ORIGINAL ARTICLE

Procalcitonin (PCT) as a biomarker in hospital acquired septicemia in neonatal intensive care unit (NICUS) and pediatric intensive care unit (PICUS) in Aswan university hospital

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ABSTRACT

Keywords: Sepsis; procalcitonin; CRP.

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Background: Sepsis is a life-threatening illness that results from an infection that damages tissues and organs. Sepsis causes shock, multiple organ failure, and death, especially if not detected and treated swiftly. The most recent definition of sepsis is infection with evidence of organ failure. **Objectives:** to evaluate the use of biomarkers for early detection of septicemia and its prognosis in order to guide antibiotic treatment. **Patient and methods:**This is a case control study that was done at the pediatric intensive care unit and the neonatal intensive care unit in Aswan University Hospital for 20 patients with suspected hospital-acquired septicemia and 20 non-septic patients as a control. **Results:** The mean levels of procalcitonin (PCT) and C-reactive protein (CRP) were considerably greater in the sepsis group than the control group, with a consistent decline over time in accordance with treatment and strong predictive power.**Conclusion:** Procalcitonin (PCT) and C-reactive protein (CRP) may be useful biomarkers for detecting nosocomial sepsis and predicting its outcome.

INTRODUCTION

Sepsis remains a major cause of mortality in infants and children, with approximately three million deaths per year. The World Health Organization and the Global Sepsis Alliance have emphasized the need for early detection of sepsis. Despite advancements in medical care, sepsis continues to have a significant impact on global health. Prompt management is crucial for improving outcomes, as delays in identification and treatment are associated with higher mortality rates.[(1)]

Despite breakthroughs in medical technology and care, sepsis continues to have a substantial worldwide health impact. Time-sensitive management improves outcomes in sepsis, with delays in detection and treatment associated with increased mortality. [(2)]

Sepsis biomarkers may provide information that is not obtainable through other measures, perhaps aiding in clinical decision-making and improving patient treatment. For example, if there were biomarkers that could accurately and specifically diagnose sepsis at an early stage, it would enable the administration of antibiotic therapy in a more timely and suitable manner while also avoiding unnecessary medications. [(3)]

Of the various biomarkers for sepsis, the acute phase reactant C reactive protein (CRP) is considered a valuable and often used biomarker for assessing the severity and prognosis of sepsis as well as monitoring the response to treatment. Recently, there have been suggestions that procalcitonin (PCT), along with other chemicals related to antimicrobial immunity, could serve as potential biomarkers for diagnosing and predicting the outcome of sepsis. [(4)]

PATIENTS AND METHODS

This is a case control study that was done at the pediatric intensive care unit and the neonatal intensive care unit in Aswan University Hospital for 20 patients with suspected hospital-acquired septicemia and 20 non-septic patients as a control.

Ethics and Research committees at Aswan University Hospital gave their approval to the project.

All patients were be subjected to:

- Complete history
- Complete clinical examinations.
- Laboratory investigations:

- CBC with differential on day 1.
- Blood cultures on day 1 and day 4 (For negative cultures on day 1).
- CRP on day 1, day 4 and day 7.
- Procalcitonin on day 1, day 4 and day 7.

- Blood sampling:

Blood samples were collected for biomarkers detection and for culture and sensitivity from all subjects on days 1, 4, and 7 after the start of septicemia, where the first day is the clinical suspicion of hospital-acquired septicemia or clinical manifestations after 48 hours of admission.

8 ml of blood was collected from every suspected case on the first, fourth, and seventh days as:

- **Blood culture**: 2-5 ml aseptically collected for blood culture: The sample was incubated at BD BACTEC 9050 system (Becton Dickinson) as soon as possible for incubation and monitoring, positive samples were processed according to standards**.** [(5)]

After being gram-stained, positive samples were subcultured on blood, MacConkey, and chocolate agar plates, which were then incubated at the proper temperatures. Using the Vitek 2 Compact (bioM'erieux), all of the organisms were fully identified.

- **CRP:**2 ml collected,10µl serum or plasma samples were measured with iChroma II (boditech) immunofluorescence assay analyzer.

- **Procalcitonin (PCT):** 2 ml for procalcitonin, The samples were tested on VIDAS (Biomerieux) according to the manufacture.

200µl serum or plasma were kept at -25 until analysis, procalcitonin was measured with VIDAS® B·R·A·H·M·S PCT™ (bioM´erieux), a compact automated immune analyzer based on an assay that combines a one-step immunoassay sandwich method with a final fluorescent detection (ELFA).

- **Complete blood count (CBC):**The differential count was performed using a Coulter LH750 Analyzer (Beckman Coulter Inc., USA), as well as peripheral blood smears.

Patient selection:

Inclusion criteria:

Any patient admitted to NICU or PICU with any diagnosis other than septicemia aged less than 12 years for a period of one year.

