The association between plasma furin and cardiovascular events after acute myocardial infarction

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Research article

Keywords: Furin, acute myocardial infarction, major adverse cardiac events.

DOI: https://doi.org/10.21203/rs.3.rs-41696/v2

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Abstract

Background: Furin is the key enzyme to cleave pro-BNP and plays a critical role in the cardiovascular system through its involvement in the lipid metabolism, blood pressure and formation of atheromatous plaques. NT-proBNP and recently corin, which is also a key enzyme to cleave pro-BNP, have been approved as predictors of prognosis after acute myocardial infarction (AMI). We here conducted this cohort study to investigate the relationship between plasma furin and the prognosis outcome in patients after AMI.

Methods: We enrolled 1100 AMI patients and measured their plasma furin concentration. The primary endpoint was the major adverse cardiac events (MACE), a composite of cardiovascular (CV) death, non-fatal myocardial infarction or non-fatal stroke. The association of plasma furin concentration with AMI outcomes was explored by using Kaplan–Meier curve and multivariate Cox regression analysis.

Results: Our results showed that slight increase of mean cTNT in patients with higher furin concentration (P=0.016). Over a median follow-up of 31 months, multivariate Cox regression analysis suggested that plasma furin was not associated with MACE (HR: 1.01; 95% CI: 0.93-1.06; P=0.807) after adjustment for potential conventional risk factors. However, plasma furin was associated with non-fatal MI (HR: 1.09; 95% CI: 1.01-1.17; P=0.022) after fully adjustment. Subgroup analysis indicated no relationship between plasma furin and MACE in different subgroup populations.

Conclusions: Our study demonstrated that plasma furin was not associated with risk of MACE and may not be used as a predictor of poor prognosis after AMI. But higher levels of plasma furin may be associated with higher risk of non-fatal MI.

Introduction

Cardiovascular diseases (CVD) remain a major cause of premature death and chronic disability for all regions in the world[1]. Acute myocardial infarction is one of the severe CVD. To help select treatment strategy for AMI patients at an early stage, current tools available to clinicians involve scoring systems such as Global Registry of Acute Coronary Events (GRACE) scores and Thrombolysis In Myocardial Infarction (TIMI)[2, 3]. Troponin I and NT-proBNP also have been approved as predictors of prognosis after AMI[4, 5]. However new biomarkers are still needed to predict poor prognosis, or help us better understand pathological process in AMI patients.

A recent study has shown that corin can be used as an independent predictor of prognosis in patients with AMI[6]. Furin, as another core enzyme which cleaves proBNP into active BNP fragment together with corin[7], may be associated with poor prognosis after AMI.

Furin is a mammalian subtilisin/kex2p-like endoprotease, which involved in processing of various precursor proteins[8]. Many studies have shown that furin plays an important role in cardiovascular system through regulation of lipid and cholesterol metabolism, blood pressure and the formation of
atherosclerotic lesions[9]. Michael T et al. found that circulating furin cleaved proprotein convertase subtilisin/kexin type 9 (PCSK9), of which the latter regulates LDL receptor and serum atheromatous plaque[10, 11]. Furin is also involved in BP regulation by shedding endogenous (pro)renin receptor[12], promoting migration and proliferation of vascular smooth muscle cells[13] and activation of Epithelial Na+ channel[14]. Moreover, Gopala K et al. observed that inhibition of furin in the atherosclerotic segment of mice decreased vascular remodeling and atherosclerosis[15]. Another study has found that furin is a better predictor compared with BNP and corin for CV outcomes in type 2 diabetes patients[16]. However, studies focus on the association of furin and cardiovascular events after AMI are lacking. We therefore carried out such analysis in our study to evaluate the prognostic utility of plasma furin in AMI patients.

Methods

Study population

A total of 1100 AMI patients were consecutively admitted to the People's Liberation Army General Hospital (PLAGH) between January 2013 and September 2017. All participants provided written informed consent. This study was completely approved by the institutional review board of the PLAGH and was in accordance with the Declaration of Helsinki. AMI was diagnosed if a patient had a cardiac troponin I level exceeding the 99th percentile of a normal reference population with at least one of the following: chest pain lasting>20 min, diagnostic serial electrocardiographic changes consisting of new pathologic Q waves, or ST-segment and T-wave changes[17].

