The results of the present experiments suggest that the normal intracellular pH of the rabbit erythrocyte is 7.27. This value is consistent with previous estimations of red cell pH in the rat[4] and in man[3]. Both these studies were based on the equilibration of non-labelled DMO with red cells in vitro. On the other hand, measurements based on equilibration with DMO in vivo[2] or on the pH of haemolysates[3–5] suggest that erythrocyte pH is 0.10–0.20 pH units lower. These latter values are not consistent with the membrane potential of the erythrocyte[7,8]. Estimation of the pH of haemolysates is unlikely to reflect true red cell pH, since the measured potential is biassed by the liquid junction potential[3,11] and the effect of hydration on electron donor and electron acceptor groups in the intact erythrocyte[12].

The effect of non-labelled DMO on the distribution of DMO-2-C^{14}

Blood cell suspension +	DMO-2-C^{14} in cell suspension (dpm/ml) (x)	DMO-2-C^{14} in separated buffer (dpm/ml) (y)	Distribution $\frac{(x)}{(y)}$
DMO-2-C^{14} (10^6 dpm)	278,300	349,600	0.80
DMO-2-C^{14} (10^6 dpm) + 0.1 mg DMO	294,800	349,300	0.84
DMO-2-C^{14} (10^6 dpm) + 1.0 mg DMO	258,500	319,700	0.81
DMO-2-C^{14} (10^6 dpm) + 10.0 mg DMO	249,900	297,200	0.84

The amount of DMO-2-C^{14} equilibrated with the blood cell suspensions was equivalent to 1–10 µg of the compound.

Zusammenfassung. Die pH-Werte von Kaninchen-Erythrozyten wurden mit Hilfe der DMO-Verteilungsmethode untersucht und das intrazelluläre pH in Übereinstimmung mit dem Membranpotential gefunden.

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Rate of Oxidation and Tissue Concentrations of Ethanol in Mice

We have previously shown that a s.c. injection of the median lethal dose of ethanol in mice produced a tissue concentration that reduced by about one-third the thermodynamic activity of the intercellular water 2 h later without a compensating decrease in electrolyte concentration[1]. We became interested to find out precisely the concentrations of ethanol in various organs of the mouse at a given time after the injection and any significant changes in the weight of the organs.

The coefficient of ethyl oxidation (CEO), a useful indicator of the speed and nature of ethanol oxidation in a given species, is quite variable in the literature. In man the normal value for the CEO is 90 mg/kg/h[2]. In mice KINARD[3] obtained the following values: 455 ± 173.3 mg/kg/h during the first $^1/_2$ h after the injection, 555.5 ± 73.1 mg/kg/h for the first h and 595.5 ± 68.3 mg/kg/h for the first 4 h. MARSHALL and OWENS[4] reported 1322 mg/kg/h, 1123 mg/kg/h and 691 mg/kg/h respectively for the 3 periods. Finally, NELSON et al.[5] obtained a CEO of 625 mg/kg/h for the first h and 598 mg/kg/h for the first 3 h. In this paper we have determined the CEO in a group of mice of the same stock.

Ethanol at a dose of 10 g/kg was injected as a 25% solution with NaCl at 0.9% into 30 Swiss male albino mice. The animals were on a commercial diet ad libitum and had free access to water. They were not fasted before the experiments. Ethanol was given s.c. in keeping with the same conditions as our previous experiments. The mice were killed by decapitation 1, 2 and 4 h respectively after the injection. The blood was collected in heparinized conical tubes and centrifuged for 10 min at average speed. The brain, heart, liver, uro-genital fat, kidneys and carcass were rapidly excised and immersed in saturated picric acid solution in previously weighed vials. After weighing, the tissues were transferred to microdistillation flasks and ethanol in each organ estimated according to LE BRETON et al.[6]. Control animals received identical treatment except that they were injected with NaCl only. Endogenous alcohol values were obtained by distilling 3 mice at a time. 3 determinations were made in each case. The CEO was obtained by estimating the alcohol in the distillate from a single mouse at a time. After injection, the animals were placed on a wire mesh resting on a glass trough and the whole covered by a large open bell-jar to minimize the loss by respiration. Urine and feces were quantitatively transferred to the distillation flask.

