A novel H-FABP assay and a fast prognostic score for risk assessment of normotensive pulmonary embolism

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Summary
We tested whether heart-type fatty acid binding protein (H-FABP) measured by a fully-automated immunoturbidimetric assay in comparison to ELISA provides additive prognostic value in patients with pulmonary embolism (PE), and validated a fast prognostic score in comparison to the ESC risk prediction model and the simplified Pulmonary Embolism Severity Index (sPESI). We prospectively examined 271 normotensive patients with PE; of those, 20 (7%) had an adverse 30-day outcome. H-FABP levels determined by immunoturbidimetry were higher (median, 5.2 [IQR, 2.7–9.8] ng/ml) than those by ELISA (2.9 [1.1–5.4] ng/ml), but Bland-Altman plot demonstrated a good agreement of both assays. The area under the curve for H-FABP was greater for immunoturbidimetry than for ELISA (0.82 [0.74–0.91] vs. 0.78 [0.68–0.89]; P=0.039). H-FABP measured by immunoturbidimetry (but not by ELISA) provided additive prognostic information to other predictors of 30-day outcome (OR, 12.4 [95% CI, 1.6–97.6]; P=0.017). When H-FABP determined by immunoturbidimetry was integrated into a novel prognostic score (H-FABP, Syncope, and Tachycardia; FAST score), the score provided additive prognostic information by multivariable analysis (OR, 14.2 [3.9–51.4]; P<0.001; c-index, 0.86) which were superior to information obtained by the ESC model (c-index, 0.62; net reclassification improvement (NRI), 0.39 [0.21–0.56]; P<0.001) or the sPESI (c-index, 0.68; NRI, 0.24 [0.05–0.43]; P=0.012). In conclusion, determination of H-FABP by immunoturbidimetry provides prognostic information superior to that of ELISA and, if integrated in the FAST score, appears more suitable to identify patients with an adverse 30-day outcome compared to the ESC model and sPESI.

Introduction
Pulmonary embolism (PE) is the third most common cause of death from acute cardiovascular disease after myocardial infarction and stroke (1) with a mortality rate of approximately 5–17% during the first 1–3 months (2–4) and 36% over four years (5). Current guidelines recommend to risk stratify patients into high- and non-high-risk PE in order to tailor medical and interventional therapy (6). Since acute pressure overload in PE may lead to right ventricular (RV) failure (7) research has focused on the assessment of RV dysfunction (8-11). Based on the available evidence, the European Society of Cardiology (ESC) recommended the combination of RV dysfunction, detected by imaging procedures or elevated levels of natriuretic peptides, with evidence of myocardial injury indicated by elevated troponin levels for risk stratification of PE (so-called ESC model) (6). Additionally, risk prediction models which include clinical information have been tested to assess the risk of PE patients. In particular, Jiménez et al. (12) proposed a simplified version of the Pulmonary Embolism Severity Index (sPESI) which reduces the technical complexity of the original score (13) by focusing on six equally weighted variables.

We and others have reported that heart-type fatty acid binding protein (H-FABP), a small (15 kDa) marker of myocardial ischaemia with favourable release kinetics, may perform better than NT-proBNP and cardiac troponins in predicting an adverse 30-day outcome and long-term mortality, both in unselected (14, 15) and in normotensive patients with PE (16). In those studies, quantitative H-FABP measurements have been performed either by a time-consuming enzyme linked immunosorbent assay (ELISA) without approval for clinical routine (14-16) or semiquantitatively by a point-of-care test (17, 18). More recently, an automated chemistry assay permitted rapid and precise measurement of H-FABP...
in clinical routine. However, it remains unknown whether it identifies normotensive patients at increased risk for an adverse outcome as reliably as the ELISA approach. Moreover, a positive H-FABP test included in a simple clinical score might further improve risk stratification and especially help to identify PE patients of the intermediate-risk group (18).

