THE ROLE OF HEPARIN – BINDING PROTEIN IN THE DIAGNOSIS AND PROGNOSIS OF SEPSIS SYNDROME IN PEDIATRIC PATIENTS AT THE PHILIPPINE CHILDREN'S MEDICAL CENTER

PAULA PILAR G. EVANGELISTA, MD AND JESUS NAZARENO J. VELASCO, MD

ABSTRACT

BACKGROUND: The burden of sepsis is global despite measures to improve its prompt recognition. However, there is no single reliable parameter for its early detection. Heparin-binding protein (HBP) is a new and promising biomarker for sepsis. Presently, there are no published reports in children apart from a limited study on UTI.

OBJECTIVE: To evaluate the role of HBP as a diagnostic tool and prognostic marker of sepsis syndrome among pediatric patients.

METHODS: This prospective cohort study enrolled pediatric patients who were categorized as SIRS or sepsis syndrome. HBP assay was determined on Day1. Likewise, blood culture was taken. A 7-day observation period using PELOD scoring was done. Final category as SIRS or sepsis syndrome was done on Day7. Statistical analysis was done to know relationship of HBP level to SIRS and sepsis.

RESULTS: 106 patients were included in this study. There was statistical significance in the correlation of HBP assay with presence of growth in blood culture and toxic granulations, length of ventilator support, and development of complications including mortality. The cutoff point was >125ng/mL. Sensitivity and specificity for HBP in sepsis syndrome were 98.31% and 97.87% respectively. Positive predictive value was 98.3%. Negative predictive value was 97.9%. Positive likelihood ratio was 46.2. Negative likelihood ratio was 0.017. Risk ratio was 47.6. Subjects with HBP level of >125 ng/mL had 47.6 times the risk of having sepsis syndrome as compared to those with level <125 ng/ml.

CONCLUSION & RECOMMENDATIONS: Elevated HBP level is a useful diagnostic and prognostic marker for childhood sepsis syndrome. Determination of HBP levels at different time intervals within a longer observation period may give a more accurate description of subject's clinical improvement or progression to MODS or mortality.

KEYWORDS: Heparin-binding protein, HBP, Pediatric Sepsis, Sepsis, SIRS

INTRODUCTION

Sepsis is the body's systemic inflammatory response to infection and can progress to severe sepsis, septic shock and ultimately multiple organ dysfunction syndrome.[1] The burden of sepsis is felt globally in both developed and developing countries with an estimate of 20 to 30 million patients afflicted every year. It is a significant cause of morbidity and mortality not only in adults but also in children. In the developing world, sepsis accounts for 60-80% of lost lives in childhood, with more than 6 million neonates and children affected annually[2]. In the Pediatric Critical Care Unit of Philippine Children's Medical Center, sepsis, septic shock and MODS are consistent entries in top 10 morbidity and mortality lists. As the early presentation of childhood sepsis is often difficult to distinguish from less serious viral illnesses, numerous studies have attempted to identify parameters to distinguish children at risk from sepsis.

Several biomarkers have been used to assess the risk of sepsis. These include white blood cell count (WBC), C-reactive protein (CRP) and erythrocyte sedimentation rate (ESR). Likewise, several clinical scales have also been tried to assess the risk of sepsis. However, these have not shown to be adequately sensitive nor specific tools in determining sepsis in the pediatric population. Numerous published reports mostly in adults have identified novel biomarkers as promising laboratory parameters for predicting the risk of sepsis. These include lactate, prolactin and interleukin. Unfortunately, there is no single reliable parameter to assess the risk of sepsis in children.

