Original Research Article

A study of the microbial spectrum involved in skin and soft tissue infections in infants at tertiary health care center

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Abstract

Background: Skin and soft tissue infections (SSTIs), most commonly cellulitis or cutaneous abscess, are one of the most common infections encountered in the pediatric age group, especially in the developing world. If diagnosed early and treated appropriately, these infections are almost always curable, but some have the potential to cause serious complications such as septicemia, nephritis, carditis and arthritis if diagnosis is delayed and/or treatment is inadequate. Present study was undertaken to study microbial spectrum in skin and soft tissue infections in infants at tertiary health care center. **Material and Methods:** Present prospective study was undertaken in children less than one year of age with clinical features suggestive of skin and soft tissue infection. **Results:** A total of 250 children were included in the present study. 143 (57.2%) were male and 107 (42.8%) were female. Heat 248 (99.2%), redness 246 (98.4%) and localized swelling 240 (96%) were the commonest clinical signs present. The other signs present were localized tenderness 209 (83.6%), purulent drainage 177 (70.8%), fever 170 (68%) and abscess 74 (29.6%) of cases. CRP was positive in 38 (15.2%) cases and negative in majority, i.e., 212 (84.8%) of the cases. The blood culture was positive in only 32 (12.8%) cases whereas in remaining 218 (87.2%) cases it was negative. Pus culture was positive in 145 (58%) cases and there was no growth in 105 (42%) samples. In one case (0.4%) acid fast bacilli were seen. Out of 152 infections in the cases of SSTIs in present study, 74 (48.68%) were gram positive and 78 (51.31%) were gram negative bacteria. Polymicrobial infection was noted in 4 cases. Out of 152 bacterial isolates, 59 (38.81%) were *Staphylococcus aureus*, 25 (16.44%) were *Escherichia coli*, 14 (9.21%) were *Enterobacter* spp. and 13 (8.55%) were *Pseudomonas aeruginosa*. Of the 74 gram positive organisms, 59 (79.72%) were *Staphylococcus aureus*, 5 (6.75%) were *Streptococcus* spp., 6 (8.13%) were Micrococci, 3 (4.05%) were Diphtheroids and one (1.35%) was *Enterococcus* spp. Among 59 *Staphylococcus aureus* isolates, 33 (55.93%) were found to be methicillin resistant and 26 (44.06%) were methicillin sensitive. **Conclusion:** In present study the main pathogens involved in these infections are Staphylococcus aureus and gram negative enteric organisms.

Keywords: Staphylococcus aureus, gram negative enteric organisms, MRSA, SSTIs

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Received Date: 08/01/2021 Revised Date: 12/03/2021 Accepted Date: 28/04/2021
DOI: https://doi.org/10.26611/10081811

Access this article online	
Quick Response Code:	Website: www.medpulse.in
	Accessed Date: 04 April 2021

INTRODUCTION

Skin and soft tissue infections (SSTIs), most commonly cellulitis or cutaneous abscess, are one of the most common infections encountered in the pediatric age group, especially in the developing world.[1,2] SSTIs can be defined as an inflammatory microbial invasion of the epidermis, dermis and subcutaneous tissues.[3] It can be classified as 'superficial' (epidermis and dermis) and 'deep' (hypodermis, fascia and muscle).[4] Bacterial skin and skin structure infections commonly encountered in children include impetigo, folliculitis, furunculosis, carbuncles,

How to cite this article: Ravidas Arjun Vasave, Shripad Taklikar, Sujata Baveja. A study of the microbial spectrum involved in skin and soft tissue infections in infants at tertiary health care center. *MedPulse International Journal of Microbiology.* April 2021;18(1): 01-06. https://www.medpulse.in/Microbiology/

wound infections, abscesses, cellulitis, erysipelas, and staphylococcal scalded skin syndrome. These infections in infants occur when skin integrity is compromised as a result of change in bacterial load on the skin, the availability of bacterial nutrients, excessive skin moisture due to daily oil massage. Poor hygiene, physical contacts and crowded conditions in day care centers facilitate the spread of contagious infections such as furuncles, carbuncles, and impetigo.[2] Colonization with Staphylococcus aureus in the anterior nares and Streptococcus pyogenes on the skin increases the risk of skin infections. Skin-to-skin contact from attendance at day care is a risk factor for community-acquired methicillin resistant Staphylococcus aureus (CA-MRSA) skin infections. Risk factors for health care–associated methicillin-resistant Staphylococcus aureus(HA-MRSA) skin infections include recent exposure to antibiotics or the health care system.[5] If diagnosed early and treated appropriately, these infections are almost always curable, but some have the potential to cause serious complications such as septicemia, nephritis, carditis and arthritis if diagnosis is delayed and/or treatment is inadequate. Therefore, this study was undertaken to study microbial spectrum in skin and soft tissue infections in infants at tertiary health care center

MATERIAL AND METHODS

Present prospective study was undertaken at Lokmanya Tilak municipal Medical College Sion Mumbai, over a period of 1 year 6 months. Study was approved by institutional ethical committee.

