Case Report

Single-photon Emission Computed Tomography–Computed Tomography Using ^{99m}Tc -labeled Leukocytes for Evaluating Infection Associated with a Cranial Implant in a Rhesus Macaque (*Macaca mulatta*)

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An adult male rhesus macaque (*Macaca mulatta*) that was enrolled in a study evaluating cognition and memory presented with suppurative exudate along the margins of a long-standing cranial implant that included a stainless-steel head post, plastic left-sided recording cylinder, and acrylic over a previously placed right-sided recording cylinder. Both cylinders were located at the level of the prefrontal cortex. After treatment comprising systemic antibiotics and daily cleaning with povidone-iodine for several months, the macaque underwent single-photon emission computed tomography-computed tomography (SPECT-CT) in which his neutrophils were labeled with ^{99m}Tc -hexamethylpropylene amine oxime (^{99m}Tc -HMPAO) to evaluate for active infection below the implant. Soft tissue inflammation and osteomyelitis were found at the site of the previous right-sided recording cylinder. Cephalosporin and tetracycline antibiotics were administered for 12 wk. Follow-up SPECT-CT imaging was then performed to evaluate response to medical treatment. Results indicated no change in the degrees of soft tissue inflammation and osteomyelitis associated with the right-sided recording cylinder site. SPECT-CT imaging was used to guide the surgical removal of the implant and debridement of the infected tissue. On removal of the entire cranial implant, the osteomyelitis and soft tissue inflammation observed on the pre- and posttreatment SPECT-CT scans were confirmed. In addition, a large cavitory defect through the calvarium with suppurative exudate was discovered below the base of the head post. Infection in this defect was not apparent on SPECT; however, the bony defect was confirmed on reevaluation of the CT images. We concluded that the infection in this defect was silent on SPECT due to the limited vascularization of the surrounding bone and the chronicity of the infection. This case study is the first to describe the use of SPECT-CT for evaluating soft tissue inflammation and osteomyelitis beneath a cranial implant in a NHP.

Abbreviations: SPECT-CT, single-photon emission computed tomography–computed tomography; ^{99m}Tc -HMPAO, technetium-hexamethylpropylene amine oxime

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Chronic cranial implants are commonly used for neurophysiologic and behavioral studies in conscious animals. This equipment has revolutionized the methods by which investigators can record neuronal activity from single neurons, deliver neuromodulatory experimental agents by microelectrophoresis, and detect responses to a variety of learning and memory paradigms. Bacterial and fungal contamination is one of the most common complications associated with cranial implants, with localized infections that have the potential to spread deeper into the cranium, dura, and brain tissue.^{2,4,8,17} The initial clinical presentation of a contaminated implant may include suppurative

and hemorrhagic discharge or crusting at the skin-implant margins. Subsequent extension of infection below the cranial implant over time can lead to infection of the calvarium and osteomyelitis, compromising the stability of the implant. Infections that spread to the dura can lead to meningitis, brain abscessation,¹⁵ neurologic deficits,¹⁷ and ultimately death. Although antibiotic treatment and a strict cleaning regimen may enervate the emerging infection, removal of the implant is almost always necessary to definitively resolve the infection.

Large cranial implants, with multiple recording cylinders or head posts, pose a specific challenge in the ability to localize infection below the implant. This localization is important for planning treatment of implant-associated infections, evaluating the response to treatment, and surgical planning for partial or full removal of the implant with debridement of the infected tissue. Historically, radiographs have been used to detect infection in the bone, but the 2-dimensionality of this imaging modality

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limits its value in surgical planning. Moreover, because cranial implants often contain screws, head posts, and cylinders made of metal, these devices may obscure visualization of the bone on radiographs. MRI prior to cranial implant surgery provides the 3-dimensionality necessary to accurately guide positioning and shaping to match the contour of the skull.¹¹ However, follow-up MR imaging of cranial implants is often not possible because the stainless steel components have the potential to be damaged or harm either the animal or personnel handling the animal. In addition, the titanium components of cranial implants can distort MR images.¹²

