Research Article

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Biomarkers in Medicine

# Prognostic performance of pancreatic stone protein in critically ill patients with sepsis

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**Aim:** To assess the prognostic value for 28-day mortality of PSP in critically ill patients with sepsis. **Material & methods:** 122 consecutive patients with sepsis were enrolled in this study. Blood samples were collected on admission and day 2. **Results:** On admission, the combination of PSP and lactate achieved an area under the receiver operating characteristic (AUC-ROC) of 0.796, similar to sequential organ failure assessment score alone (AUC-ROC: 0.826). On day 2, PSP was the biomarker with the highest performance (AUC-ROC: 0.844), although lower (p = 0.041) than Sequential organ failure assessment score (AUC-ROC: 0.923). **Conclusion:** The combination of PSP and lactate and PSP alone, on day 2, have a good performance for prognosis of 28-day mortality and could help to identify patients who may benefit most from tailored intensive care unit management.

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**Keywords:** 28-day mortality • C-reactive protein • lactate • pancreatic stone protein • procalcitonin • prognosis • sepsis • septic shock • serial biomarker measurements • SOFA score

Sepsis is a life-threatening condition caused by a dysregulated host response to infection resulting in multiple organ dysfunctions. Despite significant improvements in clinical management, sepsis remains a leading cause of death worldwide among critically ill patients [1]. Etiologically, sepsis caused by Gram-positive organisms has increased in frequency over time, now being almost as frequent as Gram-negative infections, likely due to the increase of hospital-acquired infections. The respiratory tract, urinary tract and abdomen are the most common sites of infection [1].

Risk stratification is key for the decision-making in the management of patients with sepsis [2]. Risk stratification is currently calculated using scoring systems, using laboratory findings and physiologic measures to determine the degree of organ dysfunction or severity [3]. However, their use to guide decision-making and their applicability in the real-life clinical practice have been questioned [4,5]. Circulating biomarkers would assist physicians in improving risk stratification. Despite extensive research, no biomarker has been universally accepted to support the prognosis of septic patients. The predictive ability of conventional biomarkers, such as CRP and PCT, in critically ill septic patients is controversial [6–11]. The research to identify new biomarkers is a priority for sepsis and septic shock [12].

PSP has recently emerged as a promising sepsis biomarker. It is a protein belonging to the family of lectin-binding proteins [13] secreted by pancreatic acinar cells into pancreatic juice – along with zymogens. PSP is also secreted by subsets of intestinal and gastric cells [14]. Studies focusing on infection and inflammation have postulated that PSP is an acute-phase protein with pro-inflammatory activity, which is released mainly from pancreas in response to injury in the early phase of infection [15,16]. It has been shown to have diagnostic value for sepsis in emergency department [17] and in intensive care unit (ICU) [18] patients, and capacity to predict mortality in ICU patients with sepsis, with a moderate but better accuracy than PCT or CRP [19], improving when combined with severity

scores [20]. However, to our knowledge, no study has either selected cut-off values for prognosis or evaluated the utility of follow-up sampling during the first hours of ICU.

Therefore, this study has evaluated the prognostic ability of baseline and follow-up sampling (days 0 or baseline and 2) of PSP to predict 28-day mortality in patients with sepsis admitted to the ICU from the emergency department or other wards. We have compared PSP with conventional biomarkers, such as lactate, CRP and PCT, and sequential organ failure assessment (SOFA) score.

## Material & methods

This single-center, prospective and observational study was performed at the ICU of Santa Lucía Hospital University Hospital (Cartagena, Spain). This is a 18-bed ICU with both medical and surgical patients. The study was approved by the Hospital Ethics Committee (no. TI. 14/12) and a written informed consent was obtained from all participants or from their close relatives.

### Study subjects

Eligible patients were adult patients (age ≥14 years) consecutively admitted to the ICU of our hospital from May 2013 to May 2014 meeting criteria for severe sepsis or septic shock, based on the Sepsis-2 definition [21]. Enrolled patients also had SOFA score ≥2 and therefore met criteria for sepsis according to the new Sepsis-3 definition for sepsis [22]. Exclusion criteria were: age under 14 years; patients developing sepsis while on ICU but admitted to the ICU for other reasons; patients missing data regarding the primary outcome (28-day mortality); pregnancy; requirement of limitation of therapeutic effort; lack of informed consent; and patients transferred from other ICUs.

Clinical data on admission and during the hospital stay were obtained from the medical records, including demographics, comorbidities, microbiological and biochemical test and SOFA score. For SOFA score calculation, the baseline value was assumed to be zero in patients not known to have pre-existing organ dysfunction, according with Sepsis-3 definition [22]. Patients were treated according to the Survival Sepsis Campaign 2012 guidelines [23]. All patients were followed-up to 28 days for the outcome of all-cause mortality.

### Sample collection & biomarker assays

Baseline blood samples were collected within 6 h after diagnosis of sepsis or septic shock and then on day 2 of ICU stay. Baseline blood lactate levels were measured on ICU and serum PCT and CRP levels were measured in the central laboratory within 1 h. Leftover serum for PSP was immediately frozen and stored to -80°C until testing.

Blood lactate levels were measured by amperometry on an ABL 90 FLEX point-of-care analyzer (Radiometer Medical ApS, Brønshøj, Denmark), with a detection limit of 0.1 mmol/l (from package insert). Serum CRP levels were measured by nephelometry on a Dimension Vista analyzer (Siemens Healthcare Diagnostics, CA, USA), with a detection limit of 0.29 mg/dl. Serum PCT levels were analyzed by electrochemiluminescent immunoassay, on a Cobas e411 analyzer (Roche Diagnostics, Mannheim, Germany), with a detection limit of 0.02 ng/ml and a functional sensitivity of 0.06 ng/ml (from package insert). Serum PSP levels were measured on microplate assays (duplicate runs), using a sandwich isoform-specific ELISA, as previously described [15].

