Fibroblast activation protein imaging in reperfused ST-elevation myocardial infarction: comparison with cardiac magnetic resonance imaging

Boqia Xie · Jiaxin Wang · Xiao-Ying Xi · Xiaojuan Guo · Bi-Xi Chen · Lina Li · Cuncun Hua · Shihua Zhao · Pixiong Su · Mulei Chen · Min-Fu Yang

Received: 7 October 2021 / Accepted: 29 December 2021 / Published online: 5 January 2022
© The Author(s), under exclusive licence to Springer-Verlag GmbH Germany, part of Springer Nature 2022

Abstract
Purpose The aim of this study was to explore the correlation of 18F-labeled fibroblast activation protein inhibitor (FAPI) and cardiovascular magnetic resonance (CMR) parameters in ST-elevation myocardial infarction (STEMI) patients with successful primary percutaneous coronary intervention (PPCI) and to investigate the value of FAPI imaging in predicting cardiac functional recovery, as well as the correlation between FAPI activity and circulating fibroblast activation protein (FAP) and inflammatory biomarkers.

Methods Fourteen first-time STEMI patients (11 men, mean age: 62 ± 11 years) after PPCI and 14 gender-matched healthy volunteers (10 men, mean age: 50 ± 14 years) who had completed FAPI imaging and blood sample collection were prospectively recruited. All patients underwent baseline FAPI imaging (6 ± 2 days post-MI) and CMR (8 ± 2 days post-MI). Ten patients had follow-up CMR (84 ± 4 days post-MI). Myocardial FAPI activity was analyzed for extent (the percentage of FAPI uptake volume over the left ventricular volume, FAPI%), intensity (target-to-background uptake ratio, TBRmax), and amount (FAPI% × TBRmax). Late gadolinium enhancement (LGE), T2-weighted imaging (T2WI), extracellular volume (ECV), microvascular obstruction (MVO), and cardiac function from CMR imaging were analyzed. Blood samples obtained on the day of FAPI imaging were used to assess circulating FAP, TGF-β1, TNF-α, IL-6, and hsCRP in STEMI patients and controls.

Results Localized but inhomogeneous FAPI uptake was observed in STEMI patients, which was larger than the edematous and infarcted myocardium, whereas no uptake was detected in controls. The MVO area showed lower FAPI uptake compared with the surrounding myocardium. FAPI activity was associated with the myocardial injury biomarkers T2WI, LGE, and ECV at both per-patient and per-segment levels (all \( p < 0.05 \)), but was not associated with circulating FAP, TGF-β1, TNF-α, IL-6, or hsCRP. Among the CMR parameters, T2WI had the greatest correlation coefficient with both FAPI% and FAPI% × TBRmax. Baseline TBRmax was inversely correlated with the follow-up left ventricular ejection fraction (LVEF) (\( r = -0.73, p = 0.02 \)).

Conclusion FAPI imaging detects more involved myocardium than CMR in reperfused STEMI, and is associated with myocardial damage and follow-up LVEF.

Keywords Fibroblast activation protein inhibitor · Fibroblast · Cardiovascular magnetic resonance · ST-elevation myocardial infarction · Cardiac functional recovery

Introduction
ST-elevation myocardial infarction (STEMI) triggers an orchestrated and complicated series of events from the initial sterile inflammation to the subsequent (myo)fibroblast proliferation and scar formation [1]. Appropriate fibrotic response following MI preserves the structural integrity, while excessive or prolonged fibrogenic activation leads to adverse ventricular remodeling.
Cardiac magnetic resonance (CMR) is a reliable technique for the evaluation of pathological changes of post-MI myocardium using different sequences and modalities: late gadolinium enhancement (LGE) for scar and focal fibrosis [2, 3], T2-weighted imaging (T2WI) and T2 mapping for edema, and extracellular volume (ECV) derived from T1-weighted imaging for the total interstitial space [2]. However, these imaging sequences reflect the increase of extracellular matrix components but not the central cellular effectors of fibrosis: the fibroblasts.

Fibroblasts undergo dramatic phenotypic changes following MI; in addition to their traditional role as the main source of extracellular matrix proteins, activated fibroblasts may regulate inflammation, modulate cardiomyocyte survival, and mediate angiogenesis [4–6]. These features make the fibroblast the crucial cell in both repair and pathogenesis of adverse remodeling. Fibroblast activation protein (FAP) is a membrane-anchored peptidase that is specifically expressed by activated fibroblasts [7], and lately, radiolabeled FAP inhibitor (FAPI)—targeting tracers have been developed to detect the activated fibroblasts [8]. These tracers have been widely used for positron emission tomography/computed tomography (PET/CT) imaging in various diseases, especially cancer, and recently, in cardiac diseases [9, 10]. Several pre-clinical and clinical studies reported that MI would induce enhanced FAPI uptake, which extended over the culprit territory and the LGE area [11–14]. These findings suggest that FAPI imaging may have a unique advantage in detecting the affected myocardium.

Nevertheless, some concerns of FAPI imaging in MI remain. First, previous comparison with CMR was limited to LGE, and only one case report adopted T1 and ECV mapping [12]. As new sequences have proved to be more accurate in evaluating myocardial injury, their comparisons with FAPI should be clarified. Second, the prognostic value of FAPI imaging in post-MI cardiac recovery needs to be explored. Third, circulating FAP and inflammatory biomarkers have been reported to be related to myocardial injury and STEMI prognosis [15–17], so whether these markers are related to FAPI activity is worth investigating.

Therefore, we (1) explored the correlation of FAPI imaging and CMR parameters in reperfused STEMI patients; (2) investigated the prognostic value of FAPI imaging in cardiac recovery 3 months post-MI; and (3) evaluated the correlation between FAPI activity and circulating FAP and inflammatory biomarkers.

Materials and methods

Study population

This prospective study was approved by the Institutional Ethical Committee of Beijing Chaoyang Hospital (2020-ke-225) and was performed in agreement with the Declaration of Helsinki. From January to February 2021, we recruited 14 consecutive first-time STEMI patients (11 men, mean age: 62 ± 11 years) successfully treated by primary percutaneous coronary intervention (PPCI) <12 h from the onset of symptoms at Beijing Chaoyang Hospital. Diagnosis and treatment of STEMI were as per current guidelines [18, 19]. Exclusion criteria were <18 years of age, previous cardiac events (myocardial infarction, cardiac surgery, PCI), valvular diseases, cardiomyopathies, abnormal renal function, or contraindications to the use of gadolinium-based contrast agents or CMR.

