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Overview

Magnetic Resonance Imaging of Reptiles, Rodents, and Lagomorphs for Clinical Diagnosis and Animal Research

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MRI is a great diagnostic tool for evaluating exotic animals, particularly in clinical practice and in research with animal models. Here we review various aspects of MRI of reptiles, rodents, and lagomorphs, including the indications for this modality, the preparation of subjects, and protocols for imaging various organs and the musculoskeletal system. Protocols for the anesthesia and immobilization of subjects to facilitate their imaging are discussed also.

Increasingly advanced imaging methods are introduced to the treatment of exotic animals and rodents to improve diagnostic accuracy, determine prognosis, and select the optimal treatment. Most of those methods have already been used in companion animals, but they have to be modified for use in small mammals and reptiles due to differences in anatomy and physiology.[27] The low body weight (240 to 850 g) of these animals also influences MR image quality.[26] MRI and CT are popular 3D imaging methods in experiments involving animal models (rats, mice, rabbits) to investigate the pathophysiology of human diseases.[25] For example, MRI has been used experimentally in small mammals to study pyelonephritis, spinal abscesses, synovitis, and bacterial sinusitis in rabbits.[23,34] In research, MRI is regarded as a transitional stage between in vitro and in vivo studies.[14]

Pet owners' interest in both CT and MRI for birds, amphibians, fish, and small mammals has increased, although the owners were more likely to opt for CT due to the lower cost and greater availability of the procedure.[27] CT supports the acquisition of 3D images and the visualization of early pathologic changes that cannot be detected by conventional radiography. CT is a highly useful technique, in particular in dentistry, because it prevents skull bone structures from overlapping and obstructing dental evaluations. In addition, contrast media are used during CT to diagnose tumors, including those involving the skull.[27] However, the dose of ionizing radiation generated by a CT scanner is much higher than that emitted by an X-ray machine, and in this regard, MRI, which does not use ionizing radiation, offers a considerable advantage over CT. Due to the growing availability of MRI scanners in veterinary medicine, including private clinics,[16] MRI is a preferred imaging method for exotic animals (Figure 1).

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Subject Preparation

MRI involves long scanning times, and the animal typically is immobilized by using general anesthesia.[37] Alternatively, sedation minimizes respiratory movements and the number of heart contractions and reduces the risk of artifacts.[25,41] General anesthesia poses a risk in all animal species,[20,33] but the risk is greater in small animals, such as reptiles, rodents, and rabbits, than in cats and dogs.[7,39] The anesthesia-associated risk in small animals is exacerbated by stress responses during the induction of anesthesia; their high surface-area:volume ratio, which can lead rapidly to hypothermia; technical challenges associated with intubation; frequent vascular inflammation associated with venous access; and subclinical respiratory disorders.[39] Species-specific and interindividual differences may require the adaptation of standard anesthesia protocols. Protocols for the anesthesia and immobilization of animals are available in the literature, and they can be modified to suit individual needs.

For example, guinea pigs have been sedated for MRI by inhalation of a 2% to 3% mixture of isoflurane, followed by the administration of intravenous buprenorphine at 0.05 mg/kg.[13] By using a mixture of intramuscular xylazine (5 mg/kg) and ketamine (35 mg/kg), rabbits can be immobilized for 30 to 45 min, which is generally sufficient to complete the examination.[5,38] Nembutal (pentobarbital sodium) can be administered intraperitoneally to hamsters at 70 to 90 mg/kg body weight.[18] In mice, rats, and rabbits, a propofol injection can be used to induce short-term sedation lasting around 5 min;[34] however, this protocol is not highly effective for MRI studies because the sedative has to be administered several times, thus increasing the risk of displacement inside the coil.