Biochemical measurements

Researchers who were blinded to patients' characteristics and outcomes conducted biochemical measurements. Blood samples were collected from AMI patients on the first morning after admission. Plasma was obtained by centrifugation for 10 min at 3,000 rpm and then stored at -80°C until further analyses. Plasma furin concentration were determined in EDTA-treated plasma samples using commercial available kit (Catalog # EHFURIN, ThermoFisher, USA) according to manufactural instruction.

Outcome events and follow-up

Clinical, demographic and biochemical data of patients were recorded from hospital files and computer records. The primary endpoint for this study was major adverse cardiac events (MACE), a composite of cardiovascular (CV) death, non-fatal myocardial infarction or non-fatal stroke. Other endpoints of interest included hospitalization for heart failure (HF), non-CV death and all death. Hospitalization for HF was defined as a hospital readmission mainly due to heart failure. Recurrent MI was diagnosed in accordance with established criteria as described[17]. Endpoints were obtained by reviewing clinical records of re-admitted patients, or by contacting each patient individually.

Statistical analyses
Continuous variables were compared using the Kruskal-Wallis test. Categorical variables were expressed as counts (percentages) and compared using the Chi-square test. The correlation analysis was performed using Spearman method. The association of plasma furin concentration with AMI outcomes was explored by using Kaplan–Meier methods with stratifications by furin tertiles, and results were also evaluated with Cox proportional hazard regression models. Adjusted covariates included in the multivariate models have been previously proved to associate with MACE. Model 1 adjusted for age and sex. Model 2, the fully adjusted model, additional adjusted for: eGFR, BMI, smoking, history of diabetes, hypertension and myocardial infarction, STEMI/non-STEMI. Subgroup analysis was undertaken to determine whether furin was associated with MACE in different age, gender, BMI, smoking status, diabetes, hypertension and STEMI / NSTEMI subgroups. Multiple linear regression analysis was performed to identify variables that may independently associated with furin in all study population. All statistical tests were double-tailed and a P value less than 0.05 was considered significant. All analyses were performed with SAS version 9.4.

Results

Baseline data

The mean age of 1,100 study participants was 61 ± 13 years; 77% were male. The distribution of plasma furin is left-skewed (sFigure 1). The median plasma furin levels were 156.6 (interquartile range, 102.4-228.8) pg/ml. There was no significant difference between male and female patients (158.5 [103.4-226.9] pg/ml for the male versus 145.9 [103.4-226.9] pg/ml for the female; P= 0.360), between diabetic and non-diabetic patients (160.9 [104.2-231.0] pg/ml for the diabetic versus 155.1 [101.6-224.9] pg/ml for the non-diabetic; P= 0.535), between hypertensive and non-hypertensive patients (154.0 [101.7-222.6] pg/ml for the hypertensive versus 160.8 [103.0-232.1] pg/ml for the non-hypertensive; P= 0.233) and between STEMI and non-STEMI patients (160.7 [105.3-231.4] pg/ml for STEMI versus 147.2 [94.9-220.1]pg/ml for NSTEMI; P=0.079).

sFigure1. Distribution of Plasma furin in our population.

Association between plasma furin levels and clinical parameters

The baseline characteristics of the study population listed in Table 1. AMI patients were divided into 3 groups according to tertile of plasma furin (≤117.5pg/ml, 117.5-200pg/ml, ≥200pg/ml). Slightly increase of mean cTNT were in patients with higher furin levels (P = 0.016). There is no significant increase of NT-proBNP as furin increases (Table 1).

