

agglutination reactions of red cells have stressed the role of sialic acid. An increase in the serum level of similar substances might therefore alter the sedimentation rate not only by affecting the physical properties of the serum itself but by actual combination with like structures on the erythrocyte membrane.

Riassunto. Gli autori riscontrano nel siero di pazienti leucemici un aumento della reazione con difenilamina rispetto al siero di soggetti normali. Questo aumento è discusso alla luce di precedenti risultati che collegano questa reazione con i mucosaccaridi del siero.

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¹⁰ We thank the British Empire Cancer Campaign for an award to one of us (D.L.), and are grateful to Professor C. RIMINGTON for helpful comments on the manuscript. The *N*-acetyl-neuraminic acid used for the calibration curve was very kindly supplied by Miss CARROLL of The Medical Research Council, Mill Hill.

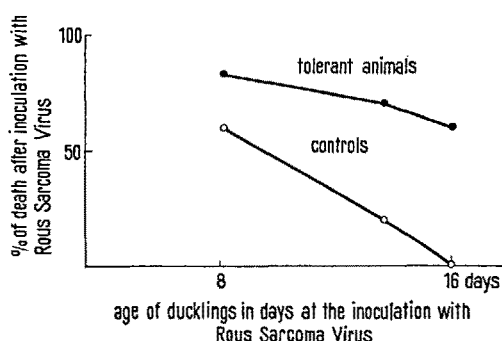
Immunological Tolerance to Rous Sarcoma Virus in Ducks

We have succeeded¹ in eliciting immunological tolerance to Rous sarcoma virus in ducks, just as it was produced in turkeys^{2,3}. Intraembryonic or better repeated post-embryonic injections of chicken blood or lyophilized blood were used for the induction of tolerance. A comparison of susceptibility to Rous virus in control and tolerant ducks of different ages is shown in the Figure. The distribution of both curves suggests that although the differences in susceptibility of the two groups are apparent on the 8th day, 16 day-old control birds are resistant, whereas tolerant ducks succumb in more than 50% to inoculation with cell-free filtrates from Rous sarcoma. (Extracts were prepared by homogenization of 20% suspension of chicken Rous sarcoma tissue (stored in frozen state) in isotonic potassium citrate at pH 7, by centrifugation for 20 min at 8000 *g* and by filtration of supernatant through procelain candles 'Selas 02'.) The state of tolerance depends on repeated administration of antigen during the adaptive period. A single injection of 0.3 ml of chicken blood on the first day after hatching is capable of eliciting a weak degree of tolerance (10%), whereas the same dose divided into 3 injections administered on the first, third, and fifth day induces a clear-cut tolerance (30%). The same dependance was found to exist with the dose of 0.9 ml. The postnatal period during which it is possible to induce tolerance in ducks is relatively long—injections of chicken blood commenced on the 14th day after hatching are still fully effective whereas those commenced later are ineffective⁴. Nuclear nucleoprotein appeared to be a highly effective chicken antigenic material for induction of tolerance to Rous virus, whereas the deoxyribonucleic acid itself was ineffective⁵.

The original explanation of tolerance to Rous virus, indicating that this is the tolerance displayed directly to the Rous virus particle, which therefore contains normal chicken antigens, is in conflict with more recent information concerning the nature of Rous virus. This is firstly, because it has been found that the Rous virus does not contain normal chicken antigens as an integral part of its functional surface⁶, that it penetrates into the cell⁷ much earlier than actively acquired immunity could play a role and that the presence itself of virus-neutralizing antibodies does not profoundly affect the tumour growth⁸.

Secondly, our results given in the Table also show that virus-neutralizing capacity of sera from tolerant animals bearing large tumours does not differ from sera of resistant control animals, although in case of direct tolerance to the virus particle it must have been lower. Similar results obtained PRINCE³ in tolerant turkeys.

All the results obtained with tolerance to Rous virus in ducks, as well as those obtained in turkeys^{3,9} are in agree-



● ○ each point represents percentage of death in a group of 8 to 10 animals.

Natural susceptibility and actively acquired tolerance to Rous sarcoma Virus in ducklings of different ages.

