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CASE REPORT

Bacteriological Profile of Osteomyelitis in Drepanocytic Children at the David Bernardino Pediatric Hospital from May to August 2017

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ABSTRACT

Introduction: Osteomyelitis is defined as the infection of the entire bone structure caused by a pyogenic infectious agent. Depending on the evolution time, clinical and radiological findings can be classified as acute or chronic. The present study aimed to describe the bacteriological profile of osteomyelitis in patients with sickle cell disease, admitted to the Pediatric Hospital David Bernardino, in Luanda/Angola, from June 1 to October 31, 2017.

Methodology: We carried out a descriptive, longitudinal and prospective study in a universe consisting of 33 children with sickle cell anemia admitted to the Surgery Service of this Hospital. Sample collection was performed during the surgical procedure.

Results: We identified a higher percentage of cases in male children with 58% and in the age group from 5 to 10 years with 63.6%. The most affected bone segment was the tibia with 33.4%, followed by the humerus and femur with 30.3% each. Most patients, 66.7% of cases, were admitted to the Surgery Service before the surgical procedure. Of the samples collected for bacteriological study, only 31.3% were positive, with the most identified germs being *Citrobacter spp*, *Enterobacter species* and *Proteus species*, with 6.1% each. The sensitivity profile of Gram positive cocci was 100% for Ofloxacin and Cloxacillin and gram negative cocci had 100% sensitivity for Ofloxacin, Amikacin and Minocycline. There was total resistance in Gram positives to Ampicillin and 80% in Gram negatives. Most children had severe anemia (hemoglobin below 9 g), in 17 cases (69.69%), Erythrocyte Sedimentation

Velocity levels between 60 to 83 mm, in 16 cases (48.5%), and levels of C-Reactive Protein (CRP) between 40 to 50 mg/L in 8 cases (24.3%). Cloxacillin was used in 60.7% of cases, followed by Cloxacillin + Gentamicin in 7 children (21.1%). Twenty-seven children (82%) underwent surgical treatment, with fenestration being the most used procedure.

Conclusion: The characteristics of the patients and the disease regarding sex, age and the most affected bone are in accordance with those described in the literature. Of the 10 patients without previous antibiotic therapy had samples with bacterial growth, with identification of *Citrobacter spp* (2), *Enterobacter sp* (2) *Proteus spp* (2), *E.Coli* (1), *S.Aureus* (1), Proteus Vulgaris (1) and Proteus Mirabilis (1). Patients were hospitalized for more than seven (7) days, due to other pathologies during hospitalization. About 60.7% of the children were treated with Cloxacillin and 21.1% with an association of Cloxacillin with Gentamicin, with surgical debridement and fenestration being the surgical procedure performed in 82% of patients.

Patients operated on and undergoing targeted antibiotic therapy had good results in 87.87% of cases; One case (3.03%) required readmission and underwent a new surgical approach.

INTRODUCTION

Bone and joint infections are common problems that affect individuals of all ages. Osteomyelitis is defined as the infection of the entire bone structure caused by an infectious pyogenic agent [1–5]. Depending on the evolution time; clinical and

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radiological findings can be classified as acute or chronic. This pathology represents a great challenge; both diagnostic and therapeutic; for health professionals of various specialties that make a detailed approach to the problem (Pediatrician; Surgeon; Orthopedist; Imaging and Pathologist) [1,6-9]. Osteomyelitis is one of the main complications of patients with sickle cell anemia and it is very frequent in sickle cell patients assisted at the David Bernardino Pediatric Hospital; The bacterial etiology of childhood osteomyelitis has changed in recent decades; with advances in culture techniques; introduction of Polymerase Chain Reaction (PCR) and increased antimicrobial resistance. *Staphylococcus aureus* is the most common causative agent; according to the literature. Accurate knowledge of the causative bacteria is fundamental for the decision to institute the antibiotic; which in most cases is initiated empirically. However; specific antibiotic therapy should be started after obtaining the results of cultural exams and antibiotic sensitivity tests; which should be performed to identify the bacteria causing osteomyelitis.

Several factors can interfere with the outcome of empirical antibiotic treatment: nutritional status and anemia of patients; laboratory parameters; bacteriological agents and antibiotic sensitivity [10-16].

