ent in sufficient quantities in the purified casein rations.

Summary. A vit. B_{12} requirement was not demonstrated for the Syrian hamster when fed either a corn-soybean oil meal ration with or without iodinated casein, or a casein basal ration with or without iodinated casein. No increase in the rate of gain was observed when 2% whole liver substance was added to these rations. Parallel experiments were conducted with rats fed the corn-soybean oil meal ration plus iodinated casein in which a vit. B_{12} requirement was demonstrated. A reduction in food efficiency was observed in rats attributable to the vit. B_{12} deficiency.

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Anticonvulsant Properties of Benadryl and Pyribenzamine.* (18157)

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It is well known that antihistaminic drugs, such as diphenhydramine (Benadryl) and tripelennamine (Pyribenzamine), produce side effects by acting on the central nervous system. According to Feinberg(1) the most consistent side effect of all antihistaminic drugs is sedation. The clinical implication of this effect has been recognized, particularly in relation to the use of antihistaminic agents in the treatment of diseases referable to the central nervous system.

The salutary effect of Benadryl in paralysis agitans was reported by McGavack and coworkers(2). Subsequently, Budnitz(3) called attention to the beneficial effect of this drug in Parkinson's disease. These observations have since been confirmed(4-7) and extended to include other antihistaminic agents(4,6,7). McGavack and coworkers(2) also tried Benadryl in 3 patients with grand mal epilepsy, and reported an increased frequency of seizures as a result of this medication. More recently, Churchill and Gammon(8) have shown that Benadryl diminished the electrical abnormality and decreased the incidence of seizures in petit mal; on the other hand, both Pyribenzamine and Benadryl induced seizures in some patients with grand mal epilepsy. Such reports on the use of antihistaminic drugs are particularly interesting when one considers that convulsions may occur when large doses of these agents are ingested(9-11).

In view of the wide use of antihistaminic agents and because of the limited information on their central nervous system actions, it was thought important to study their anticonvulsant effects in experimental animals. It was anticipated that such a study might not only provide information on the anticonvulsant properties of these drugs but might also indicate the validity of employing them in the treatment of allergic manifestations in epileptic patients.

Methods. Male albino rats obtained from

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TABLE I. Anticonvulsant Potency of Benadryl and Pyribenzamine in Comparison with Dilantin.*

Drug	Route	Toxicity (TD_{50}) mg/kg	Max. electroshock test†	
			Effective dose (ED_{50}) mg/kg	Protective index (P.I.)
Benadryl	Oral	193 (146-255)	125 (112-140)	1.5 (1.1-2.1)
,,	i.p.	18.5 (13.7-24.9)	11.0 (10.2-11.8)	1.7 (1.2-2.3)
Pyribenzamine	Oral	450 (378-535)	143 (124-164)	3.1 (2.5-3.9)
,,	i.p.	29.0 (26.4-31.9)	13.0 (9.7-17.4)	2.2 (1.7-3.0)
Dilantin	Oral	>3200	30.0 (20.0-45.0)	>100
,,	i.p.	82 (55-122)	11.3 (8.8-14.5)	7.3 (4.5-11.8)

* All values in parentheses indicate 95% confidence limits.
† 150 m A., 0.2 second, alternating current, corneal electrodes.
Dilantin and Benadryl were kindly supplied by Dr. Oliver Kamm, Parke, Davis and Co.; Pyribenzamine, by Dr. Jock L. Graeme, Ciba Pharmaceutical Products.

the Sprague-Dawley farm were used as experimental animals. The substances examined in this study were given both orally and intraperitoneally. When the oral route of administration was employed, the drug was suspended in 10% acacia mucilage and given by stomach tube. In either case the assay procedures were carried out at the previously determined time of peak drug effect. The details of the assay procedures have been previously published(12). Briefly, anticonvulsant potency (ED_{50}) of each drug was determined by its ability to abolish the tonic extensor component of maximal seizures (maximal electroshock seizure test) and to protect the animals from the subcutaneous administration of 70 mg/kg of pentylenetetrazol (Metrazol test). In addition, minimal neurological toxicity (such as loss of placing responses, ataxia, depression, etc.,) was determined (TD_{50}), and protective indices ($P.I.=TD_{50}/ED_{50}$) were calculated. To permit critical comparison of potencies, the data obtained were evaluated by the method of Litchfield and Wilcoxon(13).

Results. The results obtained are shown in Table I.

It may be seen that Benadryl and Pyribenzamine are more toxic than diphenylhydantoin (Dilantin). Benadryl is the most toxic of the 3 compounds studied. When given by the intraperitoneal route, 18.5 mg/kg produced minimal toxic symptoms in 50% of rats as compared to 29.0 mg/kg for Pyribenzamine and 82 mg/kg for Dilantin. It was impossible to estimate toxicity after the oral administration of Dilantin because of poor absorption of this drug from the gastro-intestinal tract. We have previously called attention to the fact that only a small percentage of the maximum dose administered (3200 mg/kg) is actually absorbed when the oral route of administration is employed(14).

No remarkable difference was observed in the antielectroshock potency of Benadryl and Pyribenzamine as compared to Dilantin. Thus, when administered intraperitoneally, 11.0 mg/kg of Benadryl abolished the tonic extensor component in 50% of rats as compared to 13.0 mg/kg for Pyribenzamine and 11.3 mg/kg for Dilantin. By the oral route Dilantin was found to be 4 to 5 times as potent as Benadryl or Pyribenzamine.

