Molecular Imaging of Infectious and Inflammatory Diseases: A Terra Incognita

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Interest in molecular imaging of infections and inflammation has dramatically increased as its clinical relevance has become unambiguously strengthened by the need to clearly distinguish one from the other. This need relates both to the impact of overtreatment (e.g., immune suppression in inflammation) and the induction of resistance (i.e., patterns of increasing antibiotic resistance in infections). In recent years, our understanding and knowledge of the similarities and differences in the pathophysiology of infectious and inflammatory diseases has significantly improved. Molecular imaging offers unique and unsurpassed possibilities for better separating inflammatory processes from infection-related processes. Together with the recent developments in hybrid multimodality imaging techniques, accompanied by new radiopharmaceuticals and smart chemistry, nuclear medicine has positioned itself clearly as a key player in the field of infection and inflammation.

Clinicians often struggle with questions about presumed or established infectious or inflammatory disorders: Is there an infectious focus, or is it an inflammatory sign? What is the extent of the lesion? Are there infectious metastases? Are the therapeutic drugs entering and penetrating the infection site? Is the therapy successful? Can I withdraw the treatment and switch to other therapeutic options, or is continuation needed? We have to provide answers to these questions, supporting the clinician in individual-therapy decision making and preventing over- or undertreatment and its potential effects on the patient and on society, such as the spread of highly antibiotic-resistant bacterial strains. However, we

are not there yet. There are still questions to be answered and hurdles to be overtaken, but at the end of the tunnel is a clear and bright future for molecular imaging.

^{18}F-FDG PET/CT is the most commonly used diagnostic imaging tool worldwide for infection and inflammation. The accumulation of leukocytes, macrophages, monocytes, lymphocytes, and giant cells constitutes the body's response to injury and infection. Upregulation of glucose transporters has been demonstrated in all these individual cell types and contributes to increased accumulation and detection of ^{18}F-FDG in infection and inflammation. In 2013, a guideline on the use of ^{18}F-FDG in infection and inflammation was published jointly by the Society of Nuclear Medicine and Molecular Imaging and the European Association of Nuclear Medicine. On the basis of a cumulated reported accuracy of more than 85% and the opinions of leading international experts, the guideline concluded that the major indications for ^{18}F-FDG PET/CT in infection and inflammation were sarcoidosis, peripheral bone osteomyelitis, suspected spinal infection, fever of unknown origin, metastatic infection, bacteremia in high-risk patients, and vasculitis (1). For other indications, not enough evidence-based data were available to draw a conclusion, or it was still unclear whether ^{18}F-FDG PET/CT offered any significant advantage over other imaging options. The molecular imaging community has the responsibility of aiming high at performing large prospective studies to gain evidence-based data for such diagnostic applications and even more for therapy evaluation. Furthermore, for several applications we need clear, standardized interpretation criteria for negative and positive imaging findings, as could be developed by, for example, providing a large, public-domain dataset gathered from multiple clinical studies. Highly relevant applications for such research are fungal infections, vascular graft infections, joint prosthesis infections, infections of the diabetic foot, endocarditis, inflammatory bowel diseases, and infectious and inflammatory diseases in children.

In this issue of *The Journal of Nuclear Medicine,* Tseng et al. (2) focus on the role of ^{18}F-FDG PET/CT in a specific application: the detection of infectious foci of unknown origin in septic patients with end-stage renal disease undergoing maintenance hemodialysis. In a group of 104 patients, ^{18}F-FDG PET/CT findings were positive in 73 (70.2%), and a total of 95 major infectious foci were identified. Treatment regimens were modified in 28 patients. Positive ^{18}F-FDG PET/CT results were strongly related

to mortality. The authors concluded that ^{18}F-FDG PET/CT was clinically useful in this application, having led to a significant change in patient management. This was the first study for this application, but when the results are confirmed by randomized prospective trials, it could prove to be another major use for ^{18}F-FDG PET/CT. The relevance of the study data translates to many other invasive techniques by which artificial devices that carry an increased risk of infection—such as vascular-access ports, vascular and orthopedic inserts, and cardiac devices—are implanted in either elective settings or emergency settings with a higher possibility of contamination.

