HHS Public Access

Author manuscript

Circ Cardiovasc Imaging. Author manuscript; available in PMC 2019 November 01.

Published in final edited form as:

Circ Cardiovasc Imaging. 2018 November ; 11(11): e008457. doi:10.1161/CIRCIMAGING.118.008457.

Dual-Contrast $^{19}F/^{1}H$ MRI to Characterize Myocardial Infarct Healing: Advancing the Horizon for MR Microscopy with Clinical MR Scanners

Gregory M. Lanza, MD PhD

Department of Medicine, Washington University School of Medicine, St. Louis, MO

Keywords

editorial; magnetic resonance imaging; myocardial infarction; macrophage; Magnetic Resonance Imaging (MRI); Myocardial Infarction

MRI imaging techniques to noninvasively interrogate and characterize injured myocardium continue to evolve beyond viability or "scar imaging" [1, 2]. Ramos et al[3] present proof-of-concept data demonstrating a multi-nuclear ($^{19}F/^{1}H$) MRI imaging protocol intended to characterize myocardial inflammation and extracellular matrix remodeling following permanent coronary occlusion in mice. Dual contrast agents are employed to characterize macrophage accumulation within the inflamed myocardium (i.e., perfluorocarbon nanoparticles [4]), and to assess extracellular matrix stabilization reflected by progressive elastin/tropoelastin content [5]. Collectively, the dual biomarker MR imaging offers an approach to quantitative characterization of inflammation persistence and myocardial remodeling as noninvasive metrics to support the development of post-MI cardioprotective therapies.

Myocardial Infarction Pathophysiology

Ischemic insult from total coronary occlusion in mammalian hearts produces profound cardiomyocyte death clinically assessed by electrocardiographic findings, imaging studies, and detection of highly sensitive troponin biomarkers in blood. The metabolic and microscopic consequences of ischemia are reversible over the first 20 min, but longer times lead to irreversible cardiomyocyte and noncardiomyocyte apoptotic and necrotic death associated with enzymatic matrix dissolution [6]. Cells dying due to necrosis release intracellular contents which serve as "alarmins" to activate innate immune pathways and cells expressing toll-like receptors (TLR) or receptors for advanced glycation and end-products (RAGE) among others and trigger inflammation. Infarcted heart repair proceeds through sequential overlapping phases: an inflammatory phase characterized by induction of innate immune pathways and inflammatory leukocyte recruitment to eliminate dead cells, a

Correspondence: Gregory Lanza MD PhD, Professor of Medicine and Bioengineering, James R. Hornsby Family Professorship in Biomedical Sciences, Division of Cardiology, Washington University Medical School, CORTEX Building, Suite 101, 4320 Forest Park Ave, Saint Louis, MO 63110, Tel: 314-454-8813, Fax: 314-454-5265, greg.lanza@mac.com

Disclosures: None

proliferative phase during which anti-inflammatory pathways are activated to suppress inflammatory reactions, and a maturation phase following the formation of a structural matrix network characterized by changes in scar cellular elements and extracellular matrix composition [6].

Post-MI resolution of inflammation is a dynamic orchestrated process involving neutrophils and macrophages. Overactive neutrophils and their secretion of proinflammatory mediators directly promote cardiomyocyte death. Cessation of the overactive priming of neutrophils is critical to infarct healing and amelioration of adverse myocardial remodeling [7,8]. Specialized pro-resolving lipid mediators (SPM) including lipoxins, resolvins, protectins, and maresins are induced through neutrophil-monocyte-platelet interactions. In combination with IL-10, TGF-β, SPMs limit neutrophil recruitment and promote acute inflammation resolution, in part by recruitment of anti-inflammatory macrophages that passivate the neutrophil-driven inflammatory phase[7,8]. In mice, Ly6C^{hi} proinflammatory monocytes migrate into the injured tissues expressing high levels of CCR2 and low levels of the fractalkine CXC3CR1 (Ly6C^{hi}, CCR2^{hi}, CXC3CR1^{low}) and differentiate into inflammatory M1 macrophages [9,10]. Ly6C^{low}/CCR2$^{low/neg}$/CX3CR1^{hi} monocytes migrate into the wound providing surveillance and anti-inflammatory activity as M2 macrophages [9,10]. In the complex infarcted myocardium macrophages likely exist as a spectrum of nuanced phenotypes with distinct functional properties rather than as distinct M1/M2 cells, a classification based on in vitro experimentation.