Single-photon emission computed tomography–computed tomography (SPECT-CT) scans fuse images from SPECT and CT scans to provide more precise colocalization of physiologic or molecular information (for example, perfusion, metabolism, inflammation, angiogenesis) with anatomic or structural features. The CT scan creates a 3D radiograph to visualize precise anatomic features or structural abnormalities, whereas the SPECT scan utilizes a radiopharmaceutical to evaluate functional information.^{6,23} In the current application, the coregistration of SPECT and CT data allowed for accurate functional anatomic mapping of infection or inflammation.^{20,21} Similar to MR imaging, distortion of CT images can occur in the presence of implanted metal components,³ however, unlike MR imaging, magnetic forces are not present to cause damage to the implant or harm to the animal.

^{99m}Tc-hexamethylpropylene amine oxime (^{99m}Tc-HMPAO) is a radiopharmaceutical that is commonly used in nuclear medicine for labeling neutrophils to localize inflammation and infection. ^{99m}Tc is advantageous over other radiolabels, such as indium-111, in that ^{99m}Tc has a shorter half-life of 6 h and lower radiation dose¹ and can, therefore, be administered at higher doses for better imaging quality. HMPAO acts as a chelating agent for ^{99m}Tc, and when combined, a lipophilic ^{99m}Tc-HMPAO complex is formed. This complex can be taken up by all WBC but is selectively retained in neutrophils due to a chemical change in the complex that prevents it from exiting the cell. This feature is useful for evaluating neutrophil-mediated processes of inflammation, such as bacterial infections. However, the ^{99m}Tc-HMPAO complex is not specific for uptake in leukocytes, meaning that leukocytes must be isolated from whole blood prior to being labeled and administered back into the patient,¹ a process that can last as long as 2 h. Nonetheless, previous studies have shown that ^{99m}Tc-HMPAO is a valuable tool in diagnosing osteomyelitis and orthopedic implant-associated infections in humans.^{5,6,10} Moreover, ^{99m}Tc-HMPAO has been shown to be more precise at differentiating soft tissue from bone infection, as compared with other radiopharmaceuticals used for scintigraphy, such as ^{99m}Tc-labeled antigranulocyte antibodies,^{6,18} therefore, the clinical use of ^{99m}Tc-HMPAO continues to be widespread. We here report the use of SPECT-CT imaging with ^{99m}Tc-HMPAO-labeled leukocytes as a novel diagnostic tool for evaluating ongoing and active infection below a cranial implant in a rhesus macaque.

Case Report

A 22-y-old male rhesus macaque (*Macaca mulatta*) was enrolled in a long-term study for evaluating cognition and memory. All experimental procedures performed on this macaque were approved by the Yale University IACUC. In accordance with the protocol, the macaque had a stainless steel head post surgically implanted along the midline of the caudal skull at 7 y of age. The head post was secured to the skull with stainless steel bolts and screws as well as dental acrylic. Approximately 4

y later, a craniotomy was performed to place a plastic recording cylinder (diameter, 2 cm) above the right prefrontal cortex. The recording cylinder was secured to the skull with stainless steel screws and dental acrylic. Additional dental acrylic was applied to secure the head post and recording cylinder together. Approximately 7 y later, the right-sided recording cylinder was removed for research purposes, and the defect was covered with dental acrylic. At that same time, a craniotomy was performed above the left prefrontal cortex to place a plastic recording cylinder (diameter, 2 cm). A single stainless steel screw was placed around the left recording cylinder, and dental acrylic was applied to the inside and outside of the recording cylinder to bond it to the existing implant.

The implant was routinely cleaned 2 or 3 times weekly by using 10% povidone–iodine and sterile saline (Hospira, Lake Forest, IL). Since the initial placement of the head post, the margins on the right side of the cranial implant had been observed to have mild, intermittent purulent and hemorrhagic discharge, and these margins were repeatedly swabbed for bacterial culture and antibiotic sensitivity over a 15-y period. According to sensitivity results, the macaque was placed intermittently over the 15-y period on a variety of short-term antibiotics, including enrofloxacin, doxycycline, and amoxicillin–clavulanic acid.