MATERIAL AND METHODS

We carried out a descriptive; longitudinal and prospective study on the bacteriological profile of osteomyelitis in children with sickle cell anemia; admitted to the Surgery Service of the Pediatric Hospital David Bernardino; from June 1 to October 31; 2017 [17,18].

The work universe consisted of thirty (33) children with sickle cell anemia; admitted to the Orthopedics Service of this institution; diagnosed with osteomyelitis; during the study period.

The study included sickle cell children under 16 years old; hospitalized with osteomyelitis; in the wards of the David Bernardino Pediatric Hospital; Luanda/Angola; from 1 June to 31 October 2017. All samples for bacteriological examination were collected; during the surgical procedure. Children with osteomyelitis without sickle cell disease were excluded from the study [19-21].

RESULTS

The age group with the highest number of cases was 5 to 10 years old; with 63.6%; followed by those under five years old with 30.3% of cases; with a predominance of males with 58% of cases (Table 1).

Thirty-three percent (33.3%) of the patients came from the Municipality of Luanda and 24.3% (Tables 2,3) from the Municipality of Cazenga; a fact associated with the geographical proximity of the hospital unit; in the center of the city. We found that the most affected bone was the tibia with 33.4%; followed by the humerus and femur with 30.3% each. Of the 33 bacteriological tests performed; 69.7% had no bacterial growth; and 10 cases (31.3%) were positive; with identification of the agents *Citrobacter spp*; Enterobacter species and Proteus species; with 6.1% each (Tables 4,5) [22-27]. Gram positive stained bacteria showed full sensitivity to Ofloxacin and Cloxacillin and resistance to Ampicillin.

As for antibiotic sensitivity tests (TSA); in 69.7% of the cases no etiological agent was identified (Table 6); a fact associated with prolonged hospital stay; other pathologies (Table 7) and empirical treatment with antibiotics. We found that in the cases of prolonged hospitalization and previous antibiotic therapy; bacteriology resulted in no identification of any bacteriological agent; especially in those hospitalized with more than 41 days (12 cases). Cases with positive bacteriology had a hospital stay of less than 20 days; without having undergone previous antibiotic therapy; before collecting the purulent exudate [22,28-32].

We found patients with associated pathologies; especially pneumonia (6 cases); followed by malaria (5 cases); mainly

Table 1: Distribution of children according to sex and age group.

Age group (years)	M	F	Total%	
< 5	5	5	10	30.3
5 - 10	12	9	21	63.6
11 - 15 years	2	0	2	6.1
Total	19	14	33	10%

Table 2: Distribution of children by area of residence (Municipalities).

Residency	Frequency	%
Luanda	11	33.30
Cacuaco	5	15.20
Cazenga	8	24.30
Viana	4	12.20
Belas	2	6
Icolo e Bengo	3	9
Total	33	100

Table 3: Distribution of children, according to the affected bone.

Bone	D	E	%
Humerus	6	4	30.3
Rádio	1	0	3
Ulna	1	0	3
Fémur	6	4	30.3
Tíbia	6	5	33.4
Total	20	13	100

Table 4: Distribution according to identified bacteriological agents.

Etiological agent	Frequency	%
Citrobacter Spp	2	6.10
E.Coli	1	3.00
Enterobacter Spp	2	6.10
Não identificado	23	69.70
Proteus Mirabilis	1	3.00
Proteus Spp	2	6.10
Proteus Vulgaris	1	3.00
S.Aereus	1	3.00
Total	33	100.00

Table 5: Distribution according to sensitivity of gram positive cocci.

Antibiotics	Sensitive	Resistant
Ofloxacin	100	0
Kanamycin	60	40
Amikacin	60	40
Cloxacillin	100	0
Tetracycline	40	60
Tobramycin	40	60
Gentamicin	80	10
Azithromycin	60	40
Minocycline	40	60
Cefazolin	40	60
Ampicillin	0	100

Table 6: Relation of length of hospital stay and early antibiotic therapy with bacteriological results

Hospitalization time	Identified	Without identification	Previous antibiotic therapy
Less than 10 days	3	0	0
From 10 to 20 days	7	0	0
From 21 to 30 days	0	6	6
From 31 to 40 days	0	7	7
More than 41 days	0	11	11

cases with bacteriology without bacterial growth; while in the identified cases; there was no associated pathology (Table 7).