According to MI-animal studies, cardiac fibroblast proliferation peaked from days 2 to 4 and declined through day 7, and the peak FAPI uptake was observed on day 6 after coronary ligation [11, 20]. However, the post-MI dynamic FAPI uptake pattern in humans remains unclear. To obtain the FAPI uptake characteristics during the acute phase, the imaging time point in this study was scheduled around day 7. Patients underwent baseline myocardial FAPI PET/CT imaging at 6 ± 2 days post-MI and CMR at 8 ± 2 days. Ten patients had follow-up CMR scans (84 ± 4 days post-MI). Patients or their guardians signed the informed consent before enrollment in the study.

Controls

We recruited a control group of 14 gender-matched healthy volunteers (10 men, mean age: 50 ± 14 years) who had completed FAPI imaging and blood sample collection. The inclusion criteria were as follows: (1) no history of cardiovascular disease, (2) no history of malignancy, and (3) no abnormal findings on FAPI PET/CT imaging.

Blood analysis

Serial blood samples were tested in all patients at admission and in the cardiac care unit before transferring the patients to ordinary wards, including complete blood count, metabolic panel (troponin I, creatine kinase-MB [CK-MB], creatinine kinase [CK], B-type natriuretic peptide [BNP], lactate dehydrogenase [LDH], cholesterol and triglyceride levels, liver and renal function tests), and arterial blood gas tests. On the day of FAPI imaging, venous blood samples were obtained from the cubital vein from both STEMI patients and healthy controls. Serum was separated by centrifugation at 4 °C for 10 min at 4000 rpm within 20 min of collection and was stored at −80 °C until analysis. Circulating FAP, tumor necrosis factor-α (TNF-α), transforming growth factor-β1 (TGF-β1), interleukin-6 (IL-6), and hypersensitive C-reactive protein (hsCRP) were analyzed using commercially available ELISA kits (CUSABIO, Wuhan, China) in accordance with the manufacturer’s recommendations.
**FAPI image acquisition and interpretation**

Al\(^{18}\)F-NOTA-FAPI was radiolabeled as previously described [21]. PET/CT images were acquired 60 min after injection (2.5–3.0 MBq/kg) using a 16-slice PET/CT scanner (Discovery STE, GE, USA). CT parameters were 140 kV, 120 mA, pitch 1.375, 16×0.625 mm collimation, and section width of 5 mm. For STEMI patients, two beds of PET images (5 min/bed, 3D mode) were acquired, and the heart was set in the center of the view. For controls, whole-body PET images (2.5 min/bed, 3D mode) were acquired from the base of the skull to mid-thigh. Attenuation-corrected PET images (voxel size 2.5 min/bed, 3D mode) were acquired, and the heart was set in the center of the view. For controls, whole-body PET images (2.5 min/bed, 3D mode) were acquired from the base of the skull to mid-thigh. Attenuation-corrected PET images (voxel size 3.9×3.9×3.3 mm) were reconstructed from the CT data using 3D ordered-subset expectation maximization algorithm (14 subsets, 2 iterations). Integrated PET and CT images were obtained automatically on AW VolumeShare 2 (GE Healthcare).

FAPI images were independently evaluated by two nuclear physicians (XYX and MFY) blinded to the CMR and clinical information. Disagreements were resolved by consensus. Myocardial FAPI uptake was analyzed in the global and segmental manner, respectively. For global analysis, a volume of interest (VOI) was manually drawn around the myocardium with FAPI uptake, and a region grow algorithm with a threshold of 50% of the maximum uptake was set to determine the FAPI volume. The FAPI volume was further expressed as a percentage of left ventricular (LV) volume derived from MRI, which was defined as the FAPI extent (FAPI%). Within this VOI, the maximum standardized uptake values (SUVmax) in the infarct area were automatically derived, and a target-to-background ratio (TBRmax) was calculated by dividing myocardial SUVmax by blood pool SUVmean to indicate the FAPI intensity. Finally, the total FAPI was calculated by the product of FAPI% and FAPI TBRmax. SUVmax values in the remote area and normal control group were obtained by manually drawing a circle 10 mm in diameter in the myocardium opposing the infarct zone and in the mid-anterior wall, on three consecutive slices oriented by the PET/CT fusion image, and TBRmax was calculated.

For segmental analysis, the American Heart Association 17-segment model was applied using QPS software (version 3.1, Cedars-Sinai Medical Center, Los Angeles, CA, USA). Segmental FAPI uptake was automatically calculated by the software and expressed as a percentage of the LV SUVmax, and the product of this percentage and LV TBRmax was further calculated to represent the segmental TBRmax.

**CMR image acquisition and interpretation**

CMR was performed on a 3.0-T scanner (Prisma, Erlangen, Siemens Healthcare, Germany) using the following protocol: balanced steady-state free precession (bSSFP) acquisitions for long- and short-axis cine; short time inversion recovery (STIR) prepared T2WI of three short-axis slices (basal, mid-ventricular and apical); standard LGE imaging matching long- and short-axis bSSFP cine slices for the quantification of scar burden; and modified Look-Locker inversion recovery (MOLLI) sequence for pre- and post-contrast T1 mapping of 3 short-axis slices. LGE and post-contrast T1 mapping images were collected 10–15 min after the administration of 0.1 mmol/kg Gd-DOTA (Dotarem™, Guerbet S.A., Paris, France). Typical acquisition parameters for bSSFP were TE/TR = 1.4/44 ms, flip angle 44°, voxel size 0.8×0.8×8.0 mm, and matrix 192×256. Parameters for STIR-prepared T2WI were TE/TR = 50/2100 ms, voxel size 1.4×1.4×8.0 mm, and matrix 135×256. The MOLLI acquisition followed the 5(3)3 protocol for pre-contrast and the 4(1)3(1)2 protocol for post-contrast T1 mapping with typical parameters of TE/TR = 1.12/283 ms, voxel size 1.4×1.4×8.0 mm, and matrix 148×256. The LGE parameters were TE/TR = 1.96/450 ms, flip angle 20°, and voxel size 1.4×1.4×8.0 mm.

