Journal of the American College of Cardiology
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Published by Elsevier Inc.

Vol. 58, No. 6, 2011
ISSN 0735-1097/$36.00
doi:10.1016/j.jacc.2011.04.021

EDITORIAL COMMENT

Imaging Atherosclerosis With F18-Fluorodeoxyglucose Positron Emission Tomography

What Are We Actually Seeing?*

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Coronary heart disease remains the leading cause of death in the U.S. Despite advances in diagnostic imaging and therapy, identification of patients at risk remains challenging. Anatomical imaging of luminal narrowing and determination of plaque size alone have proven to be inadequate (1). In contrast, molecular imaging of plaque composition and metabolism may not only identify vulnerable patients, but also enable therapeutic discovery and monitoring (2).

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Over the past 2 decades, it has become clear that inflammation plays a critical role in plaque destabilization, a process that is also an obvious target for molecular imaging. Plaques vulnerable to rupture have a large hypoxic, metabolically active core, containing lipid, oxidized lipid, and numerous inflammatory cells (primarily foam cells or macrophages). Inflammatory cells secrete proteolytic enzymes and cytokines, which weaken the thin fibrous cap of these vulnerable plaques (3). Disruption of fibrous cap allows thrombotic and inflammatory material in the lesion to contact blood and form a critical thrombus, occluding coronary flow and resulting in acute coronary syndrome and sudden death (4).

Although only largely circumstantial evidence exists so far, F18-fluorodeoxyglucose (FDG) positron emission tomography (PET) imaging has emerged as the most promising tool for imaging plaque inflammation and vulnerability (5). Studies in inflammatory diseases have found that activated inflammatory cells have increased expression of glucose transporters and FDG uptake (6). Similarly, in clinical and preclinical studies of atherosclerosis, areas of high macrophage density correlate with enhanced FDG uptake in vessels with plaque compared to the contralateral vessels without plaque (7,8). In addition, FDG uptake is increased after in vitro stimulation of macrophages with inflammatory cytokines and growth factors, which are present in heightened levels in atheroma (9–11). Finally, statins, which have anti-inflammatory properties, reduce FDG signal (12). Together, these data tentatively suggest that FDG signal in plaque is caused by inflammation.

What Is the Source and Mechanism of Increased FDG Uptake?

Important questions regarding the source and mechanism of FDG uptake in the atherosclerotic plaque remain unanswered. Are macrophages the only cells capable of taking up FDG? What is the relative contribution of different cells to the FDG signal? What causes cells to increase their FDG uptake (e.g., inflammation or hypoxia or both)? Knowing the source and mechanism of increased FDG signal will help us understand the meaning of changes in FDG signal and its value as a surrogate endpoint.

In this issue of the *Journal*, Folco et al. (13) help answer some of these important questions. Using both an in vitro culture system and ex vivo carotid endarterectomy specimens, they evaluated the effects of hypoxia and inflammation on FDG uptake in cells specific to atherosclerotic lesions. They found that both smooth muscle and endothelial cells increased glucose uptake when exposed to inflammatory cytokines. Surprisingly, macrophages increased glucose uptake in response to hypoxia but not to inflammatory cytokines. They further demonstrated that the primary mechanism for increased glucose uptake was induction of hexokinase 2, an enzyme that phosphorylates glucose and enables cell entry. Interestingly, administration of statins reduced basal as well as hypoxia induced glucose uptake, suggesting that statins have direct effects on macrophages independent of their anti-inflammatory effects. These findings challenge the prevailing belief that FDG uptake is a measure of inflammation.

This paper has several strengths. First, the authors carefully controlled for confounding factors that might affect FDG uptake, including glucose concentration and FDG incubation duration (10). Cells were depleted of endogenous glucose before labeled FDG was added. The FDG incubation duration was limited to 5 min to ensure linear uptake of glucose. The short incubation time could explain why minimal uptake was seen after stimulation with inflammatory cytokines. In contrast, after a 30 min FDG incubation time, a previous study showed that >250% FDG uptake occurred in monocytes stimulated with IFNγ compared with controls. Second, the authors carefully regulated the amount and duration of hypoxia and cytokines, 2 factors that may affect the degree of cell activation and glucose

*Editorials published in the *Journal of the American College of Cardiology* reflect the views of the authors and do not necessarily represent the views of *JACC* or the American College of Cardiology.

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uptake (14). Third, the authors investigated the mechanism by which hypoxia increased FDG uptake by analyzing the expression and activity of glucose transporters and hexokinases. Finally, the authors confirmed the viability of inflammatory, endothelial, and smooth cells, which ensures cell death did not confound rates of glucose metabolism.

