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Case report

Cerebral thromboembolism secondary to infective endocarditis in a pet rabbit

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ABSTRACT

Central vestibular disease is one of the most common problems in pet rabbits. *Encephalitozoon cuniculi* infection appears to be the most frequent etiology, but other causes have been described. It is known that infective endocarditis can cause thromboembolic disease affecting multiple organ systems in dogs. Secondary embolism from bacterial endocarditis commonly occurs, mainly in the spleen and kidneys; but cerebral thromboembolism disease secondary to infective endocarditis has rarely been reported in dogs. In this case, an 8-year-old, neutered male rabbit (*Oryctolagus cuniculus*) presented with mild hyperthermia, left head tilt and rolling. Radiographic examination, complete blood count, serum biochemistry and abdominal ultrasound were unremarkable. Magnetic resonance imaging did not show any abnormalities in the central nervous system. An *Encephalitozoon cuniculi* serology was performed with elevated IgM and IgG titers (1:320; 1:1280 respectively), the patient was hospitalized and treated for encephalitozoonis but the rabbit died 8 days after presentation and the body was submitted for necropsy. Histopathological examination revealed multifocal necrotizing and suppurative myocarditis, with multiple emboli in kidneys, lungs and central nervous system. Polymerase chain reaction, immunohistochemistry, and Ziehl-Neelsen staining of samples from the central nervous system and kidneys yielded negative results for *Encephalitozoon cuniculi*. Due to the positive predictive value of 92% of indirect fluorescent antibody technique *Encephalitozoon cuniculi* serology, in this case a false positive was suspected. Based on the results, final diagnosis of cerebral thromboembolism secondary to infective endocarditis was emitted. This is the first report of this process naturally occurring in rabbits, and it should be included in the differential diagnosis of central vestibular syndrome in this species.

Introduction

Vestibular signs are the most common presentation of neurologic disease in pet rabbits [1,2]. *Encephalitozoon cuniculi* infection appears to be the most frequent cause of central vestibular disease, but other etiologies have been described. Bacterial otitis media/interna is the main causes of peripheral vestibular disease [4]. There are no reports of cerebral thromboembolisms secondary to infective endocarditis (IE) in exotic pets. Infective endocarditis is uncommon in dogs and thromboembolism occurs in 70–80% the reported incidence of emboli involving the brain is 0.1–1% [5–7].

Case presentation

An 8-year-old neutered male rabbit (*Oryctolagus cuniculus*) presented with acute left head tilt and whole body rolling. The rabbit was housed in a large commercial cage with access to the room, and was fed a commercial pellet diet for pet rabbits, hay and carrots. No other pets were housed at home.

On physical examination, the animal was alert and weighed 2.3 kg. Persistent mild hyperthermia (39.7°C) was also noted [8]. The neurological examination revealed left head tilt and whole body rolling, horizontal nystagmus with the fast phase away from the head tilt, although placing and postural reactions were apparently normal. Initial

Abbreviations: CNS, Central nervous system; IE, Infective endocarditis; MRI, Magnetic resonance imaging; E. cuniculi, Encephalitozoon cuniculi; PCR, Polymerase chain reaction

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differential diagnosis included *E. cuniculi,* bacterial infection, trauma and neoplasia. Complete blood count and serum biochemistry were performed to rule out inflammatory/infectious process. They were within normal ranges [9].

Imaging was performed in order to determine if medical condition, especially related to encephalitozoonosis, other infectious process such as pneumonia, neoplasia and any sign of a secondary gastrointestinal ileus were present. Total body radiographs showed increased urinary bladder opacity consistent with hypercalciuria. Urinalysis, collected by cystocentesis, revealed proteinuria (+1) and hematuria (+1); then, an abdominal ultrasound was performed, and a microbiological culture was taken. Abdominal ultrasound was unremarkable. The urine microbiological culture was negative.