More recently, a few studies have shown the usefulness of another biomarker in predicting sepsis and mortality. Heparin Binding Protein is a new marker that has demonstrated utility in identifying patients at risk of developing severe sepsis. Heparin-Binding Protein (HBP), also known as CAP37 and azurocidin, is synthesized in neutrophils.[4] Once released from activated neutrophils, it induces a rearrangement of the endothelial cell cytoskeleton, resulting in increased permeability of the endothelium resulting to vascular leakage. At the site of infection, HBP is responsible for the recruitment and activation of monocytes and other inflammatory mediators. It is also internalized by monocytes to prolong survival and enhance cytokine production.[5] HBP therefore directly contributes to the maintenance and progression of inflammation.[6]

In a comprehensive review of the utility of biological mediators in the diagnosis of sepsis, HBP is considered one of the emerging biomarkers. The non-specific physiologic criteria of sepsis syndrome do not adequately identify patients who might benefit from either conventional anti-infective therapies or from novel therapies that target specific mediators of sepsis. The utility of a biomarker is a function of the degree to which it adds value to the available clinical information in the domains of screening, diagnosis, risk stratification and monitoring of the response to therapy. Validated biomarkers of sepsis may improve diagnosis and therapeutic decision making for these high risk patients, but will require an unprecedented degree of systematic investigation and collaboration.[8]

In a prospective study of 233 adult febrile patients with suspected sepsis, high plasma levels of HBP helped to identify patients with an imminent risk of developing sepsis with circulatory failure. HBP was shown to be the best predictor of severe sepsis with Receiver-operating characteristic (ROC) plot showing an Area Under Curve (AUC) of 0.95, exceeding that of Procalcitonin, IL-6 and Lactate. A plasma HBP level $\geq$ 15 ng/mL was a better indicator of severe sepsis (with or without septic shock) than any other laboratory parameter investigated. Thirty-two of the 70 patients with severe sepsis were sampled up to 12 hours before signs of circulatory failure appeared and in 29 of these patients, HBP plasma concentrations were already elevated.[9]

Another prospective study was conducted of two patient cohorts totaling 179 patients treated in the ICU at Karolinska University Hospital Huddinge in Sweden in 2012. Plasma HBP levels were significantly higher in patients with severe sepsis or septic shock compared to patients with a non-septic illness in the ICU. HBP was associated with severity of disease and an elevated HBP at admission was associated with an increased risk of death. HBP that rises over time may identify patients with a deteriorating prognosis.[10]

Recently, Lertdumrongluk et al. investigated heparin-binding protein as a diagnostic tool in acute pyelonephritis (APN). Urine HBP (UHBP) levels were measured at baseline and 1 month after antimicrobial therapy in children suspected with APN vs. controls. Results showed levels of 47.0 $\pm$ 8.4 and 16.6 $\pm$ 3.8 vs. 15.0 $\pm$ 2.9 ng/mL respectively. Test performance characteristics were calculated against a gold standard of positive urine cultures and compared with leukocyte esterase (LE) and nitrite measured by dipsticks and pyuria by microscopy. The sensitivity and specificity for UHBP levels $\geq$ 34 ng/mL were 100% and 100%.[13]

Despite the growing interest in HBP as a diagnostic tool in severe infections and inflammatory conditions, most studies are done in adults. The limited data on children were focused on urinary tract infection. At present, there are no published reports on the usefulness of HBP in childhood sepsis.

The mortality associated with sepsis is still substantial despite the increasing awareness of the diagnosis and recent advancements in treatment strategies. An important task for the clinician is to recognize sepsis before it progresses into a more severe state with signs of circulatory failure. Thus, a reliable molecular tool identifying patients who are at risk of developing severe sepsis among patients presenting with fever and signs of systemic inflammatory response would decrease the critical span to adequate treatment and be of considerable clinical value.[9]

At the Pediatric Critical Care Unit of the Philippine Children's Medical Center, there are approximately 40-50 admissions and referrals each month. Around 30-50% of them are due to sepsis syndrome.

