ORIGINAL ARTICLE

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Validation and comparison of the PECARN rule, Step-by-Step approach and Lab-score for predicting serious and invasive bacterial infections in young febrile infants

Natalia Sutiman [1]*MD*, Zi Xean Khoo [2]*MRCPCH*, Gene Yong-Kwang Ong [1]*MRCPCH*, Rupini Piragasam [3], Shu-Ling Chong [1,4] *MRCPCH*

ABSTRACT

Introduction: Differentiating infants with serious bacterial infections (SBIs) or invasive bacterial infections (IBIs) from those without remains a challenge. We sought to compare the diagnostic performances of single biomarkers (absolute neutrophil count [ANC], C-reactive protein [CRP] and procalcitonin [PCT]) and 4 diagnostic approaches comprising Lab-score, Step-by-Step approach (original and modified) and Pediatric Emergency Care Applied Research Network (PECARN) rule.

Method: This is a prospective cohort study involving infants 0–90 days of age who presented to an emergency department from July 2020 to August 2021. SBIs were defined as bacterial meningitis, bacteraemia and/or urinary tract infections. IBIs were defined as bacteraemia and/or bacterial meningitis. We evaluated the performances of Lab-score, Step-by-Step (original and modified) and PECARN rule in predicting SBIs and IBIs.

Results: We analysed a total of 258 infants, among whom 86 (33.3%) had SBIs and 9 (3.5%) had IBIs. In predicting SBIs, ANC ≥4.09 had the highest sensitivity and negative predictive value (NPV), while PCT ≥1.7 had the highest specificity and positive predictive value (PPV). CRP ≥20 achieved the highest area under receiver operating characteristic curve (AUC) of 0.741 (95% confidence interval [CI] 0.672–0.810). The Step-by-Step (original) approach had the highest sensitivity (97.7%). Lab-score had the highest AUC of 0.695 (95% CI 0.621–0.768), compared to PECARN rule at 0.625 (95% CI 0.556–0.694) and Step-by-Step (original) at 0.573 (95% CI 0.502–0.644). In predicting IBIs, PCT ≥1.7 had the highest sensitivity, specificity, PPV and NPV. The Step-by-Step (original and modified) approach had the highest sensitivity of 100%. Lab-score had the highest AUC of 0.854 (95% CI 0.731–0.977) compared to PECARN rule at 0.589 (95% CI 0.420–0.758) and Step-by-Step at 0.562 (95% CI 0.392–0.732).

Conclusion: CRP strongly predicted SBIs, and PCT strongly predicted IBI. The Step-by-Step approach had the highest sensitivity and NPV, while Lab-score had the highest specificity and AUC in predicting SBIs and IBIs.

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Keywords: Biomarkers, diagnostic approaches, febrile infants, Lab-score, PECARN rule, Step-by-Step approach

INTRODUCTION

The diagnostic approach and management of febrile infants <90 days of age remain a challenge, given that the majority of these infants have no localising signs and symptoms, and may appear clinically well at presentation.[1] In addition, the majority of these infants have benign viral illnesses, for which hospitalisation and antibiotics may not be warranted. Identifying infants with serious bacterial infections (SBIs) or invasive bacterial infections (IBIs) based on clinical assessment alone may lead to delayed or missed diagnosis.[2]

Previous diagnostic approaches include the Rochester criteria, which take into account clinical findings, white blood cell (WBC) and urinalysis,[3] as well as the

[1] Department of Emergency Medicine, KK Women's and Children's Hospital, Singapore
[2] Department of Pediatric Medicine, KK Women's and Children's Hospital, Singapore
[3] KK Research Centre, KK Women's and Children's Hospital, Singapore
[4] Duke-NUS Medical School, Singapore
Correspondence: Dr Shu-Ling Chong, Children's Emergency Department, KK Women's and Children's Hospital, 100 Bukit Timah Rd, Singapore 229899.
Email: Chong.Shu-Ling@kkh.com.sg

CLINICAL IMPACT

What is New

- This study compared the discriminative ability of single biomarkers (absolute neutrophil count, C-reactive protein and procalcitonin) and predictive tools comprising Lab-score, Step-by-Step approach (original and modified) and Pediatric Emergency Care Applied Research Network (PECARN) rule in differentiating infants with serious bacterial infections (SBIs) and invasive bacterial infections.
- Step-by-Step (original) approach had the highest sensitivity and negative predictive value, resulting in the fewest missed cases of SBIs.

Clinical Implications

- Future research should study the application of a modified Step-by-Step algorithm as a safe tool for use in Singapore.