The patient was hospitalized with intravenous fluid therapy with balanced crystalloid fluid (Ringer Lactate Braun® 500 mL, Braun Veterinaria, Barcelona, Spain) supplemented with 5% of dextrose at 100mL/kg/day, and ranitidine 2mg/kg IV every 12 hours (Ranitidina Normon, Laboratorios Normon SA, Madrid, Spain), meloxicam 0.2mg/kg IV every 12 hours (Metacam®, Boehringer, Ingelheim, Germany). Assisted oral feeding with critical care formula diet for herbivores (Oxbow Animal Health, Murdock, NE, U.S.A) was administered at 15–20 mL/kg every 8 hours mixing one part of the powder with two parts of warm water.

Due to the suspicion of a central nervous system (CNS) infectious and/or neoplastic process, a low-field magnetic resonance imaging (MRI) scan of the brain (0.4 Teslas Hitachi Aperto, Tokyo, Japan) was performed the following day. The patient was premedicated with midazolam 0.5mg/kg IV (Midazolam Normon, Laboratorios Normon SA, Madrid, Spain) and butorphanol 0,5mg/kg IV (Torbugesic®Vet, Zoetis Spain SL, Madrid, Spain). General anesthesia was induced with propofol 5mg/kg IV (Propovet®, Zoetis Spain SL, Madrid, Spain). The patient was intubated with a 2.0 uncuffed endotracheal tube and maintained with sevoflurane 3–4% (Sevoflo® sevo- flurane 100%, Abbott) and 100% oxygen during the MRI.

MRI showed a mild presence of fluid in the right tympanic bulla compatible with otitis media, and fluid in both external ear canals compatible with otitis externa or ceruminous debris. No intracranial lesions were found.

An *E. cuniculi* serology with indirect fluorescent antibody technique showed a marked increase in IgM and IgG antibodies (1:320; 1:1280 respectively) [10] and a urine *E. cuniculi* polymerase chain reaction (PCR) (Vetgenomics, Barcelona, Spain) was performed and yielded negative results.

Due to suspected encephalitozoonosis and secondary complications stemming from the hematuria, fenbendazole 20mg/kg PO every 24 hours (Panacur® 100mg/ml, MSD Animal Health, Salamanca, Spain), enrofloxacin 10mg/kg IV every 12 hours (Baytril® 50mg/ml injectable solution, Bayer Hispania SL, Barcelona, Spain) were added to the initial treatment. The rabbit's condition did not improve and it was discharged as per owner's wishes with the same medication as described above to be given orally. It died 48 hours later and was submitted for necropsy.

Gross postmortem examination showed bilateral cranioventral pulmonary consolidation, white millimetric nodules throughout the myocardium, and similar nodules were found in both kidneys. No remarkable lesions were identified in the mitral valve. The presence of ceruminous debris was observed in the bilateral external auditory canal, although not in the tympanic bulla. Additionally, the right kidney showed two large flat areas of pale discoloration on the renal surface, measuring 1 and 3 cm in diameter, surrounded by a red halo, which extended radially to the medulla. No changes were found in other internal organs and neither on gross examination of the CNS. Tissues from all the internal organs were formalin-fixed paraffin-embedded for 24 hours and processed routinely for histopathology. Fresh or frozen samples were not collected.

Histopathological examination revealed multifocal necrotizing and suppurative myocarditis (Fig. 1), as well as fibrinosuppurative mitral endocarditis (Fig. 2). The lungs showed suppurative bronchopneumonia,