We aimed **t**o evaluate the role of heparin-binding protein as a diagnostic tool and prognostic marker of sepsis syndrome among pediatric patients less than 19 years old admitted at the Philippine Children's Medical Center. Specifically, we wished to ascertain the relationship of heparin-binding protein and the following sepsis categories:

a) SIRS
b) Sepsis Syndrome**;**
to compute for the overall sensitivity, specificity, predictive value and likelihood ratio of heparin-binding protein in detecting the following:

a) SIRS
b) Sepsis Syndrome**;**
and to determine the cutoff value of heparin binding protein for

a) SIRS
b) Sepsis Syndrome

METHODOLOGY

This is a prospective cohort study. Pediatric patients less than 19 years old and fulfilling the SIRS and sepsis syndrome based on the International Consensus Conference on Pediatric Sepsis were included in the study.[16] Those with febrile neutropenia and obvious viral infection were excluded from this study.

Each subject was initially categorized as to SIRS or Sepsis Syndrome. After extraction of blood samples, a 7-day observation period ensued. Subjects were then reclassified as to SIRS or Sepsis Syndrome as final category. Subjects under the Sepsis Syndrome were further subcategorized as Sepsis, Severe Sepsis, Septic Shock or MODS.

Demographic profile was tabulated including name, age and sex. Laboratory profile was tabulated including white blood cell count, actual neutrophilic count, presence of toxic granules, actual platelet count, blood culture and other cultures taken, biomarkers lactate and procalcitonin when available and the HBP assay in ng/ml. Clinical profile was tabulated including initial category, number of hospital days, use of ventilatory support, outcome (no complications, with complications, mortality or readmission) and final category after the 7-day observation period.

A daily PELOD scoring was done for seven days from extraction of HBP level to monitor possible development or progression of condition to a sepsis spectrum like MODS. After seven days from enrolment to the study, a diagnosis with the worst sepsis syndrome category was labeled as the final category. This final category was compared to the actual HBP level. Clinical and Laboratory Outcome measurements such as the length of hospital stay and ventilator days, outcome, WBC count, blood culture result, presence or absence of toxic granules were also correlated with the HBP levels.

Data were described using means, standard deviations, percentages and frequency counts. T-test for independent samples was used to determine significant difference between

means of two groups, ANOVA one way to compare three or more groups. Chi-square and Fischer's exact test were used to determine significant difference in the distribution of categorical variables. Odds ratio was computed where feasible. Minitab Ver 17 was used as statistical software. ROC Analysis using MedCalc software was done to determine optimum cutoff points, after which accuracy parameters were determined such as sensitivity, specificity, predictive values and likelihood ratios. For all tests, a 95% confidence level was considered significant ($p<0.05$).

RESULTS

One hundred and nine (109) subjects were enrolled in the study. However, only 106 fulfilled the inclusion criteria. All 106 subjects were categorized as to SIRS vs. Sepsis Syndrome twice. Initially, upon enrolment to the study and prior to the HBP assay which was based on history, clinical parameters and basic laboratory exams including blood counts. Finally, after the 7-day observation period when the HBP assay was run which was based on further clinical findings as well as other diagnostic parameters including blood culture. Final category that was correlated with the HBP assay was the post-7-day observation period.

Table 1 shows the demographic profile of the subject population. Mean age was more than 3.5 to less than 4.5 years which did not show statistical significance on both categories. Gender likewise did not show statistical significance with male predominance on both categories. Of the 106 subjects, 44% (N = 47) was classified as SIRS while 56% (N = 59) was classified as Sepsis Syndrome.

TABLE 1. DEMOGRAPHIC PROFILE

	SIRS (N = 47)	**Sepsis Syndrome** (N = 59)	**P Value**
Age			0.461
Mean+/SD	*4.45+/5.13*	*3.72+/4.91*	
Sex			0.845
Male	*28 (60%)*	*34 (57.6%)*	
Female	*19 (40%)*	*25 (42.4%)*	

The SIRS category included the following diseases: 61.7% (29 of 47) was pneumonia; 6.4% (3 of 47) was acute gastroenteritis; 4.3% (2 of 47) each for systemic lupus erythematosus in flare, rheumatic heart disease in failure and enterocolitis; and 2.1% (1 of 47) each for typhoid fever, shunt malfunction, cor pulmonale, lymphoma, cellulitis, rhabdomyosarcoma, ear trauma with hemophilia B, fatal arrhythmia with electrolyte imbalance and thalassemia and abscess.