Biomarker level changes (Δ Biomarker), expressed as percentage (%), were calculated according to the following formula: Δ Biomarker (%) = 100 × ([baseline biomarker level - biomarker level on day 2]/baseline biomarker level).

### Statistics

Data were tested for normality. Because continuous variables were not normally distributed, they were reported as median and interquartile range and nonparametric tests were used to compare them using the Mann–Whitney U test for independent data and Wilcoxon's test for paired data. Categorical variables were expressed as frequencies and percentages with differences analyzed with the $\chi^2$ test. We used different measurements of performance to test the potential prognostic value of PSP and the other circulating markers included in the study. Discriminatory ability was evaluated by calculating the area under the receiver operating characteristic curve (AUC-ROC). The DeLong method was used to compare the AUC-ROCs. We additionally calculated the optimal ROC-derived cutoffs (Youden Index – corresponding to the maximum of the sum 'sensibility + specificity') and sensitivity, specificity and predictive values of them. The model calibration was evaluated using the Hosmer–Lemeshow test. Finally, the value of biomarkers over SOFA score in term of reclassification was evaluated by calculating the net reclassification

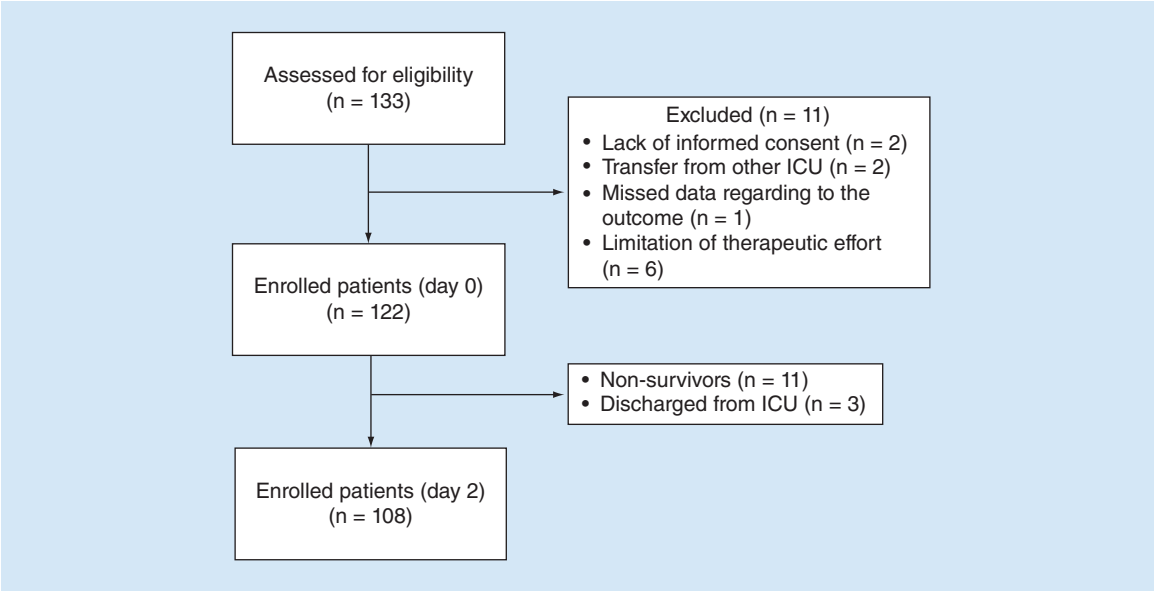


**Figure 1. Flowchart of patients' enrollment in the study.**
ICU: Intensive care unit.

index (NRI). Because NRI requires a previous definition of risk categories, we used the selected cutoffs according to Youden Index. The association between biomarkers and 28-day mortality was assessed by Cox regression analysis; for this analysis, biomarkers were treated as continuous variables. Variables yielding a p-value of <0.15 in the univariate regression analysis were further included in the multivariate analysis, similarly to previous studies [24], and a backward selection was used for the covariate selection in the multivariate model. Covariates were retained in the final model if they were statistical significant with a p-value of <0.05. Kaplan–Meier curves were used to analyze the survival and the Mantel–Kaenszel log-rank test for comparison among survivors and nonsurvivors. We performed analyses by using the software packages SPSS 21.0 (SPSS Inc., IL, USA) and MedCalc 15.0 (MedCalc Software, Ostend, Belgium). In all tests, a two-sided p-value of <0.05 was considered significant.

## Results

### Characteristics of patients

In the study period, 133 patients with sepsis were admitted to the ICU. Total 11 patients were excluded according to exclusion criteria (Figure 1). Consequently, 122 patients (median age: 62 years [interquartile range: 52–72]; 68 [55.7%] male), 64 (52.5%) with sepsis and 58 (47.5%) with septic shock were included in the study. Total 97 (79.5%) and 25 (20.5%) patients presented with community-acquired and nosocomial infections, respectively. The most common source of infection was abdominal (32%), followed by respiratory (26.2%) and urinary (20.5%). Infection was microbiologically documented in 89 (73.0%) patients; in these patients, Gram-negative bacteria were the main causative agent for infection (n = 49; 52.1%). Bacteremia was detected in 47 (38.5) patients, being *Escherichia coli* the most frequent isolate (n = 13). The 28-day mortality was 27% and it was higher in patients with septic shock (50%) in comparison with patients with sepsis (6.7%). Patients' characteristics according to survival status are presented in Table 1.

