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The effect of ethanol on phenobarbitone and pentobarbitone absorption into rat blood and brain

B. B. COLDWELL, H. L. TRENHOLM, B. H. THOMAS, AND S. CHARBONNEAU

Research Laboratories, Food and Drug Directorate, Department of National Health and Welfare, Ottawa, Canada

Male rats administered [^{14}C]phenobarbitone (50 mg/kg) or [^{14}C]-pentobarbitone (30 mg/kg) simultaneously with either 15% ethanol (3 g/kg) or saline intraperitoneally were killed 5, 10 or 20 min after injection. The radioactivity in the blood, whole brain and different brain areas was measured. Phenobarbitone was absorbed more slowly into the blood and brain than pentobarbitone. Ethanol-treated rats had significantly higher phenobarbitone concentrations than the saline-treated controls in the blood, whole brain, cerebrum and cerebellum up to 10 min after injection. Pentobarbitone concentrations were not significantly altered by ethanol. Barbiturate concentrations in the cerebral cortex were lower than in other regions of the brain. The brain:blood barbiturate ratios were not appreciably changed by ethanol. It is concluded that ethanol (15%) given intraperitoneally aided the transport of phenobarbitone across the peritoneum and hence increased the rate of its absorption into the blood and brain.

The reasons for the severe toxicity resulting from the simultaneous ingestion of ethanol and barbiturates remain obscure. Recently, ethanol has been shown to inhibit the degradation of several drugs, including pentobarbitone, in microsomal preparations and in liver slices and *in vivo* slows the rate of disappearance of drugs from the blood (Rubin & Lieber, 1968; Rubin, Bacchin & others, 1970; Rubin, Gang & others, 1970). It appears unlikely that the immediate pharmacological effects of ethanol–barbiturate interaction are due to changes in metabolism since they are most pronounced with the least metabolized barbiturates (Wiberg, Coldwell & Trenholm, 1969). Seidel (1967) observed with mice that pentobarbitone concentrations in several tissues including blood and brain, were enhanced by ethanol but phenobarbitone concentrations remained unaffected. In rats, ethanol increased the brain concentrations of phenobarbitone and barbitone, whilst those of pentobarbitone appeared unaffected (Coldwell, Wiberg & Trenholm, 1970). Subsequently, using [^{14}C] compounds, it was found that the concentrations of phenobarbitone and pentobarbitone in several tissues including the brain, were elevated in the presence of ethanol (Coldwell, Trenholm & others, 1971). This paper describes the results of a detailed investigation of the effect of ethanol on the distribution of [^{14}C]phenobarbitone and [^{14}C]pentobarbitone in the rat brain during the immediate post-injection period.

MATERIALS AND METHODS

Male rats of the Wistar strain, 175 to 225 g, were housed 8–10 per cage and acclimatized one week to the environment. After overnight starvation the animals were randomly divided into 4 groups of 36 each and treated intraperitoneally (20 ml/kg)

with either pentobarbitone, (30 mg/kg) or phenobarbitone, (50 mg/kg) in saline or 15% ethanol (3 g/kg). The barbiturates were ring-labelled with ^{14}C in the two position; the specific activity of each dose of pentobarbitone was 500 μCi/g and of phenobarbitone was 175 μCi/g.

Twelve animals of each group were decapitated 5, 10 and 20 min after drug injection. The brains were removed and sectioned into the half-brain, cerebellum, pons plus medulla, thalamus and cerebral cortex. Triplicate specimens of the half-brain and all of the other sections were weighed immediately and added to Soluene (1 ml/100 mg, Packard Instrument Co.) in scintillation counter vials. The blood was collected and triplicate 10 μl aliquots were treated similarly. After incubation at 37° for 18 h, a toluene-based scintillation fluid containing 0·6% 2,5-diphenyloxazole (PPO) and 0·02% *p*-bis-(2-4-methyl-5-phenyloxazolyl)-benzene (dimethyl POPOP) was added to each vial, in the proportion of 10 ml/ml of Soluene. The radioactivity was measured with a liquid scintillation counter. All vials were counted three times. Standards were similarly processed to obtain measurements of machine efficiency, background count, and specific activity of the injected drug. Quenching was corrected by the external standard method. All calculations were made on an IBM-360 computer. The radioactivity was expressed as unchanged barbiturate.