Philadelphia criteria, which incorporate clinical criteria, WBC, urinalysis, chest X-ray and lumbar puncture for cerebrospinal fluid (CSF) analysis.[3,4] A retrospective cohort study on infants in Singapore showed that the Rochester criteria performed with a sensitivity of 96% but had a low specificity of 15.7%, and classified up to 88% of febrile infants as high risk.[5] Newer diagnostic algorithms and clinical prediction rules have since been developed to incorporate newer biomarkers such as C-reactive protein (CRP) and procalcitonin (PCT).[6] These include Lab-score,[7] Step-by-Step approach[8] and Pediatric Emergency Care Applied Research Network (PECARN) rule,[9] which have proven to be more accurate in predicting SBIs and IBIs than single biomarkers.

The first of these prediction models, Lab-score, was developed and validated in 2008 to identify febrile infants at risk of SBIs. Lab-score is based on 3 predictive variables, which have been independently associated with SBIs, namely CRP, PCT and urinary dipstick results.[10] The Step-by-Step approach was subsequently developed as an algorithm that sequentially evaluates infants' general appearance, age, urinalysis, and markers—including absolute neutrophil count (ANC), CRP and PCT—in predicting IBIs.[11] Kuppermann et al. developed and internally validated a promising prediction tool, the PECARN rule, which takes into consideration urinalysis, ANC and PCT, to aid the identification of SBIs among febrile infants.[9]

However, the diagnostic performances of these prediction rules in identifying SBIs and IBIs in our population remain unknown. Therefore, we seek to evaluate and compare the diagnostic performances of single biomarkers, Lab-score, Step-by-Step approach and PECARN rule in predicting SBIs and IBIs in febrile infants.

METHOD

Study design

This prospective observational study was conducted between July 2020 and August 2021 at KK Women's and Children's Hospital in Singapore, a tertiary paediatric hospital where the pediatric emergency department (ED) is visited by approximately 150,000 children aged 0–17 years annually.

Patient population

In our institution, all febrile infants aged 0–90 days are admitted for investigations and further inpatient management. Patients were recruited based on the following inclusion criteria: 0–90 days of age (actual age), and fever, defined as axillary or rectal temperature of at least 38°C either in the ED or prior healthcare settings within the preceding 24 hours. We excluded the following patients: pre-term infants aged <35 weeks; those with significant neonatal complications requiring prolonged perinatal hospitalisation of >7 days; those receiving prior intravenous antibiotics within 48 hours of presentation; and those with underlying genetic, chromosomal, immunological and haematological diseases. We excluded these infants because they innately formed a higher-risk population that would receive closer attention. We also excluded those with incomplete medical records or biochemistry results that would preclude the validation of the prediction rules.

Clinical and biochemical evaluation

Demographic data, clinical history and physical examination findings were recorded for all enrolled patients. We defined infants who were not well appearing as those with ≥1 abnormal component of the paediatric assessment triangle (appearance, work of breathing and circulation to the skin). In our institution, while haemodynamically unstable febrile infants are resuscitated in the ED, stable febrile infants receive their investigations after admission to the hospital. A comprehensive workup including blood, urine and cerebrospinal fluid (CSF) cultures is performed for all neonates 0–28 days old. Among infants 29–90 days old, the extent of investigations is decided at the discretion

of the treating physicians based on clinical assessment. Infants who are subsequently afebrile in the wards and examine well may not undergo all investigations. Therefore, we excluded those without the relevant blood test results since we could not analyse them. Blood samples were taken for WBC, CRP and PCT, and urine specimens were collected for urinalysis. A positive urinalysis was defined as positive for leucocyte esterase, nitrite or pyuria at >5 WBCs.

Classification, definitions and outcome measures

The definitions used to classify patients in the SBI group were: (1) bacterial meningitis, defined as growth of a single bacterial pathogen in the CSF (with culture-proven diagnosis for both bacteraemia and urinary tract infections [UTIs]); (2) bacteraemia, defined as growth of a single bacterial pathogen in blood (excluding growth of bacteria considered a priori as contaminants, e.g. coagulase-negative *Staphylococcus*); and/or (3) UTIs.[9] UTIs were defined by growth of single urine pathogen with at least 50,000 colony forming units (CFU)/mL from catheterised urine specimen; or 10,000–50,000CFU/mL from a catheterised specimen with positive urinalysis (positive for leucocyte esterase, nitrite or pyuria at >5 WBCs; or at least 100,000CFU/mL from urine collected via voided specimens).[9] All other patients were classified into the non-SBI group. IBI was defined by bacteraemia and/or bacterial meningitis.