For diagnosis, staging, and evaluation of therapy for infections, the clinical impact of ^{18}F-FDG PET/CT is already high and certainly is destined to increase in the future (3). However, ^{18}F-FDG has major limitations and pitfalls. In particular, specificity is a major issue. Because of overlapping pathophysiologic mechanisms, this technique is unable to reliably discriminate infection from sterile inflammation, malignancies, and physiologic wound healing. Logically then, we need other, more specific, targeted radiopharmaceuticals and imaging techniques. A more classic technique, the application of labeled white blood cells, is specific for infection and still is preferred in Europe for the detection of peripheral osteomyelitis, joint prosthesis infections, and diabetic foot infections. This approach, however, requires a laborious preparation under sterile conditions, may be hazardous to technicians and patients since it involves potentially contaminated blood, and is not convenient for patients since they have to visit the nuclear medicine department at least 4 times (collection of blood, reinjection, and at least dual-time-point imaging). Alternatives such as [99mTc]-labeled interleukin-2, specific for inflammation by visualization of the interleukin-2 receptor, which is overexpressed by activated T lymphocytes during inflammation, have been successfully used in small patient groups with several autoimmune and inflammatory diseases (4). Here, again, we need larger prospective patient studies and a cutoff value for positivity regarding uptake. We look forward to the first results for the PET tracer ^{18}F-interleukin-2.

As has been shown in the literature over the last few decades, many other forms of targeted imaging are feasible using a wide variety of probes, resulting in more or less successful visualization of structures or pathways of interest. Some of these tracers allow for relatively straightforward implementation in the clinic, whereas others are more complicated or even impossible to implement. One important strategy with a high chance of straightforward clinical implementation is the use of tracers based on agents already applied in health care and therefore with a known toxicologic profile (5). Indeed, most tracers that have been described over the past few years for molecular imaging in patients were based on molecules with a known toxicologic profile, that is, therapeutic agents or humanized antibodies such as bevacizumab or trastuzumab (6). Besides having a straightforward implementation, the tracer needs to have several other qualities of importance to the clinician, such as high sensitivity and specificity, a short time between administration and imaging, easy preparation and application, and preferably low cost. In an effort to create an infection-specific imaging tracer, the antibiotic ciprofloxacin was labeled with [99mTc] and ^{18}F to specifically target bacteria. However, in a large clinical trial with [99mTc]-ciprofloxacin, low specificity (81.7%) was the major limitation (7). Despite that suboptimal result, the use of antibiotic-based tracers might be the solution to infection-specific imaging and could serve the clinician

in solving some of the above-mentioned questions. Other agents that are highly promising for infection-specific imaging are antimicrobial peptides, antigen-specific antibodies and antibody fragments, and compounds that are specifically bound or metabolized by bacterial cells (prokaryotic) but not by mammalian cells (eukaryotic).

One step further, and a challenging one, would be to use imaging not only to distinguish inflammation from infection but to determine the particular microorganism that has caused an infection or become colonized. For example, early detection of *Streptococcus pyogenes* in necrotizing fasciitis might be of great use in decreasing the often medieval mutilating surgical procedures that are needed because of the lack of a reliable diagnostic instrument. Similarly, early detection of *Pseudomonas aeruginosa* as a cause of infection would allow for early adjustment of antibiotics to improve the overall outcome. Not only is early identification of the infectious species of great use, but detection of specific resistance mechanisms might also be highly practical (such as methicillin-resistant *Staphylococcus aureus* or extended-spectrum β-lactamase–producing bacteria). This ability would aid decision making and infection prevention at both the individual and the community levels by, for example, helping to prevent cross-patient contamination within an intensive care unit or among different hospital wards.

