

International Journal of Pharmacology and Toxicology

Journal home page: www.sciencepubco.com/index.php/IJPT doi: 10.14419/ijpt.v2i2.3303 Research Paper



Isolation of Streptococcus species from throat swabs of school going children

Shobha K.L¹*, Kusumakshi², Anand K.M¹, Gowrish Rao.S¹, Jessica D'souza¹, Ramachandra L³

¹ Department of Microbiology, Melaka Manipal Medical College (Manipal Campus), Manipal University, Manipal
² Department of Medical Laboratory Technology, Manipal College of Allied health Sciences, Manipal University, Manipal
³ Department of Surgery, Kasturba Medical College, Manipal University, Manipal

*Corresponding author E-mail: shobha.kl@manipal.edu

*Corresponding duinor E-mail: shoond.ki@manipai.ed

Abstract

Introduction : Group A Streptococcus (Streptococcus pyogenes), is a Gram positive coccus appearing in chains and causes an incredible history of changing disease pattern. It has numerous virulence factors that helps in the adherence of the tissues, destruction of tissues and resulting in causing autoimmune complications. An attempt was made to screen for Streptococcus pyogenes throat carriers among school going children of Udupi district, Karnataka State, India.

Materials and Methods: Throat swabs were collected from posterior pharynx. Swabs were immediately plated onto a sheep blood agar (Hi-Media,Mumbai,India) and transported to the laboratory within one hour of collection. The inoculated plates were incubated at 37° C for 24- 48 hours in carbondioxide incubator. Organism grown on colonies were identified up to species level.

Result: A total of 235 children were screened for Streptococcus pyogenes. They were in the age group of 6-11 years. None of the children had Streptococcus pyogenes.

Conclusion: Streptococcus pyogenes was not found from the throat samples in school going children More studies are required to establish an accurate value of prevalence with a larger sample size.

Key words: Gram positive cocci in chains, Streptococcus pyogenes, school going children, throat swab

1. Introduction

Group a Streptococcus (Streptococcus pyogenes), is a Gram positive coccus appearing in chains and causes an incredible history of changing disease pattern (Cunningham MW et al, 2000; 13:470-511). It has numerous virulence factors that helps in the adherence of the tissues, destruction of tissues and resulting in causing autoimmune complications (Heath A et al, 1999; 67:5298-305) .Proteomic detection of Streptococcus pyogenes has identified 79 proteins on its surface and 21 cytoplasmic proteins (Severin A et al 2007, 189; 1514-22) .Streptococcal adherence to host pharyngeal epithelium is the basic step in colonization. Environmental conditions, cell density and growth phase are known to influence expression of virulence factors (McIver KS et al 1995; 63; 4540-2 & Chaussee MA et al 2008; 189; 27-41). High prevalence of rheumatic fever/rheumatic heart disease in India and the association of pharyngitis that ultimately leading to RF/RHD is not studied from this place. Therefore, an attempt was made to screen for Streptococcus pyogenes throat carriers among school going children of Udupi district, Karnataka State, India.

2. Materials and the methods

2.1. Materials

The swabs that were used were prepared as follows. Pre-sterilized wooden sticks were spun with sterile cotton manually. The swabs

were sterilized in hot air oven (holding time of 2 hours at 160 °C)[.] The sterility of swabs was ensured by plating randomly selected swabs to Sheep blood agar. No growth was seen after 72 hrs of incubation at 37^0 C.

Consent from the parents was taken for the collection of the throat samples. Exclusion criteria included children whose parents did not give consent to collect the throat sample and children, who did not co-operative for the sample collection. Throat swabs were collected from posterior pharynx as suggested by H. Nsanze, et al; 1998; 260-264). Swabs were immediately plated onto a sheep blood agar (Hi-Media,Mumbai,India) and transported to the laboratory within one hour of collection. The inoculated plates were incubated at 37^oC for 24- 48 hours in carbondioxide incubator at 5% carbondioxide. A bacitracin disc (0.04 units) (Hi Media,Mumbai,India) was placed in the plate directly on the culture plate (Dawson KT et al; 1996,16:123-7) before placing the blood agar plate in the carbon-di-oxide incubator. Colonies were identified for beta haemolysis (Webcls procedure manual 2010).

Gram stain smears were made from the colonies (Hucker's modification). The colonies showing morphology of streptococcus were further identified to species level and production of PYRase was tested. Grouping of Streptococcus was done using Slidex Strepto-Kit (BioMeieux,France) (Koneman et al;2006)

2.2. Results

A total of 235 children were screened for Streptococcus pyogenes. They were in the age group of 6- 11 years. 132 (56.17%) were

Copyright © 2014 Shobha K.L et al. This is an open access article distributed under the <u>Creative Commons Attribution License</u>, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

male children and 103(43.82%) were female children None of the samples collected from these children showed any growth on sheep blood agar and the prevalence estimated was zero. Fig 1; Distribution of male and female children

