

**Sensitive cardiac troponin I predicts poor outcomes in pulmonary arterial
hypertension**

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ABSTRACT

Circulating cardiac troponins are markers of myocardial injury. We sought to determine if cardiac troponin I (cTnI), measured by a sensitive assay, is associated with disease severity and prognosis in pulmonary arterial hypertension (PAH).

cTnI was measured in 68 patients with PAH diagnostic category 1 in a research-based sensitive immunoanalyzer with a lower limit of detection of 0.008 ng/ml. The associations between cTnI and PAH severity and clinical outcomes were assessed using Chi square and Wilcoxon rank sum tests, Kaplan-Meier analysis, and Cox regression models.

cTnI was detected in 25% of patients. Patients with detectable cTnI had more advanced functional class symptoms, shorter 6-minute walk distance, more pericardial effusions, larger right atrial area, and higher B-type natriuretic peptide and C-reactive protein levels. 36-month transplant-free survival was 44% in patients with detectable cTnI versus 85% in those with undetectable cTnI. cTnI was associated with a 4.7 fold increased risk of death related to right ventricular failure or transplant (hazard ratio 4.74, 95% CI 1.89 - 11.89, $p < 0.001$), even when adjusted individually for known parameters of PAH severity.

Elevated plasma cTnI, even at subclinically detectable levels, is associated with more severe disease and worse outcomes in patients with PAH.

KEYWORDS

Biomarkers

Cardiac troponin

Outcomes

Prognosis

Pulmonary arterial hypertension

INTRODUCTION

Pulmonary arterial hypertension (PAH) is characterized by luminal obliteration and remodeling of the pulmonary vasculature leading to progressive increments in pulmonary vascular resistance. This leads to overload of the right ventricle (RV), right ventricular failure and premature death [1]. Despite impressive advancements in the understanding of the pathobiology of PAH and the introduction of targeted medical therapies, the mortality associated with this condition remains substantial [2]. While guidelines suggest how to tailor different therapeutic approaches according to the severity of the disease [3], risk stratification is difficult, and often relies on suboptimal tools like the assessment of symptoms via the New York Heart Association (NYHA) functional classification. Non-invasive biomarkers of the disease offer tremendous potential in aiding the difficult therapeutic decisions [4].

The detection of circulating cardiac troponins reflects myocardial necrosis, and is diagnostic of acute myocardial infarction in the presence of ischemic signs and symptoms [5]. In recent years it has been recognized that elevated cardiac troponins may be detected in other conditions, including stable coronary artery disease [6], left ventricular failure [7], chronic kidney disease [8], sepsis [9] and mixed critical care populations [10]. Elevated cardiac troponins have also been noted in situations of right ventricular overload, both acute as in acute pulmonary embolism [11], and chronic, as in pulmonary hypertension [12, 13]. Mechanisms other than myocardial necrosis may be responsible for this “troponin leak” in these clinical scenarios, even though detectable cardiac troponin is specific for ongoing myocardial damage and generally associated with worse prognosis.

In this study we sought to determine if cardiac troponin I (cTnI), measured with a

sensitive assay, is associated with severity and prognosis in diagnostic category 1 PAH patients treated with contemporary PAH-targeted therapy.

METHODS AND MATERIAL

Study population and clinical data collection

Patients were recruited from the Cleveland Clinic Pulmonary Vascular Program. Eligible subjects included patients with PAH diagnostic category 1 according to the updated Dana Point clinical classification [14]. Acute coronary syndromes and advanced renal disease defined as a serum creatinine more than 2 mg/dl were further exclusion criteria. Non-consecutive patients were enrolled after their peripheral blood was collected for study purposes. Venous blood was obtained at the time of right heart catheterization in 17 incident (newly diagnosed) patients, or during a routine clinical visit where six-minute walk distance (6MWD), NYHA functional class, Doppler echocardiography, B-type natriuretic peptide (BNP) measurements were performed in the remaining 51 patients with prevalent disease. Blood was processed and samples were stored in a biobank until biomarker measurements were performed. Time to clinical outcomes was determined since the time of blood sampling. The clinical outcome of interest was transplant-free survival. Events for this outcome were either lung transplantation or death by right ventricular (RV) failure. Patients who were alive at the end of follow up and those who died of causes other than RV failure were censored. The cause of death was determined by author GAH by careful review of the medical records and, when needed, discussions with the treating clinician. Clinical events and data were recorded in a prospective fashion in 48 patients, and retrospectively in the remaining 20. Six-minute

walk distance, NYHA functional class, echocardiographic data, serum creatinine and BNP levels were measured the same day as study blood sampling. Right heart catheterization data were obtained closest to study blood sampling. The median time between diagnostic right heart catheterization and blood sampling for study purposes was 240 days. Other data collected included demographic information, traditional cardiovascular risk factors (diabetes mellitus, systemic arterial hypertension, smoking status), coronary artery disease, serum creatinine levels and the use of PAH-targeted therapies. Healthy volunteers were used as controls. This study was approved by the Cleveland Clinic Institutional Review Board. All study subjects gave written informed consent.

