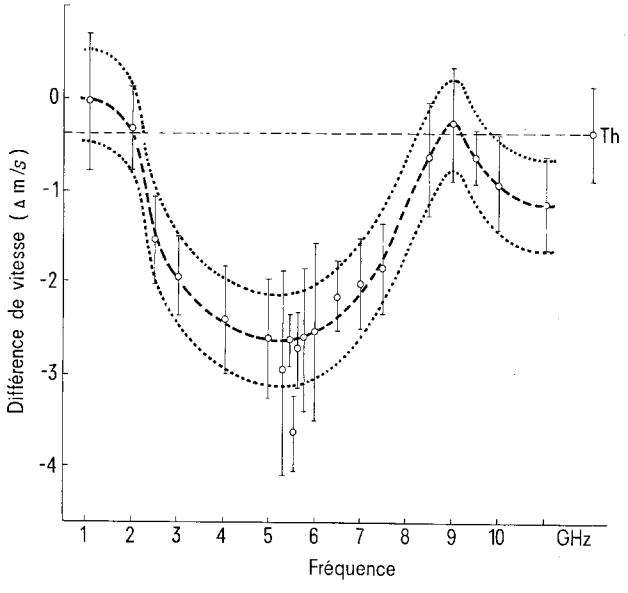
méthode nous a conduit à rechercher les fréquences qui ont un effet sur la vitesse de conduction, la rhéobase et la chronaxie du nerf.

Techniques expérimentales. Dès le prélèvement, le nerf est placé dans un tube à hémolyse contenant de l'huile de paraffine. Ce milieu assure convenablement sa conservation, permet de bonnes conditions d'irradiation en raison de sa faible absorption des micro-ondes, et enfin dissipe une partie des effets thermiques.

Les stimulations sont des chocs rectangulaires dont le temps de montée est de 5 microsec. La stimulation et le potentiel d'action sont visualisés sur oscilloscope à entrées différentielles, de sensibilité de 10 micro-volts/cm. Les irradiations sont effectuées dans des cavités en laiton, le flux d'hyperfréquences étant fourni par un générateur associé à un amplificateur à TOP. Nous procédons selon l'ordre chronologique suivant: prélèvement du nerf, repos de 30 min à 20 ± 2 °C, mesure des divers paramètres, repos de 10 min, irradiation de 5 min, repos de 10 min, nouvelle mesure des mêmes paramètres. Chaque série de mesures est réalisée sur 12 préparations nerveuses (ce nombre assurant une distribution statistique convenable). Tous nos résultats sont programmés et les tests statistiques de «Fischer-Student» χ^2 sont calculés par ordinateur.

Essais préliminaires. Ils nous permettent d'apprécier les variations des paramètres dans le temps et en fonction de la température. Lorsque le nerf n'est soumis à aucun traitement, les divers paramètres sont stables de 30 à 90 min après le prélèvement. L'élévation de température du nerf, mesurée au thermocouple en plusieurs points, était de 2° à 3 °C à la fin d'une irradiation, nous avons recherché l'influence éventuelle de cet échauffement sur ces paramètres en augmentant la température de l'ensemble de la préparation nerveuse de 4° à 5 °C au bain-marie; après le repos habituel de 10 min, nous n'avons constaté aucune variation des paramètres mesurés. Ces témoins (Th dans la figure) nous permettent d'éliminer le facteur thermique des micro-ondes.



Variation moyenne de la vitesse de conduction dans le nerf sciatique de grenouille sous l'effet des micro-ondes. Abscisse: fréquence en GHz. Ordonnée: différence (△) de vitesse (en *m/s*) avant et après irradiation. Th: essais thermiques. Le pointillé(·····) montre la corrélation effet/fréquence.