Tests for virus-neutralizing capacity of sera from tolerant and control ducklings (tests were made on 10 day-old Leghorn chicks)

ID 50% after incubation of sera from tolerant animals with Rous virus	ID 50% after incubation of sera from control animals with Rous virus
10 ^{-2,66}	10 ^{-2,5}
10 ^{-2,6}	10 ^{-2,33}
10 ^{-2,5}	10 ^{-2,16}
10 ^{-2,7}	10 ^{-2,54}
10 ^{-2,33}	10 ^{-3,0}
10 ^{-3,16}	10 ^{-3,23}

¹ J. SVOBODA, *Folia biol. (Praha)* 4, 205 (1958).

² M. SIMONSEN, *Nature* 175, 763 (1955). – R. J. C. HARRIS, *Proc. Roy. Soc., B*, 146, 59 (1956). – J. SVOBODA and M. HAŠEK, *Folia biol. (Praha)* 2, 256 (1956). – R. J. C. HARRIS, *J. chron. Dis.* 3, 58 (1958). – R. J. C. HARRIS and P. J. SIMONS, *Nature* 181, 1485 (1958).

³ A. M. PRINCE, Personal communication (1960).

⁴ J. SVOBODA, *Folia biol. (Praha)* 6, 21 (1960).

⁵ J. SVOBODA, *Folia biol. (Praha)* 6, 96 (1960).

⁶ H. RUBIN, *Virology* 2, 545 (1956). – T. BORSOS, *J. Nat. Cancer Inst.* 20, 1215 (1958). – M. RYCHLÍKOVÁ and J. SVOBODA, *Virology* 10, 545 (1960).

⁷ H. TEMIN and H. RUBIN, *Virology* 6, 669 (1958).

⁸ P. VIGIER, *Bull. du Cancer* 45, 460 (1958).

⁹ J. HORT, J. SVOBODA, and M. HAŠEK, *Folia biol. (Praha)* 6, 371 (1960).

ment with DULBECCO's suggestion¹⁰ that the Rous virus – simultaneously with the malignant transformation of foreign cell – is capable of inducing the formation of some chicken antigenic complex in this cell by a process which might be analogous to the transduction or better conversion in phages¹¹. In this connection, the finding of GUSYEV¹² is of great importance, namely that the Rous virus induces the formation of specific non-virus antigen, even in chicken cells, and that it produces the morphological conversion of chicken fibroblasts¹³.

These foreign tumour cells containing some chicken antigens may exhibit progressive growth only in hosts which are immunologically not fully mature or are tolerant to chicken antigens, because, in normal animals which are immunologically mature, they are destroyed like all cells of antigenically foreign tissues by immunological reaction.

The present interpretation is in agreement with our previous findings¹ that in control ducks, after inoculation with Rous virus, palpable tumours occur which regress in contrast to those in tolerant animals. The difference between the two groups is not manifest until in the reaction to virus-induced tumour cells.

PRO EXPERIMENTIS

Benützung der oszillographischen Polarographie in der Mikrobiologie

Im folgenden sind einige Möglichkeiten der analytischen Ausnützung der oszillographischen Polarographie mit Wechselstrom¹ für den Nachweis der Utilisation und des Metabolismus einiger Aminosäuren und der Bestandteile der Nucleinsäuren bei *Escherichia coli* entwickelt. Es wurde das «Polaroskop P-524» (Fa. Křížlk, Prag) mit Quecksilber-Tropfenelektrode benützt, welches die zeitliche Änderung der Spannung $dE/dt = f_1(E)$ demonstriert (Fig. 1).

Mit Hilfe dieser Apparatur wurde der Einfluss gewachsener bakterieller Zellsuspensionen und auch wachsender Kulturen von *E. coli* auf einige Aminosäuren und Bestandteile der Nucleinsäuren in vollsynthetischen flüssigen Nährböden erforscht (Tab.). Die analysierten Substrate wurden in gepufferten (Michaelis m/15 Phosphatpuffer) physiologischen Lösungen verschiedener pH-werte gelöst und mit der *E. coli*-Kultur beimpft oder aber mit 1% der gewaschenen Zellen von *E. coli* gemischt. Nach 1–24 h Inkubation bei 37°C wurden die bakteriellen Kulturen und Zellsuspensionen in verschiedenen Grundelektrolyten oszillographisch analysiert^{2,3} (Tab.).