Sepsis Identification:

Any patient admitted to NICU or PICU with any diagnosis other than sepsis who develop within 48 hours at least 3 of the following signs:

- Fever: Temperature >38.0°c
- Bradycardia: Heart rate < 100 beats/min in infants <1year old.
- Apnea: Transient cessation of breathing.
- Oliguria: Low output of urine less than (0.5 ml/kg/h).

Exclusion criteria:

- Any patient admitted to NICU or PICU with diagnosis of sepsis.

Statistical analysis

Researcher verified, coded, and analyzed data using IBM-SPSS 24.0 (IBM-SPSS Inc., Chicago, IL, USA). Statistics by type: Calculated mean, SD, median, range, frequency, and percentages. Kolmogorov-Smirnov and Shapiro-Wilk tests were used to determine continuous variable normality. A chi-square or Fisher's exact test was used to compare group frequency distributions. Student/Mann Whitney t-test Comparisons of dichotomous means and medians were done using U analysis. RM-ANOVA measured mean differences for continuous variables

with more than two categories and repeated assessments. Bonferroni adjustments were used for post-hoc tests. The ROC curve with AUC, SE, and 95% CI was used to compare how well different markers and combinations of markers can diagnose and predict disease. The p-value was considered significant at <0.05**.**

RESULTS:

A total of 40 participants were included in the current study (20 cases diagnosed with sepsis and were eligible for inclusion, along with 20 control individuals).

Table 1 showed the baseline characteristics of the studied groups. Regarding patient's age, cases were significantly (p = 0.044) younger (4 months (7 days-10 years) compared with control (27 months (3 days-10 years). However, sex distribution was comparable in the two groups with a p-value of 0.342 (male/female ratio was 9/11 in control and 12/8 in cases)(Fig. 1).

Respecting the mortality was significantly higher in the septic group (15%) than the control (0%) and this was statistically significant (P = 0.036). Additionally, length of stay at ICU in days, cases had significantly (p<0.001) longer ICU stay duration (7.8 ± 2.1 days) than control (3.4 ± 1.7 days) (Fig. 1).

Cases had significantly (p<0.001) higher mean body temperature (37.3 ± 0.3) than control (38.5 ± 0.5) (Fig. 2). Similarly, sepsis cases had significantly (p<0.001 and 0.003) higher RR and HR (66.5 ± 12.3 cycle/min. and 121.4 ± 19.1 beat/min.) compared with control (42.6 ± 12.8 cycle/min. and 91.7 ± 15.5 beat/min.) (Fig. 3).

The study found that the mean CRP level increased significantly in the sepsis group over time, with a mean increase of 33.7 ± 6.6 at the first day, 37.5 ± 10.9 at the fourth day, and 34.3 ± 12.3 at the seventh day, compared to a steady reduction in the control group (Table 2, Fig. 4).

Concerning the validity of CRP as a tool for prediction of sepsis among the study cohort. The predictive power was excellent for the three-time intervals at cutoff of 12 mg/dl. The CRP test showed 90% accuracy at the first day, 85% specificity, 86% precision, and 94% NPV at the fourth day. It had 85% accuracy at the seventh day, with the lowest predictive power at the seventh day. The test met empirical benchmarks for sepsis prediction at the first, fourth, and seventh days (Table 3, Fig. 5).

The study found a significant decrease in the mean PCT level in the sepsis group over time, with a decrease from the first day to the seventh day. Interestingly, the control group showed no significant reduction in PCT levels. The sepsis group had significantly higher mean PCT levels from baseline to the study end.(Table 4, Fig. 6)**.**

Regarding the validity of PCT as a tool for prediction of sepsis among the study cohort. The study found that PCT is an excellent tool for predicting sepsis, with excellent predictive power at the first day, 4th day, and 7th day. The test met all validity criteria, with 85% sensitivity, 95% specificity, 94.5% PPV, 86% NPV, and 90% accuracy. The Youden index confirmed the predictive power, meeting empirical benchmarks for sepsis prediction(table 5, Fig. 7).

DISCUSSION

Pediatric sepsis is a major cause of morbidity and mortality in babies and children all over the world. Pediatric sepsis has become more common in the last 20 years, and an increasing number of these patients have co-morbid illnesses. [6]

Our study found a higher mortality rate (15%) in septic cases compared to controls ($p = 0.036$), reflecting the severity of this condition.

The mortality rate in our study was close to what was found by **Humoodi et al.** [7] A retrospective cohort research at King Abdulaziz Medical City, Jeddah (KAMC-J), Saudi Arabia, found a 28-day PICU death rate of 23.9%. However, it was higher than that reported by **Hartman et al.** [8], who found a case-fatality rate of 8.9% in retrospective observational cohort data from seven U.S. states.

C-reactive protein (CRP) and procalcitonin (PCT) have shown potential as sepsis biomarkers for early diagnosis and treatment response.