CMR images were analyzed on a commercial software (Cvi42, Version 5.12.2, Circle Cardiovascular Imaging Inc., Calgary, Canada) by two experienced CMR operators (JXW and SHZ) who were blinded to both clinical and FAPI data; disagreements were resolved by consensus. Volumetric and functional parameters were calculated from the cine images. The infarcted myocardium was identified on LGE images using 5 standard deviations (SD) above the remote myocardium [14]. Myocardial edema was defined by a high signal intensity on T2WI that was 2 SD above the signal intensity of the remote myocardium. The extent of T2WI and LGE was expressed as the percentage of LV mass (T2WI% and LGE%). Microvascular obstruction (MVO) was identified in LGE images as hypointense recesses within the hyper-enhanced myocardium. ECV was quantified according to the established formula [22]: ECV = (1 − hematocrit) × (1/T1myopost − 1/T1myopre) / (1/T1bloodpost − 1/T1bloodnative) and expressed as ECV%. The apex segment was excluded in the segment-to-segment comparative analysis between FAPI and CMR images.

**Statistical analysis**

SPSS Statistics (Version 24; IBM) was adopted. The normality of distribution was assessed by Kolmogorov–Smirnov test. Continuous variables were described as mean (SD) or medians (ranges), depending on the normality of distribution. Categorical variables were expressed as absolute numbers and percentages. A paired t test was used to compare the different imaging parameters in the same individual. ANOVA and non-parametric tests were used to compare the differences of parameters among segments. Pearson/Spearman’s correlation analysis was conducted to explore the
relationship. A boxplot was used to determine the outliers. A $p$-value $< 0.05$ was considered to be statistically significant.

**Results**

**Patients’ characteristics**

Detailed characteristics of the 14 patients are presented in Table 1. Culprit lesion was determined at the left main coronary artery in 1 patient (7.10%), left anterior descending coronary artery in 10 patients (71.40%), and right coronary artery in 3 patients (21.40%). The pain-to-balloon time was 6 h (3–11 h). Four patients' peak troponin levels exceeded the measurable upper limit and were reported as $> 500$ ng/ml. The remaining 10 patients' peak troponin level was $158 \pm 40$ ng/ml. All patients were taking aspirin, clopidogrel/ticagrelor, and statin at discharge.

**FAPI activity and blood tests**

Strong correlation was demonstrated between FAPI% and CKMBmax, maximum white blood cell count (WBCmax), and LDHmax ($r$ values of 0.79, 0.65, and 0.62; all $p < 0.05$), followed by a moderate correlation between FAPI% × TBRmax and CKMBmax, WBCmax, LDHmax, and BNPmax ($r$ values of 0.56, 0.55, 0.56, and 0.59; all $p < 0.05$) (Table 2). TBRmax was only related to WBCmax ($r = 0.59$, $p = 0.03$).

Decreased FAP (44.62 ± 15.67 ng/ml vs. 87.79 ± 18.67 ng/ml, $p < 0.001$) and TGF-β1 levels [1.42 (0.93–3.83) ng/ml vs. 3.88 (2.08–5.32) ng/ml, $p = 0.04$], increased hsCRP [11210 (7859–18,169) ng/ml vs. 1671 (610–3131) ng/ml, $p < 0.001$] and IL-6 levels [7.93 (5.98–19.59) pg/ml vs. 2.80 (2.01–4.78) pg/ml, $p = 0.01$], and a similar TNF-α level (22.06 ± 15.12 pg/ml vs. 23.97 ± 15.33 pg/ml, $p = 0.74$) were identified in STEMI patients compared with normal controls (Table 3). None of these markers was related to the concurrent FAPI activity, either in the infarct region or in the remote area (Table 2).

**Comparison of FAPI and CMR**

Localized but inhomogeneous FAPI uptake was observed in all STEMI patients (Fig. 1), while no visible uptake was detected in normal controls (Supplement Fig. S1). TBRmax was higher in the infarct region than in the remote area in STEMI patients (9.68 ± 2.61 vs. 1.07 ± 0.25, $p < 0.001$), and was higher than the normal controls (0.96 ± 0.20, $p < 0.001$). Comparison of TBRmax in the remote area in STEMI patients and TBRmax in normal controls yielded no difference (1.07 ± 0.25 vs. 0.96 ± 0.20, $p = 0.23$) (Supplement Fig. S1 and S2). FAPI% varied to a larger scale (20.82–78.74%), even in patients with similar culprit lesions. Notably, the FAPI% was larger than T2WI% (45.03 ± 16.23% vs. 24.80 ± 6.73%, $p < 0.001$) and LGE% (19.79 ± 8.26%, $p = 0.03$) (Fig. 2B). When FAPI% increased, the differences between FAPI% and T2WI%, LGE%, and ECV% increased (all $p < 0.001$) (Fig. 2C).

At the patient level, FAPI% had a stronger correlation with CMR parameters than FAPI% × TBRmax and TBRmax (Table 4). Among the CMR parameters, T2WI% had the greatest correlation coefficient with both FAPI% and FAPI% × TBRmax ($r$ values of 0.83 and 0.83; both $p < 0.05$).

At the segment level (Fig. 3), significant correlations were demonstrated between segmental TBRmax and T2WI%, LGE%, and ECV% ($r$ values of 0.62, 0.79, and 0.71; all $p < 0.001$). MVO was observed in 21 segments in 4 patients. FAPI uptake in the MVO area was lower than in the surrounding tissue (Fig. 4).

**Follow-up**

Improvement was recorded in LVEF (47.84% ± 12.10% vs. 55.39% ± 7.93%, $p = 0.01$). After exclusion of one outlier in the follow-up LVEF in patient No. 11, baseline TBRmax, ECV%, and T2WI% were found related to the follow-up LVEF ($r$ values of $-0.73$, $-0.74$, and $-0.70$; all $p < 0.05$) (Table 5 and Fig. 5). TBRmax in the remote area, circulating FAP, hsCRP, TNF-α, TGF-β1, and IL-6, was not associated with follow-up LVEF (all $p > 0.05$).