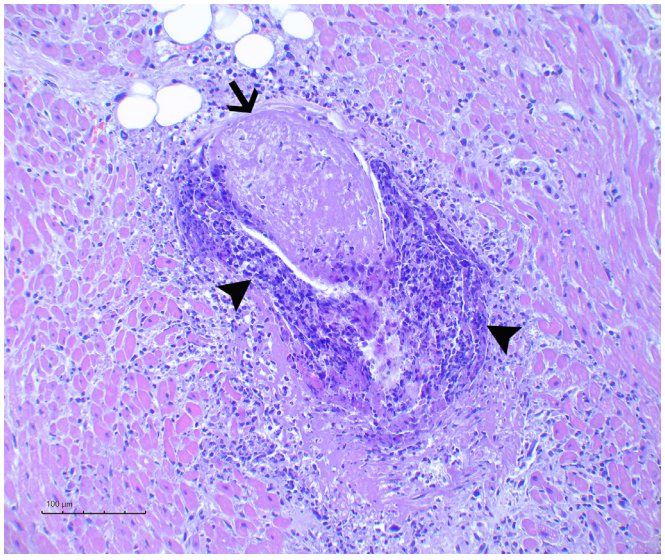

Fig. 1. Histopathology of the heart of the rabbit with cerebral thromboembolism secondary to infective endocarditis. A fibrin thrombus (arrow), and large amounts of degenerated leukocytes (arrowheads) within the myocardial arteriolar lumen were observed. The vascular wall was circumferentially and transmurally effaced by fibrin (fibrinoid change) and degenerated leukocytes (vasculitis).

and the kidneys showed multifocal embolic nephritis (Fig. 3). All these lesions were accompanied by deeply basophilic bacterial colonies showing a coccoid morphology. Examination of the CNS revealed multifocal and bilateral histological lesions. The leptomeninges, choroid plexus, white and gray matter of the cerebellum and cerebrum, and to a lesser extent, the brainstem, showed well demarcated and nonencapsulated inflammatory infiltrates, composed of markedly degenerated heterophils and a few macrophages, with abundant necrotic debris and similar bacterial colonies to those observed in the other organs (Figs. 4, 5 and 6A). Vasculitis was observed in some affected areas, capillaries and arterioles presented necrotic intramural leucocytes (vasculitis), and their lumen was occasionally occluded by the fibrin thrombi and/or coccoid bacteria. No foci of nonsuppurative inflammation which could be suggestive

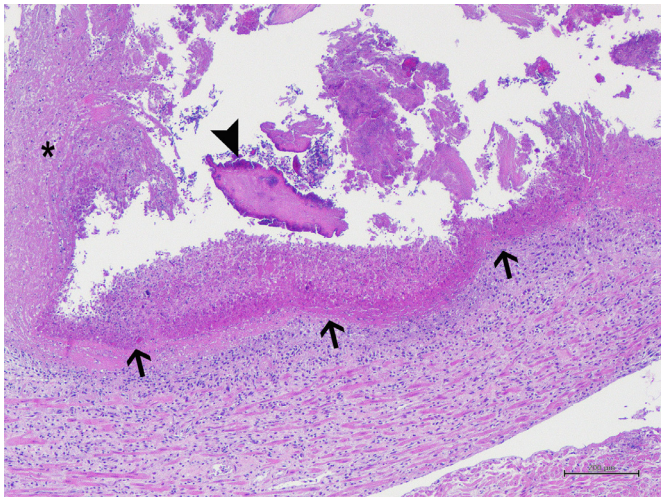

Fig. 2. Histopathology of the heart of the rabbit with cerebral thromboembolism secondary to infective endocarditis. It showed a fibrinosuppurative mitral endocarditis. The mitral valve (asterisk) and the adjacent endocardium (arrows) were replaced by abundant amounts of fibrin, degenerated heterophils and coccoid bacterial aggregates, which further extended to the underlying myocardium. Septic thrombi (arrowhead) were detached and could be seen within the ventricular lumen.