Regarding CRP and for comparison between cases and controls as regards the effect of sepsis on the CRP, our study found significantly elevated CRP levels in the sepsis group compared to controls at all time intervals (1st day $p < 0.001$, 4th day $p = 0.012$, 7th day $p = 0.026$).

These results align with the findings of **Sakyi et al.** [9], who similarly observed significantly elevated levels of CRP in septic patients compared to the control group ($p < 0.0001$).

Concerning the validity of CRP as a tool for prediction of sepsis among the study cohort, the present study detected that the predictive power was excellent for the three-time intervals at a cutoff of 12 mg/dl, best results were botained in the first day.

Although the predictive power of CRP decreasesover time, the predictive value of CRP serial measurements is superior to a single dose (the p value was < 0.001 in the three measurements). This was in line with **McWilliam and Riordan, 2010** [10]which found that a single CRP measurement is not sufficient for detecting serious bacterial illness, but serial measurements can help monitor treatment effectiveness or make a comparison between cases and controls.

Similarly, our study found significantly higher PCT levels in the sepsis group compared to controls at all time intervals.

This is in line with the findings of **Downes et al.'s** [11] prospective cohort study, which found that PCT was a better predictor of bacterial infections compared to CRP as a single biomarker ($p = 0.01$).

The predictive power of PCT was also excellent in our study cohort, with slowly declining levels and perfect predictive values on the first day of evaluation. This highlights the potential of PCT as a valuable tool for predicting sepsis in pediatric patients.

The decrease in PCT levels can be attributed to the observed impact of antibiotics, as it has been demonstrated that marker plasma concentrations decrease following the administration

of antibiotics. This advantage renders it more precise compared to CRP, aligning with the findings of **Fioretto et al.** [12].

Also in accordance with**Schuetz et al.** [13]. PCT has demonstrated value in directing antibiotic therapy. When PCT levels fall to less than 80% of the peak value or below 0.5 g/L in these individuals, stopping antibiotic medication should be taken into consideration.

In contrast to our study, **Nellis et al.** [14], found no statistically significant differences ($p = 0.12$) between the accuracy of PCT for bacteremia and CRP in their population. The accuracy of PCT alone revealed no change with the rate of rise of PCT ($p = 0.81$) at a ROC curve threshold of 2 ng/dL. PCT's sensitivity for bacteremia was 69.2%, specificity was 74.4%, PPV was 28.6%, NPV was 94.2%, and total accuracy was 73.8%.

CONCLUSION:

Early detection and prompt management are crucial for improving outcomes in pediatric sepsis. Biomarkers such as CRP and PCT have shown promise for early diagnosis and monitoring treatment response. Combination and serial measurements may be more effective than single measurements in predicting sepsis and monitoring treatment effectiveness. Further research is needed to fully understand the role of these biomarkers in precision medicine for sepsis management.

REFERENCES:

1.Gilholm P, Gibbons, K, Lister, P, Harley, A, Irwin, A, Raman, S, et al. (2023): Validation of a paediatric sepsis screening tool to identify children with sepsis in the emergency department: a statewide prospective cohort study in Queensland, Australia. BMJ open, 13(1):e061431.

2. Oruganti S, Evans, J, Cromarty, T, Javaid, A & Roland, D (2022): Identification of sepsis in paediatric emergency departments: A scoping review. Acta Paediatrica, 111(12):2262-2277.

3. Pierrakos C, Velissaris, D, Bisdorff, M, Marshall, JC & Vincent, J-L (2020): Biomarkers of sepsis: time for a reappraisal. Critical care medicine, 24(1):1-15.

4. El Shabrawy RM, Gawish, A, Elgabry, R, Nasr, FM, Diab, M & Gamal, D (2021): Presepsin, procalcitonin and C reactive protein as diagnostic biomarkers of sepsis in intensive care unit patients. Microbes Infectious Diseases, 2(1):119-129.

5. Atreya MR & Wong, HR (2019): Precision medicine in pediatric sepsis. Current opinion in pediatrics,

31(3):322-335.

6. Centre for Disease Research and Control (2015): CDC.

7. Humoodi MO, Aldabbagh, MA, Salem, MM, Al Talhi, YM, Osman, SM, Bakhsh, M, et al. (2021): Epidemiology of pediatric sepsis in the pediatric intensive care unit of king Abdulaziz Medical City, Jeddah, Saudi Arabia. BMC pediatrics, 21(1):222-229.

8. Hartman ME, Linde-Zwirble, WT, Angus, DC & Watson, RS (2013): Trends in the epidemiology of pediatric severe sepsis. Pediatric critical care medicine: a journal of the Society of Critical Care Medicine, 14(7):686-693.

9. Sakyi SA, Enimil, A, Adu, DK, Ephraim, RD, Danquah, KO, Fondjo, L, et al. (2020): Individual and combined bioscore model of presepsin, procalcitonin, and high sensitive C-reactive protein as biomarkers for early diagnosis of paediatric sepsis. Heliyon, 6(9):e04841.