### Biomarker levels

Biomarker levels are presented in Table 2 according to survival status. When the relation between biomarker levels and 28-day mortality was evaluated, baseline PSP and lactate levels were significantly higher in nonsurvivor patients compared with survivors, whereas CRP and PCT remained similar in both groups (Table 3). On day 2, PSP levels were also significantly higher in nonsurvivors, difference not observed for the other tested biomarkers, PCT and CRP (Table 2).

Changes in biomarker levels from baseline to day 2 were also analyzed. In survivors, levels of CRP, PCT and PSP on day 2 were lower than baseline levels and a significant decrease trend was observed for all of them. In

**Table 1. Patient characteristics by survival status.**

| Characteristic | Total (122) | Survivors; 89 (73%) | Nonsurvivors; 33 (27%) | p-value |
|---|---|---|---|---|
| Age (years) | 65 (52–72) | 64 (49–73) | 70 (56–81) | 0.030 |
| Male | 68 (55.7) | 47 (52.8) | 21 (63.6) | 0.285 |
| Type of patient: | | | | 0.212 |
| – Medical | 94 (77) | 66 (74.2) | 28 (84.8) | |
| – Surgical | 28 (23) | 23 (25.8) | 5 (15.2) | |
| Patient origin: | | | | 0.573 |
| – Emergency department | 86 (70.5) | 64 (71.9) | 22 (66.7) | |
| – Ward | 36 (29.5) | 25 (28.1) | 11 (23.3) | |
| SOFA, median: | | | | |
| – Baseline | 8 (6–10) | 7 (5–9) | 11 (9–14) | <0.001 |
| – On day 2[†] | 6 (4–9) | 5 (3–7) | 12 (9–15) | <0.001 |
| Severity: | | | | <0.001 |
| – Sepsis | | 60 (67.4) | 4 (12.1) | |
| – Septic shock | | 29 (32.6) | 29 (87.9) | |
| Comorbidities: | | | | |
| – Diabetes | 28 (23.0) | 23 (25.8) | 5 (15.2) | 0.212 |
| – Immunosuppression | 19 (15.6) | 11 (12.4) | 8 (24.2) | 0.108 |
| – Chronic renal failure | 16 (13.1) | 10 (11.2) | 6 (18.2) | 0.313 |
| – COPD | 13 (10.7) | 10 (11.2) | 3 (9.1) | 0.733 |
| – Neoplasia | 25 (20.5) | 16 (18) | 9 (27.3) | 0.259 |
| – Heart failure | 8 (6.6) | 5 (5.6) | 3 (9.1) | 0.491 |
| – Ischemic cardiopathy | 12 (9.8) | 10 (11.2) | 2 (6.1) | 0.394 |
| AKI (stages 2 and 3 KDIGO) | 64 (52.5) | 37 (41.6) | 27 (81.8) | <0.001 |
| Replacement renal therapy | 29 (23.8) | 9 (10.1) | 20 (60.6) | <0.001 |
| IMV | 52 (42.6) | 27 (30.3) | 25 (75.8) | <0.001 |
| NIMV | 32 (26.2) | 19 (21.3) | 13 (39.4) | 0.044 |
| ICU LOS (days) | 4 (2–9) | 4 (3–7.5) | 4 (1.5–10.5) | 0.580 |
| Biochemical parameters (on admission to ICU): | | | | |
| – Creatinine (mg/dl) | 1.90 (1.10–3.00) | 1.70 (1.04–2.82) | 2.60 (1.56–3.48) | 0.011 |
| – Bilirubin (mg/dl) | 1.0 (0.6–1.9) | 1.0 (0.5–1.4) | 1.1 (1.0–3.1) | 0.002 |
| – Platelet count (/μl) | 140 (104–206) | 139 (108–232) | 143 (70–209) | 0.387 |
| Type of infection: | | | | 0.904 |
| – Community acquired infection | 97 (79.5) | 71 (79.8) | 26 (78.8) | |
| – Nosocomial infection | 25 (20.5) | 18 (20.2) | 7 (21.2) | |
| Infection focus: | | | | 0.364 |
| – Abdominal | 39 (32.0) | 30 (33.7) | 9 (27.3) | |
| – Respiratory | 32 (26.2) | 21 (23.6) | 11 (33.3) | |
| – Urinary | 25 (20.5) | 21 (23.6) | 4 (12.1) | |
| – Skin and soft tissues | 9 (7.4) | 7 (7.9) | 2 (6.1) | |
| – Isolated bacteremia | 9 (7.4) | 4 (4.5) | 5 (15.2) | |
| – Other | 4 (3.3) | 3 (3.4) | 1 (3.0) | |
| – Unknown | 4 (3.3) | 3 (3.4) | 1 (3.0) | |
| Etiology: | | | | 0.182 |
| – Gram- bacteria | 49 (52.1) | 37 (59.7) | 12 (44.4) | |
| – Gram+ bacteria | 29 (32.6) | 20 (32.3) | 9 (33.3) | |
| – Polymicrobial | 9 (10.1) | 4 (6.5) | 5 (18.5) | |
| – Fungal | 1 (1.1) | – | 1 (3.7) | |
| – Other | 1 (1.1) | 1 (1.6) | – | |

Data shown as n (%) or median (IQR) unless otherwise stated.

[†] Data available in 108 patients (86 survivors and 22 nonsurvivors). Ten patients who died and four who were discharged to ward before collecting blood on day 2 were excluded, being 108 patients still in ICU on day 2 finally used for this analysis.