Table 1. Baseline variables according to tertile of plasma furin in AMI patients.
|                                | Overall | ≤117.5 | 117.5-200 | ≥200 | P value  |
|--------------------------------|---------|--------|-----------|------|----------|
| Patients, n                    | 1100    | 356    | 374       | 370  |          |
| Anterior MI, n (%)             | 313 (29.6%) | 100 (29.1%) | 116 (32.4%) | 97 (27.2%) | 0.300    |
| STEMI, n (%)                   | 747 (69.6%) | 231 (66.8%) | 256 (70.7%) | 260 (72.2%) | 0.265    |
| Age, year                      | 61.0 (13.4) | 60.8 (13.8) | 61.6 (13.0) | 60.7 (13.4) | 0.489    |
| Male, n (%)                    | 817 (77.0%) | 262 (75.9%) | 275 (76.6%) | 280 (78.4%) | 0.718    |
| Current smoker, n (%)          | 370 (41.9%) | 127 (44.1%) | 116 (38.5%) | 127 (34.1%) | 0.345    |
| Medical history, n (%)         |         |        |           |      |          |
| Diabetes mellitus              | 394 (37.1%) | 122 (35.7%) | 140 (38.8%) | 132 (36.9%) | 0.690    |
| Hypertension                   | 456 (43.2%) | 150 (44.0%) | 159 (44.5%) | 147 (41.2%) | 0.624    |
| MI                             | 45 (4.1%) | 14 (3.9%) | 13 (3.5%) | 18 (4.9%) | 0.623    |
| CKD                            | 20 (1.9%) | 8 (2.4%) | 7 (2.0%) | 5 (1.4%) | 0.654    |
| AF                             | 10 (0.9%) | 4 (1.1%) | 4 (1.1%) | 2 (0.5%) | 0.648    |
| LIPID                          | 121 (11.0%) | 40 (11.2%) | 38 (10.2%) | 43 (11.6%) | 0.828    |
| HF                             | 5 (0.5%) | 4 (1.1%) | 0 (0.0%) | 1 (0.3%) | 0.090    |
| Clinical assessment            |         |        |           |      |          |
| BMI, kg/m²                     | 25.3 (3.6) | 25.4 (3.8) | 25.5 (3.7) | 24.9 (3.3) | 0.079    |
| HBA1C, %                       | 6.7 (1.6) | 6.7 (1.6) | 6.8 (1.6) | 6.6 (1.6) | 0.465    |
| Glucose, mmol/ L               | 8.5 (3.9) | 8.3 (3.5) | 8.4 (4.0) | 8.7 (4.3) | 0.944    |
| Cr, umol/ L                    | 78.5 (68.0, 94.7) | 79.1 (67.6, 95.6) | 78.1 (67.6, 93.9) | 78.0 (68.7, 93.8) | 0.912    |
| LVEF, %                        | 50.5 (9.0) | 51.0 (9.0) | 50.1 (9.3) | 50.5 (8.7) | 0.509    |
| cTNT, pg/mL                    | 1.8 (0.5, 4.9) | 1.4 (0.4, 3.9) | 1.7 (0.5, 4.6) | 2.1 (0.6, 6.2) | 0.016    |