Approximately 15 y after initial head post placement, the macaque was evaluated for a recent increase in the amount of purulent and hemorrhagic discharge along the margins of the cranial implant. Infection of unknown severity was suspected below the acrylic located on the right side of the implant at the previous site of the recording cylinder. Bacterial culture of the discharge yielded the growth of *Staphylococcus aureus* and *Corynebacterium ulcerans*. According to antibiotic sensitivity results, the macaque was started on a 10-d course of ceftiofur sodium (2.2 mg/kg SC daily; Naxcel, Zoetis, Kalamazoo, MI) and the frequency of implant cleaning was increased to once daily. Given a lack of decrease in the amount of purulent and hemorrhagic discharge at the implant margins after treatment, SPECT-CT imaging was proposed to determine the extent and source of infection associated with the implant.

Briefly, the macaque was immobilized and induced with ketamine (5 mg/kg IM; Henry Schein Animal Health, Dublin, OH) and dexmedetomidine (0.005 mg/kg IM; Dexdomitor, Orion, Kalamazoo, MI), and 22-gauge catheters were placed in the right and left cephalic veins as well as the left saphenous vein. Atropine (0.02 mg/kg IM; Henry Schein Animal Health) was administered prior to endotracheal intubation. General anesthesia was maintained with isoflurane (Henry Schein Animal Health, Dublin, OH) in oxygen for the duration of the procedure. Approximately 60 mL of blood was collected from the macaque through the intravenous catheters by using 20-mL syringes containing heparin (670 U per 20-mL syringe; Sagent Pharmaceuticals, Schaumburg, IL), and leukocytes from the blood were labeled with ^{99m}Tc-HMPAO, as described later. To replace the blood volume lost from blood collection, intravenous lactated Ringers solution (Hospira, Lake Forest, IL) was administered at a rate of 10 mL/kg/h for 2 h.

Leukocytes were labeled by using a Ceretec kit (GE Healthcare, Arlington Heights, IL) according to the manufacturer's instructions. Briefly, 30 mCi of ^{99m}Tc in 5 mL of sterile 0.9% sodium chloride was added to a sterile, nonpyrogenic lyophilized vial containing Ceretec (HMPAO solution containing 0.5 mg exametazime, 7.6 µg stannous chloride dehydrate, and 4.5 mg sodium chloride). The mixture was assayed for total activity, and the dose to be injected was calculated as 8.76 mCi. The mixture was used within 30 min of preparation. The blood was

allowed to rest 60 min for the RBC to sediment. The leukocyte-rich plasma supernatant was collected and processed according to the kit instructions for labeling with ^{99m}Tc -HMPAO, and approximately 5 mL of ^{99m}Tc -HMPAO-labeled leukocyte suspension was administered intravenously to the macaque.

After a 2-h incubation period to allow systemic distribution of labeled leukocytes, the SPECT-CT scan was performed. The macaque was positioned in ventral recumbency, with only the head imaged for the scan. The imaging was performed by using a hybrid SPECT-CT system (Discovery 570c NM/CT, GE Healthcare) equipped with a cadmium zinc telluride detector capable of high-resolution SPECT imaging paired with 64-slice CT. The acquisition parameters for CT were 350 mA, 120 kV, and a slice thickness of 0.625 mm, and for SPECT a 15-min single isotope acquisition with a standard energy window of $140\% \pm 5\%$ was used. The SPECT was reconstructed and visualized by using Xeleris (GE Healthcare), and CT was analyzed on an Advantage Workstation (GE Healthcare) with SPECT and CT images fused by using Xeleris. After the scan, the macaque recovered from anesthesia without complication.

Culture of the implant margins at the time of the SPECT-CT scan indicated growth of *Escherichia coli*, *Corynebacterium* spp., *Staphylococcus aureus*, and *Klebsiella pneumoniae*. According to antibiotic sensitivity results and in consultation with an infectious disease specialist, the macaque was started on long-term doxycycline (2.5 mg/kg PO once daily; Wedgewood Pharmacy, Swedesboro, NJ) and ceftiofur crystalline free acid suspension (20 mg/kg SC once weekly; Excede, Zoetis). In addition, a probiotic (one tablet daily; CulturelleKids, i-Health, Cromwell, CT) was started to prevent potential dysbiosis. After 12 wk of treatment with doxycycline and ceftiofur, a follow-up SPECT-CT study with ^{99m}Tc -HMPAO labeled leukocytes was performed as described earlier. The labeled leukocyte preparation and scan parameters for this follow-up scan were the same as for the initial scan, except that a dose of 4.6 mCi of ^{99m}Tc was calculated and used.