The aim of the present study was to investigate whether elevated H-FABP values measured with the new fully-automated immunoturbidimetric assay in comparison to ELISA have additive prognostic value with regard to 30-day outcome in addition to other risk factors. Furthermore, we aimed to validate a novel, fast and simple combined prognostic score integrating the information obtained from this assay for reliable identification of normotensive patients at increased risk for an adverse outcome and compared its performance with established risk prediction models such as the ESC model and the simplified Pulmonary Embolism Severity Index (sPESI).

Material and methods

Patient population and study design

We prospectively included and followed consecutive patients who were diagnosed with acute symptomatic PE (symptom onset, ≤4 weeks) at the University Hospital of Göttingen between October 2005 and August 2011. Of 319 patients with confirmed PE, 48 were not considered for further analysis because they met at least one of the following exclusion criteria: (i) haemodynamic instability (as described previously [16, 19]) (40 patients); (ii) incomplete data (2 patients); (iii) unexpected or accidental diagnosis of PE (patients undergoing diagnostic tests for another suspected disease), or PE coinciding with acute decompensation of left ventricular failure or acute myocardial infarction (4 patients); (iv) recurrent PE during the study period (only the first event was included; 2 patients). Thus, a total of 271 patients were eventually included in the study. There was an overlap of patients who were also included in our recent studies for investigation of the predictive value of H-FABP measured by ELISA (n=45; 16%) in normotensive patients with acute PE (16) and for derivation of a novel prognostic score using a point-of-care test for H-FABP (n=113; 42%) in normotensive patients with acute PE (18).

In all cases, blood was drawn immediately for measurement of baseline (admission) biomarker levels prior to further diagnostic workup. Following confirmation of the diagnosis, patients were asked to sign the informed consent form. Subsequently, complete data on baseline clinical, haemodynamic, and laboratory parameters were obtained using a standardised questionnaire by investigators unaware of the patients’ biomarker levels.

The diagnostic workup for patients with suspected acute PE complied with existing guidelines during the study period. Patients with high clinical (pre-test) probability of PE based on the standardised Wells Score and those with intermediate or low clinical probability and a positive D-dimer test underwent an imaging procedure, preferably multidetector-row (64-slice) CT, to confirm the diagnosis (244 patients; 90% of the study population). Alternatively, diagnosis was confirmed by ventilation/perfusion lung scan in 26 patients (10%) and by pulmonary angiography in one patient. A transthoracic echocardiogram was performed in 183 patients (68% of the study population) within 48 hours after admission. RV dysfunction (as described previously [14]) was diagnosed in 100 patients (55%). The sPESI was calculated as suggested by Jiménez et al. (12) and patients were classified into either a “low-risk” (0 points) or a “high-risk” (≥1 point(s)) group. Missing data were considered to be normal (12, 13, 19). Patients were stratified as recommended by the current ESC guideline (6) in patients with “intermediate-risk” if they had signs of RV dysfunction on echocardiography and/or elevated troponin T plasma concentrations on admission and in patients with “low-risk” if both tests were normal. Overall, stratification was feasible for 224 patients (83%) including all patients who were examined by echocardiography (n=183) and those without an echocardiogram, if they had elevated troponin values (n=41).

Thirty-day clinical follow-up data were obtained from all patients included in the study. An adverse 30-day outcome was defined as all-cause death or at least one of the following major complications: (i) need for catecholamine administration (except for dopamine at a rate of ≤5 µg/kg/minute [min]) to maintain adequate tissue perfusion and prevent or treat cardiogenic shock; (ii) mechanical ventilation; or (iii) cardiopulmonary resuscitation.

Biomarker levels (if determined for study purpose) were not communicated to the clinicians involved in the care of the study patients, and they were not used to guide patient management or to monitor the effects of treatment during the hospital stay or at any time during the follow-up period. Treatment decisions were made by the physicians caring for the patient and not influenced by the study protocol. The study protocol was approved by the ethics committee of the University of Göttingen, Germany.