The Sepsis Syndrome Category is broken down into the following subcategories: 23.7% (14 of 59) was sepsis; 20.3% (12 of 59) was severe sepsis; 47.5% (28 of 50) was septic shock; and 8.5% (5 of 59) was MODS.

Table 2 shows the clinical profile for each category. All parameters including the hospital stay, ventilator days and outcome were statistically significant. For purposes of this study, "complications" included worsening of clinical symptoms or development of conditions that can affect prognosis like pneumothorax, effusions; change of current antibiotic due to worsening condition or non-response; increase in oxygen requirement, or need for ventilatory support.

There were more subjects with $\geq$ 7 days hospital stay in sepsis syndrome at 73% compared to SIRS at 27%. Comparatively, more subjects had shorter hospital stay of < 7days in SIRS at 47%. Expectedly, fewer subjects required ventilator support in SIRS at 85.1% compared to sepsis syndrome at 35.6%. On subject outcome, majority of those under the SIRS category showed no complications at 72.3%. In contrast, 62.7% showed complications, 33.9% expired and 1.7% was readmitted under the sepsis syndrome category.

TABLE 2. CLINICAL PROFILE

	SIRS (N = 47)	**Sepsis Syndrome** (N = 59)	**P Value**
Hospital Stay			0.043
< 7 days	*22 (47%)*	*16 (27%)*	
≥ 7 days	*25 (53%)*	*43 (73%)*	
Ventilator Days			**< 0.0001**
<7 days	*3 (6.4%)*	*20 (33.9%)*	
≥ 7 days	*4 (8.5%)*	*18 (30.5%)*	
Not ventilated	*40 (85.1%)*	*21 (35.6%)*	
Outcome			**< 0.0001**
No complications	*34 (72.3%)*	*1 (1.7%)*	
With complications	*9 (19.2%)*	*37 (62.7%)*	
Expired	*4 (8.5%)*	*20 (33.9%)*	
Readmitted	*0 (0%)*	*1 (1.7%)*	

Table 3 shows the laboratory profile of the subjects. White blood cell counts under SIRS are slightly lower than in the sepsis syndrome with mean values of 14.8 and 18.1 respectively. While there was leukocytosis in both categories, there was no statistical significance. Likewise, platelet counts in both categories did not show statistical significance. Blood cultures and appearance of toxic granules, however, showed statistical significance. Blood cultures showed positive growth on 30.5% of subjects under sepsis syndrome and 0% growth on SIRS. Toxic granules appeared on 13.6% of subjects under sepsis syndrome and none under SIRS.

TABLE 3. LABORATORY PROFILE

	SIRS (N = 47)	**Sepsis Syndrome** (N = 59)	**P Value**
WBC			0.075
Mean +/SD	*14.84+/8.48*	*18.07+/9.66*	
Platelet Count			0.249
Mean +/SD	*357.34+/209.85*	*309.95+/207.18*	
ANC			0.210
Mean+/SD	*9,935.85+/7054.56*	*11,743.42+/ 7571.10*	
Culture			**0.00003**
Positive	*0 (0%)*	*18 (30.5%)*	
Negative	*47 (100%)*	*41 (69.5%)*	
Toxic granules			**0.0084**
Positive	*0 (0%)*	*8 (13.6%)*	
Negative	*47 (100%)*	*51 (86.4%)*	

Table 4 shows the relationship of clinical and laboratory parameters with HBP levels. Although there was an increase in the mean HBP level with hospital stay ≥ 7 days, this was not statistically significant. The length of ventilator days was significantly associated with HBP levels. Mean HBP level of subjects without ventilator support was significantly lower than those with ventilatory support. However, mean HBP level of subjects with ventilator use of < 7 days and ≥ 7 days use was not statistically significant.

Outcome was also significantly associated with HBP levels. Subjects without complications had significantly lower mean HBP level compared to those with complications and those who expired. However, the mean HBP level of the group with complications did not differ significantly from that of the group with subjects who died. The outcome variable "readmitted" was not included in the analysis since it only had one subject, thus, mean and standard deviation could not be computed.

