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Original Article

Soluble urokinase plasminogen activator receptor: a novel biomarker of pediatric community-acquired and hospital-acquired pneumonia

Muhammad Said El-Mekkawy[1], Shimaa El-Shafey Soliman[2,3], Nagwan Yossery Saleh[1]

Departments of [1]Pediatrics and [2]Medical Biochemistry and Molecular Biology, Menoufia University Faculty of Medicine, Shebin El-kom, Egypt; [3]Department of Pathology, Medical Biochemistry Unit, Qassim University College of Medicine, Saudi Arabia.

ABSTRACT

Background. Soluble urokinase plasminogen activator receptor (suPAR) is an emerging biomarker in different clinical disorders but data in pediatric pneumonia is scarce. Our objective was to assess utility of suPAR in pediatric community-acquired and hospital-acquired pneumonia.

Methods. A prospective observational study including 120 hospitalized pneumonia patients and 55 healthy controls. Patients fell into two groups: community-acquired pneumonia (CAP) group (75 patients) and hospital-acquired pneumonia (HAP) group (45 patients). CAP severity scores were calculated, including Predisposition, Insult, Response, Organ dysfunction modified (PIROm) score and Pediatric Respiratory Severity (PRESS) Score. suPAR was measured to CAP patients on admission and to HAP patients on the day of pneumonia diagnosis. suPAR was also measured to controls.

Results. suPAR was higher among the whole patient cohort compared with controls ($p<0.001$) and higher among CAP group compared with both controls ($p<0.001$) and HAP group ($p<0.001$). No significant difference was found between HAP and control groups. suPAR was higher among CAP patients with shock, PICU admission, mechanical ventilation, and death ($p=0.013$, 0.044, 0.019, 0.049 respectively). Among CAP patients, suPAR correlated with oxygen saturation, pulse rate, respiratory rate, PRESS, and PIROm. suPAR had area under Receiver Operating Characteristic Curve=0.68 for prediction of severe CAP. Among HAP group, suPAR was negatively correlated with oxygen saturation ($rs=-0.31$; $p=0.048$) and was higher among patients with shock ($p=0.005$) and among those with increased pediatric Sequential Organ Failure Assessment (pSOFA) score ($p=0.034$).

Conclusions. suPAR is promising for diagnosing pediatric CAP but not HAP. suPAR predicted illness severity in both CAP and HAP but performed better in the former.

Key words: urokinase plasminogen activator receptors, pediatrics, biomarkers, pneumonia, ventilator-associated pneumonia, healthcare-associated pneumonia.

Pneumonia is an infection in the lower respiratory tract in which the inflammatory process leads to accumulation of fluid in the airspaces which interferes with gas exchange, leading to the typical symptoms of tachypnoea, increased work of breathing, hypoxia, and cough.[1]

Pediatric pneumonia is associated with significant morbidity and mortality worldwide, accounting for around 15% of deaths in children under the age of five years.[2]

Pneumonia is classified into community-acquired pneumonia (CAP) and hospital-acquired pneumonia (HAP). HAP is defined as pneumonia which is not incubating at the

✉ Muhammad Said El-Mekkawy
mohamed.elmakawi@med.menofia.edu.eg

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time of hospital admission and occurs more than 48 hours after admission.[3] Usage of the term HAP is inconsistent: some use it to refer to any pneumonia that develops in the hospital, including ventilator-associated pneumonia (VAP)[4], while others use the term to refer specifically to pneumonia that develops in the hospital without an association with mechanical ventilation.[3] According to the latter view, HAP and VAP are mutually exclusive entities.

Although both CAP and HAP represent infections of the lung parenchyma, major differences exist between them in terms of the causative pathogens, risk factors, diagnostic criteria, treatment options, and prognosis.

Physicians need objective tools for diagnosing pneumonia, assessing its severity, and predicting its outcome. Soluble urokinase plasminogen activator receptor (suPAR) has recently emerged as a promising candidate.