AKI: Acute kidney injury; COPD: Chronic obstructive pulmonary disease; ICU: Intensive care unit; IMV: Invasive mechanical ventilation; IQR: Interquartile range; KDIGO: Kidney disease improving global outcome; LOS: Length of stay; NIMV: Noninvasive mechanical ventilation; SOFA: Sequential organ failure assessment.

Table 2. Biomarker levels according to survival status.

| Characteristic | Total (122) | Survivors; 89 (73%) | Nonsurvivors; 33 (27%) | p-value |
|---|---|---|---|---|
| **Baseline biomarkers, median (IQR)** | | | | |
| CRP (mg/dl) | 21.0 (13.8–32.0) | 22.0 (13.5–32.0) | 20.0 (130–31.5) | 0.901 |
| PCT (ng/ml) | 12.6 (5.4–6.6) | 11.3 (5.4–32.7) | 15.0 (5.6–71.0) | 0.370 |
| Lactate (mmol/l) | 2.2 (1.3–4.0) | 2.0 (1.1–3.0) | 4.0 (2.2–6.9) | <0.001 |
| PSP (ng/ml) | 436 (217–621) | 381 (180–535) | 604 (415–738) | <0.001 |
| **Biomarkers on day 2[†], median (IQR)** | | | | |
| CRP (mg/dl) | 18.0 (11.2–278) | 17.0 (11.0–25.3) | 24.5 (15.8–31.3) | 0.061 |
| Δ CRP (%) | 17 (-14–44) | 19 (-16–47) | 5 (-14–33) | 0.259 |
| PCT (ng/ml) | 5.8 (2.2–15.4) | 5.0 (2.2–12.8) | 13.2 (2.1–32.5) | 0.053 |
| Δ PCT (%) | 61 (23–73) | 62 (44–75) | 24 (-93–61) | <0.001 |
| PSP (ng/ml) | 204 (68–442) | 134 (60–368) | 547 (310–727) | <0.001 |
| Δ PSP (%) | 40 (-5–71) | 52 (12–73) | -3 (-17–34) | <0.001 |

[†]Data available in 108 patients (86 survivors and 22 nonsurvivors). Total ten patients who died and four who were discharged to ward before collecting blood on day 2 were excluded, being 108 patients still in ICU on day 2 finally used for this analysis.

Table 3. Accuracy of biomarkers for the prognosis of 28-day mortality.

| Baseline | Cut-off | Se (%) | Sp (%) | PPV (%) | NPV (%) |
|---|---|---|---|---|---|
| Lactate (mmol/l) | 3.2 | 63.6 (45.1–79.6) | 79.8 (69.9–87.6) | 53.8 (37.2–69.9) | 85.5 (76.1–92.3) |
| PSP (ng/ml) | 581 | 57.6 (39.2–74.5) | 82.0 (72.5–89.4) | 54.3 (36.6–71.2) | 83.9 (74.5–90.9) |
| **Day 2** | | | | | |
| PSP (ng/ml) | 201 | 95.5 (77.2–99.9) | 59.3 (48.2–69.8) | 37.5 (24.9–51.5) | 98.1 (89.7–100.0) |
| Δ PCT | 40 | 68.2 (45.1–86.1) | 77.9 (67.7–86.1) | 44.1 (27.2–62.1) | 90.5 (81.5–96.1) |
| Δ PSP | 23 | 72.7 (49.8–89.3) | 70.9 (60.1–80.2) | 39.0 (24.2–55.5) | 91.0 (81.5–96.6) |

Data shown as n (%) or median (IQR) unless otherwise stated.
Brackets indicate 95% CI. In this table, only those biomarkers with a significant area under the receiver operating characteristic curve ($p < 0.05$) to predict 28-day mortality were included.
NPV: Negative predictive value; PPV: Positive predictive value; Se: Sensitivity; Sp: Specificity.

nonsurvivors, no significant change in biomarker levels was found (Figure 2). When the differences in the dynamic change of biomarker levels between survival and nonsurvival groups were compared, only the changes in PCT and PSP from baseline to day 2 reached statistical significance (Table 2).

## Baseline biomarker levels for prediction of 28-day mortality

Figure 3 shows the ROC curves of SOFA score and the different tested biomarkers, measured on admission, to predict 28-day mortality. Lactate and PSP were the only biomarkers with a significant discriminative value to predict 28-day mortality. A 28-day mortality prediction model including PSP and lactate had an AUC-ROC of 0.796, which was similar to that achieved by SOFA score alone (0.826). Of note, when both biomarkers were added to the SOFA score, AUC-ROC increased to 0.866, with a difference close to the statistical significance ($p = 0.080$) between both AUC-ROCs. The Hosmer–Lemeshow $\chi^2$ test was 9.32 ($p = 0.316$) for the model including PSP and lactate and 3.24 ($p = 0.919$) for the model including SOFA score and both biomarkers, indicating a good calibration of both. Optimal cutoffs derived from ROC curves are listed in Table 3. The reclassification analysis showed that NRI favored the combination of PSP and lactate when compared with SOFA score to identify nonsurvivors (NRI: 2.7; $p = 0.008$).

In the Kaplan–Meier analysis, mortality was significantly higher in the patients with increased PSP and lactate levels (Figure 4A). The 28-day mortality rates for patients with PSP and lactate levels above the selected cutoffs were 54.3 and 53.8%, significantly higher than in patients with PSP and lactate level below the cutoffs, with 28-day mortality rates of 16.1 and 14.5%, respectively.