Turtles typically do not require pharmacologic sedation, and only the head and limbs have to be mechanically immobilized during an examination. However, anxious turtles require pharmacologic sedation, to prevent the head and limb movements that can produce artifacts,[27,37] and all turtles undergoing direct MRI examinations of the head or limbs must be sedated.[27] The

Class	Family	Species
Reptilia	Pythonidae	*Python molurus*
	Cheloniidae	*Caretta caretta*
	Testudinidae	*Testudo graeca*
		Testudo hermanni
		Testudo horsfieldii
		Kinixys spp
	Emydidae	*Trachemys scripta*
Mammalia	Caviidae	*Cavia porcellus*
	Cricetidae	*Mesocricetus auratus*
	Muridae	*Rattus norvegicus*
		Mus musculus
	Leporidae	*Oryctolagus cuniculus*

Figure 1. Species mentioned in the text.

recommended analgesia protocol involves propofol (5 mg/kg) or a mixture of ketamine (5 mg/kg IV) and medetomidine (50 µg/kg IV).[29] All snakes have to be sedated to prevent movement inside the coil;[1,17,27] the recommended ketamine dose for snakes is 22 to 44 mg/kg IM.[4]

Subjects are positioned in accordance with the protocols developed for dogs and cats;[34] dorsal and ventral positions are most common. Reptiles should not be examined in the lateral view, which causes organ displacement and prevents correct interpretation of results.[27] The imaged area or organ should be positioned centrally inside the coil.[22] The coil should fully accommodate the subject's size to deliver high-quality images that are characterized by a satisfactory signal-to-noise ratio and spatial resolution. In addition, coils that are precisely adapted to the subject's size minimize the frequency of artifacts.[3] For example, small rodents can be examined effectively in a human wrist coil.[22]

Contrast Agents

Animals are often examined with the use of contrast agents, despite the risk of adverse reactions.[37] At present, 3 groups of contrast media are used in MRI: gadolinium chelates (gadopentetate dimeglumine, gadoteridol, gadodiamide, and gadoterate meglumine), manganese chelate (mangafodipir trisodium) and iron oxide particles (superparamagnetic iron oxide). Gadolinium chelates are widely available, and gadopentetate dimeglumine is used most frequently.[35] Gadolinium (gadopentetate dimeglumine) is delivered intravenously[25,31] or injected directly into joints[13,24] to visualize individual organs and joints. However when administered intravenously to rats, gadolinium chloride reportedly accumulated in lung and kidney capillaries and led to hepatocellular and splenic necrosis.[36] Because free gadolinium (III) ions are toxic, contrast media are rendered chemically stable by complexing Gd(III) ions with various compounds; the stability of the resulting complex is determined by the physicochemical properties of associated compound.[15] Anaphylactic shock is the key risk associated with the administration of contrast media. However, the frequency of adverse reactions is much lower for the gadolinium chelates used during MRI than for the iodinated contrast media often used for CT.[30]

Manganese-enhanced MRI, an imaging method that relies on Mn^{2+} ions, is successfully used in experiments targeting the brain. Due to their physicochemical similarity to Ca^{2+} ions, Mn^{2+} ions flow through calcium channels and are capable of binding to their receptors. Mn^{2+} ions shorten T1 and T2 relaxation times and supports mapping of functional brain activity. The key limitation of manganese-enhanced MRI is the potential hepatotoxicity and cardiotoxicity of Mn^{2+} ions.[6]

MRI of Reptiles

MRI has potential as an effective tool for reptile medicine and has been used for turtles and snakes in clinical practice. Radiology has limited diagnostic potential in turtles due to the low contrast between soft tissues and shell scales.[37] Similarly, ultrasound scanning is not highly effective in turtles because it does not visualize deep organs or those shielded by bones or gas.[2] MRI appears to be an advantageous method in turtles because it is by far more effective in cross-sectional examinations of internal organs than is CT. The intravascular injection of an MRI contrast agent (gadolinium) further supports the visualization of organ vasculature.[25] Turtles can be placed in the dorsoventral position inside a human knee coil.[32] In low-field MRI, organs should be scanned in T1-, T2- and proton-density–weighted sequences in the sagittal, dorsal, and transverse planes. Slice thickness should not exceed 5 mm to enhance the visualization of the heart muscle and major vessels, stomach, intestines, liver, urinary bladder, and kidneys. Lung tissues are characterized by very low signal intensity, so only the pulmonary septa is visualized effectively, but mild inflammatory reactions in the lung can be diagnosed in MRI scans.[37] Low-field MRI supports the detection of pathologic changes, including tumors, in the abdominal cavity. In a clinical study of a tortoise (body weight, 1803 g), MRI (0.5 T; T1-spin echo and T2-fast spin echo sequences; dorsal and transverse planes) revealed a hyperintense (T2-weighted) and hypointense (T1-weighted) mass, which was identified as cancerous during postmortem histopathologic analysis.[32] In high-field MRI, a 3D gradient-echo sequence with 0.5-mm slice thickness is recommended.[25]