suPAR is produced by cleavage of urokinase plasminogen activator receptor (uPAR) from cell surface.[5] uPAR is a membrane receptor expressed on monocytes, macrophages, activated T-lymphocytes, and natural killer cells. It binds urokinase plasminogen activator (uPA) and its precursor (pro-uPA). After association with uPAR, pro-uPA is converted to the active enzyme uPA which subsequently activates plasminogen to generate plasmin, producing broad-spectrum proteolytic activity.[6]

uPAR was found to play a role in cell adhesion, chemotaxis, and migration. It can form complexes with the β2-integrin CD11b/ CD18, thereby modulating the migration-promoting activity. suPAR, similarly, has chemotactic properties.[5,7]

suPAR was found to possess diagnostic and prognostic roles in various clinical disorders associated with immune system activation like liver diseases, renal diseases, systemic lupus erythematosus, psoriasis, and malignancy.[8] Furthermore, suPAR proved to be a useful biomarker in sepsis, tuberculosis, malaria, and human immunodeficiency virus.[5] Nevertheless, studies investigating the role of suPAR in pediatric pneumonia are rare, particularly in pediatric HAP, where the topic has not been studied before. The aim of the present study was to assess the diagnostic and prognostic values of suPAR in pediatric pneumonia.

Material and Methods

This was a prospective observational study conducted on 120 patients admitted into the Pediatric Intensive Care Unit (PICU) and the pediatric inpatient ward of a university hospital. The study protocol was approved by the local ethical committee and a written informed consent was obtained from parents.

The study was conducted on children with pneumonia from the age of one month to 15 years. Two patient groups were recruited: the first included children hospitalized with a diagnosis of CAP while the second included children with HAP. Besides, 55 healthy children served as a control group.

CAP was diagnosed in the presence of signs and symptoms of lower respiratory tract infection in a previously healthy child, in association with pulmonary infiltrate on chest radiograph, provided that the infection was acquired outside the hospital. Patients with CAP were hospitalized and admitted into the PICU according to specific criteria.[9]

For the CAP group, exclusion criteria were age <1 month or >15 years; co-existence of another infection with CAP; cough for >14 days; acute bronchiolitis; suspected tuberculosis; chronic respiratory disorders (e.g., persistent asthma, cystic fibrosis, foreign body aspiration, congenital lung anomalies, chronic aspiration; or immune deficiency disorders). Patients who reported history of previous episodes of CAP were not excluded from the study if the number of these episodes was ≤ two and the patient had been symptom-free for> 14 days after the last episode.

CAP severity was assessed on admission according to revised World Health Organization (WHO) criteria for children aged 2 months to 5 years[10]; Respiratory Index of Severity in Children (RISC) score for children <2 years[11]; Pediatric Respiratory Severity Score (PRESS) for patients up to 15 years[12]; and Predisposition, Infection, Response and Organ failure (PIROm) score for patients up to 15 years.[13]

Patients with HAP included 2 subgroups: (1) "VAP": defined as pneumonia that occurred >48 hours after endotracheal intubation. (2) "Non-ventilator HAP": defined as pneumonia that developed >48 hours after hospital admission in the absence of endotracheal intubation.

Patients initially hospitalized for non-respiratory infections (like meningitis), then developed HAP, were not included in the study unless signs of active infection had disappeared, and their C-reactive protein (CRP) levels had become negative. Co-existence of other hospital-acquired infections with HAP was another exclusion criterion. Patients initially hospitalized for pneumonia, then improved but later developed HAP, were not included in the study except after 14 days of admission, which is the "repeat infection timeframe" necessary of an infection of the same type to be considered a new event.[14]

For patients who developed several episodes of VAP, only the first one was included in the study.

Diagnosis of HAP in the present study was made on clinical grounds, according to the Center for Disease Control (CDC) criteria for clinically defined pneumonia (PNU1).[15] Specific microbiological diagnosis of pneumonia was not thoroughly sought due to limited resources. Only blood and pleural fluid cultures were taken. Quantitative cultures from lower respiratory tract samples (e.g., bronchoalveolar lavage and tracheal aspirate) were not performed.

The diagnostic work up for patients included chest x ray; complete blood count (CBC); CRP; blood gas analysis; and serum electrolytes. Chest computed tomography was ordered in specific conditions i.e., for children with poor response to treatment; for evaluation of pleural, mediastinal, or very small parenchymal lesions; and if pneumonia is suspected despite negative or equivocal chest radiographs.

Pediatric Sequential Organ Failure Assessment (pSOFA) score was calculated for patients admitted into PICU.[16] Patients were closely monitored to assess the accuracy of suPAR in diagnosis of pneumonia and its association with hospital mortality and morbidity.

Hospital stay was considered "prolonged" if it was greater than the "median" and was considered "short" if it was≤ the "median".