Clinical prediction tools

Determination of Lab-score was based on the original study (Fig. 1A). Briefly, 1 point was attributed to positive urine dipstick; 2 points for PCT ≥0.5ng/mL or CRP ≥40mg/L; and 4 points to procalcitonin ≥2ng/mL or CRP ≥100mg/L. Lab-score ≥3 was deemed high risk. Step-by-Step approach was applied according to the original study (Fig. 1B). Additionally, in our institution, the Step-by-Step approach was modified from the original algorithm (Fig. 1C), and this modified approach is also applied in our study. Based on PECARN rule, febrile infants were classified as low-risk following negative urinalysis, ANC <4,090/mm^3 and PCT <1.7ng/mL (Fig. 1D). We note that the Step-by-Step approach was originally formulated to predict IBIs. However, we chose to study its validity for both SBIs and IBIs in order to conduct a comprehensive investigation on the performance of Step-by-Step, Lab-score and PECARN rule. Since the validation of Lab-score and its publication, multiple studies have evaluated the cutoffs at which each biomarker show the best discriminative ability.[11-15] We chose to follow the established thresholds in Step-by-Step and PECARN rule of CRP >20mg/L and PCT >0.5ng/mL and 1.ng/mL as cutoffs as these are more recently developed than Lab-score.

Statistical analysis

Statistical analyses were performed using SPSS Statistics version 22 (IBM Corp, Armonk, US). Normally distributed continuous variables were expressed as mean ± standard deviation. Non-normally distributed data were expressed as median and interquartile range (IQR). Comparisons between groups were performed using 2-sample t-tests for normally distributed data and Mann-Whitney U tests for non-normal data. Categorical variables were expressed as percentages and were compared using Fisher's Exact tests. We presented the univariate and multivariable regression for clinical and biochemical predictors of SBIs and IBIs. In the multivariable regression, we included covariates based on clinical rationale and statistical significance. In the multivariable regression, biomarkers were treated as continuous variables.

Diagnostic performance measures including area under the receiver operating characteristic curve (AUC), sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), positive likelihood ratio (LR) and negative LR were calculated. Point estimates for AUC analyses were presented with a 95% confidence interval (CI). For analysis on diagnostic performance, all biomarkers were categorised using cutoffs derived from the literature (see section on Clinical prediction tools). Statistical significance was defined as P value <0.05.

The study protocol was approved by the ethics review committee of KK Women's and Children's Hospital (IRB: 2017/2680).

RESULTS

Patient population and demographics

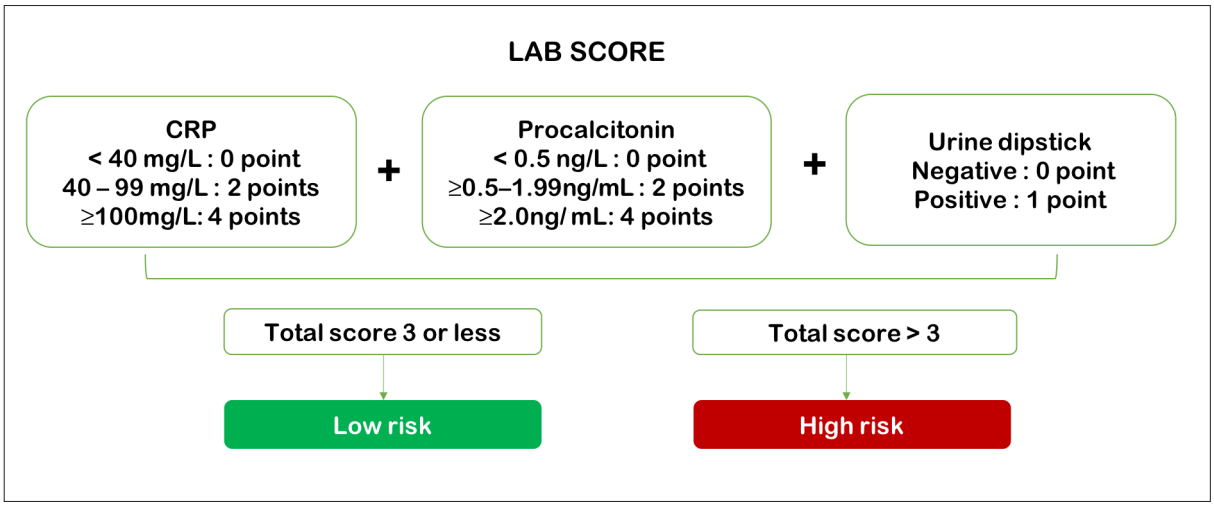
Among 468 febrile infants, we analysed 258 infants who met the inclusion criteria (Fig. 2). The overall median age was 47 days (IQR 14–64 days) and 100 patients (38.8%) were neonates. The median temperature at triage was 38.7°C (IQR 38.2–39.0°C). Sixty-one patients (29.3%) were born to mothers with group B *Streptococcus*. Eighty-six patients (33.3%) had SBIs and nine patients (3.5%) had IBIs. Table 1 shows the demographic and clinical characteristics of the study population in SBI, non-SBI, IBI and non-IBI groups.