There was no difference in the degree or location of uptake of labeled leukocytes between before and after treatment, therefore, only images from the posttreatment scan are shown. Post-treatment SPECT-CT imaging revealed significant accumulation of labeled leukocytes in soft tissue and bone at the location of the previous right-sided recording cylinder above the level of the prefrontal cortex and extended caudally near the head post (Figure 1 A through C). The intensity of the uptake was increased centrally within the region of uptake and indicated soft tissue inflammation with underlying osteomyelitis. In addition, the bone associated with this increased region of uptake was thin and irregular (Figure 1 A). Labeled leukocytes also were present under the left-sided recording cylinder but to a much lesser degree (not shown).

Given the lack of response of the infection under the implant to long-term antibiotics, surgical removal of the implant was elected. At 2 wk prior to surgical removal of the cranial implant, the macaque was observed having a generalized tonic-clonic seizure. Three additional seizures were observed during the 2 wk prior to the implant removal surgery; consequently the macaque was started on diazepam (1 mg/kg PO twice daily; Teva Pharmaceuticals, North Wales, PA). At 1 wk prior to implant removal surgery, the macaque was anesthetized with ketamine (7 mg/kg IM; Henry Schein Animal Health) and midazolam (0.05 mg/kg IM; West-Ward) for preoperative diagnostics; CBC, serum biochemistry profile, and thoracic and abdominal radiographs were unremarkable. In addition to SPECT-CT scan results, skull radiographs were performed to assist in surgical planning (Figure 2 A). Radiographs confirmed 4 bolts and 8

screws in the bone. Because preoperative blood cultures yielded *Enterococcus faecalis* that was resistant to ceftiofur and doxycycline, both of these antibiotics were discontinued just prior to surgery.

On the day of surgery to remove the cranial implant, the macaque was immobilized and induced with ketamine (7 mg/kg IM; Henry Schein Animal Health) and midazolam (0.05 mg/kg IM; West-Ward). General anesthesia was maintained with isoflurane (Henry Schein Animal Health) in oxygen. The surgical site was aseptically cleaned. Preoperative buprenorphine (0.02 mg/kg SC; Par Pharmaceutical, Chestnut Ridge, NY) and cefazolin (25 mg/kg IV; Wg Critical Care, Paramus, NJ) were administered. The implant acrylic was drilled away to reveal the 4 stainless steel bolts and 8 stainless steel screws, which were removed from the cranium; the surgeon noted that the anterior bolt on the right (no. 4) was loose (Figure 2 A), and exudate at this site was continuous with the right-sided suppurative lesion (Figure 1). The head post and recording cylinder on the left side were removed also.

Evaluation of the cranium revealed extensive purulent discharge and osteomyelitis on the right rostral area at the location of the initial right-sided recording cylinder that extended throughout subcutis and superficially into the bone (Figure 2 B). In addition, a pocket (diameter, 1.5 cm) of purulent discharge that extended full thickness through the calvarium was present under the base of the head post (Figure 2 B). Swabs for aerobic and anaerobic bacterial culture and antibiotic sensitivity collected from the sites of infection on the right cranium and below the head post base yielded *Corynebacteria ulcerans*, *Staphylococcus aureus*, *Enterococcus faecalis*, *Klebsiella oxytoca*, and *Serratia marcescans*. The lytic bone at the infections sites was debrided and flushed with copious amounts of sterile saline. The surgical site was redraped, and the surface of the cranium and head post defect was cultured once more. The skin margins were undermined to sufficiently allow apposition of the skin edges and closure. The macaque remained stable throughout the majority of the procedure but went into cardiac arrest as the final skin sutures were being placed. Cardiopulmonary resuscitation was initiated, and epinephrine (1 mL; Henry Schein Animal Health) and atropine (0.025 mg/kg IV; Henry Schein Animal Health) were administered intravenously but were unsuccessful.