Laboratory method

For CAP patients, 2 mL blood sample for serum suPAR measurement was withdrawn within 24 hours of hospital admission. For HAP patients, samples were withdrawn on the day when pneumonia was diagnosed. suPAR was also measured in children in the control group. Blood samples were withdrawn in plain vacutainer tubes, incubated for 15 minutes, centrifuged, separated into aliquots, and stored until the test was performed. Serum suPAR levels were measured by Human soluble urokinase type plasminogen activator receptor, suPAR ELISA kit (Chongqing Biospes Co., Ltd, China) which uses double-antibody sandwich enzyme-linked immunosorbent assay (ELISA) according to the manufacturer's protocol.

Statistical method

Qualitative data was expressed as numbers and percents. Non-normally distributed continuous variables were presented as median and range (minimum–maximum). Chi-square test or Fisher exact test were used to assess the association between qualitative variables. Mann-Whitney U test was used for comparing non-normally distributed continuous variables. Correlations between continuous variables were determined by Spearman correlation coefficient (r_s). Receiver Operating Characteristic (ROC) curve analysis

was used to evaluate the performance of suPAR in discriminating patients from controls and discriminating severe pneumonia from non-severe pneumonia. Two-tailed p-value<0.05 was considered statistically significant. Data was analyzed using SPSS version 23 (Statistical Package for Social Science) (Chicago, Inc, Illinose).

Results

Characteristics of the study population

120 patients were recruited along with 55 age and sex-matched controls. The control group consisted of 30 males and 25 females; their median age was 36 months (range 2–144 months) which was not significantly different from that of the patient group (P=0.11). The characteristics of patients are shown in Table I in which baseline clinical and laboratory data of CAP patients were recorded on admission, while baseline data of HAP patients were recorded on the day of pneumonia diagnosis.

Among the CAP patients, 16 (21.3%) reported having had previous episodes:15 patients had one previous CAP episode and one patient had two previous CAP episodes that were treated at home.

The primary reasons for hospitalization of HAP patients included, neurological (40%), gastrointestinal (17.8%), cardiac (11.1%), traumatic (8.9%), respiratory (6.7%), metabolic (2.2%), renal (2.2%), surgical (2.2%), and toxicological (snake envenomation: 2.2%) disorders. Additionally, three patients (6.7%) were admitted for sepsis without focus.

Overall, 17 patients (37.7%) from the HAP group were initially admitted for various infections which improved, but the patients later developed HAP and were included in the study. These infections included central nervous system infections (7 patients), gastroenteritis (4 patients), and sepsis without focus (3 patients). In addition, three patients had been initially hospitalized for pneumonia that improved but HAP developed after 14 days of admission, so they were deemed to have new pneumonia episodes, and were included in the HAP group.

Most HAP patients (75.6%) had VAP. None of the remaining HAP patients needed mechanical ventilation while 13.3% of CAP patients needed mechanical ventilation. HAP patients had significantly higher pulse rate and CRP levels; longer hospital stay; and higher frequency of hypoxia, shock, lobar consolidation, and mortality compared with CAP patients.

Pathogenic bacteria were isolated from 4 patients with CAP: *Staphylococcus aureus* (2 patients, both from pleural fluid), *Streptococcus pneumonia* (one patient, from blood), and group A *Streptococci* (one patient, from blood). As for the HAP group, pathogenic bacteria were isolated from 8 patients: *Pseudomonas aeruginosa* (3 patients, from blood), *Staphylococcus aureus* (2 patient, one from pleural fluid and one from blood), *Acinetobacter* (2 patients, from blood), and *Klebsiella pneumoniae* (one patient, from blood).

Diagnostic value of suPAR

Figure 1 shows that suPAR levels were higher in the whole patient cohort compared with controls (*p<0.001*). suPAR levels were higher among CAP group compared with controls (*p<0.001*), but no significant difference was found between HAP group and controls (p=0.065). suPAR levels were higher among CAP group compared with HAP group (p<0.001). No significant difference in suPAR levels were found between patients with VAP and those with non-VAP HAP (*p=0.31*).

ROC curve analysis revealed that suPAR had an AUC of 0.76 for discriminating the whole patient group from controls [a cutoff level of ≥594.4 pg/ml had a sensitivity of 64.2% and a specificity of 89.1%; p<0.001].

suPAR had an AUC of 0.98 for discriminating CAP patients from controls [a cutoff level of ≥ 504.3 pg/ml had a sensitivity of 92% and a specificity of 90.9%; p<0.001].

Table I. Demographic, clinical, and laboratory characteristics of patients.