Laboratory parameters and biomarker testing

Venous plasma and serum samples were collected on admission and immediately stored at −80°C. Samples were later analysed in batches after a single thaw. Plasma levels of H-FABP were measured by ELISA (HyCult Biotechnology, Uden, The Netherlands). The same samples were also analysed by an immunoturbidimetric chemistry assay (Randox Cardiology H-FABP; Randox Laboratories Ltd, Crumlin, UK) that enables the quantitative measurement of H-FABP via a range of non-proprietary automated clinical chemistry analysers. The assay utilises latex particles coated with monoclonal anti-H-FABP antibodies in order to generate turbidity, measured as an absorbance change at 659 nm using the Cobas Integra 800 analyser. It measures H-FABP in a range of 0.747–120 ng/ml. Elevated H-FABP levels were defined as concentrations ≥26 ng/ml as described previously (14-16).

Standard laboratory measurements were performed at the Department of Clinical Chemistry, University of Göttingen, using standard laboratory techniques. Cardiac troponin T plasma concentrations on admission were determined using a 4th generation (cTnT) and a highly sensitive (hsTnT) quantitative electrochemiluminescence immunoassay (Elecsys 2010 analyzer, Roche Diag-
nistics, Mannheim, Germany). Elevated levels were prospectively defined as ≥20.03 μg/l for cTnT and ≥14 pg/ml for hS-TnT (19, 20). Renal insufficiency was defined as a glomerular filtration rate (GFR) < 60 ml/min/1.73 m². GFR was calculated from the “four-variable” MDRD Study equation (21).

Statistical analysis
Using the modified Kolmogorov-Smirnov test (Lilliefors test), the continuous variables tested in the present study were found not to follow a normal distribution; therefore, they are presented as medians with corresponding 25th and 75th percentiles and were compared using the unpaired Mann-Whitney U-test. Categorical variables were compared using Fisher’s exact test. Receiver operating characteristics (ROC) curve analysis was used to determine the area under the curve (AUC) of H-FABP with regard to 30-day outcome. The optimal cut-off value for H-FABP measured by the immunoturbidimetric assay was defined as the cut-off value at which 0.5 x (sensitivity + specificity) was maximal using ROC analysis. The inter assay comparison of the immunoturbidimetric assay and the ELISA was performed by Bland-Altman plot. The prognostic relevance of baseline parameters (those suspected of being associated with an adverse outcome such as age, chronic heart failure, renal insufficiency, malignant disease, pulmonary disease, coronary artery disease, syncope, and tachycardia [defined as heart rate ≥100 beats per min]) with regard to an adverse 30-day outcome (binary) were estimated using multivariable logistic regression models with forward stepwise selection (inclusion criterion: p-value of the score-test ≤5%, exclusion criterion: p-value of the likelihood-ratio test ≥10%). The results are presented as odds ratios (OR) with the corresponding 95% confidence intervals (95% CI). Evidence of RV dysfunction on echocardiography was not included in these multivariable models due to the limited number of echocardiographic examinations performed. The two main hypotheses of this study are that elevated H-FABP values ≥6 ng/ml measured with (i) immunoturbidimetry and (ii) the ELISA have additive prognostic value with regard to 30-day outcome in addition to the other risk factors. To assess the two main questions both binary H-FABP levels measured with ELISA and immunoturbidimetry were entered into the multivariable logistic regression model described above in a second step. To evaluate which assay provides higher prognostic value for 30-day-outcome c-indices were used. For calculation of a novel score (18), elevated H-FABP values ≥6 ng/ml measured by immunoturbidimetry weighted 1.5 points, syncope 1.5 points, and tachycardia 2.0 points, and a cut-off value of 3.0 points was used (Table 1). To determine the additive prognostic relevance of the novel prognostic score with regard to an adverse 30-day outcome, it was entered into a multivariable logistic regression model together with other predictors identified by forward stepwise selection as described above (but without considering syncope and tachycardia in the first step). For comparison of the novel score with the sPESI and the ESC model, c-indices and the net reclassification improvement (NRI) were calculated (22). Of note, for the sPESI and the ESC model the OR could not be evaluated since none of the patients with a sPESI of 0 points or patients classified as “low-risk” by the ESC model had an adverse 30-day outcome.

