Circulating fibrocyte levels correlate with infarct size in patients with ST elevation myocardial infarction treated with primary percutaneous coronary intervention☆

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ABSTRACT

Study objective: Infarct size is a strong predictor of outcomes after ST elevation myocardial infarction (STEMI). Circulating fibrocytes are bone marrow-derived progenitor cells associated with fibrotic processes. We tested whether fibrocytes correlate with infarct size in STEMI patients treated with primary percutaneous coronary intervention (PCI).

Design: Prospective observational study.

Setting: Academic medical center.

Participants: Subjects with STEMI treated with primary PCI.

Interventions: Peripheral blood draw and cardiac magnetic resonance imaging (CMR).

Main outcome measure: Correlation of fibrocyte levels with infarct size.

Methods: Peripheral blood fibrocytes were quantified at discharge from STEMI hospitalization and at 6 months follow-up using flow cytometry. Infarct size was determined within 2 weeks of discharge and at 6 months follow-up using late gadolinium enhancement on CMR.

Results: Among 14 patients (median age 54 years, 79% men) with STEMI, there was a statistically significant positive correlation between fibrocyte levels at 6 months and 6-month infarct size on CMR (r = 0.58, p = 0.031). In addition, there was a positive correlation between peak troponin I level (r = 0.85, p < 0.001) and white blood cell count (r = 0.55, p = 0.042) during the hospital stay and 6-month infarct size on CMR.

Conclusions: Circulating fibrocytes measured 6 months after STEMI positively correlate with 6-month infarct size assessed by CMR.

1. Introduction

In patients with ST elevation myocardial infarction (STEMI) treated with reperfusion therapy, the infarct area initially contains infarcted tissue, edema, swollen cardiomyocytes, and variable areas of microhemorrhage. Over several weeks, inflammation resolves and the infarcted tissue is replaced by extracellular matrix proteins that are then cross-linked, forming the mature myocardial scar [1]. The size of this fibrotic area, as detected by late gadolinium enhancement on cardiac magnetic resonance (CMR) imaging, is a strong predictor of clinical outcomes [2]. While CMR is a powerful tool to assess this process [3], it is not routinely performed following STEMI in clinical practice [4].

Circulating fibrocytes are bone marrow-derived progenitor cells that are released into the bloodstream in response to tissue injury and mediate tissue fibrosis [5–7] and can be quantified as an easily measurable biomarker of fibrosis. Fibrocytes have been shown to home to the injured myocardium in patients with myocardial infarction [8], and the concentration of circulating fibrocytes is elevated in several...
fibrotic heart diseases [9,10]. We thus hypothesized that, in patients with STEMI, circulating fibrocyte levels correlate with infarct size on CMR at 6 months.

2. Material and methods

2.1. Study design

This study was carried out in concordance with the principles of the Declaration of Helsinki and with approval of institutional review boards. We enrolled consecutive patients presenting with STEMI treated with primary PCI from August 1, 2014 to September 30, 2019. Inclusion criteria were: (1) age older than 18 years; (2) diagnosis of STEMI; and (3) treatment with primary PCI. Exclusion criteria were: (1) history of prior acute coronary syndrome; (2) known fibrotic disease or malignancy; (3) active infection; (4) surgery or trauma within the prior 6 weeks; (5) renal insufficiency (glomerular filtration rate < 0.05. 

2.2. Fibrocyte measurement

Peripheral venous blood samples were collected in heparinized tubes prior to hospital discharge and at 6 months follow-up, and the concentration of fibrocytes was measured as previously described [11,12]. Briefly, the buffy coat was filtered and the concentration of leukocytes quantified by enumerating live cells under a hemocytometer. Fibrocytes were identified by flow cytometry on a FACS Canto-II flow cytometer using Diva software (version 5.0.3; BD Biosciences), defined as cells co-expressing the common leukocyte antigen CD45, CD34, and intracellular staining for collagen-1 (CD45+ CD34+ CD105+). The absolute concentration was determined as the product of the ratio to parent CD45+ population and the concentration of live leukocytes in each sample.

2.3. Cardiac magnetic resonance imaging

Patients underwent CMR imaging within 2 weeks of discharge and at 6 months follow-up using previously published techniques [13]. Cine left ventricular function was assessed from end-diastolic and end-systolic volumes. Late gadolinium enhancement (LGE) imaging was obtained 10–15 min after injection of 0.15 mmol/kg of gadolinium contrast. Images were analyzed by MRI-trained cardiologists (MS and MA) blinded to patient fibrocyte data. For quantification of LGE, endocardial and epicardial borders were traced in a semi-automated fashion. Hyper-enhanced areas were detected in comparison with remote segments using full-width-half-maximum method. Manual correction was used to avoid blood pool detection and to include areas of microvascular obstruction. Infarct size was then recorded as the percentage of LGE areas from total left ventricular mass.

