ELSEVIER

Seminars in
NUCLEAR
MEDICINE

Pathogen-Specific Bacterial Imaging in Nuclear Medicine

Alvaro A. Ordonez, MD*,† and Sanjay K. Jain, MD*,†

When serious infections are suspected, patients are often treated empirically with broad-spectrum antibiotics while awaiting results that provide information on the bacterial class and species causing the infection, as well as drug susceptibilities. For deep-seated infections, these traditional diagnostic techniques often rely on tissue biopsies to obtain clinical samples which can be expensive, dangerous, and has the potential of sampling bias. Moreover, these procedures and results can take several days and may not always provide reliable information. This combination of time and effort required for proper antibiotic selection has become a barrier leading to indiscriminate broad-spectrum antibiotic use. Exposure to nosocomial infections and indiscriminate use of broad-spectrum antibiotics are responsible for promoting bacterial drug-resistance leading to substantial morbidity and mortality, especially in hospitalized and immunosuppressed patients. Therefore, early diagnosis of infection and targeted antibiotic treatments are urgently needed to reduce morbidity and mortality caused by bacterial infections worldwide. Reliable pathogen-specific bacterial imaging techniques have the potential to provide early diagnosis and guide antibiotic treatments.

Semin Nucl Med ■■:■■–■■

Introduction

Bacterial infections are a major cause of morbidity and mortality worldwide. However, with antimicrobial resistance emerging and spreading globally, multidrug resistant organisms are an increasingly serious threat to human health. In the United States alone, at least 2 million people become infected with multidrug resistant bacteria each year, with at least 23,000 of these patients dying as a direct result of these infections, leading to significant social and economic costs.[1-3] Although antimicrobial resistance can occur naturally, the misuse and overuse of antibiotics is accelerating this process.[4-8] Improved diagnostic accuracy of pathogen-specific imaging techniques could curb the inappropriate use of antibiotics for noninfectious entities, streamline empiric antibiotic selection to target the class of the bacteria causing the infection, and provide a way to rapidly monitor antibiotic treatments.

A common clinical problem for practicing infectious disease specialists and radiologists is differentiating active infection from other causes of inflammation. Current practice in microbiology (microbiologic cultures or molecular techniques) relies on the availability of a clinical sample (blood, urine, etc.). However, when a deep-seated infection is suspected, a biopsy is often required, leading to costly, invasive, and sometimes dangerous procedures, prone to sampling errors. Therefore, clinicians frequently use conventional radiological techniques such as CT, MRI, x-ray, and ultrasound, to aid in the diagnosis and localization of deep-seated infections.[9] However, these techniques rely on the presence of structural abnormalities in tissues and organs caused by the infection and inflammatory response of the host, and are unable to reliably differentiate infection from inflammation or oncologic processes. To try to address this clinical challenge, multiple molecular imaging techniques have been developed to detect bacteria in vitro, in animal models, and for clinical studies. Extensive research has been done using optical imaging techniques with promising results,[10-14] and although these studies have helped significantly in understanding disease pathogenesis and in developing therapeutics for infections, their clinical evaluation for bacterial infections has been limited by tissue penetration. Conversely, nuclear imaging techniques such as PET and SPECT are not limited by tissue depth and use very small amounts of radioactive tracers that reflect the physiological and biochemical changes associated with the infection process.

*Center for Infection and Inflammation Imaging Research, Johns Hopkins University School of Medicine, Baltimore, MD
†Department of Pediatrics, Johns Hopkins University School of Medicine, Baltimore, MD.
Address reprint requests to Sanjay K. Jain, MD, 1550 Orleans St, Rm 109, Baltimore, MD 21287. E-mail: sjain5@jhmi.edu

https://doi.org/10.1053/j.semnuclmed.2017.11.003
0001-2998/

Table Desirable Properties of an Ideal Bacteria-specific PET Imaging Tracer (Adapted From Gemmel et al[17] and Johnson et al[18])

Sensitive	High target to background signal ratio. Low limit of detection.
Specific	Probe targets infectious, not inflammatory lesions.
Quantitative	Signal proportionate to infectious burden.
Rapid	Fast localization to the site of infection.
Stable	Probe retained at the site of infection. No degradation of the probe by the host.
Safe	Acceptable radiation dose. Repeat injection feasible, without pharmacologic or immunologic effect.
Manufacturable	Uncomplicated synthesis with reasonable expense.