Laboratory measurements

Plasma levels of cTnI were measured using the *STAT* Troponin I assay (Abbott Laboratories, Inc., Abbott Park, IL) in a research-based immunoanalyzer that provides a 3-decimal point readout from venous blood samples collected by EDTA-tubes. Levels \geq 0.03 ng/ml are considered to be diagnostic of myocardial infarction in the right clinical context. We have previously shown that this assay provides highly sensitive analytical measurement of cTnI with a limit of detection of 0.008 ng/mL [15]. However, the assay detected cTnI levels between 0.001 and 0.008 ng/mL in 1291 subjects out of a total of 3828 stable patients undergoing an elective cardiac evaluation. Importantly, cTnI levels between 0.001 and 0.008 ng/mL were associated with an increased risk of death, myocardial infarction or stroke, compared to levels below 0.001 ng/mL [15]. This assay has been also shown to be comparable to other high-sensitivity assays [16]. Thus, for the purpose of this study, we defined detectable cTnI as any level $>$ 0.001 ng/ml. In the PAH

cohort we also measured high sensitivity C-reactive protein (CRP) and high-density lipoprotein cholesterol (HDL-C) using the Abbott ARCHITECT platform. We also measured cardiac troponin T (cTnT) with the Elecsys Troponin T assay (Roche Diagnostics, Indianapolis, IN) in 62 out of the 68 patients. This assay provides a 2-decimal readout with a limit of detection of 0.01 ng/ml

Statistical Analysis

Demographic, clinical and laboratory data were summarized as frequency (%), and mean \pm standard deviation (SD). cTnI levels were compared between PAH patients and controls by means of Wilcoxon rank sum and Chi-square tests. Patient groups defined by cTnI status were compared with respect to categorical study variables using Chi-square and Fisher's exact test, and with respect to quantitative and ordinal variables using Wilcoxon rank sum tests. Correlations between cTnI and variables of interest were estimated by the Spearman correlation coefficient.

The association between transplant-free survival and cTnI was assessed using log-rank tests, with Kaplan-Meier estimation used to describe likelihood of the outcomes during the follow-up period. Patients who died of causes other than RV failure were censored for this analysis. Cox regression models were used to provide estimates of hazard ratios (HR) and 95% confidence intervals (CI) for transplant-free survival with respect to their associations with cTnI. Cox regression was also used in a multivariable fashion to explore the covariate-adjusted associations between transplant-free survival and cTnI. We adjusted for selected covariates individually since the sample size and number of events did not allow for building models with multiple covariates. Results for

covariate adjusted associations were summarized using Wald p-values and covariate-adjusted HR estimates. Analyses were performed using R version 2.8.1 [17].

RESULTS

Study population and cTnI levels

We studied 68 PAH patients (age 47 ± 13 , female 91%) and 29 controls (age 35 ± 6 , female 75%). PAH categories were as follows: 41 idiopathic PAH, 1 heritable PAH (hereditary hemorrhagic telangiectasia), and 26 associated PAH (20 connective tissue disease, 3 congenital heart disease, 2 chronic hemolytic anemia, and 1 portopulmonary). At the time of study enrollment, 51 patients (75%) were receiving PAH-targeted therapies: 15 intravenous (IV) prostacyclin, 7 phosphodiesterase inhibitor (PDE5i), 4 endothelin receptor antagonist (ERA), 13 IV prostacyclin plus ERA, 3 IV prostacyclin plus PDE5i, 5 ERA plus PDE5i, and 4 IV prostacyclin plus ERA plus PDE5i. The 17 patients naive to therapy at enrollment (25% of the cohort) were eventually treated as follows: 2 IV prostacyclin, 4 PDE5i; 4 IV prostacyclin plus PDE5i, 3 ERA plus PDE5i, 2 IV prostacyclin plus PDE5i; and 2 IV prostacyclin plus ERA plus PDE5i. Table 1 details functional, echocardiographic and hemodynamic characteristics of the PAH cohort. No PAH patient had diabetes, 15 (22%) had systemic hypertension, and 27 (30%) were either former or active smokers. Three patients (4%) had coronary artery disease, all of whom had non-detectable cTnI.