Shedding Light on Current Controversies

Although the studies described in the preceding text were performed in vitro, the findings nevertheless may help resolve current controversies for the application of in vivo FDG PET imaging in atherosclerosis. The first controversy is why FDG signal correlates mainly with macrophage density in studies using endarterectomy specimens and balloon-injured hypercholesterolemic rabbits even though endothelial and smooth muscle cells can also metabolize glucose in vitro (9–11,15). This may occur because macrophages are more abundant than these other cell types in atheroma. In addition, they may preferentially uptake FDG because these cells appear to respond to both inflammatory cytokines and hypoxia. The second controversy is why FDG signal appears to vary with plaque composition. A previous study has shown that plaques with a predominant lipid necrotic component and, therefore, a more hypoxic environment have higher uptake than those with fibrous or calcified plaques (16). Interestingly, FDG uptake also colocalizes with calcifications, which may develop secondary to chronic hypoxia. Furthermore, FDG uptake correlates with microvessel density and angiogenesis, both of which are related to the degree of hypoxia (17). Finally, the percentage of FDG-positive vascular segments and the amount of uptake increase with age, which has also been linked to hypoxia (18). Thus, plaques with varying degrees of hypoxia may have different amounts of FDG uptake. The final, and perhaps the most important unresolved issue, is why results from FDG PET in vivo imaging studies of atherosclerosis are often inconsistent. For example, although the majority of clinical studies have found that FDG signal is increased in vessels with plaque compared with the contralateral vessel, other studies have not found such an association (19). The presence of FDG has been associated with age, hypercholesterolemia, and the metabolic syndrome, but not with smoking, diabetes mellitus, hypertension, and obesity. In addition, the extent and intensity of the FDG signal do not necessarily correlate with cardiovascular risk factors (19). Because FDG uptake appears to depend on multiple factors, the inconsistencies found in clinical studies are not surprising.

Implications for In Vivo FDG PET Imaging for Atherosclerosis

Taken together, the study by Folco et al. (13) also raises important questions for in vivo FDG PET imaging for atherosclerosis. The first question is whether FDG signal can still be used as a measure of plaque vulnerability. Previous studies have shown that hypoxia exists in macrophage-dense regions of animal and human atherosclerosis (20). Hypoxia may result from 2 primary causes: 1) limited oxygen supply, which occurs when the intimal thickness exceeds the maximal oxygen diffusion capacity; or 2) increased demand due to high metabolic activity within macrophages (21,22). Hypoxia in macrophages has been shown to increase angiogenesis as well as cytokine production, low-density lipoprotein oxidation, and lipid loading in vitro, all of which contribute to plaque destabilization (22–24). Hence, it is likely that FDG PET can still evaluate plaque vulnerability, as both inflammation and hypoxia are associated with plaque growth and instability.

The second question is whether FDG signal is an accurate measure of vulnerability. Macrophages activated by inflammatory cytokines may not be measured if cells do not have enough time to take up FDG. In addition, FDG has been shown to underestimate hypoxia compared to more specific hypoxic markers, based on findings from a previous oncologic study (25).

The third question that arises from these findings is the value of FDG signal in monitoring therapy. Although changes in FDG signal can still be used to monitor therapy, reduction in FDG signal is not necessarily due to reduction in inflammation, as shown in this study, where statins reduced the sensitivity of macrophages to hypoxia but not because of their anti-inflammatory effects. To use FDG PET for monitoring, it will be important to differentiate the effects of a potential drug on decreasing macrophage number, reaction to hypoxia, and response to inflammatory cytokines.

Finally, perhaps the most important question is whether FDG PET is a valuable surrogate endpoint. Findings from 2 prospective studies with 6-month follow-up suggest that high FDG signal is associated with higher incidence of clinical events (26,27). The clinical value of FDG PET imaging will be better elucidated after completion of the High Risk Plaque Initiative, a prospective event-driven study involving 6,500 volunteers at risk for cardiovascular disease (5). It will also be important to perform clinical trials to determine whether changes in FDG signal after medical therapy are associated with improvements in morbidity and mortality.

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REFERENCES

1. Topol EJ, Nissen SE. Our preoccupation with coronary luminology. The dissociation between clinical and angiographic findings in ischemic heart disease. Circulation 1995;92:2333–42.
2. Chen IY, Wu JC. Cardiovascular molecular imaging: focus on clinical translation. Circulation 2011;123:425–43.