2.4. Statistical analysis

Data were analyzed in SPSS (version 26 for Mac, IBM, Armonk, New York, USA) or Prism (version 9 for Mac, GraphPad, San Diego, CA, USA) software. Continuous variables were summarized as median and inter-quartile range, and compared using the Mann-Whitney U test. Correlations were assessed using Pearson’s correlation coefficient (r). Results were considered statistically significant if two-sided p value was less than 0.05.

### Table 1
Baseline characteristics of 14 patients with ST elevation myocardial infarction.

| Demography          | Baseline | 6 month follow-up | p-value |
|---------------------|----------|-------------------|---------|
| Age, years          | 54 [50-57] | 52 [48-56]      | 0.626   |
| Male sex            | 11 (79%) | 12 (86%)         | 0.818   |
| Race                |          |                   |         |
| White               | 12 (86%) | 12 (86%)         | 0.974   |
| Black               | 2 (14%)  | 0 (0%)           | 0.137   |
| Co-morbidities      |          |                   |         |
| Diabetes mellitus   | 3 (21%)  | 3 (21%)          | 0.974   |
| Hypertension        | 5 (36%)  | 4 (29%)          | 0.657   |
| Hyperlipidemia      | 7 (50%)  | 6 (43%)          | 0.505   |
| Tobacco use         | 11 (79%) | 11 (79%)         | 0.974   |
| Laboratory studies  |          |                   |         |
| Peak troponin level | 66.4 [27.2-88.1] | 12.8 [10.6-15.8] | 0.256   |
| Left ventricular function |            |                   |         |
| Cardiogenic shock on admission | 3 (21%) | 6 (43%) | 0.328   |
| Diastolic dysfunction | 3 (21%) | 3 (21%) | 0.974   |
| Coronary anatomy    |          |                   |         |
| Infarct-related artery |       |                   |         |
| Left anterior descending | 7 (50%) | 5 (36%) | 0.657   |
| Circumflex          | 2 (14%)  | 2 (14%)          | 0.974   |
| Right coronary      | 5 (36%)  | 5 (36%)          | 0.974   |
| Number of vessels with ≥70% luminal narrowing | | | |
| 1-vessel            | 10 (71%) | 5 (36%)         | 0.256   |
| 2-vessel            | 4 (29%)  | 2 (14%)          | 0.657   |
| 3-vessel            | 0        | 0                | 0.974   |
| Number of drug-eluting stents deployed | | | |
| 1                  | 13 (93%) | 5 (36%)         | 0.256   |
| 2                  | 1 (7%)   | 0                | 0.974   |
| Medications on discharge | | | |
| Angiotensin converting enzyme inhibitor | 13 (93%) | 3 (21%) | 0.256   |
| Beta-blocker        | 11 (79%) | 7 (50%)         | 0.256   |
| Dual anti-platelet therapy | 14 (100%) | 14 (100%) | 0.974   |
| Oral hypoglycemic medication | 3 (36%) | 3 (36%) | 0.974   |
| Statin              | 14 (100%) | 14 (100%) | 0.974   |

Data presented as median [inter-quartile range].

### Table 2
Cardiac functional assessment, myocardial scar, and fibrocyte levels at baseline and 6 month follow-up.

| CMR cardiac functional assessment | Baseline | 6 month follow-up | p-value |
|-----------------------------------|----------|-------------------|---------|
| LV end-diastolic volume, ml       | 150.72 [114.58-180.43] | 147.50 [103.18-162.49] | 0.369   |
| LV end-systolic volume, ml        | 79.10 [45.65-104.55] | 85.20 [48.61-160.50] | 0.626   |
| Stroke volume, ml                 | 73.83 [65.79-79.57] | 49.48 [43.14-87.08] | 0.362   |
| Ejection fraction, %              | 48.08 [38.65-62.02] | 45.97 [30.35-66.79] | 0.657   |
| Cardiac output, L/min             | 4.07 [3.40-5.12] | 3.58 [2.68-6.11] | 0.743   |
| CMR myocardial scar extent        |          |                   |         |
| Total left                        | 118.89 [110.69-145.93] | 121.80 [95.75-151.00] | 0.724   |
| ventricular mass, g               | 23.02 [15.34-31.46] | 19.47 [7.75-31.26] | 0.328   |
| Percent scar, %                   | 19.74 [11.91-24.77] | 17.07 [7.84-23.96] | 0.256   |
| Microvascular obstruction, g      | 1.85 [1.00-2.82] | 1.46 [0.08-2.43] | 0.283   |
| Fibrocyte level, cells/μl         | 1.71 × 10^7 [6.40 × 10^6-3.04 × 10^7] | 1.84 × 10^7 [6.23 × 10^6-3.43 × 10^7] | 0.818   |