10. Mcwilliam S & Riordan, A (2010): How to use: C-reactive protein. Archives of Disease in Childhood-Education Practice, 95(2):55-58.

11. Downes KJ, Fitzgerald, JC & Weiss, SL (2020): Utility of procalcitonin as a biomarker for sepsis in children. Journal of Clinical Microbiology, 58(7):e01851-19

12. Fioretto JR, Martin, JG, Kurokawa, CS, Carpi, MF, Bonatto, RC, De Moraes, MA, et al. (2010): Comparison between procalcitonin and C-reactive protein for early diagnosis of children with sepsis or septic shock. Inflammation research, 59(2):581-586.

13. Schuetz P, Albrich, W & Mueller, B (2011): Procalcitonin for diagnosis of infection and guide to antibiotic decisions: past, present and future. BMC medicine, 9(1):1-9.

14. Nellis ME, Pon, S, Giambrone, AE, Coleman, NE, Reiss, J, Mauer, E, et al. (2016): The diagnostic accuracy of serum procalcitonin for bacteremia in critically ill children. Infectious diseases in clinical practice, 24(6):343-348.

Table (1): Baseline Characteristics of the studied groups

	Control (n = 20)	**Sepsis (n = 20)**	**P-value**
Age/months			
• **Mean SD**	41.93 ± 35.5	28.36 ± 28.7	**= 0.041***
• **Median (Range)**	27 (3 days-10 years)	3.8 (7 days-10 years)	
Sex			
• **Female**	9 (45%)	12 (60%)	= 0.342**
• **Male**	11 (55%)	8 (40%)	
ICU Stay/days			
• **Mean SD**	3.40 ± 1.7	7.80 ± 2.1	**< 0.001***
• **Median (Range)**	4 (2 - 8)	8 (4 - 11)	
Mortality			
• No	20 (100%)	17 (85%)	**0.036*****
• Yes	0 (0%)	3 (15%)	
Convulsion	1 (4%)	5 (20%)	0.171***
Body Temperature			
• **Mean SD**	37.30 ± 0.3	38.49 ± 0.5	**< 0.001***
• **Median (Range)**	37 (36.9 - 38)	38.5 (38 - 40)	
Organomegaly	0 (0%)	2 (10%)	0.244***
RR (cycle/min.)			
• **Mean SD**	42.60 ± 12.8	66.50 ± 12.3	**< 0.001***
• **Median (Range)**	42.5 (20 - 63)	68 (47 - 86)	
HR (beat/min.)			
• **Mean SD**	91.70 ± 15.5	121.40 ± 19.1	**= 0.003***
• **Median (Range)**	90 (69 - 115)	140.5 (70 - 170)	

***Mann Whitney U-test was employed to compare the median differences across groups**

****Chi-square test was employed to compare the frequency differences across groups**

****Fisher's exact test was employed to compare the frequency differences across groups**

Table (2): Differences in the CRP of the studied groups

	Control (n = 20)	Sepsis (n = 20)	P-value
CRP			
• 1st day	8.12 ± 0.9	33.65 ± 6.6	**< 0.001***
• 4th day	7.03 ± 0.8	37.50 ± 10.9	**= 0.012***
• 7th day	5.71 ± 0.7	34.30 ± 12.3	**= 0.026***
P-value**	**< 0.001**	**= 0.002**	

Table (3): Diagnostic criteria of CRP for Sepsis Prediction

Diagnostic criteria	CRP-1	CRP-4	CRP-7
• AUC *	0.973	0.938	0.874
• 95% CI**	0.933 - 1.000	0.860 - 1.000	0.767 - 0.980
• SE***	0.020	0.039	0.054
• P-value****	< 0.001	< 0.001	< 0.001
• Cut-off	12 mg/dL		
• Accuracy	90%	85%	77.5%
• Sensitivity%	95%	90%	80%
• Specificity%	85%	80%	75%
• PPV%	86%	82%	76%
• NPV%	94%	89%	79%
• Youden's J	0.8	0.7	0.55

***AUC=Area under the Curve**

****CI=Confidence Interval**

*****SE=Standard Error CI=Confidence Interval**

******Null hypothesis: true area=0.5**

----Sensitivity (true positives/all diseased); specificity (true negatives/all non-diseased);

PPV (true positives/all test positives); NPV (true negatives/all test negatives).

