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Characterisation of Streptococcal Isolates from a Tertiary Care Centre –A One Year Study

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ABSTRACT
Streptococci are significant human pathogens causing wide spectra of disease ranging from uncomplicated infections to serious life threatening diseases. 212 isolates were studied from various clinical specimens from March 2012 to February 2013. The epidemiology of infections caused by β-hemolytic streptococci has shown a major change with increasing incidence of invasive and soft tissue infections. The emergence of antimicrobial resistance of streptococci and enterococci pose a major challenge to treating physicians.
KEY WORDS: *Streptococci, Enterococci.*

Introduction

Streptococcal infections are highly prevalent among all age groups throughout the world. They represent an important health and economic problem. This ubiquitous organism is responsible for a wide array of local and systemic infections. Streptococcus produces a wide range of invasive infection like streptococcal toxic shock syndrome (STSS), necrotizing fasciitis, pneumonia, and bacteremia and non invasive infections like pharyngitis and impetigo.

Aim of the study was

1. To study the isolation rate of Streptococci from various specimens & to characterize them.
2. To analyze the resistance pattern to antibiotics among various species of Streptococci.

Streptococci isolated from various clinical specimens in Central Microbiology Laboratory, Medical College Hospital and, Sree Avittom Thirunal (SAT) Hospital, Thiruvananthapuram during the period March 2012 to February 2013 were included in the study.

Materials and Methods

Streptococci (isolated from various clinical specimens) were inoculated into 5% Sheep blood agar, Mac Conkey agar, Tellurite blood agar & Glucose broth.

Identification was based on the growth characteristics on culture medium, Gram staining, Catalase test, Bacitracin sensitivity test, PYR Test, CAMP test and Sugar fermentation tests. Aesculin hydrolysis, Salt tolerance test, Heat resistance test & Arginine Hydrolysis test were done for Enterococci isolated.[1]

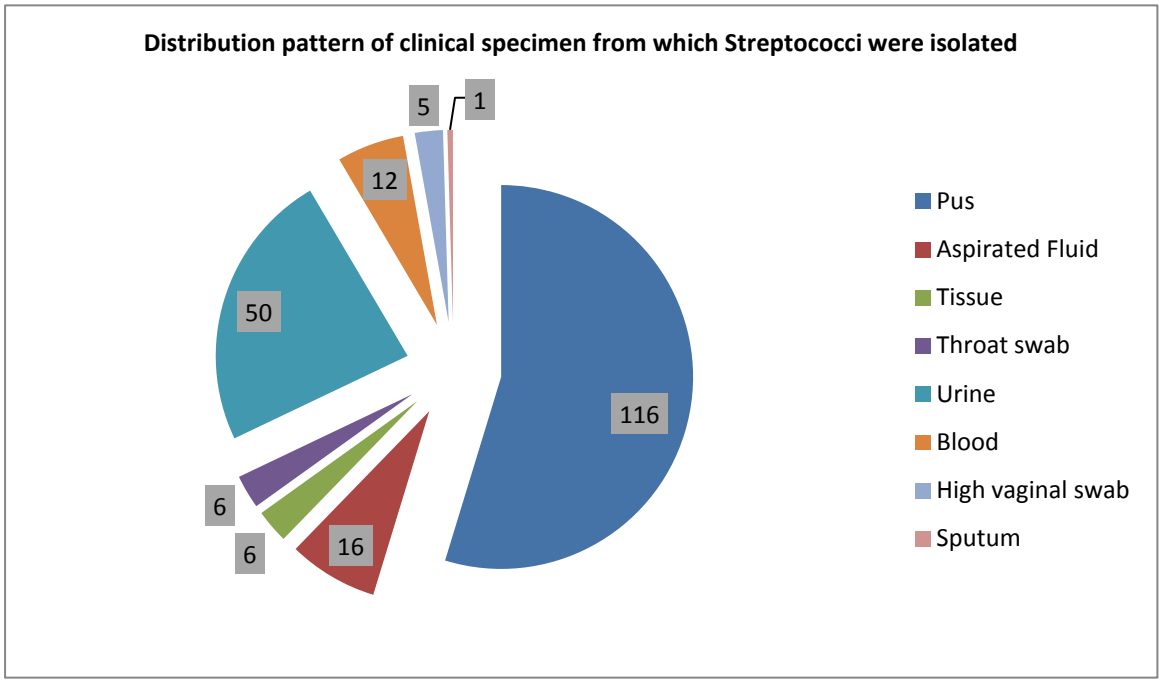
Results

The total number of streptococci isolated during the study period was 212. Among the isolates, Group D Streptococci (GDS) predominated 94(44.3%), followed by Group A Streptococci (GAS) 47 (22.2%), (GBS) 31 (14.6%), Ungroupable Streptococci 24 (11.3%) and the rest Group G Streptococci (GGS) 8 (3.8%), Group F Streptococci (GFS) 7 (3.3%) and Group C Streptococci (GCS) 1 (0.5%).