Variable	All patients (n=120)	CAP††† (n=75)	HAP‡‡‡ (n=45)	P value
Age, months	18 (1.5 – 168)	18 (1.5 – 108)	12 (2 – 168)	0.012*
Male sex	65 (54.2%)	48 (64%)	17 (37.8%)	0.77
Weight/age z-score				
- 2 to +2 SD	95 (79.2%)	69 (92%)	26 (57.8%)	
< -2 to -3 SD	13 (10.8%)	6 (8%)	7 (15.6%)	<0.001*
< -3SD	12 (10%)	0 (0%)	12 (26.7%)	
Temperature, °C	38.5 (35 – 41)	38.5 (36.5 – 39.5)	38.8 (35 – 41)	0.078
Respiratory rate/minute	53.5 (30 – 93)	48 (33 – 78)	62 (30 – 93)	0.068
Pulse rate/minute	140 (86 –186)	127 (86 – 178)	160 (90 – 186)	<0.001*
Minimum SPO2†, %	92.5 (51 – 99)	95 (65 – 99)	75 (51 – 88)	<0.001*
Hypoxia (SPO2<94%)	30 (25%)	29 (38.7%)	34 (75.6%)	<0.001*
Type of consolidation				
Lobar consolidation	70 (58.3%)	42 (56%)	28 (62.2%)	
Patchy	31 (25.8%)	14 (18.7%)	17 (37.8%)	<0.001*
Interstitial	19 (15.8%)	19 (25.3%)	0 (0%)	
Pleural effusion	11 (9.2%)	7 (9.3%)	4 (8.8%)	0.93
Shock	15 (12.5%)	8 (10.7%)	7 (15.6%)	<0.001*
Invasive MV‡	44 (36.7%)	10 (13.3%)	34 (75.6%)	<0.001*
PICU patients§	56 (46.7%)	20 (26.7%)	36 (80%)	<0.001*
Length of hospital stay, days	8 (5 – 90)	7 (5 – 13)	29.5 (6 – 90)	<0.001*
PIROm¶	NA	1 (0 – 5)	NA	NA
PRESS††	NA	3 (2 – 5)	NA	NA
RISC‡‡	NA	3 (1 – 6)	NA	NA
Hospital mortality	27 (22.5%)	5 (6.7%)	22 (48.9%)	<0.001*
Serum sodium, mEq/L	136 (119 – 167)	134 (119 – 141)	137 (128 – 167)	0.002*
WBC§§ (1000/μL)	9 (1.8 – 41.1)	8 (3 – 41)	12.5 (1.8 – 34.9)	0.39
Hemoglobin, g/dL	10.7 (7.4 – 16)	10.7 (7.4 – 13.5)	10 (7.4 – 16)	0.55
Platelets (1000/μL)	300 (8 – 731)	300 (48 – 731)	254.5 (8 – 661)	0.95
CRP¶¶, mg/dL	26 (0 – 385.4)	24 (0 – 385.4)	48 (12 – 160)	<0.001*

Data is expressed as median (minimum - maximum) and number (percentage); *statistically significant
†Saturation of peripheral Oxygen; ‡Mechanical ventilation; §Pediatric intensive care unit; ¶Predisposition, Insult, Response, Organ dysfunction modified score; ††Pediatric Respiratory Severity Score; ‡‡Respiratory Index of Severity Score; §§White blood cell count; ¶¶C-reactive protein; †††Community-acquired pneumonia; ‡‡‡Hospital-acquired pneumonia, NA: Non-applicable

Association of suPAR with CAP severity

suPAR levels increased significantly in parallel with higher PRESS and PIROm scores (Table II). suPAR levels were significantly higher among CAP patients who died as well as among those who had shock; required PICU admission; or required mechanical ventilation (Table III).

suPAR was positively correlated with pulse rate, respiratory rate, PRESS, and PIROm but negatively correlated with peripheral oxygen saturation (SPO2) [Table IV].

ROC curve analysis revealed that suPAR had an AUC of 0.68 for prediction of severe CAP (as classified by PRESS score) but this was smaller than that of White Blood cell Count (WBC) (Table V). When CAP severity was alternatively defined according to WHO, PRIROm, and RISC; suPAR had an AUC of 0.65 (p=0.043), 0.74 (p=0.11), and 0.69 (p=0.018) respectively.

