ORIGINAL ARTICLE

Rapid Diagnosis of Bacteraemia in Hospitalized Infants and Children in Chhattisgarh

Madhuri Agnihotri [1], **Animesh Panda** [2], **Sitholay Pooja** [3], **Vacchani Rashim** [4], **Anil Rahule** [5]
[1] *Professor & HOD of Biochemistry, C.M. Medical College, Bhilai (C.G)*
[2] *Assistant Professor of Biochemistry, C.M. Medical College, Bhilai (C.G)*
[3] *Tutor in Biochemistry, C.M. Medical College, Bhilai (C.G)*
[4] *Tutor in Anaesthesia, C.M. Medical College, Bhilai (C.G)*
[5] *Associate Professor of Anatomy, GMC Rajnandgaon*
http://dx.doi.org/10.18049/jcmad/225

Abstract

Bacteraemia is frequently seen with serious complication and the problem is more severe in children as it is associated with high morbidity and mortality. Most of the time, it is difficult to diagnose due to presence of non specific clinical features with no noticeable focus of infection. Hence the present study was carried out to identify the rapid diagnostic methods. ***Materials and Methods****: Clinically suspected 330 cases of bacteraemia in neonates, infants and children admitted as inpatients at CM Medical College and Hospital, Durg and 25 healthy children as control were included in the present study. The cases were investigated by blood culture and 5 rapid tests Viz total leucocyte count (TLC), immature to total neutrophil (I:T) ratio, C – reactive protein (CRP), ESR and Grams smears of Buffy coat for organisms.* ***Results:*** *Blood cultures were positive in 141 (42.7%) cases including 55.3% gram negative and 44.6% of gram positive. The most common isolates were Staphylococcus epidermides (25.5%) and Staphylococcus aureus (17.0%) with overall staphylococcal prevalence of 42.5%. CRP yielded maximum sensitivity of 80.5%, Specificity of 77.7% and positive predictive accuracy of 73.0%.* ***Conclusion:*** *Blood cultures and a battery of rapid tests could be carried out depending upon the amount of blood drawn from children of different age groups. And if blood drawn is around 1ml only CRP test could be preferred since it is a sensitive indicator of bacteraemia.*
Keywords: *Bacteraemia, blood cultures, neotal sepsis, rapid diagnostic test.*

Address for correspondence: *Dr. Maduri A Agnihotri, Professor & HOD of Biochemistry, C.M. Medical College, Kachandur, Bhilai (CG), shrishivhar@rediffmail.com, animeshpanda48@yahoo.com, rahuleanil@yahoo.co.in, Mob: 08600975700, 09730630600, 07898968485*

Introduction

In under developed and developing countries like India infectious diseases in infants and children continue to be of common occurrence and among them bacteraemia is not uncommon. Bacteraemia is frequently seen with serious complications. The problem is more prominent in children, since high fever usually associated with high morbidity and mortality. [1] Neonates are immunocompromised even at term gestation since the neonatal immune system is functional at birth but not mature which is the reason for more common occurrence of neonatal sepsis. Infections spreads through transplacental route after maternal infections and the bloodstream is the usual rout by which the foetus becomes infected. Most of the times, clinically bacteraemia is difficult to diagnose due to presence of non specific clinical features with no noticeable focus of infection. [2] Since blood cultures are difficult in neonates, infants and very young children and usually require 2-3days for diagnosis, some rapid tests can be used for early diagnosis of bacteraemia to facilitate prompt treatment. [3] Some relevant clinical features and laboratory tests can help us to determine that which children are at low risk of occult bacteraemia and need not have their blood cultures and which children needs blood culture for accurate diagnosis. [4] Hence the present study was carried out to identify the

rapid diagnostic methods in this resource deficient region of a developing country.

Materials and Methods

It was a prospective hospital based study which was conducted at CM Medical College and Hospital, Durg, Chhattisgarh, India. 330 suspected cases of bacteraemia in neonates, infants and children included in the study. 25 healthy children were also enrolled as controls for the study. The cases were investigated by blood culture and 5 rapid tests Viz total leucocyte count (TLC), immature to total neutrophil (I:T) ratio, C – reactive protein (CRP), ESR and Grams smears of Buffy coat for organisms. Blood samples were collected aseptically for different tests depending upon the age of the child. About 3-5ml blood was drawn from children of age 6 months and above. One ml of blood was put into a bottle containing 2mg/ml EDTA as anticoagulant for TLC, I:T neutrophil ratio and buffy coat. 1ml was allowed to clot in a sterile bottle for CRP and 0.5ml was collected in a sterile small test tube with 0.2ml 3.8% citrate solution for ESR. Remaining 2.3ml blood was inoculated into 20ml Trypticase soy broth (TSB) containing 0.05% liquid in Mccartney bottle. In neonates and infants finger prick blood was used for TLC, I:T ratio and at least 1ml blood was collected and clotted in a bottle for CRP and clot culture in 10ml TSB.