A Cox regression analysis was performed to evaluate variables independently associated with 28-day mortality. In the multivariate analysis, after adjusting for age, SOFA score, AKI and immunosuppression, PSP was the only independent predictor biomarker for 28-day mortality (Table 4).

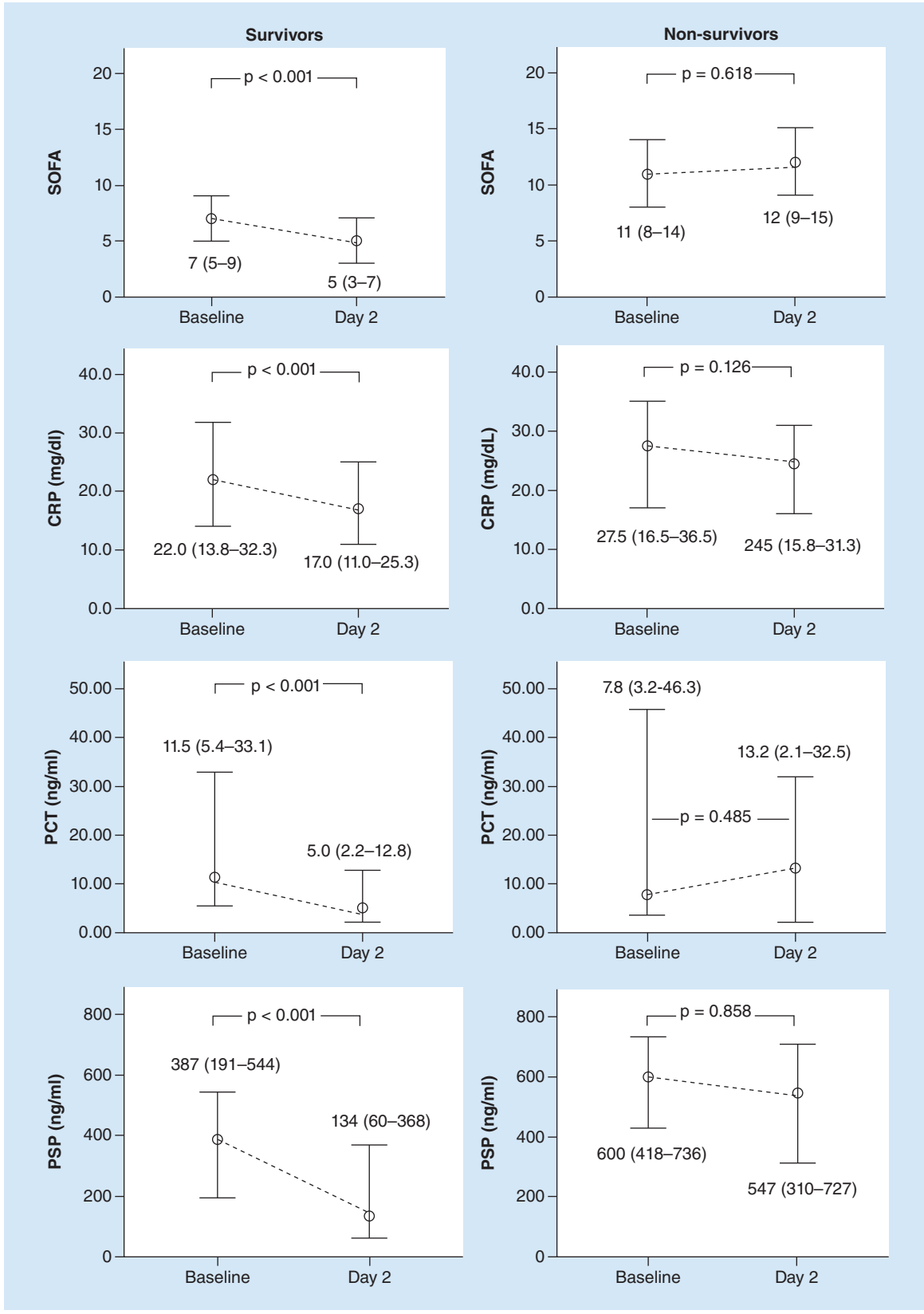


**Figure 2. Changes in sequential organ failure assessment score and biomarker levels from baseline 0 to day 2.**
SOFA: Sequential organ failure assessment.

| | AUC-ROC | 95% CI | p-value | p* |
|---|---|---|---|---|
| SOFA | 0.826 | 0.747–0.889 | <0.001 | – |
| Lactate | 0.743 | 0.656–0.818 | <0.001 | 0.102 |
| PSP | 0.724 | 0.636–0.801 | <0.001 | 0.103 |
| PCT | 0.553 | 0.460–0.643 | 0.420 | <0.001 |
| CRP | 0.507 | 0.415–0.599 | 0.904 | <0.001 |
| PSP + Lactate | 0.796 | 0.710–.881 | <0.001 | 0.565 |
| SOFA+ Lactate | 0.841 | 0.764–0.901 | <0.001 | 0.177 |
| SOFA + PSP | 0.859 | 0.784–0.915 | <0.001 | 0.140 |
| SOFA + PSP + Lactate | 0.866 | 0.792–0.921 | <0.001 | 0.080 |