Nuclear imaging of bacterial infections has been a developing field for more than 50 years and provides valuable insight not only in the diagnosis and monitoring of infections, but it can also help understand the disease pathogenesis in both preclinical and clinical settings.[15] Although most of the published literature in this area involves nonspecific imaging agents such as ^{18}F-fluorodeoxyglucose (^{18}F-FDG), ^{67}Ga-citrate, and in vitro-radiolabeled leukocytes, significant efforts have been made to develop bacteria-specific radiolabeled tracers that can bind or accumulate only in bacteria.[16] This is a challenging task because the ideal bacteria-specific imaging compound should be both sensitive and specific for detection of the infection site, in addition to being nontoxic, affordable, widely available, and easily and rapidly prepared (Table). These compounds should have ideal biochemical properties (eg, moderate lipophilicity, low plasma protein binding, and metabolic stability),[18,19] as well as optimal pharmacokinetic and pharmacodynamic features for maximum performance and usability.[17] The tracer should be able to rapidly localize to the infection site with little nonspecific binding and have an optimal clearance rate that allows sufficient time for it to reach its target but is rapid enough to excrete unbound probe and minimize radiation exposure.[20] The search for the best bacteria-specific imaging tracers is still an ongoing challenge and this review summarizes the efforts made to achieve this goal.

Metabolism-based Tracers

Based on the principles utilized in clinical microbiology, which uses selective metabolism to differentiate microbes,[21] bacteria-specific imaging tracers based on selective metabolic pathways expressed only in bacteria (or a specific class of bacteria) could be developed not only to detect the presence of bacterial infection, but also to identify the class of bacteria causing the infections.[22] Metabolism-based tracers, such as radiofluorinated sugars and sugar alcohols, combined with favorable radiation dosimetry due to short half-life isotopes (eg, ^{18}F), provide multiple advantages such as easy penetration into diseased tissues, possible accumulation inside bacteria, and rapid clearance from nontarget tissues. However, because these tracers are designed to only be incorporated into bacteria, there would be limitations on their use for diagnosis of pathogens with a predominantly intracellular lifecycle (eg, *Listeria monocytogenes*). Similarly, the incorporation rate of these tracers into the cells could be affected by the metabolic state of the bacteria.

Nucleoside Analogs

The thymidine analog FIAU (1-(2'-deoxy-2'-fluoro-β-D-arabinofuranosyl)-5-iodouracil) was initially developed as a noninvasive agent to monitor tumor-targeting bacteria. FIAU is a substrate for the native thymidine kinases (TKs) of herpes viruses and a wide variety of bacterial species. Once inside the bacterial cell, FIAU gets phosphorylated by TKs and subsequently trapped. However, bacteria that do not possess the TK enzyme (eg, *Pseudomonas aeruginosa*), would not retain FIAU.[23] In 2004, Bettegowda et al reported successful in vivo imaging of bacterial infections in mice using ^{125}I-FIAU.[24] Using a lung infection murine model, Pullambhatla et al also showed that this tracer can be used to monitor the efficacy of antimicrobial therapy.[25] Subsequent studies with ^{124}I-FIAU reported promising results in patients with musculoskeletal infections (Fig. 1).[26] However, a recent study found that ^{124}I-FIAU lacks specificity in patients with prosthetic joint infections, with high background signal in uninfected muscle presumably due to host mitochondrial metabolism.[27] Although the synthesis methods and dosimetry of ^{18}F-FIAU in animal models have also been reported, data for bacterial imaging are not available.[28]

Sugars

While widely employed in clinical oncology, ^{18}F-FDG is presently the most used PET tracer for imaging bacterial infections by identifying infection-associated inflammatory responses.[29,30] Immune cells increase the use of glucose as an energy source during metabolic bursts associated with inflammatory responses due to infection.[31] Even during chronic inflammation, increased cell glycolysis persists,[32] giving ^{18}F-FDG the potential to image infections with high sensitivity.[31] Current guidelines recommend ^{18}F-FDG-PET imaging in clinical cases of peripheral bone osteomyelitis, suspected spinal infection, and fever of unknown origin (FUO),[33] and suggest it can be used in other infection settings. ^{18}F-FDG-PET has also been used to monitor tuberculosis (TB) patients,[34,35] evaluate patients with inconclusive diagnosis of infective endocarditis,[36] help distinguish between septic and aseptic loosening in joint prosthesis infection,[37] and diagnose diabetic foot infections.[38] However, because ^{18}F-FDG is metabolized by the same pathways as glucose, and given the widespread expression of the glucose surface transporters in eukaryotic cells, ^{18}F-FDG is not able to differentiate true infection from sterile inflammation. Mills et al developed the phosphate analog ^{18}F-FDG-6-P in an effort to facilitate specific uptake of ^{18}F-FDG by bacterial hexose phosphate transporters.[39] However, in vivo PET imaging with ^{18}F-FDG-6-P in a foreign body *Staphylococcus aureus* infection mouse model demonstrated that the biodistribution was similar to that of ^{18}F-FDG, therefore limiting its use for bacteria-specific imaging.