Clinical characteristics and causative organisms

Among the 258 analysed infants, 77 (29.8%) had UTIs, 3 (1.2%) had bacteraemia and 1 (0.4%) had bacterial

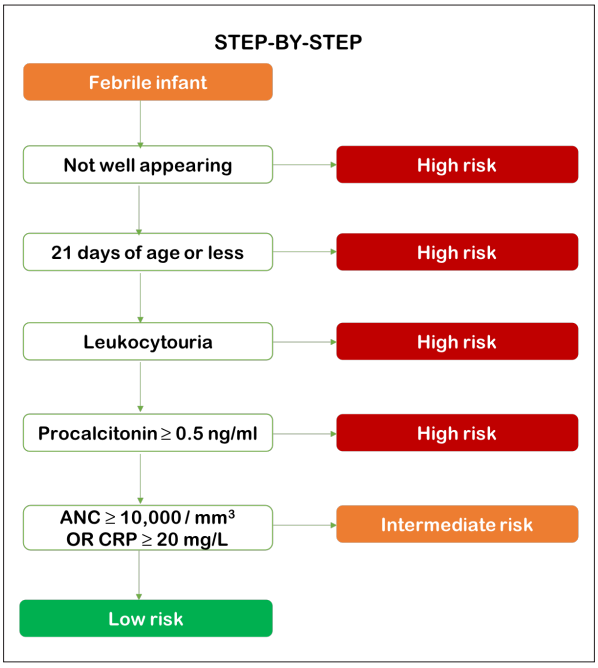
Fig. 1. Risk stratification of febrile infants according to (A) Lab-score, (B) Step-by-Step (original) approach, (C) Step-by-Step (modified) approach, and (D) Pediatric Emergency Care Applied Research Network (PECARN) rule.

(A)

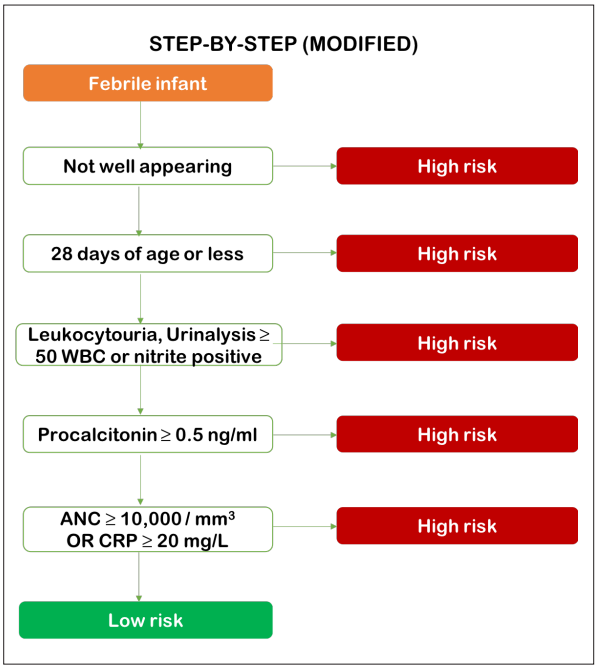


CRP: C-reactive protein
Adapted from Galetto-Lacour A, Zamora SA, Andreola B, et al. Validation of a laboratory risk index score for the identification of severe bacterial infection in children with fever without source. Arch Dis Child 2010;95:968-73.

(B)



(C)

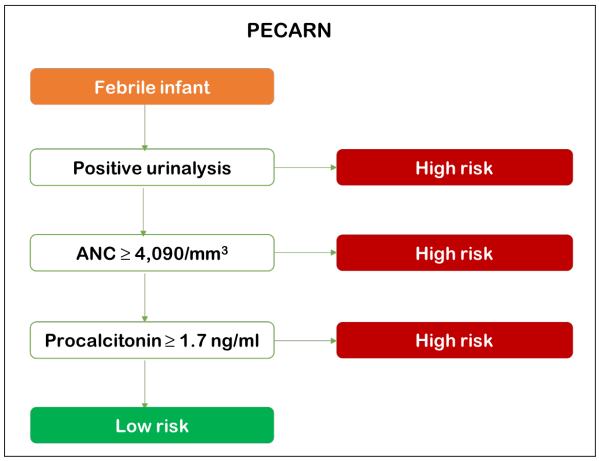


ANC: absolute neutrophil count; CRP: C-reactive protein; WBC: white blood cell

Adapted from Gomez B, Mintegi S, Bressan S, et al. Validation of the "Step-by-Step" approach in the management of young febrile infants. Pediatrics 2016;138:e20154381.

meningitis. In addition, 4 (1.6%) had concomitant UTI with bacteraemia and 1 (0.4%) had meningitis with bacteraemia (Table 2). The CSF of the child with meningitis grew group B *Streptococcus*. The CSF of the child with both bacteraemia and meningitis also grew group B *Streptococcus*. *Escherichia coli* was the most common pathogen causing both bacteraemia (2, 28.6%) and UTI (53, 65.4%).