**Discussion**

Noninvasive imaging plays an essential role in the monitoring of post-MI myocardial remodeling. Using gallium-68 (68 Ga)–labeled FAPI, Varasteh et al. demonstrated excellent in vivo imaging quality for the detection of post-MI fibroblast activation in an MI rat model [11]. More recently, the feasibility of 68 Ga-FAPI imaging in MI patients was reported by Notohamiprodjo [12], Kessler [13], and Diekmann [14], who identified intense FAPI uptake in the infarcted territory. These data suggest that FAPI imaging is a reliable technique in delineating the temporospatial status of activated fibroblasts and may help to better explain post-MI pathophysiologic evolution and monitoring of the efficacy of antifibrotic therapies.

**Reperfused MI induced an extending FAPI uptake over the infarct region**

Previous reports [12, 14] demonstrated an extended fibroblast activation over the perfusion defect and LGE area. In the current study, by comparing with multiple CMR sequences, we further identified that the FAPI uptake extent was even larger than the edematous area. Our finding is in
Table 1 Patients' characteristics

| Patient No | Male | Age | BMI (kg/m²) | Culprit vessel | Pain to balloon (h) | Peak cTnI (ng/ml) | Peak CK (U/l) | Peak CKMB (ng/ml) | WBCmax (*10⁹/l) | Peak LDH (U/l) | LDL (mmol/l) | Peak BNP (pg/ml) | DM | HTN | Current smoker | Family history | MI to PET (days) | MI to CMR (days) |
|------------|------|-----|-------------|----------------|-------------------|-------------------|--------------|------------------|----------------|---------------|--------------|----------------|------|------|---------------|---------------|----------------|----------------|
| 1          | Yes  | 82  | 33.78       | LAD            | 8                 | 110               | 2319         | 291.0           | 18.70          | 469           | 4.45         | 329           | No   | No   | Yes           | Yes           | 6               | 7               |
| 2          | Yes  | 55  | 23.51       | RCA            | 2                 | 59                | 991          | 72.2            | 9.17           | 312           | 5.02         | 134           | No   | No   | Yes           | Yes           | 8               | 8               |
| 3          | Yes  | 54  | 25.66       | LAD            | 3                 | 177               | 2475         | 174.0           | 8.26           | 459           | 3.64         | 72            | No   | Yes  | Yes           | No            | 3               | 9               |
| 4          | Yes  | 52  | 24.68       | RCA            | 8                 | >500              | 6418         | 177.4           | 12.13          | 1126          | 2.57         | 444           | Yes  | No   | Yes           | No            | 8               | 11              |
| 5          | No   | 63  | 23.53       | LAD            | 3                 | 227               | 1583         | 116.3           | 12.69          | 444           | 4.89         | 88            | No   | Yes  | No            | Yes           | 4               | 8               |
| 6          | Yes  | 75  | 25.01       | LAD            | 5                 | >500              | 3554         | 395.0           | 11.29          | 584           | 3.35         | 495           | No   | Yes  | No            | No            | 3               | 8               |
| 7          | No   | 59  | 23.73       | RCA            | 12                | >500              | 3554         | 395.0           | 11.29          | 584           | 3.35         | 495           | No   | Yes  | No            | No            | 6               | 7               |
| 8          | Yes  | 54  | 29.65       | LAD            | 4                 | 349               | 5983         | 134.5           | 14.39          | 1145          | 3.61         | 99            | Yes  | No   | Yes           | Yes           | 5               | 7               |
| 9          | Yes  | 72  | 29.05       | LAD            | 12                | >500              | 4763         | 337             | 14.73          | 845           | 2.57         | 380           | Yes  | Yes  | Yes           | No            | 6               | 7               |
| 10         | Yes  | 46  | 31.14       | RCA            | 10                | 19                | 222          | 29.7            | 12.24          | 252           | 3.45         | 121           | No   | Yes  | Yes           | No            | 5               | 5               |
| 11         | Yes  | 64  | 22.31       | LAD            | 12                | 373               | 4167         | 338             | 16.22          | 1289          | 2.65         | 720           | No   | No   | No            | No            | 9               | 9               |
| 12         | No   | 53  | 30.92       | LAD            | 3                 | 53                | 1636         | 160             | 13.87          | 510           | 4.25         | 214           | No   | No   | Yes           | No            | 6               | 8               |
| 13         | Yes  | 69  | 26.83       | LM             | 2                 | 171               | 1486         | 104             | 11.17          | 1289          | 4.95         | 794           | No   | No   | Yes           | No            | 7               | 6               |
| 14         | Yes  | 74  | 22.68       | LAD            | 6                 | 45                | 964          | 84              | 9.58           | 383           | 3.39         | 732           | Yes  | Yes  | Yes           | No            | 12              | 11              |
| Total      |     | 62±11| 26.61±3.66 | NA              | 6 (3–11)         | NA               | 286.5±1925.73 | 200.58±125.34  | 12.55±2.84     | 3.72±0.86     | 365.50±256.51 | 4 (29%) | 6 (43%) | 11 (79%) | 11 (79%) | 6±2 | 8±2 |

BMI, body mass index; LAD, left anterior descending coronary artery; RCA, right coronary artery; cTnI, cardiac troponin I; CK, creatine kinase; CKMB, creatine kinase-MB; WBCmax, maximum white blood cell; LDH, lactate dehydrogenase; LDL, low-density lipoprotein; BNP, B-type natriuretic peptide; DM, diabetes mellitus; HTN, hypertension; MI, myocardial infarction; PET, positron emission tomography; CMR, cardiac magnetic resonance
Table 3 Comparisons of serum biomarkers and FAPI activity in the remote area between STEMI patients and normal controls

| Serum biomarkers | STEMI patients (n=14) | Normal controls (n=14) | p value |
|------------------|-----------------------|------------------------|---------|
| FAP, ng/ml       | 44.62±15.67           | 87.79±18.67            | <0.001  |
| TGF-β1, ng/ml    | 1.42 (0.93–3.83)      | 3.88 (2.08–5.32)       | 0.04    |
| hsCRP, ng/ml     | 11,210 (7859–18,169)  | 1671 (610–3131)        | <0.001  |
| TNF-α, pg/ml     | 22.06±15.12           | 23.97±15.33            | 0.74    |
| IL-6, pg/ml      | 7.93 (5.98–19.59)     | 2.80 (2.01–4.78)       | **0.01**|
| Peak CK, U/l     | 1.07±0.25             | 0.96±0.20              | 0.23    |

STEMI, ST-elevation myocardial infarction; FAP, fibroblast activation protein; TGF-β1, transforming growth factor-β1; hsCRP, hypersensitive C reactive protein; TNF-α, tumor necrosis factor-α; IL-6, interleukin-6; TBRmax, maximum target-to-background ratio. Bold p values, statistically significant (p < 0.05).