Résultats. Vitesse de conduction: La stimulation avait une intensité de 120 mV et une durée de 0,4 msec. La figure nous montre que la vitesse de conduction décroît d'une façon significative sous l'effet d'irradiation à certaines fréquences: c'est dans la zone de 5 à 6 GHz que la diminution est la plus marquée: 2,5 à 3 *m/s*, soit un écart de 8 à 10% par rapport à la moyenne avant irradiation. On note également une étroite corrélation effet/fréquence pour les longueurs d'ondes comprises entre 2,5 et 9 GHz. La chronaxie et la rhéobase ne sont que faiblement influencées par l'irradiation.

Discussion. D'après nos résultats les micro-ondes ont un effet sur la propagation du potentiel d'action dans le nerf sciatique isolé de la grenouille. Cependant il faut bien remarquer que cette action ne se manifeste que pour certaines longueurs d'onde. Ceci nous amène à penser qu'une étude des effets biologiques des micro-ondes réalisée à une seule fréquence ne peut pas permettre de conclure valablement et qu'il convient donc d'effectuer des recherches systématiques avec toute une gamme de fréquences. Une telle différence de réponses permet en outre, de parler d'effet spécifique des micro-ondes.

Summary. With the study of the reaction of batracian peripheral nervous system subject to 1 to 10 GHz irradiations, the authors show that the systematic frequency study is the more appropriate method for determining the microwave specific effects.

A. Deficis[4], A. Delfour[4], J.C. Dumas[5] et S. Laurens[6]

DERMO-CERT, 2 avenue Edouard-Belin, Toulouse-Lespinet (France); et Laboratoire de Physiologie, U.E.R. des Sciences Pharmaceutiques, 31, allées Jules-Guesde, F-31400 Toulouse (France), 21 février 1974.

[4] Ingénieurs au DERMO-CERT.
[5] Maître de Conférences de Physiologie à l'U.E.R. des Sciences Pharmaceutiques, Toulouse.
[6] Maître-Assistant de Physiologie à l'U.E.R. des Sciences Pharmaceutiques, Toulouse.

Effect of Cortisol on Phosphorylation of Acidic Proteins in Liver Nuclei from Adrenalectomized Rats

Acidic chromosomal proteins are thought to be involved in the control of genetic expression[1]. These proteins are actively phosphorylated[2–6] and their rates of phosphorylation are increased during times of gene activation [7–9]. Hormones have been observed to increase phosphorylation of acidic nuclear proteins and have been suggested to influence genetic activity via this increased phosphorylation[10–13]. We have studied in the rat liver how early after the administration of cortisol increased phosphorylation of acidic proteins does occur, and whether it is selective or generalized with respect to the various acidic nuclear proteins.

Effect of cortisol on incorporation of ^{32}P into phenol-soluble acidic proteins and histones from rat liver

Before sacrifice (h)	Cortisol	Alkalilabile ^{32}P (cpm/mg protein $\times 10^2$)			
		Acidic proteins		Histones	
1	—	254 ± 15		80 ± 29	
	+	267 ± 7	NS	75 ± 8	NS
5	—	247 ± 21		109 ± 26	
	+	236 ± 32	NS	117 ± 31	NS
7	—	237 ± 15		102 ± 16	
	+	302 ± 6	$P < 0.01$	160 ± 15	$P < 0.01$
9	—	348 ± 6		100 ± 14	
	+	445 ± 7	$P < 0.01$	152 ± 24	$P < 0.01$
11	—	369 ± 9		102 ± 9	
	+	477 ± 50	$P < 0.01$	171 ± 26	$P < 0.01$

+, Each of these figures is the average of 6 assays, performed in 2 experiments, and consisted of 3 rats, respectively. In each experiment all liver tissue was put in 1 pool. Assays on each pool were conducted in triplicate. The SD and p-values (calculated by the t-test) are shown.

Materials and methods. Most materials used are listed in a previous report[13]. Hydrocortisone sodium succinate was purchased from the Upjohn Company, Kalamazoo, Michigan.