Seventeen PAH patients (25%) had detectable cTnI levels (above 0.001 ng/ml), compared to 1 control (3.4%) ($p = 0.011$). Two patients (2.9%) had cTnI levels between 0.001 and 0.008 ng/ml. Nine patients (13.2%) had cTnI levels between 0.009 and 0.029

ng/ml, as did the 1 control with detectable cTnI. Six patients (8.8%) had levels higher 0.030 ng/ml, none of whom had coronary artery disease. In patients with detectable cTnI, mean levels were 0.037 ± 0.009 ng/mL, with a range of 0.003 to 0.140 ng/mL. The detection rate for cTnT was only 4 out of 62 (6.5%, $p = 0.006$ for the comparison with the 25% detection rate of cTnI). All patients with detectable cTnT had also a detectable cTnI. Conversely, 12 patients with detectable cTnI had undetectable cTnT.

cTnI and PAH severity

cTnI concentrations had significant positive correlations with the NYHA functional class, right atrial (RA) area measured with echocardiography, and BNP and CRP levels (Table 2). There was a borderline positive correlation with RA pressure ($r = 0.24$, $p = 0.06$). Negative correlations with the 6MWD, the mixed venous oxygen saturation, and HDL-C levels ($r = -0.42$) were observed (Table 2).

Patients with detectable cTnI were more likely to have associated PAH, more advanced functional class symptoms, and more pericardial effusions; they also had shorter 6MWD, higher RA pressure, larger RA area, higher BNP and CRP levels, and lower HDL-C concentrations (Table 1). Both subgroups were similar in age, gender, heart rate, and systemic blood pressure, as well as proportion with systemic hypertension and tobacco use.

Clinical outcomes

22 (32.4%) patients died and 2 (2.9%) underwent lung transplantation. Nine out of 17 patients (53%) with detectable cTnI died, compared to 13 out of 51 (25%) with undetectable cTnI ($p = 0.041$). All of the 9 deaths in patients with detectable cTnI were due to RV failure. Among the 13 deaths with undetectable cTnI, 3 were secondary to

severe infections (2 pneumonias and 1 catheter-related blood stream infection), 1 was related to pancreatic cancer, and 1 could not be ascertained; the remainder 8 deaths were secondary to RV failure. There were no sudden deaths in either group. The proportion of transplant or death related to RV failure in patients with undetectable cTnI was 17.6% (9 out of 51) compared to 58.8% (10 out of 17) in patients with detectable cTnI ($p = 0.003$). Fifteen out of 58 patients (26%) with undetectable cTnT died of RV failure at the end of follow up, compared to 3 out of 4 patients (75%) with detectable cTnT ($p=0.07$).

Detectable cTnI was associated with a 4.7 fold increased risk of death related to RV failure or transplant: HR 4.74, 95% CI 1.89 - 11.89, $p < 0.001$) (Figure 1 and Table 3). In patients with detectable cTnI, 12-month, 24-month, and 36-month transplant-free survival was 64.7%, 52.9% and 44.1% respectively, compared to 96.1%, 90% and 84.7% in patients with undetectable cTnI. Table 3 shows that age, type of PAH, NYHA functional class, 6MWD, pericardial effusion, RAP, and BNP were associated with the risk of death by RV failure or transplant. We also found prognostic value for HDL-C and creatinine, but not for CRP. In this cohort, there was no association between prevalent versus incident cases and different PAH-targeted therapies with clinical outcomes (Table 3). The association between cTnI and transplant-free survival remained unchanged when adjusted individually for age, PAH subcategory (idiopathic vs. associated), NYHA functional class, 6MWD, the presence pericardial effusion, RA pressure, cardiac index, the use prostacyclin therapy, HDL-C, CRP, and creatinine concentrations (all $p < 0.05$) (Table 4). Adjustment for the presence of systemic HTN, coronary artery disease, statin therapy, and body mass index did not affect this association either. There was a significant interaction between cTnI and BNP ($p = 0.006$). Patients with both low BNP

levels and undetectable cTnI had longer transplant-free survival, while those with both detectable cTnI and supra-median BNP had the worst outcomes, but small numbers in each group do not allow for firm conclusions. .When analyzing all-cause mortality and transplantation, detectable cTnI was still highly predictive of worse outcomes (HR 3.21, 95% CI 1.40 – 7.37, p = 0.004). When excluding the 2 patients with cTnI levels between 0.001 and 0.008 ng/mL, detectable cTnI was still highly associated death by RV failure (HR 4.07, 95% CI 1.54 – 10.78, p<0.001) (see Figure and Table in online supplementary appendix).

DISCUSSION

The main finding of our study is that cTnI detected by a sensitive assay even at subclinical levels is associated with more severe disease and worse transplant-free survival in patients with PAH. The current study uses a sensitive troponin assay and provides the largest cohort with a homogenous category 1 PAH population treated with contemporary PAH-targeted therapies for the longest follow up time.