FAPI uptake was associated with the degree of myocardial injury

Our data show that both the extent and amount of FAPI uptake were strongly correlated with myocardial injury assessed by serum biomarkers, CMR-derived parameters, and systemic inflammation. As discussed above, upregulated chemokines and increased mechanical stress induced by MI are strong stimulators of fibroblasts, and inflammatory cells as macrophages and mast cells also play an important role in fibroblast activation in the non-culprit territory.

Fibroblasts are sensitive to local chemical and mechanical stimulations. Acute MI induces an intense inflammatory reaction in the infarcted region and upregulates the levels of TGF-β1, IL-1β, and IFNγ in the injured area along with increased mechanical load [23, 24] contributing to fibroblast activation. Although the abundant cardiac resident fibroblasts located close to the vessels are important cellular effectors that respond to the activating signals following MI, recent data from animal experiments suggest that the intense upregulation of chemokines induced by MI may drive recruitment of non-resident fibroblasts, which may significantly contribute to the expansion of activated fibroblasts. As reported by Nagaraju et al. in an MI pig model [25], upregulated expression of TGF-β1, a strong stimulator for fibroblast differentiation and positive FAP-α staining, was seen in the border and remote myocardium in addition to the scar region, suggesting that the activation of fibroblasts can occur in the non-infarcted area. Future studies are needed to explore the signal pathways mediating fibroblast activation in the non-culprit territory, and to illustrate their functions.

FAPI uptake was associated with the degree of myocardial injury

Table 2 Correlation analysis between FAPI activity and blood results (n=14)

| Blood results | FAPI% × TBRmax-infarct | p value |
|---------------|-------------------------|---------|
| FAP, ng/ml    | 0.44                    | 0.26    |
| TGF-β1, ng/ml | 0.32                    | 0.09    |
| hsCRP, ng/ml  | 0.01                    | 0.66    |
| TNF-α, pg/ml  | 0.16                    | 0.001   |
| IL-6, pg/ml   | 0.06                    | 0.59    |
| Peak CKMB, U/l| 0.25                    | 0.23    |
| Peak LDH, U/l | 0.02                    | 0.44    |
| LDL, mmol/l   | 0.31                    | 0.02    |
| Peak BNP, pg/ml| 0.53                  | 0.05    |

FAPI, fibroblast activation protein; TBRmax, maximum target-to-background ratio; TGF-β1, transforming growth factor-β1; hsCRP, hypersensitive C reactive protein; TNF-α, tumor necrosis factor-α; IL-6, interleukin-6; FAPI, fibroblast activation protein; BNP, B-type natriuretic peptide. Bold p values, statistically significant (p < 0.05).
Fig. 1 Detailed images of the 14 patients. Coronary angiographic images before (first column) and after PCI (second column) with the treated culprit vessel (white arrows). Polar maps (third column) and the selected short-axis images of FAPI (fourth column), LGE (fifth column), and T2WI (sixth column). FAPI uptake was observed in the culprit territory, which was larger than the corresponding edematous and infarcted area (yellow arrows). PCI, percutaneous coronary intervention; FAPI, fibroblast activation protein inhibitor; SA, short-axis; LGE, late gadolinium enhancement; T2WI, T2-weighted imaging.

Fig. 2 Comparison of FAPI uptake extent (FAPI%) and CMR parameters. A A representative case and post-myocardial infarction illustration indicating that FAPI uptake extent exceeded that of CMR parameters. B FAPI uptake extent was larger than the edematous and infarcted area. C The differences between FAPI% and LGE%, T2WI%, and ECV% increased with the FAPI uptake extent. FAPI, fibroblast activation protein inhibitor; CMR, cardiac magnetic resonance; LGE, late gadolinium enhancement; T2WI, T2-weighted imaging; ECV, extracellular volume.
in fibroblast activation by secreting various bioactive mediators [26]. These facts explain the tight association between myocardial damage and FAPI uptake, and FAPI imaging is a reliable modality in evaluating reperfused myocardial damage. Although our findings concerning the correlation of FAPI uptake and serum biomarkers are in line with Kessler’s results [13], they are different from those of Diekmann [14]. A more detailed data presentation from the latter may help in better interpretation of the differences.

**Cardiac FAPI activity and circulating biomarkers**

MI induces dynamic alterations of serum FAP and inflammatory biomarkers, each with a specific kinetic and variable peak time: FAP concentrations declined in the first 5 days, IL-6 and hsCRP reached a peak within 48–72 h, and TNF-α increased until day 6. Furthermore, the decline of FAP concentration, TNF-α measured at day 1, hsCRP, and IL-6 were all reported to be significantly correlated with increased myocardial damage [15, 27, 28].

Our findings are congruent with these reports by determining a decreased FAP concentration and an increased hsCRP and IL-6 level in STEMI patients at day 6 ± 2 days. However, none of these markers was related to cardiac FAPI activity or to the follow-up LVEF, even though the decline of FAP concentration, TNF-α, hsCRP, and IL-6 were all reported to be significantly correlated with myocardial damage [15, 29]. One possible explanation is that a one-time value cannot reflect the dynamic changes and peak values of the biomarkers, which may be related to FAPI activity. This assumption needs to be verified in future studies by kinetic assessment at different time points.

Although enhanced FAPI activity was identified in our study, and previous studies demonstrated increased TGF-β1 expression in the post-MI myocardium [20, 27], circulating FAP and TGF-β1 levels significantly declined in the current study. This inconsistency of the same biomarker between tissue-level and circulating-level raises the possibility that the peripheral biomarkers may not be reliable in reflecting the tissue status, no matter whether the difference is attributed to local consumption during cardiac wound healing or because the circulating biomarkers are prone to be affected by various factors other than MI [30]. We suggest that FAPI imaging is more direct and reliable than serum biomarkers in the evaluation of post-MI myocardial damage and cardiac functional recovery.

