## ANTI-LCMV IMMUNE SERUM PREPARATION AND ITS TESTING BY COMPLEMENT FIXATION TEST AND IMMUNOELECTROOSMOPHORESIS

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There have been reports (1) of considerable difficulties in the production of an anti-LCMV hyperimmune serum designed to have a high CF antibody titre. In preparing an anti-LCMV immune serum by immunization mice were allotted to 4 groups each immunized with a certain antigen dilution (ranging from  $10^{-1}$  to  $10^{-4}$ ). As an antigen, a suspension of guinea pig brain tissue infected with WE-123 strain of LCMV (Colindale) and complete Freund adjuvant were used. The suspension was centrifuged for 20 min at 2 000 rev/min and the supernatant was inactivated by means of 0.2% formalin, and kept for 3 days at 4 °C. The antigen was administered on 4 occasions according to the following immunization schedule:

The first injection (I ml dosis) was administered by subcutaneous (s.c.) route; it contained a given dilution of antigen and adjuvant in proportion 1:1. This was followed by second and third doses which were also given by s.c. route (at a 4-day interval apart) containing antigen and adjuvant in the same amount and identical proportion. The fourth injection, centaining 0.5 ml of not inactivated antigen, was administered by i. p. route 7 days later. Mice were bled 21, 25, 30, 35 and 40 days starting immunization. The sera were investigated by CF and immunoelectroosmophoresis (IEOP) (2). The findings are given in the Table.

Antigen dilution	21		25		30		35		40	
	CF	IEOP	CF	IEOP	CF	IEOP	CF	IEOP	CF	IEOF
10-1	_	_	1:4	_	_		_	_		_
$10^{-2}$	1:2	-	1:8	+	1:8	+	1:4	_	_	-
$10^{-3}$	1:8	+	1:16	+	1:64	+	1:16	+	-	_
$10^{-4}$		_	1:8	+	1:8	+	1:4	_	-	-

The results show that complement-fixing and precipitation antibodies first occured in mice which had received the antigen diluted 1:1000 with the highest complement-fixing antibody titre (1:64) attained in this group of mice.

## References

- E. H. Lenette, N. J. Schmidt (1979): Diagnostic Procedures for Viral, Rickettsial, and Chlamydial Infections, Fifth Ed., American Public Health Association, Washington DC 20005.
- 2. D. Čečuk, Z. Žerjav (1984): Countercurrent immunoelectrophoresis-IEOP as one of the possible methods for ROTA virus detection (in Croatian). Mikrobiologija, 21 (2), 97-101.