

# Microbiological Identification of Bacteria with Leukemic Children

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## Abstract

The most common form of childhood cancer is leukemia, Calculation for more than one third of all childhood cancers among those ages 1 day – 14 years. The diseases of leukemia are worldwide, it occurred in both genders from male and females and in all age. A total of 80 different samples of patient children were collected from the central teaching hospital of pediatric in Baghdad. Obtained during the period from 1st Sept. 2019 till the 31 th of Aug. 2020. Each isolates identification by using Vitek 2. Isolated organism in leukemic children show 18 (22.5 %) gram positive and 62 (77.5 %) gram negative from the total sample 80 (100.0 %). The percent of male is 45 more than female which represents 35 in most age groups between age (1 day-3 year) 33 (100.0 %). In most distribution of organism according to the type of sample for gram positive is *Staphylococcus aureus* 13 (16.2 %) and gram negative *Escherichia coli* 21 (26.3 %), *Klebsiella pneumoniae* 13 (16.2 %), *Pseudomonas aeruginosa* 11 (13.8 %) from the total results 80 (100.0 %). In Conclusion, the gram negative bacteria is more than gram positive bacteria and especially effect with *Escherichia coli*, and *Staphylococcus aureus*, in males for the age group 1 day to 3 years. And the most antibiotic sensitive to *Escherichia coli* is Imipenem, Amikacin, Gentamycin, Piperacillin/Tazobactam and resist to Cefotazidime, Tobramycin, Cefepime, Ceftriaxone, Ciprofloxacin and *Staphylococcus aureus* most sensitive to Vancomycin, Gentamycin, Clindamycin, and resist to Penicillin, Cefotaxime, Ceftriaxone, Oxacillin.

**Keywords:** Microbiological Identification, Leukemic diseases, Children, bacteria, Antibiotic Sensitivity

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## INTRODUCTION

The most common form of childhood cancer is leukemia, Calculation for more than one third of all childhood cancers among those ages 1 day – 14 years.<sup>1</sup> The diseases of leukemia are worldwide, it occurred in both genders from male and females and in all age.<sup>2</sup> Determined the clinical presentations of infections through a complex interaction between the virulence, pathogen, and the defense mechanisms of the host and the degree to impaired.<sup>3</sup> Patients with previously untreatable illness may now receive therapy and survive, due to large advances have been done in the management of patients with cancers.<sup>4</sup> Most important reasons for morbidity and mortality in leukemia is bacterial infections.<sup>5</sup> A divers set of chromosomal and molecular changes originate in pediatric leukemia, most of proof are acquired not inherited as only 5% of leukemia are associated with inherited syndromes.<sup>6</sup> Leukemic relapses in the early decades of therapy for childhood acute lymphoblastic leukemia (ALL) was recognized in central nervous system.<sup>7</sup> In patient who receives cancer treatment, blood stream infection is one of the major causes of morbidity and mortality.<sup>8</sup> In leukemic patients receiving chemotherapy, particularly bacteremia is the basic factor for life-threatening, In spite of development in antibiotics therapy and health care; Especially in malignant disease including leukemia; Because of complication resulted from these infection, which still one of the therapeutics as a result challenges to immune system defect which causing deficiency in immunity, also because of little using of prophylactics therapy.<sup>9-11</sup> Although evolution in antibiotics therapy, it remain the basic reason for mortalities in hospitalized patients chiefly critically ill patients like leukemia patients.<sup>12</sup> In acute medical condition the semi-pathogen and normal microflora organisms can play important role.<sup>13</sup> Higher mortality associated with polymicrobial was found 15-25% of bacteremia infections in cancer patients.<sup>8</sup> Fever during chemotherapy induce neutropenia can be the first sign of bacterial infection.<sup>14</sup> The neutropenic patients developing fever during neutropenia is a recurrent complication, Affecting 80% of those with hematological malignancies.<sup>15</sup> In cancer patients for the choice of an effective empirical

therapy or prophylaxis the important result is to increase of resistant bacteria.<sup>16,17</sup> Excessive antibiotic exposure is also the most important agents influencing the emergence and prevalence of antibiotic resistance.<sup>18</sup>

## METHODS

A total of 80 different samples of patient children were collected from the central teaching hospital of pediatric in Baghdad. Obtained during the period from 1<sup>st</sup> Sept. 2019 till the 31<sup>st</sup> of Aug. 2020. Data was collected by using questionnaire sheet concerning their [ages from (1 days -15 years), sex, type of sample, diagnosis and antibiotic sensitivity.

