Abstract

Leishmania siamensis is a newly emerged parasitic protozoan causing autochthonous infection in human in Thailand as well as in cattle and horse in Europe and USA. Since fundamental pathologic research of *L. siamensis* infection using murine experimental model is lacking, the aim of this research was to study parasite burden and pathology in BALB/c mice experimentally inoculated with $5x10^6$ *L. siamensis* promastigotes via subcutaneous (s.c.), intraperitoneal (i.p.) and intravenous route (i.v.). Control group was injected with 1x PBS via the same routes. On 7, 14, 28 and 112 days post-infection, dpi, microscopic examination of Giemsa-stained impression smear and molecular detection of *L. siamensis* DNA of liver and spleen were conducted to evaluate parasite burden. Hematocrit values, weights of livers and spleens were determined and comparatively evaluated among different routes and time points. Histopathology of liver, spleen and kidney was performed from formalin-fixed, paraffin-embedded tissue sections and H&E staining. By enumeration of amastigote-positive tissues from impression smear, levels of parasite burden in liver and spleen on 7, 14 and 28 dpi via i.v. route were found significantly higher than did other treatment and control groups ($p<0.05$). On 112 dpi via i.v. route, no parasite was detected in liver whereas significantly high level of amastigotes was still persistent in spleen ($p<0.05$). These results were also correlated with findings of *L. siamensis* DNAs in both organs during these time points. Compared to the control group, hematocrit values of intravenously inoculated mice were significantly lower on 7 and 14 dpi ($p<0.05$). The weights of liver in such group were significantly higher on 7, 14, 28 dpi similar to the weights of spleen in which they were significantly higher in all time points ($p<0.05$). The numbers of liver granuloma were significantly greater in all time points ($p<0.05$). Infection with *L. siamensis* via i.p. route was the only route of inoculation capable of inducing serological responses using direct agglutination test on 112 dpi. This study is the first to demonstrate that *L. siamensis* is pathogenic in experimental BALB/c mouse via i.v. and i.p. routes hence serving as a murine model for visceral leishmaniasis.

Keywords: *Leishmania siamensis*; BALB/c mouse; experimental infection; pathology