The samples were immediately processed in the laboratory. Blood cultures and clot cultures were incubated at 37^0C for 10days with subcultures at 3 days intervals on blood agar and Macconkey agar. The bacterial isolates were identified by biochemical reactions and special tests. [5] TLC was done using improved Newbaur's counting chamber and WBC pipette. I:T ratio was done in leishman's blood smears by making differential count of 100 successive neutrophils to determine nuclear indices according number of lobes of nuclei. Grams smears of buffy coat obtained by centrifugation of EDTA blood in wintrobe tube at 2500 rpm were examined microscopically for organisms. CRP estimation was done by latex agglutination using reagents obtained from Tulip Diagnostics limited.

The cut off values for positive tests were TLC less than 5000 and more than 20000 / cmm; I:T neutrophil ratio, 0.2 and above; ESR more than 10mm of first hour; CRP more than 6mg/ml. The results of all the rapid tests were analyzed singly or in combination of 2 to assess their sensitivity, specificity and positive predictive accuracy.

Results

Out of 330 cultures which include blood and clot cultures, 141 (42.7%) yielded growth while 189 (57.2%) did not show growth. All positive blood cultures showed mono-bacterial isolates. Out of 141 cultures positive, 78 (53.3%) showed growth of gram negative bacteria and 63 (44.6%) yielded gram positive organisms. All blood cultures of control group were negative for bacterial growth. The most common isolates were Staphylococcus epidermides 36 (25.5%) and Staphylococcus aureus 24 (17.0%) giving overall Staphylococcal prevalence of 42.25% followed by gram negative bacterias which includes 21 (14.8%) Salmonella Typhi, 15 (10.6%) E.coli and 15 (10.6%) Pseudomonas auroginosa (Table- 1).

Table- 1: Isolated bacterias from culture

Organisms	Number	%
Staphylococcus epidermides	36	25.5
Staphylococcus aureus	24	17
Candida albicans	3	2.1
Salmonella typhi	21	14.8
Escherichia coli	15	10.6
Pseudomonas aeruginosa	15	10.6
Proteus mirabilis	12	8.5
Klebsiella aerogenes	9	6.3
Citrobacter freundii	6	4.2
Total	**141**	**100**

Result of rapid diagnostic tests in 141 culture positive and 189 cultures negative cases are shown in table- 2. Gram smears of buffy coat were negative for organisms in both cultures positive and negative cases. But out of 141 blood cultures positive cases abnormal values of TLC in 84 (59.5%), I:T ratio in 72 (51.0%), ESR in 75 (53.0%) and CRP in 114 (80.9%) were observed. On the other hand, 189 cultures negative cases, abnormal values TLC in 84 (44.4%), I:T ratio 102 (53.9%), ESR 54 (28.5%)

and in 147 (77.7%) were noticed. It is evident that CRP revealed sensitivity of 80.8%, specificity of 77.7% and positive predictive accuracy of 73.0% when compared with other tests either alone or in combination of 2 tests (Table- 3).

Table- 2: Result of rapid diagnostic tests

Test	Positive (A)	(%)	Negative (C)	(%)	Negative (B)	(%)	Positive (D)	(%)
TLC	84	59.5	57	40.5	105	55.5	84	44.4
I :T Ratio	72	51	69	48.9	87	46	102	53.9
ESR	75	53	66	46.8	135	71.4	54	28.5
CRP	114	80.9	27	19	42	22.2	147	77.7
Buffy coat smear	00	00	00	00	00	00	00	00

(A) True Positive, (B) True Negative, (C) False Negative, (D) False Positive

Table- 3: Sensitivity, Specificity and Positive Predictive accuracy

Test	Sensitivity (Ax100/A+C)	Specificity (Dx100/B+D)	+ve predictive accuracy (Ax100/A+B)
TLC	59.5	44.4	44.4
I : T ratio	51	53.9	45.2
ESR	53.1	28.5	35.7
CRP	80.8	77.7	73
CRP+TLC	70.2	61.1	57.3
CRP+I:T ratio	65.9	65.8	59
CRP+ESR	67	53.1	51.6