The elevated pulmonary vascular resistance that characterizes PAH leads to increase intramural RV tension and eventually decreased cardiac output, which in turn can lead to lower systemic pressures. This situation is conducive to RV myocardial ischemia and necrosis, which may account for the troponin leak observed in a quarter of our patients. Our data support this notion, as we observed more right heart chamber dilation and dysfunction in patients with detectable cTnI. Mechanisms other than myocardial ischemia are also possible. We found a positive correlation between cTnI and CRP levels, suggesting that inflammatory mechanism may play a role. In the present

study, a 1 SD increase in CRP levels was not associated with death by RV failure (Table 3). This contrasts with the report by Quarck et al showing decrease survival with CRP levels above the upper limit of normal of 5 mg/L [18]. We have previously reported a similar association with regards to clinical worsening, but not survival [19]. While differences in the statistical methods may account for these discrepancies, the relationship between CRP and clinical outcomes deserves further study.

The previous 2 studies of cardiac troponin in pulmonary hypertension are limited by a number of issues: the use of a lower sensitivity assay [12], the inclusion of heterogeneous populations with pulmonary hypertension secondary to chronic thromboembolic disease and interstitial lung disease [12, 13], low numbers of category 1 PAH [13], short term follow up [13] with small numbers of clinical events [13], and non-contemporary management of PAH [20]. In the present study, we used an assay that provides highly sensitive analytical measurement of cTnI with a 3-decimal-point read out, which has been found to be comparable to other high-sensitivity assays [16]. We detected cTnI in 25% of the patients with the sensitive assay, and cTnT in 6.5% of patients, compared to a detection rate for cTnT of 13-14% in the previous studies by the Polish group [12, 21], and 4.3% in a study of chronic thromboembolic pulmonary hypertension by Lankeit and colleagues [22]. Our lower detection rates for cTnT may be explained by the inclusion of a majority of patients receiving PAH-targeted therapies. Our findings for cTnT mirror the results of Lankeit and colleagues [22] (very low detection rate and poor outcomes in all patients with detectable cTnT), and suggest that using an assay with a 2-decimal readout for cTnT does not provide enough prognostic

sensitivity. We show that the association between cardiac troponins and poor outcomes extends to lower troponin levels not detected with assays in current clinical use.

We report on a cohort that includes only diagnostic category 1 PAH according to the recent classification [14], who were treated with contemporary PAH management [20]. The latter may explain the 52.9% 24-month survival rate in our cohort with detectable cTnI, compared to 29% in the report by Torbicki and colleagues published 8 years ago [12]. Remarkably, even with improved outcomes, cTnI was predictive of transplant-free survival in our cohort. The sensitive assay used for this study allowing for the detection of subclinical levels of cTnI may have contributed to our ability to show this predictive ability. A recent French study failed to show any association between cTnI and survival in patients with acutely worsened PAH, with the authors noting that more sensitive assays for troponin measurements should be used [23].

Detectable cTnI was associated with more severe PAH, as suggested by more advanced functional class symptoms, more pericardial effusions, shorter 6MWD, higher RA pressure, larger RA area, higher BNP and CRP levels, and lower HDL-C concentrations. The detection of cTnI by our sensitive assay carried a 4.7 fold increase risk of death related to RV failure or transplant, with 36-month transplant-free survival of 44.1%, compared to 84.7% in patients with undetectable cTnI. cTnI was predictive of worse outcomes when adjusted individually for several covariates known to influence survival in PAH, including age, idiopathic versus associated PAH, functional class, 6MWD, pericardial effusion, RA pressure, cardiac index, creatinine, CRP and HDL-C. As they both reflect RV strain, it is not surprising that we found a strong correlation between cTnI and BNP. Furthermore, we found a significant interaction between these

markers. The addition of cTnI to BNP may allow for better risk stratification however (Figure 2).

The study of a relatively small and heterogeneous PAH population, including idiopathic disease and cases associated with connective tissue disease constitutes a limitation of our study, one that is common to single center biomarker investigations. Nevertheless, when adjusted for PAH category (idiopathic versus associated disease), cTnI remained associated with worse outcomes. Another limitation of this study is the inclusion of incident and prevalent cases, where 75% of the patients were receiving PAH-targeted therapies when cardiac troponins were measured. While this may have biased the results and also explained the very low detection rate for cTnT, it is noteworthy that a more sensitive troponin assay still provided prognostic information in patients receiving contemporary PAH therapies. Before a novel biomarker can be recommended for widespread clinical use, it needs to be evaluated in a rigorous fashion [24]. While biologic plausibility certainly exists for the use of circulating troponins in PAH, and the available evidence suggests a potential role, further studies with larger numbers of patients are needed to confirm their utility. For example, a recent systematic review of the literature suggested that troponins may not be discriminative enough to be used in the risk stratification of patients with normotensive acute pulmonary embolism [25]. As cardiac troponin assays become more sensitive, cutoffs associated with meaningful clinical outcomes will have to be defined to avoid diagnostic confusion. For instance, in patients suspected of having acute myocardial infarction, the higher-sensitivity assays improve diagnostic sensitivity, but at the expense of lower specificity [16, 26]. Recently, cTnT has been detected in 25% of the general population [27] and 66% in people older than 65

years [28] using highly sensitive assays with a limit of detection of 0.003 ng/ml. Rates of detection are much lower in female and young people [27], relevant to patients afflicted with PAH.