Inclusion Criteria

- Children less than one year of age with clinical features suggestive of skin and soft tissue infection.

Exclusion Criteria

- The participant may not enter the trial if hospital Acquired Infections occurring after 48 hours of admission.

Study was explained to parents and a written informed consent was taken for participation. Diagnosis of skin and soft tissue infection was made on the basis of following clinical features - local signs of redness, warmth, swelling, pain with or without dysfunction, with presence of bullae, haemorrhage, rapidly progressive in nature, crepitus.

Pus or exudate was collected from the depth of the lesion by either aspiration or using at least two sterile cotton swabs after cleaning the wound with sterile normal saline and surrounding skin with alcohol. Care was taken not to touch the adjacent skin margins. Then the swabs were transported in sterile cotton plugged test tubes.

After reaching the laboratory the swab was inoculated on Blood agar, and MacConkey agar plates and these plates were incubated aerobically at 37 for 24-48 hours. After 24 hrs. of incubation, plates were examined for any growth and plates showing no growth were incubated further for next 24 hrs. The plates are discarded if there was no growth after 48 hrs. From the plates showing growth, secondary smears were prepared and biochemical tests were put up for identification. All the aerobic bacteria were identified by Gram stain morphology, cultural characteristics, pigment production, haemolysis on blood agar, motility and conventional biochemical tests for aerobic bacteria as per standard guidelines For lactose fermenting and non-lactose fermenting colonies, further Hanging drop for motility and catalase and oxidase tests were done. After which the following biochemical tests were performed. Indole test, Methyl Red test, Voges Proskauer test, Citrate utilisation test, Urease hydrolysis test, Nitrate reduction test, Phenylalanine deaminase test, Triple sugar iron agar test, Hugh-Leifson oxidation fermentation test, Sugar fermentation tests. For gram-positive cocci catalase test was done first. For catalase positive cocci Slide and tube coagulase tests, Mannitol fermentation test, Hugh-Leifson oxidation fermentation test, Furazolidone sensitivity tests, Phosphatase test were done. For gram positive cocci in pairs, short chains and catalase negative bacilli, tests such as growth at 45^0c, growth in 6.5% NaCl, bile esculin hydrolysis, bacitracin sensitivity, optochin sensitivity were done. The blood that was sent in brain heart infusion broth was incubated at 37ºC for 24 hours. Subculture was done on Blood agar and MacConkey's agar after 24 hours incubation. These plates were incubated at 37ºC for 24 hours. If no growth, subculture was repeated for 5 days Identification of isolates was done by cultural characteristics and standard biochemical tests. The isolates were subjected for antibiotic susceptibility testing by employing Kirby Bauer disc diffusion technique as recommended by Clinical and Laboratory Standards Institute (CLSI). Patients were observed till discharge from the hospital in case admitted for the procedure for removal of pus. Data was entered into Microsoft excel data sheet and was analyzed using SPSS 22 version software. Categorical data was represented in the form of Frequencies and proportions. Chi-square test or Fischer's exact test was used as test of significance for qualitative data, p value of <0.05 was considered as statistically significant.

RESULTS

A total of 250 children were included in the present study. 143 (57.2%) were male and 107 (42.8%) were female. Heat 248 (99.2%), redness 246 (98.4%) and localized swelling 240 (96%) were the commonest clinical signs present. The other signs present were localized tenderness 209 (83.6%), purulent drainage 177 (70.8%), fever 170 (68%) and abscess 74 (29.6%) of cases.

Table 1: General characteristics

Characteristics	No. of cases	Percentage
Sex		
Male	143	57.2
Female	107	42.8
Clinical signs		
Heat	248	99.2
Redness	246	98.4
Localized swelling	240	96
Localized tenderness	209	83.6
Purulent discharge	177	70.8
Fever	170	68
Abscess	74	29.6

Routine laboratory investigations were done in all cases of SSTIs to predict the risk of development of fulminant soft tissue infection. It was found that all laboratory values were within normal ranges and none of the patient had a risk of development of life threatening infection. CRP was positive in 38 (15.2%) cases and negative in majority, i.e., 212 (84.8%) of the cases. The blood culture was positive in only 32 (12.8%) cases whereas in remaining 218 (87.2%) cases it was negative. Pus culture was positive in 145 (58%) cases and there was no growth in 105 (42%) samples. In one case (0.4%) acid fast bacilli were seen. Out of 152 infections in the cases of SSTIs in present study, 74 (48.68%) were gram positive and 78 (51.31%) were gram negative bacteria. Polymicrobial infection was noted in 4 cases.