(D)



ANC: absolute neutrophil count; PECARN: Pediatric Emergency Care Applied Research Network
Adapted from Kuppermann N, Dayan PS, Levine DA, et al. A Clinical Prediction Rule to Identify Febrile Infants 60 Days and Younger at Low Risk for Serious Bacterial Infections. JAMA Pediatr 2019;173:342-51.

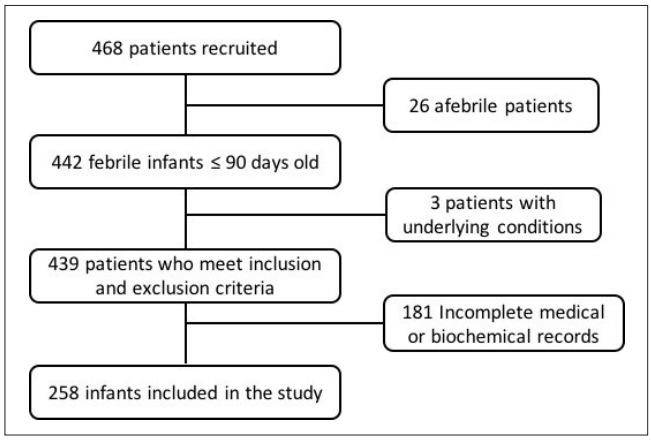


Fig. 2. Patient flow diagram.

Univariate and multivariate analyses for predictors of SBIs and IBIs

Median values for WBC, ANC, CRP and PCT were significantly higher in infants with SBI versus without SBI: WBC (15.8 vs 12.6, P<0.001), ANC (8.14 vs 5.38, P<0.001), CRP (37.4 vs 11.1, P<0.001), and PCT (0.46 vs 0.23; P<0.001). Only PCT was significantly higher in infants with IBI vs without IBI (9.27 vs 0.27, P<0.001) (Table 1).

In the univariate analysis, age, sex, temperature, positive urinalysis, WBC, ANC and CRP were significantly associated with SBIs (online Supplementary Materials, Supplementary Table S1). In the multivariable analysis, only temperature at triage and positive urinalysis were significantly associated with SBIs (online Supplementary Table S1). Male sex, temperature at triage, duration of fever, ANC, CRP and PCT were significantly associated with IBIs (online Supplementary Table S2). Only WBC and ANC were significantly associated with IBIs in the multivariate analysis (online Supplementary Table S2).

Diagnostic performance of single biomarkers, PECARN rule, Step-by-Step approach and Lab-score in predicting SBIs

We compared the diagnostic performances of single biomarkers ANC, CRP and PCT with cutoff levels at recommended thresholds as well as the 4 approaches. Notably, among the 3 single biomarkers, ANC ≥4,090/mm^3 had the highest sensitivity (80.2%) and NPV (80.2%) in predicting SBIs. PCT ≥1.7ng/mL had the highest specificity (95.9%) and PPV (77.4%) in predicting SBIs (Table 3). Among the 4 diagnostic approaches, the Step-by-Step (original) approach had the highest sensitivity in predicting SBIs (97.7%), which is notably higher than the sensitivity achieved with single biomarkers (Table 3). The Step-by-Step (original) approach had the highest NPV of 93.5% compared to PECARN rule and Lab-score (NPV of 86.3% and 77.6%, respectively).

CRP ≥20mg/L achieved the highest AUC of 0.741 (95% CI 0.672–0.810), higher than the AUCs achieved by the 4 approaches. Among the 4 approaches, Lab-score was found to have the highest AUC of 0.695 (95% CI 0.621–0.768), compared to PECARN rule with AUC of 0.625 (95% CI 0.556–0.694) and Step-by-Step with AUC of 0.573 (95% CI 0.502–0.644).

Diagnostic performance of single biomarkers, PECARN rule, Step-by-Step approach and Lab-score in predicting IBIs

In predicting IBIs, PCT ≥1.7ng/mL had the highest sensitivity, specificity, PPV and NPV among the single biomarkers. Additionally, it also achieved the highest AUC of 0.898 (95% CI 0.778–1.00), higher than those achieved by the 4 approaches (Table 4). Among the 4 approaches, Step-by-Step approach (both original and modified) had the highest sensitivity for predicting IBIs (sensitivity of 100%) whereas the PECARN rule and Lab-score both achieved a sensitivity of 88.9% (Table 4). Step-by-Step approach also had the highest NPV of 100% compared to the PECARN rule and Lab-score (Table 4). Lab-score had the highest specificity of 81.9%, compared to PECARN rule and Step-by-Step (original) approach (28.9% and 12.4%) (Table 4). Among the 4 approaches, the Lab-score had the highest AUC of 0.854 (95% CI 0.731–0.977) compared to PECARN rule with AUC of 0.589 (95% CI 0.420–0.758) and Step-by-Step (original) with AUC of 0.562 (95% CI 0.392–0.732).