The mean HBP level of subjects found with toxic granules was significantly higher than the subjects without toxic granules. Similarly, the mean HBP level of subjects with growth on blood culture was significantly higher than those without growth. There was increase in the mean HBP level for age group 1 month to 1 year with WBC > 17.5 as well as for age group 6 to 12 years with WBC > 13.5, In contrast, the mean HBP level was close for age group of 2 to 5 years and for age group 13 to 19 years with varying WBC counts. Despite these findings, there was no statistical significance across all age groups.

TABLE 4. RELATIONSHIP OF CLINICAL AND LABORATORY PARAMETERS WITH HBP LEVELS

	HBP (ng/ml)
Hospital Stay (days)	
< 7 days (N = 38)	125.6+/91.0
≥ 7 days (N = 68)	150.5 +/73.4
P value	*0.126*
Ventilator Days	
No ventilator (Group 1: N = 61)	110.7 +/75.7
< 7 days (Group 2: N = 23)	201.3 +/64.7
≥ 7 days (Group 3: N = 22)	164.8 +/67.3
P value	***< 0.0001***
Post-hoc T-test after a significant ANOVA	*Group 1 vs. Group 2 = SIG* *Group 1 vs. Group 3 = SIG* *Group 2 vs. Group 3 = NS*
Outcome*	
No complications (Group 1: N = 35)	57.8 +/39.3
With complications (Group 2: N = 46)	177.2 +/57.1
Mortality (Group 3: N = 24)	194.3 +/74.4
P value	***< 0.0001***
Post-hoc T-test after a significant ANOVA	*Group 1 vs. Group 2 = SIG* *Group 1 vs. Group 3 = SIG* *Group 2 vs. Group 3 = NS*
Toxic Granules	
With (N = 8)	202.8+/28.8
Without (N = 98)	136.6 +/81.5
P value	***< 0.0001***
Blood Culture	
No growth (N = 88)	127.5 +/79.3
With growth (N = 18)	210.3+/45.0
P value	***< 0.0001***
WBC	
For age group 1 month - 1 year	
> 17.5 or < 5 (N = 26)	163.3 +/61.2
5 - 17.5 (N = 29)	131.1+/82.4
P value	*0.115*
For age group 2 years - 5 years	
> 15.5 or < 6 (N = 13)	131.8+/75.6
6 - 15.5 (N = 8)	131.7+ 91.6
P value	*1.00*
For age group 6 - 12 years	
> 13.5 or < 4.5 (N = 12)	154.7+/88.6
4.5 - 13.5 (N = 8)	113.7 +/94.9
P value	*0.335*
For age group 13 – 19 years	
>11 or < 4.5 (N = 5)	143.6+/100.7
4.5-11 (N = 5)	142.4+/104.4
P value	*0.984*

**One subject readmitted, not included in outcome.*

Table 5 shows the mean HBP levels of SIRS and Sepsis Syndrome and the sub-categories of Sepsis Syndrome. Statistical significance was seen between the values in SIRS vs. Sepsis Syndrome with mean HBP level of 62.7 ng/mL and 204.5 ng/mL respectively. Statistical significance was likewise seen in SIRS vs. the different sub-categories of Sepsis Syndrome. Among the different sub-categories, the statistical difference of Category 2 (Sepsis) vs. Categories 3 (Severe Sepsis), 4 (Septic Shock) and Category 5 (MODS) was significant. The statistical differences among Categories 3, 4 and 5 were not significant.