In the present study as the duration of ischemia persists due to total non-reperfusion, the heterogenous resident macrophages in the heart were destroyed and replaced with circulating monocytes originating from the bone marrow and particularly the spleen. Monocyte uptake of PFC nanoparticles peripherally was used to assess the macrophage influx into the heart during the inflammatory phase [11]. Macrophage accumulation increased steadily during the first week of post infarction followed by a marked reduction on day 14 and negligible levels thereafter based on ^{19}F MRI normalized to scar volume. These cells participate in the clearance of dead cells and matrix debris during the inflammatory phase of the infarction. As a single parameter the accumulation of PFC laden macrophages within the scar did not correlate with LV remodeling changes. During the proinflammatory phase, reparative macrophages and lymphocyte subsets infiltrate the infarction site by secreting cytokines and growth factors that modulate resident fibroblasts, fibroblast progenitor cells, cardiac pericytes, and vascular smooth muscle cells into synthetic myofibroblast-like cells producing structural and matricellular extracellular matrix proteins [6]. Whether these reparative macrophages within the scar bear a PFC cell-tracking label in the present study is unclear.

Concurrently, extracellular matrix composition is remodeled with reconstitution of collagens, reticulins, and elastins that were degraded along with cardiomyocytes during the acute ischemic insult [6,12,13]. Collagen I/III production constitutes the predominant extracellular myocardium matrix component, previously reported to increase progressively to 4X normal heart content over 4 weeks following acute infarction in rats [13]. While providing structural integrity, increased myocardial collagen content of the remodeled heart relative to normal cardiac muscle yields a stiffer, less compliant heart characterized by

diastolic and systolic dysfunction. Conversely, elastin, a relatively small component of extracellular matrix, provides elasticity to the myocardium preserving systolic and diastolic function and decreasing ventricular dilation. The importance of elastin in the post-acute MI heart is illustrated by the effectiveness of intra-myocardium cell therapy of bone marrow stromal cells that were genetically modified to over-express elastin[14]. In that study enhanced elastic structure of the injured myocardial extracellular matrix improved diastolic function, ameliorated ventricular dilation and preserved cardiac ejection fraction. In Ramos et al[3] the expression of elastin/tropoelastin was most prominent at 14- and 21-days post infarction using Gd-ESMA T1w imaging and mapping, which is consistent with the known pathological evolution of myocardial infarction repair [6]. While elastin replacement improves myocardial compliance and helps preserve ventricular ejection fraction, this metric alone did not predict changes in post infarction EDV. Together, the PFC signal and elastin on day 7 were significantly but weakly correlated with cardiac remodeling outcomes. Ramos et al[3] have elegantly demonstrated the use of dual contrast imaging techniques to characterize mid to late post-MI pathophysiology, and perhaps this noninvasive interrogation technique could be applied to earlier inflammatory resolution events with development of new biomarker probes.

$^{19}F/^{1}H$ MRI imaging

Ramos et al[3] utilize ^{19}F imaging to assess macrophage migration into infarcted myocardium as a wise alternative to more commonly used iron oxide nanoparticles, avoiding magnetic susceptibility artifacts that might otherwise interfere with the Gd-ESMA elastin probe results. The history of MR fluorine (^{19}F) imaging and spectroscopy began during the infancy of ^{1}H MR imaging and was recently comprehensively reviewed[15].