The global significance level was set to α_{global}=5%. In order to test the main hypothesis, we performed a Bonferroni correction, resulting in local significance levels of α_{local}=2.5%. All tests were performed two-sided and all other tests are performed exploratory. Analyses were performed by R (version 2.15.1, The R Foundation for Statistical Computing c/o Institute for Statistics and Mathematics, Wirtschaftsuniversität Wien, Vienna, Austria) and the SPSS software (version 20.0, SPSS Inc., Chicago, IL, USA).
Results

Baseline clinical and laboratory findings

The baseline clinical characteristics of the study population are summarised in Table 2. Overall, 50 patients (18.5%) received early recanalisation treatment (thrombolysis, inclusion in the Pulmonary Embolism Thrombolysis – PEITHO study [23], or surgical embolectomy). In more detail, 22 patients (8.1%) received early (within 24 hours of diagnosis) thrombolysis, 25 patients (9.2%) were included in the PEITHO study and randomised in a double-blind fashion to placebo versus single-bolus tenecteplase. Moreover, seven patients (2.6%) underwent surgical embolectomy (of those, 4 patients after failure of initial thrombolysis).

The immunoturbidimetric assay yielded H-FABP values between 0.1 and 1134.36 ng/ml (median, 5.16 ng/ml) which were higher than those obtained from the ELISA method (0.39–164.58 ng/ml; median, 2.94 ng/ml). The correlation coefficient between both approaches was 0.71 (Spearman-Rho; p<0.001). The Bland-Altman plot demonstrated a good agreement of both assays for small H-FABP values, while the difference between both assays became larger for higher H-FABP values (Figure 1).

H-FABP measured by immunoturbidimetry is superior to levels measured by ELISA for predicting 30-day outcome

During the first 30 days, 20 patients (7.4%) had an adverse outcome: 12 patients died (of those, 5 directly related to PE), six underwent cardiopulmonary resuscitation, and 15 required catecholamines and/or mechanical ventilation. The calculated AUC for H-FABP levels on admission regarding 30-day outcome was 0.78 for the ELISA-based approach and 0.82 for values determined by the immunoturbidimetric assay (p=0.039; Table 3, Figure 2). Using ROC analysis, a concentration of 6.6 ng/ml was identified as the optimal cut-off level for H-FABP measured with the immunoturbidimetric assay in our study population. This value is similar to the previously proposed cut-off value of 6 ng/ml in PE patients [14-16], which was subsequently used as threshold for elevated H-FABP values. H-FABP values were ≥6 ng/ml in 56 patients (20.7%) if determined by ELISA and in 123 patients (45.4%) if determined by the immunoturbidimetric assay (p<0.001; Table 3). Only one patient with low H-FABP values on admission determined by the immunoturbidimetric assay had an unfavourable clinical course (this patient died due to a recurrent PE on day three after admission), whereas the ELISA measured normal values in 8 patients who had an adverse 30-day outcome. This translated into a higher negative predictive value (NPV) (99%) and sustainability (95%) for the fully automated H-FABP measurement compared to the ELISA approach (Table 3). In addition, patients with an adverse outcome had higher H-FABP values determined by either method. By ELISA, median H-FABP was 2.8 (25th–75th percentile, 1.0–5.0) ng/ml and 8.5 (4.0–13.4) ng/ml in patients without and with an adverse outcome, respectively (p<0.001); by the immunoturbidimetric assay, median H-FABP was 4.9 (2.6–9.0) vs 15.7 (8.5–24.1) ng/ml, respectively (p<0.001).