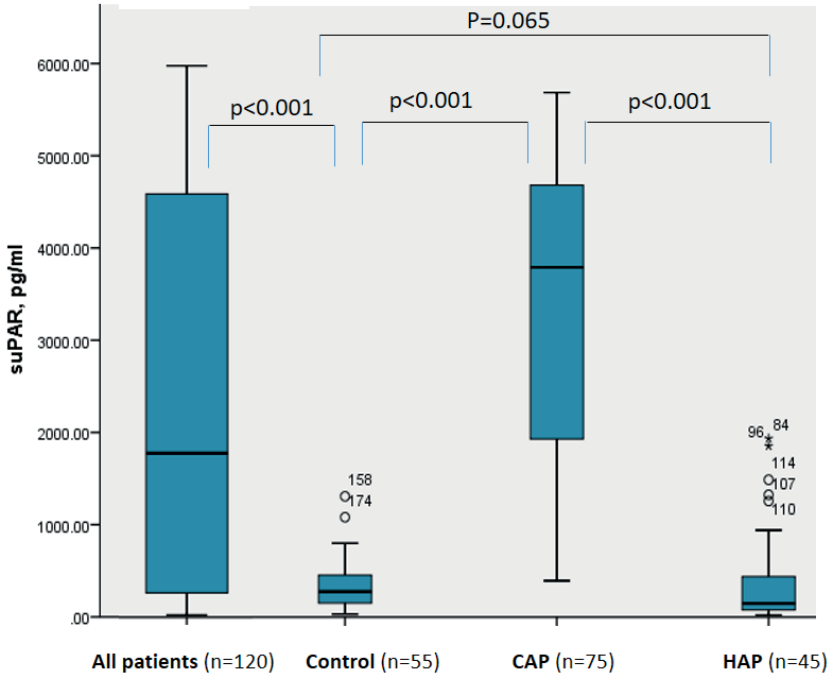

Fig. 1. Serum suPAR levels in patients and controls.

The median and range of suPAR level (pg/mL) in the whole patient cohort, CAP patients, HAP patients, and controls was 1693.7 (22.4–5694.9), 3798 (395–5694.9), 148 (22.4–1939.7), 276.1 (31.6–1311.8), respectively.
In this boxplot, the bold black lines corresponds to the median in each group. The top and bottom of the box represent the 75th and 25th percentiles respectively. The lower whisker corresponds to the 25th percentile minus 1.5 times the interquartile range while the upper whisker corresponds to the 75th percentile plus 1.5 times the interquartile range. Open circles represent "outliers". Asterisks represent "extreme outliers" i.e. data points higher than the 75th percentile plus 3 times the interquartile range.
CAP: Community-acquired pneumonia; HAP: Hospital-acquired pneumonia; suPAR: Soluble urokinase plasminogen activator receptor

Table II. The relation of suPAR levels to different community-acquired pneumonia classification systems.

Pneumonia severity classification	suPAR††, pg/ml	P value
WHO† classification		
Pneumonia (n=52)	3617.4 (395 – 5694.9)	0.11
Severe pneumonia (n=20)	4338 (586.7 – 5540.3)	
RISC‡ score		
< 4 points (n=24)	4393 (489.9 – 5694.9)	0.19
≥ 4 points (n=17)	4433 (586.7 – 5540.3)	
PRESS score§		
Mild: 0-1 points (n=0)	NA‡‡	
Moderate: 2-3 points (n=56)	3518.1 (395 – 5694)	0.02*
Severe: 4-5 points (n=19)	4430 (586.7 – 5540.2)	
PIROm score¶		
Mild: 0-2 points (n=55)	3338.6 (395 – 5694.9)	0.017*
Moderate/severe:3-6 points (n= 20)	4430 (586.7 – 5540.3)	

*Statistically significant
Data is expressed as the median (minimum - maximum)
†World health organization; ‡Respiratory Index of Severity Score; §Pediatric Respiratory Severity Score; ¶Predisposition, Insult, Response, Organ dysfunction modified score; ††Soluble urokinase plasminogen activator receptor; ‡‡Non-applicable

Table III. Soluble urokinase plasminogen activator receptor level in different patient subgroups.