Data presented as median [interquartile range], CMR = cardiac magnetic resonance imaging; LV = left ventricular.
3. Results

Among 19 enrolled patients, 5 patients did not complete CMR due to claustrophobia (n = 1), COVID-19 restrictions regarding clinical research (n = 1), and relocation (n = 3). A total of 14 patients completed peripheral blood sampling and CMR imaging at baseline and follow-up and were included in the analysis (Table 1). CMR data and fibrocyte levels at baseline and 6 months are summarized in Table 2. No patients were rehospitalized or experienced major adverse cardiac events during the study period.

The concentration of circulating fibrocytes 6 months after STEMI had no detectable relationship with peak troponin I level, peak white blood cell count or left ventricular function during the hospital stay (Table 3), but positively correlated with 6-month infarct size on CMR (Fig. 1, panel A). Peak troponin I level (Fig. 1, panel B) and peak white blood cell count (Fig. 1, panel C) during the hospital stay both positively correlated with infarct size at 6 months, although left ventricular function did not (Table 4). The presence of left anterior descending artery as a culprit for STEMI also positively correlated with infarct size at 6 months (r 0.57, p 0.034). There was no correlation between the change in EF and change in fibrocyte level over time (r 0.27, p 0.354).

4. Discussion

We found that the concentration of circulating fibrocytes 6 months after STEMI correlated with the size of infarction on CMR. Consistent with prior literature, we also found a positive correlation between peak troponin and white blood cell count during hospital stay with 6-month infarct size [14–16], consistent with the concept that, in the days following STEMI, higher white blood cell counts and troponin levels are indicative of a larger infarction.

Fibrocytes have been linked to normal wound healing as well as numerous fibrotic disorders [5]. As related to the heart, circulating fibrocytes correlated with LGE on CMR in patients with hypertrophic cardiomyopathy [9] and hypertensive heart disease [10], left atrial fibrosis in patients with atrial fibrillation [17], and myocardial infarction in murine models [18,19]. In an autopsy study of patients who died of sudden death, investigators detected fibrocytes in the myocardium in areas of scar suggesting that fibrocytes migrate to the heart and

| Table 3 |
|---|
| Correlation of variables with fibrocyte levels at baseline and 6 month follow-up. |
| Fibrocyte level | Fibrocyte level |
| baseline | 6 month follow-up |
| r | p-value | r | p-value |
| CMR cardiac functional assessment | | | |
| Left ventricular ejection fraction, % | 0.51 | 0.065 | 0.30 | 0.306 |
| Angiography results | | | |
| Left anterior descending culprit | 0.09 | 0.771 | 0.51 | 0.063 |
| Peak laboratory values | | | |
| Troponin I (ng/mL) | 0.27 | 0.355 | 0.23 | 0.430 |
| White blood count (cells/mm³) | 0.11 | 0.706 | 0.43 | 0.125 |

CMR = cardiac magnetic resonance imaging; r = correlation coefficient.

Fig. 1. Scatterplots showing correlations between fibrocyte levels and infarct size at 6 months (r = 0.58, p = 0.031) (panel A), peak troponin I level and infarct size (r = 0.85, p < 0.001) (panel B), and peak white blood cell count and infarct size (r = 0.55, p = 0.042) (panel C). LVEF = left ventricular ejection fraction.
levels at 6 months have larger infarct size on CMR at 6 months adds to peripheral vein [8].

with subsequent myocardial scar formation. While left ventricular systolic ejection fraction is an important predictor of clinical outcomes in STEMI patients undergoing primary PCI and found that fibrocytes were related artery at the time of reperfusion and from a peripheral vein in investigators measured fibrocytes in thrombi aspirated from the infarct-related artery in STEMI patients treated with primary PCI, circulating levels of fibrocytes at 6 months positively correlate with infarct size on CMR at 6 months. If validated in larger studies, circulating fibrocyte levels may be useful as a risk stratification tool, or as a target of response to therapy.

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Declaration of competing interest

No authors reported conflict of interest.

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