*Comparison vs. SOFA

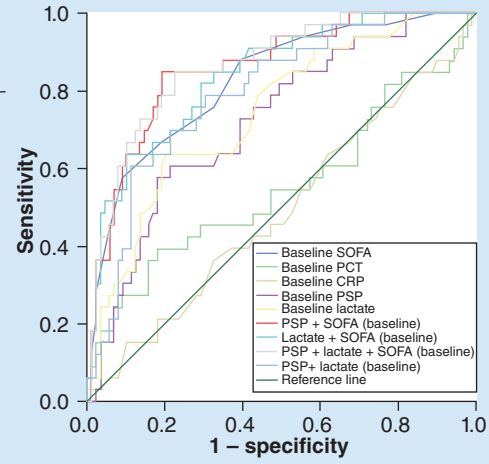


**Figure 3. Receiver operating characteristic curves of sequential organ failure assessment score and biomarker levels on admission to intensive care unit to predict 28-day mortality.** Other comparisons between biomarkers: CRP vs. PCT p = 0.613; CRP vs. lactate p < 0.001; CRP vs. PSP p = 0.008; PCT vs. Lactate p = 0.006; PCT vs. PSP p = 0.011; Lactate vs. PSP p = 0.789.
AUC-ROC: Area under the receiver operating characteristic curve; CI: Confidence intercal; SOFA: Sequential organ failure assessment.

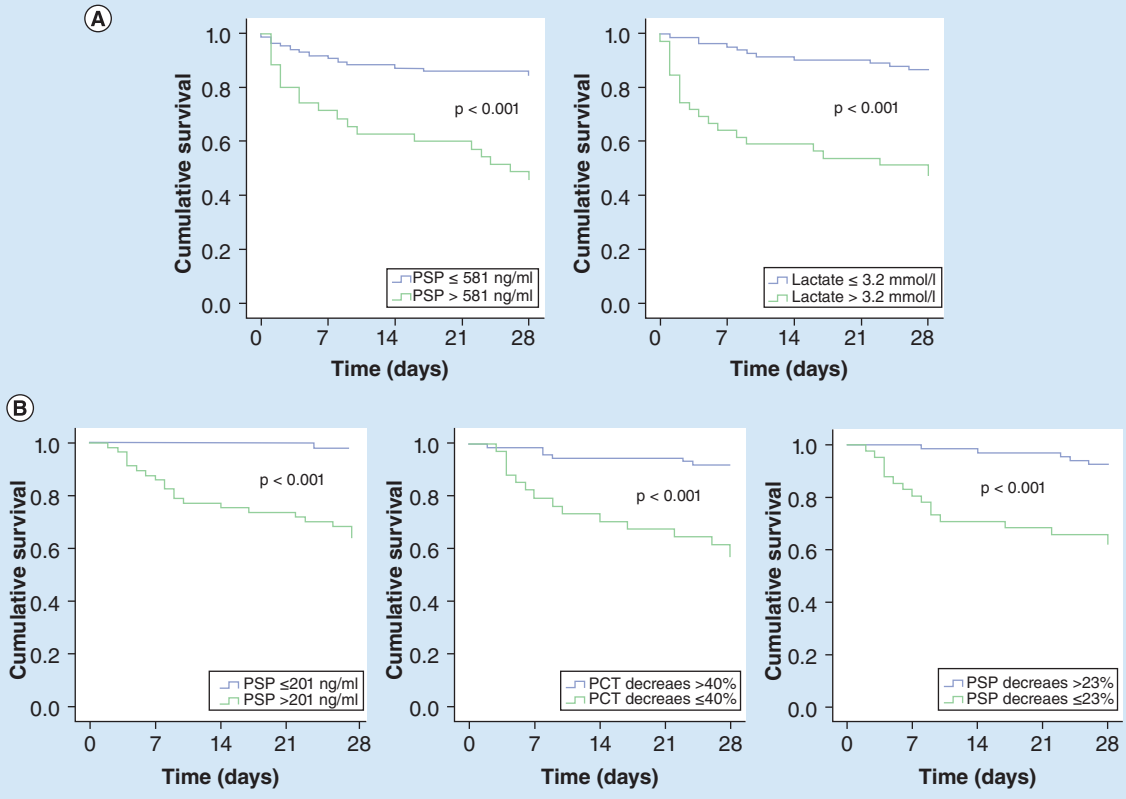


**Figure 4. Kaplan–Meier curves showing the survival according to increased lactate and PSP levels on admission (A) and increased PSP levels on day 2 and PSP and PCT decreases in the first 48 hours (B).**

**Table 4. Univariate and multivariate Cox proportional hazards analysis of 28-day mortality.**

| Variable | Univariate | | Multivariate | |
|---|---|---|---|---|
| | HR (95% CI) | p-value | HR (95% CI) | p-value |
| Age (years) | 1.03 (1.00–1.05) | 0.069 | 1.02 (0.98–1.05) | 0.395 |
| Male gender | 1.56 (0.69–3.56) | 0.287 | NI | |
| SOFA score | 1.46 (1.25–1.71) | <0.001 | 1.43 (1.22–1.68) | <0.001 |
| Surgical patient | 1.67 (0.14–20.40) | 0.689 | NI | |
| Immunosupression | 2.27 (0.82–6.27) | 0.114 | 3.36 (0.75–15.01) | 0.112 |
| AKI | 2.68 (0.85–8.41) | 0.092 | 0.31 (0.06–1.71) | 0.178 |
| Nosocomial infection | 1.06 (0.40–2.84) | 0.904 | NI | |
| Biomarkers (on admission to ICU): | | | | |
| – CRP | 1.00 (0.97–1.03) | 0.939 | NI | |
| – PCT | 1.01 (1.00–1.02) | 0.034 | 1.00 (0.99–1.01) | 0.647 |
| – PSP | 1.34 (1.12–1.62) | <0.001 | 1.27 (1.08–1.58) | <0.001 |
| – Lactate | 1.26 (1.09–1.45) | 0.001 | 1.14 (0.96–1.36) | 0.126 |

AKI: Acute kidney injury; HR: Hazard ratio; ICU: Intensive care unit; NI: Not included; SOFA: Sequential organ failure assessment.

