In Experiment 2, ϱ correlations were calculated between 1. rank on substrate level and rank on analgesia, 2. rank on substrate level and rank on brain morphine level and 3. rank on analgesia and rank on brain morphine level.

Results. Experiment 1. The ϱ correlations are shown in Table 1. In addition, average blood levels for each substrate are shown. These levels are similar to literature values[10]. As can be seen, blood levels of pyruvate and lactate correlated with analgesia, while no substrate correlated with pre-drug reaction time.

Experiment 2. Table II shows that pyruvate and lactate, as well as citrate levels correlated with analgesia. Only malate and α-ketoglutarate correlated with whole-brain morphine levels. The average brain level of morphine is also shown in Table II. The correlation between whole brain morphine level and analgesia was $+0.47$ ($p < .05$). The average brain level of morphine was 0.06 ± 0.005 µg/g (S. E.).

Discussion. The finding of a positive correlation between blood pyruvate, lactate, and possibly citrate with analgesia suggests that high glycolytic flux (as measured by high pyruvate levels) may be a component of analgesia. Since there were no significant correlations between substrate levels and pre-drug reaction time, it appears unlikely that peripheral intermediary levels are associated with basic sensitivity to pain. High levels of malate and α-ketoglutarate were associated with high levels of brain morphine, suggesting the these substrates may be important in morphine uptake.

The finding of a relatively low (0.47) but statistically significant correlation between brain morhpine level and analgesia is in agreement with the findings of MULÉ et al.[11] and MULÉ[12]. This suggests that the intensity of the metabolic change produced by morphine may be more important in determining the level of analgesia than the amount of drug present.

The data also suggest that some of the metabolic changes involved in morphine-induced analgesia may be mediated through the citric acid cycle.

Zusammenfassung. Es konnte gezeigt werden, dass bei Ratten eine Beziehung besteht zwischen der analgetischen Wirkung von Morphin und den Blut-Konzentrationen einiger Metaboliten des Krebs-Zyklus.

A. D. SHERMAN and C. L. MITCHELL

Department of Pharmacology,
College of Medicine,
University of Iowa,
Iowa City (Iowa 52240, USA), 3 July 1972

[10] M. WEIL and A. ABDELOMONEN, Circulation *41*, 989 (1970).
[11] S. MULÉ, C. REDMAN and J. FLESHER, J. Pharmac. exp. Ther. *157*, 459 (1967).
[12] S. MULÉ, Archs int. Pharmacodyn. Thér. *173*, 201 (1968).

Selective Suppression of Rapid Eye Movement Sleep (REM) by Fusaric Acid, an Inhibitor of Dopamine-β-Oxidase

Several converging lines of evidence point to an important participation of noradrenaline (NA) in the production of REM. However, existing data are still conflicting. In the cat, lesioning of the locus coeruleus rich in the NA-containing neurons[1] has been reported by some[2] to suppress REM selectively, while others[3] have been unable to confirm this finding. Experiments performed with inhibitors of enzymes at different steps of biosynthesis of NA have shown that decreased concentration of NA in the brain leads to reduction in the amount of REM[4,5]. This is in line with the fact that administration of DOPA restores reserpine-suppressed REM[6]. However, inhibition of tyrosine hydroxylase by α-methyl-*p*-tyrosine has been shown to increase the REM amount, while decreasing the concentration of NA in the brain[7].

It seems that, with respect to the relationship between NA and REM, conflicts among the results obtained through pharmacological interventions may arise from coincidental involvement of factors other than NA. For this reason we have been prompted to study the effect on REM of fusaric acid (5-butylpicolinic acid), which possesses a very pertinent property without unfavorable complications. This drug has no reported action other than inhibition of dopamine-β-oxidase and consequent reduction of the concentration of NA in the central nervous system as well as in the periphery[8].

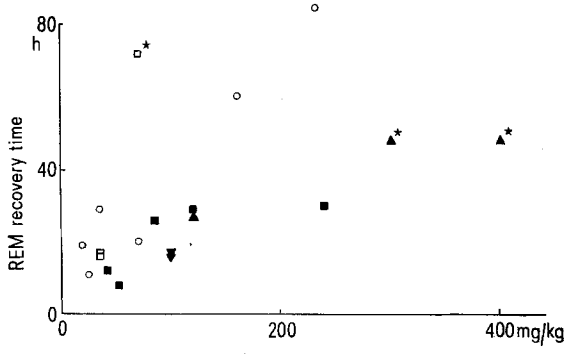

Fig. 1. REM recovery time after single administrations of fusaric acid or its calcium salt in 4 cats. Ordinate; time in h for the cumulative amount of REM to catch up to the control level after drug administration. Abscissa; the amount of drug administered (open marks, peritoneal injection of fusaric acid; filled marks, oral administration of the calcium salt). Marks accompanied with asterisk indicate failure to catch up to the control level within the follow-up period.