Table (4): Differences in the PCT of the studied groups

	Control (n = 20)	Sepsis (n = 20)	P-value
PCT			
• 1st day	0.14 ± 0.03	4.13 ± 0.7	**< 0.001***
• 4th day	0.11 ± 0.02	2.61 ± 0.9	**= 0.019***
• 7th day	0.07 ± 0.02	2.39 ± 0.3	**= 0.024***
• **P-value****	= 0.195	**= 0.007**	

***Independent t-test was used to compare the mean differences between groups**

****Repeated Measure One-way ANOVA test was used to compare the mean within group**

Table (5): Diagnostic criteria of PCT for Sepsis Prediction

Diagnostic criteria	PCT-1	PCT-4	PCT-7
• **AUC ***	**1.000**	**0.956**	**0.901**
• **95% CI****	**1.000 - 1.000**	**0.901 - 1.000**	**0.795 - 1.000**
• **SE*****	**0.000**	**0.028**	**0.054**
• **P-value********	**< 0.001**	**< 0.001**	**< 0.001**
• **Cut-off**	**0.58 ng/ml**	**0.33 ng/ml**	**0.11 ng/ml**
• **Accuracy**	**100%**	**90%**	**87.5%**
• **Sensitivity%**	**100%**	**85%**	**85%**
• **Specificity%**	**100%**	**95%**	**90%**
• **PPV%**	**100%**	**94.5%**	**89.5%**
• **NPV%**	**100%**	**86%**	**86%**
• **Youden's J**	**1**	**0.8**	**0.75**

***AUC=Area under the Curve**

****CI=Confidence Interval**

*****SE=Standard Error**

******Null hypothesis: true area=0.5**

----Sensitivity (true positives/all diseased); specificity (true negatives/all non-diseased);

PPV (true positives/all test positives); NPV (true negatives/all test negatives).

