

## Original research article

## A research to determine the utility of Hs CRP in the Sepsis Spectrum from neonate upto 2 months

Dr. Prabudh Paritosh Mishra<sup>1</sup>, Dr. Bhupendra Narain<sup>2\*</sup>

<sup>1</sup>Assistant Professor, Upgraded Department of Pediatrics, Patna Medical College and Hospital, Patna, Bihar, India

<sup>2</sup>Associate Professor, Upgraded Department of Pediatrics, Patna Medical College and Hospital, Patna, Bihar, India

Corresponding Author: Dr. Bhupendra Narain

### Abstract

**Aim:** The aim of the present study to evaluate the role of Hs CRP in Sepsis Spectrum from neonate upto months

**Methods:** This prospective observational study was carried out in the Upgraded Department of Pediatrics, Patna Medical College and Hospital, Patna, Bihar, India for 6 months. Each paediatric patient who came to our Hospital was initially categorized according to age till 2 months of age. Neonates were graded according to the signs and symptoms of FIMNCI. FIMNCI considers bacterial infections in young infants when signs or symptoms of sepsis, pneumonia or meningitis are present. 2 ml of blood was again collected from the corresponding patient in a plain bulb under all aseptic precautions. The sample was then sent laboratory for CRP testing.

**Results:** Out of 110, 105 cases were successfully followed up to 48 hours as was aimed at the beginning of the study and 5 cases failed to complete a 48 hour study period due to various reasons. Out of 105 newborns studied, 69 were male and 36 were female babies. Clinical improvement was assessed by hemodynamic profile, absence of presenting complaint/s and ability to tolerate feed and absence of blood culture positivity of first culture. 33 babies improved clinically, whereas 40 babies had almost similar clinical profile. 32 babies showed clinical downward status in spite of starting empirical antibiotics and supportive treatment. Majority of babies had shown positivity in HS-CRP at admission (94/105) as well as after 48 hours (99/105). Mean and median of all babies as shown in Table 3 was not conclusive about severity of infection and CRP values. It seems that Hs CRP is very sensitive indicator for neonatal sepsis.

**Conclusion:** We concluded that the HS CRP levels were increased in all cases of suspected neonatal sepsis.

**Keywords:** C-reactive protein (CRP), Hs CRP, IMNCI, Neonatal sepsis.

### Introduction

Neonatal sepsis is defined as invasive bacterial infection occurring in first 4 weeks of life [1]. The incidence of neonatal sepsis is lower in developed countries (2.7/1000 live birth) compared to developing countries (10-15/1000 live birth).<sup>1</sup> the incidence of neonatal sepsis varies from 11-24.5/1000 live births in India. Neonatal sepsis causes high morbidity and mortality during neonatal period, and is a thrust area for research in neonatal medicine. Many of the manifestations of the sepsis have their counterparts in non-infectious neonatal disorder.<sup>1</sup> The difficulty in early diagnosis of neonatal sepsis coupled with non-specific signs of life threatening illness during neonatal period has warranted widespread antibiotic use in many clinical settings leading to excess use of antibiotics and the resulting antibiotic resistance.<sup>2</sup>

C-reactive protein (CRP) is generally considered a helpful marker for diagnosis of sepsis used in addition to blood culture, but sometimes it may be inadequate for sepsis diagnosis, so more rapid efficient markers are needed.<sup>3</sup> CRP is an acute phase protein synthesized in liver in the presence of infectious and/or inflammatory stimuli, correlating with the disease severity,<sup>4</sup> but its production significantly varied during the neonatal period,<sup>5</sup> with lower production in preterm compared to full term neonates.<sup>6</sup> The high sensitivity assays of CRP (hsCRP) could detect lower grade of inflammation.<sup>7</sup> Cluster differentiation 14 (CD14) is the receptor for lipopolysaccharide-lipopolysaccharide binding protein (LPSLBP) complexes.<sup>8</sup> CD14 has two types: mCD14 (membrane bound) which is expressed on the cell surface of monocytes/macrophages and neutrophils; and sCD14 (soluble) which is present in the plasma, mediating the immune response and called presepsin. Although plasma presepsin level may increase in response to sepsis, its increase has been reported in many other conditions such as liver cirrhosis, diabetes mellitus and heart failure. Interleukin6 (IL-6), one among the most important cytokines, is secreted in response to infection, inducing the B-lymphocytes to secrete antibodies and enhance cytotoxic T-cells differentiation.

