methylcytosine, each value being the mean of six estimations.

It appears, then, that the deoxyribonucleic acid prepared by the above methods from *T2r* bacteriophage contains two fractions, one containing approximately 30 per cent of the total deoxyribonucleic acid phosphorus, with the ratio adenine + thymine/guanine + 5-hydroxymethylcytosine approximately equal to 1·9, and the remaining 70 per cent with that ratio approximately equal to 2·15. As it is unlikely that these distinct fractions would be produced by degradation of a larger single molecule by the preparative methods used, this means that the *T2r* bacteriophage particle contains at least two different species of deoxyribonucleic acid molecule which differ in their ease of dissociation from basic protein and their content of purine and pyrimidine bases.

Geoffrey L. Brown
Angela V. Martin

Medical Research Council
Biophysics Research Unit,
Wheatstone Physics Laboratory,
King's College, London, W.C.2.
Sept. 6.

[1] Hershey, A. D., and Chase, M., *J. Gen. Physiol.*, **36**, 39 (1952). Hershey, A. D., Cold Spring Harbor Symp. Quant. Biol., **18**, 135 (1953). Hershey, A. D., Dixon, J., and Chase, M., *J. Gen. Physiol.*, **36**, 777 (1953). Hershey, A. D., *Virology*, **1**, 108 (1955).
[2] Brown, G. L., and Watson, M., *Nature*, **172**, 339 (1953). Chargaff, E., Crampton, C. F., and Lipshitz, R., *Nature*, **172**, 289 (1953). Elson, D., Gustafson, T., and Chargaff, E., *J. Biol. Chem.*, **209**, 285 (1955).
[3] Herriott, R. M., and Barlow, J. L., *J. Gen. Physiol.*, **36**, 17 (1952)
[4] Crampton, C. F., Lipshitz, R., and Chargaff, E., *J. Biol. Chem.*, **206**, 499 (1954).
[5] Hoffpauir, C. L., and Guthrie, J. D., *Textile Res. J.*, **20**, 617 (1950).
[6] Campbell, D. H., Leuscher, E., and Lerman, L. S., *Proc. U.S. Nat. Acad. Sci.*, **37**, 575 (1951).
[7] Anderson, T. F., *Ann. Inst. Pasteur*, **84**, 5 (1953).
[8] Wyatt, G. R., and Cohen, S. W., *Biochem. J.*, **55**, 774 (1953).

Crystalline Inhibitors of Trypsin from Potato

Recently, Sohonie and Ambe[1] have reported the preparation of crystalline inhibitors of trypsin from field-bean and double-bean. Unlike these inhibitors, which are comparatively stable to heat treatment, the acid extracts of potato (*Solanum tuberosum*) have been found to contain a thermolabile inhibitor[2]. We have now examined the possibility of preparing this inhibitor in quantity in a crystalline form.

A method similar to that described for the crystallization of field-bean and double-bean inhibitors[1] was successfully employed, with the difference that all the operations were carried out in the cold to minimize any loss of activity in the course of preparation. The crude inhibitor, precipitated by ammonium sulphate, yielded two fractions. One was extracted by 2·5 per cent trichloroacetic acid, while the other, insoluble in trichloroacetic acid, was removed from the residue, in 1 per cent saline. Further purification, lyophilization and crystallization were carried out essentially as described in the previous communication.

Both fractions are dirty white in colour, soluble in water and twice as potent as the inhibitors from field-bean and double-bean. These were also found to have a powerful anticoagulant action on citrated human plasma.

Heat treatment resulted in 80–90 per cent destruction of the activity. Both components gave characteristic protein reactions and were found to differ widely in lysine, histidine and methionine contents.

A detailed report of the study of the different physico-chemical properties, etc., of the potato inhibitor fractions will be published in the *Journal of Scientific and Industrial Research* (India). We are grateful to the Council of Scientific and Industrial Research (India) for financing this work and for a personal grant to one of us (K. S. A.).

Kamala Sohonie
K. S. Ambe

Department of Biochemistry,
Institute of Science,
Bombay 1. July 7.

[1] Sohonie, K., and Ambe, K. S., *Nature*, **175**, 508 (1955).
[2] Sohonie, K., and Bhandarkar, A. P., *J. Sci. Indust. Res.*, **13**, 500 (1954).
[3] Kazal, L. A., Spicer, D. S., and Brahinsky, R. A., *J. Amer. Chem. Soc.*, **70**, 3034 (1948).

Interference between Variants of Pseudorabies Virus demonstrable in Tissue Culture

The results of a series of investigations dealing with the cytopathogenic activity of pseudorabies virus have recently been reported from this Institute[1]. In these studies a strain of pseudorabies virus was serially transferred in tissue suspensions derived from chick embryos, and these served as a source of virus for the experiments. Destructive effects of the virus on tissue cultures made from fragments of embryonic chicken heart were recorded, and the cytopathogenic titre of virus material was used as a measure of its activity. Our present experiments have demonstrated that the virus became modified in its cytopathogenic activity for chicken cells by passages carried out in tissue suspensions of mouse embryo. The change in cytopathogenicity set in after four or five serial transfers of the virus in this tissue suspension. The characters of virus materials serially passed in suspensions of chicken and mouse embryonic tissue respectively are given in Table 1.

As is seen, the most striking feature of mouse (*MEC*) virus is its low cytopathogenic effect on chicken cells. It should be noted that the change in properties of virus cultivated in mouse tissue was not permanent. When *MEC* virus was serially transferred in embryonic chicken tissue, its destructive effect for chicken cells gradually increased, and it had regained its original properties within three to five passages.

The low cytopathogenic effect of *MEC* virus on chicken explants suggests a potential ability of this

Table 1. Characters of Pseudorabies Virus Propagated in Tissue Suspension either of Chick Embryo (*CEC* Virus) or Mouse Embryo (*MEC*) Virus

Mode of titration of virus	Virus material			
	CEC		*MEC*	
	No. of materials tested	Geom. mean of titres	No. of materials tested	Geom. mean of titres
Cytopathogenic effect of virus on chicken cells	45	$10^{-6\cdot5}$	46	$10^{-1\cdot15}$
Cytopathogenic effect of virus on mouse cells	4	$10^{-3\cdot5}$	11	$10^{-5\cdot8}$
In young chickens intracerebrally	1	$10^{-5\cdot3}$ (*LD*50)	1	10^{-4} (*LD*50)
In mice intracerebrally	20	$10^{-4\cdot2}$ (*LD*50)	34	$10^{-3\cdot8}$ (*LD*50)