The rats were all male, Sprague Dawley, weighing 120–160 g, adrenalectomized 7 days before the experiments, maintained at 78 °C, fed Purina chow and had free access to water containing 0.9% NaCl solution. After a 10-hour fast, 7.5 mg hydrocortisone sodium succinate/100 g body weight was injected i.p. 1, 5, 7, 9 and 11 h before sacrifice. Controls received an equivalent volume of 0.9% NaCl solution. ^{32}P-labelled *ortho*phosphate (2 mCi/150 g body wt.) was injected i.v. 60 min before sacrifice. After cervical section, the livers were removed to isolate liver cell nuclei according to the method of McGREGOR and MAHLER[14]. Phenol-soluble acidic proteins from isolated nuclei were prepared, and polyacrylamide gel electrophoresis was performed in the presence of sodium dodecyl sulfate, according to TENG et al.[2]. Each 0.2 ml sample had 90 µg protein, 0.1 *M* sodium phosphate buffer, 0.2 mg sodium dodecyl sulfate, 200 mg succrose, and 10 µg bromphenol blue, at pH 7.4. Electrophoresis conditions: 16 h at 4 mA/column (10×0.6 cm). The gels were stained with Amido black 10 B; 1 mm transverse slices of the gels were counted[2]. Histones were obtained as described by AHMED and ISHIDA[12]. Alkalilabile ^{32}P radioactivity was measured according to MEISLER and LANGAN[15], protein by the LOWRY method[16].

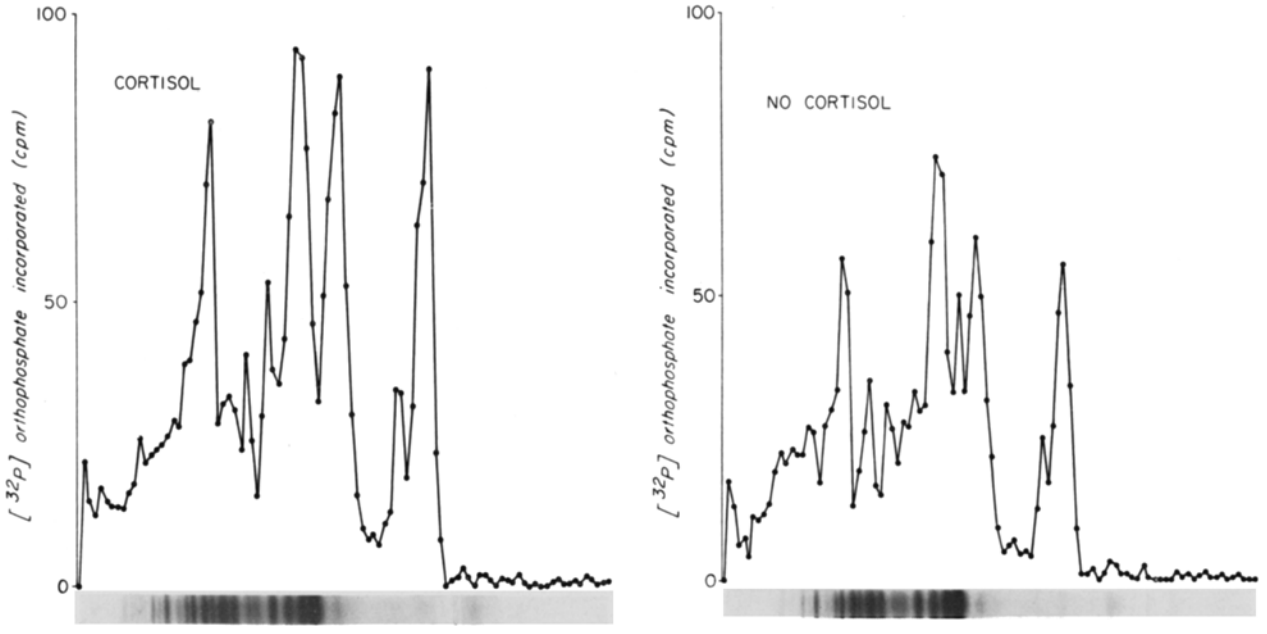
Results. Cortisol did stimulate, in livers from adrenalectomized rats, the phosphorylation of phenol-soluble nuclear acidic proteins (Table). This stimulation did occur with a lag period of more than 5 h. It was not restricted to acidic proteins since phosphorylation of histones was stimulated as well.

