PET tracers and techniques for measuring myocardial blood flow in patients with coronary artery disease

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Abstract
Assessment of the relative distribution of myocardial flow with myocardial perfusion imaging (MPI) is methodologically limited to predict the presence or absence of flow-limited coronary artery disease (CAD). This limitation may often occur, when obstructive lesions involve multiple epicardial coronary arteries or disease-related disturbances of the coronary circulation coexist at the microvascular level. Non-invasive assessment of myocardial blood flow in absolute units with position emission tomography (PET) has been positioned as the solution to improve CAD diagnosis and prediction of patient outcomes associated with risks for cardiac events. This article reviews technical and clinical aspects of myocardial blood flow quantitation with PET and discusses the practical consideration of this approach toward worldwide clinical utilization.

Keywords: myocardial blood flow, PET, coronary artery disease

PET MYOCARDIAL PERFUSION TRACERS
So far, there are several myocardial perfusion radiopharmaceuticals qualified for PET flow imaging, such as $^{15}$O-water, $^{13}$N-ammonia and $^{82}$Rb. Differences in the first-pass extraction of these tracers determine their regional myocardial uptake in relation to regional blood flow (Fig. 1). Resting myocardial blood flow (MBF) measured with these tracers in healthy human is approximately 1.0 mL/(min • g) while MBF increases three fold or higher than 3.0 mL/(min • g) under pharmacological stress with adenosine or dipyridamole[1-3]. $^{15}$O-water as an ideal myocardial flow tracer exhibits a linear relation to MBF over a wide range of flow rates while $^{13}$N-ammonia and $^{82}$Rb as two more commonly used tracers in routine clinical environment do not exhibit such a linear property[4]. Because of the non-linearity, roll-off of tracer uptake in the myocardium can result in underestimated calculation of regional myocardial blood flow at high flow levels. To accurately quantify MBF, it is necessary to apply proper physiological compensation for the non-linear relation between uptake and MBF.

Post intravenous injection, myocardial extraction fraction of $^{15}$O-water approaches unity since its net uptake (the product of the first-pass tracer extraction) tracks linearly with myocardial blood flow[5-7]. To permit clinical utilization of $^{15}$O-water with a short physical half-life of 125 seconds, an on-site cyclotron near a PET imaging system is required. Perfusion images of $^{15}$O-water normally present low target to background ratios because of the phenomenon of equilibrated diffusion between adjacent water spaces (e.g. myocyte...
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...and blood pool). $^{15}$O-water is not typically used for assessment of myocardial perfusion alone, but mostly with measurements of MBF. Technically, the correction for high blood pool activity can be achieved by subtracting the images acquired from labeled arterial blood pool with inhalation of $^{15}$O or $^{11}$C carbon monoxide. However, the complexities of this procedure still encounter practical limitations for routine clinical utilization. Alternatively, novel analytical approaches based on factor analysis methods can be employed to simplify the process.$^{[8-10]}$ Since the measurement of MBF with $^{15}$O-water PET is fairly accurate, it has been widely accepted as a non-invasive gold standard for flow assessment over several decades.

$^{13}$N-ammonia is another myocardial perfusion tracer delivered to the myocardium with intravenous injection and retained metabolically in the myocardium in proportion to MBF.$^{[11,12]}$ $^{13}$N-ammonia exchanges across the capillary wall and transits through the interstitial spaces to reach the myocardial cell. Its first capillary transient retention fraction reaches about 85% for rest MBF but progressively and nonlinearly declines with increased blood flows. After entering myocyte, a fraction of tracer can diffuse back from tissue into blood while another fraction becomes metabolically trapped and retained in the myocardium through the α-ketoglutarate-to-glutamate and the glutamate-to-glutamine reactions.$^{[11]}$ Since the tracer is retained in the myocardium with rapid clearance from blood pool, perfusion images with high diagnostic quality can be obtained. High uptake in the lung and liver may also be observed in patients. In general, over a few to ten minutes, $^{13}$N-ammonia concentration in the myocardium remains retained before the loss of $^{13}$N-labeled glutamine from the myocardium may occur. $^{13}$N-ammonia’s physical half-life of 10 minutes permits repeated evaluations of rest and stress MBF at relatively short time intervals (about 30-40 minutes). Additionally, the tracer allows assessment of stress perfusion and left ventricular function with imaging protocols of treadmill exercise. Like $^{15}$O-water, flow quantitation with $^{13}$N-ammonia PET has also been widely accepted as another non-invasive gold standard for flow assessment. In fact, $^{13}$N-ammonia is usually preferred rather than $^{15}$O-water for qualitative evaluations of myocardial flow and perfusion images because of its superior image quality and simplified imaging protocols for clinical utilization. Nonetheless, because of the short physical half-life, $^{13}$N-ammonia production still requires an on-site cyclotron.