Discussion

Bacteraemia is a serious problem of all age group and is more dangerous in paediatric age group specifically neonatal sepsis. Early diagnosis is difficult but some specific salient clinical features and few rapid laboratory tests often help to make early diagnosis easy. [4] In the present study 42.7% blood cultures were positive for various bacteria. In the present study prevalence of gram positive bacteria as high as 53.3% when compared to 44.6% prevalence of gram negative organisms. Furthermore, we observed staphylococcal predominance (42.5%) with prevalence of staphylococcal epidermides and staphylococcal aureus being 25.5% and 17.0% followed by salmonella typhi (14.8%), E. coli and pseudomonas aeruginosa (10.6%) each indicating importance of these pathogens in bacteraemia. These findings are different from various other Indian authors. UK Namdev et al in1985 found culture positivity in 50% neonatal cases. [6] N Mehrotra et al in 1985 observed that 60% blood cultures were positive when they studied correlation between maternal and neonatal factors responsible for neonatal sepsis. [7] Khatua SO et al found 59.9% culture positivity during their study about neonatal sepsis in 1990. [8] But least number of cases (32%) was observed by Bhakoo in Chandigarh region of India. [9]

Out of 5 rapid tests, significantly Gram smears of buffy coat were negative in all cases. CRP showed maximum sensitivity of 80.8%, specificity of 77.7% and positive predictive accuracy of 73.0% and these findings are in agreement with the reports of other workers. Kari Saraswathi [10], Anuradha DE et al in 1998 [11], Anita Sharma in 1993 [12] also found the similar pattern of culture results. On the other hand other rapid tests singly or in combinations of two did not show any advantage when compared to CRP test alone.

Conclusion

Based on our observations, we are of the opinion that blood cultures and a battery of

rapid tests could be carried out depending upon the amount of blood drawn from children of different age groups. And if blood drawn is around 1ml only CRP test could be preferred since it is a sensitive indicator of bacteraemia in the absence of blood cultures. Clot cultures could be done whenever possible.

Acknowledgements

We are extremely thankful to the superintendent and teaching staff of the department of pediatrics, CM Medical College and Hospital, Durg for their kind permission to carry out this study. Technical help from the department of Microbiology is also appreciated.

Source(s) of support: *Nil*
Conflict of Interest: *None declared*

References

1. Goldman DA, FreemanJ, Durbin WA. Nosocomial infection and death in a neonatal intensive care unit. J Inf Dis 1983;147:635-639.
2. Siegel JD, Mc Craken Gh. Sepsis neonatorum. N Eng J Med 1981;304:642-645. [PubMed]
3. Kite M, Miller MR, Gorham P, Congclon P. Comparison of files test in diagnosis of neonatal bacteaemia. Arch Dis child 1988;63:639-643. [PubMed]
4. Mc Lellan D, Glebink GS. Perspectives of occult bacteraemia in children. J Paediatr 1986;109:1-8.
5. Collee JG, Miles RS. Mackie and McCartney Practical medical Microbiology, test for identification of bacteria 13th ed (Churchill livingston 1989;P:141-165.
6. Namdev UK, Singh HP, Rajput VJ, Kushwaha JS. haematological indices for early diagnosis of neonatal septicaemia. Ind J Paediatr 1985;22:287-292.
7. Mehrotra N, Kumar A chansoria M, Kaul KK. Neonatal Sepsis, Correlation of maternal and neonatal factors to positive bacterial cultures. Ind J Paediatr 1985;22:275-280.
8. Khatua SO, Das Ak, Chatterijee BD, Khatua S, Ghose B, Saha A. Neonatal Sepsis. Ind J Paediatr 1986;53:509-514.
9. Bhakoo ON. Neonatal bacterial infections in chandigarh; a decade of experience. Ind J Paediatr 1980;47:419-424.
10. Kari Saraswathi, Anuradha DE, Alka Gogate, Armida R, Fernandes. Citrobacter Sepsis in infants. Ind Paediatr 1955;32:359-361.
11. Anuradha DE, Kari Saraswathi, Alka Gogate, Kalayani Raghavan; C-reactive protein and buffy coat smear in early diagnosis of neonatal Septicaemia. Ind J Pathal Microbiol 1998;41:23-26.
12. Anita Sharma CV, Krebhina Kutty, Uma Sabharwal, Sushila Rathee, Harsha Mohan. Evalution of Sepsis serene for diagnosis of neonatal Septicaemia. Ind J Paediatr 1993;60:559-563.