The identification of each isolates by using the Vitek 2 which is automated microbiology system and its applications. The reagent cards used that are incubated and expound automatically. The Vitek 2 system was used according to the manufacturer's instructions; The combination of automated Vitek 2 Technology ready-to-use Vitek 2 identification (ID) and susceptibility (AST) cards provide dependable, accurate ID and AST results for clinically important Gram-positive cocci, Gram-negative cocci, Gram-positive bacilli, Gram-negative bacilli. The sample was culture on special media according to the type of sample on blood agar, MacConkey agar, Chocolate agar and incubated at 35-37°C at 18-24 hours, bacteria were suspend in 2.5 ml of 0.45% sodium chloride solution. The suspension used in the Vitek2 system was adjusted to a McFarland standard of 0.5 by using a Densicheek (bioMerieux).

### Statistical Analysis

Used in order that analyze and assess the results they including:

Binomial (0.50) = Frequencies / Descriptive statistics

### Descriptive statistics

statistical tables contain observed frequencies with their percentages.

### Inferential statistics

The accept or reject show by used statistical hypotheses, 0.05 level of significance represent the Persons Chi-Square test ( $\chi^2$ ).

P value < 0.05 level of significance was considered statistically significant.

P-value < 0.025 in 2-sided.

**RESULTS**

In this study, Isolated organism in leukemic children show 18(22.5%) gram positive and 62(77.5%) gram negative from the total sample 80(100.0%) as shown in Table (1).

**Table 1.** Isolated Organism in Leukemic Children

Isolated Organism	No.	%	P-Value
Gram Positive	18	22.5	0.00
Gram Negative	62	77.5	
Total	80	100.0	

P- Value < 0.05

**Table 2.** Age and Sex Distribution in Leukemic Children

Age Group	Sex		Total	P- Value
	Male No. %	Female No. %		
1 day - 3 Y	19 (57.6 )	14 (42.4 )	33 (100.0 )	0.326
4 Y - 6 Y	12 (63.2 )	7 (36.8 )	19 (100.0 )	
7 Y – 9 Y	7 (53.8 )	6 (46.2 )	13 (100.0 )	
10 Y – 12 Y	6 (66.7 )	3 (33.3 )	9 (100.0 )	
13 Y- 15 Y	1 (16.7 )	5 (83.3 )	6 (100.0 )	
Total	45 (56.2 )	35 (43.8 )	80 (100.0 )	

P-Value > 0.025 in 2- sided.

**Table 3.** The Distribution of Organism according to the type of Sample

Organisms	Total No.%	Type of Sample							
		Blood No.%	Urine No.%	Pus No.%	Wound Swab No.%	Sputum No.%	Ear Swab No.%	Skin Swab No.%	Abscess No.%
<b>Gram Positive</b>									
<i>Staph. aureus</i>	13(16.2)	10(76.9)	0(0.0)	0(0.0)	1(7.7)	1(7.7)	1(7.7)	0(0.0)	0(0.0)
<i>Staph. hominis</i>	1(1.3)	0(0.0)	1(100.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)
<i>Strpto. faecalis</i>	2(2.5)	1(50.0)	0(00.0)	1(50.0)	(00.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)
<i>Strepto. pneumoinae</i>	2(2.5)	1(50.0)	0(0.0)	0(0.0)	0(0.0)	1(50.0)	0(0.0)	0(0.0)	0(0.0)
<b>Gram Negative</b>									
<i>Escherichia coli</i>	21(26.3)	4(19.0)	15(71.4)	1(4.8)	1(4.8)	0(0.0)	0(0.0)	0(0.0)	0(0.0)
<i>K. pneumonia</i>	13(16.2)	5(38.5)	6(46.2)	0(0.0)	2(15.4)	0(0.0)	0(0.0)	0(0.0)	0(0.0)
<i>P. aeruginosa</i>	11(13.8)	5(45.5)	3(27.3)	0(0.0)	0(0.0)	0(0.0)	2(18.2)	0(0.0)	1(9.1)
<i>Acinetobacter</i>	8(10.0)	6(75.0)	1(12.5)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	1(12.5)	0(0.0)
<i>Enterobacter</i>	5(6.2)	3(60.0)	2(40.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)
<i>Proteus mirablis</i>	4(5.0)	0(0.0)	4(100.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)
Total (Gram positive+ Gram Negative)	80 (100.0)	35(43.8)	32(40.0)	2(2.5)	4(5.0)	2(2.5)	3(3.8)	1(1.2)	1(1.2)