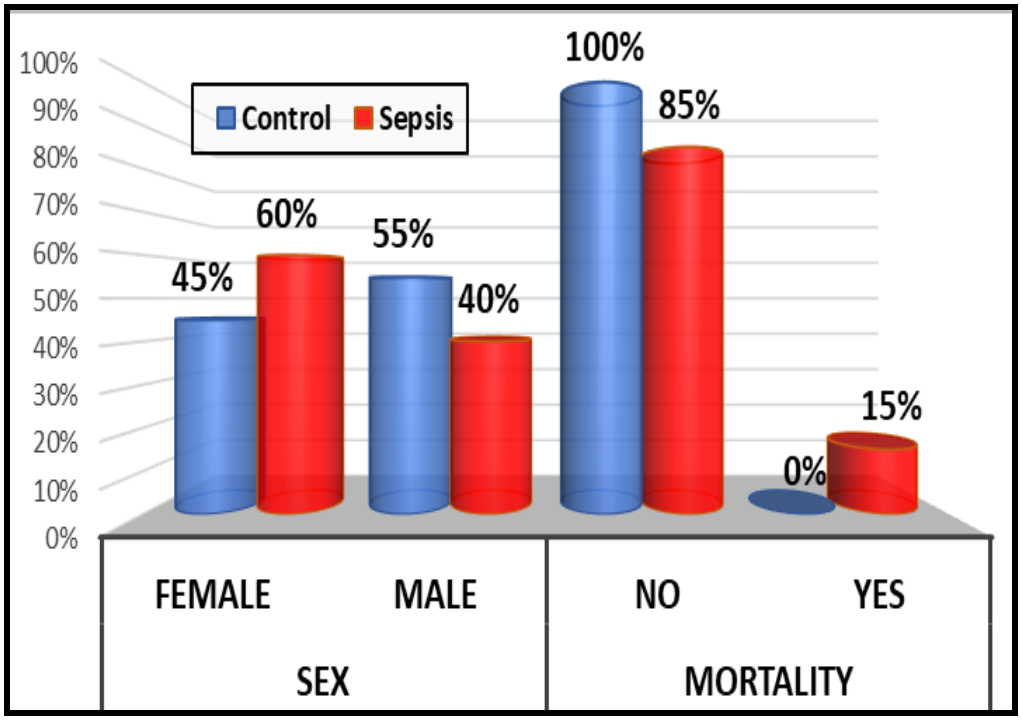

Fig. (1): Sex/Mortality Distribution of the studied Groups

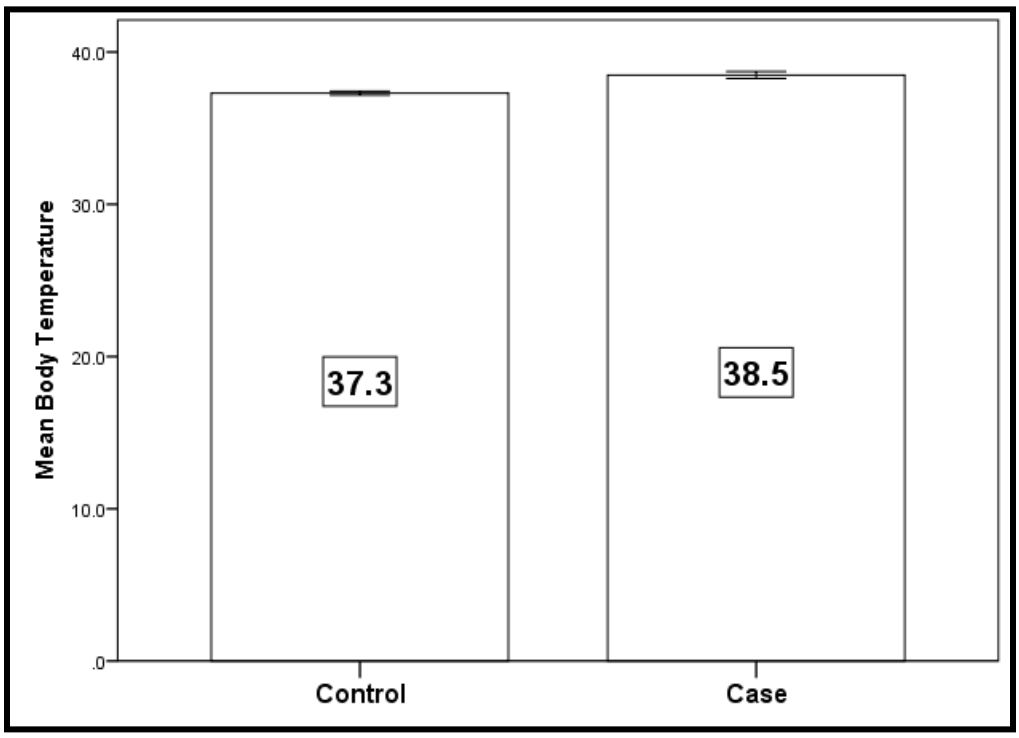

Fig. (2): Difference in the Mean Body Temperature between the studied Groups