Molecular imaging is not limited to imaging with radionuclides but can be performed using the elegant method of optical imaging as well (8). In optical imaging, fluorophores are used to generate an optical signal on excitation by an external light source of a specific wavelength, such as a laser. For untargeted imaging, an unconjugated fluorophore such as indocyanine green can be used, or the fluorophore can be conjugated to a targeting molecule to image specific areas of interest. The emitted signal is captured by a specialized, sensitive charge-coupled-device camera. The main advantage of optical imaging is that it allows for fast imaging with an extremely high resolution. This technique does not require any radioactivity, is cheaper and more flexible than radionuclide imaging, and can be repeated without increasing the ionization burden. The major limitation is that the fluorescence signal can penetrate tissue to a maximum depth of only 1 cm. As a consequence, optical imaging currently is applied mainly for superficial imaging, such as for intraoperative guidance or endoscopic imaging. A new technique—optoacoustic or photoacoustic imaging—expands this maximum penetration depth and, therefore, the number of potential clinical applications. Optoacoustic imaging is based on thermal expansion of a tissue of interest caused by absorption of short laser pulses by intrinsic fluorophores that occur naturally in the body or extrinsic fluorophores that are exogenously administered. However, instead of detecting the emitted optical signal, specialized detectors register ultrasonic pressure waves generated through thermoelastic expansion of the excited fluorophores (9). Because the ultrasonic pressure waves have a lower wavelength than optical waves, the maximum penetration in tissue is substantially deeper and is expected to be in the same range as ultrasound imaging (∼3–5 cm). Both the technology and the targeted optoacoustic probes are currently in development. It is expected that the field of molecular optical and optoacoustic imaging will rapidly evolve within the next decade and come into more general daily use in close synergy with modalities such as radionuclide imaging (PET and SPECT), MR imaging, and ultrasound.

In addition to the developments in targeted imaging probes, developments in camera systems have taken place or are to be

expected. Simultaneous PET/MR imaging has major theoretic advantages over PET/CT that could be of interest to the whole medical community. The hybrid modality can provide an absolute match between the two separate modalities, allowing tissue characterization under comparable physiologic conditions and better colocalization of the PET signal within soft tissues (10). Furthermore, the fact that MR imaging imposes no radiation burden could benefit patients (especially children) who require serial scanning to evaluate the success of therapy. Indeed, PET/MR imaging is expected to have a clinical impact in several infectious and inflammatory diseases, such as insulitis, spondylodiskitis, inflammatory bowel diseases, soft-tissue infection, neurologic inflammation and infection, the infected diabetic foot, and infectious and inflammatory diseases in children.

Ultrasound is often used in the clinical setting for noninvasive diagnosis of infectious abscesses. Therapeutic ultrasound, on the other hand, has been applied mainly for its thermal or mechanical effects. Recently, the application of nonthermal ultrasound energy as a combination treatment with drugs has been launched for various diseases. Furthermore, the introduction of microbubbles, molecule-targeted echo contrast agents, and carriers or enhancers of drugs has added a new dimension to diagnosis and therapy. Ultrasound irradiation of tissues and cells enhances drug targeting, lowers systemic drug toxicity, and improves drug absorption rates—all topics of interest in infectious diseases. Efficient delivery of a drug into target cells or tissues for therapeutic purposes has always been a challenge, requiring the combined effort of experts in different scientific fields.

Advances in techniques and translational medicine have brought molecular imaging into a new diagnostic era. Oncology is in the lead, with large prospective trials that investigate the role of molecular imaging for personalized or precision medicine. Obviously, this approach is highly translatable to infectious and inflammatory diseases, but it currently represents a terra incognita in this arena. Molecular imaging techniques can characterize pathophysiologic processes, highlight the cell types and pathogens involved, detect the presence of a potential target, and quantify the pathogenic bacteria and biologically active molecules both for diagnosis and for treatment monitoring. By implementing all these options, we will one day be able to provide clinicians with clear answers to their urgent questions, as well as providing infectious/inflammatory disease patients with optimal personalized treatment.

DISCLOSURE

No potential conflict of interest relevant to this article was reported.

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