Patient subgroup	CAP‡		HAP¶	
	suPAR§, pg/ml	P value	suPAR, pg/ml	P value
Hypoxia	4246 (586.7 – 5540.3)	0.053	190.3 (22.4 – 1939.7)	0.31
No hypoxia	2899.5 (395 – 5694.9)		138.7 (23.7 – 944.9)	
PICU† patients	4338 (586.7 – 5540.3)	0.044*	160.4 (22.4 – 1939.7)	0.55
Ward patients	3518 (395 – 5694.9)		145.4 (23.8 – 944.9)	
Shock	4808.6 (3246.7 – 5540.3)	0.013*	1260 (96.4 – 1939.7)	0.005*
No shock	3716.6 (395 – 5694.9)		138.9 (23.7 – 728.8)	
Mechanical ventilation	4609.5 (2240 – 5540.3)	0.019*	190.3 (22.4 – 1939.7)	0.31
No mechanical ventilation	3518 (395 – 5694.9)		138.9 (23.7 – 944.9)	
Lobar consolidation	4146.8 (489.7 – 5540.3)		160.4 (28.5 – 1939.7)	
Patchy consolidation	3757.7 (395 – 4975)	0.41	145.4 (22.4 – 1491.5)	0.85
Interstitial	2346.7 (651.9 – 5694.9)		NA	
Prolonged Hospital stay††	4265.5 (586.7 – 5540.3)	0.13	168.6 (22.4 – 1939.7)	0.37
Short hospital stay	3518.1 (395.1 – 5694.9)		148 (23.7 – 1857.4)	
Pleural effusion	3742.3 (727.4 – 4736.4)	0.76	78 (28.5 – 1327.9)	0.39
No effusion	3916.3 (395 – 5694.9)		160.4 (22.4 – 1939.7)	
Non-survivors	4828.3 (3773.5 – 5540.3)	0.049*	138.9 (22.4 – 1939.7)	0.31
Survivors	3729.4 (395 – 5694.9)		206.1 (28.5 – 1857.4)	

*Statistically significant
Data is expressed as the median (minimum - maximum).

†Pediatric intensive care unit; ‡Community-acquired pneumonia; §Soluble urokinase plasminogen activator receptor; ¶Hospital-acquired pneumonia.
††Hospital stay was considered "prolonged" if it was greater than the "median" (>7 days for CAP and > 29.5 days for HAP) and was considered "short" if it was≤ the median.

When CAP patients admitted into PICU were subgrouped according to the median pSOFA score, no significant difference in suPAR levels were found (*p=0.24*)

Association of suPAR with HAP severity

suPAR levels were significantly higher among HAP patients with shock. It was also higher among non-survivors and among those with hypoxia, acidosis, mechanical ventilation, prolonged hospital stay, but without statistical significance (Table III). A negative correlation was found between suPAR and minimum SPO2 (Table IV).

When HAP patients were subgrouped according to the median pSOFA score (9 points) on the day of pneumonia diagnosis, suPAR was higher among the subgroup with elevated pSOFA [408 pg/ml (28.5–1939.7) vs 138.9 (22.4–1491.5); *p=0.034*].

Discussion

suPAR is a novel biomarker that has attracted attention in recent years through demonstrating prognostic value in diverse clinical disorders. In the present study, suPAR proved to have a diagnostic value for pediatric pneumonia since its level was significantly elevated among the whole patient cohort compared with controls.

Pneumonia can be diagnosed by clinical and radiological criteria and physicians usually do not need biomarkers for diagnosing it. However, this is not always the case; for instance, suPAR, can be useful for uncovering the nature of a lung's opacity e.g., differentiating

Table IV. Correlations of suPAR with other variables among patients.

Variable	HAP¶¶ patients		CAP‡‡‡ patients	
	suPAR†††		suPAR	
	Spearman correlation coefficient (r_s)	P value	Spearman correlation coefficient (r_s)	P value
Age	-0.13	0.39	-0.31	0.007*
Weight	-0.19	0.21	-0.32	0.004*
Temperature	-0.23	0.12	0.037	0.75
Respiratory rate	0.16	0.29	0.24	0.041*
Heart rate	0.13	0.41	0.26	0.027*
SPO2†	-0.31	0.048*	-0.24	0.041*
RISC‡	NA	NA	0.11	0.51
PRESS§	NA	NA	0.24	0.043*
PIROm¶	NA	NA	0.29	0.012*
Length of hospital stay	-0.08	0.57	0.19	0.1
MV†† duration	-0.23	0.19	0.23	0.53
Sodium	0.07	0.63	0.08	0.47
CRP‡‡	0.14	0.36	-0.01	0.93
WBC§§	0.08	0.62	-0.09	0.41
Platelets	0.10	0.52	0.11	0.37

*statistically significant
†Saturation of peripheral Oxygen; ‡Respiratory Index of Severity Score; §Pediatric Respiratory Severity Score; ¶Predisposition, Insult, Response, Organ dysfunction modified score; ††Mechanical ventilation; ‡‡C-reactive protein; §§White blood cell count; ¶¶Hospital-acquired pneumonia; †††Soluble urokinase plasminogen activator receptor; ‡‡‡Community-acquired pneumonia

Table V. Prediction of severe community-acquired pneumonia* by Soluble urokinase Plasminogen Activator receptor and other biomarkers.