3. Burke AP, Farb A, Malcolm G, et al. Coronary risk factors and plaque morphology in men with coronary disease who died suddenly. N Engl J Med 1997;336:1276–82.
4. Kovanen PT. Mast cells and degradation of pericellular and extracellular matrices: potential contributions to erosion, rupture and intraplaque haemorrhage of atherosclerotic plaques. Biochem Soc Trans 2007;35:857–61.
5. Rudd JH, Narula J, Strauss HW, et al. Imaging atherosclerotic plaque inflammation by fluorodeoxyglucose with positron emission tomography: ready for prime time? J Am Coll Cardiol 2010;55:2527–35.
6. Love C, Tomas MB, Tronco GG, Palestro CJ. FDG PET of infection and inflammation. Radiographics 2005;25:1357–68.
7. Rudd JH, Warburton EA, Fryer TD, et al. Imaging atherosclerotic plaque inflammation with [18F]-fluorodeoxyglucose positron emission tomography. Circulation 2002;105:2708–11.
8. Tawakol A, Migrino RQ, Hoffmann U, et al. Noninvasive in vivo measurement of vascular inflammation with F-18 fluorodeoxyglucose positron emission tomography. J Nucl Cardiol 2005;12:294–301.
9. Fukuzumi M, Shinomiya H, Shimizu Y, Ohishi K, Utsumi S. Endotoxin-induced enhancement of glucose influx into murine peritoneal macrophages via GLUT1. Infect Immun 1996;64:108–12.
10. Deichen JT, Prante O, Gack M, Schmiedehausen K, Kuwert T. Uptake of [18F]fluorodeoxyglucose in human monocyte-macrophages in vitro. Eur J Nucl Med Mol Imaging 2003;30:267–73.
11. Paik JY, Lee KH, Choe YS, Choi Y, Kim BT. Augmented 18F-FDG uptake in activated monocytes occurs during the priming process and involves tyrosine kinases and protein kinase C. J Nucl Med 2004;45:124–8.
12. Tahara N, Kai H, Ishibashi M, et al. Simvastatin attenuates plaque inflammation: evaluation by fluorodeoxyglucose positron emission tomography. J Am Coll Cardiol 2006;48:1825–31.
13. Folco EJ, Sheikine Y, Rocha VZ, et al. Hypoxia but not inflammation augments glucose uptake in human macrophages: implications for imaging atherosclerosis with FdG-positron emission tomography. J Am Coll Cardiol 2011;58:603–14.
14. Matsui T, Nakata N, Nagai S, et al. Inflammatory cytokines and hypoxia contribute to 18F-FDG uptake by cells involved in pannus formation in rheumatoid arthritis. J Nucl Med 2009;50:920–6.
15. Maschauer S, Prante O, Hoffmann M, Deichen JT, Kuwert T. Characterization of 18F-FDG uptake in human endothelial cells in vitro. J Nucl Med 2004;45:455–60.
16. Silvera SS, Aidi HE, Rudd JH, et al. Multimodality imaging of atherosclerotic plaque activity and composition using FDG-PET/CT and MRI in carotid and femoral arteries. Atherosclerosis 2009;207:139–43.
17. Laitinen I, Marjamaki P, Haaparanta M, et al. Non-specific binding of [18F]FDG to calcifications in atherosclerotic plaques: experimental study of mouse and human arteries. Eur J Nucl Med Mol Imaging 2006;33:1461–7.
18. Bural GG, Torigian DA, Chamroonrat W, et al. FDG-PET is an effective imaging modality to detect and quantify age-related atherosclerosis in large arteries. Eur J Nucl Med Mol Imaging 2008;35:562–9.
19. Sheikine Y, Akram K. FDG-PET imaging of atherosclerosis: do we know what we see? Atherosclerosis 2010;211:371–80.
20. Sluimer JC, Gasc JM, van Wanroij JL, et al. Hypoxia, hypoxia-inducible transcription factor, and macrophages in human atherosclerotic plaques are correlated with intraplaque angiogenesis. J Am Coll Cardiol 2008;51:1258–65.
21. Torres Filho IP, Leunig M, Yuan F, Intaglietta M, Jain RK. Noninvasive measurement of microvascular and interstitial oxygen profiles in a human tumor in SCID mice. Proc Natl Acad Sci U S A 1994;91:2081–5.
22. Murdoch C, Muthana M, Lewis CE. Hypoxia regulates macrophage functions in inflammation. J Immunol 2005;175:6257–63.
23. Bostrom P, Magnusson B, Svensson PA, et al. Hypoxia converts human macrophages into triglyceride-loaded foam cells. Arterioscler Thromb Vasc Biol 2006;26:1871–6.
24. Rydberg EK, Krettek A, Ullstrom C, et al. Hypoxia increases LDL oxidation and expression of 15-lipoxygenase-2 in human macrophages. Arterioscler Thromb Vasc Biol 2004;24:2040–5.
25. Christian N, Deheneffe S, Bol A, et al. Is (18)F-FDG a surrogate tracer to measure tumor hypoxia? Comparison with the hypoxic tracer (14)C-EF3 in animal tumor models. Radiother Oncol 2010;97:183–8.
26. Arauz A, Hoyos L, Zenteno M, Mendoza R, Alexanderson E. Carotid plaque inflammation detected by 18F-fluorodeoxyglucose-positron emission tomography. Pilot study. Clin Neurol Neurosurg 2007;109:409–12.
27. Rominger A, Saam T, Wolpers S, et al. 18F-FDG PET/CT identifies patients at risk for future vascular events in an otherwise asymptomatic cohort with neoplastic disease. J Nucl Med 2009;50:1611–20.

Key Words: atherosclerosis ■ hypoxia ■ inflammation ■ positron emission tomography.