[1] N. E. ANDÉN, A. DAHLSTRÖM, K. FUXE, K. LARSSON, L. OLSON and U. UNGERSTEDT, Acta physiol. scand. *67*, 313 (1966). – U. UNGERSTEDT, Acta physiol. scand. Suppl. *367*, 1 (1971).
[2] M. JOUVET and F. DELORME, C. r. Séanc. Soc. Biol., Paris *159*, 895 (1965). – B. ROUSSEL, A. BUGUET, P. BOBILLIER and M. JOUVET, C. r. Séanc. Soc. Biol., Paris *161*, 2537 (1967).
[3] G. CARLI and A. ZANCHETTI, Archs ital. Biol. *103*, 751 (1965). – K. HENLEY and A. R. MORRISON, Psychophysiol. *6*, 245 (1969).
[4] D. PEYRETHON-DUSAN and J. L. FROMENT, C. r. Séanc. Soc. Biol. *162*, 2141 (1968).
[5] F. DELORME, Monoamines et sommeil, dissertation of medicine, University of Lyon (1966). – D. PEYRETHON-DUSAN, J. PEYRETHON and M. JOUVET, C. r. Séanc. Soc. Biol., Paris *162*, 116 (1968).
[6] J. MATSUMOTO and M. JOUVET, C. r. Séanc. Soc. Biol., Paris *158*, 2137 (1964).
[7] C. D. KING and R. E. JEWETT, J. Pharmac. exp. Ther. *177*, 188 (1971). – E. HARTMANN, T. J. BRIDWELL and J. J. SCHILDKRAUT, Psychopharmacologia *21*, 157 (1971).
[8] T. NAGATSU, H. HIDAKA, H. KUZUYA, K. TAKEYA, H. UMEZAWA, T. TAKEUCHI and H. SUDA, Biochem. Pharmac. *19*, 35 (1970).

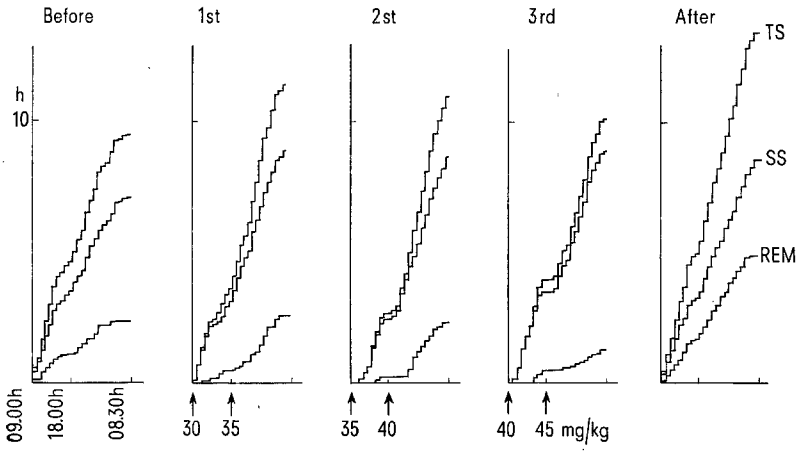

Fig. 2. Effect of peritoneal injection of fusaric acid for 3 consecutive days on total sleep (TS), SS and REM, each being shown in cumulative amounts for 1 h periods. Drug or placebo was given twice a day at 09.00 h and 18.00 h. The amounts of SS in a day were within the normal range (494 min ± 68 S.D. in this cat) in all 5 days, while those of REM were out of the normal (162 min ± 40 S.D.) in the last days.

Experiments were performed on 4 freely moving cats carrying chronically implanted electrodes. Starting from 3 or 4 days before administration of the drug, the baseline sleep pattern was recorded continuously except for regular interruptions twice a day. Fusaric acid was injected i.p. or its calcium salt was given p.o. Recording was continued for 2 to 4 days after the drug administration. Placebos were given on the days when the drug was not administered.