| NT-proBNP, pg/mL               | 1566 (668, 3929) | 1427 (610, 3490) | 1587 (712, 4098) | 1645 (709, 4049) | 0.361    |
| HR                             | 77.6 (14.9) | 76.5 (13.5) | 77.3 (14.2) | 78.8 (16.7) | 0.591    |
| CHOL, mmol/ L                  | 4.3 (1.1) | 4.2 (1.1) | 4.4 (1.1) | 4.2 (1.1) | 0.601    |
| TRIG, mmol/ L                  | 1.3 (0.9, 1.8) | 1.3 (0.9, 1.8) | 1.3 (1.0, 1.9) | 1.3 (0.9, 1.8) | 0.453    |
| LDL, mmol/ L                   | 2.7 (0.9) | 2.6 (0.9) | 2.7 (1.0) | 2.7 (0.9) | 0.588    |
| HDL, mmol/ L                   | 1.1 (0.3) | 1.1 (0.3) | 1.1 (0.3) | 1.1 (0.3) | 0.771    |
| AST, U/ L                      | 45.6 (24.5, 108.3) | 42.1 (24.3, 91.1) | 43.9 (24.4, 100.3) | 52.0 (26.6, 132.7) | 0.112    |
| ALT, U/ L                      | 30.8 (19.6, 52.5) | 31.6 (19.4, 51.7) | 30.7 (19.7, 51.1) | 31.1 (19.6, 55.1) | 0.898    |
| GGTV, U/ L                     | 29.1 (19.3, 47.9) | 28.4 (19.3, 48.5) | 28.9 (19.3, 47.1) | 29.7 (19.4, 51.0) | 0.772    |
| PT, s                          | 14.1 (2.0) | 14.0 (2.1) | 14.0 (1.3) | 14.3 (2.3) | 0.114    |
| APTT, s                        | 39.4 (35.3, 46.8) | 38.8 (35.3, 45.6) | 39.3 (35.1, 45.9) | 39.9 (35.4, 51.5) | 0.304    |
| DDIMER, ng/ L                  | 0.4 (0.3, 0.8) | 0.4 (0.3, 0.8) | 0.4 (0.3, 0.8) | 0.4 (0.3, 0.9) | 0.653    |
| Medications, n (%)             |         |        |           |      |          |
| Aspirin                        | 1021 (96.3%) | 334 (97.1%) | 347 (96.7%) | 340 (95.2%) | 0.392    |
| ACEI/ARB                       | 434 (40.9%) | 146 (42.4%) | 158 (44.0%) | 130 (36.4%) | 0.093    |
| Statin                         | 1033 (97.5%) | 338 (98.3%) | 351 (97.8%) | 344 (96.4%) | 0.251    |
| DIURETIC                      | 572 (52.0%) | 186 (52.2%) | 193 (51.6%) | 193 (52.2%) | 0.996    |
| CABBLOCKER                     | 121 (11.0%) | 45 (12.6%) | 41 (11.0%) | 35 (9.5%) | 0.394    |
| BETABLOCKER                    | 528 (48.0%) | 187 (52.5%) | 178 (47.6%) | 163 (44.1%) | 0.070    |
| GLP1                           | 22 (2.0%) | 6 (1.7%) | 8 (2.1%) | 8 (2.2%) | 0.875    |
| INSULIN                        | 531 (48.3%) | 158 (44.4%) | 182 (48.7%) | 191 (51.6%) | 0.146    |
| DPP4                           | 49 (4.5%) | 15 (4.2%) | 16 (4.3%) | 18 (4.9%) | 0.895    |