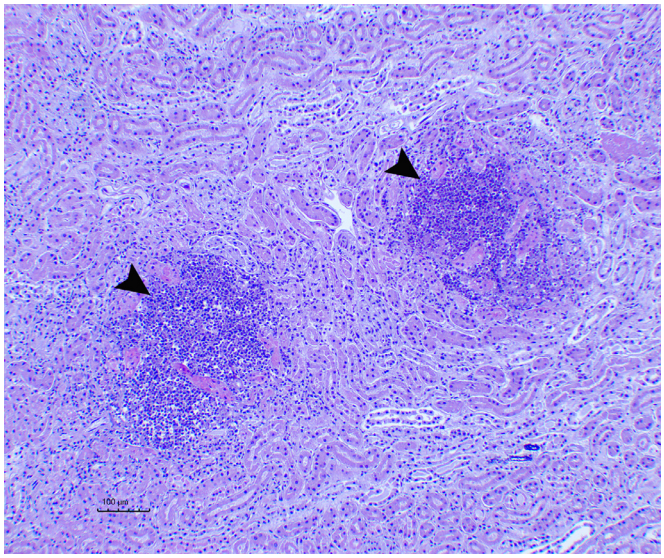

Fig. 3. Histopathology of the kidney of the rabbit with cerebral thromboembolism secondary to infective endocarditis. Multifocal areas of lytic necrosis with degenerated heterophils (microabscesses) could be seen scattered throughout renal parenchyma (arrowheads).

of *E. cuniculi* could be observed in any of the brain sections assessed. Gram staining of the tissue sections of the CNS confirmed the presence of Gram positive cocci (Fig. 6B).

An immunohistochemistry analysis using a polyclonal antiserum against *E. cuniculi* (Medicago, Quebec, Canada) and Ziehl-Neelsen staining were performed. Both tests yielded negative results for *E. cuniculi* compared to positive controls from previously diagnosed cases. *Encephalitozoon cuniculi* was targeted in FFPE kidneys and CNS tissue samples using Vetgenomics PCR, as described in urine. It yielded a negative result.

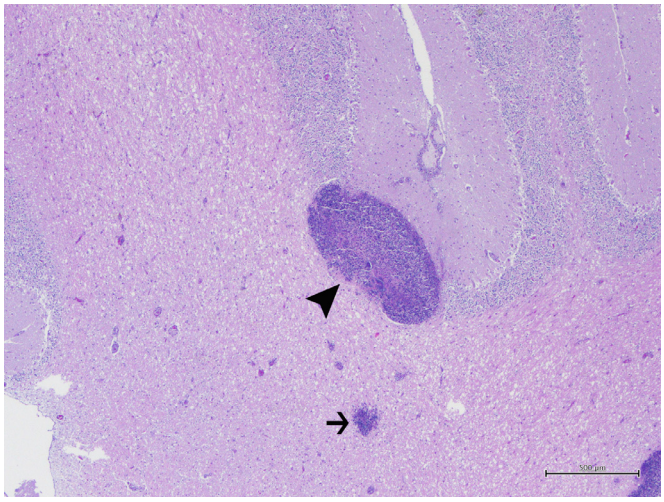

Fig. 4. Histopathology of the cerebellar cortex of the rabbit with cerebral thromboembolism secondary to infective endocarditis. A large area of lytic necrosis with high amounts of degenerated heterophils (microabscess) measuring approximately 600um in diameter is observed disrupting the molecular, Purkinje and granular layers of the cerebellar cortex (arrowhead). Similar lesions of smaller caliber can be found within cerebellar white matter (arrow).

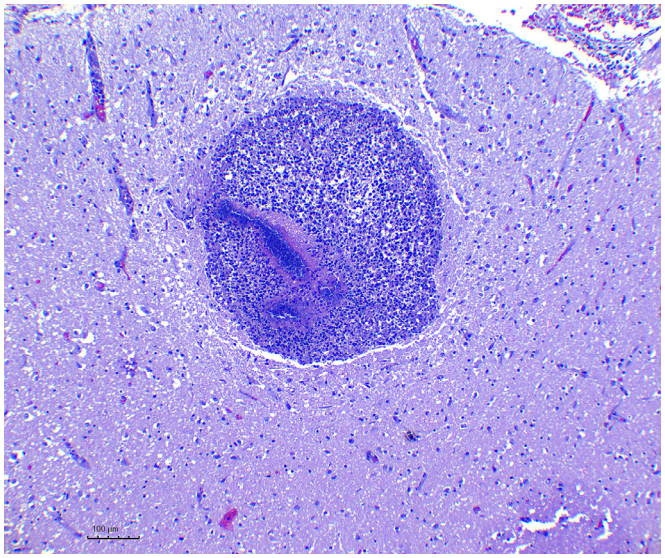

Fig. 5. Histopathology of the cerebral cortex of the rabbit with cerebral thromboembolism secondary to infective endocarditis. A large area of lytic necrosis with high amounts of degenerated heterophils (microabscess) measuring approximately 400um in diameter was observed featuring a necrotic blood vessel in the center with abundant coccoid bacteria in its lumen.

