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Citation	Japanese Journal of Veterinary Research, 48(1), 89-89
Issue Date	2000-05-31
Doc URL	https://hdl.handle.net/2115/2845
Type	departmental bulletin paper
File Information	KJ00003408179.pdf



Evaluation of IgG/IgM-ELISA as a serodiagnostic method of tick-borne encephalitis
in comparison with neutralization test.

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In October 1993, a human case of encephalitis was diagnosed as tick-borne encephalitis (TBE) in Kamiiso, Hokkaido. The viruses were isolated from sentinel dogs, *I. ovatus* ticks, and wild rodents in the same area. These isolates were identified as TBE viruses of the Far Eastern subtype [Russian spring summer encephalitis (RSSE)]. Since the *I. ovatus* tick is a common species throughout Japan, TBE virus may be endemic not only at the area where a patient was found but also in other parts of Japan. It is possible that TBE may not have been reported because the clinical symptoms are similar to those of Japanese encephalitis (JE), which is endemic in Japan, and a high degree of cross-reactivity is seen in TBE and JE viruses in serological tests. But it was difficult to perform large scale epidemiological surveys in different areas, because neutralization test (NT), in which dangerous live viruses are used, was the only method for serological diagnosis of TBE. To specify the geographical distribution of TBE endemic foci, epidemiological surveys must be expanded using simple serological methods. In this study, sera from patients clinically diagnosed with TBE in Khabarovsk, the endemic area of the Far Eastern subtype, were examined by NT and IgM/IgG-ELISA. Since this kit was originally developed for diagnosis of TBE virus of Western subtype [Central European tick-borne encephalitis (CEE)], results of the two methods were

compared to see the applicability of the kit for the diagnosis of the Far Eastern subtype TBE.

First, sera of patients clinically diagnosed with TBE were examined by NT. And 16 of 31 (52%) of patients with paired sera were diagnosed as TBE cases, because NT antibody titers to TBE virus increased more than 4 times from the acute to the convalescent phase. Although in 13 of 31 (42%) of patients with paired sera with specific NT titer ($> 1 : 20$) to TBE virus, no significant increase of titer was observed. So those sera could not be diagnosed as TBE by NT. However, all could be diagnosed as TBE or non-TBE by IgM-ELISA test. More than 90% of TBE patient sera obtained 10 days or later from the onset of illness had TBE virus-IgM antibody. Therefore, even a single serum is useful for the diagnosis of TBE using IgM-ELISA test, if this serum is collected 10 days or later after disease onset.

The above results indicate that this IgM-ELISA test can be applicable for the serological diagnosis of TBE virus of the Far Eastern subtype. This ELISA is simpler and a more useful method than the NT test.

One virus strain was isolated by intracerebral inoculation method of suckling mice from the brain sample of a patient who was diagnosed with TBE using NT test. The isolated virus strain was identified as TBE virus by indirect immunofluorescent antibody (IFA) test using monoclonal antibodies specific to TBE virus.