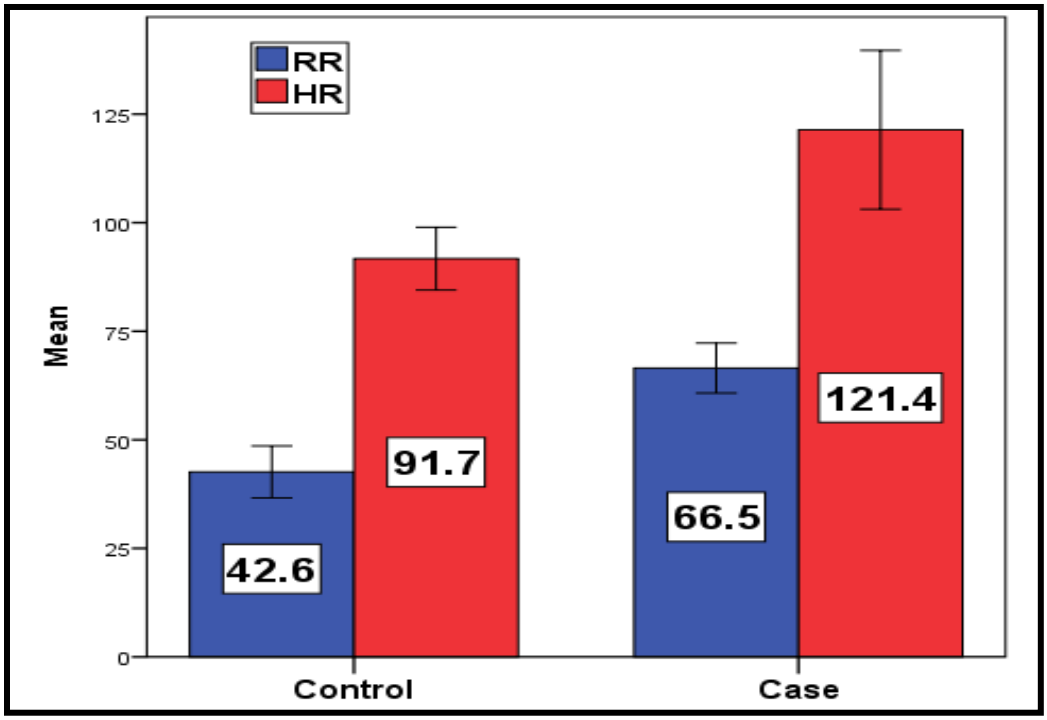

Fig. (3): Difference in the Mean RR/HR between the studied Groups

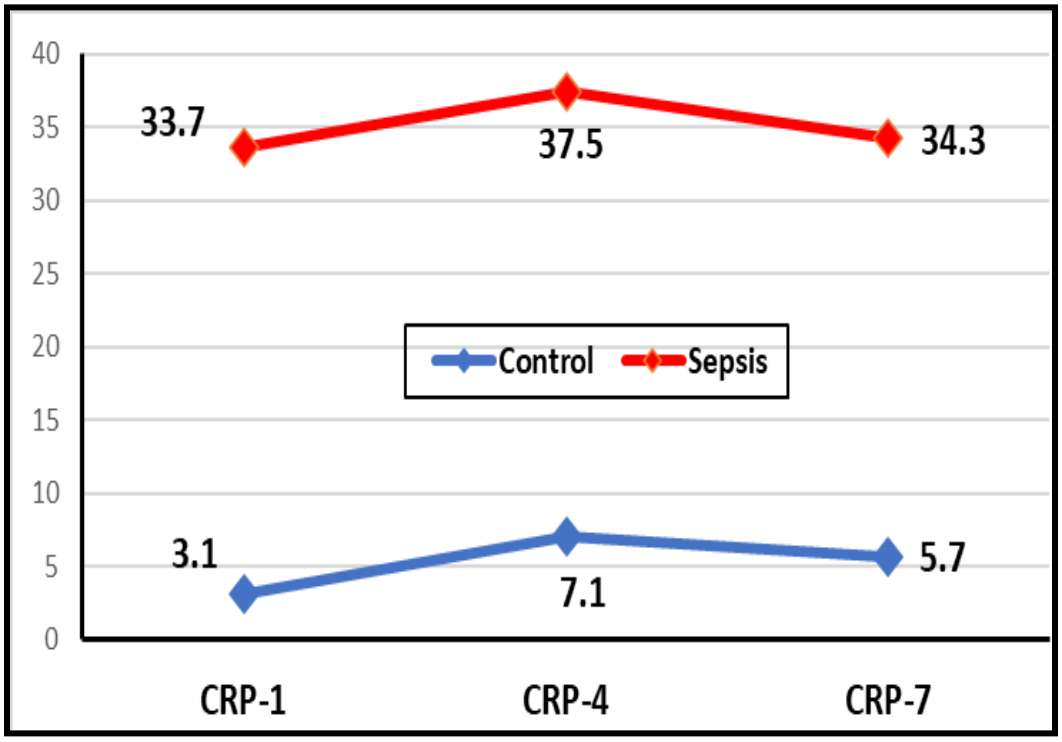

Fig. (4): Mean CRP level Difference between Groups over Time

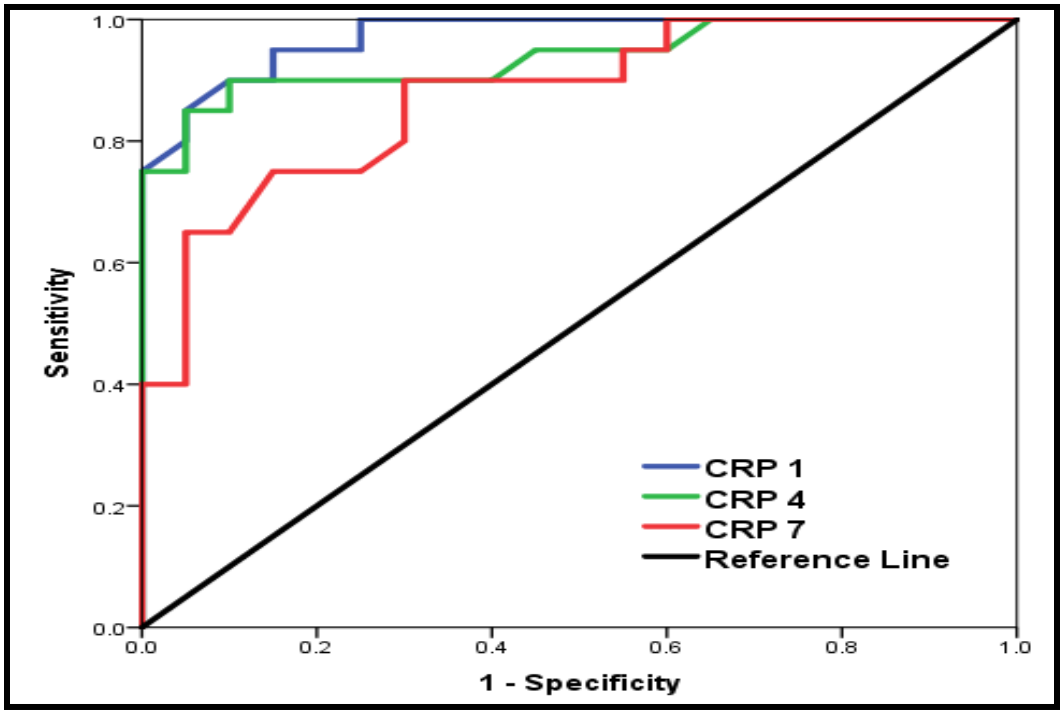

