Original Paper

Troponin I, but not BNP, is Associated with Phosphorus, Calcium and Vitamin D in Stable Coronary Artery Disease

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Key Words

Troponin • Phosphorus • Calcium • Vitamin D • BNP

Abstract

Background: Elevated plasma cardiac troponin, elevated plasma phosphorus and decreased plasma vitamin D have been shown to be associated with negative outcomes. **Methods and results:** Troponin I, calcium, phosphorus and 25-OH vitamin D were studied in a cohort of 60 patients with stable coronary heart disease and preserved left ventricular function. Using a cut-off value of 0.012 ng/mL for plasma troponin I, patients with higher values (18 patients), when compared to the other patients (n=42), had higher mean values for plasma phosphorus (3.42 ± 0.45 mg/dL vs 3.17 ± 0.45 mg/dL, $p = 0.041$) and calcium (5.08 ± 0.23 mEq/L vs 4.92 ± 0.18 mEq/L, $p = 0.016$) and lower values for 25-OH vitamin D (14.2 ± 5.6 ng/mL vs 19.4 ± 8.8 ng/mL, $p = 0.032$). Binary logistic regression analysis showed that troponin I > 0.012 ng/mL is associated with increased phosphorus, increased calcium and decreased 25-OH vitamin D concentrations. A similar analysis using BNP > 100 pg/mL failed to show significant associations with phosphorus, calcium and 25-OH vitamin D concentrations. **Conclusions:** In patients with stable coronary artery disease and preserved left ventricular function, those having cardiac troponin I > 0.012 ng/mL, but not those having BNP > 100 pg/mL, had higher plasma phosphorus, higher plasma calcium and lower plasma 25-OH vitamin D concentrations than those having cardiac troponin I ≤ 0.012 ng/mL (or BNP ≤ 100 pg/mL).

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Introduction

Plasma cardiac troponin and brain natriuretic peptide (BNP) are major cardiac biomarkers. Cardiac troponin has been shown to be elevated due to myocardial necrosis in myocardial infarction [1], but is also elevated in other acute and chronic settings [2, 3]. Brain natriuretic peptide is a recognized marker for cardiac dysfunction with fluid overload [4].

In patients with stable coronary artery disease, plasma cardiac troponin has been shown to be associated with the incidence of cardiovascular death and heart failure, but not with myocardial infarction, [5]. Similar findings were described in a cohort of adults aged 65 years or older without prior heart failure [6].

In a cohort of individuals with prior myocardial infarction, serum phosphate was shown to be associated with total mortality, new heart failure, new myocardial infarction, but not to the risk of stroke [7]. In the Framingham Offspring study, an increased cardiovascular disease risk was associated with serum phosphorus, but not with serum calcium [8].

Low levels of 25-OH vitamin D were shown to be associated with all-cause and cardiovascular mortality [9]. Vitamin D deficiency has been presented as a possible cardiovascular risk factor [10].

In the present report, plasma cardiac troponin I and BNP were studied in a cohort of 60 patients with stable coronary artery disease and preserved left ventricular function, in search for a possible association to plasma phosphorus, calcium and 25-OH vitamin D levels.

Materials and Methods

A cohort of 60 patients (41 male, 19 female) with coronary heart disease, aged 65.5 years (60.0-73.8; median and interquartile range), was the subject of an observational study. All patients were Caucasian. Patients were observed at an outpatient Cardiology clinic. The study had an observational character.

The research protocol (including the informed consent forms) was evaluated and approved by an independent ethics committee (Comissão de Ética do Centro de Saúde São João, Porto, Portugal), as previously reported [11]. All participants provided a written informed consent, a consent procedure approved by the ethics committee. All consent forms were kept on file. All calculations were made using a database with anonymous data.

Diagnosis was established by coronary angiography, except in one case (with angina, left bundle branch block and suggestive findings in cardiac perfusion imaging). Angiography was performed after myocardial infarction or during assessment of angina pectoris. Exclusion criteria included serious systemic diseases, such as cancer and systemic infections. All patients included in the study had left ventricular ejection fraction, evaluated by echocardiography and/or nuclear scan, greater or equal to 45%.

Patients were evaluated using a protocol which included electrocardiogram, echocardiogram, and plasma levels of creatinine, calcium, phosphorus, 25-OH vitamin D, cardiac troponin I and brain natriuretic peptide (BNP). Biochemical measurements were performed under routine conditions, immediately after collection of blood and blinded for the other patient characteristics. Additional diagnostic tests were performed if indicated.