No protection was afforded by these compounds against Metrazol convulsions. Indeed, all animals receiving Pyribenzamine or large doses of Benadryl appeared exquisitely sensitive to Metrazol and had severe, recur-

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14. Swinyard, E. A., and Toman, J. E. P., *J. Pharm. and Exp. Therap.*, in press.

rent and often fatal seizures.

Discussion. Everett(15) reported that Benadryl and Pyribenzamine did not effect the "electroshock threshold for full tonic clonic seizures." The observations were made on mice given intraperitoneal doses of 10 to 50 mg/kg. Our data were obtained in rats and indicate that these same drugs have a potency almost as great as Dilantin when tested for their ability to modify maximal electroshock seizure pattern. The reason for the difference in the results reported here and those of Everett is unknown, unless it be a species difference.

It is interesting to compare the observed differences between Benadryl and Pyribenzamine. Benadryl is approximately twice as toxic as Pyribenzamine but the toxicity patterns differ for the two drugs. Benadryl has a wide range between the doses causing hyperexcitability and spontaneous seizures. Some differences were also observed when these drugs were tested against Metrazol. Neither Benadryl nor Pyribenzamine had the ability to prevent Metrazol convulsions, but small doses (5 to 10 mg/kg) of Benadryl did modify seizure pattern. In contrast, Benadryl in large doses and Pyribenzamine in all doses employed appeared to synergize with Metrazol to cause severe, protracted and often lethal convulsions.

The data indicate that Benadryl, Pyribenzamine and Dilantin have several properties in common. All 3 drugs modify maximal electroshock seizure pattern in non-toxic doses but do not prevent Metrazol convulsions. Toxic doses of all three drugs produce increased excitability and convulsions. We have previously called attention to the fact that clinically useful antiepileptic hydantoins and barbiturates are characterized in the laboratory by their ability to modify maximal electroshock seizure pattern(16). Inasmuch as Benadryl and Pyribenzamine have a maximal electroshock seizure potency approaching that of Dilantin, it would be of interest to know whether they are effective clinically in grand mal. The only data we have been able to find concerning the effects of antihistaminics in grand mal are those of McGavack and coworkers(2) and Churchill and Gammon(8). McGavack and coworkers tried Benadryl in 3 cases of grand mal and reported a "definite aggravation of status" in each instance. Churchill and Gammon employed Benadryl in 4 cases of grand mal and Pyribenzamine in one case of grand mal but concluded that the patients had "major seizures so infrequently" that the effect of the medication could not be evaluated. It would be most interesting to have additional clinical data on a larger series of patients with grand mal in order more precisely to determine the effect of these drugs. If such data substantiated the limited results reported to date it would then appear that Benadryl and Pyribenzamine have no value in grand mal and may actually aggravate such cases. This would suggest that the maximal electroshock seizure test may have questionable predictive value for antiepileptic efficacy when employed to measure anticonvulsant action of antihistaminic derivatives of ethanolamine and ethylenediamine.

It is tempting to speculate on the mechanism whereby Benadryl depresses the spike-wave abnormality, as reported by Churchill and Gammon(8). It has long been known that certain central stimulants, such as ephedrine, amphetamine and caffeine are effective in some cases of petit mal. Indeed, before the discovery of Trimethadione, Paradione and Phenurone, these agents were practically the only drugs useful in petit mal. The mechanism of antiepileptic action of central stimulants is unknown, but it is probably entirely unlike that of the central depressants. It is well established that sensory stimulation may occasionally abort cortical seizures(17,18) and that seizure discharges are more common in sleep than during the waking state(19). In

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view of these facts we have previously postulated that "it is not unlikely that a drug-induced increase in activity of normal brain may inhibit discharges from seizure foci"(20). The greater ability of Benadryl, as compared with Pyribenzamine, to stimulate the central nervous system in subconvulsive doses could account for the salutary effect of this drug in petit mal.

Summary. Benadryl, Pyribenzamine and Dilantin were tested in rats for toxicity and for ability to modify maximal electroshock seizure pattern and to prevent Metrazol convulsions. The data indicate that all three drugs modify maximal electroshock seizure pattern but do not prevent Metrazol convulsions. The significance of the data is discussed.

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Influence of Methadone on Oxygen Uptake and Glycogenolysis of Rat Liver Slices.* (18158)

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The effects of morphine-like analgetics, including methadone, on the metabolism of surviving tissues and isolated enzyme systems have been studied by several workers(1-4), in an attempt to explain the mechanism of action of analgetics. Although these studies have not accomplished their primary purpose, they show that analgetics affect tissue metabolism in a manner different from anesthetics, and they indicate the usefulness of methadone as a tool in the study of oxidative processes.

The present study was undertaken as a result of the observation that the same concentration of methadone increased the oxygen uptake of liver slices from fed rats but decreased that of slices from rats starved overnight. Since the effect was the same whether or not glucose was included in the suspending medium, the possibility of increased glycogenolysis by liver slices in the presence of methadone has been investigated by biochemical and histological techniques.

Methods. Liver slices were obtained from adult albino rats of the Slonaker-Wistar strain. When high initial liver glycogen values were desired, the animals were starved for twenty-four hours and then given a mixture of half glucose, half ground stock diet overnight before use. Animals which had been starved overnight provided liver slices low in glycogen content. The rats were sacrificed by a blow on the head, the livers quickly removed, placed in a cold box(5) and with a Martin slicer(6) cut into slices approximately 0.5 mm thick. The slices were randomized and used either for measurement of oxygen uptake, chemical determination of glycogen or histologic examination. Oxygen consumption was measured by the direct method of Warburg(7) at 37.2°C. The liver slices which weighed 50-100 mg were suspended in 2 ml of a modified Krebs-Ringer phosphate buffer(4) with or

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