TABLE 5. MEAN HBP LEVEL OF SIRS AND SEPSIS SYDROME AND SUB-CATEGORIES OF SEPSIS SYNDROME

	Mean	**SD**
HBP Determination Post-7 Days Observation Period		
Sepsis Syndrome	204.5	42.0
SIRS	62.7	34.4
P value	***< 0.0001***	
***Sub-Categories of Sepsis Syndrome**		
Category 1: SIRS (N = 47)	62.7	34.4
Category 2: Sepsis (N = 14)	161.4	26.4
Category 3: Severe Sepsis (N = 12)	210.6	33.5
Category 4: Septic Shock (N = 28)	219.6	37.6
Category 5: MODS (N = 5)	226.1	43.9
P value	***< 0.0001***	

**Significant Pairs: Category 1 vs. 2; Category 1 vs. Category 3; Category 1 vs. Category 4; Category 1 vs. Category 5; Category 2 vs. Category 3; Category 2 vs. Category 4; Category 2 vs. Category 5.*
**Category 3 vs. Category 4 and Category 5, and Category 4 vs. Category 5 not significantly different.*

The line diagram in Figure 2 illustrates the diagnostic role of HBP assay in determining Sepsis Syndrome. The HBP cutoff point for this study was determined by receiver operating characteristic (ROC) analysis.

FIGURE 2.

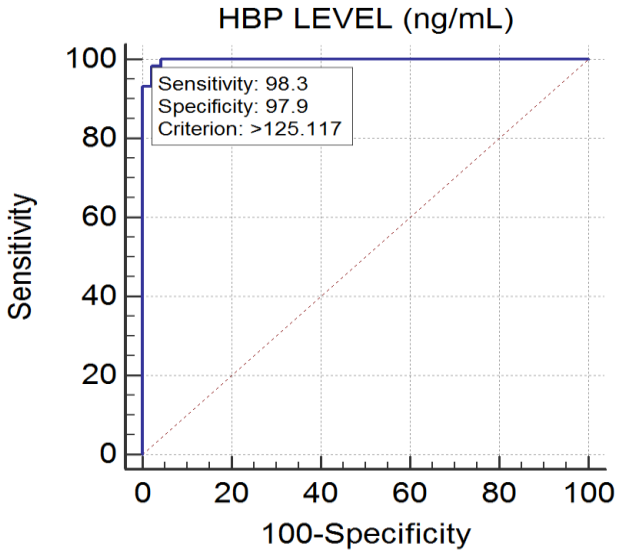

Table 6-A shows that for Day 1, HBP level for low risk subjects based on PELOD score was significantly lower than those categorized as medium to high risk with computed difference of 89.6 ng/ml. For Day 7, HBP level for low risk was likewise significantly lower than those categorized as medium to high risk with computed difference of 94.2 ng/ml.

TABLE 6-A. ASSOCIATION OF HBP WITH PELOD SCORES AT DAY 1 AND DAY 7

PELOD SCORE DAY 1				
	LOW RISK < 10 (N=80)	MEDIUM TO HIGH RISK * ≥ 10 (N=26)	P VALUE	Mean A – Mean B and 95% CI of the difference
Mean HBP ± SD	119.6 ± 77.3	209.2 ± 46.8	< 0.0001	- 89.6 (± 24.96)
PELOD SCORE DAY 7				
	LOW RISK <10 (N=60)	MEDIUM TO HIGH RISK * ≥10 (N=46)	P VALUE	Mean A – Mean B and 95% CI of the difference
Mean HBP ± SD	100.7 ± 67.4	194.9 ± 63.9	< 0.0002	- 94.2 (± 25.5)

**Only 2 subjects with Medium Risk for Day 1 and 3 subjects for Day 7, hence, pooled with High Risk category. T-test for independent samples used.*

In Table 6-B, the correlation coefficient between values of HBP and PELOD scores was high and significant, with R higher at Day 7.

TABLE 6-B. CORRELATION OF HBP LEVELS WITH PELOD SCORES (HBP AS DEPENDENT VARIABLE) AT DAY 1 AND DAY 7

	PEARSON'S R OR CORRELATION COEFFICIENT	CRITICAL VALUE FOR SIGNIFICANCE	P VALUE
DAY 1	0.440	0.196	< 0.0001
Day 7	0.527	0.196	< 0.0001

Categories based on pre-HBP determination were significantly associated with PELOD score at Day 1. That is, with a higher proportion of subjects categorized as medium to high risk associated with having sepsis syndrome (85%) than those categorized as low risk (40%). The computed odds ratio was 8.25 indicating that the odds or chance of a subject categorized as medium to high risk for progression to MODS was 8.25x higher among those with sepsis syndrome than those with SIRS. At Day 7, the association was also highly significant between the two variables, with computed odds ratio of 15.2 indicating that those with sepsis syndrome have 15x greater odds or chance to progress to MODS than those with SIRS.

