In Vivo Pathological Mapping of the Rat Brain Infected with Angiostrongylus Cantonensis using MRI

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Introduction

Angiostrongylus cantonensis (A. cantonensis) is the most common cause of eosinophilic meningoencephalitis in Taiwan. This parasitic infection is endemic in the Southeast Asian and Pacific region, but it becomes a global infection in recent years. The infection in the final host, rats, or non-permissive host, including human, is acquired by ingesting contaminated raw snails. The third-stage larvae migrate to the brain and develop into the fifth stage with twice molts. The worms then migrate to lung and heart and develop into adult worm. The typical clinical presentation is acute eosinophilic meningoencephalitis frequently accompanied by brain and spinal cord disorders, and other symptoms of central nervous system (CNS) [1]. The features of the pathological changes in the brain were previously limited to a few case reports and techniques [2, 3]. Previously the diagnosis was established by immunodiagnosis, lumbar puncture and eosinophilia examination. Fourth- or fifth-stage larvae could be found in the cerebrospinal fluid (CSF) with lumbar puncture. Improper puncture and false positive response of immune resulted in an erroneous diagnosis. Therefore, the purpose of this study was to determine the lesion localization, pathological changes and angiostrongyliasis characterization of rat brain infected with larvae of *A. cantonensis* by magnetic resonance imaging (MRI) techniques. The results were verified with histopathological study. Rats were infected with different numbers of *A. cantonensis* larvae and their brains were diagnosed continuously with MRI and histopathological study. The association between the clinical features of the rats and MRI findings was also addressed.

Materials and Methods

In parasite infection, third-stage larvae of *A. cantonensis* were collected from infected *Achatina fulica* snails in Taiping, Taichung, and were isolated by artificial digestion using 0.08% pepsin in 0.7% HCl for 1 hr at 37°C. A total of six male Wistar rats weighing 250 - 300 g (12 weeks old) were used. Three of them were orally inoculated with 100 larvae using a metal fedding tube, and the other three was orally inoculated with 300 larvae, respectively. MR scans were performed before and after infection of *A. cantonensis*. In order to determine the permeability changes of blood-brain barrier after infection, gadodiamide was given by intraperitoneally injecting 151 mg/kg of a 0.5 M Gd-DTPA solution. T1W imaging was performed immediately after T1-shortening contrast agent (gadodiamide) administration. During the MR scanning, the rats were anesthetized with 2% isoflurane mixed with O_2, maintained with 1.5% isoflurane. Rat body temperature was maintained at 37 °C using warm water circulation.

The experiment was performed on a 1.5T MRI system (Magnetom Sonata; Siemens Medical Systems, Erlangen, Germany). A surface coil was used for RF reception. Three imaging sequences were performed to acquire whole brain T2 weighted (T2W) images as follows: Multi-slice turbo spin echo (TSE) sequence was performed to obtain T2W images with TR/TE = 3760/114 ms; Fluid attenuation inversion recovery (FLAIR) was performed to obtain T2W images with TR/TE/TI = 8420/155/2500 ms; Half fourier acquisition single shot turbo spin echo (HASTE) was performed to obtain T2W images with TR/TE = 2000/95 ms. All sequences were performed with in-plane resolution = 195μm x 390μm and slice thickness = 1.5 mm. To improve detection sensitivity over the full extent of T2 changes caused by the infection of *A. cantonensis*, image data for R2 mapping were acquired. To obtain R2 mapping, single-slice multi-echo spin echo sequence with half spatial resolution was performed to acquire 32 sets of images corresponding to 32 different TEs, ranging from 15 to 480 ms, to sample along the decay of transverse magnetization. A multi-slice fast spin echo sequence was also performed to obtain contrast-enhanced T1W images in the end of scans with TR/TE = 513/46 ms, in-plane resolution = 195μm x 390μm and slice thickness = 1.5 mm.