Troponin I was measured using the ARCHITECT STAT system, of Abbott Diagnostics (Abbott Park, Illinois, USA). The 99th percentile of troponin I in a normal population with this assay was established at 0.012 ng/mL. All troponin I values <0.01 ng/mL (detection limit) were taken as 0.009 ng/mL for the purpose of calculation.

Estimated glomerular filtration rate (eGFR; mL/min per 1.73 m²) was calculated according to the (updated) Modification of Diet in Renal Disease (MDRD) study equation: $GFR = 175 \times (\text{serum creatinine})^{-1.154} \times (\text{age})^{-0.203} \times 0.742$ (in women) [12] (IDMS-traceable assay).

Statistical analysis

Data are presented as either median and interquartile range (whole group) or arithmetic mean and standard deviation (sub-group analysis). Correlations between the various parameters under study were calculated by using the Spearman correlation coefficient (r_s). Pairs of means were compared by using the Mann-Whitney U test. P-values <0.05 were considered significant.

Binary logistic regression, taking as dependent variable troponin I (cut-off value at 0.012 ng/mL) and as independent variables plasma eGFR, phosphorus, calcium, and 25-OH vitamin D (variables entered in the model in this order), was performed. Statistical calculations were made by using the Statistical Package for the Social Sciences software (SPSS, version 20, IBM Corp., Armonk, NY, USA).

Table 1. Median and interquartile range for a number of parameters measured in 60 patients with coronary heart disease

	Median	Interquartile range
Age (years)	65.5	60.0-73.8
Troponin I (ng/mL)	0.009	0.009-0.014
Brain natriuretic peptide (pg/mL)	58.4	31.3-106.4
Phosphorus (mg/dL)	3.30	2.90-3.60
Calcium (mEq/L)	5.00	4.80-5.10
25-OH vitamin D (ng/mL)	17.5	12.3-23.5
Estimated glomerular filtration rate (mL/min per 1.73 m ²)	83.2	66.1-96.8

Table 2. Plasma values for calcium (mEq/L), phosphorus (mg/dL), 25-OH vitamin D (ng/mL) and estimated glomerular filtration rate (eGFR; mL/min per 1.73 m²), in 60 stable

outpatients with coronary heart disease, divided by high and low plasma levels of troponin I and BNP. Data are expressed as means $\pm$ standard deviation. p – probability (Mann-Whitney U test)

	Troponin I ≤ 0.012 ng/mL (n=42)	Troponin I > 0.012 ng/mL (n=18)	BNP ≤ 100 pg/mL (n=41)	BNP > 100 pg/mL (n=19)
Calcium	4.92 $\pm$ 0.18	5.08 $\pm$ 0.23 (p 0.016)	4.96 $\pm$ 0.19	4.98 $\pm$ 0.25 (p 0.83)
Phosphorus	3.17 $\pm$ 0.45	3.42 $\pm$ 0.45 (p 0.041)	3.19 $\pm$ 0.50	3.37 $\pm$ 0.34 (p 0.11)
25-OH Vitamin D	19.4 $\pm$ 8.8	14.2 $\pm$ 5.6 (p 0.032)	18.6 $\pm$ 7.5	16.2 $\pm$ 9.9 (p 0.25)
eGFR	83.2 $\pm$ 18.0	80.2 $\pm$ 21.4 (p 0.33)	83.7 $\pm$ 16.5	79.4 $\pm$ 23.7 (p 0.26)

Results

Median values for the parameters under study are shown in Table 1. Cardiac troponin I was ≤ 0.012 ng/mL in 42 patients and even below detection in 34 cases. BNP levels > 100 pg/mL were seen in 19 cases (Table 2).

In the 18 patients having troponin I > 0.012 ng/mL, plasma values of phosphorus, calcium, and 25-OH vitamin D differed significantly from those in the remaining 42 patients (Table 2).

As can be seen in Fig. 1, no plasma troponin I values > 0.020 ng/mL were seen in patients with a plasma phosphorus ≤ 3.3 mg/dL, or with a plasma 25-OH vitamin D level > 20 ng/mL.

Binary logistic regression study, taking troponin I as dependent variable (cut-off level at 0.012 ng/mL), and eGFR, calcium, phosphorus and 25-OH vitamin D as independent variables, yielded significant contributions for phosphorus (p= 0.021), calcium (p= 0.026) and 25-OH vitamin D (p= 0.026), but not for eGFR (p= 0.688).