Table 1. Distribution pattern of various groups of Streptococci in the study group

Group	Number	Percentage
A	47	22.2%
B	31	14.6%
C	1	0.5%
D	94	44.3%
F	7	3.3%
G	8	3.8%
Ungroupable	24	11.3%
Total	212	100%

Out of total 212 isolates, 116(54.71%) were from pus and 50 (23.58%) from urine and the rest were from aspirated fluid, tissue, throat swab, blood and vaginal swab and sputum (Figure1).



Most of GAS 35 (74.46%) and GBS 13 (41.93%) were isolated from skin and soft tissue infection and five (10.63%) were isolated from tonsillitis.

Out of 31 GBS, 13(41.93%) were from skin and soft tissue infections. (Fig2)

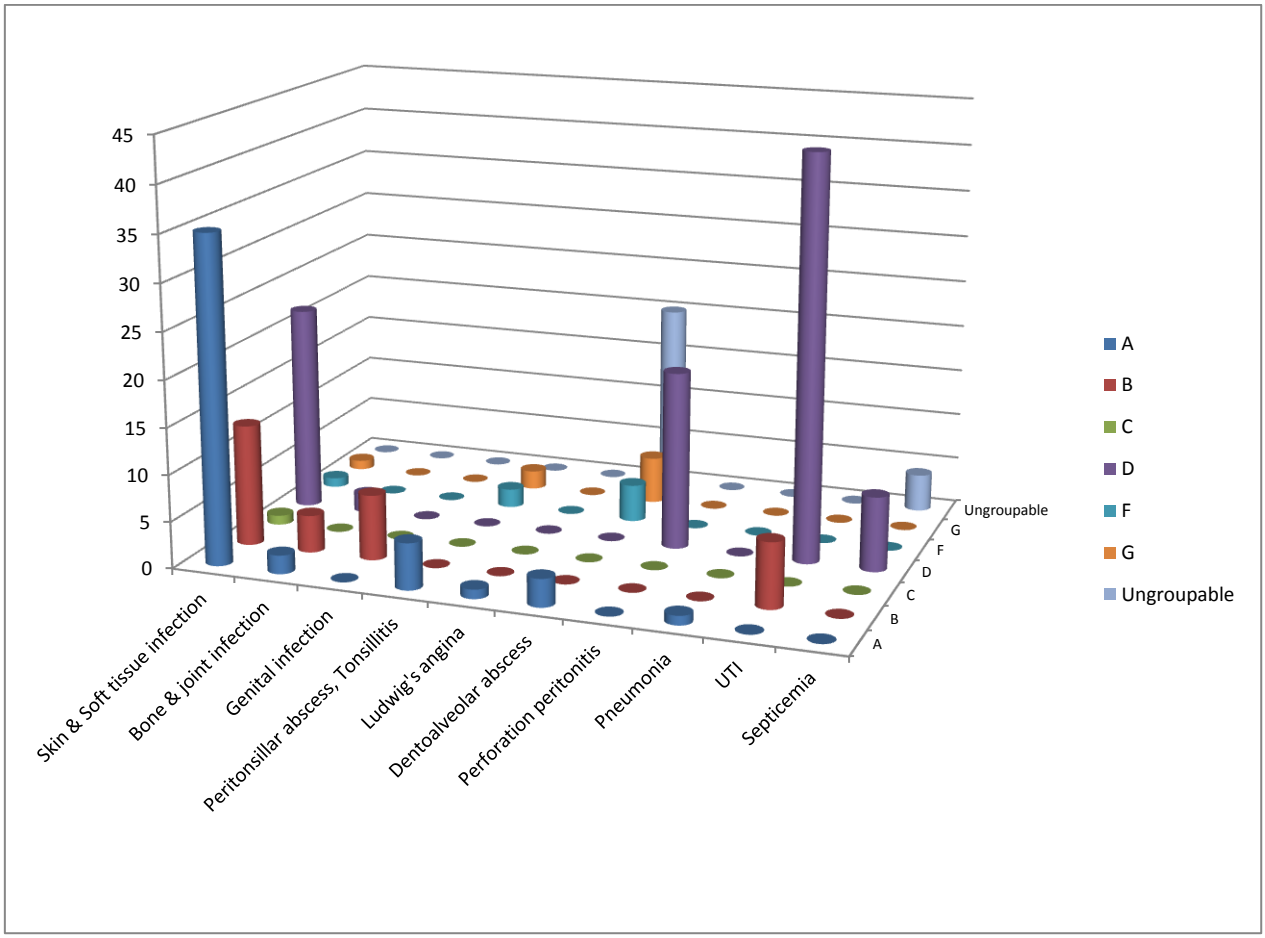


Figure 2: Distribution of Streptococci from various clinical conditions.

Streptococcal infections were more common in males than in females. Out of 212 isolates 131(61.79) were isolated from male patients and 81(38.21) were isolated from female patients. Among the Enterococci isolated, 80(85.10) belonged to Enterococcus faecalis and 14(14.89) belonged to Enterococcus faecium**.**

Table 2. Bacitracin sensitivity of Streptococci

Group	No. of strains	Bacitracin 0.04U Sensitive	Bacitracin 0.04U Resistant
A	47	47	0
B	31	0	31
C	1	1	0
D	94	0	94
F	7	0	7
G	8	2	6
Ungroupable	24	0	24

All GAS were bacitracin sensitive. One GCS and two GGS were also sensitive to bacitracin (Table2). All GBS were CAMP test positive. All strains were sensitive to Penicillin and Cephalosporin. 85.10% of group A Streptococci, 90.32% of group B Streptococci, 85% of group D Streptococci and 87.5% of ungroupable Streptococci were sensitive to Erythromycin. 85% of group D Streptococci were sensitive to Ampicillin and Erythromycin, 97.87% were sensitive to Vancomycin. (Table 3)

Table 3: Antibiotic sensitivity of the streptococcal isolates

Groups	No. of strains tested	Penicillin	%	Ampicillin	%	Cephalosporin	%	Erythromycin	%	Vancomycin	%
A	47	47	100	NA		47	100	40	85.10	NA	NA
B	31	31	100	NA		31	100	28	90.32	NA	NA
C	1	1	100	NA		1	100	1	100	NA	NA
D	94	NA	NA	80	85	NA	NA	80	85	92	97.87
F	7	7	100	NA		7	100	7	100	NA	NA