RESULTS

The concentrations of [^{14}C]pentobarbitone in the blood, brain, and the various component areas of the latter, in the presence and absence of ethanol, are shown in Fig. 1A. Peak concentrations were reached in all tissues, except the half-brain, within 10 min of the drug being given. Ethanol had no significant effect on pentobarbitone uptake. Brain concentration of pentobarbitone reflected those in blood

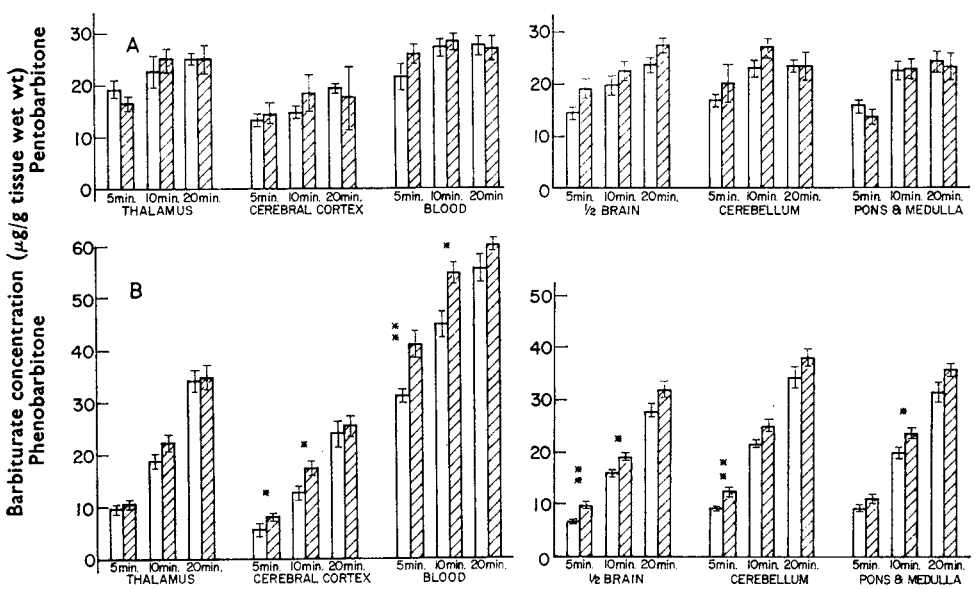

FIG. 1. Barbiturate concentrations in blood and brain tissue at 5, 10 and 20 min intervals after administrtion (i.p.) of (A) pentobarbitone (30 mg/kg), (B) phenobarbitone (50 mg/kg) (open columns) in saline and in ethanol (3 g/kg, 15% w/v soln) (hatched columns). * $P<0·05$; ** $P<0·01$.

and the brain:blood pentobarbitone ratios were not appreciably altered by ethanol. Concentrations in the cerebral cortex were significantly lower than in other areas of the brain.

Phenobarbitone concentrations in the tissues increased continuously during the 20 min after injection (Fig. 1B), however, that in the brain (27·7 $\pm$ 1·6 μg/g) lagged behind that in the blood (55·7 $\pm$ 2·8 μg/g) at 20 min. The uptake of the barbiturate in blood and brain was more rapid in the presence of ethanol, especially during the first 10 min after injection. This was reflected in all areas of the brain except the thalamus. Concentrations in the cerebral cortex were significantly lower than in the thalamus and cerebellum 20 min after drug administration. The brain:blood phenobarbitone ratios were unaffected by ethanol.

DISCUSSION

Since pentobarbitone is more lipid soluble than phenobarbitone, under physiological conditions it is more rapidly absorbed by tissues (Goodman & Gilman, 1965). At physiological concentrations ethanol slightly increased the lipid solubility of both barbiturates (Thomas, Coldwell, Trenholm & Wiberg, unpublished); in 15% w/w ethanol their solubilities are several times greater than in water (Breon & Paruta, 1970). Our findings suggest that ethanol increased the *in vivo* lipid solubility of phenobarbitone and its rate of transport across the peritoneum and uptake into the blood. This effect would be less pronounced on pentobarbitone uptake because of its inherent high lipid solubility and rapid absorption.

There was no indication that ethanol (3 g/kg) caused a perturbation of the blood-brain barrier. Unpublished data from our laboratory has established that pentobarbitone metabolites are absent from rat brain 3 h after dosing the animals with the barbiturate in saline or 15% ethanol. The similarity in the brain:blood barbiturate ratios in the presence and absence of ethanol and the non-specific nature of the higher tissue concentration of barbiturate when ethanol is administered simultaneously (Coldwell & others, 1971) indicate that in the rat the blood-brain barrier is relatively unaffected by intoxicating amounts of ethanol.

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