| Table 4 | Correlation analysis between FAPI and baseline CMR parameters at patient level (n = 14) |
|---|---|---|
| CMR parameters | FAPI% | TBRmax-infarct | FAPI% × TBRmax-infarct |
| | r | 95%CI | p value | r | 95%CI | p value | r | 95%CI | p value |
| T2WI, % | 0.83 | 0.54–0.95 | <0.001 | 0.58 | 0.21–0.83 | 0.03 | 0.83 | 0.48–0.97 | <0.001 |
| LGE, % | 0.72 | 0.32–0.94 | 0.004 | 0.54 | 0.11–0.81 | 0.048 | 0.81 | 0.46–0.96 | <0.001 |
| ECV, % | 0.78 | 0.16–0.95 | 0.002 | 0.50 | −0.15–0.85 | 0.09 | 0.72 | 0.19–0.92 | 0.006 |
| LVEF, % | −0.72 | −0.90 to −0.24 | 0.005 | −0.62 | −0.86 to −0.21 | 0.02 | −0.73 | −0.92 to −0.33 | 0.003 |
| EDV/BSA, ml/m² | 0.34 | −0.27–0.75 | 0.23 | 0.11 | −0.57–0.70 | 0.71 | 0.15 | −0.40–0.66 | 0.62 |
| ESV/BSA, ml/m² | 0.44 | −0.14–0.80 | 0.12 | 0.42 | −0.10–0.78 | 0.14 | 0.50 | −0.02–0.80 | 0.07 |

**CMR**, cardiac magnetic resonance; **FAPI**, fibroblast activation protein inhibitor; **TBRmax**, maximum target-to-background ratio; **T2WI**, T2-weighted imaging; **LGE**, late gadolinium enhancement; **ECV**, extracellular volume; **LVEF**, left ventricular ejection fraction; **EDV**, end diastolic volume; **ESV**, end systolic volume; **BSA**, body surface area; bold p values, statistically significant (p < 0.05)
FAPI and follow-up cardiac function

To the best of our knowledge, our study was the first to show that FAPI imaging had prognostic value in post-MI cardiac recovery. The correlation coefficient of FAPI uptake intensity was not inferior to well-established CMR predictors. However, our data cannot explain why baseline FAPI uptake extent was not associated with follow-up LVEF even though its relationship was stronger than FAPI uptake intensity with the CMR predictors. The traditional concept of cardiac fibroblast as the manufacturer of extracellular matrix proteins has been challenged by growing evidence suggesting that fibroblasts are functionally and phenotypically heterogeneous, and exerts diverse effects in the complex post-MI signal pathways [2–4]. Therefore, we presume that fibroblasts in the infarct, border, and remote myocardium may be functionally and phenotypically distinct, thus leading to inconsistent or even contrary effects on post-MI prognosis, which may partially explain why the extent of FAPI uptake was not predictive of cardiac recovery.

Although animal experiments showed that myocardial FAPI uptake declined significantly at 6 days post-MI [11], human-based clinical studies [13] show that FAPI activity can be detected on the 66th day post-infarction. Continued activation of fibroblasts may play an essential role in the cardiac remodeling. Unfortunately, repeated FAPI imaging was not performed in the follow-up phase in our patients. The evolution of FAPI uptake in STEMI patients with PPCI in the real world needs further exploration. Future studies aimed at the temporal alterations of post-MI FAPI imaging and its correlation to the cardiac remodeling will provide useful theoretical evidence and improve our knowledge of post-MI remodeling.

MVO could be visualized by FAPI imaging

Ischemia–reperfusion injury could result in coronary microvascular obstruction, leading to a significant reduction or occlusion of effective blood flow to the affected regions [31]. Accordingly, we assume that the delivery of FAPI tracer to
the MVO area also declined and resulted in the hypo-uptake region within the culprit territory.

MVO is an important predictor of cardiac mortality and the therapeutic target in STEMI patients undergoing PPCI. First-pass perfusion and LGE by CMR were two techniques used to detect MVO [32]. However, first-pass perfusion is limited by the low spatial resolution and signal-to-noise ratio, and LGE is limited in terms of longer examination time and multiple breath holds. This study, to our knowledge, proposed for the first time that MVO could be visualized by FAPI imaging. However, given that we were restricted to the limited number of patients, the feasibility of FAPI imaging in detecting MVO needs further validation.

Limitations

This study had several limitations. First, the sample size was small: only 14 patients were included in the FAPI-CMR comparison, and only 10 patients had repeated CMR scans. Second, FAPI and CMR were not simultaneously performed, which may have affected the comparability of the results. Third, FAPI imaging was not repeated in the follow-up period. Fourth, although studies that focused on STEMI patients receiving primary PCI [33, 34] showed that the majority of patients experienced LV remodeling in the first 3 months, a longer follow-up period may yield more evident LV remodeling. Fifth, histological evidence was not available in the present study, and FAPI activity could not be directly validated to represent fibroblast activation, even though this has been verified in animal models [11]. A multicentered cohort study enrolling more STEMI patients with an extended follow-up period may yield more information on post-MI FAPI imaging.

Conclusions

In reperfused STEMI patients, by comparing FAPI imaging and CMR, we demonstrated that activated fibroblasts existed in the non-edematous or infarcted myocardium. Thus, as a new modality, FAPI imaging is feasible in assessing myocardial damage and has prognostic value in post-MI cardiac recovery, which is more direct and reliable than serum biomarkers. These advantages make it a potential technique in the evaluation of myocardial remodeling after MI and a favorable method in analyzing fibroblast-targeted antifibrotic therapies.

Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1007/s00259-021-05674-9.

Author contribution

Boqia Xie, Jiaxin Wang, Xiao-Ying Xi, Mulei Chen, and Min-Fu Yang wrote the draft of the manuscript; Boqia Xie, Jiaxin Wang, Xiao-Ying Xi, Xiaojuan Guo, Bi-Xi Chen, Cuncun Hua, and Pixiong Su collected and analyzed the clinical data; Jiaxin Wang, Xiaojuan Guo, and Shihua Zhao analyzed the CMR data; Xiao-Ying Xi, Bi-Xi Chen, and Min-Fu Yang analyzed the PET/CT data; Lina
Li collected and analyzed the blood samples and drafted the related discussion; Boqia Xie, Pxiong Su, Mulei Chen, and Min-Fu Yang conceived the study and interpreted the results. All authors contributed to the article’s revision, agreed to its submission, and had full access to original data.

**Funding** This work was supported by Beijing Hospitals Authority Clinical Medicine Development of Special Funding Support (ZYLX202105).

**Availability of data and material** The data underlying this article will be shared on reasonable request to the corresponding author.