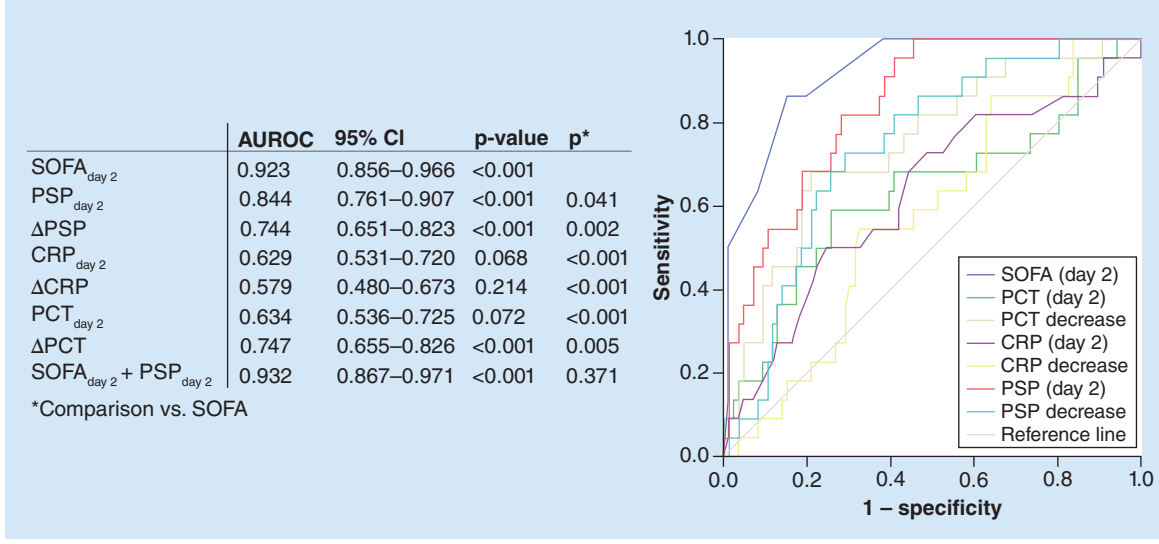


**Figure 5. Receiver operating characteristic curves of sequential organ failure assessment. score, biomarker levels on day 2 and changes of biomarker levels from baseline to day 2 to predict 28-day mortality.**
AUROC: Area under the receiver operating characteristic curve; CI: Confidence interval; SOFA: Sequential organ failure assessment.

### Biomarker levels on day 2 & changes from baseline to day 2 for prediction of 28-day mortality

Figure 5 illustrates the ROC curves of SOFA score and biomarkers on day 2 and the changes in biomarker levels from baseline to day 2 to predict 28-day mortality. PSP levels and the changes in PCT (ΔPCT) and PSP (ΔPSP) levels showed a significant predictor capacity, with PSP measured on day 2 achieving the highest discriminatory value. Optimal cutoffs are listed in Table 3; according to these cut-off points, a long-rank test showed a significant difference for PSP, ΔPCT and ΔPSP between survival curves in the two groups (Figure 4B).

## Discussion

Sepsis is still a major challenge in critical care. Because it is a time-sensitive condition, early diagnosis and implementation of appropriate antibiotic and supportive therapy, source control and close monitoring remain the cornerstones to decrease the morbidity and mortality related to sepsis [2]. However, the early risk stratification of septic patients still remains an unsolved issue and novel strategies are required [12]. In this sense, biomarkers are

emerging tools to help monitor patient response to treatment and guide therapeutic decisions in individual patients. However, the ability of conventional biomarkers for risk stratification in patients with sepsis is limited.

Recent studies have reported the value of a single measurement of PSP on admission to ICU for prediction of in-hospital mortality in ICU patients with sepsis, with AUC-ROCs ranging from 0.65 to 0.665 [19,20]. In our present study, baseline PSP showed a slightly higher predictive accuracy, similar to other prognostic biomarkers commonly used in critically ill patients, such as lactate, and better than other traditional infection biomarkers, such as CRP and PCT. For the other tested biomarkers, similar results have been previously reported for CRP [6,19,20,25–27], PCT [19,20,25–28] and lactate [26,29,30]. Of note, AUC-ROC of PSP to predict 28-day mortality was slightly higher than those achieved by other emerging biomarkers measurable in automated analyzers, such as presepsin [27,28], NGAL [31] and GDF15 [32] and similar to that reported for MR-proADM [26]. Besides, baseline PSP was the only independent predictor biomarker for 28-day mortality. To our knowledge, no study has previously evaluated the association between PSP and mortality by using Cox regression analysis

In the present study, SOFA score achieved the highest performance for 28-day mortality. This score is a well-established and validated tool for prognosis of critically ill patients, but its calculation is time consuming and requires a significant data collection burden in a setting where delays in clinical decision-making increase the risk for adverse outcomes [33]. It is remarkable that the combination of lactate and PSP achieved a very similar performance than SOFA score alone and the reclassification analysis showed a higher NRI to identify nonsurvivors. The combined measurement of both biomarkers, easily measurable with point-of-care testing (POCT) devices [34] as rapid bedside tests, as the European Society of Clinical Microbiology and Infectious diseases has recently recommended [35], could lead to a rapid decision-making for the attending physician confronted with the septic patient.