The overall experimental approach by Ramos et al[3] has a strong translational bent, utilizing a clinical 3T scanner in conjunction with a custom $^{19}F/^{1}H$ coil. As common in many preclinical ^{19}F studies, the investigators utilized perfluoro-15-crown-5 ether (PFCE), which offers 20 chemically equivalent fluorine atoms to improve contrast signal-to-noise ratio (SNR) and detectability[16,17]. Similar perfluorocarbon compounds, such as perfluoropolyether (PFPE) with up to 40 chemically equivalent fluorine atoms have been used for clinical cell tracking studies[18–21]. However, in those studies PFC cell labeling occurred ex vivo and the total amount of PFC administered was very small. Ramos et al[3] used large doses of PFC nanoparticles to achieve in vivo monocyte labeling, comparable to the levels used for artificial blood substitute applications. PFCs are biologically inert and bioeliminated in man through pulmonary exhalation. PFCE has a prolonged biological persistence and at high volumes is associated with reduced pulmonary compliance and residual alveolar gas trapping as well as adverse effects from reticuloendothelial organ congestion and cytokines released from engorged phagocytic cells. Developers of artificial blood substitutes used perflubron (perfluoro-bromo-octane, PFOB) and similar fluorocarbons[22–24] in part to lessen the respiratory effects of PFCs. Although PFOB has a long clinical trial record, its multispectral ^{19}F signal complicates MR data acquisition.

However, the proof-of-concept of application demonstrated Ramos et al[3] for PFCE emulsions could be extended to multispectral compounds like PFOB for human use. Full

spectral information MR acquisition technique have been proposed and demonstrated but they are generally complicated or simply slow[25–28]. A preferred clinical imaging approach could be ultra-short echo time (UTE) imaging which ultra-rapidly interrogates 12 of 17 F nuclei of PFOB before significant signal loss occurs[29–31]. Ultrafast imaging capability on modern clinical MR instrumentation could enable a renaissance and translation of ^{19}F imaging.

Ramos et al[3] utilized independent scans to acquire ^{1}H (i.e., elastin) and ^{19}F (i.e., macrophage) image datasets, which were then co-registered retrospectively. ^{1}H MR data acquisition is achieved quickly due to the rich abundance of water protons, but longer scan times are required to accumulate data for sparser ^{19}F nuclei. In mice imaged with a clinical 3T scanner, cardiac motion is typical less than the voxel size, which helps to minimize mis-registration and blurring of the ^{19}F data relative to the proton image. However, in man cardiac motion will be a significant translational challenge dual-contrast MR tissue characterization. Fortunately, techniques for simultaneous ^{1}H and ^{19}F imaging using dual tuned coils have been developed and implemented on clinical 3T scanners similar to the one used by Ramos et al.[3] One-to-one correspondence of ^{1}H and ^{19}F acquisitions achieved with simultaneous imaging allows motion adjustments based on ^{1}H image dataset to correct for motion in the ^{19}F images[32]. With clinical availability of Gd-ESMA and PFC nanoparticles and the updating of MR scanners for robust $^{9}F/^{1}H$ MR imaging, new personalized approaches to patient management draw closer to reality.

Conclusion

Ramos et al[3] offer a multinuclear proof of concept molecular imaging report performed on a 3T clinical MR scanner demonstrating the noninvasive diagnostic potential of MR to characterize the pathophysiology of post infarction myocardium with quantitative metrics. Their approaches captured the temporal and spatial dynamics of inflammatory macrophage accumulation in healing ischemic tissues as well as the reconstitution of stiff collagen-rich scar with elastic fibers to retain ventricular compliance and elasticity. Overall, the report reflects the continuing interest and efforts of MR scientists to utilize noninvasive imaging to personalize and improve patient outcomes.

Acknowledgments

Sources of Funding: This study was supported by R01 CA216840 from the National Institutes of Health.