Phenol-soluble acidic proteins from liver of adrenalectomized rats had identical electrophoretic profiles (Figure), regardless of whether cortisol was administered or not. The cortisol-induced enhanced phosphorylation was generalized and not specific with respect to the various acidic nuclear proteins.

Discussion. These data suggest that there is a generalized stimulation in phosphorylation of acidic nuclear proteins from rat liver after the administration of cortisol. This stimulation does occur within 7 h after treatment, when cortisol-mediated enzyme induction[17–19] and liver growth[20] are apparent. Increased phosphorylation might therefore be related to both of these processes.

In view of the reports on increased incorporation of amino acids into nuclear acidic proteins from rat liver after administration of cortisol[21,22], alterations in phosphorylation could represent altered synthesis of these proteins and/or specific increases in phosphorylation. As suggested for other systems of gene activation [23–25], enzymatic modification of existing acidic nuclear proteins

[1] R. BASERGA and G. STEIN, Fedn. Proc. *30*, 1752 (1971).
[2] C. S. TENG, C. T. TENG and V. G. ALLFREY, J. biol. Chem. *246*, 3597 (1971).
[3] L. J. KLEINSMITH, V. G. ALLFREY and A. E. MIRSKY, Proc. natn. Acad. Sci., USA *55*, 1182 (1966).
[4] L. J. KLEINSMITH and V. G. ALLFREY, Biochim. biophys. Acta *175*, 123 (1969).
[5] L. J. KLEINSMITH and V. G. ALLFREY, Biochim. biophys. Acta *175*, 136 (1969).
[6] T. A. LANGAN, in *Regulation of Nucleic Acid and Protein Biosynthesis* (Eds. V. V. KONINGSBERGER and L. BOSCH; Elsevier, Amsterdam 1967), p. 233.
[7] L. J. KLEINSMITH, V. G. ALLFREY and A. E. MIRSKY, Science *154*, 780 (1966).
[8] J. ILAN and J. ILAN, Biol. Bull. *139*, 425 (1970).
[9] K. AHMED, Biochim. biophys. Acta *243*, 38 (1971).
[10] R. W. TURKINGTON and M. RIDDLE, J. biol. Chem. *244*, 6040 (1969).
[11] K. AHMED and H. ISHIDA, Molec. Pharmac. *7*, 323 (1971).
[12] R. A. JUNGMANN and J. S. SCHWEPPE, J. biol. Chem. *247*, 5535 (1972).
[13] P. SCHAUDER, B. J. STARMAN and R. H. WILLIAMS, Proc. Soc. exp. Biol. Med. *145*, 331 (1974).
[14] R. R. MCGREGOR and H. R. MAHLER, Arch. Biochem. Biophys. *120*, 136 (1967).
[15] M. H. MEISLER and T. A. LANGAN, J. biol. Chem. *244*, 4961 (1969).
[16] O. H. LOWRY, N. J. ROSEBROUGH, A. L. FARR and R. J. RANDALL, J. biol. Chem. *193*, 265 (1951).
[17] F. T. KENNEY, J. biol. Chem. *242*, 4367 (1967).
[18] A. GROSSMAN and C. MAVRIDES, J. biol. Chem. *242*, 1398 (1967).
[19] C. B. HAGER and F. T. KENNEY, J. biol. Chem. *243*, 3296 (1968).
[20] P. FEIGELSON, M. FEIGELSON and O. GREENGARD, Recent Progr. Horm. Res. *18*, 491 (1962).
[21] K. SHELTON and V. G. ALLFREY, Nature, Lond. *228*, 132 (1970).
[22] M. D. BUCK and P. SCHAUDER, Biochim. biophys. Acta *224*, 644 (1970).
[23] W. B. BENJAMIN and R. M. GOODMAN, Science *166*, 629 (1969).
[24] E. M. JOHNSON and V. C. ALLFREY, Arch. Biochem. Biophys. *152*, 786 (1972).
[25] J. BANKENSHIP and E. BRESNICK, Biochim. biophys. Acta *340*, 218 (1974).



