Streptococcus pyogenes orophanynx research in indigenous village of the West of Parana

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Abstract

Objectives: Execute orophanynx excretion culture in Tekohá Jevy indigenous from village located in Guaíra City Paraná State, aiming to provide the presence of Streptococcus pyogenes. Method: Cross-sectional and observational study on which 127 indigenous orophanynx excretion sample had taking through sterile swab, 5% sheep blood agar sown and from 24 to 48 hours incubated. The beta-hemolysis standard colonies were submitted to sensitivity test of sulfamethoxazole-trimethoprim, bacitracin, catalase and 1Pyrrolidonyl-Beta-Naphthylamide (PYR) quick test. The following variables were also rated: sex, age and running orophanynx excretion in the sample taking moment. Results: The higher sample taking were obtained from 4 to 11 years old children (45,7%) and from female gender (58,3%). None fo the participants presented pharyngotonsillitis signs in the material sample taking moment and none of the samples had compatible culture with the researched bacteria. Conclusion: There wasn’t presence of S. Pyogenes in the studied population. This can indicate lower contamination rate and bacteria dissemination in isolated population.

Keywords: Prevalence, Indigenous Population (Public Health), Streptococcus pyogenes.
INTRODUCTION

Streptococcus pyogenes is a catalase-negative bacteria with morphological and staining characteristics of gram-positive cocci, classified as a Lancefield Group A beta hemolytic streptococcus (GABS)\(^1\). They may be part of the palatine tonsils’ microbiota as colonizers in the absence of clinical manifestations, and as potential pathogens when causing diseases in the bearer or in other susceptible individuals\(^1\). It is highly important in clinics for being an agent of pharyngitis, impetigo, erysipelas, cellulitis, puerperal sepsis, necrotizing fasciitis, toxic shock syndrome, endocarditis, septic arthritis, acute diffuse glomerulonephritis and acute rheumatic fever\(^1\).\(^2\).\(^3\). \(S. pyogenes\) is responsible for 30-40% of all acute pharyngotonsillitis, especially those in children and young adults, thus being deemed a public health issue. Infection may happen by direct contact through secretions (cough, sneeze), making it easy to spread it in crowded areas and places with much interpersonal contact\(^4\). The etiology of pharyngitis is based on lab tests and in the oropharyngeal secretion culture - which is the diagnostic gold standard, with 90 to 95% sensitivity.\(^5\).

In Brazil it is very difficult to establish the incidence of bacterial pharyngotonsillitis caused by GABS. Following a projection from the World Health Organization (WHO), and according to the census made by the Brazilian Institute of Geography and Statistics (IBGE), it is estimated that in Brazil there are about 10 million cases of streptococci pharyngotonsillitis per year\(^6\).\(^7\).

The indigenous population has several characteristics that make them vulnerable to infection and oropharynx colonization by \(S. pyogenes\), such as low socioeconomic status, crowding, difficulty in accessing healthcare and difficulty in understanding health and disease.

This study aimed to investigate the presence of \(S. pyogenes\) in the oropharynx of indigenous people from Tekohá Jevy village, located in Guaíra-PR.

METHODS

Observational cross-sectional study, carried out with indigenous people from the Tekohá Jevy village in the municipality of Guaíra-PR. The initially estimated population was 250 inhabitants and after considering the exclusion criteria (under four years, over 60 years, physical impossibility of opening one’s mouth, people with diagnosed infectious disease and refusal to participate), the study population consisted of 127 native Brazilians. We chose to exclude individuals under four years and those over 60 years from the study because of the low incidence of bacteria in these excluded age groups, reinforcing the higher incidence in children older than 3 years and especially those in school age\(^8\).\(^9\). Prior to harvesting, we explained verbally and visually (through banner images made by the team), how the test would be performed. The free and informed consent form was signed by the participant or guardian. Children under 18 years of age required the signature of a responsible adult, and adolescents between 12 and 18 also signed an informed consent form.

The samples were collected in two stages, on May 22, 17 and May 29, 2017, quickly and non-invasively, with the aid of a tongue depressor gently pressed over the tongue and a sterile swab that was passed over the tonsils and near the uvula. The material was transported in a tube with the swab in sterile Stuart transport medium (model K41-01-02, OLEN), packed in a thermally insulated box and sent to the Clinical Analysis laboratory of the University Hospital of Western Paraná, Bacteriology department. In order to reduce errors in harvesting, transport and storage, the researchers assigned to the harvesting were trained to obtain the sample in an appropriate place, avoiding contamination, proper preservation and transportation of the material.