P-Value = 0.003; P-Value < 0.025 in 2- sided

**Table 4.** Antimicrobial Susceptibility Pattern of Isolated Gram Positive Bacteria

Antibiotic	<i>Staph aureus</i>		<i>Staph. hominis</i>		<i>Strepto. faecalis</i>		<i>Strept. pneumoniae</i>	
	(13)		(1)		(2)		(2)	
	S No. %	R No. %	S No. %	R No. %	S No. %	R No. %	S No. %	R No. %
CTX	3(23.1)	5(38.1)	0(0.0)	0(0.0)	1(100.0)	0(0.0)	0(0.0)	0(0.0)
Van	12(92.3)	1(7.7)	1(100.0)	0(0.0)	1(100.0)	0(0.0)	2(100.0)	0(0.0)
FOX	4(30.8)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)
CAZ	2(15.4)	1(7.7)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)
P	0(0.0)	7(53.8)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	2(100.0)
CD	6(46.2)	2(15.4)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	3(100.0)	0(0.0)
AMP	2(15.4)	0(0.0)	0(0.0)	0(0.0)	1(100.0)	0(0.0)	0(0.0)	0(0.0)
Gen	7(53.8)	2(15.4)	0(0.0)	1(100.0)	2(100.0)	0(0.0)	1(100.0)	0(0.0)
Li	4(30.8)	2(15.4)	0(0.0)	0(0.0)	1(100.0)	0(0.0)	0(0.0)	0(0.0)
CRO	2(15.4)	6(46.2)	0(0.0)	0(0.0)	1(100.0)	0(0.0)	0(0.0)	0(0.0)
PI	1(7.7)	1(7.7)	0(0.0)	0(0.0)	1(100.0)	0(0.0)	0(0.0)	0(0.0)
IPM	1(7.7)	0(0.0)	0(0.0)	0(0.0)	1(100.0)	0(0.0)	0(0.0)	0(0.0)
E	2(15.4)	1(7.7)	0(0.0)	1(100.0)	0(0.0)	2(100.0)	0(0.0)	0(0.0)
TM	3(23.1)	0(0.0)	1(100.0)	0(0.0)	1(100.0)	0(0.0)	0(0.0)	0(0.0)
Cip	5(38.1)	2(15.4)	1(100.0)	0(0.0)	1(100.0)	0(0.0)	0(0.0)	0(0.0)
OX	0(0.0)	4(30.8)	0(0.0)	1(100.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)
TE	2(15.4)	2(15.4)	0(0.0)	1(100.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)
TEC	0(0.0)	0(0.0)	0(0.0)	0(0.0)	1(100.0)	0(0.0)	0(0.0)	0(0.0)
Tig	3(23.1)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)
FA	1(7.7)	1(7.7)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	1(100.0)