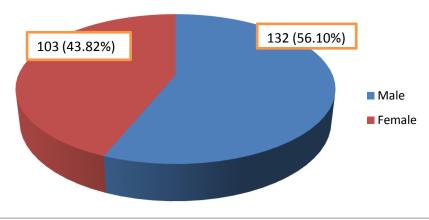


Fig. 1: Distribution of male and female children

Table 1: Age Wise Distribution of Children		
Age in Years	Male children	Female children
6-7	35	20
7-8	28	21
8-9	26	23
9-10	20	19
10-11	23	20
Total; 235	132	103

3. Discussion

The surprising finding was that all the screened subjects were negative for the Streptococcus pyogenes throat carriers. A study in India showed 8 of 310 subjects (2%) was throat carriers. The same study also concluded that the prevalence was high with 0.6% prevalence of rheumatic heart disease (Thangam Menon et al 2004; 171-173). In comparison our prevalence was nil in the group studied. The possible reason could be that our study had only healthy students in comparison to their study on subjects who had acute respiratory infections. Axena Haggar et al ;2012 in their study regarding the Streptococcus pyogenes causing invasive spread found that North India had higher incidence than South India.(Kumar R et al;2009) in their study reported in 1.3% of children between the age group of 5-15 years, again this study was from North India. Our study did not have any case of Streptococcus carriers which indicates that the organism is not found frequently as carriers.

4. Conclusion

We conclude that the prevalence of Streptococcus pyogenes in the school going children in this area of Udupi District is possibly zero. More studies are required to establish an accurate value of prevalence with a larger sample size.

References

- [1] Cunningham MW. Pathogenesis of group A streptococcal infections. Clin Microbiol Rev. 2000; 13:470–511. http://dx.doi.org/10.1128/CMR.13.3.470-511.2000.
- [2] Heath A, Rita VJ, Barg NL, Engleberg NC. A two component regulatory system, CsrR-CsrS, represses expression of three Streptococcus pyogenes virulence factors, hyaluronic acid capsule, slo and pyrogenic exotoxin B. Infect Immun. 1999; 67:5298–305.
- [3] Severin A, Nickbarg E, Wooters J, Quazi SA, Matsuka YV, Murphy E, et al. Proteomic analysis and identification of Streptococcus pyogenes surface-associated proteins. J Bacteriol. 2007; 189:1514–22 http://dx.doi.org/10.1128/JB.01132-06.

- [4] McIver KS, Heath AS, Scott JR. Regulation of virulence by environmental signals in group a streptococci: influence of osmolarity, temperature, gas exchange and iron limitation of emm transcription. Infect Immun. 1995;63:4540–2
- [5] Chaussee MA, Dmitriev AV, Callegari EA, Chaussee MS. Growth phase-associated changes in the transcriptome and proteome of Streptococcus pyogenes. Arch Microbiol. 2008; 189:27–41 http://dx.doi.org/10.1007/s00203-007-0290-1.
- [6] H. Nsanze, K. Dawson, A.S. Ameen and N. Mustafa. Group A streptococcal antigen detection in schoolchildren; Eastern Mediterranean Health Journal, Volume 4 issue 8, 1998, Page 260-264.
- [7] Bacitracin (taxo a) disk for beta-hemolytic streptococcus, WEBCLS procedure manual; http://webcls.utmb.edu/samplecourses/bact/Bact/procedures/bacitracin diskgrpastrep.htm.2010.
- [8] Dawson KP et al. The prevalence of group a streptococcal throat carriage in Al-Ain, United Arab Emirates. Annals of tropical pediatrics, 1996, 16:123-7.
- [9] Henry D Isenberg, Clinical Microbiology procedure handbook; 2010; ASM press. http://biology.clc.uc.edu/fankhauser/Labs/Microbiology/Gram_Stain/
- http://biology.clc.uc.edu/fankhauser/Labs/Microbiology/Gram_Stain/ Gram_stain.htm.
- [10]Koneman's color atlas and textbook of diagnostic microbiology;Lippincott Williams.2006; 6th Edition;674-688.
- [11] Thangam Menon, S. Shanmugasundaram*, M. Palani Kumar & C.P. Girish Kumar; Group A streptococcal infections of the pharynx in a rural population in south India, Indian J Med Res 119 (Suppl) May 2004, pp 171-173.
- [12] Axana Haggar, Andreas Nerlich, Rajesh Kumar, Vinod JAbraham, Kootallur N Brahmadathan, Pallab Ray, Vanita Dhanda, John Melbin Jose Joshua, Narinder Mehra, Rene Bergmann, G. Singh Chhatwal and Anna Norrby-Teglund; Clinical and Microbiologic characteristics of invasive Streptococcus pyogenes in North and South India; Journal of Clin Microbiol. May 2012, Vol 50, no5 1626-1631 http://dx.doi.org/10.1128/JCM.06697-11.
- [13] R.Kumar, H.Vohra, A.Chakraborty, Y.P.Sharma, S.Bandhopadhya, V.Dh anda, V.Sagar, M.Sharma, B.Shah and N.K Ganguly; Epidemiology of group A Streptococcal pharyngitis and impetigo; A cross-sectional and follow up study in a rural community of Northern India; Indian Journal Of Medical research; 130, December 2009, 765-771.