In summary, using a sensitive assay we found a 25% detection rate for cTnI in a cohort of category 1 PAH. Detectable cTnI was associated with more severe PAH and worse clinical outcomes. The measurement of circulating cTnI with a sensitive assay may help clinicians to identify high risk patients that could benefit from more aggressive therapies.

REFERENCES

1. Bogaard HJ, Abe K, Vonk Noordegraaf A, Voelkel NF. The right ventricle under pressure: cellular and molecular mechanisms of right-heart failure in pulmonary hypertension. *Chest* 2009; 135(3): 794-804.
2. Humbert M, Sitbon O, Yaici A, Montani D, O'Callaghan DS, Jais X, Parent F, Savale L, Natali D, Gunther S, Chaouat A, Chabot F, Cordier JF, Habib G, Gressin V, Jing ZC, Souza R, Simonneau G. Survival in incident and prevalent cohorts of patients with pulmonary arterial hypertension. *Eur Respir J* 2010; 36(3): 549-555.
3. Badesch DB, Abman SH, Simonneau G, Rubin LJ, McLaughlin VV. Medical therapy for pulmonary arterial hypertension: updated ACCP evidence-based clinical practice guidelines. *Chest* 2007; 131(6): 1917-1928.
4. Warwick G, Thomas PS, Yates DH. Biomarkers in pulmonary hypertension. *Eur Respir J* 2008; 32(2): 503-512.
5. Morrow DA, Cannon CP, Jesse RL, Newby LK, Ravkilde J, Storrow AB, Wu AH, Christenson RH. National Academy of Clinical Biochemistry Laboratory Medicine Practice Guidelines: Clinical characteristics and utilization of biochemical markers in acute coronary syndromes. *Circulation* 2007; 115(13): e356-375.
6. Eggers KM, Lagerqvist B, Venge P, Wallentin L, Lindahl B. Persistent cardiac troponin I elevation in stabilized patients after an episode of acute coronary syndrome predicts long-term mortality. *Circulation* 2007; 116(17): 1907-1914.
7. Horwich TB, Patel J, MacLellan WR, Fonarow GC. Cardiac troponin I is associated with impaired hemodynamics, progressive left ventricular dysfunction, and increased mortality rates in advanced heart failure. *Circulation* 2003; 108(7): 833-838.

8. Apple FS, Murakami MM, Pearce LA, Herzog CA. Predictive value of cardiac troponin I and T for subsequent death in end-stage renal disease. *Circulation* 2002; 106(23): 2941-2945.
9. Ammann P, Fehr T, Minder EI, Gunter C, Bertel O. Elevation of troponin I in sepsis and septic shock. *Intensive care medicine* 2001; 27(6): 965-969.
10. Quenot JP, Le Teuff G, Quantin C, Doise JM, Abrahamowicz M, Masson D, Blettery B. Myocardial injury in critically ill patients: relation to increased cardiac troponin I and hospital mortality. *Chest* 2005; 128(4): 2758-2764.
11. Janata KM, Leitner JM, Holzer-Richling N, Janata A, Laggner AN, Jilma B. Troponin T predicts in-hospital and 1-year mortality in patients with pulmonary embolism. *Eur Respir J* 2009; 34(6): 1357-1363.
12. Torbicki A, Kurzyna M, Kuca P, Fijalkowska A, Sikora J, Florczyk M, Pruszczyk P, Burakowski J, Wawrzynska L. Detectable serum cardiac troponin T as a marker of poor prognosis among patients with chronic precapillary pulmonary hypertension. *Circulation* 2003; 108(7): 844-848.
13. Filusch A, Giannitsis E, Katus HA, Meyer FJ. High-sensitive troponin T: a novel biomarker for prognosis and disease severity in patients with pulmonary arterial hypertension. *Clin Sci (Lond)* 2010; 119(5): 207-213.
14. Simonneau G, Robbins IM, Beghetti M, Channick RN, Delcroix M, Denton CP, Elliott CG, Gaine SP, Gladwin MT, Jing ZC, Krowka MJ, Langleben D, Nakanishi N, Souza R. Updated clinical classification of pulmonary hypertension. *J Am Coll Cardiol* 2009; 54(1 Suppl): S43-54.