Of the patients studied; all had severe anemia; in which we highlight; about 51.61% with values above level (9gr/dL) and 15.15% with hemoglobin values below five (5 gr/dL) and 33.33% with values between 5 and 9 gr/dL. Of the infection markers studied; we highlight the levels of sedimentation velocity (V.S.); in which about 48.48% of the cases; with values between 60 to 83 mm in the first hour (Table 9); followed in the range from 84 to 107 and 108 to 131 with 5 cases (15.2%) each (Table 9). In terms of CRP; we found that 24.24% (Table 10) had values in the range of 40 to 50 mm; followed in the range of 6.12 to 7.12 with 7 cases (21.2%). Regarding antibiotic therapy; 20 (60.7%) children were treated only with Cloxacillin; and 7 (21.1%) with the association of Cloxacillin with Gentamicin (Table 11).

Of the thirty-three cases studied; all benefited from surgical treatment; in which surgical debridement; associated with fenestration was performed in about 81.90 of the cases (Tables 12,13); followed by debridement with fenestration and sequestrectomy; in 12.10% of the cases. Of the patients operated on and treated with antibiotic therapy; 29 were discharged with good results; and only one patient (3.03%) required readmission and reoperation.

DISCUSSION

The distribution of patients by gender; identified mostly in males (58%) and the predominant age group was from 5 to 10 years of age with 63.6% of cases; followed by those under 5 years of age with 30.3%). Our findings do not differ from those referenced in literature of pediatric orthopedics; which refers more to the number of cases in males in a ratio of 3:1. (Tachdjian; Pediatric Orthopedics). Similar

Table 7: Associated pathologies in sickle cell children with osteomyelitis.

Associated pathologies	In the identified	In the unidentified
exposed fracture of the femur	0	1
Pneumonia	1	6
Malária	0	5
Urinary infection	0	3
Pulmonary abscess + lithiasic cholecystitis	0	3
Allithiatic Cholecystitis	2	0
None	7	5
Total	10	23

Table 8: Distribution of children according to hemoglobin levels.

HB	Frequency	%
Under 5	5	15.20%
5 to 8	11	33.30%
More than 9	17	51.50%
Total	33	100%

Table 9: Distribution according to Sedimentation Velocity levels, in mm in the 1st hour.

Values	Frequency	%
13 a 36	3	9,09
37 a 59	0	0
60 a 83	16	48.48
84 a 107	5	15.15
108 a 131	5	15.15
132 a 155	4	12.12
Total	33	

Table 10: Distribution of patients according to CRP levels, in mg/L.

Values	Frequency	%
18 a 28	3	9.09
29 a 39	4	12.12
40 a 50	8	24.24
51 a 60	6	18.18
61 a 70	7	21.21
71 a 80	5	15.15
Total	33	100

Table 11: Distribution according to antibiotic treatment performed.

ATB. Administered	Frequency	%
Cloxacillin	20	60.70
Cloxacillin + Ceftriaxone	1	3.00
Cloxacillin + Ceftriaxone + Metronidazole	1	3.00
Cloxacillin + Gentamicin	7	21.20
Cloxacillin + P.crystalline; Ciprofloxacin	2	6.10
Cloxacillin; Ceftriaxone + Metronidazole	1	3.00
Cloxacillin + Gentamicin	1	3.00
Total	33	100.00

Table 12: Distribution according to surgical treatment performed.

Surgical treatment	Frequency	%
Debridement and fenestration	27	81.90
Debridement and sequestrectomy	2	6
Debridement + fenestration and sequestrectomy	4	12.10
Total	33	100

Table 13: Distribution according to the patient's condition at the time of discharge.

State	Frequency	%
Well	29	87.87
Reasonable	3	9.09
Bad	1	3.03
Total	33	100

studies carried out in Brazil; identified it in children over five years of age. However; Mayitondolo Eduardo in 2015; at the Josina Machel Hospital (Angola) and Dady Netemo in 2004 at the David Bernardino Pediatric Hospital (Angola); found a greater predominance in this age group; with values of 72.4% and 50 %; respectively. Both Pedro Puccini and Mayitondolo Eduardo in 2015 obtained values very close to our study (66.7%).

As for the origin; the largest number of children lived in the Municipality of Luanda with 33.3%; a fact that we associate with the proximity of the Hospital; which is located in the same municipality; and direct access; while the municipality of Cazenga with 24; 3%.; because access is done by referral from a secondary-level health unit.

