

INFLUENCE OF ACTINOMYCIN D ON MULTIPLICATION OF LEDNICE VIRUS

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The multiplication of Lednice virus, strain 6118 (1, 2), in the presence of actinomycin D (AD) was followed to obtain indirect evidence of the type of nucleic acid of the virus. The method using AD is based on the fact that AD blocks the synthesis of DNA-dependent RNA (3), which causes inhibition of DNA viruses (4). The multiplication of model RNA (Ťahyňa) and DNA (pseudorabies) viruses was assayed in parallel.

The experiment was done on chick embryo cells (CEC). Cell monolayers in Miller's bottles were inoculated with 1 ml of diluted viral suspension. After 1 hr at 37 °C the inoculum was removed, the monolayer washed 3 times in phosphate buffered saline (PBS) and further incubated at 37 °C with medium containing AD (Calbiochem) in concentrations of 2, 0.2 and 0.05 µg/ml respectively. Virus multiplication was checked 12, 18, 24 and 48 hr after inoculation by intracerebral titration in suckling mice.

The concentration of 2 µg/ml of AD in maintenance medium was highly cytopathic and therefore unsuitable for further assay. The concentration of 0.2 µg/ml of AD was suitable for use during 24 hr p.i., when the cell damage was minimal, as was the case with 0.05 µg/ml AD during 48 hr p.i. The effect of AD on virus multiplication as follows (ND = not done; — = no virus detected):

Virus	MOI	AD µg/ml	Virus titre at hr p. i. (log)			
			12	18	24	48
6118	0.001	0.2	<0.5	0.8	1.4	<0.5
		0.05	ND	0.7	2.0	3.3
		0 (control)	<0.5	0.9	2.1	3.8
Ťahyňa	0.1	0.2	2.4	4.0	5.8	3.6
		0	2.5	3.6	6.3	6.4
		0.2	—	—	—	—
Pseudorabies	0.1	0.2	—	—	—	—
		0	1.5	3.2	6.5	6.6

It is evident that, in spite of the low titre increases at the first intervals p.i. due to low multiplicity of infection (MOI), the Lednice virus multiplied in the presence of AD. The use of a low MOI was necessary with regard to high autointerference of Lednice virus in CEC (5). A comparison of the multiplication of Lednice virus with that of model viruses showed a similarity between Lednice and Ťahyňa viruses on the one hand and a clear difference between Lednice and pseudorabies viruses on the other. Based on these results, Lednice virus can be assumed to be an RNA virus.

References

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