Data are presented as mean (SD), median (interquartile range) or numbers (percentages). ACEI, angiotensin converting enzyme inhibitors; ARB, angiotensin receptor blocker; BMI, body mass index; cTNT, cardiac troponin T; eGFR, estimated Glomerular Filtration Rate; LVEF, left ventricular ejection fraction; MI, myocardial infarction; STEMI, ST-elevation myocardial infarction.
In Spearman correlation analysis, the results showed that log furin did not correlate with age, blood glucose, HbA1c, left ventricular ejection fraction, log eGFR, log cTNT, log CKMB, log NT-proBNP (sTable 1).

**Table 1. Spearman correlation analysis of log furin with covariates.**

| Age   | Glucose | HbA1c | Log eGFR | LVEF | Log CKMB | Log cTNT | Log NT-proBNP |
|-------|---------|-------|----------|------|----------|----------|---------------|
| Log Furin | 0.007   | 0.022 | -0.007   | 0.035| -0.003   | 0.051    | 0.065         | 0.018         |

**Kaplan-Meier analysis**

Over a median follow-up of 31 month, 133 cases of cardiovascular death, 37 cases of non-cardiovascular death, 26 cases of recurrent non-fatal MI, 22 cases of non-fatal stroke and 27 cases of hospitalization for heart failure occurred in this population. Kaplan-Meier survival analysis results suggested that furin was not associated with composite CV outcomes (Figure 1).