**Code availability** Not applicable.

**Declarations**

**Conflict of interest** The authors declare no competing interests.

**Ethics approval** All procedures involving human participants were carried out in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards.

**Consent to participate** Informed consent was obtained from all individual participants included in the study.

**Consent for publication** Patients signed informed consent regarding publishing their data and photographs.

**References**

1. Prabhu SD, Frangogiannis NG. The biological basis for cardiac repair after myocardial infarction: from inflammation to fibrosis. Circ Res. 2016;119:91–112. https://doi.org/10.1161/CIRCRESAHA.116.303577.

2. Kali A, Cokic I, Tang RL, Yang HJ, Sharif B, Marbán E, et al. Determination of location, size, and transmurality of chronic myocardial infarction without exogenous contrast media by using cardiac magnetic resonance imaging at 3T. Circ Cardiovasc Imag. 2014;7:471–81. https://doi.org/10.1161/CIRCIMAGING.113.001541.

3. Gupta S, Ge Y, Singh A, Gráni C, Kwong RY. Multimodality imaging assessment of myocardial fibrosis. JACC Cardiovasc Imag. 2021;14:2457–69. https://doi.org/10.1016/j.jcmg.2021.01.027.

4. Shinde AV, Frangogiannis NG. Mechanisms of fibroblast activation in the remodeling myocardium. Curr Pathobiol Rep. 2017;5:145–52. https://doi.org/10.1007/s40319-017-0132-z.

5. Nakaya M, Watari K, Tajima M, Nakaya T, Matsuda S, Ohara H, et al. Cardiac myofibroblast engulfment of dead cells facilitates recovery after myocardial infarction. J Clin Invest. 2017;127:383–401. https://doi.org/10.1172/JCI83822.

6. Ubil E, Duan J, Pillai IC, Rosa-Garrido M, Wu Y, Bargiacchi F, et al. Mesenchymal-endothelial transition contributes to cardiac neovascularization. Nature. 2014;514:585–90. https://doi.org/10.1038/nature13839.

7. Toms J, Kogler J, Maschauer S, Daniel C, Schmidkonz C, Kuwert T, et al. Targeting fibroblast activation protein: radiosynthesis and preclinical evaluation of an 18F-labeled FAP inhibitor. J Nucl Med. 2020;61:1806–13. https://doi.org/10.2967/jnumed.120.242958.

8. Altmann A, Haberkorn U, Siveke J. The latest developments in imaging of fibroblast activation protein. J Nucl Med. 2021;62:160–7. https://doi.org/10.2967/jnumed.120.244806.

9. Tillmanns J, Hoffmann D, Habbaba Y, Schmitt JD, Sedding D, Fraccarollo D, et al. Fibroblast activation protein alpha expression identifies activated fibroblasts after myocardial infarction. J Mol Cell Cardiol. 2015;87:194–203. https://doi.org/10.1016/j.yjmcc.2015.08.016.

10. Tallquist MD, Molkentin JD. Redefining the identity of cardiac fibroblasts. Nat Rev Cardiol. 2017;14:484–91. https://doi.org/10.1038/nrcardio.2017.37.

11. Varasteh Z, Mohanta S, Robu S, Braeuer M, Li Y, Omidvari N, et al. Molecular imaging of fibroblast activation after myocardial infarction using a 68Ga-labeled fibroblast activation protein inhibitor, FAPi-04. J Nucl Med. 2019;60:1743–9. https://doi.org/10.2967/jnumed.119.226993.

12. Notohamiprodjo S, Nekolla SG, Robu S, Villagran Asaires A, Kupatt C, Ibrahim T, et al. Imaging of cardiac fibroblast activation in a patient after acute myocardial infarction using 68Ga-FAPI-04. J Nucl Cardiol. 2021. https://doi.org/10.1007/s12350-021-02603-z.

13. Kessler L, Kapusovic J, Ferdinandus J, Hirmas N, Umutlu L, Zarrad F, et al. Visualization of fibroblast activation after myocardial infarction using 68Ga-FAPI PET. Clin Nucl Med. 2021;46:807–13. https://doi.org/10.1097/RLU.0000000000003745.

14. Diekmann J, Koenig T, Zwadlo C, Derlin T, Neuser J, Thackeray JT, et al. Molecular imaging identifies fibroblast activation beyond the infarct region after acute myocardial infarction. J Am Coll Cardiol. 2021;77:1835–7. https://doi.org/10.1016/j.jacc.2021.02.019.

15. Tillmanns J, Fraccarollo D, Galuppo P, Wollert KC, Bauersachs J. Changes in concentrations of circulating fibroblast activation protein alpha are associated with myocardial damage in patients with acute ST-elevation MI. Int J Cardiol. 2017;232:155–51. https://doi.org/10.1016/j.ijcard.2017.01.037.

16. Raposeiras-Roubin S, Barreiro Pardal C, Rodiño Janeiro B, Abussi E, García-Acuña JM, González-Juantey JR. High-sensitivity C-reactive protein is a predictor of in-hospital cardiac events in acute myocardial infarction independently of GRACE risk score. Angiology. 2012;63:30–4. https://doi.org/10.1177/0003319711406502.

17. Genrawsingh RM, Cheng JM, Akkerhuis KM, Kardys I, Degertekin M, van Geuns RJ, et al. High-sensitivity C-reactive protein predicts 10-year cardiovascular outcome after percutaneous coronary intervention. EuroIntervention. 2016;12:345–51. https://doi.org/10.4244/EIJY15M07_04.

18. Levine GN, Bates ER, Blankenship JC, Bailey SR, Bittl JA, Cercek B, et al. 2015 ACC/AHA/SCAI focused update on primary percutaneous coronary intervention for patients with ST-elevation myocardial infarction: an update of the 2011 ACCP/AHA/SCAI guideline for percutaneous coronary intervention and the 2013 ACCF/AHA guideline for the management of ST-elevation myocardial infarction: a report of the American College of Cardiology/American Heart Association Task Force on Clinical Practice Guidelines and the Society for Cardiovascular Angiography and Interventions. Catheter Cardiovasc Interv. 2016;87:1001–19. https://doi.org/10.1002/ccd.26325.