Validation of a novel fast combined prognostic score

Recently, our group demonstrated that a novel clinical score based on elevated H-FABP values (determined with a point-of-care test), syncope, and tachycardia provides superior prognostic information for the risk stratification of non-high-risk PE patients compared to the FAST score [19]. However, ELISA is not available in the majority of hospitals, whereas the immunoturbidimetric assay is. Therefore, we wanted to analyse whether the FAST score can be translated into a valid score if determined by the immunoturbidimetric assay (Figure 1).

Using univariable logistic regression, elevated H-FABP values determined by the immunoturbidimetric assay were associated with a nearly 27-fold increased risk for an adverse 30-day outcome (p<0.001) compared to a 7.1-fold increased risk (p<0.001) if measured by ELISA (Table 3). Other baseline parameters univariately found to predict an adverse outcome were tachycardia (OR 4.0 [1.5–10.3]; p=0.005), renal insufficiency (OR 4.3 [1.7–11.0]; p=0.002), RV dysfunction (on echocardiography; OR 7.2 [1.6–32.2]; p=0.011), and syncope (OR 4.5 [1.7–11.4]; p=0.002). When tested by multivariable analysis (including all variables suspected of being associated with an adverse outcome as described in Methods), the presence of a malignant disease, chronic heart failure, renal insufficiency, syncope, and tachycardia had an impact on 30-day outcome (Table 4). H-FABP determined by the immunoturbidimetric assay showed a significant additional impact on the 30-day outcome (p=0.017) as opposed to a non-significant (using Bonferroni correction with a significance level of αcor=2.5% as defined in Methods) impact if measured with the ELISA (p=0.030; Table 4). In direct comparison, the c-index for the multivariable model was 0.84 with the immunoturbidimetric assay and 0.82 with the ELISA indicating a superiority of the immunoturbidimetric assay regarding the prognostic value.

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pared to each parameter alone and comparable prognostic information as evidence of RV dysfunction on echocardiography (18) (Table 1). In the present study, we validated this clinical score using the immunoturbidimetric assay based on its superior performance over ELISA.

Table 3: Comparison of H-FABP values determined by ELISA or immunoturbidimetry.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>H-FABP (ELISA)</th>
<th>H-FABP (immunoturbidimetry)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Median (25th-75th percentile)</td>
<td>2.94 (1.13–5.37)</td>
<td>5.16 (2.68–9.75)</td>
</tr>
<tr>
<td>Range</td>
<td>0.39–164.58</td>
<td>0.10–1134.36</td>
</tr>
<tr>
<td>AUC regarding 30-day outcome</td>
<td>0.78 (0.68–0.89)</td>
<td>0.82 (0.74–0.91)</td>
</tr>
<tr>
<td>H-FABP ≥6 ng/ml</td>
<td>n= 56 (21%)</td>
<td>n= 123 (45%)</td>
</tr>
</tbody>
</table>

Adverse 30-day outcome (n= 20)
- Patients with H-FABP ≥6 ng/ml and adverse 30-day outcome: n= 12
- Sensitivity (95% CI): 60% (39–78) vs. 95% (76–99)
- Specificity (95% CI): 82% (77–87) vs. 59% (52–64)
- PPV (95% CI): 21% (12–34) vs. 15% (10–23)
- NPV (95% CI): 96% (93–98) vs. 99% (96–100)
- OR (95% CI; p-value): 7.1 (2.7–18.3; p<0.001) vs. 26.9 (3.5–203.8; p<0.001)

Abbreviations: H-FABP, heart-type fatty acid-binding protein; ELISA, enzyme linked immunosorbent assay; AUC, area under the curve; PPV, positive predictive value; NPV, negative predictive value; OR, odds ratio; CI, confidence interval.