Discussion

In a retrospective study of rabbits with neurological disease, the most commonly observed clinical signs were vestibular signs [1]. Differentiating peripheral from central vestibular syndrome can be challenging, as it is difficult to perform an accurate neurological examination on pet rabbits, due to their prey response [11–13]. In addition, *E. cuniculi* central nervous system infections cause signs resembling peripheral dysfunction in this species [14]. In rabbits *E. cuniculi* infections are commonly chronic and subclinical, with a wide range of seroprevalence percentage, from 15% to 84,7% [15]. Other reported causes of central vestibular syndrome in rabbits include: bacterial infection; toxoplasmosis; herpes virus encephalitis; cerebrovascular accident; cerebral larva migrans; trauma; lead toxicity; neoplasia; sepsis; hepatic encephalopathy; osteomyelitis of the petrous temporal bone, rabies, hypoxia and cerebellum infarction [1,3,4,16]. On the other hand, the most frequent cause of peripheral vestibular disease is otitis media/interna, while traumatic rupture of the tympanic bulla, aminoglycoside-mediated ototoxicity, and idiopathic vestibular disease are other less common causes of peripheral vestibular disease [3,4].

In the case presented here, *E. cuniculi* infection was ruled out as a cause of neurological signs based on the absence of focal, nonsuppurative meningoencephalitis lesions on histopathological examination, these lesions being typical for encephalitozoonosis [3,4,17]. Although a serology resulted positive, a PCR, immunohistochemistry and Ziehl-Neelsen staining from samples of CNS and kidneys yielded negative results. Brain and kidneys were selected because they have the highest diagnostic value for detection of *E. cuniculi* by histology, IHC, and real-time PCR [17]. It is demonstrated that real-time PCR is the most sensitive method for the confirmation of *E. cuniculi* infection, followed by immunohistochemistry and histological spore detection. The gold standard for confirmation of the pathogenic role of *E. cuniculi* infection would be a combination of histological investigation of predilection sites with real-time PCR performed on the above-mentioned tissues [17]. Cray et al. reported a positive predictive value of a serology with IgM >1:64 and IgG >1:512 of 92%, so the remaining 8% could explain the false positive serologic determination resulting in the case here [10,18].

Reports of endocarditis in rabbits are limited to experimental infection with one exception, in which *Pasteurella multocida* was isolated

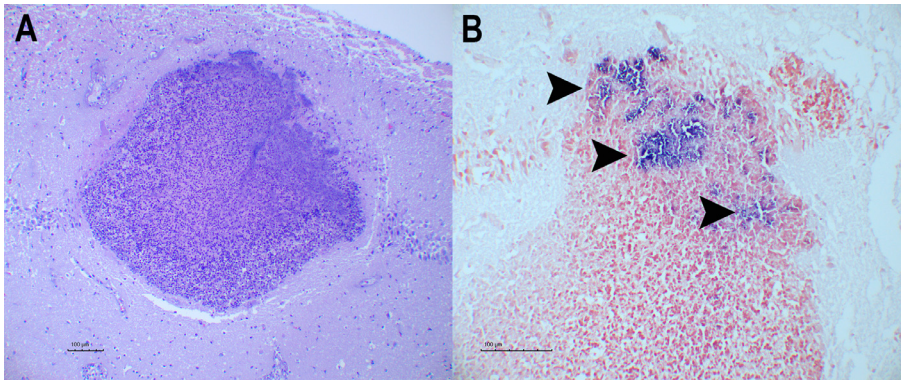

Fig. 6. Histopathology of the hippocampus of the rabbit with cerebral thromboembolism secondary to infective endocarditis. (A) A large area of lytic necrosis with high amounts of degenerated heterophils (microabscess) measuring approximately 600um in diameter was observed disrupting the hippocampus. (B) Gram stain showed coccoid Gram-positive bacterial aggregates (arrowheads).