**Figure 1. Kaplan–Meier analysis of MACE rates in AMI patients according to different furin category**

**COX regression analysis of end points**

Cox regression analysis indicated that Increasing plasma furin levels was not associated with increasing risk of MACE (HR: 1.01; 95% CI: 0.93-1.06; P=0.807). In addition, for each endpoint of CV death, non-fatal MI, non-fatal stroke, non-CV death, all death or hospitalization for HF, our findings showed that plasma furin was not associated with all these end point except possible recurrent non-fatal MI (HR: 1.09; 95% CI: 1.01-1.17; P = 0.022). (Table 2)

**Table 2. Every 50 unit increase of furin on cardiovascular outcomes**
| Event/No | Unadjusted | Model 1 | Model 2 |
|----------|------------|---------|---------|
|          | Event/No   | HR (95% CI) | P value | HR (95% CI) | P value | HR (95% CI) | P value |
| MACE     |            |          |         |            |         |            |         |
| Low (≤117.5) | 60/356 | 1.12 (0.80, 1.56) | 0.519 | 1.18 (0.84, 1.66) | 0.338 | 1.41 (0.91, 2.17) | 0.125 |
| Median (117.5-200) | 57/374 | Ref. | Ref. | Ref. |
| High (≥200) | 64/370 | 1.04 (0.74, 1.46) | 0.824 | 1.08 (0.77, 1.53) | 0.655 | 1.20 (0.76, 1.90) | 0.433 |
| 50 pg/mL increase | 181/1100 | 1.01 (1.00, 1.03) | 0.084 | 1.02 (1.00, 1.03) | 0.030 | 1.01 (0.96, 1.06) | 0.807 |
| CV death |            |          |         |            |         |            |         |
| Low (≤117.5) | 46/356 | 1.17 (0.77, 1.78) | 0.465 | 1.21 (0.79, 1.86) | 0.376 | 1.29 (0.75, 2.22) | 0.350 |
| Median (117.5-200) | 42/374 | Ref. | Ref. | Ref. |
| High (≥200) | 45/325 | 1.10 (0.72, 1.67) | 0.661 | 1.20 (0.78, 1.84) | 0.410 | 1.02 (0.57, 1.83) | 0.939 |
| 50 pg/mL increase | 133/1100 | 1.01 (1.00, 1.03) | 0.078 | 1.02 (1.00, 1.04) | 0.013 | 0.99 (0.92, 1.06) | 0.709 |
| Non-fatal MI |            |          |         |            |         |            |         |
| Low (≤117.5) | 7/356 | 1.16 (0.41-3.30) | 0.786 | 1.20 (0.42, 3.45) | 0.734 | 1.68 (0.36, 7.90) | 0.509 |
| Median (117.5-200) | 7/374 | Ref. | Ref. | Ref. |
| High (≥200) | 12/370 | 1.67 (0.66, 4.25) | 0.280 | 1.73 (0.68, 4.45) | 0.253 | 5.12 (1.24, 21.2) | 0.024 |
| 50 pg/mL increase | 26/1100 | 1.02 (0.98, 1.07) | 0.394 | 1.02 (0.98, 1.07) | 0.316 | 1.09 (1.01, 1.17) | 0.022 |
| Non-fatal Stroke |            |          |         |            |         |            |         |
| Low (≤117.5) | 7/356 | 1.04 (0.38, 2.86) | 0.945 | 0.99 (0.35, 2.74) | 0.976 | 1.34 (0.41, 4.40) | 0.625 |
| Median (117.5-200) | 8/374 | Ref. | Ref. | Ref. |
| High (≥200) | 7/370 | 0.88 (0.32, 2.43) | 0.806 | 0.83 (0.29, 2.41) | 0.731 | 0.62 (0.15, 2.65) | 0.521 |
| 50 pg unit increase | 22/1100 | 0.92 (0.76, 1.11) | 0.389 | 0.91 (0.74, 1.12) | 0.358 | 0.85 (0.64, 1.14) | 0.277 |
| Hospitalized HF |            |          |         |            |         |            |         |
| Low (≤117.5) | 10/356 | 1.05 (0.44, 2.53) | 0.908 | 1.08 (0.45, 2.60) | 0.860 | 1.85 (0.53, 6.39) | 0.333 |
| Median (117.5-200) | 10/374 | Ref. | Ref. | Ref. |
| High (≥200) | 7/370 | 0.71 (0.27, 1.85) | 0.479 | 0.74 (0.28, 1.94) | 0.540 | 1.71 (0.48,6.10) | 0.405 |
| 50 pg/mL increase | 27/1100 | 0.99 (0.91, 1.09) | 0.862 | 0.99 (0.90, 1.09) | 0.876 | 1.03 (0.94,1.14) | 0.490 |
| Non-CV death |            |          |         |            |         |            |         |
| Low (≤117.5) | 16/356 | 1.68 (0.76, 3.71) | 0.197 | 1.88 (0.85, 4.15) | 0.121 | 1.70 (0.60, 4.79) | 0.316 |
| Median (117.5-200) | 10/374 | Ref. | Ref. | Ref. |
| High (≥200) | 11/370 | 1.11 (0.47, 2.61) | 0.817 | 1.15 (0.49, 2.71) | 0.097 | 0.58 (0.14, 2.39) | 0.449 |
| 50 pg/mL increase | 37/1100 | 1.01 (0.99, 1.04) | 0.305 | 1.02 (1.00, 1.05) | 0.093 | 0.94 (0.75,1.17) | 0.561 |
| All death |            |          |         |            |         |            |         |
| Low (≤117.5) | 62/356 | 1.28 (0.89, 1.85) | 0.191 | 1.36 (0.93, 1.98) | 0.112 | 1.37 (0.85, 2.22) | 0.195 |
| Median (117.5-200) | 52/374 | Ref. | Ref. | Ref. |
| High (≥200) | 56/370 | 1.11 (0.76, 1.62) | 0.600 | 1.21 (0.83, 1.78) | 0.327 | 0.94 (0.55, 1.59) | 0.804 |
Model 1 adjusted for age and sex;  
Model 2 adjusted for model 1 plus eGFR, BMI, smoking, history of diabetes, hypertension or myocardial infarction, STEMI/non-STEMI.