Overall, 77 patients (28.4%) had a score above the predefined cut-off value of ≥3 points and of those, 17 patients (6.3%) had an adverse outcome while of 194 patients (71.6%) with a score <3 points only three patients (1.1%) had an unfavourable clinical course. Thus, the sensitivity and specificity were 85% and 76%, respectively, and the positive predictive value (PPV) and the NPV were 22% and 99%, respectively. Using ROC analysis, the AUC for the score was 0.82 (0.73–0.92) and the predefined cut-off value of 3 points was confirmed as optimal cut-off-value to distinguish between patients with an adverse and a favourable 30-day outcome.

Patients with a score ≥3 points had an 18-fold increased risk of an adverse 30-day outcome (OR 18.0 [5.1–63.7]; p<0.001). In comparison, evidence of RV dysfunction on echocardiography was associated with an only 7.2-fold increased risk of an adverse 30-day outcome (1.6–32.2; p=0.011). If the clinical score was entered into a multivariable model with malignant disease, chronic heart failure, and renal insufficiency (identified by multivariable logistic regression model using forward stepwise selection as described in Methods), the clinical score using the immunoturbidimetric assay provided "additive" prognostic information (OR 14.2; p<0.001; Table 5).

Of note, the results could also be confirmed if 113 patients (41.7%) who were included in the derivation cohort of the score (19) were excluded from the current analysis: in 158 patients (of those, 11 patients (7.0%) had an adverse 30-day outcome), univariable logistic regression analysis could demonstrate that a score of ≥3 points was associated with an adverse 30-day outcome (OR 8.5 [2.1–33.9]; p=0.002).

Additionally, we compared the performance of the novel score with the model proposed by the ESC and the sPESI.

The novel score stratified a lower proportion of patients to a higher risk class (77 patients, 28.4%; see above) compared to the ESC model (175 patients, 78.1%) and the sPESI (179 patients, 66.1%; p<0.001 each; Table 2). Moreover, the novel score was associated with a higher specificity and PPV compared to the ESC model and the sPESI (Table 6). This was also reflected by a positive likelihood ratio (LR) of 3.56 and negative LR 0.20, indicating high diagnostic evidence of the novel score (Table 6). Within the group of the patients classified as intermediate-risk by the ESC model, of 83 patients (37.1%) with RV dysfunction on echocardiography or troponin elevation, five patients (6.0%) had an adverse 30-day outcome, compared with 14 (15.2%) of 92 patients (41.1%) with RV dysfunction on echocardiography plus troponin elevation. In direct comparison, using univariable logistic regression analysis, the novel score (c-index, 0.81; OR 18.0 [5.1–63.7]; p<0.001) provided superior prognostic information for the identification of patients with an adverse 30-day outcome compared to the ESC model (c-index, 0.62) and the sPESI (c-index, 0.68). Finally, reclassification of patients stratified by the ESC model or the sPESI by the use of the novel score provided better risk prediction: 60% of the patients classified as intermediate-risk by the ESC model were reclassified as low-risk by the novel score and 4% of the patients classified as low-risk by the ESC model were reclassified as higher risk by the novel score (net reclassification improvement [NRI], 0.39 [0.21–0.56]; p<0.001; Table 7) and 63% of the
patients classified as high-risk by the sPESI were reclassified as low-risk by the novel score and 12% of the patients classified as low-risk by the sPESI were reclassified as higher risk by the novel score (NRI 0.24 [0.05–0.43]; p=0.012; ▶ Table 7).

**Discussion**

Our findings in 271 normotensive patients with acute PE can be summarised as follows: (i) H-FABP values were higher if measured with immunoturbidimetry compared to ELISA; however, Bland-Altman plot demonstrated a good agreement of both assays especially for small H-FABP values; (ii) only one patient with normal H-FABP concentrations determined by the immunoturbidimetric assay had an unfavourable clinical course (NPV, 99%; sensitivity 95%); (iii) H-FABP levels ≥6 ng/ml determined with immunoturbidimetry (and not if determined with ELISA) showed a significant additional impact on the 30-day outcome in multivariable analysis (OR 12.4); (iv) validation of a novel fast and simple combined prognostic score (▶ Table 1) (18) by the use of H-FABP measured by immunoturbidimetry provided additional prognostic information (OR 14.2); and finally (v) the prognostic information obtained from the novel prognostic score were superior to the ones obtained by the ESC model and the sPESI especially for identification of intermediate-risk patients.

