

ISOLATION OF INFLUENZA VIRUS IN HUMAN EMBRYO
LUNG DIPLOID CELL CULTURES

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In the course of an epidemic due to influenza virus types A(H3N2) and B during January — April 1980 in the North-Bohemian Region (Czechoslovakia), most of the 80 influenza virus isolates were gained by cultivation in embryonated eggs or in secondary monkey (MK) cell cultures, but a part of them in human embryonic lung diploid cell (HEL; commercially available from SEVAC, Prague) cultures, about which few experiences are available.

The growth medium was removed from the HEL cell monolayer tube cultures and replaced by maintenance medium (synthetic medium SEVAC, Prague, with 2 % calf serum); then the examined nasopharyngeal swab material was inoculated, the tubes tightly stoppered and without further medium exchange kept stationary at 36 °C for 3—4 weeks. The cultures were repeatedly examined for adenoviral, enteroviral, or herpesviral cytopathic effect (CPE). In some cultures, during the 2nd—3rd week, a different finely granular CPE was observed; in further passages in HEL cell cultures it appeared only exceptionally and with a delay. This fine CPE was also produced by a number of samples, which yielded influenza viruses in simultaneously inoculated MK cell cultures or when influenza viruses were recovered from the nasal swab of the same patient on inoculation of embryonated eggs. Therefore, one of these positive nasopharyngeal swabs and 30 of the samples negative in isolation attempts in embryonated eggs and MK cells were inoculated into the amniotic cavities of embryonated eggs and 4-egg passages were performed. Ten of these 30 samples, yielded haemagglutinating activity (titres 32—265) in the 2nd—4th passage; the adapted virus strains were identified as influenza viruses A(H3N2), antigenically related to A/Bangkok/79, and B, related to B/Singapore 222/79.

The occasional finding of type B virus was probably due to the relatively high temperature of incubation of HEL cell cultures aimed at isolation of viruses other than those of influenza. The presence of influenza virus in HEL cell cultures was proved by immunofluorescence.

Cultivation on	Swab from	No. of isolates	Influenza virus			
			A(H3N2)	B	A(H3N2)	+B
Embryonated eggs	nose	42	31	10		1
MK cells	nasopharynx	27	15	12		0
HEL cells	nasopharynx	10	9	1		0
Embryonated eggs	nose	1	1	0		0
+	+					
HEL cells	nasopharynx	1	1	0		0

Recently several authors (1, 2, 3) studied the possibility of isolating influenza virus in systems more universal for respiratory and other viruses than are embryonated eggs; our report is a contribution to their studies.

The isolation of influenza virus in HEL cell cultures, of course, is more time-consuming than classical methods, but as an accessory finding at a complex investigation of infectious material it increased the isolation rate from otherwise negative samples.

References

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