Subgroup analysis

Subgroup analysis according to age, gender, BMI, history of smoking, diabetes, hypertension, types of MI (STEMI / NSTEMI), showed that the association between furin and MACE did not differ in these subgroups (figure 2).

Figure 2. Furin on cardiovascular outcomes subgroup patients.

Finally, we constructed univariable cox regression analysis to identify variables that may independently associated with MACE in our population (table 3). The results showed that NT-proBNP (P < 0.001), age (P < 0.001), creatinine (P < 0.001), cTnT (P = 0.001), blood glucose (P = 0.001), diabetes history (P = 0.010), CKD history (P=0.023) and STEMI (P = 0.039) were positively associated MACE. In contrast, LVEF (P < 0.001), usage of aspirin (P < 0.001), ACEI / ARB (P < 0.001) and women (P = 0.001) were negatively associated with MACE.

Table 3. Univariable predictors of MACE after MI in all study population

| Predictors                          | Chi-Square | HR (95% CI)     | P     |
|-------------------------------------|------------|-----------------|-------|
| NT-proBNP (1000 pg/ml greater)      | 240.4268   | 1.09 (1.08, 1.11) | <0.001 |
| Age (year older)                    | 120.0124   | 1.07 (1.05, 1.08) | <0.001 |
| LVEF (1% greater)                   | 94.6846    | 0.93 (0.91, 0.94) | <0.001 |
| Creatinine (10 unit increase)       | 21.6745    | 1.02 (1.01-1.03)  | <0.001 |
| Aspirin (Yes/No)                    | 14.0069    | 0.38 (0.23, 0.63)  | <0.001 |
| ACEI/ARB (Yes/No)                   | 12.1239    | 0.57 (0.42, 0.78)  | <0.001 |
| cTnT (1 μg/L greater)               | 11.4552    | 1.03 (1.01, 1.04)  | 0.001  |
| Glucose (1 mg/dL greater)           | 11.4426    | 1.05 (1.02, 1.07)  | 0.001  |
| Male                                | 10.3531    | 0.61 (0.45, 0.82)  | 0.001  |
| Diabetes (Yes/No)                   | 6.6461     | 1.45 (1.09, 1.93)  | 0.010  |
| CKD (Yes/No)                        | 5.1332     | 2.40 (1.13, 5.10)  | 0.023  |
| STEMI                               | 4.2433     | 1.36 (1.01, 1.82)  | 0.039  |
| Hypertension (Yes/No)               | 2.6197     | 1.27 (0.95, 1.69)  | 0.106  |
| Furin (50 pg/mL greater)            | 2.1521     | 1.01 (1.00, 1.03)  | 0.142  |
| Hba1c (1 unit greater)              | 1.7690     | 1.07 (0.97, 1.19)  | 0.184  |
| Statin (Yes/No)                     | 0.0710     | 0.90 (0.40, 2.02)  | 0.790  |

Discussion
Our study including 1,100 consecutive AMI patients demonstrated that plasma furin was not associated with MACE events, but may be associated with higher risk of non-fatal MI.

Furin can convert many inactive protein precursors into their active forms. Of these proteins, some play protective roles, while others play harmful roles. In lipid metabolism, furin cleaved PCSK9, which increases the LDL receptor, leading to decrease of LDL-C[11, 18]. On the other hand, ANGPTL 3 and 4, which have a consensus FURIN-recognition site can mediated endothelial lipase (EL) and lipoprotein lipase (LPL) inactivation[9, 19]. Renin receptor (RR), which transformed to its activated form by furin, binds renin or prorenin and consequently increases blood pressure[20]. Epithelial Na+ channel (ENaC), which could be active by furin, associates with increased blood pressure[14, 21]. However, transforming growth factor (TGF-β) is also activated by furin but it contributes to lower blood pressure[22, 23]. BNP activated by furin associates with low blood pressure through its diuretic and vasodilatory actions. In a word, the underlying mechanisms of plasma furin in cardiovascular system may be complex and contradictory.

Studies investigated the role of furin in cardiovascular system also appears complex. Li et al. suggested that the furin gene may be a candidate gene involved in human hypertension as the G allele of 1970C > G is a modest risk factor for hypertension[24]. On the contrary, a genome-wide association human study found that the genotype AA of rs4702 in the furin gene, which marked decreased with furin expression, was associated with both elevated SBP and DBP[25]. In addition, a study including 4678 relative-healthy European adults found that a higher baseline plasma furin was significantly associated with higher BMI, blood glucose and blood pressure[26]. However, the other study including 2312 relative-healthy Chinese adults indicated that a lower level of furin at baseline was significantly associated with higher blood glucose and blood pressure[27]. Our study showed that there is no significant relationship between plasma furin and BMI, blood pressure or blood glucose. These findings suggest a complicated role of furin may exists in cardiovascular system. A recent paper has found that plasma furin is positively associated with MACE after MI, however a detailed method of blood sample collection is not available from the method that forbid a direct comparison with our study[28].

Thus furin involves in a variety of complex mechanisms that interact with each other, which may finally causes the neutral effect to cardiovascular system. Mechanistic studies of furin on cardiovascular diseases should further clarify its activity and regulatory factors.

NT-proBNP can provide prognostic value for MACE in patients with AMI[4]. Our study has also verified this. A recent study has demonstrated two major post-translational modifications including reduction in proBNP glycosylation and increase in furin activity[29]. These synergistically lead to increase in circulating BNP and NT-proBNP[29], but neither concentrations of corin/furin nor corin activity increases. It is possible that an increase in furin activity but no plasma furin concentration are pivotal to the increase in circulating BNP in circumstances of AMI.

