

## MECHANISM OF THERMAL INACTIVATION OF FREEZE-DRIED ECBO VIRUS SEROTYPE VG(5) 27

D.F. McCafferty, Department of Pharmacy, The Queen's University of Belfast,  
Belfast BT7 INN

The thermal inactivation mechanisms of liquid virus suspensions have been widely reported (Simpson and Hauser, 1968). However, the same is not true for freeze-dried viruses. Therefore, in the present study ECBO virus serotype VG(5)27 freeze-dried in SPG (Bovarnick, Miller and Snyder, 1950) was stored at a variety of temperatures (30 - 51°C) and the rate constants for virus degradation obtained for each temperature. Statistical analysis employing the method of least squares was used to obtain the best estimates for the rate constants and also to ensure that they were significantly different from one another. An Arrhenius plot was then drawn from which two inactivation energies ( $E_a$ ) were obtained. The entropy of activation change ( $\Delta S^*$ ) and the Gibb's function ( $\Delta G^*$ ) were calculated by use of appropriate equations (Fleming, 1971).

Table 1. Thermodynamic parameters for the inactivation of ECBO virus serotype VG(5)27 freeze-dried in SPG

Temperature (°C)	$E_a$ (kJ mol <sup>-1</sup> )	$\Delta G^*$ (kJ mol <sup>-1</sup> )	$\Delta S^*$ (J mol <sup>-1</sup> K <sup>-1</sup> )
30 to 45	82.2	79.4 to 77	21.4
45 to 51	344.4	77 to 74.2	845.6

Although these results (Table 1) were obtained for a freeze-dried virus, a similar trend has been reported for many liquid virus preparations (Dimmock, 1967). The thermal inactivation of viruses is frequently attributed to a degradation of the nucleic acid at low temperatures and to a breakdown of structure at high temperatures. In this investigation, the difference between these two reactions is demonstrated by the four-fold increase in  $E_a$  at high temperature inactivation (45 - 51°C) as well as the 50 fold rise in  $\Delta S^*$  (Table 1). This latter phenomenon is due to the much greater change in the conformation of virus structure at high temperature inactivation. The similarity between the thermodynamic parameters for low temperature inactivation (30 - 45°C) in this study, and for the breakage of phosphodiester bonds, suggests the involvement of RNA at these temperatures (Eigner, Boedtker and Michaels, 1961). Furthermore, the activation energy for high temperature VG(5)27 virus degradation (Table 1) corresponds to those for the denaturation of many proteins (Woese, 1960).

Bovarnick, M.R., Miller, J.C. and Snyder, J.C. (1950). *J. Bact.*, 59, 509-522.

Dimmock, N.J. (1967). *Virology*, 31, 338-353.

Eigner, J., Boedtker, H. and Michaels, G. (1961). *Biochem. biophys. Acta*, 51, 165-168.

Fleming, P. (1971). *J. gen. Virol.*, 13, 385-391.

Simpson, R.W. and Hauser, R.E. (1968). *Virology*, 34, 361-367.

Woese, C. (1960). *Ann. N. Y. Acad. Sci.*, 83, 741-751.