The present study is carried to ascertain usefulness of estimation of CRP with technologically better method called High Sensitivity C-Reactive Protein (Hs CRP) estimation, in the spectrum of infection, ranging from infection to septic shock. The age group included is from birth to 2 months of age. An effort has been made to correlate values of Hs CRP with respect to severity of infection.

### **Material and methods**

This prospective observational study was carried out in the Upgraded Department of Pediatrics, Patna Medical College and Hospital, Patna, Bihar, India for 6 months, after taking the approval of the protocol review committee and institutional ethics committee.

Total 110 Newborns and children upto 2 months of completed age and admitted in paediatric department, Suspicion of possible infections in newborns were included in this study. Newborns and children with definitive viral and fungal infections at admission, newborns and children with MODS, children with known immunodeficiency and autoimmune disorders were excluded from this study.

### **Methodology**

Each paediatric patient who came to our Hospital was initially categorized according to age till 2 months of age. Neonates were graded according to the signs and symptoms of FIMNCI. FIMNCI considers bacterial infections in young infants when signs or symptoms of sepsis, pneumonia or meningitis are present.

For our study, following signs and symptoms were considered for possible infections in neonates to enrol patients: unable to feed, fast breathing (RR >60/min) severe retractions, lethargic or unconsciousness, bulging fontanelle, convulsions, nasal flaring, Grunting, less than normal movements, axillary temperature 37.5 C or above; or less than 35.5 C, Painful joints, joint swelling, reduced movements around a particular joint and irritability, many skin pustules/ big boils, Umbilical redness extending to the periumbilical skin or umbilicus draining pus, Meningitis is considered in neonates if one or more of the following signs are present: drowsiness, lethargy or unconsciousness, persistent irritability and high pitched cry.

The high sensitivity application was used for hsCRP testing.

2 ml of blood was again collected from the corresponding patient in a plain bulb under all aseptic precautions. The sample was then sent laboratory for CRP testing. This sample was labelled as the 2nd Hs CRP sample. It was run on the same machine to avoid technological bias. The 2nd Hs CRP value was noted too.

### Statistical analysis

The means and standard deviations (SDs) for continuous variables. We also estimated the proportions for the categorical variables. The means between three groups were compared using the unpaired t-test. We estimated the various SDs of the Hs CRP values at the time of admission and after 48 hours.

### Results

A total 110 cases were enrolled in the study in which infection was suspected at the time of admission based on the category in IMNCI/FIMNCI for suspicion of infection. Out of these, 105 cases were successfully followed up to 48 hours as was aimed at the beginning of the study and 5 cases failed to complete a 48 hour study period due to various reasons. Out of 105 newborns studied, 69 were male and 36 were female babies. Clinical improvement was assessed by hemodynamic profile, absence of presenting complaint/s and ability to tolerate feed and absence of blood culture positivity of first culture. 33 babies improved clinically, whereas 40 babies had almost similar clinical profile. 32 babies showed clinical downward status in spite of starting empirical antibiotics and supportive treatment.

Surprisingly, majority of babies had shown positivity in HS-CRP at admission (94/105) as well as after 48 hours (99/105). Mean and median of all babies as shown in Table 3 was not conclusive about severity of infection and CRP values. It seems that Hs CRP is very sensitive indicator for neonatal sepsis.

Neonates were further categorised in 3 groups depending on clinical improvement and fall or rise of Hs CRP level was studied. It was seen that Hs CRP levels have decreased in group which had shown clinical improvement, however in both other groups it was showing rise.