G	8	8	100	NA		8	100	8	100	NA	NA
Un-group able	24	24	100	NA		24	100	21	87.5	NA	NA

Discussion

Streptococci are significant human pathogens causing a wide spectrum of disease, ranging from uncomplicated infections to serious life threatening diseases. 212 Streptococcal strains were collected and characterized in the study. GDS predominated among the streptococcal isolates in contrast to a study done by Dr.Sarada Devi, in the same setting in 1994 which showed the following percentage of isolates; GAS 15.55%, GBS 4.4%, GCS 1.1%, GDS 8.33%, GFS 5%,GGS 3.88%, Ungroupable 15% and not grouped (46.6%)[2].

The distribution of isolates in clinical specimens in this study almost correlates with the study done by Le Hello S etal at New Caledonia 2006. Skin and soft-tissue infections such as necrotizing fasciitis, myositis, erysipelas, and bacteremia were the most frequent clinical manifestations.[3]

In the present study two cases were isolated from Varicella zoster skin infection. Tyrrell GJ et al in his study isolated 14 cases of invasive GAS in children with Varicella.[4] In the present study GAS accounted for 40 cases of septic arthritis by Streptococci.

In the present study out of 31GBS, 41.93% were from skin and soft tissue infection, 12.9% from bone and joint infection, 22.58% from genital infection and 22.58% from UTI. Studies conducted by Lee et al and Farley et al also observed skin and soft tissue infections as the most common presentations of invasive Streptococcus agalactiae infections.[5,6] In the present study GBS were also isolated from 7 cases of urinary tract infections. Girgitzova B et al (1991) reports group B Streptococci as an important cause of urinary tract infection after group D Streptococci in adult patients.[7]

Group C and Group G streptococci together accounted for about 4.8 percent of skin and soft tissue infections. This is almost in accordance to the study done by Mathur Pet al at AIIMS, New Delhi in 2003, where they reported Group C and G Streptococcal skin infection as 3 percent[8].