MRI is a minimally invasive imaging method that supports repeated measurements of organ size in the same subject. This feature was used in a study[19] of a Burmese python (*Python molurus*) that ranged in weight from 227 to 635 g, and the antemortem MRI results were compared with postmortem findings. The snake was placed inside the coil of a 1.5-T MRI scanner in a ventral position, with distal parts of the body folded parallel to the central part of the body. The entire body was scanned simultaneously, and images were compiled with the use of computer software. Imaging involved 2D proton-density multislice spin-echo (slice thickness, 1 mm; repetition time, 4220 ms; echo time, 73 ms, including a prepulse for fat suppression) and 3D T1-weighted gradient-echo (repetition time, 9.3 ms; echo time, TE 3.3 ms) sequences. These sequences supported measurements of the heart, kidneys, liver, pancreas, and small bowel.[19]

MRI in Rodents

Mice and guinea pigs are the most popular animal models for osteoarthritis research, but symptoms of the disease are manifested earlier in guinea pigs. MRI is popularly used to diagnose musculoskeletal and joint disorders.[34] It is a highly effective and minimally invasive diagnostic method that supports evaluations of articular cartilage degradation. In addition, MRI is used to assess the effectiveness of pharmacologic treatment for synovial inflammation and cartilage degradation.[31] In the past, macroscopic and histopathologic analyses were the 'gold standards' regarding evaluations of articular cartilage damage in animals, but those methods are highly invasive and often require euthanasia. Alternatively, radiologic images do not enable accurate diagnosis of soft tissues.[3] Dunkin–Hartley guinea pigs are a good model for research into spontaneous osteoarthritis; the disease progresses similarly in Dunkin–Hartley guinea pigs as in humans.[13,14] The disease initially was diagnosed according to differences between the T1 and T2 times, but this method is not highly effective in early stages of disease. The administration of the gadodiamide contrast agent directly into joints produces much better results, but the injection makes the procedure more invasive.[13]

MRI is a highly accurate diagnostic tool in dental disorders which are very common in rodents. In those animals, the oral cavity is very difficult to examine because of its small size and the specific anatomy of the temporomandibular joint; these features typically allow the evaluation of superficial changes only.[27] Furthermore, 2D radiologic images often do not reveal bone loss or osteomyelitis in areas that are shielded by other bone structures and soft tissues.[8] In contrast, MRI is highly effective in visualizing soft tissues and, unlike conventional radiology, can be used to diagnose even small dental abscesses.[9]

MRI is an excellent diagnostic tool for examinations of the rodent abdominal cavity. Turbo spin-echo and T2-weighted, fat-attenuation sequences in 3 planes were effective in cross-sectional studies imaging the livers of 6-wk-old (weight, 120 to 150 g) golden hamsters.[18] Those sequences effectively highlighted small structures, such as bile ducts, including their dilation, inflammation, and fibrosis. In the T2-weighted sequence, the signal intensity of the liver is lower than that of the spleen and kidneys, and the liver appears darker in scans.[18]

Mice and rats are among the most popular laboratory animals, and they are widely used as animal models in research. In a study of the pathogenesis of inflammatory brain changes that accompany Parkinson disease, rat brains were analyzed by using high-field MRI (9.4 T); the rats weighed 200 to 250 g. After striatal injection of the neurotoxin 6-hydroxydopamine, multilayered foci of decreased signal intensity were observed in T2-weighted gradient-echo images, and the signal intensity during the T1-weighted spin-echo sequence increased after intravenous administration of a contrast agent (Gd-DTPA).[40]