15. Tang WH, Wu Y, Nicholls SJ, Brennan DM, Pepoy M, Mann S, Pratt A, Van Lente F, Hazen SL. Subclinical myocardial necrosis and cardiovascular risk in stable patients undergoing elective cardiac evaluation. *Arteriosclerosis, thrombosis, and vascular biology* 2010; 30(3): 634-640.
16. Reichlin T, Hochholzer W, Bassetti S, Steuer S, Stelzig C, Hartwiger S, Biedert S, Schaub N, Buerge C, Potocki M, Noveanu M, Breidthardt T, Twerenbold R, Winkler K, Bingisser R, Mueller C. Early diagnosis of myocardial infarction with sensitive cardiac troponin assays. *The New England journal of medicine* 2009; 361(9): 858-867.
17. R Development Core Team (2008). R: A language and environment for statistical computing. R Foundation for Statistical Computing V, Austria. ISBN 3-900051-07-0, URL <http://www.R-project.org>.
18. Quarck R, Nawrot T, Meyns B, Delcroix M. C-reactive protein: a new predictor of adverse outcome in pulmonary arterial hypertension. *Journal of the American College of Cardiology* 2009; 53(14): 1211-1218.
19. Heresi GA, AYTEKIN M, Newman J, DiDonato J, Dweik RA. Plasma levels of high-density lipoprotein cholesterol and outcomes in pulmonary arterial hypertension. *American journal of respiratory and critical care medicine* 2010; 182(5): 661-668.
20. Galie N, Hoeper MM, Humbert M, Torbicki A, Vachiery JL, Barbera JA, Beghetti M, Corris P, Gaine S, Gibbs JS, Gomez-Sanchez MA, Jondeau G, Klepetko W, Opitz C, Peacock A, Rubin L, Zellweger M, Simonneau G, Vahanian A, Auricchio A, Bax J, Ceconi C, Dean V, Filippatos G, Funck-Brentano C, Hobbs R, Kearney P, McDonagh T, McGregor K, Popescu BA, Reiner Z, Sechtem U, Sirnes PA, Tendera M, Vardas P, Widimsky P, Sechtem U, Al Attar N, Andreotti F, Aschermann M, Asteggiano

R, Benza R, Berger R, Bonnet D, Delcroix M, Howard L, Kitsiou AN, Lang I, Maggioni A, Nielsen-Kudsk JE, Park M, Perrone-Filardi P, Price S, Domenech MT, Vonk-Noordegraaf A, Zamorano JL. Guidelines for the diagnosis and treatment of pulmonary hypertension: The Task Force for the Diagnosis and Treatment of Pulmonary Hypertension of the European Society of Cardiology (ESC) and the European Respiratory Society (ERS), endorsed by the International Society of Heart and Lung Transplantation (ISHLT). *European heart journal* 2009; 30(20): 2493-2537.

21. Fijalkowska A, Kurzyna M, Torbicki A, Szewczyk G, Florczyk M, Pruszczyk P, Szturmowicz M. Serum N-terminal brain natriuretic peptide as a prognostic parameter in patients with pulmonary hypertension. *Chest* 2006; 129(5): 1313-1321.

22. Lankeit M, Dellas C, Panzenbock A, Skoro-Sajer N, Bonderman D, Olschewski M, Schafer K, Puls M, Konstantinides S, Lang IM. Heart-type fatty acid-binding protein for risk assessment of chronic thromboembolic pulmonary hypertension. *Eur Respir J* 2008; 31(5): 1024-1029.

23. Sztrymf B, Souza R, Bertoletti L, Jais X, Sitbon O, Price LC, Simonneau G, Humbert M. Prognostic factors of acute heart failure in patients with pulmonary arterial hypertension. *Eur Respir J* 2010; 35(6): 1286-1293.