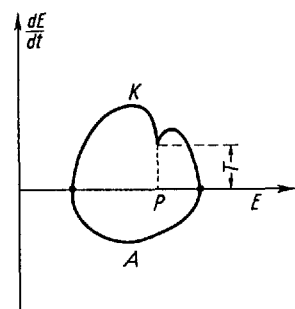


Fig. 1. Oszillogramm $dE/dt = f_1(E)$. Die Qualität der analysierten Stoffe ist durch das Potential des Einschnittes (P) charakterisiert (das gewissermaßen mit dem polarographischen Halbstufenpotential analog ist) und die Quantität durch die Tiefe (T) des Einschnittes. (K) kathodischer Zweig, (A) anodischer Zweig.

Résumé. Tolérance immunologique au virus du sarcome de Rous chez les canards. Les résultats obtenus ont été interprétés comme suit: Le virus de Rous par l'action maligne sur les cellules d'autres espèces y provoque la formation d'un complexe antigénique de poulet et rend ces cellules capables d'une croissance progressive, chez les animaux qui tolèrent les antigènes de poulet.

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¹⁰ R. DULBECCO, in *Symposium on Latency and Masking in Viral and Rickettsial Infections* (Ed. D. L. WALKER, R. P. HANSON, A. S. EVANS, published by Burgess Publishing Co., Minneapolis), p. 46.

¹¹ H. UETAKE, S. E. LURIA, and J. W. BURROUS, *Virology* 5, 68 (1958).

¹² A. I. GUSYEV, *Byull. Eksptl. Biol. Med.* 47, 70 (1960).

¹³ H. TEMIN, *Virology* 10, 182 (1960).

Aktivitätsbestimmungen der bakteriellen Asparaginase⁴, der Nucleodesaminasen^{5,6} sowie der Phosphorylasen der Pyrimidinnucleosiden^{7,8} sind vorgenommen worden. Es ergaben sich zahlreiche Möglichkeiten für die praktische Ausnützung der Oszillopolarographie in der Mikrobiologie: a) vor allem die Geschwindigkeit der Manipulation; b) relative experimentelle Einfachheit der Oszillographie; c) gleichzeitige Orientierung über Ernährungsprozesse und Metabolismus der Mikroorganismen; d) Applikation auf dem Gebiete der mikrobiologischen Enzymo-

Substrat	Grundelektrolyt (1 Mol)		
	NaOH	H ₂ SO ₄	HCOONH ₄ /HCOOH
Asparagin	+		
Glutaminsäure	+		
Adenin		+	
Guanin			+
Adenosin		+	
Desoxyadenosin		+	
Guanosin			+
Adenylsäure		+	
Desoxyadenylsäure		+	
Guanylsäure			+
Desoxyguanylsäure			+
Cytidin		+	
Cytidylsäure		+	
Thymin	+		
Uridin	+		

¹ J. HEYROVSKÝ und R. KALVODA, *Oszillographische Polarographie mit Wechselstrom* (Akad. Verlag, Berlin 1960).

² D. KALÁB, *Pharmazie* 10, 81 (1955).

³ E. PALEČEK, *Naturwiss.* 45, 186 (1958).

⁴ D. KALÁB, (Vortrag) I. Tschechoslowakisches oszillopolarographisches Symposium in Smolentza, April 1960.

⁵ C. LUTWAK-MANN, *Biochem. J.* 30, 1495 (1936).

⁶ R. J. MANS und A. L. KOCH, *J. biol. Chem.* 235, 450 (1960).

⁷ L. M. PAEGE und F. SCHLENK, *Arch. Biochem.* 28, 348 (1950).

⁸ L. M. PAEGE und F. SCHLENK, *Arch. Biochem. Biophys.* 40, 42 (1952).