In rats, Mn^{2+}-enhanced MRI supports detailed analysis of brain structures. The images obtained 24 h after intraperitoneal injection of a Mn^{2+} solution (dose, 100 mg/kg; body weight, 275 to 385 g) were characterized by higher signal intensity in specific brain regions on T1-weighted FLASH scans in comparison with the same areas visualized by using a T2-weighted RARE fast spin-echo sequence.[6] The Mn^{2+} solution has also been used to examine the structure and function of the optical nerve in rats, including during pathologic conditions, such as glaucoma.[10] A T1-weighted FLASH MRI sequence was applied to assess heart function, measure Ca^{2+} ion flow, and determine the cardiac inotropic state in mice.[21]

MRI in Lagomorphs

Rabbits are excellent animal models for studies of human skeletal and nervous system disorders. In rabbits with surgically and immunologically induced osteoarthritis, articular cartilage was assessed in a 1.5-T MRI scanner (field of view, 8 cm; thickness of the sagittal section – 0.7 mm) after intravenous administration of the gadolinium contrast.[31] The results revealed that normal articular cartilage in adult rabbits is characterized by a short T2 time.[12,31] Increases in signal intensity at the sites of articular cartilage damage corresponded to changes that were observed postmortem in histopathologic examinations.[31] The recommended protocol for visualizing articular cartilage in 3-kg rabbits comprises 3D spoiled gradient–recalled echo (SPGR) and 2D fast spin-echo (FSE) sequences. In 4-T MRI, the recommended 3D SPGR sequence was applied in 2 variants: with and without attenuation of adipose tissue.[3] The scan was performed in the sagittal plane by using a transmit–receive cylindrical high-pass radio frequency birdcage coil. The results were characterized by high sensitivity and specificity, suggesting that the described sequences might be used to visualize articular cartilage in other animal species other than rabbits.[3]

MRI was a highly useful tool in diagnosing infectious and noninfectious arthritis in rabbits.[38] The T1-weighted localizer sequence in the coronal plane was followed by a T2-weighted sequence and, before and after contrast administration, the T1-weighted sequence in the sagittal plane with adipose tissue attenuation.[38]

New Zealand rabbits also were used as animal models in a study in which brain changes that accompany Alzheimer disease were diagnosed by MRI.[28] Specifically, atrophy of temporal lobe cells and enlargement of lateral cerebral ventricles were evaluated. Lateral ventricles were analyzed with the use of standard tools programmed in the scanner software. The MRI protocol (0.2 T) involved T1, T2, and 3D gradient–echo sequences in the sagittal plane, which promoted optimal visualization of the examined structures.[28] In addition, rabbits are useful models in studies of spine conditions. In particular, rabbits were used to investigate the pathogenesis of degenerative lumbar spine disease resulting from ischemia. [11] The size of the analyzed structures (rabbit weight, 3200 g) supported the evaluation of microcirculatory problems. T1-weighted, fat-suppressed T1-, and T2-weighted sequences were used to detect hematomas that exerted pressure on spinal cord nerves.[11]

Conclusions

Reptiles, rodents, and lagomorphs are increasingly popular companion animals. The awareness and expectations of animal owners have increased considerably in recent years, thus prompting veterinary professionals to incorporate increasingly advanced diagnostic methods and treatments into routine practice. MRI imparts high contrast to soft tissues; it therefore is highly effective in diagnosing musculoskeletal disorders and visualizing internal organs that cannot be accessed by ultrasonography. For this reason, MRI can be regarded as the ‘gold standard’ diagnostic imaging modality for reptiles, rodents. and lagomorphs. Very few publications discuss the use of MRI in amphibians, and further research is required in this area. The development of protocols for visualizing various organs and systems in laboratory animals will contribute to the standardization of methods in animal research to bridge the gap between in vitro and in vivo studies.

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