A diagnostic necropsy was performed immediately. Tissues were fixed in 10% formalin, embedded in paraffin, and sectioned. Sections were stained with hematoxylin and eosin, with Gram stain, or for glial fibrillary acid protein, a nonspecific marker for glial cell damage. The right side of the calvarium at the level of the previous cylinder was thin and had eroded (Figures 1 D and 2 B). The lesion extended from the prefrontal cortex to bolt 4 above the parietal cortex (Figure 2 B). A penetrating cranial defect under the head post was encountered. This previously exudate-filled cavity was partially surrounded by bone growth that enclosed a bony cavity that extended from the right cortex and across the midline to end over the left parietal cortex (Figure 1 E and F). The caudal aspect of this bony cavity was open (lacking bone and dura) and coincided with a parenchymal defect of the left posterior parietal cortex (Figure 1 G). Repeat review of SPECT-CT imaging at this same level confirmed this bony defect and thickened cortical bone (Figure 1 B), which resulted in considerable depression (by approximately 2 cm) of the underlying parietal cortex on the right (Figure 2 E).

The midline deviated markedly to the left (Figures 1 G and 2 C). Coronal sections taken at the level of parietal cortex (Figure 2 E) revealed a focal chronic fibrotic subdural lesion associated with a bolt on the right side (bolt 3 in Figure 2 E). Numerous

Figure 1. Comparison of (A through C) SPECT-CT images after long-term treatment with (D and E) coronal sections of skull. (A) SPECT-CT and (D) gross sections taken just anterior to the head post (asterisk), at the level of the (C) strong SPECT signal corresponding to exudate from beneath the acrylic cap (arrow). (A) The calvarium in this location is thin and irregular and has a strong SPECT signal (arrow). (B) SPECT-CT and (E) gross sections just posterior to the head post. Both images show a cavitory lesion enclosed by bone (asterisks). This imaging feature corresponded to the pus-filled cavity discovered beneath the head post at surgery and did not label on SPECT-CT. The right side of the calvarium is markedly thickened by proliferative bone. (F) Gross section of calvarium, taken posterior to the site shown in panel E. The bony cavitory lesion extends to the left of the midline, where it opens onto the surface of the brain (G, arrow). (G) Fresh brain, with the locations of calvarial sections shown in panels D, E, and F corresponding to levels i, ii, and iii, respectively. Bar, 1 cm.

Figure 2. Comparison of radiographs, dorsal aspects of cranium and brain, and coronal sections of brain. (A) Preoperative radiograph, dorsal view. Bolts are numbered and surround the head post. The black arrow indicates the position of the current left cylinder. (B) Calvarium, dorsal view. The bone beneath the previous cylinder and right sided exudate is eroded; this erosion extends caudally to bolt 4. A penetrating cavity in the skull is located under the head post (asterisk). (C) Fixed brain, dorsal view. A depression of the prefrontal cortex beneath the previous cylinder is present (arrow). Depressions created by bolts are numbered. Depression of the parietal cortex created by hypertrophic calvarium is shown by an asterisk. The midline is deviated to the right. The levels of the coronal sections illustrated in panels D and E are shown in parentheses. (D) Fixed brain, coronal section at the level of the arcuate sulcus (site of the previous cylinder). The cortex over this area is depressed and covered by thickened hemosiderin-stained dura (arrow). The brain beneath the left cylinder appears grossly unremarkable. (E) Fixed brain, parietooccipital cortex. Depression of the brain beneath hypertrophic calvarium is shown by an asterisk. A chronic abscess is located beneath bolt 3 on the right, and a region of superficial cerebral necrosis and hemorrhage is located beneath the cavitary bony lesion located close to bolt 2 on the left. Bar, 1 cm.

macrophages containing gram-positive intracytoplasmic coccobacilli were embedded within the dural scar (Figure 3 E). Dural cultures yielded *Corynebacterium* spp. from this site and from the right anterior bone lesion with which this lesion was continuous. Contralateral parietal cortex (located beneath the interior opening of the bony cavity) demonstrated focal cortical necrosis, hemorrhage, and gliosis (Figure 3 B and C). The site of exudate associated with the previous right-sided prefrontal cylinder exhibited bone atrophy, dural fibrosis, and suppurative inflammation of the subcutis (Figure 3 A). Underlying brain exhibited modest flattening and local gliosis, but no evidence of inflammation. Bone over the existing left-sided prefrontal cylinder was thin but otherwise normal, with no gross abnormality of the underlying brain.