References:

1. Lima JA, Judd RM, Bazille A, Schulman SP, Atalar E, Zerhouni EA. Regional heterogeneity of human myocardial infarcts demonstrated by contrast-enhanced MRI. Potential mechanisms. Circulation. 1995;92:1117–25. [PubMed: 7648655]
2. Kim RJ, Fieno DS, Parrish TB, Harris K, Chen EL, Simonetti O, Bundy J, Finn JP, Klocke FJ, Judd RM. Relationship of MRI delayed contrast enhancement to irreversible injury, infarct age, and contractile function. Circulation. 1999;100:1992–2002. [PubMed: 10556226]
3. Ramos IT, Henningsson M, Nezafat M, Lavin B, Lorrio S, Gebhardt P, Protti A, Eykyn TR, Andia ME, Flögel U, Phinikaridou A, Shah AM, Botnar RM. Simultaneous Assessment of Cardiac Inflammation and Extracellular Matrix Remodeling after Myocardial Infarction. Circ Cardiovasc Imaging. 2018;11:e007453. [PubMed: 30524648]

4. Flogel U, Ding Z, Hardung H, Jander S, Reichmann G, Jacoby C, Schubert R, Schrader J. In vivo monitoring of inflammation after cardiac and cerebral ischemia by fluorine magnetic resonance imaging. Circulation. 2008;118:140–148. [PubMed: 18574049]
5. von Bary C, Makowski M, Preissel A, Keithahn A, Warley A, Spuentrup E, Buecker A, Lazewatsky J, Cesati R, Onthank D, Schickl N, Schachoff S, Hausleiter J, Schomig A, Schwaiger M, Robinson S, Botnar R. MRI of coronary wall remodeling in a swine model of coronary injury using an elastin-binding contrast agent. Circ Cardiovasc Imaging. 2011;4:147–55. [PubMed: 21378029]
6. Frangogiannis NG. Pathophysiology of Myocardial Infarction. Compr Physiol 2015;5:1841–75. [PubMed: 26426469]
7. Serhan CN, Petasis NA. Resolvins and protectins in inflammation resolution. Chem Rev. 2011;111:5922–43. [PubMed: 21766791]
8. Serhan CN. Pro-resolving lipid mediators are leads for resolution physiology. Nature. 2014;510:92–101. [PubMed: 24899309]
9. Lai L, Alaverdi N, Maltais L, Morse HC, 3rd. Mouse cell surface antigens: nomenclature and immunophenotyping. J Immunol. 1998;160:3861–8. [PubMed: 9558091]
10. Geissmann F, Jung S, Littman DR. Blood monocytes consist of two principal subsets with distinct migratory properties. Immunity. 2003;19:71–82. [PubMed: 12871640]
11. Temme S, Bonner F, Schrader J, Flogel U. 19F magnetic resonance imaging of endogenous macrophages in inflammation. Wiley Interdiscip Rev Nanomed Nanobiotechnol. 2012;4:329–43. [PubMed: 22354793]
12. Mizuno T, Yau TM, Weisel RD, Kiani CG, Li RK. Elastin stabilizes an infarct and preserves ventricular function. Circulation. 2005;112:I81–8. [PubMed: 16159870]
13. Yu Y, Yin G, Bao S, Guo Z. Kinetic alterations of collagen and elastic fibres and their association with cardiac function in acute myocardial infarction. Mol Med Rep. 2018;17:3519–3526. [PubMed: 29286107]
14. Li SH, Sun Z, Guo L, Han M, Wood MF, Ghosh N, Vitkin IA, Weisel RD, Li RK. Elastin overexpression by cell-based gene therapy preserves matrix and prevents cardiac dilation. J Cell Mol Med. 2012;16:2429–39. [PubMed: 22435995]
15. Schmieder AH, Caruthers SD, Keupp J, Wickline SA, Lanza GM. Recent advances in ^{19}Fluorine magnetic resonance imaging with perfluorocarbon emulsions. Engineering. 2015;1:475–489. [PubMed: 27110430]
16. Ahrens ET and Bulte JWM. Tracking immune cells in vivo using magnetic resonance imaging. Nat Rev Immunol. 2013;13:755–763. [PubMed: 24013185]
17. Zhong J, Mills PH, Hitchens TK, Ahrens ET. Accelerated fluorine-19 MRI cell tracking using compressed sensing. Magn Reson Med. 2013;69:1683–1690. [PubMed: 22837054]
18. Srinivas M, Morel PA, Ernst LA, Laidlaw DH, Ahrens ET. Fluorine-19 MRI for visualization and quantification of cell migration in a diabetes model. Magn Reson Med. 2007;58:725–34. [PubMed: 17899609]
19. Janjic JM, Srinivas M, Kadayakkara DKK, Ahrens ET. Self-delivering nanoemulsions for dual fluorine-19 MRI and fluorescence detection. J Am Chem Soc. 2008;130:2832–2841. [PubMed: 18266363]
20. Ahrens ET, Helfer BM, O'Hanlon CF, Schirda C. Clinical cell therapy imaging using a perfluorocarbon tracer and fluorine-19 MRI. Magn Reson Med. 2014;72:1696–1701. [PubMed: 25241945]
21. Kadayakkara DK, Damodaran K, Hitchens TK, Bulte JWM, Ahrens ET. ^{19}F spin-lattice relaxation of perfluoropolyethers: Dependence on temperature and magnetic field strength (7.0–14.1 T). J Magn Reson. 2014;242:18–22. [PubMed: 24594752]
22. Leach CL, Greenspan JS, Rubenstein SD, Shaffer TH, Wolfson MR, Jackson JC, DeLemos R, Fuhrman BP. Partial liquid ventilation with perflubron in premature infants with severe respiratory distress syndrome. The LiquiVent Study Group. N Engl J Med. 1996;335:761–7. [PubMed: 8778584]
23. Lowe KC. Perfluorinated blood substitutes and artificial oxygen carriers. Blood Rev. 1999;13:171–84. [PubMed: 10527269]

