mechanism for the mucosal protection of these patients.

MATERIAL AND METHODS

Thirty saliva samples were collected from children and adolescents aged between 3 and 13 years and 4 from adults aged between 20 and 33 years at Instituto da Criança of Hospital das Clínicas and Escola Paulista de Medicina, São Paulo, Brazil.

The deficiency diagnosis was: X-linked Agammaglobulinemia (8 cases), Ataxia Telangiectasia Syndrome (5 cases), Common Variable Immunodeficiency (6 cases), Polysaccharide Antibody Deficiency –which represent normal serum levels of immunoglobulins and IgG subclasses, good antibody response to proteic antigens and severely impaired antibody response to polysaccharide antigens– (4 cases), Hyper-IgM Syndrome (2 cases), Chronic Granulomatous Disease (2 cases), IgG2 Deficiency and Down Syndrome (1 case), Silver-Russel Syndrome with Common Variable Immunodeficiency (1 case), Chédiak Higashi Syndrome (1 case), Selective IgA Deficiency (1 case), Hyper IgE Syndrome (1 case), and Agammaglobulinemia with partial deletion of chromosome 2 (1 case).

Saliva samples were also collected from 49 healthy 3-15 year-old children and adolescents and from 11 of 20-42 year-old adults who were used as controls. The lysoplate method was utilied to determine the lysozyme concentrations in saliva samples. This test is based on the agar plate diffusion