**Table 5.** Antimicrobial Susceptibility Pattern of Isolated Gram Negative Bacteria

Antibiotic	<i>E. coli</i> (21)		<i>K. Pneumonia</i> (13)		<i>P. Aeruginosa</i> (11)		<i>Acinetobacter</i> (8)		<i>Enterobacter</i> (5)		<i>Proteus mirabilis</i> (4)	
	S No. %	R No. %	S No. %	R No. %	S No. %	R No. %	S No. %	R No. %	S No. %	R No. %	S No. %	R No. %
Total	7(33.3)	1(4.8)	2(15.4)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	1(20.0)	0(0.0)	0(0.0)	0(0.0)
Ni	18(85.7)	1(4.8)	8(61.5)	1(7.7)	9(81.8)	1(29.1)	6(75.0)	2(25.0)	3(60.0)	2(40.0)	3(75.0)	1(25.0)
IPM	14(66.6)	5(23.8)	7(53.8)	5(88.5)	9(81.8)	1(29.1)	5(62.5)	3(37.5)	3(60.0)	1(20.0)	1(25.0)	1(25.0)
G	2(9.5)	6(28.6)	1(7.7)	5(38.5)	0(0.0)	0(0.0)	1(12.5)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)
AMP	10(47.6)	4(19.0)	3(23.1)	6(46.2)	2(18.2)	4(36.4)	1(12.5)	4(50.0)	2(40.0)	1(20.0)	2(50.0)	2(50.0)
PI	1(4.8)	3(14.3)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	1(12.5)	0(0.0)	0(0.0)	0(0.0)	0(0.0)
CFM	1(4.8)	4(19.0)	0(0.0)	1(7.7)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)
TMP	15(71.4)	1(4.8)	7(53.8)	5(38.5)	10(90.9)	1(29.1)	3(37.5)	2(25.0)	1(20.0)	1(20.0)	2(50.0)	0(0.0)
AK	1(4.8)	10(47.6)	3(23.1)	9(69.3)	4(36.4)	3(27.3)	1(12.5)	5(62.5)	1(20.0)	4(80.0)	0(0.0)	3(75.0)
CAZ	1(4.8)	6(28.6)	2(15.4)	4(30.8)	4(36.4)	3(27.3)	0(0.0)	4(50.0)	1(20.0)	1(20.0)	1(25.0)	1(25.0)
CTX	2(9.5)	9(42.9)	2(15.4)	6(46.2)	4(36.4)	5(45.5)	1(12.5)	1(12.5)	3(60.0)	1(20.0)	0(0.0)	1(25.0)
CRO	2(9.5)	8(38.1)	1(7.7)	6(46.2)	0(0.0)	3(27.3)	0(0.0)	6(75.0)	4(80.0)	0(0.0)	0(0.0)	2(50.0)
FEP	4(19.0)	10(47.6)	1(7.7)	4(30.8)	3(27.3)	1(29.1)	3(37.5)	2(25.0)	2(40.0)	1(20.0)	1(25.0)	3(75.0)
TM	3(14.3)	1(4.8)	1(7.7)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	1(25.0)
FOX	5(23.8)	7(33.3)	2(15.4)	4(30.8)	0(0.0)	2(18.2)	2(25.0)	0(0.0)	2(40.0)	0(0.0)	2(50.0)	1(25.0)
Cip	1(4.8)	1(4.8)	3(23.1)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	1(12.5)	0(0.0)	0(0.0)	0(0.0)	0(0.0)
Tig	2(9.5)	2(9.5)	2(15.4)	1(7.7)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	2(40.0)	0(0.0)	1(25.0)	0(0.0)
LEV	0(0.0)	0(0.0)	0(0.0)	0(0.0)	1(29.1)	1(29.1)	2(25.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)
CS	0(0.0)	0(0.0)	0(0.0)	3(23.1)	3(27.3)	2(18.2)	2(25.0)	3(37.5)	1(20.0)	2(40.0)	0(0.0)	2(50.0)
TCC	0(0.0)	1(4.8)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	1(25.0)	0(0.0)
TE												

Sensitive=S, Resistance=R  
 Cefotaxime (CTX), Vancomycin (Van), Cefixime (FOX), Cefotazidime (CAZ), Penicillin (P), Clindamycin (CD), Ampicillin (AMP), Gentamycin (G), Lincomycin (Li),  
 Ceftriaxone (CRO), Piperacillin/Tazobactam (PI), Imipenem (IPM), Erythromycin(E), Tobramycin (TM), Ciprofloxacin(Cip), Oxacillin (OX), Tetracycline (TE), Teicoplanin (TEC), Tigecycline (Tig),  
 Fucidic Acid (FA), Nitrofurantoin (NI), Cefixime (CFM), Trimethoprim/Sulfamethoxazole (TMP), Amikacin (AK), Cefepime (FEP), Colistin (CS), Ticarcillin (TCC), Levofloxacin (LEV).