Concerning BNP, no significant difference was seen in the same parameters, however a trend in the same direction was noted for phosphorus (Table 2). Regression study similar to the one described for troponin I yielded negative results (data not shown). Troponin I and BNP were not significantly correlated ($r_s = 0.05$, p= 0.69). Troponin I was significantly correlated to calcium ($r_s = 0.31$, p= 0.02) and to 25-OH vitamin D

Fig. 1. Relations between plasma phosphorus and cardiac troponin I (top), and between 25-OH vitamin D and cardiac troponin I (bottom) in 60 outpatients with stable coronary artery disease.

($r_s = -0.30$, $p = 0.02$), but not to phosphorus ($r_s = 0.21$, $p = 0.11$). Phosphorus was not significantly correlated to either calcium ($r_s = -0.02$, $p = 0.88$) or 25-OH vitamin D ($r_s = 0.08$, $p = 0.53$).

Discussion

In a cohort of 60 patients with stable coronary heart disease and preserved left ventricular function, those having troponin I > 0.012 ng/mL had higher levels of plasma phosphorus and calcium, and lower levels of 25-OH vitamin D than those having troponin I ≤ 0.012 ng/mL. Binary logistic regression analysis also showed an association between troponin I and the three plasma parameters.

Cardiac troponin I plasma levels were initially studied in the context of acute myocardial infarction [1]. However, subsequent research showed that elevated plasma troponin I was often seen in several different acute and chronic contexts, leading to the view that cardiac strain could be the mechanism for increased plasma troponin I in some cases [2, 13, 14]. Hessel *et al.* [15] showed that intact troponin I can be released from viable cardiomyocytes by stimulation of stretch-responsive integrins. More recently, using a high-sensitivity assay for plasma troponin T, Omland *et al.* showed that cardiac troponin T concentrations were significantly associated with the incidence of cardiovascular death and heart failure but not with myocardial infarction in patients with stable coronary artery disease [5]. deFilippi *et al.* showed incident heart failure and cardiovascular death to be associated with troponin T values in a cohort of adults aged 65 years or older without prior heart failure [6].

In a cohort of patients with prior myocardial infarction serum phosphate elevations were associated with increased risks of total mortality, new heart failure, new myocardial infarction, but not of stroke [7]. In the Framingham Offspring study, an increased cardiovascular disease risk was shown by Dhingra *et al.* to be associated with serum phosphorus, but not to serum calcium [8].

On the other hand, Dobnig *et al.* showed low levels of 25-OH vitamin D to be associated with all-cause and cardiovascular mortality [9].

Phosphate excretion in the kidney is regulated by fibroblast growth factor 23 (FGF-23), interacting with a cofactor, klotho. Kuro-o *et al.* have shown mutated klotho gene to cause a syndrome resembling human ageing, including a short lifespan, infertility, arteriosclerosis, skin atrophy, osteoporosis and emphysema [16]. Holden *et al.*, studying patients with chronic kidney disease, have shown troponin T to be associated with FGF-23 [17]. In patients with chronic renal failure, serum FGF-23 concentrations were correlated with serum phosphate concentrations [18]. In the present investigation, the hypothesis is raised that deleterious effects of higher phosphorus and/or calcium and of lower vitamin D levels on the heart are reflected by an increase in plasma troponin I. The precise mechanisms underlying this possible association are not yet fully elucidated. Renal insufficiency could be viewed as a possible mechanism, however the data now presented does not support this possibility. An impact of cardiac integrins on mechanisms involving phosphorus/ calcium/ vitamin D [15, 19] would be another theoretical possibility. This phenomenon needs further study in larger samples of individuals both with and without coronary artery disease.

BNP levels were not shown to be associated with phosphorus/calcium/vitamin D in this cohort, leading to the view that volume overload and phosphorus/calcium/vitamin D metabolism could be relatively independent phenomena.

Study limitations – the major limitation of this study is the limited number of patients under study; another limitation, in what concerns calculations with troponin I, is the relatively high detection limit causing undetectable troponin I levels in 34 patients.

Conclusion

In patients with stable coronary artery disease having preserved left ventricular function, plasma levels of troponin I > 0.012 ng/mL, but not those having BNP > 100 pg/mL, had higher plasma phosphorus, higher plasma calcium, and lower plasma 25-OH vitamin D concentrations than those having troponin I ≤ 0.012 ng/mL (or BNP ≤ 100 pg/mL).

Conflict of Interests

None.

