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Fourteen Years' Surveillance of Coxsackievirus Group A in Kyoto 1996–2009 Using Mouse, RD-18S, and Vero Cells

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Coxsackievirus group A (CA) is known to cause flaccid paralysis in infant mice (mouse) as well as different arrays of clinical manifestations in humans, such as herpangina, hand, foot, and mouth disease, aseptic meningitis, and encephalitis. At the Kyoto City Institute of Health and Environmental Sciences, laboratory diagnosis of enteroviruses, including CA, is carried out by virus isolation and neutralization and/or complementary fixation (CF) assay as this has been found to be the most reliable method. However, although virus isolation using infant mice is time-consuming and requires specific expertise, this technique is still useful for the isolation of some human enterovirus A isolates.

The RD-18S cell line was cloned to efficiently isolate CA by cell culture (1). In this study, which was conducted as part of the infectious agent surveillance activities in Japan, we compared the sensitivities of mouse, RD-18S, and Vero cells for CA.

CA isolates were obtained from patients with suspected enterovirus infection in Kyoto (population, 1.46 million) between 1996 and 2009. A total of 8,993 clinical samples were

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Table 1. Isolation of coxsackievirus group A using mouse, RD-18S, and Vero cells from 8,993 clinical samples

Species	Serotype	No. of isolates[1)]				Isolation rate (%)			
		Mouse	RD-18S	Vero	Total	Mouse	RD-18S	Vero	Total
Human enterovirus A	CA2	37	26	0	39	0.41	0.29	0	0.43
	CA3	13	14	1	15	0.14	0.16	0.01	0.17
	CA5	17	5	0	17	0.19	0.06	0	0.19
	CA8	11	2	1	12	0.12	0.02	0.01	0.13
	CA10	60	37	1	63	0.67	0.41	0.01	0.70
	CA12	13	0	0	13	0.14	0	0	0.14
	CA16	19	0	3	19	0.21	0	0.03	0.21
Human enterovirus B	CA9	0	24	5	25	0	0.27	0.06	0.28
Tentative	CA4	89	46	0	89	0.99	0.51	0	0.99
	CA6	54	33	0	55	0.60	0.37	0	0.61
Total		313	187	11	347	3.48	2.10	0.12	3.86

[1)]: Multiple isolates from the same patient were counted as one if they were identified as having the same CA serotype.

collected from 7,724 patients. Virus isolation was conducted using infant mice and the above-mentioned cell lines to give a total of 347 CA strains. Multiple isolates from the same patient were counted as one if they were subsequently identified as having the same CA serotype. CA isolates were identified as type 2, 3, 4, 5, 6, 8, 9, 10, 12, and 16 using the neutralization and/or CF test (2) (Table 1).

A total of 313 (90.2%), 187 (53.9%), and 11 (3.2%) of these 347 CA isolates were isolated using mice, RD-18S, and Vero cells, respectively, thus showing that infant mice are most susceptible, except for types 3 and 9. Only CA9 of the CA types isolated belongs to human enterovirus B. A total of 11 CA strains isolated in Vero cells were identified as CA3, 8, 9, 10, and 16.

The rates of virus isolation by mouse and RD-18S were compared, and mice were found to be more susceptible than RD-18S for CA2, 4, 5, 6, 8, 10, 12, and 16. In contrast, RD-18S is more susceptible than mice for isolating CA3 and 9. In summary, the use of mice is critical for conducting sensitive infectious agent surveillance for CA.

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Conflict of interest None to declare.

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