[19]. In the case presented here, *Pasteurella multocida* was ruled out because the histopathological examination revealed gram positive cocci colonies. Clinical signs of IE may include detectable heart murmur and fever in 74% and 80–90% in dogs, respectively [7]. In the case of the rabbit here, no murmur was detected but the animal presented with mild hyperthermia. Unfortunately, a blood culture was not performed, as IE was not suspected.

Central nervous system thromboembolic disease secondary to bacterial endocarditis has been described in cattle, rats, seals, pigs, horses and ibis; neurological clinical signs were only observed in cattle and horses [20–26].

This is the first case report of a naturally occurring cerebral thromboembolism secondary to IE in a pet rabbit. In summary, cerebral thromboembolism should be included in the differential diagnosis of central vestibular syndrome in rabbits. Given the results of this case report, it may be important to recommend considering blood culture or thoracic imaging in the case of patients that present with acute central vestibular signs and are febrile.

Authors' contributions

FS was the primary clinician and wrote the article. TB was an assistant clinician of the case and reviewed the article. AC performed the necropsy and histopathology, informed the necropsy and reviewed the article. MF performed the magnetic resonance, informed the images and reviewed the article. JM supervised the case, wrote and reviewed the article.

Declaration of interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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References

[1] Gruber A, Pakozdy A, Weissenböck H, Csokai J, Künzel F. A retrospective study of neurological disease in 118 rabbits. J Comp Pathol 2009;140(1):31–7. doi: 10.1016/j.jcpa.2008.09.009.
[2] Künzel F, Doz P, Small D. Clinical signs, diagnosis and treatment of encephalitozoon cuniculi infection in rabbits. Vet Clin NA Exot Pet 2018;21(1):69–82. doi: 10.1016/j.cvex.2017.08.002.
[3] Meredith AL, Richardson J. Neurological diseases of rabbits and rodents. J Exot Pet Med 2015;24(1):21–33. doi: 10.1053/j.jepm.2014.12.007.
[4] Latney L V, Bradley CW, Wyre NR. Encephalitozoon cuniculi in pet rabbits: diagnosis and optimal management. Vet Med Res Rep 2014;5:169–80. doi: 10.2147/VMRR.S49842.
[5] Sykes JE, Kittleson MD, Chomel BB, Macdonald KA, Pesavento PA. Clinicopathologic findings and outcome in dogs with infective endocarditis: 71 cases (1992–2005). J Am Vet Med Assoc 2006;228(11):1735–47. doi: 10.2460/javma.228.11.1735.
[6] MacDonald K. Infective endocarditis in dogs: diagnosis and therapy. Vet Clin North Am - Small Anim Pract 2010;40(4):665–84. doi: 10.1016/j.cvsm.2010.03.010.
[7] Cook LB, Coates JR, Dewey CW, Gordon S, Miller MW, Bahr A. Vascular encephalopathy associated with bacterial endocarditis in four dogs. J Am Anim Hosp Assoc 2005;41(4):252–8. doi: 10.5326/0410252.
[8] Di Girolamo N, Toth G, Selleri P. Prognostic value of rectal temperature at hospital admission in client-owned rabbits. J Am Vet Med Assoc 2016;248(3):288–97. doi: 10.2460/JAVMA.248.3.288.
[9] Leineweber C, Müller E, Marschang RE. Blood reference intervals for rabbits (Oryctolagus cuniculus) from routine diagnostic samples. Tierarztl Prax Ausgabe K Kleintiere - Heimtiere 2018;46(6):393–8. doi: 10.1055/s-0038-1677403.