Variable	AUC (95% CI)¶	Cutoff	P-value	Sensitivity	Specificity
suPAR†, pg/mL	0.68 (0.54 – 0.82)	≥4784	0.021*	84.2%	46.4%
CRP‡, mg/dL	0.42 (0.29 – 0.59)	≥ 30.55	0.31	36.8%	66.1%
WBC§, 1000/μL	0.79 (0.64 – 0.94)	≥12.9	<0.001*	78.9%	91.1%
Platelets, 1000/μL	0.39 (0.22 – 0.55)	≤267	0.15	10.5%	78.6%

*Pneumonia severity is diagnosed by PRESS score
†Soluble urokinase plasminogen activator receptor; ‡C-reactive protein; §White blood cells; ¶Area under the Receiver Operating Characteristic curve and 95% confidence interval.

pneumonia from atelectasis or differentiating current pneumonia from opacity persisting from previous pneumonia episodes. Our current study did not specifically address these prospects, but they can be the subject of further research. Furthermore, suPAR can be utilized for monitoring the response of pneumonia to treatment as suggested by a previous adult study.[17]

For better clarification of the role of suPAR, we included both patient with CAP and those with HAP. suPAR was promising in CAP diagnosis, demonstrating high sensitivity and specificity in discriminating patients from controls. Moreover, suPAR was associated with indicators of CAP severity, including respiratory rate, heart rate, SPO2, shock, PICU admission, mechanical ventilation, and mortality.

As far as we know, only two previous pediatric studies, conducted by the same authors, evaluated the role of suPAR in CAP.[18,19] Consistent with our findings, one of these studies[18] reported a significant elevation of suPAR among CAP patients compared with controls, and showed correlation of suPAR with capillary blood saturation, fever, length of hospital stay, and time for defeverscence.

In addition to the relation of suPAR to individual indicators of CAP severity, we found positive correlation between suPAR and both PIROm and PRESS scores, a finding not reported by previous studies.

Of note, we found no correlation between suPAR and CRP which might be due to a difference in the onset or peak of serum level elevation of these markers. Nevertheless, the latter pediatric studies reported correlations of suPAR with CRP and Procalcitonin.[18,19]

Adult studies on the role of suPAR in CAP are also few, but have similarly shown diagnostic and prognostic values, including association of suPAR with mortality and illness severity. Importantly, the AUC for predicting CAP severity by suPAR in our study was 0.68, compared with a value of 0.71 to 0.84 in adult studies.[20,21] A difference in the type of suPAR assay might underlie that variation, as suggested by a previous study which measured suPAR for the same cohort by two different assays, detecting a difference in the AUC for prediction of mortality (0.80 vs 0.68).[22]

The ability of suPAR to diagnose CAP and predict its outcome raises the question of whether a similar association exists between suPAR and other inflammatory respiratory disorders. We did not explore this issue, but previous studies demonstrated a value for suPAR in predicting bronchopulmonary dysplasia[23] and assessment of asthma control.[24]

The inclusion of patients with recurrent CAP in the present study is unlikely to have affected our results since we stipulated that such patients be symptom-free for >14 days after the last episode. In addition, we excluded patients with chronic respiratory disorders who might have had prior subclinical inflammation affecting suPAR levels. Of note, almost all patients with recurrent CAP had only one previous episode that was diagnosed mostly by doctors in other healthcare institutions.

Unlike the diagnostic role demonstrated by suPAR in CAP, we failed to find a significant differences in suPAR levels between HAP patients and controls. Nevertheless, suPAR was inversely correlated with SPO2 and associated with shock and higher pSOFA score, suggesting a prognostic value in HAP.

Studies on the role of suPAR in HAP are scarce. Moreover, they are generally small and conducted on non-pediatric patients with VAP. In contrast to our findings, a small study of adult mechanically ventilated patients showed that suPAR was significantly higher among patients who developed VAP, both on the day of VAP diagnosis and 3 days before.[25] Another study of 180 adults with VAP and sepsis revealed a significant elevation of suPAR levels among patients compared with controls and among non-survivors compared with survivors.[26] Likewise, another study reported a significant elevation of suPAR among adult ICU patients who developed VAP, compared with those who didn't develop it, but suPAR was not associated with mortality.[27]

Undoubtedly, the small sample size precludes us from drawing firm conclusions regarding the role of suPAR in pediatric HAP. However, if our current findings are confirmed by future studies, this will imply that suPAR has a prognostic, rather than diagnostic, value in pediatric HAP.