However; Eduardo M.T; in 2015 (7) at Hospital Josina Machel (Luanda); had about 24.1% of children from the former Municipality of Kilamba Kiaxi; currently a district belonging to the Municipality of Luanda.

The location of patients before the surgical procedure was mostly in the Orthopedics Service with 66.7% of cases; as it is a service where they are transferred or hospitalized; and in the Pulmonology services with 21.2% of cases in which they present pulmonary complications.

In the present study; we found that the tibia (33.4%) was the most affected bone; followed by the humerus and femur with 30.3% each; a finding that coincides with the results of Mayitondolo Eduardo; and Dady Netemo; in 2004 [14]; in which they had the tibia as the most compromised bone in 39.1% and 21%; respectively.

In the distribution of patients according to the etiological agent; we found that 69.7% of the collected samples did not show bacterial growth; a fact that we empirically associate with the early institution of antibiotics. However; 31.3% of the cases (10 patients) had positive bacteriology; where Citrobacter spp; Enterobacter species and Proteus species were detected with 6.1% each. These findings differ from those described in the literature that refer *Staphylococcus aureus* as the most common agent in approximately 40% of cases of common osteomyelitis and *Salmonella spp* in 90% of children with sickle cell anemia (Yamada and Patrícia 2014) [10,33-35].

Gram positive cocci showed 100% sensitivity to Ofloxacin; Cloxacillin; and 100% of resistance to Ampicillin. The sensitivity of Gram negative bacilli was 100% for Ofloxacin; Amikacin; Minocycline and a resistance in 80% of cases to Ampicillin. Calvo; Cristina et al; in 2016; in a study published in The Pediatric Infectious Disease Journal; where 241 children less than 14 years of age were studied; Spain; 2008 to 2012; 60% of children were treated with Cefotaxime+ Cloxacillin and 40% only with Cloxacillin.

The relationship between length of hospital stay and previous antibiotic therapy; we found that cases without bacterial growth were empirically submitted to antibiotic therapy prior to purulent exudates collection; and had a hospital stay of more than 41 days (12 cases). Cases with positive bacteriology did not receive antibiotic therapy before purulent exudate collection; and were hospitalized for 10 to 20 days (7 cases) and less than 10 days (3 cases). This finding may be related to the fact that patients are admitted to the hospital by other specialties due to infectious pathology and with the institution of antibiotic treatment; which probably ended up interfering with the laboratory results of the purulent exudate.

There are several causes that can cause a microorganism not to grow in cultures even when the infection is present; among them; the most common is the use of antimicrobial during or its suspension shortly before harvest [20].

The patients studied had several associated pathologies; with emphasis on pneumonia (6 cases); followed by malaria (5 cases); in those patients who had positive bacteriology. Patients with bacterial samples without growth had no associated pathologies. These aspects may be associated with the occurrence of several acute sickle cell crises; with different manifestations; from pain crises to acute chest syndrome.

Although most patients have anemia; we found that 66.69% of the cases had hemoglobin levels below 9 gr/dL. These values; although low; are tolerable in these patients and all benefited from serial blood transfusions in the preoperative and post-treatment surgical procedures. Of the infection markers; we found that 48.5% presented the sedimentation velocity with levels between 60 and 83 mm; in the first hour; a common aspect in situations of severe infection and in line with reports in the literature. The same occurs with CRP values; another important infection marker; in which 24.3% of cases presented levels of 30 to 40 mg/L. The antibiotic therapy performed in these patients in 60.7% of the cases was with Cloxacillin; alone; and in 21.1%; the association of Cloxacillin and Gentamicin. Orrego and Moran in 2014; recommended an antibiotic therapy regimen with Cefazolin or Cloxacillin+Gentamicin; however the regimen could vary according to the results of the samples and the TSA; respectively. The patients included in this study were all surgically treated in which the surgical procedure generally included surgical debridement associated with other acts such as fenestration in 82%; and in 12% the association of fenestration and sequestrectomy. According to Orrego and Moran; surgical treatment is absolutely indicated in subperiosteal abscess; in the absence of response to antibiotic treatment after 48 hours. About 29 of the patients were cured with good results after the surgical procedure associated with antibiotic therapy according to the TSA and only one patient needed to be readmitted and reoperated. Patients had results considered good in 87.87% of cases and only one patient (3.03%) needed readmission and reoperation.

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