[10] Cray C, McKenny S, Perritt E, Arheart KL. Utility of IgM titers with IgG and C-reactive protein quantitation in the diagnosis of suspected encephalitozoon cuniculi infection in rabbits. J Exot Pet Med 2015;24(3):356–60. doi: 10.1053/j.jepm.2015.06.017.
[11] Warnefors E, Rueløkke ML, Gredal H. Results of a modified neurological examination in 26 healthy rabbits. J Exot Pet Med 2019;30:54–9. doi: 10.1053/j.jepm.2018.01.010.
[12] Mancinelli E. Neurologic examination and diagnostic testing in rabbits, ferrets, and rodents. J Exot Pet Med 2015;24(1):52–64. doi: 10.1053/j.jepm.2014.12.006.
[13] Vernau KM, Osofsky A, LeCouteur RA. The neurological examination and lesion localization in the companion rabbit (oryctolagus cuniculus). Vet Clin North Am - Exot Anim Pract 2007;10(3):731–58. doi: 10.1016/j.cvex.2007.05.003.
[14] Künzel F, Gruber A, Tichy A, et al. Clinical symptoms and diagnosis of encephalitozoonosis in pet rabbits. Vet Parasitol 2008;151(2-4):115–24. doi: 10.1016/j.vetpar.2007.11.005.
[15] Latney L V, Bradley CW, Wyre NR. Encephalitozoon cuniculi in pet rabbits: diagnosis and optimal management. Vet Med Res Reports 2014;5:169–80. doi: 10.2147/VMRR.S49842.
[16] García R, Añor S, de la Fuente C, Novellas R, Soler V, Martorell J. Paradoxical vestibular syndrome caused by a presumptive cerebellar infarction in a rabbit. Top Companion Anim Med 2021;43:0–2. doi: 10.1016/j.tcam.2021.100509.
[17] Leipig M, Matiasek K, Rinder H, et al. Value of histopathology, immunohistochemistry, and real- time polymerase chain reaction in the confirmatory diagnosis of Encephalitozoon cuniculi infection in rabbits. J Vet Diagnostic Investig 2013;25(1):16–26. doi: 10.1177/1040638712466394.
[18] Cray C, Liebl MP, Arheart K, Peschke R, Frank K, Joachim A. Comparison of ELISA and IFAT for determination of anti-Encephalitozoon cuniculi antibodies in sera from rabbits with different clinical and histopathological presentations. J Exot Pet Med 2020;32:39–42. doi: 10.1053/j.jepm.2019.11.003.
[19] Vigneshwar R, Imayarasi K, Divisha R. A rare case report of Pasteurella multocida infective valvular endocarditis in a laboratory rabbit. Pharma Innov J 2020;9(12):82–5 SP.
[20] Henderson B, Diaz M, Martins C, Kenney D, Baird JD, Arroyo LG. Valvular endocarditis in the horse: 20 cases (1993–2020). Can Vet J 2020;61:1290–4.
[21] Greenwood AG, Marshall J, Tinsley EGF. Vegetative endocarditis in a Waldrapp ibis. Avian Pathol 1996;25(2):387–91. doi: 10.1080/03079459608419149.
[22] Chinnadurai A, Sathya K, Brigid V, Karen N, Septicemia Ryan S. Endocarditis and cerebral infarction due to staphylococcus aureus in a harp seal (phoca groenlandica). J Zoo Wildl Med 2009;40(2). doi: 10.1638/2008-0176.1.
[23] Delbosc S, Desilles J, Journé C, Diallo D. Neurologic complications of infective endocarditis: a joint model for a septic thromboembolism and inflammatory small vessel disease. Crit Care Med 2019;47(8):685–92. doi: 10.1097/CCM.0000000000003796.
[24] Karstrup CC, Jensen HE, Aalbæk B, Leifsson PS, Boye M. Endocarditis-associated brain lesions in slaughter pigs. J Comp Pathol 2011;144(4):289–95. doi: 10.1016/j.jcpa.2010.10.009.
[25] Maxson AD, Reef VB. Bacterial endocarditis in horses: ten cases (1984-1995). Equine Vet J 1997;29(5):394–9. doi: 10.1111/j.2042-3306.1997.tb03146.x.
[26] Revue LA, Canadienne V. Infectious thromboembolic meningoencephalitis in Ontario cattle. Can Vet J 1970;11(7):125–30.