Single administration of a moderate dose of fusaric acid (30–40 mg/kg) or its calcium salt (50–200 mg/kg) suppressed REM for 5–10 h. Thereafter REM showed a rebound, and the cumulative amount of REM usually caught up with the normal level between 10 and 30 h after drug administration (REM recovery time in Figure 1). Large doses (over 70 mg/kg of fusaric acid or over 300 mg/kg of the calcium salt) suppressed REM for a much longer period and prolonged its recovery time, in some cases, in excess of 2 or 3 days.

The administration of moderate doses for several consecutive days was also conducted to avoid gastrointestinal disturbances and hypotension[9] following a single large dose. As illustrated in Figure 2, with increasing the dose, the suppression of REM was more prolonged and the cumulative REM record failed to catch up to the control value by the next morning. On the following day with placebo, REM rebounded far above the control value.

In contrast to its effect on REM, the drug had no significant influence on slow wave sleep (SS). This is consistent with the current concept of the SS mechanism, which is said to be dependent not on a noradrenergic mechanism, but primarily on a serotonergic one[10]. Only with large doses, SS was depressed for a few hours following drug administration. This is presumed to be secondary to gastrointestinal and circulatory perturbations, since recovery was always complete within a day.

The suppression of REM by fusaric acid is similar to the effect of another dopamine-β-oxidase, disulfiram which, however, causes a slight increase in the amount of SS and elicits abnormal movements[4]. Since the concentration of dopamine in the brain is not altered by fusaric acid[8], the differences in the action between the 2 drugs might be explained by the accumulation of dopamine in the brain[11], resulting from inhibition by disulfiram of aldehyde dehydrogenase[12], required for the degradation of dopamine.

Although fusaric acid selectively suppressed peripheral manifestation of REM, the biological need for REM seems not to be suppressed by the drug, because in most cases REM rebounded after a period of drug-induced suppression. Therefore it is conceivable that a noradrenergic mechanism is heavily involved in the transmission of information from the structures which elaborate the biological need for REM to the individual sites which have been claimed to be localized in the lower brain stem and to be directly related to the peripheral manifestations of REM[2,13].

In some experiments with large doses, no appreciable REM rebound was observed. This might be due to a shift in the level of need for REM to a lower one, the level being determined by many biological parameters, including the concentration of NA in the body that can be manipulated by the drug. Alternatively, the lack of rebound might suggest the presence in the neuronal structures elaborating the need for REM of a noradrenergic mechanism which is less vulnerable to the action of fusaric acid than the above-posturated transmission system[14].

Résumé. L'administration d'acide fusarique, un inhibiteur de la dopamine-β-oxidase, a supprimé électivement le sommeil REM, sans influence significative sur le sommeil lent. Le rôle de noradrénaline dans le mécanisme central du sommeil REM est discuté.

T. SATOH and R. TANAKA

Department of Physiology, School of Dentistry, Aichi-Gakuin University, 2-11, Suemori-dori, Chikusa-ku, Nagoya, 464 (Japan), 4 July 1972.

[9] H. HIDAKA, T. NAGATSU, K. TAKEYA, T. TAKEUCHI, H. SUDA, K. KOJIRI, M. MATSUZAKI and H. UMEZAWA, J. Antibiot. Tokyo *22*, 228 (1969).

[10] W. P. KOELLA, A. FELDSTEIN and J. S. CZICMAN, Electroenceph. clin. Neurophysiol. *25*, 481 (1968). – M. JOUVET, Science *163*, 32 (1969).

[11] M. GOLDSTEIN and K. NAKAJIMA, J. Pharmac. exp. Ther. *157*, 96 (1967).

[12] E. RACKER, in *Methods in Enzymology* (Eds. S. P. COLOWICK and N. O. KAPLAN; Academic Press, New York 1955), Vol. 1, 514.

[13] O. POMPEIANO and A. R. MORRISON, Archs ital. Biol. *103*, 569 (1965). – M. MATZSUZAKI, Brain Res. *13*, 247 (1969).

[14] Fusaric acid was available through the courtesy of Dr. T. NAGATSU, Department of Biochemistry, School of Dentistry, Aichi-Gakuin University. Thanks are due to Dr. L. M. VERNON for his reading of the manuscript. This work was supported in part by a grant from Japan Society for Promotion of Science under contract No. 5R023.