We have calculated independent t-value also for each group and it was seen that change in Hs CRP values after 48 hours was significant.

**Table 1: Gender distribution of patients**

Gender	No. of patients =105	Percentage
Male	69	65.71
Female	36	34.29

**Table 2: Clinical condition after 48 hr**

Improved	33	31.43
Same	40	38.10
Deteriorated	32	30.47

**Table 3: Clinical characteristics and values of hs-CRP**

Positive on admission	94
Positive after 48 hr	99
At admission / 48hrs(n=97)	
Mean	60.22/66.32
Mode	39.5/75.31
Median	55/60.5
Standard deviation(admission /48hours)	39.58/22.69
hsCRP comparison against age	1.69
Paired t-test value (p-value)	(0.191)

**Table 4: Comparison of hs-CRP in Same, Deteriorated & Improved group**

	Mean At admission/ 48 hours	Median At admission/ 48hours	
Same	30.78/45.67	27.02/38.97	
Deteriorated	49.68/72.78	50.36/82.37	
Improved	82.96/55.97	75.32/52.69	
Standard deviation (admission /48hours)	31.62/39.76	29.03/35.06	
Independent t value			
	Same	Deteriorated	Improved
Mean (rise/ fall)	Rise	Rise	Fall
Median (rise/ fall)	Rise	Rise	Fall
Standard deviation (rise/ fall)	Rise	Rise	Fall
Average fall /rise	Rise	Rise	
change in hsCRP values after 48 hours	p- value 0.001	Significant	

## Discussion

Serum concentrations of CRP increase several hundredfold in response to bacterial infection, making it an attractive diagnostic test for neonatal sepsis. Because many of the more than 70 publications on this subject that have appeared during the past 30 years were flawed by imprecise diagnostic criteria, absent or inappropriate controls (eg, healthy neonates), incomplete description of results, or inadequate sample sizes, the role of this test in evaluation of neonates remains controversial. Early reports described a high prevalence of elevated CRP levels in infected infants, but levels are elevated in only 35% to 65% of neonates with bacterial infection at the onset of illness. Recognition that a delay of at least several hours is intrinsic to the cascade of events leading to elevation of serum CRP levels (including activation of neutrophils, elaboration of interleukin-6, and induction of hepatic synthesis of CRP) led to appropriate criticism of this test as having insufficient sensitivity to guide therapy either by reliably diagnosing or excluding bacterial infection. Noting that CRP levels are consistently elevated 24 to 48 hours after the onset of infection.<sup>9</sup>

In our study, total 105 cases were enrolled in the study in which infection was suspected at the time of admission based on the category in IMNCI/FIMNCI for suspicion of infection. Out of these, 105 cases were successfully followed up to 48 hours to find the correlation of Hs CRP with clinical condition of the patient. Bacterial sepsis considered a life-threatening emergency with high mortality and morbidity in neonates. Although blood culture was considered as the golden standard for diagnosis of neonatal sepsis, it is time consuming, with lower sensitivity and often high false-negative results.<sup>10</sup> For example, in a series of 50 critically ill sepsis patients, Schmit, et al. observed that CRP on admission was  $16.7 \pm 10.6$  mg/dL and that the magnitude of CRP decrease was associated with response to antimicrobial therapy.<sup>11</sup>

Povoa, Schmit and colleagues continued their longstanding work on C-reactive protein (CRP) kinetics by evaluating the patterns of evolution of CRP in patients with severe community-acquired pneumonia (CAP).<sup>12,13</sup> In a study conducted by LOBO SM et al., CRP is an acute-phase protein synthesized by the liver after stimulus by cytokines and its serum levels increase markedly within hours after the onset of infection, inflammation or tissue injury. Decreasing plasma concentrations of this biomarker have been used as an indicator for resolution of infection or sepsis.<sup>14</sup> Coelho et al. studied 891 intensive care unit patients with community-