Fig. (5): ROC curve for CRP for Sepsis Prediction

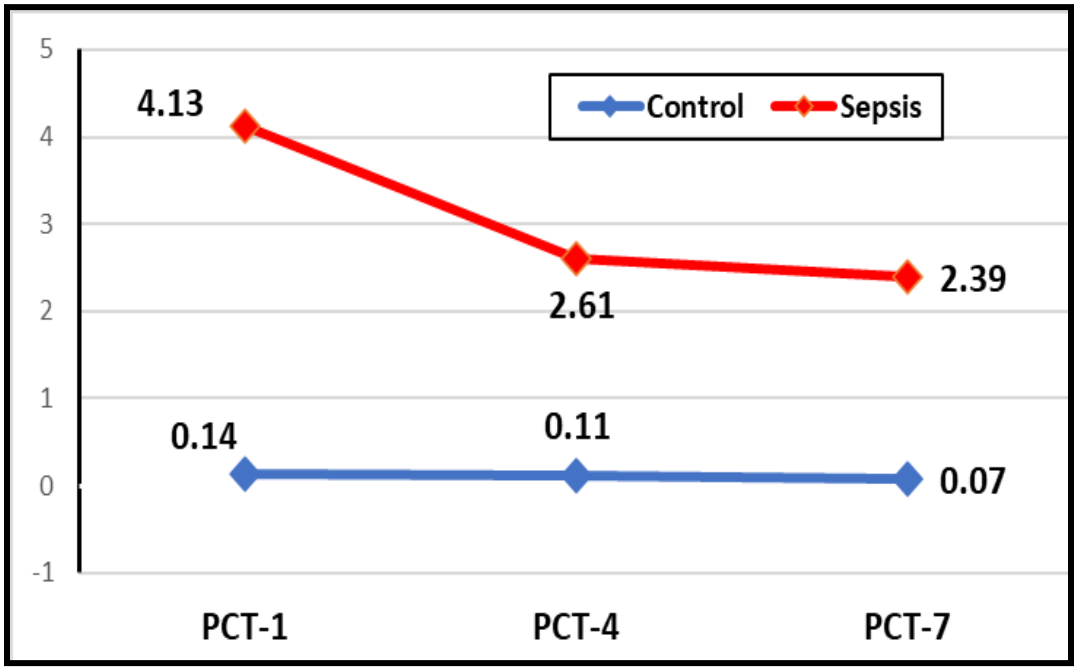

Fig.(6): Mean PCT level Difference between Groups over Time

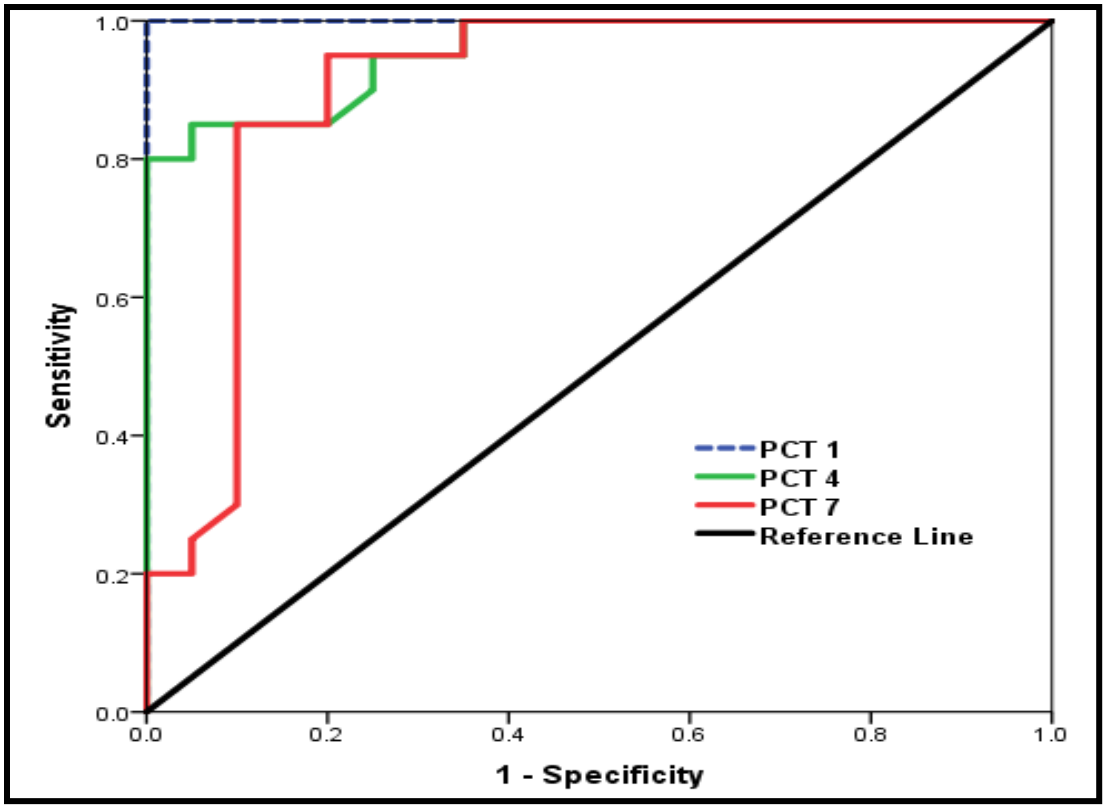

Fig. (7): ROC curve for PCT for Sepsis Prediction