SIRS is significantly associated with PELOD score at day 7, with SIRS 5.2% less likely to occur with a PELOD Score of ≥ 10 or medium to high risk for progression to MODS. Sepsis and severe sepsis, on the other hand did not show significant association with PELOD. Septic shock was significantly associated and is 15.3x more likely to occur with a PELOD Score of ≥ 10. MODS was also significantly associated with PELOD but the odds ratio could not be computed in the presence of a zero value. However, an estimate of RR showed that subjects with MODS were 2.5x more likely to get a PELOD Score of ≥ 10.

DISCUSSION

The diagnosis of sepsis and its progression to MODS remain elusive despite several published guidelines and the introduction of novel biomarkers. It is for this reason that studies have continually been conducted to find more reliable parameters.

The data and findings of this study showed that clinical parameters alone for sepsis might not be sufficient in its early stages. Likewise, even standard laboratory parameters including blood culture are not totally reliable. Hence, HBP as an early diagnostic marker for sepsis is very useful and potentially lifesaving.

Upon initial enrolment to the study, the diagnosis of SIRS was 49% (52 of 106) of the subject population while Sepsis Syndrome stood at 51% (54 of 106). In the 7-day observation period, 27% (14 of 52) of patients with initial diagnosis of SIRS progressed to Sepsis Syndrome. Significantly, all of the 14 subjects had an HBP level of > 125 ng/ml with values ranging from 142.92 – 275.49 ng/ml. In the same manner, 17% (9 of 54) of Sepsis Syndrome turned out to be SIRS. And all of the 9 subjects had an HBP level < 125.11 ng/ml with values ranging from 15.74 – 89.03 ng/ml. These findings highlight the limitation of clinical findings alone in the diagnosis of sepsis syndrome. Out of the 14 subjects with initial SIRS, 6 turned out to be Sepsis (42.8%) with HBP levels ranging from 143.22 – 209.58 ng/ml; 2 progressed to Severe Sepsis (14.3%) with HBP levels ranging from 208.716 – 275.49 ng/ml; and 6 progressed to Septic Shock (42.8%) with HBP levels ranging from 142.92 – 228.132 ng/ml.

Out of the 45 subjects with initial and post-7 days observation period diagnosis of Sepsis Syndrome, 8 turned out to be Sepsis (17.8%) with HBP levels ranging from 117.32 – 197.827 ng/ml; 10 progressed to Severe Sepsis (22.2%) with HBP levels ranging from 173.4 – 264.87 ng/ml; 22 progressed to Septic Shock (48.9%) with HBP levels ranging from 168.06 – 283.96 ng/ml; and 5 progressed to MODS (11.1%) with HBP levels ranging from 170.58 – 289.62 ng/ml. Although there was no statistical significance among the subcategories of Sepsis Syndrome, it is interesting to note that there was progressive increase of HBP levels as the severity of Sepsis Syndrome increases. In this group, the Sepsis subcategory had the lowest HBP level at 117.32 ng/ml while the MODS subcategory had the highest HBP level at 289.62 ng/ml.

There were 26 mortalities in the study population giving a mortality rate of 24.5%. Out of these, 84.6% (22 of 26) belonged to the category of Sepsis Syndrome. Sepsis accounted for 1 mortality (3.8%) with HBP level of 181.309 ng/ml. Severe Sepsis also accounted for 1 mortality (3.8%) with HBP level of 173.4 ng/ml. Likewise, MODS accounted for 1 mortality (3.8%) with HBP level of 242.226 ng/ml. Septic Shock accounted for 19 mortalities (86.4%) with HBP levels ranging from 168.06 – 283. 96 ng/ml. Interestingly, the highest HBP level of 283.96 ng/ml in this study belonged to the MODS subcategory who expired on the 3rd day of observation period.