Comparisons of the effect of cortisol, 11 h after administration, on the electrophoretic pattern of phenol-soluble nuclear acidic proteins from rat liver with amounts of ^{32}P incorporated in them. Results with cortisol on the left portion and without cortisol on the right portion. For each electrophoretic analysis radioactivity is plotted as a function of the distance of migration and is aligned with the banding pattern shown immediately below.

via phosphorylation is a likely mechanism by which cortisol might influence genetic control, thereby affecting enzyme induction and liver growth. However, the biological significance of increased phosphorylation of nuclear proteins during times of gene activation is still unclear.

Cortisol did influence, with a lag period of about 7 h, phosphorylation of histones and the bulk of acidic proteins, i.e. preferential phosphorylation of single acidic nuclear proteins did not occur. Therefore, the cortisol-mediated enzyme induction and/or liver growth does not seem to be initiated by the specific phosphorylation of a single acidic nuclear protein. Further investigations are needed for clarification of the role of phosphorylation of nuclear acidic proteins occurring during hormone-induced increase in genetic activity.

Zusammenfassung. Nach Gabe von Cortisol war in adrenalektomierten Ratten ein gesteigerter Einbau von ^{32}P-*Ortho*phosphat in saure Kernproteine und Histone der Leber nachweisbar. Diese gesteigerte Phosphorylierung erfolgte mit einer Verzögerung von mehr als 5 h post injectionem gleichförmig in den verschiedenen sauren Proteinen. Das elektrophoretische Muster (Polyacrylamid) saurer Proteine blieb durch Cortisolgabe unbeeinflusst.

P. Schauder[26], Barbra Starman and R. H. Williams

Department of Medicine, Division of Endocrinology, University of Washington, Seattle (Washington 98195, UAS), 27 May 1974.

[26] Present address for reprint requests: Division of Gastroenterology and Metabolism, Department of Medicine, University of Göttingen, D-34 Göttingen (German Federal Republic, BRD).

How Well does Man Thermoregulate During Sleep?

Baker and Hayward have suggested that in active (REM) sleep thermoregulatory and other autonomic control is inhibited[1]. Panting and shivering in cats are indeed suppressed during active sleep[2,3]. In men sleeping at 'comfortable' environmental temperatures, sweating measured over small areas of the hand or chest is depressed or absent during the active phase[4]. If the sweat depression is general over the entire body and if it persists in warm environments then it would seem that active sleep and precise thermoregulation are mutually exclusive phenomena in normal human function.

In order to investigate whether such a conflict exists we exposed a sleeping man to 3 warm environments in a human calorimeter. The calorimeter[5,6] provided accurate control of the thermal environment and allowed direct measurements of sensible (radiant plus convective) and evaporative heat transfer with a precision of better than

[1] M. A. Baker and J. N. Hayward, Science *157*, 1586 (1967).
[2] P. L. Parmeggiani and L. Sabattini, Electroenceph. clin. Neurophysiol. *33*, 1 (1972).
[3] P. L. Parmeggiani, C. Franzini, P. Lenzi and G. Zamboni, Brain Res., *52*, 189 (1973).
[4] T. Ogawa, T. Satoh and K. Takagi, Jap. J. Physiol. *17*, 135 (1967).
[5] J. Visser and T. Hodgson, S. Afr. mech. Engr. *9*, 243 (1960).
[6] J. W. Snellen and P. Baskind, Research report 24/69, Chamber of Mines of South Africa, Johannesburg (1969).