Table 1. Patient characteristics and univariable analysis

Characteristics	Overall (n=258)	SBI (n=86)	Non-SBI (n=172)	*P* value	IBI (n=9)	Non-IBI (n=249)	*P* value
Age, median (IQR), days	47 (14–64)	55 (28–69)	36 (11–62)	**0.005**	31 (26–45)	51 (14–65)	0.590
Age at presentation, no. (%)				**0.041**			0.432
0–28 days	100 (38.8)	24 (9.3)	76 (29.5)		4 (1.6)	96 (37.2)	
29–60 days	81 (31.4)	32 (12.4)	49 (19.0)		4 (1.6)	77 (29.8)	
61–90 days	77 (29.8)	30 (11.6)	47 (18.2)		1 (0.4)	76 (30.5)	
Sex, no. (%)				**0.002**			0.582
Male	149 (57.8)	61 (23.6)	88 (34.1)		6 (2.3)	143 (55.4)	
Female	109 (42.2)	25 (9.7)	84 (32.6)		3 (1.2)	106 (41.1)	
Prematurity, no. (%)				**0.044**			0.482
No	245 (95.0)	85 (32.9)	160 (62.0)		9 (3.5)	236 (91.5)	
Yes	13 (5.0)	1 (0.4)	12 (4.7)		0	13 (5.0)	
Maternal GBS, no. (%)				0.622			0.190
No	147 (70.7)	51 (24.5)	96 (46.2)		4 (1.9)	143 (68.8)	
Yes	61 (29.3)	19 (9.1)	42 (20.2)		4 (1.9)	57 (27.4)	
Comorbidities, no. (%)				0.210			0.500
No	246 (95.3)	80 (31.0)	166 (64.3)		9 (3.5)	237 (91.9)	
Yes	12 (4.7)	6 (2.3)	6 (2.3)		0	12 (4.7)	
Well appearing, no. (%)							
No	17 (6.6)	6 (5.7)	11 (11.3)	0.859	2 (3.3)	15 (16.4)	0.543
Yes	241 (93.4)	80 (80.3)	161 (160.7)		7 (8.4)	234 (232.6)	
Temperature at triage, median (IQR), °C	38.7 (38.2–39)	38.9 (38.4–39.3)	38.5 (38.1–38.7)	**<0.001**	39.2 (38.7–38.9)	38.7 (38.2–39)	**0.002**
Duration of fever, mean (IQR), days	1 (0–4)	1 (0–4)	1 (1–2)	**0.034**	1 (1–4)	1 (0–3)	**0.024**
Positive urinalysis, no. (%)				**<0.001**			0.119
No	176 (68.2)	27 (10.5)	149 (57.8)		4 (1.6)	172 (66.7)	
Yes	82 (31.8)	59 (22.9)	23 (8.9)		5 (1.9)	77 (29.8)	
White blood cell count, median (IQR), $\times 10^9$/L	14.3 (3–31)	15.8 (3–31)	12.6 (4–30)	**<0.001**	11.7 (3–28)	14.3 (4–31)	0.796
Absolute neutrophil count, median (IQR), mg/L	6.8 (1–20)	8.14 (1–20)	5.38 (2–17)	**<0.001**	7.88 (1–20)	13.5 (2–20)	0.241
C-reactive protein, median (IQR), mg/L	21.1 (1–245)	37.4 (2–245)	11.1 (1–178)	**<0.001**	59.9 (3–194)	20.2 (1–245)	0.069
Procalcitonin, median (IQR), ng/mL	0.29 (0–121)	0.46 (0–86)	0.23 (0–121)	**<0.001**	9.27 (0–82)	0.27 (0–121)	**<0.001**

GBS: group B *Streptococcus;* IBI: invasive bacterial infection; IQR: interquartile range; SBI: serious bacterial infection

Table 2. Bacterial infections by age groups

Bacterial infections, no. (%)	Overall (n=258)	0–28 days (n=100)	29–60 days (n= 81)	61–90 days (n=77)
UTI	77 (29.8)	20 (20.0)	28 (34.6)	29 (11.2)
Bacteraemia	3 (1.2)	1 (1.0)	1 (1.2)	1 (0.4)
Bacterial meningitis	1 (0.4)	0	1 (1.2)	0
Bacteraemia and UTI	4 (1.6)	3 (3.0)	1 (1.2)	0
Bacteraemia and meningitis	1 (0.4)	0	1 (1.2)	0

UTI: urinary tract infection

Table 3. Diagnostic performance of single biomarkers, Pediatric Emergency Care Applied Research Network (PECARN) rule, Step-by-Step (original) approach, Step-by-Step (modified) approach and Lab-score for predicting serious bacterial infections