There were 4 mortalities in the SIRS category with HBP levels ranging from 19.45 – 70.88 ng/ml. While these 4 subjects only fulfilled the SIRS criteria and had HBP levels below the cutoff value established in this study, their mortality outcome was surprising. Within the 7-day observation period, their progression of illness was confirmed as due to non-septic causes. One mortality was diagnosed with Rhabdomyosarcoma and expired from Stage 4 Disease. Another mortality was diagnosed with Thalassemia and died of fatal arrhythmia secondary to hypokalemia. Another mortality was diagnosed with Rheumatic Heart Disease, Mitral Stenosis Severe who died of low cardiac output syndrome. While the last mortality was diagnosed with Prader Willi Syndrome, Obstructive Sleep Apnea who expired from complications of Cor Pulmonale. These findings add credibility to the usefulness of HBP assay in ruling out sepsis as an initial diagnosis especially since the above HBP levels fall way below the cutoff level of > 125.11 ng/ml established in this study. Moreover, it helps the physicians to consider causes other than sepsis given the limitation of clinical findings. This

may prove timely and lifesaving in the management of other illnesses mimicking or overlapping with sepsis syndrome. In addition the cost of sepsis burden is minimized when the diagnosis is early and management is prompt.

There were 35 subjects out of the total population (33%) who did not develop complications with HBP levels ranging from 5.9 – 140.319 ng/ml. Thirty-four out of 35 (97.1%) belonged to the SIRS category while only 1 (2.9%) belonged to the Sepsis Syndrome category. Out of the 34 SIRS, 19 subjects (55.88%) did not require ventilator support and were discharged in less than seven days with HBP levels ranging from 5.9 – 125.117 ng/ml. Out of the 34 SIRS 13 subjects (38.23%) did not require ventilator support but stayed in the hospital for more than seven days for varying reasons with HBP levels ranging from 8.08 – 106.27 ng/ml. Only 2 subjects out of the 34 SIRS (5.88%) required ventilatory support and stayed in the hospital for more than seven days with HBP levels ranging from 67.79 – 76.54 ng/ml. Of the 2 SIRS requiring ventilator support, one subject was diagnosed with Hemophilia A Severe with Intracranial Bleed and developed healthcare-associated infection while the other subject was diagnosed with Congestive Heart Failure. These diagnoses could explain the low HBP levels noticeably falling below the cutoff value of > 125.11 ng/ml established in this study. The lone subject belonging to the Sepsis Syndrome Category stayed in the hospital for more than seven days but did not require ventilatory support had an HBP level of 140.319 ng/ml which was the highest for the group that did not develop complications.

These findings confirmed the utility of HBP as a credible biomarker for sepsis syndrome and its vast potential for predicting progression to MODS. The disparity between published HBP cutoff point for adults in the diagnosis of sepsis with highest cutoff level > 50 ng/ml and this study with cutoff level >125 ng/ml might be due not only to age difference but also to ethnicity and geographic location.

CONCLUSION

This study showed the usefulness of heparin binding protein in the diagnosis of sepsis syndrome. The HBP cutoff level of > 125.11 ng/ml gave a sensitivity of 98.3% with specificity of 97.87%, PPV of 98.3 with NPV of 97.9 and LR+ of 46.2 with LR- of 0.017. The computed risk ratio revealed 47.6 times the risk of having sepsis syndrome at the same cutoff level. While this study did not establish a statistically significant cutoff point for progression to MODS, the mean HBP levels progressively rose to > 200 ng/ml with progression to MODS.

This study was limited to a one-time determination of HBP level. Determining the HBP levels at different time intervals within a longer observation period may give a more accurate description of the subject's clinical improvement or progression to MODS.

The population distribution as to the different sepsis syndrome categories was also limited in number. It may be helpful to do a study focusing on the different sepsis syndrome subcategories each with a larger population size to produce a more reliable comparison and analysis.

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