Table 2: Laboratory reports

Laboratory signs	Mean ± SD	
Hb (gm/dl)	10.86 ± 0.86	
TLC (per mm^3)	6404.24 ± 1133.47	
Platelet count (per µl)	249500 ± 79661.8	
BSL (mg/dl)	81.54 ± 9.97	
Serum Sodium (mEq/L)	139 ± 0.86	
Serum potassium (mEq/L)	4.01 ± 0.20	
Total protein (g/dl)	4.78 ± 0.31	
Albumin (g/dl)	3.04 ± 0.21	
Globulin (g/dl)	1.71 ± 0.30	
A/G ratio	1.64 ± 0.29	
CRP - Positive	38	15.2
Blood culture Positive	32	12.8
Pus culture Positive	145	58
AFB positive	1	0.4
Gram reaction	Frequency	Percentage
Gram positive	74	48.68
Gram negative	78	51.31

The three cases with history of animal bites showed growth of gram positive organisms in two cases and gram negative in one case. Trauma cases (n=199) showed growth of gram positive organisms in 56 cases and gram negative in 60 cases, 83 cases showed no growth on culture media. Out of 48 cases with history of body piercing, 22 were sterile, 11 showed growth of gram positive and 14 showed growth of gram negative organisms. Majority of the cases had history of daily massage with oils (n=237), of that 101 were sterile and 67 and 68 cases showed growth of gram positive and negative organisms respectively. Cases with history of previous hospitalization (n=35) showed growth of gram positive and negative organisms in seven and eight cases respectively.

Table 3: Distribution of risk factors and growth on culture media

Risk factors	No growth	Gram positive	Gram negative
Daily massage with oils	101	67	68
Trauma	83	56	60
Body piercing	22	11	14
Previous hospitalization	19	07	08
Animal bites	00	02	01

Chi Square=12.24; df=12; p>0.05; Not significant

Out of 152 bacterial isolates, 59 (38.81%) were *Staphylococcus aureus*, 25 (16.44%) were *Escherichia coli*, 14 (9.21%) were *Enterobacter* spp. and 13 (8.55%) were *Pseudomonas aeruginosa*. Of the 74 gram positive organisms, 59 (79.72%) were *Staphylococcus aureus*, 5 (6.75%) were *Streptococcus* spp., 6 (8.13%) were Micrococci, 3 (4.05%) were Diphtheroids and one (1.35%) was *Enterococcus* spp. Among 59 *Staphylococcus aureus* isolates, 33 (55.93%) were found to be methicillin resistant and 26 (44.06%) were methicillin sensitive. Among all the gram negative organisms (n=78) isolated, *Escherichia coli* were the commonest (32.05%) followed by *Enterobacter* spp. (17.94%), *Pseudomonas aeruginosa* (16.66%), *Klebsiella pneumoniae*. and *Acinetobacter* spp. (14.10% each) and *Citrobacter* spp., *Proteus mirabilis* and *Serratia* spp. (1.28% each). The Micrococci and Diphtheroids were considered as commensals and not processed further.

Table 5: Frequency of Microorganisms isolated

Organisms	Frequency	Percentage
Gram positive (n=74)		
Staphylococcus aureus	59	79.72
MRSA	33	55.93
MSSA	26	44.06
Streptococcus spp.	05	6.75
Enterococcus spp.	01	1.35
Micrococcus	06	8.13
Diphtheroids	03	4.05
Gram negative (n=78)		
Escherichia coli	25	32.05
Enterobacter spp.	14	17.94
Pseudomonas aeruginosa	13	16.66
Klebsiella pneumoniae.	11	14.10
Acinetobacter spp.	11	14.10
Citrobacter spp.	01	1.28
Proteus mirabilis	01	1.28
Serratia spp.	01	1.28
Mixed growth	04	5.12

DISCUSSION

The skin has an extremely diverse ecology of organisms that may produce infection. Microbial invasion of skin and underlying soft tissues leads to SSTIs. The clinical manifestations of SSTIs could be due to microbial invasion or an interaction of bacteria with host defense mechanisms. SSTIs are suppurative microbial invasions of the epidermis, dermis and subcutaneous tissues characterized by induration, erythema, warmth, and pain or tenderness. Local manifestations may be accompanied by systemic signs and symptoms, such as fever, chills, malaise and, at times, haemodynamic instability. Emergency department (ED) visits for skin and soft tissue infections in children have increased dramatically in the last decade.[6] Accordingly, from 1997 to 2009, hospital admission for pediatric patients with skin and soft tissue infections increased from 1.9 to 3.4 million annually.[7] During that same time, pediatric patients requiring incision and drainage have doubled.[7] This growing volume of patients is thought to be largely due to the emergence of community-acquired methicillin-resistant Staphylococcus aureus (CA-MRSA). In present study, out of 250 cases, 143 (57.2%) were male and 107 (42.8%) were female. Rani *et al.*[8] found SSTIs more commonly among males (56%) than females. In several other studies conducted by Ghadage DP *et al.*[9] and Andrews RM *et al.*[10] similar pattern of gender distribution was found. In the present study 94.8% cases had history of daily massage with some oils. Topical application of oils has not been shown to reduce the risk of infection or its associated morbidity or mortality, and may increase the risk of infection with coagulase-negative staphylococci in a study by Cleminson *et al.*[11] A wide range of signs and symptoms can be seen in patients with skin and soft tissue infections. Systemic signs