acquired sepsis, observing a mean hospital admission CRP level of  $20.1 \pm 13.9$  mg/dL and finding association between rates of CRP decline and hospital survival.<sup>15</sup> A time-dependent analysis was performed and CRP ratios were calculated daily in relation to the CRP concentration on day 0, considered equal to 1. They showed that survivors of CAP had a continuous decrease of the CRP ratio during the first week of antibiotic therapy.<sup>13</sup> Along with cases showing a clinical improvement in our study, 25 cases showed a fall in Hs CRP values after 48 hours. Hence a decreasing trend of Hs CRP was seen in majority of cases from admission to 48 hours of admission. Only patients with severe sepsis failed to show a significant change in Hs CRP values i.e. 30%.

The secretion of CRP begins within 4–6 h of the stimulus, doubling every 8 h and peaking at 36–50 h. With a very intense stimulus, the CRP concentration can rise above 500 mg/l, i.e. more than 1000 times the reference value.<sup>16,17</sup> After disappearance or removal of the stimulus, CRP falls rapidly, as it has a half-life of 19 h. However, CRP can remain elevated, even for very long periods, if the underlying cause of the elevation persists. With the exception of severe hepatic failure, CRP rises whenever an inflammatory process is present; its serum concentration only depends on the intensity of the stimulus and on the rate of synthesis. The CRP level is independent of the underlying pathology and is not modified by any therapy or intervention such as renal replacement therapy. Only those interventions affecting the inflammatory process responsible for the acute phase reaction can change the CRP level.

In a study by Suprin et al., mean values were 70 mg/l in systemic inflammatory response syndrome (SIRS) patients, 98 mg/l in sepsis, 145 mg/l in severe sepsis and 173 mg/l in septic shock, probably reflecting different degrees of inflammatory response.<sup>16</sup>

Our study also is similar values at admission, but as shown in above discussion it can be deduced that fall or rise documentation was not important in all groups and clinical acumen is more important. In fact, it is not worthy to prick the child frequently to show that child has deteriorated or improved.

Recent study by Nagwan I. Rashwan et al states that CRP could be a helpful prognostic marker in late onset neonatal sepsis. Hs CRP and PCT have higher diagnostic accuracy in neonatal sepsis in comparison to other studied markers. Both IL-6 and presepsin have equal diagnostic utility in neonatal sepsis, but presepsin could be helpful diagnostic marker in early onset neonatal sepsis.<sup>18</sup> In a study for Evaluation of IL-6, CRP and Hs-CRP as Early Markers of Neonatal Sepsis by Purushothaman Ganesan et al in India it was concluded that IL-6 is a highly sensitive marker and CRP is a more specific marker for the diagnosis of neonatal sepsis. Hs-CRP is a less reliable marker. So, the combination of IL-6 and CRP are the better predictors of neonatal sepsis. However, the sample size of this study was only 40, so may have biased reporting. However, it is noteworthy that in this study, Hs CRP showed sensitivity of 92% and specificity of 34.24%.<sup>19</sup> In extensive work done by William E. Benitz et al Serial Serum C-Reactive Protein Levels in the Diagnosis of Neonatal Infection, it was observed that Serial CRP levels are useful in the diagnostic evaluation of neonates with suspected infection. Two CRP levels  $< 1$  mg/dL obtained 24 hours apart, 8 to 48 hours after presentation, indicate that bacterial infection is unlikely. The sensitivity of a normal CRP at the initial evaluation is not sufficient to justify withholding antibiotic therapy. The positive predictive value of elevated CRP levels is low, especially for culture-proven early-onset infections. This was very old study of 1998 and since now Hs CRP has altered the values but principle remains same.<sup>20</sup>

In our study also similar results are found and it can be concluded that serial Hs CRP may not be helpful if it is already high initially.

## Conclusion

The present study concluded that the hs-CRP levels were increased in all cases of suspected neonatal sepsis. They remained high in neonates who had deteriorated or remained same clinically at 48 hours of follow-up. However, they significantly reduced in neonates showing clinical improvement.

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