Unlike $^{15}$O-water and $^{13}$N-ammonia, $^{82}$Rb with an ultra short physical half-life of 75 seconds is a generator-produced myocardial perfusion PET tracer.$^{[13]}$ It is the decayed product from $^{82}$Sr with a physical half-life of 28 days, which allows the clinical use of a generator system to produce $^{82}$Rb for as long as 4 to 5 weeks. Since the biologic properties of $^{82}$Rb as a positron-emitting cation are similar to those of potassium, intracellular uptake of $^{82}$Rb across the sarcolemmal membrane can reflect the activity of cation transport via the Na-K ATPase transport system. When transported into myocyte, $^{82}$Rb is retained in the myocardium in proportion to MBF. The first-pass retention fraction of $^{82}$Rb reaches 65% for MBF at

![Fig. 1 Schematic illustration of radiotracer uptake in relation to regional myocardial blood flow.](image)
rest but declines corresponding to higher flow rates. In patients with chronic CAD, myocardial uptake of \(^{82}\text{Rb}\) is preserved in viable regions and largely reduced in scarred regions. In the setting of acute myocardial injury and reperfusion, initial uptake of \(^{82}\text{Rb}\) can reflect the amount of recovered blood flow in revived area. Since necrotic myocardium cannot retain \(^{82}\text{Rb}\), the kinetics of \(^{82}\text{Rb}\) washout may be utilized as an index of myocardial viability\(^{[19]}\). In the clinical setting, 40 to 60 mCi of \(^{82}\text{Rb}\) is administered intravenously with a sophisticated infusion system to deliver targeted activities from the \(^{82}\text{Sr}\) column\(^{[13]}\). After infusion of \(^{82}\text{Rb}\), data acquisition for perfusion images usually commences 60 to 120 seconds and continues for about 6 minutes\(^{[13,15]}\). Perfusion images with \(^{82}\text{Rb}\) generally have a good diagnostic quality and can detect flow abnormalities with a similar accuracy to that of \(^{13}\text{N-ammonia}\)\(^{[16,17]}\). However, patients with low left ventricular ejection fraction (LVEF) or severe lung disease may result in slower blood clearance to affect image quality. A general strategy to overcome this issue is to extend the waiting time beyond 120 seconds to collect perfusion data. Because of the ultra short physical half-life, infusion of high dose activities to attain statistically adequate images is generally necessary for \(^{82}\text{Rb}\) PET imaging, but the high dose infusion, on the other hand, generates high count rates during the acquisition of input function. The over-exceeded count rates can lead to substantial dead-time losses for many current PET or PET/CT imaging systems, especially when they are operated in a 3D acquisition mode. Nevertheless, measurements of MBF in absolute units are still possible with low-dose protocols\(^{[18,19]}\). Because of the superiority of image quality and diagnostic accuracy over the traditional MPI\(^{[20]}\) and the simplicity of on-site \(^{82}\text{Rb}\) generator for clinical utilization, \(^{82}\text{Rb}\) has been widely utilized in North America to clinically facilitate detection of CAD in patients suitable for pharmacological stress.

**IMAGING METHODS**

The high spatial and temporal resolutions of PET imaging with the capability of image quantitation are the essentials for MBF measurements. Rapidly acquired dynamic images (typically with 5-10 second frame rates) for several to ten minutes are necessary to track the initial transit of the radiotracer bolus through the central circulation and its continuous exchange from blood into the myocardium. In general, PET dynamic data acquisition should be started at least a few to ten seconds prior to the injection or infusion of PET tracers in order to establish the reference time point before tracer entering the imaging field of view. For stress flow measurement, it is crucial to inject PET tracers when the myocardium reaches the hyperemia stage with pharmacological stimulation or cold presser testing.