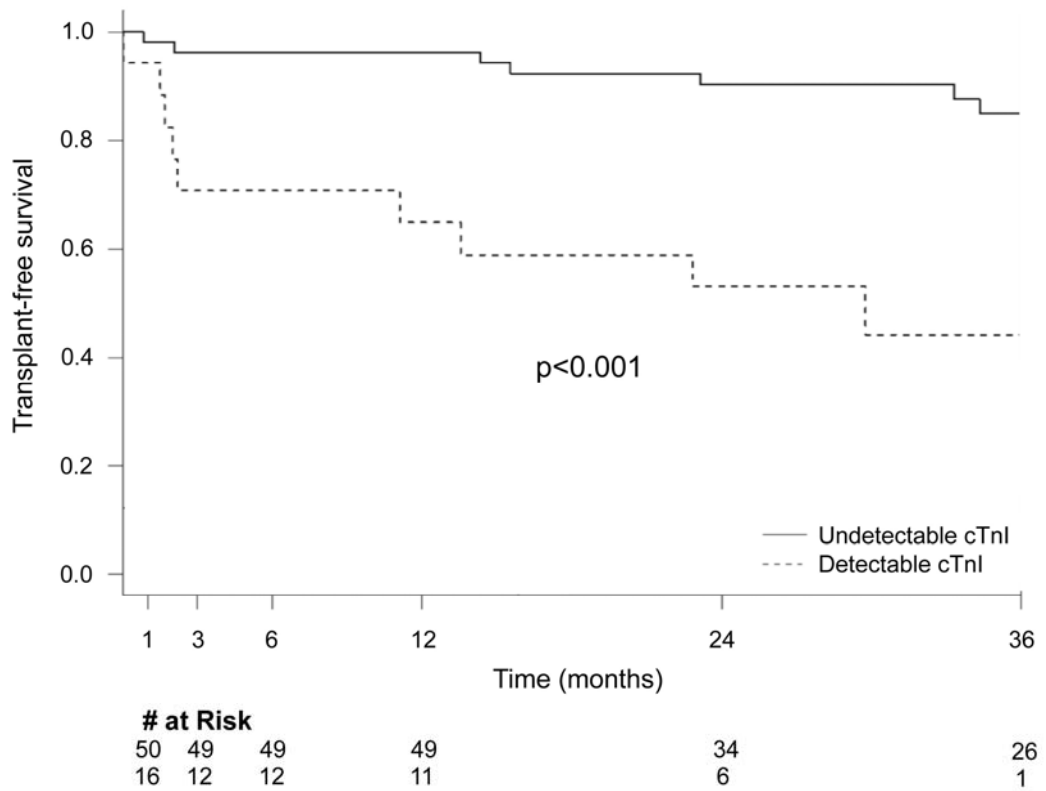
24. Heresi GA. Clinical perspective: biomarkers in pulmonary arterial hypertension. *International journal of clinical practice* 2011(169): 5-7.

25. Jimenez D, Uresandi F, Otero R, Lobo JL, Monreal M, Marti D, Zamora J, Muriel A, Aujesky D, Yusen RD. Troponin-based risk stratification of patients with acute nonmassive pulmonary embolism: systematic review and metaanalysis. *Chest* 2009; 136(4): 974-982.

26. Keller T, Zeller T, Peetz D, Tzikas S, Roth A, Czyz E, Bickel C, Baldus S, Warnholtz A, Frohlich M, Sinning CR, Eleftheriadis MS, Wild PS, Schnabel RB, Lubos E, Jachmann N, Genth-Zotz S, Post F, Nicaud V, Tiret L, Lackner KJ, Munzel TF, Blankenberg S. Sensitive troponin I assay in early diagnosis of acute myocardial infarction. *The New England journal of medicine* 2009; 361(9): 868-877.
27. de Lemos JA, Drazner MH, Omland T, Ayers CR, Khera A, Rohatgi A, Hashim I, Berry JD, Das SR, Morrow DA, McGuire DK. Association of troponin T detected with a highly sensitive assay and cardiac structure and mortality risk in the general population. *Jama* 2010; 304(22): 2503-2512.
28. deFilippi CR, de Lemos JA, Christenson RH, Gottdiener JS, Kop WJ, Zhan M, Seliger SL. Association of serial measures of cardiac troponin T using a sensitive assay with incident heart failure and cardiovascular mortality in older adults. *Jama* 2010; 304(22): 2494-2502.

FIGURE LEGENDS

Figure 1. Transplant-free survival according to cTnI status. Patients with detectable cTnI had a 4.7-fold higher risk of RV-related death or lung transplantation compared to patients with undetectable cTnI. cTnI = cardiac troponin I; RV = right ventricle.



TABLES

Table 1. Characteristics of the PAH cohort

	All patients	Undetectable cTnI	Detectable cTnI	p*
	n = 68	n=51	n=17	
Age, years	47 +/- 13	46 +/- 12	49 +/- 16	0.42
Female, n (%)	62 (91)	47 (92)	15 (88)	0.64
APAH, n (%)	27 (40)	16 (31)	11 (65)	0.019
NYHA III-IV, n (%)	44 (65%)	29 (57)	15 (88)	0.031
6MWD, meters	367 +/- 124	389 +/- 119	297 +/- 116	0.012
Pericardial eff, n (%)	22 (33)	12 (24)	10 (59)	0.011
RA area, cm ²	24 +/- 8	22 +/- 7	29 +/- 9	0.011
RA pressure, mmHg	11 +/- 6	10 +/- 6	13 +/- 6	0.06
mPAP, mmHg	52 +/- 14	51 +/- 12	56 +/- 17	0.35
CI, l/min/m ²	2.27 +/- 0.81	2.33 +/- 0.83	2.09 +/- 0.72	0.36
PVR, Wood units	11 +/- 7	10 +/- 6	14 +/- 10	0.24
BNP, pg/ml	224 +/- 314	155 +/- 280	454 +/- 321	<0.001
CRP, mg/l	12.7 +/- 20.9	7.8 +/- 8.6	27.5 +/- 35.8	0.007
HDL-C, mg/dl	35 +/- 11	38 +/- 10	27 +/- 11	0.001
Creatinine, mg/dl	0.90 +/- 0.26	0.89 +/- 0.24	0.94 +/- 0.32	0.58

APAH = associated pulmonary arterial hypertension; BNP = B-type natriuretic peptide; cTnI = cardiac troponin I; CI = cardiac index; CRP = C-reactive protein; eff = effusion; HDL-C = high-density lipoprotein cholesterol; IPAH = idiopathic pulmonary arterial hypertension, mPAP = mean pulmonary artery pressure; NYHA = New York Heart Association functional class; PAH = pulmonary arterial hypertension; PVR = pulmonary vascular resistance; RA = right atrium; 6MWD = six-minute walk distance.