Discussion

The routine use of cranial implants in animals for neurophysiologic and behavioral studies is an important advancement in

the field and has improved our understanding of cognition and neurodegenerative diseases. In NHP, cranial implants often allow the collection of longitudinal data for months to years. Cranial implants, however, act as foreign bodies and can promote chronic inflammation and serve as a nidus for infection. These devices can become infected at the time of the implant surgery or at any point after surgery, with local extension under the implant. Interestingly, a recent study evaluating metal alloys in chronic cranial implants in macaques showed that the titanium components of the implant can leach into the surrounding tissues.⁷ Whether these leached metal ions facilitate inflammation and predispose the tissue to infection or whether preexisting infection facilitates leakage of metal ions into the tissue is unclear. However, when evaluating for sources of inflammation and infection under a cranial implant, the metal components of the implant should be taken into consideration.

In the presented case, we suspect that a chronic infection associated with bolt 4 may have been a constant and continuous source of infection that began during the time of the initial head

Figure 3. Histopathology of brain. (A) Cortex beneath the previous cylinder that has been stained for glial fibrillary acidic protein (GFAP) is atrophic and gliotic. Meninges are thickened and fibrotic and contain bone fragments, but no meningitis is evident. The region within the rectangle is illustrated. (B and C). Posterior left parietal cortex, beneath the cavitary bony lesion stained (B) for GFAP and (C) with hematoxylin and eosin. Gliosis (B) accompanied by hemorrhage (C) is present. (D and E) A chronic abscess beneath bolt 3 is illustrated. Gram-positive coccobacilli are abundant within macrophages. Bar: 50 μ m (A through C), 500 μ m (D), 20 μ m (E).

post placement or during the subsequent surgery for placement of the left-sided cylinder and removal of the right-sided cylinder, when this bolt may have been reexposed during bonding of the existing head post, left-sided cylinder, and right-sided acrylic together to form a single implant. We cannot, however, rule out the possibility of other sources contributing to this chronic infection, such as translocation of skin bacteria at the margins of the implant. Reports in the literature indicate that bacterial colonization is the major source of infection, and *Staphylococcus* spp. and *Corynebacterium* spp. are commonly recovered.^{2,24} Both of these bacteria were cultured from this macaque. Moreover, the chronic localized infection associated with bolt 4 likely provided a constant source of infection that was very difficult to detect and was unresponsive to antibiotics *in vivo* despite demonstrated sensitivity after culture. Management of such chronic infections can be challenging, because infection under the implant cannot be visualized readily. Relying on the culture of the implant margins may not reflect the full spectrum of microorganisms proliferating under the implant, as was demonstrated in this case. Development of resistance to the antibiotics can occur, and the infected site may not be sufficiently exposed to bactericidal levels of antibiotic. In most cases, removal of the

entire implant is necessary to completely resolve the infection. However, given the time and cost that often go into training the animal and collection of the data, which in many cases includes years of longitudinal data, the removal of the entire implant should be carefully considered. Nonetheless, failure to remove the implant in a timely manner may result in more severe complications, such as neurologic signs, which have the potential to invalidate study results, and, as in this case, can ultimately lead to death.