24. Riess JG. Understanding the fundamentals of perfluorocarbons and perfluorocarbon emulsions relevant to in vivo oxygen delivery. Artif Cells Blood Substit Immobil Biotechnol. 2005;33:47–63. [PubMed: 15768565]

25. Reeder SB, Herzka DA, McVeigh ER. Signal-to-noise ratio behavior of steady-state free precession. Magn Reson Med. 2004;52:123–30. [PubMed: 15236375]

26. Yu J, Ishii M, Law M, Woodburn JM, Emami K, Kadlecek S, Vahdat V, Guyer RA, Rizi RR. Optimization of scan parameters in pulmonary partial pressure oxygen measurement by hyperpolarized ^{3}He MRI. Magn Reson Med. 2008;59:124–31. [PubMed: 18050348]

27. Keupp J, Mazurkewitz P, Frasslin, Schaeffter T. Simultaneous ^{19}F and ^{1}H imaging on a clinical 3T MR scanner. Proc Int Soc Magn Reson Med. 2006;15:102.

28. Lamerichs R, Yildirim M, Nederveen A, Stoker J, Lanza G, Wickline S, Caruthers S. In vivo 3D ^{19}F fast spectroscopic imaging (F-uTSI) of angiogenesis on Vx-2 tumors in rabbits using targeted perfluorocarbon emulsions. Proc Int Soc Magn Reson Med. 2010;18:457.

29. Rahmer J, Bornert P, Groen J, Bos C. Three-dimensional radial ultrashort echo-time imaging with T2 adapted sampling. Magn Reson Med. 2006;55:1075–82. [PubMed: 16538604]

30. Schmid F, Höltke C, Parker D, Faber C. Boosting ^{19}F MRI - SNR efficient detection of paramagnetic contrast agents using ultrafast sequences. Magn Reson Med. 2013;69:1056–1062. [PubMed: 22628001]

31. Goette MJ, Keupp J, Rahmer J, Lanza GM, Wickline SA, Caruthers SD. Balanced UTE-SSFP for ^{19}F MR imaging of complex spectra. Magn Reson Med. 2015;74:537–543. [PubMed: 25163853]

32. Keupp J, Rahmer J, Grässlin I, Mazurkewitz PC, Schaeffter T, Lanza GM, Wickline SA, Caruthers SD. Simultaneous dual-nuclei imaging for motion corrected detection and quantification of ^{19}F imaging agents. Magn Reson Med. 2011;66:1116–1122. [PubMed: 21394779]