Our study found that plasma furin concentration potentially associated with recurrent non-fatal MI after fully adjustment for conventional risk factors. Previous studies investigated the role of furin in
atherosclerosis could explain the association. Earlier studies in different stages identified that furin mRNA increased after myocardial infarction in rat model and the expression of furin was negatively correlated with left ventricular ejection fraction[30, 31]. In addition, Turpeinen et al. found furin overexpressed in human atherosclerotic plaque and inhibition of furin decreased vascular remodelling and atherosclerosis in mouse models, suggesting that furin may play an important role in plaque progression[32]. Several pathophysiologic mechanisms also demonstrate that furin processes and activates many pro-inflammatory cytokines, such as TNF-α, IFN-γ, which contributes to atherosclerosis[33, 34]. Furthermore, furin levels in vascular endothelial cells affect monocyte-endothelial adhesion and migration[35]. In our study, slightly increase of cTNT were found in patients with higher furin levels. Higher peak concentration of cTnT reflect larger infarct area[5]. Therefore, it is reasonable to suggest that higher levels of furin should indicate progression of atherosclerosis and more severe or vulnerable plaque lesions, resulting in higher risk of recurrent nonfatal MI.

To the best of our knowledge, our study is the first to examine the association of plasma furin with risk of MACE in AMI patients. The results showed that plasma furin concentration was not associated with risk of MACE but may be associated with non-fatal MI. Our results highlighted larger sample size studies are needed to verify and detailed mechanism studies are encouraged to explore this. Our study has some limitations. First, this cohort study was conducted in a Chinese population, the generalizability of our findings to other populations with different genetic backgrounds and health profiles should be cautiously. Second, blood samples were collected in first morning after admission. There was no samples available at other time point after MI. The dynamic changes of furin during MI and the relationship between the changes to prognosis are still unknown. Third, we did not evaluate its differences between patients with and without MI, whether MI is associated with higher or lower plasma furin levels could not be identified by this study. Forth, the plasma furin activity was not measured, which may be different from plasma furin concentration, the degradation rate of furin substrate is a potential way to detect this. Lastly, the sample size did not provide enough power to detect a difference in end-points other than composite MACE outcomes.

**Conclusion**

Our study demonstrated that plasma furin was not associated with risks of MACE, but higher levels of plasma furin may be associated with higher risk of recurrent MI in AMI patients.

**Abbreviations**

AMI: acute myocardial infarction; AST: aspartate aminotransferase; ALT: alanine transaminase; ANP: Atrial Natriuretic Peptide; BMI: body mass index; cTNT: troponin T; CK-MB: creatine kinase-MB; CKD: Chronic Kidney Disease; CV: Cardiovascular; DM: Diabetes mellitus; eGFR: estimated glomerular filtration rate; ENaC: Epithelial Sodium Channels; HBA1C: glycosylated hemoglobin A1C; HDL-C: high density lipoprotein cholesterol; LDL-C: low density lipoprotein cholesterol; LVEF: left ventricular ejection fraction; MACE: major adverse cardiac events; NSTEMI: Non-ST-segment elevation myocardial infarction; NT-
proBNP: N terminal pro B type natriuretic peptide; PCSK: proprotein convertase subtilisin/kexin; STEMI: ST-segment elevation myocardial infarction.

**Declarations**

**Ethics approval and consent to participate**

The study was reviewed and approved by the ethics committee of the PLAGH and is in accordance with the Declaration of Helsinki. Written informed consent was obtained from individual or guardian participants.

**Consent for publication**

Not applicable.

**Availability of data and materials**

The datasets that support the findings of this study are available from the corresponding author on reasonable request.

**Competing interests**

The authors declare that they have no competing interests.

**Funding**

This study was supported by the China Postdoctoral Science Foundation (2016M603025).

**Authors' contributions**

ZWL conceived the concept of the study and drafted the manuscript. ZWL and JWL performed the statistical analyses and drafted the manuscript. ZWL and QM collected baseline and follow-up data. QM and JL performed laboratory tests. YDC guided the writing of the article and is the first correspondent author. All authors have read and approved the manuscript.

**Acknowledgements**

Not applicable.

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