	Sensitivity	NPV	Specificity	PPV	Positive LR	Negative LR	AUC (CI)	*P* value
Step-by-Step (original)	97.7%	93.5%	16.9%	37.0%	1.18	0.136	0.573 (0.502–0.644)	0.057
Step-by-Step (modified)	91.9%	90.4%	38.4%	42.7%	1.49	0.211	0.651 (0.584–0.718)	<0.001
PECARN rule	88.4%	86.3%	36.6%	41.1%	1.39	0.317	0.625 (0.556–0.694)	0.001
ANC ≥4.09	80.2%	80.2%	40.1%	40.1%	1.34	0.49	0.602 (0.531–0.673)	0.008
CRP ≥20	61.6%	81.9%	86.6%	69.7%	4.61	0.44	0.741 (0.672–0.810)	<0.001
Lab-score	46.5%	77.6%	92.4%	75.5%	6.12	0.579	0.695 (0.621–0.768)	<0.001
PCT ≥0.5	41.9%	74.5%	84.9%	58.1%	2.77	0.68	0.634 (0.559–0.709)	<0.001
ANC ≥10	27.9%	72.0%	92.4%	64.9%	3.69	0.78	0.602 (0.525–0.678)	0.008
PCT ≥1.7	27.9%	72.7%	95.9%	77.4%	6.86	0.75	0.619 (0.542–0.696)	0.002

ANC: absolute neutrophil count; AUC: area under receiver operating characteristic curve; CI: confidence interval; CRP: C-reactive protein; LR: likelihood ratio; NPV: negative predictive value; PCT: procalcitonin; PPV: positive predictive value

DISCUSSION

In this prospective study, we compared the diagnostic performances of single biomarkers ANC, CRP and PCT, as well as 4 approaches—PECARN rule, Step-by-Step approach (original and modified) and Lab-score—in identifying febrile infants at risk of SBIs and IBIs. Overall, there were 86 patients (33.3%) who had SBI and 9 patients (3.5%) who had IBI. In predicting for SBI, WBC, ANC and CRP were shown to be good discriminators, with CRP ≥20mg/L achieving the highest AUC of 0.741. In predicting IBIs, PCT was shown to be a good discriminator, with PCT ≥1.7ng/mL achieving the highest AUC of 0.898. For SBI, the Step-by-Step approach (original) had the highest sensitivity and NPV, while Lab-score reported the highest specificity, PPV and AUC. For IBIs, the Step-by-Step (original) approach had the highest sensitivity and NPV, whereas the Lab-score reported the highest specificity, PPV and AUC.

Notably, the SBI rate in this study was higher compared with those reported in other centres.[10,16,17] In Singapore, KKH is one of 2 tertiary centres that receive referrals from primary care, which may account for the higher rates of disease, since otherwise well infants may be managed at the primary care level. UTIs account for the majority of the SBIs in this study, with *Escherichia coli* and *Klebsiella spp.* being the most common organisms, accounting for respectively 54 (66.7%) and 14 (17.3%) of UTI cases. The rates of UTI remain high across all age groups, including older infants 29–60 days (Table 2), emphasising the need to evaluate for UTI among older infants. *E. coli* also accounted for the majority of the cases with bacteraemia. This is consistent with previously reported studies.[10,18,19]

We found that CRP and PCT as single biomarkers were strong predictors of SBIs and IBIs. In line with the findings of previous studies,[6,20,21] CRP ≥20mg/L performed best in predicting SBIs with an AUC of 0.741.

Table 4. Diagnostic performance of the single biomarkers, Pediatric Emergency Care Applied Research Network (PECARN), Step-by-Step (original) approach, Step-by-Step (modified) approach and Lab-score for predicting invasive bacterial infections

	Sensitivity	NPV	Specificity	PPV	Positive LR	Negative LR	AUC (CI) *P* value
Step-by-Step (original)	100%	100%	12.4%	4.0%	1.14	0	0.562 (0.392–0.732) 0.526
Step-by-Step (modified)	100%	100%	29.3%	4.9%	1.41	0	0.647 (0.507–0.787) 0.135
PECARN rule	88.9%	98.6%	28.9%	4.3%	1.25	0.384	0.589 (0.420–0.758) 0.364
Lab-score	88.9%	99.5%	81.9%	15.1%	4.91	0.136	0.854 (0.731–0.977) <0.001
PCT ≥0.5	88.9%	99.5%	78.3%	12.9%	4.10	0.14	0.836 (0.712–0.960) 0.001
PCT ≥1.7	88.9%	99.6%	90.8%	25.8%	9.62	0.12	0.898 (0.778–1.00) <0.001
ANC ≥4.09	66.7%	96.5%	33.3%	3.5%	1.00	1.00	0.500 (0.308–0.692) 0.098
ANC ≥10	44.4%	97.7%	86.8%	10.8%	3.35	0.64	0.656 (0.450–0.862) 0.105
CRP ≥20	66.7%	98.4%	71.9%	7.89%	2.37	0.46	0.693 (0.511–0.874) 0.049

ANC: absolute neutrophil count; AUC: area under receiver operating characteristic curve; CI: confidence interval; CRP: C-reactive protein; LR: likelihood ratio; NPV: negative predictive value; PCT: procalcitonin; PPV: positive predictive value

CRP is a commonly available biomarker, but peaks later in the course of illness, typically 4–6 hours. In contrast, PCT levels rise quickly in response to bacterial infections, typically within 2–4 hours, but may take up to 6–12 hours to peak.[22] CRP performed better than PCT in predicting for SBIs in our population. We postulate that this could be because the majority of these infants receive their workup after hospitalisation rather than on presentation to the ED, therefore providing an adequate window for CRP to rise.