After an injection of 10 g/kg, the average ethanol concentration in all the organs studied was from 4–6 g/kg (Table I). The highest concentration was in the brain and the plasma. There was a progressive reduction in the concentration with time, the peak being obtained 1 h after the injection. There was a reduction of the wet weight of the brain, the heart and the liver (Table II). Endogenous alcohol had a value of 8.79 mg/kg. The CEO was $1578 \pm$

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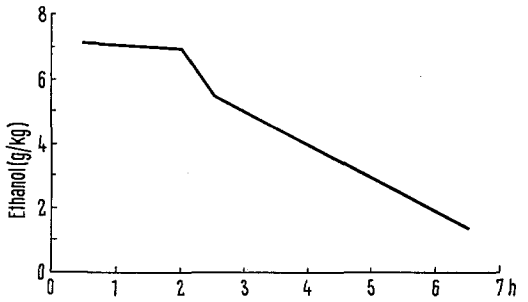

Total concentration of ethanol in mice after injection of the median lethal dose (10 g/kg).

Table I. Concentration of ethanol in various organs of the mouse after a s.c. injection of LD_{50} (10 g/kg)

Organ	Concentration (g/kg)		
	1 h (6)[a]	2 h (21)[a]	4 h (3)[a]
Brain	4.8	3.8	2.0
Heart	4.2	3.0	–
Liver	4.2	3.5	3.5
Kidney	3.0	3.0	–
Carcass	4.6	3.7	2.5
Plasma	6.0	2.0	–
Urogenital fat	0.5	0.1	–

[a] Number of animals.

Table II. Effects of ethanol at 10 g/kg on the weight of mouse organs

	Control (Mean weight of animals: 30.86 g)		Ethanol (Mean weight of animals: 35.28 g)	
	g/kg body weight	Absolute weight (g)	g/kg body weight	Absolute weight (g)
Brain	14.66 ± 0.68	0.4588 ± 0.021 (17)	12.69 ± 1.03	0.4478 ± 0.0364 (13)
Heart	5.37 ± 2.39	0.1659 ± 0.074 (17)	4.58 ± 1.26	0.1618 ± 0.0446 (13)
Liver	53.67 ± 7.41	1.6564 ± 0.229 (17)	40.81 ± 14.90	1.4398 ± 0.526 (13)
Urogenital fat	15.15 ± 9.62	0.4678 ± 0.297 (17)	17.34 ± 10.54	0.6118 ± 0.372 (13)
Carcass	656.03 ± 100.69	20.2453 ± 3.108 (17)	650.37 ± 70.90	22.9452 ± 2.502[a] (7)
Kidney[b]	3.69 ± 0.82	0.1139 ± 0.0255 (5)	3.38 ± 0.79	0.1194 ± 0.0282 (9)

Figures in parentheses indicate the total number of animals. [a] $p < 0.03$. [b] 1 Kidney.

209 mg/kg/h (average of 10 determinations). The Figure shows that the oxidation rate is constant from 2.5 h after injection. The results, like those of KINARD et al.[3,5], also show that the CEO increases with time. The results of MARSHALL and OWENS[4], however, indicate the opposite.

Hitherto it was thought that ethanol is oxidized only in the blood and in the liver due to the action of alcohol dehydrogenase. But there is strong evidence to support the oxidation of ethanol in the brain in vitro, BURBRIDGE[7], and in vivo[7,8], and in the kidney (BARTLETT and BARNET[9,10]). The brain and the blood, having the highest water content[1], will have at equilibrium the largest concentration of ethanol (Table I), while the fat and skeleton will contain the lowest. It is not surprising therefore that acetaldehyde, a product of ethanol oxidation, is itself very slowly oxidized in the brain and accumulates especially in the cerebellum[11] which controls body equilibrium. A high concentration of ethanol will result in more pyruvic acid accumulating, since oxidation of the latter is inhibited by acetaldehyde.

The reductions of the wet weight of the organs are not significant except for that of the carcass ($p < 0.03$), probably due to an elevated subcutaneous edema. A longer time and possibly chronic intoxication seems to be necessary for changes in dry weight to occur. From these and previous findings, the CEO for mice seems to be the highest although the rate of ethanol disappearance in bats (550–700 mg/kg/h) is reported to be higher than for most other species[12].

Résumé. Chez la souris Swiss albinos le CEO est de 1578 ± 209 mg/kg/h, après une injection de 10 g/kg d'éthanol. Les concentrations tissulaires sont de l'ordre de 4 à 6 g/kg. Les réductions des poids frais des organes ne sont pas significatifs.

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