The measurement of biomarkers for diagnosis or risk assessment by chemical laboratory analysers is a standard procedure in emergency units caring for patients with chest pain and dyspnoea, including patients with PE. Besides cardiac troponins and natriuretic peptides as valuable cardiac biomarkers in PE (8, 10, 11, 24-26), risk stratification can be further improved by H-FABP (14-18). Until recently, H-FABP was determined in these patients by the use of a time-consuming ELISA (14-16) without approval for clinical application, or by a semiquantitative point-of-care test (17, 18). A novel latex-enhanced immunoturbidimetric assay now allows fast and quantitative measurement of H-FABP and showed good clinical performance in a recent clinical and analytical evaluation (27). The CE-marked assay can be run on several clinical analysers with serum or plasma samples and has already been studied in patients with acute coronary syndromes for early diagnosis and the prediction of long-term mortality (28, 29).

In the present study, H-FABP values determined by immunoturbidimetry and ELISA showed a good agreement, particularly for H-FABP values below 20 ng/ml. The calculated optimal cut-off value for H-FABP measured by immunoturbidimetry in our patient cohort was 6.6 ng/ml and thereby similar to other studies (28) and other assays (16). Thus, the cut-off value of H-FABP appears stable and this may allow clear recommendations for its use in clinical routine. The prognostic value of the immunoturbidimetric assay was superior to the ELISA. More specifically, elevated H-FABP levels determined by both methods were associated with an unfavourable 30-day outcome; however, only H-FABP values determined by the immunoturbidimetric assay showed a significant additional impact on 30-day outcome besides other well-known risk factors. Given these findings, together with a larger AUC and c-index for the immunoturbidimetric assay, the novel assay simplifies the measurement of this biomarker and also seems to provide more reliable prognostic information than the ELISA method.

During the past years, it has become obvious that laboratory biomarkers alone may not be sufficient for risk stratification of acute PE. Therefore, a number of clinical scores, imaging modalities, and combination models have been developed to better distinguish intermediate- and low-risk patients (6, 12, 13, 30). In our study population, 92 patients (33.9%) had an sPESI of 0 points and importantly, none of them had an adverse 30-day outcome (NPV, 100%) which is in accordance with previous findings (19) and confirms the eligibility of the sPESI for identification of patients at low mortality risk and thus possible candidates for home treatment (30, 31).

Our group recently developed a novel simple clinical score based on H-FABP (initially determined semiquantitatively by a point-of-care test), Syncope, and Tachycardia (FAST score; ▶ Table 1), which indicated a nearly 12-fold increased risk for an
Table 6: Prognostic performance of risk prediction models with regard to an adverse 30-day outcome.

<table>
<thead>
<tr>
<th>Model</th>
<th>FAST score</th>
<th>ESC model</th>
<th>sPESI</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Sensitivity (95% CI)</strong></td>
<td>85 (64–95)%</td>
<td>100 (83–100)%</td>
<td>100 (84–100)%</td>
</tr>
<tr>
<td><strong>Specificity (95% CI)</strong></td>
<td>76 (71–81)%</td>
<td>24 (19–30)%</td>
<td>37 (31–43)%</td>
</tr>
<tr>
<td><strong>PPV (95% CI)</strong></td>
<td>22 (14–33)%</td>
<td>11 (7–16)%</td>
<td>11 (8–17)%</td>
</tr>
<tr>
<td><strong>NPV (95% CI)</strong></td>
<td>99 (96–100)%</td>
<td>100 (92–100)%</td>
<td>100 (95–100)%</td>
</tr>
<tr>
<td><strong>Positive LR (95% CI)</strong></td>
<td>3.56 (2.67–4.74%)</td>
<td>1.31 (1.22–1.42%)</td>
<td>1.58 (1.43–1.73%)</td>
</tr>
<tr>
<td><strong>Negative LR (95% CI)</strong></td>
<td>0.20 (0.07–0.56)</td>
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</tr>
</tbody>
</table>