Models combining biochemical markers and severity scores represent a new and interesting strategy to overcome the limited prognostic performance of single parameters, although the results about this approach are controversial [26,36,37]. In Que *et al.* study [20], the models combining PSP with either APACHE II or Simplified Acute Physiology Score II predicted in-hospital mortality better than those models based on the addition of CRP or PCT. In our study, the combination of SOFA score, lactate and PSP measured at baseline was the best model to predict 28-day mortality and showed a trend to improve the performance of SOFA alone.

A second strategy to improve the prognostic value of circulating biomarkers is by using serial measurements because their time courses may be more reliable than their absolute levels. Recently, the US FDA has approved the use of PCT for prognosis of 28-day mortality supported by the results reported by Schuetz *et al.* [11], which concluded that the inability of treatment to lead to PCT drops of more than 80% from baseline to day 4 correlates with poor 28-day mortality prognosis. However, the optimal definition of PCT clearance for accurate risk assessment has not been clearly defined [10].

We have also evaluated the predictive utility of serial measurements of PSP, in comparison with SOFA score, CRP and PCT. To our knowledge, the utility of follow-up sampling has not yet been studied. In this study, PSP level measured on day 2 was the biomarker with the highest performance to predict 28-day mortality, although PCT-decrease and PSP-decrease also achieved a significant, but moderate, accuracy.

On day 2, PSP showed a high discriminatory ability for 28-day mortality, higher than that reported for other emerging biomarkers, such as presepsin [28]. Although the performance of SOFA score on day 2 was higher than that of PSP, the quick measurement of this biomarker by using POCT analyzers [34] could be useful to triggering assessing the SOFA score to understand better the extend of organ dysfunction, selecting a cut-off with a high sensitivity.

## Strengths & limitations

To our knowledge, this is the first study including serial measurements of PSP and calculating PSP thresholds for prognosis of mortality in ICU patients. Besides, we have tested the performance of PSP in comparison with a conventional biomarker, such as lactate, commonly used for prognosis in ICU. Unlike our study, in many studies about emergent biomarkers [19,25,27,31,32], lactate performance for predicting mortality was not reported and compared with the tested biomarker. It does not truely reflect the possible impact of a new biochemical marker in the current clinical practice.

On the other hand, our study has several limitations. First, this study took place in one center, this limits the generality of our findings to other institutions and populations. Second, due to the relatively small sample size of patients, our results require further validation in a larger and multicentric cohort of patients with sepsis. Third, although extensive daily monitoring of circulating biomarkers is important during the early management of sepsis,

only two measurements (baseline and on day 2) were available. Finally, lactate level on day 2 was not available, although Ferreruela *et al.* [38] have recently reported that lactate clearance has a limited value to predict survival in critically ill patients.

### Future studies

Because a POCT analyzer is available for a rapid and reliable measurement of PSP [34], future studies should focus on investigating the clinical and economic impact of PSP on the clinical ICU routine.

## Conclusion

Our results confirm the value of PSP to predict mortality in patients with sepsis requiring ICU management. On admission, a model combining two biomarkers, such as lactate and PSP, measurable in a reduced turnaround time by POCT methodologies, and on day 2, the measurement of PSP, may help clinicians in the challenging task of improving the care of critically ill patients with sepsis. Further studies in larger populations are required to validate our promising results.

### Summary points

- Sepsis remains a leading cause of death worldwide among critically ill patients and risk stratification is key for the decision-making in the management of these patients.
- Blood biomarkers could be tools to improve the risk stratification, but no biomarker has been universally accepted to support it. The predictive ability of conventional biomarkers, such as CRP and PCT in critically ill septic patients is controversial.
- PSP has emerged as a new biomarker for sepsis, and previous studies have reported, when measured on admission to intensive care unit, its predictive ability for mortality in critically ill patients with sepsis. However, the value of serial measurements has not been studied.
- On admission, the combination of PSP and lactate has a good accuracy for prognosis of 28-day mortality, similar than SOFA score alone. Besides, PSP was the only independen predictor biomarker for 28-day mortality.
- On day 2, PSP was the biomarker with the highest accuracy to predict 28-day mortality.
- The combination of PSP and lactate, on admission, or PSP levels, measured on day 2, may help clinicians in the challenging task of improving the care of critically ill patients with sepsis.

### Author contributions

L García de Guadiana-Romualdo and M Albaladejo-Otón carried out the study design. E Jiménez-Santos, A Hernando-Holgado and P Esteban-Torrella recruited the subjects for the study. S Rebollo-Acebes, R Jiménez-Sánchez and A Ortín-Freire reviewed the clinical records and classified the subjects. E Jiménez-Santos was responsible for measuring P Esteban-Torrella. L García de Guadiana-Romualdo, M Albaladejo-Otón and M Berger analyzed and interpreted the data and wrote the manuscript. J Trujillo-Santos participated in the statistical analysis. All authors read and approved the final manuscript.

### Financial & competing interests disclosure

Abbott Laboratories supported the study providing reagents and other material for measurement of PSP. Abbott GmbH and Co. KG did not participate in the study design or analysis and interpretation of results. M Berger was employee of Abbott GmbH and Co. KG. Wiesbaden, Germany, when the study was performed. The authors have no other relevant affiliations or financial involvement with any organization or entity with a financial interest in or financial conflict with the subject matter or materials discussed in the manuscript apart from those disclosed.

No writing assistance was utilized in the production of this manuscript.

### Ethical conduct of research

The authors state that they have obtained appropriate institutional review board approval and have followed the principles outlined in the Declaration of Helsinki for all human or animal experimental investigations. In addition, informed consent has been obtained from the participants involved.

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