## DISCUSSION

The prevalence in patients with cancer is 5.7- 44% of bacteremia according to the global reports.<sup>19,20</sup> The percentage of gram negative bacteria is more than gram positive bacteria which represent 62(77.5%), 18(2.5%) respectively show in Table (1) and the p-value is 0.00 which represent significant. According to the study the result has slightly more incidents in males number were 45 while females number were 35 from the whole number of specimen was 80 patients and agreement with Marcotte (1), Muhammed,<sup>2</sup> Ahamed,<sup>4</sup> Martinez-Laperche,<sup>7</sup> Sevan<sup>21</sup> show in Table (2). During the last few decades have seen diagnosed with cancer in the survival of children with the 5-year survival rate approaching 80%.<sup>22</sup> Incidence rates increase to a peak around age 3-4 years and then decline.<sup>23</sup> Also, the Table (2) appear the most age group gives positive result between (1 day- 3 years) which represent 33(100.0%) this agreement with Ahamed.<sup>4</sup> The p-value is 0.326 which, is non-significant in 2-sided. In our study, the multiple samples were associated with 70.5% of positive result Hosseini.<sup>19</sup> In present study 35(43.8%) of 80 blood cultures were positive which is similar to other studies Eslami Nejad,<sup>13</sup> El-Mahallawy.<sup>24</sup> The most Gram positive bacteria was isolated is *Staphylococcus aureus* 13(16.2%) and the most gram negative bacteria from the total isolates is *Escherichia coli* 21(26.3%), *Klebsiella pneumoniae* 13 (16.2%), *Pseudomonas aeruginosa* 11(13.8%) from the total results 80(100.0%) these show in Table (3) and the result agreement with Ahmed,<sup>4</sup> Tezcan.<sup>25</sup> The p-value is 0.003, which is significant gram positive and gram negative bacteria in relation with multiple sample. In Table (4) the antibiotic susceptibility of isolated most effects to *Staphylococcus aureus* is Vancomycin 12(92.3%), Gentamycin 7(53.8%), Clindamycin 6(46.2%), Ciprofloxacin 5(38.1%), Lincomycin 4(30.8%), Cefixime 4(30.8%) and the more resistance is Penicillin 7(53.8%), Ceftriaxone 6(46.2%), Cefotaxime 5(38.1%), Oxacillin 4(30.8%) which agree with Eslami Nejad.<sup>13</sup> The most antibiotic sensitive gram negative organism is *Escherichia coli* to Imipenem 18(85.7%), Amikacin 15(71.4%), Gentamycin 14(66.6%),

Piperacillin/Tazobactam 10(47.6%) and *Klebsiella pneumoniae* sensitive to Imipenem 8(61.5%), Amikacin 7(53.8%), Gentamycin 7(53.8%), Piperacillin/Tazobactam 3(23.1%), *pseudomonas aeruginosa* sensitive to Amikacin 10(90.9%), Imipenem 9(81.8%), Gentamycin 9(81.8%). The *Escherichia coli* is resistance to Cefotazidime 10(47.6%), Tobramycin 10(47.6%), Cefepime 8(38.1%), Ceftriaxone 7(33.3%), Ciprofloxacin 7(33.3%), and *Klebsiella pneumoniae* is resist to Cefotazidime 9(69.3%), Piperacillin/Tazobactam 6(46.2%), Cefepime 6(46.2%), Ciprofloxacin 6(46.2%), Tobramycin 4(30.8%). *Pseudomonas aeruginosa* is resist to Ceftriaxone 5(45.5%), Piperacillin/Tazobactam 4(36.4%), Cefepime 3(27.3%), these result agreement with Al-Ouqaili.<sup>26</sup> these bacteria is multidrug resistant shown in Table (5).

## CONCLUSION

In conclusion, in the present study the gram negative bacteria is more than gram positive bacteria and especially effective with *Escherichia coli*, and *Staphylococcus aureus*, in males for the age group one day to three years. And the most antibiotic sensitive to *Escherichia coli* is Imipenem, Amikacin, Gentamycin, Piperacillin/Tazobactam and resist to Cefotazidime, Tobramycin, Cefepime, Ceftriaxone, Ciprofloxacin and *Staphylococcus aureus* most sensitive to Vancomycin, Gentamycin, Clindamycin, and resist to Penicillin, Cefotaxime, Ceftriaxone, Oxacillin.

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## CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

## AUTHORS' CONTRIBUTION

All authors listed have made a substantial, direct and intellectual contribution to the work, and approved it for publication.

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**DATA AVAILABILITY**

All datasets generated or analyzed during this study are included in the manuscript.

**ETHICS STATEMENT**

Not applicable.

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