19. Ibanez B, James S, Agewall S, Antunes MJ, Bucciarelli-Ducci C, Bueno H, et al. 2017 ESC guidelines for the management of acute myocardial infarction in patients presenting with ST-segment elevation: the task force for the management of acute myocardial infarction in patients presenting with ST-segment elevation of the European Society of Cardiology (ESC). Eur Heart J. 2018;39:119–77. https://doi.org/10.1093/eurheartj/ehx393.
20. Fu X, Khalil H, Kanisicak O, Boyer JG, Vagnozzi RJ, Maliken BD, et al. Specialized fibroblast differentiated states underlie scar formation in the infarcted mouse heart. J Clin Invest. 2018;128:2127–43. https://doi.org/10.1172/JCI98215.

21. Wang S, Zhou X, Xu X, Ding J, Liu S, Hou X, et al. Clinical translational evaluation of Al39F-NOTA-FAPI for fibroblast activation protein-targeted tumour imaging. Eur J Nucl Med Mol Imag. 2021;48:4259–71. https://doi.org/10.1007/s00259-021-05470-5.

22. Flett AS, Hayward MP, Ashworth MT, Hansen MS, Taylor AM, Elliott PM, et al. Equilibrium contrast cardiovascular magnetic resonance for the measurement of diffuse myocardial fibrosis: preliminary validation in humans. Circulation. 2010;122:138–44. https://doi.org/10.1161/CIRCULATIONAHA.109.930636.

23. Kawaguchi M, Takahashi M, Hata T, Kashima Y, Usui F, Morimoto H, et al. Inflammasome activation of cardiac fibroblasts is essential for myocardial ischemia/reperfusion injury. Circulation. 2011;123:594–604. https://doi.org/10.1161/CIRCULATIONAHA.110.982777.

24. Siwik DA, Chang DL, Colucci WS. Interleukin-1beta and tumor necrosis factor-alpha decrease collagen synthesis and increase matrix metalloproteinase activity in cardiac fibroblasts in vitro. Circ Res. 2000;86:1259–65. https://doi.org/10.1161/01.res.86.12.1259.

25. Nagaraju CK, Dries E, Popovic N, Singh AA, Haemers P, Rod erick HL, et al. Global fibroblast activation throughout the left ventricle but localized fibrosis after myocardial infarction. Sci Rep. 2017;7:10801. https://doi.org/10.1038/s41598-017-09790-1.

26. Saxena A, Chen W, Su Y, Rai V, Uche OU, Li N, et al. IL-1 induces proinflammatory leukocyte infiltration and regulates fibroblast phenotype in the infarcted myocardium. J Immunol. 2013;191:4838–48. https://doi.org/10.4049/jimmunol.1300725.

27. Kempf K, Halter G, Füth R, Herder C, Müller-Scholze S, Gülker H, et al. Increased TNF-alpha and decreased TGF-beta expression in peripheral blood leukocytes after acute myocardial infarction. Horm Metab Res. 2006;38:346–51. https://doi.org/10.1055/s-2006-925403.

28. Kehmeier ES, Lepper W, Kropp M, Heiss C, Hendgen-Cotta U, Balzer J, et al. TNF-α, myocardial perfusion and function in patients with ST-segment elevation myocardial infarction and primary percutaneous coronary intervention. Clin Res Cardiol. 2012;101:815–27. https://doi.org/10.1007/s00392-012-0465-x.

29. Di Stefano R, Di Bello V, Barsotti MC, Grigoratos C, Armani C, Dell’Omodarme M, et al. Inflammatory markers and cardiac function in acute coronary syndrome: difference in ST-segment elevation myocardial infarction (STEMI) and non-STEMI models. Biomed Pharmacother. 2009;63:773–80. https://doi.org/10.1016/j.biopha.2009.06.004.

30. Navarro SL, Brasky TM, Schwarz Y, Song X, Wang CY, Kristal AR, et al. Reliability of serum biomarkers of inflammation from repeated measures in healthy individuals. Cancer Epidemiol Biomarkers Prev. 2012;21:1167–70. https://doi.org/10.1158/1055-9965.

31. Judd RM, Lugo-Oliveri CH, Araï M, Kondo T, Croisille P, Lima JA, et al. Physiological basis of myocardial contrast enhancement in fast magnetic resonance images of 2-day-old reperfused canine infarcts. Circulation. 1995;92:1902–10. https://doi.org/10.1161/01.cir.92.7.1902.

32. Nijveldt R, Hofman MB, Hirsch A, Beek AM, Umans VA, Algra PR, et al. Assessment of microvascular obstruction and prediction of short-term remodeling after acute myocardial infarction: cardiac MR imaging study. Radiology. 2009;250:363–70. https://doi.org/10.1148/radiol.2502080739.

33. van der Bijl P, Abou R, Goedemans L, Gersh BJ, Holmes DR Jr, Ajmone Marsan N, et al. Left ventricular post-infarct remodeling: implications for systolic function improvement and outcomes in the modern era. JACC Heart Fail. 2020;8:131–40. https://doi.org/1016/j.jchf.2019.08.014.

34. Lustosa RP, van der Bijl P, El Mahdii M, Montero-Cabezas JM, Kostyukevich MV, Ajmone Marsan N, et al. Noninvasive myocardial work indices 3 months after ST-segment elevation myocardial infarction: prevalence and characteristics of patients with postinfarction cardiac remodeling. J Am Soc Echocardiogr. 2020;33:1172–9. https://doi.org/10.1016/j.echo.2020.05.001.

Publisher's note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

Authors and Affiliations

Boqia Xie1 · Jiaxin Wang2 · Xiao-Ying Xi3 · Xiaojuan Guo4 · Bi-Xi Chen3 · Lina Li3 · Cuncun Hua1 · Shihua Zhao2 · Pixiong Su1 · Mulei Chen1 · Min-Fu Yang3

1 Cardiac Center, Beijing Chaoyang Hospital, Capital Medical University, 8th Gontinlan Rd, Chaoyang District, Beijing 100020, China
2 MR Center, Fuwai Hospital, State Key Laboratory of Cardiovascular Disease, National Center for Cardiovascular Diseases of China, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing 100037, China
3 Department of Nuclear Medicine, Beijing Chaoyang Hospital, Capital Medical University, 8th Gontinlan Rd, Chaoyang District, Beijing 100020, China
4 Department of Radiology, Beijing Chaoyang Hospital, Capital Medical University, 8th Gontinlan Rd, Chaoyang District, Beijing 100020, China