Abbreviations: FAST, H-FABP; sPESI, simplified Pulmonary Embolism Severity Index; CI, confidence interval; PPV, positive predictive value; NPV, negative predictive value; LR, likelihood ratio.

Table 7: Reclassification of patients by the use of the FAST score.

<table>
<thead>
<tr>
<th>Model</th>
<th>FAST score &lt;3 points</th>
<th>FAST score ≥3 points</th>
<th>Reclassified</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Patients without an adverse 30-day outcome</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ESC model &quot;low-risk&quot;</td>
<td>47</td>
<td>2</td>
<td>4%</td>
</tr>
<tr>
<td>ESC model &quot;intermediate-risk&quot;</td>
<td>103</td>
<td>53</td>
<td>66%</td>
</tr>
<tr>
<td>sPESI 0 points</td>
<td>81</td>
<td>11</td>
<td>12%</td>
</tr>
<tr>
<td>sPESI ≥1 point(s)</td>
<td>110</td>
<td>49</td>
<td>69%</td>
</tr>
<tr>
<td><strong>Patients with an adverse 30-day outcome</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ESC model &quot;low-risk&quot;</td>
<td>0</td>
<td>0</td>
<td>---</td>
</tr>
<tr>
<td>ESC model &quot;intermediate-risk&quot;</td>
<td>2</td>
<td>17</td>
<td>11%</td>
</tr>
<tr>
<td>sPESI 0 points</td>
<td>0</td>
<td>0</td>
<td>---</td>
</tr>
<tr>
<td>sPESI ≥1 point(s)</td>
<td>3</td>
<td>17</td>
<td>15%</td>
</tr>
</tbody>
</table>

Abbreviations: FAST, H-FABP; syncope, and tachycardia; ESC, European Society of Cardiology; sPESI, simplified Pulmonary Embolism Severity Index.

What is known about this topic?
- Current guidelines emphasise the importance of early risk stratification of the prognostic heterogeneous group of normotensive patients with pulmonary embolism (PE) to guide therapeutic decision making. However, defining the optimal tool for identification of patients with an increased risk remains challenging.
- Heart-type fatty acid-binding protein (H-FABP) is a useful biomarker for risk stratification superior to established cardiac biomarkers; but its applicability has been limited by the lack of fully-automated assays for determination in clinical routine.

What does this paper add?
- H-FABP determined by a novel fully-automated immunoturbidimetric assay, which can be run on several clinical analysers, appears more suitable for identification of patients with an increased risk of an adverse 30-day outcome compared to ELISA.
- Moreover, a novel clinical prediction score (based on H-FABP, Syncope, and Tachycardia; FAST score) provides superior prognostic information compared to the model recommended by current guidelines of the European Society of Cardiology (ESC) and the simplified Pulmonary Embolism Severity Index (sPESI).
- Pending external validation, this study now provides the basis for routine clinical application of H-FABP – alone or integrated in a novel clinical prediction score – for risk assessment of normotensive patients with acute PE.

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Conflicts of interest
None declared.
References

23. The PEITHO Steering Committee. Single-bolus tenecteplase plus heparin compared with heparin alone for normotensive patients with acute pulmonary embolism who have evidence of right ventricular dysfunction and myocardial injury: rationale and design of the Pulmonary Embolism Thrombolysis (PEITHO) trial. Am Heart J 2012; 163: 33–38.