* p for the comparison between detectable and undetectable cTnI

Table 2. Significant correlations between cTnI and PAH indices of severity

Correlation	Variable	R	p
Positive	BNP	0.45	<0.001
	NYHA class	0.36	0.002
	CRP	0.34	0.004
	RA area	0.36	0.010
Negative	HDL-C	-0.42	<0.001
	6MWD	-0.29	0.020
	MVO ₂	-0.30	0.034

BNP = B-type natriuretic peptide; cTnI = cardiac troponin I; CI = cardiac index; HDL-C = high-density lipoprotein cholesterol; MVO₂ = mixed venous oxygen saturation; NYHA = New York Heart Association functional class; RA = right atrium; RV = right ventricle; 6MWD = six-minute walk distance.

Table 3. Predictors of the risk of transplant or death by right ventricular failure.

Variable	Univariable HR (95% CI)	p
Detectable cTnI	4.74, (1.89 - 11.89)	<0.001
Age, years*	1.70 (1.01 – 2.87)	0.048
IPAH vs. APAH	0.39 (0.15 – 0.99)	0.048
NYHA class	3.55 (1.03 – 12.2)	0.045
6MWD, meters*	0.60 (0.37 – 0.97)	0.038
Pericardial effusion	3.21 (1.28 – 8.06)	0.013
RAP, mmHg*	2.17 (1.30 – 3.61)	0.003
CI, l/min/m ² *	1.00 (0.64 – 1.56)	0.99
CRP, mg/l*	1.17 (0.83 – 1.63)	0.37
HDL-C, mg/dl*	0.45 (0.28 – 0.71)	0.001
Creatinine, mg/dl*	1.50 (1.05 – 2.14)	0.027
BNP*	2.01 (1.39 – 2.90)	<0.001
Prevalent vs. Incident disease	1.39 (0.45 – 4.22)	0.57
Prostacyclin	1.28 (0.49 – 3.30)	0.61
ERA	1.18 (0.48 – 2.91)	0.72
PDE5i	0.63 (0.25 – 1.59)	0.32
# PAH medications*	0.99 (0.62 – 1.57)	0.96

APAH = associated pulmonary arterial hypertension; BNP = B-type natriuretic peptide;

cTnI = cardiac troponin I; CI = cardiac index; CRP = C-reactive protein; ERA =

endothelin receptor antagonists; HR = hazard ratio; HDL-C = high-density lipoprotein

cholesterol; IPAH = idiopathic pulmonary arterial hypertension, NYHA = New York

Heart Association functional class; PDE5i = phosphodiesterase -5 inhibitors; RAP = right atrial pressure; 6MWD = six-minute walk distance.* For continuous variables, HRs reported corresponding to a 1SD increase in the variable.

Table 4. cTnI and risk of transplant or RV-death when adjusted individually for selected covariates.

Covariate	cTnI adjusted HR (95% CI)	p
Age, years	4.36 (1.72 - 11.02)	0.002
IPAH vs APAH	4.06 (1.57 - 10.53)	0.004
NYHA class	3.84 (1.48 - 9.96)	0.006
6MWD, meters	3.02 (1.07 - 8.53)	0.040
Pericardial effusion	3.81 (1.49 - 9.75)	0.005
RAP, mmHg	3.91 (1.47 - 10.39)	0.006
CI, l/min/m ²	4.75 (1.89 - 11.91)	0.001
Prostacyclin therapy	4.80 (1.91 - 12.06)	0.001
CRP, g/L	3.80 (1.47 - 9.84)	0.006
HDL-C, mg/dl	2.88 (1.04 - 7.97)	0.042
Creatinine, mg/dl	3.88 (1.46 - 10.31)	0.007

APAH = associated pulmonary arterial hypertension; cTnI = cardiac troponin I; CI = cardiac index; CRP = C-reactive protein; HDL-C = high-density lipoprotein cholesterol; IPAH = idiopathic pulmonary arterial hypertension, mPAP = mean pulmonary artery pressure; NYHA = New York Heart Association functional class; RA = right atrium; RV = right ventricle; 6MWD = six-minute walk distance.