The samples were seeded by depletion technique in Petri dishes containing sheep blood agar. After sowing, the bacitracin and sulfamethoxazole-trimethoprim discs were placed on the middle. This technique was performed to differentiate \(S. pyogenes\) from other group A strains and from other species with \(\beta\)-hemolytic colonies, i.e. strains of \(S. pyogenes\) are bacitracin-sensitive (halo around the disc) and resistant to sulfamethoxazole trimethoprim (do not form halo). The seeded material was incubated for 24 to 48 hours at 35-37°C at a 5% CO\(_2\) stress. After the incubation period, the plates were visually analyzed for growth of colonies that exhibited beta-hemolysis characteristics, as well as halo formation around bacitracin and sulfamethoxazole-trimethoprim discs. Then, the colonies with phenotypic characteristics compatible with \(S. pyogenes\) were tested for the production of catalase enzyme, the \(S. pyogenes\) being catalase negative. Finally, those catalase negative colonies were identified by the PYR test. This is a disk test based on the enzymatic hydrolysis of L-Pyrrolidonyl-Beta-Naphthylamide, a rapid, accurate and specific test for identifying the \(S. pyogenes\). For quality control purposes, a positive \(S. pyogenes\) ATCC 19615 Newprov` strain growth was seeded under the same conditions as the participants’ oropharyngeal secretion culture. All tests were done in duplicate.

The following variables were analyzed: age, gender and signs of pharyngotonsillitis (purulent plaques in the oropharynx). The results were quantitatively described as absolute frequency and percentage distribution.

This study was carried out with the approval of the Unioeste Research Ethics Committee (CEP) and the National Health Council (CONEP), protocol No. 1.775.058. The study’s scientific merit was obtained from the National Council for Scientific and Technological Development, permit to visit the indigenous land was given by the National Indian Foundation (FUNAI), together with a consent from the chief of the tribe and the nurse responsible for the local indigenous health to carry out this research.
RESULTS

We collected 127 samples, 53 (41.7%) from males and 74 (58.3%) from females. The largest number of samples were collected from children between four and eleven years of age (58 samples); among those between 12 and 18 years we collected 18 samples, and 51 samples from adults older than 18 years. (Table 1).

None of the participants had signs of acute pharyngotonsillitis during sample harvesting.

Of the 127 samples that were microbiologically assessed, 2 had characteristics that made us suspect of S. pyogenes, that is: beta hemolysis, resistance to sulfamethoxazole-trimethoprim, sensitivity to bacitracin and negative catalysis. Both samples were the samples submitted to the fast PYR test; however, they showed negative results, indicating the samples were not S. pyogenes.

DISCUSSION

There are no studies in Brazil showing the prevalence of S. pyogenes in the native population. This study showed that there was no S. pyogenes in the oropharynx secretion of natives from the Tekohá Jevy tribe. Notwithstanding, the outcome of this study requires caution, because the natives, although having characteristics that place them in a position of vulnerability, such as poverty, low income per capita, poor dwellings and less access to healthcare. They work and go to school in their own village, thus having less contact with non-native individuals, which may be a protection factor for the population, with a lower rate of contamination.

Studies estimate that 15% of school-age children in developed countries will develop a symptomatic case of streptococcal pharyngitis each year, while the incidence of S. pyogenes pharyngitis in developing countries may be five to ten times higher. The prevalence of S. pyogenes is admittedly higher in children and adolescents, but streptococci may occur at any age. A study conducted in two public schools in Pernambuco - Recife with 753 schoolchildren aged 5 to 19 years showed a prevalence of S. pyogenes of 0.8% in asymptomatic carriers. Another study conducted in Bamako, Mali with children aged 5 to 16 years isolated the GABS in 25.5% of pharyngitis episodes. Schools and closed institutions with large numbers of children are believed to represent a risk factor for bacterial spread and contamination, as shown in a study carried out in Roraima and São Paulo comparing the prevalence among asymptomatic and non-institutionalized children, which showed an 8% prevalence in oropharyngeal cultures of children attending day care centers and 2% in the control group (not children attending day care centers) in the city of São Paulo, which difference was statistically significant.

S. pyogenes was present in 24% of the oropharyngeal cultures of children in day care centers and in 16% of the control group cultures from the city of Porto Velho in 2006. A study carried out with 114 healthcare undergraduate students from a University Center University in the city of Maringá, Paraná, showed zero prevalence of S. pyogenes in the oropharynx secretion of the aforementioned students. Another study involving four to 15-year-old children from a rural school in Argentina found a prevalence of 13% of S. pyogenes in asymptomatic carriers, and showed that asymptomatic carriers were 5.6 times more likely to develop acute pharyngotonsillitis.

Our study concludes that the studied population of the indigenous village of Tekohá Jevy did not present S. pyogenes in their oropharynx, which may mean a lower rate of contamination and dissemination of this bacterium in the place. This matter requires further studies, since no data was found in Brazil regarding the prevalence of S. pyogenes in isolated populations such as indigenous peoples.

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Table 1. Descriptive analysis of the study variables.

| Study variables | N  | %   |
|-----------------|----|-----|
| Gender          |    |     |
| Males           | 53 | 41.7|
| Females         | 74 | 58.3|
| Age             |    |     |
| 4-11 years      | 58 | 45.7|
| 12-18 years     | 18 | 14.2|
| > 18 years      | 51 | 40.1|
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