In this report, SPECT-CT imaging with autologous ^{99m}Tc-HMPAO-labeled leukocytes was beneficial in visualizing and characterizing infection under a chronic cranial implant. The SPECT-CT scan localized soft tissue inflammation and osteomyelitis above the prefrontal cortex correlating with the previous site of the right-sided recording cylinder. Importantly, the exudate-filled lesion that penetrated through the skull at the level of the previous head post was silent on SPECT imaging, as was the localized infection beneath bolt 4. In contrast, well-vascularized soft tissue (which had manually been cleaned daily) at the site of exudation on the right implant margin was readily populated by radiolabeled leukocytes. We suspect that the chronically infected head post and bolt sites were not sufficiently vascularized

to permit adequate uptake by labeled leukocytes during the 2 h between infusion and scanning. Previous studies comparing positron emission tomography using ^{18}F -fluorodeoxyglucose and SPECT-CT with $^{99\text{m}}\text{Tc}$ -methylene diphosphonate radiolabeling in human patients with osteomyelitis suggest that positron emission tomography may provide better sensitivity regarding the detection of inflammation and infection in bone regions with decreased vascularity, compared with SPECT-CT.^{9,23} Perhaps positron emission tomography with ^{18}F -fluorodeoxyglucose radiolabeling might have revealed these areas of infection with decreased vascularity in our case. Nonetheless, CT is still necessary for precise anatomic location and when adequate blood supply is present, SPECT may have better sensitivity and specificity than positron emission tomography of the bone.^{22,23}

According to the manufacturer, optimal imaging should be performed 2 to 4 h after injection of the $^{99\text{m}}\text{Tc}$ -HMPAO-labeled leukocyte suspension into the patient. This recommendation is based on clinical trials in human patients with intraabdominal infection or inflammation showing 95% to 100% sensitivity and 62% to 85% specificity for $^{99\text{m}}\text{Tc}$ -HMPAO radiolabel leukocytes in the gut when SPECT-CT imaging is performed between 2 and 4 h, compared with imaging at 24 h after radiolabel administration. Other studies have shown comparable accuracy for imaging performed between 4 to 6 h in a patient with bone or joint infections.^{6,13,14} Although the optimal interval between administration of the $^{99\text{m}}\text{Tc}$ -HMPAO-labeled leukocyte suspension and SPECT-CT imaging may depend on the tissue type and the ability of leukocytes to accumulate in that tissue, in most studies the optimal interval appears to be within a few hours of administration. Because $^{99\text{m}}\text{Tc}$ has a short half-life of 6 h, its ability to reveal indolent infections compared with active infections may be limited. Radiolabels that have a longer half-life, such as ^{111}In (half-life, 67 h), are recommended for the diagnosis of slower developing infections.^{16,19} In our case, we suspected that the increase in purulent and hemorrhagic discharge along the right side of the implant margin indicated a more active and progressing infection; consequently $^{99\text{m}}\text{Tc}$ was an appropriate radiolabel choice to detect this infection with imaging. In contrast, the infection beneath the head post was silent on both pretreatment and posttreatment SPECT and was only discovered subsequently at the time of surgery. Because this infection appeared to be chronic and slowly progressive based on evaluation at the time of surgery and at gross necropsy, a radiolabel with a longer half-life, such as ^{111}In , may—in hindsight—have been more successful at detecting this lesion. Therefore, the activity and chronicity of a suspected cranial implant infection should be taken into consideration when determining the appropriate radiolabel to use in conjunction with SPECT-CT to evaluate for infection.

In conclusion, the 3-dimensionality of the CT combined with the ability of SPECT to detect functional inflammation and infection has proven to be very useful in localizing these inflammatory processes in soft tissue and bone. $^{99\text{m}}\text{Tc}$ -HMPAO-labeled leukocytes are readily taken up by WBC, with accumulation most prominent in neutrophils, making this radiolabel a useful tool for evaluating bacterial-associated cranial implant infections. In this case report, we used SPECT-CT imaging with $^{99\text{m}}\text{Tc}$ -HMPAO-labeled leukocytes as a novel diagnostic tool for evaluation of infection under a cranial implant in a macaque. This imaging modality was able to detect active areas of infection underneath the cranial implant but failed to detect an indolent infection below the head post. Given the widespread use of cranial implants in macaques for neurophysiology and behavioral studies, SPECT-CT with radiolabeled leukocytes should

be considered as a diagnostic tool for suspected infections, and results can be readily used to guide a medical and surgical treatment plan. However, the radiopharmaceutical appropriate for use in conjunction with this imaging modality must be considered carefully.

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