To produce quantitative dynamic images for accurate flow measurement, it is demanded to correct for physical interferences with in the PET images due to attenuation, scatter, randoms, and dead-time loss for high dose injection. Attenuation correction for dedicated PET systems are normally achieved by applying attenuation maps sequentially acquired by rotating multiple radionuclide rod sources (e.g. \(^{68}\text{Ge}\)) or a single radioactive point source (e.g. \(^{137}\text{Cs}\)) around patients. For PET/CT systems, separated low-dose CT images are acquired for the generation of attenuation maps. Quality control of misregistration and associated correction between emission images and attenuation maps acquired from either radionuclide transmission or CT is mandatory; otherwise, flow values would be systematically underestimated in regions with attenuation artifacts\(^{[21]}\). Scatter correction is generally achieved by modeling 511-keV scatter photons with reconstructed activity map of tracer distribution using the Monte-Carlo simulation process\(^{[22,23]}\). Randoms correction is usually applied to the sinogram raw data by estimating randoms in PET images based on singles rates measured for paired detector elements\(^{[24,25]}\). Compensation for the dead-time loss is important, particularly for high activity injection. It can be achieved by compensating singles count loss with the non-linear relation of singles count rate to activity presented in the imaging field of view. With all physical corrections applied, quantitative PET images with minimized physical interferences can be obtained. To depict the true regional radiotracer activity concentrations, pixel values in the corrected images are quantitatively presented in a physical unit of (Bq/mL). Quantitative images can be further employed to derive time activity curves of the arterial radiotracer input function and myocardial tissue response for flow calculation. In the process of quantitation, time activity curves are fitted with operational equations derived from tracer kinetic models (described below), which relate the externally observed tracer uptake to absolute MBF in the unit of mL/min/g through tissue kinetics. To utilize quantitative flow values for CAD detection, it is applicable to display regional estimates of MBF in the form of color-coded parametric images for clinical utilization\(^{[26]}\).

**PHYSIOLOGICAL FLOW MODELS**

Among non-invasive medical imaging modalities for flow assessment, modeling tracer kinetics is a commonly used technique to simplify physiological proc-
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ess of tracer uptake in mathematical terms for MBF calculation. Depending on the mechanism of tracer uptake from capillaries into myocyte, tracer kinetics can be described by one or two tissue compartments with multiple kinetic parameters as illustrated in Fig. 2. In general, ¹⁵O-water and ¹³⁵Rb only require two kinetic parameters, $K_1$ (mL/min/g) and $K_2$ (min⁻¹), to calculate the rate of tracer uptake from blood to tissue and the rate of tracer washout from the tissue. ¹³¹⁵N-ammonia demands an additional parameter, $K_3$ (min⁻¹), to depict the rate of metabolic process in myocyte as previously described.

In the compartmental flow model consisting of $K_1$ and $K_2$[^27,28], time activity curves derived from the left ventricular blood pool and myocardium are fitted into the flow equation as:

$$C_m(t) = FBV \cdot C_a(t) + (1-FBV) \cdot K_1 \cdot e^{-K_1 \cdot t} \cdot Ca(t)$$

where $K_1$ (ml/min/g) and $K_2$ (min⁻¹) are two kinetic parameters characterizing the rate of tracer uptake from the blood to the myocardium and the rate of tracer washout from the myocardium, respectively. $C_m(t)$ is the measured activity concentration in the myocardium obtained from PET images, assumed to consist of arterial blood input $C_a(t)$ and true myocardial uptake as a convolution of $K_1$, $K_2$ and $C_a(t)$. FBV is referred to the fractional blood volume in $C_m(t)$ coming from $C_a(t)$, and (1-FBV) is the rest of faction contributed from the myocardial uptake. By applying the curve fitting process, $K_1$, $K_2$ and FBV can be exactly solved into numerical values. MBF (mL/min/g) is then converted from $K_1$ with additional compensation for tracer extraction (E) in myocardium:

$$K_1 = MBF \cdot E = MBF \cdot (1 - \alpha \cdot e^{2/MBF})$$

where $\alpha$ and $\beta$ are physiological parameters derived from the effective capillary permeability surface (PS) area product (mL/min/g) accounting for nonlinear tracer extraction as a function of MBF[^29,30]. The assumption in this model is consistent with the observed tracer extraction, which typically decreases with flow as previously described. To compensate for the physiological variance (e.g. hypertension or hypotension), rest blood flow is corrected for baseline heart rate and blood pressure by the factor of rate-pressure product/ (10,000 bpm × mm Hg). Coronary flow reserve (CFR) as the indicator of flow augmentation from rest to stress is calculated by stress to rest flow ratio.

The techniques for noninvasive flow estimates with compartmental modeling can accurately reflect regional MBF up to 5.0 mL/(min • g). Validation studies with the arterial reference microsphere technique in animal experiments have been reported to demonstrate equally accurate flow estimates for both ¹⁵O-water and ¹³¹⁵N-ammonia[^11,32]. More recently, a study with ¹³⁵Rb shows a similar linear correlation between the flow estimates by PET imaging and microsphere blood flows[^31]. Importantly, measurements of regional MBF with PET at rest stage, as well as during pharmacologically stimulated hyperemia or with cold presser testing, are highly reproducible. This property was confirmed by repeated MBF measurements during the same study session or by repeated measurements within several days[^36-36].