Group F Streptococci were isolated from seven cases out of which 14.28% were from skin and soft tissue infections, 28.57% from peritonsillar abscess and 57.14% from dentoalveolar abscess. Dr.Sarada devi reports isolation of GFS from two cases of liver abscess, two cases of brain abscess and intraabdominal and perianal infections[2]. Out of 94 GDS, 23.34% were from skin and soft tissue infection, 45.74% from UTI, 20.3% from perforation peritonitis, 8.5% from septicemia and 2.12% from bone and joint infections.

In the present study 85.10% of Enterococci belonged to Enterococcus faecalis species and 14.89% to Enterococcus faecium species. This

almost correlates with the study by Bose S et al in which 82% were Enterococcus faecalis and 18%, Enterococcus faecium.[9]

In the present study, Streptococcal infections were more common in males than in females. Out of 212 isolates 61.79% were isolated from male patients and 38.21% were isolated from female patients. Mathur Pet al (2002) at All India Institute of Medical Sciences, Le Hello S et al, also reports higher isolation of Streptococci in males[3,8].

Bacitracin susceptibility test was commonly used for presumptive identification of Streptococcus pyogenes (group A Streptococci). The presumptive identification is improved by co-trimoxazole disc as S. pyogenes is resistant and many other Bacitracin susceptible Streptococci are susceptible to cotrimoxazole. In the present study, out of 47 group A Streptococci tested, all (100%) were susceptible to bacitracin, 25% group G Streptococci and one group C Streptococci were also bacitracin susceptible. All group A Streptococci were resistant to cotrimoxazole whereas group G and C were susceptible. This correlates with the study done by Dr. Sarada Devi in which all Group A (28) and two group G Streptococci were susceptible to Bacitracin.[2]

In the present study all GAS (47), GBS (31), GCS (1), GFS (7), GGS (8) and Ungroupable Streptococci (24) were sensitive to Penicillin and Cephalosporin. 85.10% GAS, 90.32% GBS, 100% GCS, 100% GFS, 100% GGS, 87.5% Ungroupable Streptococci were sensitive to Erythromycin. Out of 94 Enterococci 85% were sensitive to Ampicillin and Erythromycin and 97.87% were sensitive to Vancomycin.

In present study, all GAS were sensitive to Penicillin. 14.9% of GAS was resistant to Erythromycin. All GBS were sensitive to Penicillin and Erythromycin resistance was 9.68%. All group C, GFS and GGS were susceptible to Penicillin, Erythromycin and Cephalosporin. In a study done by Devi U, among total six group C Streptococci 100% were susceptible to erythromycin and penicillin.[10] In the study done by Broyles LN et al on antibiotic susceptibility testing on 31 group C and 180 group G, there was no resistance to Penicillin, Ampicillin, and first generation cephalosporins, but Erythromycin resistance was noted in 61 isolates (29%).[11]

Out of 94 Enterococci, 15% were resistant to Ampicillin and Erythromycin, and 2.13% were resistant to Vancomycin. In similar studies from Lady Hardinge Medical College, New Delhi, Chandigarh and Mumbai reported the percentage of VRE are 8, 5.5 and 23 per cent respectively [12]. Bose S et al found out that the Enterococcal isolates from various clinical samples were 100% susceptible to vancomycin[9]

Conclusion

The epidemiology of infections caused by beta hemolytic Streptococci have shown a major change in the past one decade with increase in the incidence of invasive and soft tissue infections.

All groups of Streptococci were sensitive to Penicillin and Cephalosporin reinforcing the fact that the penicillin resistance has not yet developed in this organism in spite of frequent penicillin use. 14.9% of group A Streptococci, 9.68% of group B Streptococci, Streptococci, 12.5% of Ungroupable Streptococci were resistant to Erythromycin. 15% of Enterococci were resistant to Ampicillin and Erythromycin, 2.13% were resistant to Vancomycin. The emergence of antimicrobial resistance in Streptococci to macrolides necessitates strict surveillance for these infections. Enterococci which have traditionally been regarded as low grade pathogens have emerged as an increasingly important cause of nosocomial infections in the last decade. The emergence of vancomycin-resistant Enterococcal (VRE) is a cause of concern, as once established, it is very difficult to control posing a threat to the patient safety and also challenges for the treating physicians.

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