In predicting IBIs, PCT of >1.7 had the highest AUC of 0.898, compared to a PCT of >0.5ng/mL with an AUC of 0.836. The diagnostic values of PCT in this study correspond to those previously reported,[6,16,23,24] with most studies utilising the same thresholds. Indeed, systematic reviews and recent studies have concluded that PCT was superior to CRP in identifying IBIs.[25,26] However, the PCT cutoff values have been debated and vary from 0.12 to 1.0ng/mL, as different studies tried to differentiate invasive and non-invasive bacterial infections. We found that a PCT cutoff of 1.7ng/mL yielded similar sensitivity and NPV to a cutoff of 0.5ng/mL, and performed with much higher specificity (90.8% compared to 78.3%, respectively).

The PECARN rule in the original study by Kuppermann et al. reported a sensitivity of 97.7%, specificity of 60.0% and NPV of 99.6% in identifying infants who are at risk of SBIs.[9] In comparison, its validation in our cohort showed a lower sensitivity of 88.4%, specificity of 36.6% and NPV of 86.3% in predicting SBIs. The decrease in performance could be attributed to differences in patient populations and delivery of health services. Singapore is geographically a small nation and young infants with fever tend to arrive very early in the course of illness. Our population consisted of 23 infants in whom urinalysis was negative but urine cultures were positive for urinary tract infections. These accounted for a number of false negatives when the above algorithm was applied.

Unlike in the original derivation and validation study, Lab-score showed much lower sensitivities for predicting SBIs and IBIs in our cohort (46.5% and 88.9%, respectively), compared to 94% in the original study. Lab-score, which takes into account urinalysis, also resulted in missed cases in our population whereby urinalysis was normal but urine cultures turned positive. Although the sensitivity was low, specificity for Lab-score was high at 92.4% and 81.9% for SBIs and IBIs,

respectively. This resulted in the AUCs for Lab-score being higher in predicting SBIs and IBIs, when compared to the other approaches.

Due to the gravity of missed diagnoses in this vulnerable population, frontline physicians should choose a diagnostic approach with high sensitivity and NPV. The original Step-by-Step algorithm by Gomez et al. demonstrated a sensitivity of 92% and NPV of 99.3% for IBIs in febrile infants.[11] Our study showed a similarly high sensitivity and NPV in predicting SBIs (97.7% and 93.5%, respectively) and IBIs (100% and 100%, respectively). Notably, of all the approaches, the Step-by-Step (original) approach showed the highest sensitivity and NPV in predicting both SBIs and IBIs. However, due to low specificity, the overall AUC was not optimal (AUC of 0.573 for SBIs and 0.562 for IBIs). The team is working to refine the algorithm by recruiting a larger study population to drive locally derived thresholds rather than depend on the existing published thresholds.

Going forward, we are implementing a modified Step-by-Step algorithm to examine outcomes and missed cases, towards validating it as a safe tool for use in Singapore. Further studies should include that of non-invasive biomarkers such as heart rate variability[27] that may add to the emergency physician's armamentarium in deciding which febrile infant should receive priority in early investigations and broad-spectrum antibiotics.

Limitations

We recognise that our study has limitations. At the time of this study, PCT was not routinely assessed for all febrile young infants, resulting in the need to exclude some patients who did not have complete biochemical information. Some cases of bacterial meningitis might have been missed in our cohort because we only included culture-positive bacterial meningitis. However, we believe the number is small and it would not affect our final results. In addition, study investigators who were assessing outcomes were not blinded to the investigation results, which could have resulted in bias. However, we set out clear criteria for SBIs' and IBIs' limited subjectivity in outcome assessment. In the analysis, we recognise that performing a multivariable regression with the biomarkers may have suffered from collinearity, resulting in some biomarkers being rendered not significant at the final multivariable analysis. Finally, this is a single-centre study with limited generalisability due to differences in patient populations and physician practices.

CONCLUSION

In our population, Step-by-Step (original) approach had the highest sensitivity and NPV, while Lab-score has the highest specificity and AUC in predicting both SBIs and IBIs. CRP as a single biomarker was a strong predictor of SBIs, while PCT as a single biomarker was a strong predictor of IBIs.

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