Depending on the time period to obtain sequential dynamic images, it is applicable to further simplify the compartmental model for flow calculation by utilizing image data only acquired within the early phase. This applicability is based on the assumption that the tracer washout and metabolic process do not yet occur during the early few minutes; thereby, the effects of $K_2$ and $K_3$ can be logically neglected. This simplified model is particularly suitable for ¹³⁵Rb and ¹³¹⁵N-ammonia with the given property of tracer retention in the myocardium[^37]. The flow equation only accounting for tracer retention for flow calculation is:

$$K_1 = E \cdot MBF = \int_0^t \frac{P(t)}{Ca(x)} dx$$

where $P(t)$ is tracer uptake in the myocardium after

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Fig. 2 Representative flow models. A: one-tissue compartment and two kinetic parameters as a two compartment model. B: two-tissue compartments and three kinetic parameters as a three compartment model.
time \( t \int_{0}^{t} Ca(x)dx \) represents the sum of arterial blood concentration from the start of tracer injection to time \( t \), and \( E \) is the myocardial extraction fraction of tracer. The advantage of retention flow model is its simplicity without the necessity of curve fitting process to obtain \( K_{1} \); thereby, the method enhances the applicability of

Fig. 3 An example of SPECT myocardial blood flow quantitation to detect three-vessel CAD with luminal narrowing in LAD. D1=90%; LCX: M=90%, D=90%, OM1=50%; RCA: PD=80%, confirmed by invasive coronary angiogram (upper panel). Attenuation-corrected perfusion images are interpreted to report a normal perfusion study without evidence of transient ischemia dilatation (middle panel). SPECT flow quantitation uncovers severe CAD with flow steal (CFR< 1.0) for all three territories associated with a total of 91% CAD burden throughout the whole myocardium (lower panel). LAD=left anterior descending, D1=diagonal 1, LCX=left circumflex, M=middle, D=distal, OM1=obtuse margina 1, RCA=right coronary artery, PD=posterior descending.
flow quantitation for routine clinical use. However, this simplified method utilizing a short period of data for flow calculation and lack of flexibility to accommodate for slow blood-pool clearance may be limited to calculate flow values for patients with low LVEF or severe lung problems.

**CLINICAL APPLICATIONS OF PET FLOW QUANTITATION**

Noninvasive assessment of absolute MBF in mL/(min • g) and CFR with PET is continuously emerging as a clinical tool to stratify risks for cardiac events and predict associated patient outcomes and to evaluate the early stage of asymptomatic CAD. Assessment of functional abnormalities of the coronary vessels with PET flow has an advantage over structural evaluation of the arterial wall. This advantage has been highlighted in classifying the early functional and progressive stages of coronary atherosclerosis before structural alteration within the arterial wall is magnified. Consequently, adding PET flow information to the relative perfusion imaging provides incremental diagnostic value for CAD detection and progressive stages of coronary atherosclerosis before structural alteration within the arterial wall is magnified. The clinical integration of this approach has been recommended to enhance both CAD detection and risk assessment of patients with known or suspected CAD.

**PRACTICAL CONSIDERATION**

Although PET myocardial flow quantitation has been clinically marked as a powerful tool for diagnosis and prognosis of CAD, the utilization of PET flow as a routine clinical tool has several practical challenges. The main challenges come from general accessibility of PET flow tracers which are currently restricted to certain regions (e.g., North America and Europe), and the requirement of a relatively high cost to adopt in the clinical environment. Myocardial perfusion single photon emission computed tomography (SPECT) with $^{99m}$Tc-labeled myocardial perfusion tracers, such as $^{99m}$Tc-sestamibi and $^{99m}$Tc-tetrofosmin, remains the clinical standard for MPI worldwide. Flow quantitation with SPECT, when available, may be a simple solution to overcome PET’s challenges to warrant a widespread utilization. In fact, modern SPECT instrumentation has been improved to have high temporal resolution for dynamic data acquisition. In the past this unique capability has not yet been well investigated to design clinical protocols for dynamic SPECT flow quantitation. The implementation of iterative reconstruction technique with effective physical corrections, in addition to SPECT instrumentation, collectively affirms to explore the clinical potential of flow quantitation with dynamic SPECT imaging. From the clinical standpoint, the accessibility of SPECT flow quantitation as a comprehensive clinical tool can be considerably important to areas where a proper myocardial PET tracer for flow quantitation is not available (e.g., Asian countries). From the economical standpoint, the SPECT approach for flow quantitation demands a much smaller financial overhead than the PET approach, therefore SPECT flow quantitation may also be attractive in areas, where both PET and SPECT myocardial flow tracers are available (e.g., North America and Europe).

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