Results and Discussions

Abnormal findings on MR images were observed in each rat infected with different numbers of *A. cantonensis* larvae. However, each group of the infected rats with different degrees was found to have variable pathological changes in the brain tissue. In the infection of 100 *A. cantonensis* larvae, hyperintensities near the left and right hippocampus, left and right lateral ventricles, and right dentatus gyrus were found on T2W images of the rat infected after 6 to 28 days (Fig. 1a - 1c, 1e). In the same period, the contrast-enhanced T1W images after intravenous administration of gadolinium showed hypointensities without enhancement near the left and right hippocampus, left and right lateral ventricles and right tractus (Fig. 1d). Both T2W and contrast-enhanced T1W images showed hyperintensities in subarachnoid space of the rat brain infected after 20 to 28 days (Fig. 1). In the infection of 300 *A. cantonensis* larvae, T2W and contrast-enhanced T1W images showed more obvious hyperintensities and hypointensities in the same areas of the rat brains (than infection of 100 larvae) infected after 4 to 24 days (Fig. 2). T2W and contrast-enhanced T1W images both showed hyperintensities in subarachnoid space of the rat brain infected after 14 to 24 days (Fig. 2). Our MR results were consisted with histopathological study. Moreover, fifth-stage larvae of *A. cantonensis* in the brain parenchyma near subarachnoid space were found in most rats (Fig. 3).

Fig. 1

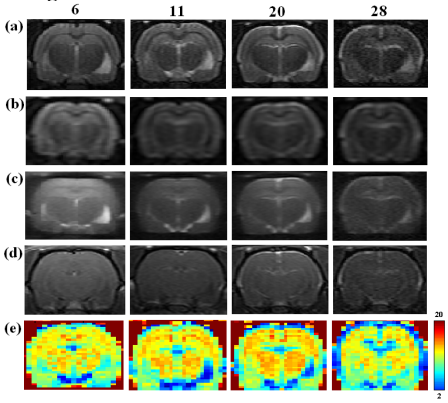

Fig. 2

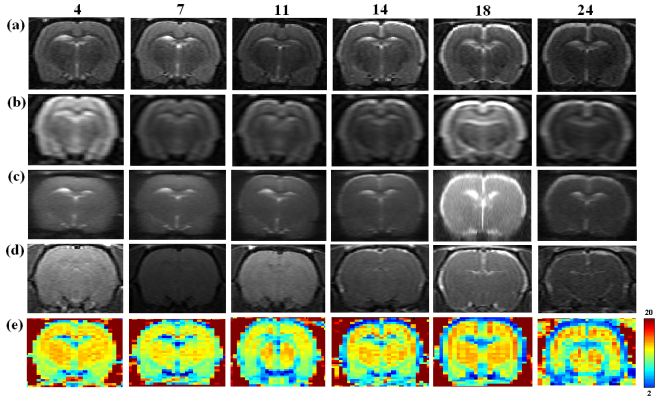

Fig. 3

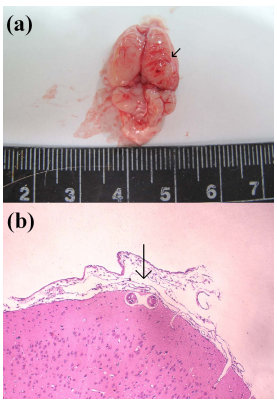

Fig. 1 (a) T2W images, (b) FLAIR, (c) HASTE, (d) contrast-enhanced T1W images and (e) R2 mapping of rat brains infected with 100 *A. Cantonensis* larvae after 6, 11, 20 and 28 days infection, respectively.

Fig. 2 (a) T2W images, (b) FLAIR, (c) HASTE, (d) contrast-enhanced T1W images and (e) R2 mapping of rat brains infected with 300 *A. Cantonensis* larvae after 4, 7, 11, 14, 18 and 24 days infection, respectively.

Fig. 3 (a) Photograph and (b) microphotograph of histopathology showed a fifth-stage larva in the brain parenchyma near subarachnoid space (arrow).

Conclusions

Our MRI results showed pathological changes in the rat brains infected with 300 *A. cantonensis* larvae were more severe than those infected with 100 *A. cantonensis* larvae. MRI was sensitive in showing tissue change and oedema, and provided higher tissue contrast and superior sensitivity in the detection of lesions. Therefore, MRI was suggested to be a non-invasive technique in localizing and characterizing lesions during the acute phase of angiostrongyliasis due to *A. cantonensis*.

References

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