include hypotension and associated findings consistent with severe sepsis/septic shock including mental obtundation, cardiovascular and/or pulmonary collapse among other organ system failures. A study by Wall *et al.*[12] compared admission variables between patients with necrotizing soft tissue infections and those with non-necrotizing soft tissue infections. Those patients with either a white blood cell (WBC) count > 15,400 or a serum Na level <135 mmol/L were at higher risk of having a necrotizing soft tissue infections. The model is very sensitive but not very specific with a negative predictive value (NPV) of 99% and a positive predictive value (PPV) of only 26%. Clearly it is a valuable tool when negative (rules out necrotizing soft tissue infections) but when it is positive it does not confirm the diagnosis. Out of 156 cases, 152 (97.43%) were monomicrobial and 4 (2.56%) were polymicrobial infections. Among 152 monomicrobial infections, 74 (48.68%) were gram positive and 78 (51.31%) were gram negative bacteria. In a study by Rani *et al.*[8] 65 90% cases yielded growth of bacteria, out of which 71.85% were monomicrobial and 28.14% were polymicrobial infections. Of the 152 bacterial isolates in present study, 59 (38.81%) were Staphylococcus aureus, 25 (16.44%) were Escherichia coli, 14 (9.21%) were Enterobacter spp. and 13 (8.55%) were Pseudomonas aeruginosa. Mohanty *et al.*[13] reported Staphylococcus aureus(38.05%), Escherichia coli(17.39%) and Pseudomonas aeruginosa(11.82%) as the top three isolates in their study. They have reported incidence of Enterobacter spp. as 2.80% in their study. Zargar *et al.*[14] from India and Rennie *et al.*[15] and Sader *et al.*[16] from outside also reported these organisms among top five pathogens isolated from skin and soft tissue infections in hospitalized patients. Staphylococcus aureus is the almost-universal cause of furuncles, carbuncles, and skin abscesses and worldwide is the most commonly identified agent responsible for skin and soft tissue infections. The typical organisms that colonize the skin above the waist are usually Gram-positive species such as Staphylococcus epidermidis, Corynebacterium species, Staphylococcus aureus and Streptococcus pyogenes. The latter two species are particularly significant because they contribute to a majority of SSTIs. Staphylococcus aureus was the commonest isolate in our study. Many other investigators such as Ghadage *et al.*,[9] Mohanty *et al.*[13] Mathew *et al.*,[17] Baslas *et al.*, [18] Ahmed *et al.*,[19] and Sugeng *et al.*, [20] have similarly found Staphylococcus aureus to be the major isolate in pediatric patients. In present study, resistance to methicillin was detected in 33 (55.93%) of Staphylococcus aureus isolates. MRSA is on the rise in SSTIs in children both in the hospital setup (HA-MRSA) and in the community. Prevalence of MRSA was found to be consistent with studies by Gupta *et al.*(54.5%),[21] Anupurba *et al.*(54.8%)[22] and by Roveta *et al.*(53%).[23] The typical organisms that colonize the skin below the waist are both Gram-positive and Gram-negative species. It is speculated that this difference is secondary to the proximity to the anorectal region. Enteric species, such as Enterobacteriaceae and Enterococcus species, gravitate to and colonize this area of the skin. Amongst gram negative bacilli, Escherichia coli was the predominant isolate in this study, other studies have also quoted Escherichia coli as the leading organism. [15] Bacterial skin infections are one of the most common causes of childhood morbidity and constitute one of the prime causes of hospital attendance in children. The diagnosis of skin and soft tissue infections is often clinical, and treatments must be tailored to the most likely organisms present in a given population of patients.

CONCLUSION

In present study the main pathogens involved in these infections are Staphylococcus aureus and gram negative enteric organisms. Among 59 Staphylococcus aureus isolates, 33 (55.93%) were found to be methicillin resistant and 26 (44.06%) were methicillin sensitive. With knowledge of likely causative organisms causing SSTIs and their sensitivity pattern, the most suitable antibiotic can be started without waiting for the result.

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Source of Support: None Declared
Conflict of Interest: None Declared