The next question will be: why did suPAR levels not rise in HAP to the same extent as in the case of CAP? One possible answer comes from the pathogen type. Pathogens causing HAP are generally distinct from those causing CAP and it was noted that different pathogens

trigger different immunological responses due to interaction with different pattern recognition receptors.[28] It is thus possible that suPAR levels vary according to the type of pathogen and, consequently, according to the type of pneumonia (CAP vs HAP).

Another interesting explanation lies in the phenomenon termed "immunoparalysis" which occurs among critically ill patients who develop severe and persistent compensatory anti-inflammatory response syndrome (CARS) that affects mainly the innate immune system.[29] Most HAP patients had been critically ill before they developed pneumonia. It is, therefore, possible that uPAR expression decreased in these patients due to depression of monocytic function in the course of immunoparalysis, with consequent failure of suPAR to rise during HAP to the high levels found in CAP; therefore, lower suPAR levels could represent a risk factor for HAP development among critically ill children.

Indeed, it has been shown that neutrophil migration from the pulmonary circulation utilizes two pathways, one of them is dependent on CD11b/ CD18 (with which uPAR forms a functional complex). Accordingly, uPAR possesses essential role in combating pulmonary infections, particularly those caused by *Pseudomonas aeruginosa*. Moreover, uPAR was shown to be important for neutrophil recruitment into the alveoli during *S. pneumoniae* infection in a CD11b/ CD18-independent way.[30]

On the other hand, it is possible that HAP patients who did not develop immunoparalysis, showed higher suPAR levels, and consequently, more severe inflammatory response, which could explain the occurrence of some aspects of illness severity in these patients.

It should be emphasized that proving the latter hypothesis requires specific laboratory tests for immunoparalysis which can be the subject of future studies.

Generally speaking, suPAR appears to possess some advantages that favor its use in routine practice, including stability in vitro in serum and plasma over time and during repeated freeze-thawing cycles. Additionally, circadian suPAR levels are stable, so the sampling schedule does not affect its measurement.[31]

Limitations of the present study include the small sample size and the limited scope. Further studies are required to determine the onset, peak, and duration of suPAR level elevation in relation to pneumonia. We also need to know whether suPAR can differentiate pneumonia from other respiratory disorders. Moreover, it is unclear whether suPAR can guide antibiotic therapy or prove superior to other markers like Procalcitonin.

Likewise, thorough specific microbiological diagnosis of pneumonia was not made, so we were not able to assess the relation of suPAR levels to the type of bacteria due to the low yield of microbiological cultures, which were taken from blood and, in few cases, from pleural fluid, but not from lower respiratory secretions. The low yield of blood culture in pneumonia is consistent with previous studies.[32]

In addition, we did not evaluate the ability of suPAR to discriminate viral from bacterial infections. Viral studies were not performed due to lack of resources. It was also not possible to diagnose a viral etiology based on chest radiograph (e.g., interstitial infiltrate) due to poor accuracy of this tool. What is more, a significant proportion of pneumonia cases result from mixed viral and bacterial pathogens.[9]

Finally, some of our patients developed HAP in the context of prior infections, including sepsis, which could have affected serum suPAR level. However, the influence of this factor is unlikely to be important since we included in the study only patients in whom the original infections had significantly improved.

suPAR is a promising marker for pediatric pneumonia. It demonstrated clear diagnostic value in CAP but not in HAP. The prognostic value of suPAR was evident in both groups

but more in CAP; among CAP patients, suPAR was clearly associated with morbidity and with mortality while among HAP patients, suPAR was not associated with mortality but was associated with some aspects of disease severity. In other words, suPAR had both diagnostic and prognostic values in CAP but was only prognostic in HAP. Further studies are needed for stringently assessing the value of suPAR.

Ethical approval

This study was approved by the ethics committee of The faculty of Medicine, Menoufia University (Number:1912/9PEDI15) and written informed consent was obtained from parents.

Author contribution

The authors confirm contribution to the paper as follows: study conception and design: MSE, NYS; data collection: MSE, NYS, SES; analysis and interpretation of results: MSE, NYS, SES; draft manuscript preparation: MSE. All authors reviewed the results and approved the final version of the manuscript.

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Conflict of interest

The authors declare that there is no conflict of interest.

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