References

- 1 Antman EM, Tanasijevic MJ, Thompson B, Schactman M, McCabe CH, Cannon CP, Fischer GA, Fung AY, Thompson C, Wybenga D, Braunwald E: Cardiac-specific troponin I levels to predict the risk of mortality in patients with acute coronary syndromes. *N Engl J Med* 1996;335:1342-1349.
- 2 Nunes JPL: Cardiac troponin I in systemic diseases. A possible role for myocardial strain. *Rev Port Cardiol* 2001;20:785-788.
- 3 Thygesen K, Alpert JS, Jaffe AS, Simoons ML, Chaitman BR, White HD: Third universal definition of myocardial infarction. *J Am Coll Cardiol* 2012;60:1581-1598.
- 4 Yoshimura M, Yasue H, Okumura K, Ogawa H, Jougasaki M, Mukoyama M, Nakao K, Imura H: Different secretion patterns of atrial natriuretic peptide and brain natriuretic peptide in patients with congestive heart failure. *Circulation* 1993;87:464-469.
- 5 Omland T, de Lemos JA, Sabatine MS, Christophi CA, Rice MM, Jablonski KA, Tjora S, Domanski MJ, Gersh BJ, Rouleau JL, Pfeffer MA, Braunwald E: A sensitive cardiac troponin T assay in stable coronary artery disease. *N Engl J Med* 2009;361:2538-2547.
- 6 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;304:2494-2502.
- 7 Tonelli M, Sacks F, Pfeffer M, Gao ZW, Curhan G: Relation between serum phosphate level and cardiovascular event rate in people with coronary disease. *Circulation* 2005;112:2627-2633.
- 8 Dhingra R, Sullivan LM, Fox CS, Wang TJ, D'Agostino RB Sr, Gaziano JM, Vasan RS: Relations of serum phosphorus and calcium levels to the incidence of cardiovascular disease in the community. *Arch Intern Med* 2007;167:879-885.
- 9 Dobnig H, Pilz S, Scharnagl H, Renner W, Seelhorst U, Wellnitz B, Kinkeldei J, Boehm BO, Weihrauch G, Maerz W: Independent association of low serum 25-hydroxyvitamin D and 1,25-dihydroxyvitamin D levels with all-cause and cardiovascular mortality. *Arch Intern Med* 2008;168:1340-1349, 2008.
- 10 Lee JH, O'Keefe JH, Bell D, Hensrud DD, Holick MF: Vitamin D deficiency: an important, common, and easily treatable cardiovascular risk factor? *J Am Coll Cardiol* 2008;52:1949-1956.
- 11 Nunes JPL, Silva JC: Systemic correlates of angiographic coronary artery disease. *Plos One* 2009;4:e4322.
- 12 Levey AS, Coresh J, Greene T, Marsh J, Stevens LA, Kusek JW, Van Lente F: Expressing the Modification of Diet in Renal Disease Study equation for estimating glomerular filtration rate with standardized serum creatinine values. *Clin Chem* 2007;53:766-772.
- 13 Logeart D, Beyne P, Cusson C, Tokmakova M, Leban M, Guiti C, Bourgoin P, Solal AC: Evidence of cardiac myolysis in severe nonischemic heart failure and the potential role of increased wall strain. *Am Heart J* 2001;141:247-253.
- 14 Nunes JPL, Macedo F: An analytical triad for the diagnosis of pulmonary embolism. *Cardiology* 2000;94:264.
- 15 Hessel MH, Atsma DE, van der Valk EJ, Bax WH, Schali J MJ, van der Laarse A: Release of cardiac troponin I from viable cardiomyocytes is mediated by integrin stimulation. *Pflugers Arch* 2008;455:979-986.
- 16 Kuro-o M, Matsumura Y, Aizawa H, Kawaguchi H, Suga T, Utsugi T, Ohyama Y, Kurabayashi M, Kaname T, Kume E, Iwasaki H, Iida A, Shiraki-Iida T, Nishikawa S, Nagai R, Nabeshima YI: Mutation of the mouse *klotho* gene leads to a syndrome resembling ageing. *Nature* 1997;390:45-51.
- 17 Holden RM, Beseau D, Booth SL, Adams MA, Garland JS, Morton RA, Collier CP, Foley RN: FGF-23 is associated with cardiac troponin T and mortality in hemodialysis patients. *Hemodial Int* 2012;16:53-58.
- 18 Unsal A, Kose Budak S, Koc Y, Basturk T, Sakaci T, Ahbap E, Sinangil A: Relationship of fibroblast growth factor 23 with left ventricle mass index and coronary calcification in chronic renal disease. *Kidney Blood Press Res* 2012;36:55-64.
- 19 Brancaccio M, Hirsch E, Notte A, Selvetella G, Lembo G, Tarone G: Integrin signalling: the tug-of-war in heart hypertrophy. *Cardiovasc Res* 2006;70:422-433.