Discussion. These results indicate that kynurenine does not appear to be an intermediate in pigment formation by skins of A^y/a mice. The fact that DL-5-hydroxytryptophan is oxidized nearly as rapidly as equimolar L-tryptophan means that L-5-hydroxytryptophan is probably oxidized more rapidly than L-tryptophan. This suggests that the hydroxylated form may be an intermediate in formation of the colored substance. In the absence of strong nitric acid, the nitrosonaphthol reagent is said to be quite specific for 5-hydroxyindoles when the violet chromophore results, and a number of compounds have been examined(8). Since supernatants after incubation with tryptophan yield a substance forming a similar but not identical chromophore, it is possible that a related hydroxyindole or a di-hydroxyindole is formed. It is unlikely that the difference in the chromophore is due to an impurity, since 5-hydroxytryptophan was added to skin suspensions before taking the spectra, and the chromophore produced was identical with that from 5-hydroxytryptophan alone. In addition, it was found that the chromophore of 5-hydroxytryptophan was not altered by excess tryptophan, and finally, an impurity might be expected to add, rather than subtract, optical density. Further experiments will be required to determine whether the colored substance formed may be due to action of an amine oxidase on a hydroxytryptamine, as has been described(9,10), or whether a di-hydroxyindole may lead to a quinone, as has been suggested(3).

Summary. 1. Skin powders from 10-day-old mice with a dominant gene for yellow hair (A^y/a) oxidize L-tryptophan, DL-5-hydroxytryptophan, and 5-hydroxytryptamine, but not D-tryptophan. Yellow solutions are formed. 2. L-kynurenine does not appear to be an intermediate, since it is not oxidized, nor does it disappear at an appreciable rate when incubated with skin powders from these mice. It does not appear when tryptophan is incubated with skin powders. 3. When tryptophan is oxidized by these skin powders, a substance appears which gives a reaction with nitrosonaphthol in the absence of strong nitric acid. The chromophore is distinctive, and differs from that formed by 5-hydroxyindoles.

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Differential Action of Styramate and Meprobamate on Spinal Reflexes. (24900)

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Styramate, (Sinaxar®) 2 phenyl-2 hydroxyethyl carbamate, a drug with central depressing action has been recently recommended by us as skeletal muscle relaxant for human use. Its pharmacological, toxicological and anticonvulsive properties have been described(1) and preliminary data on its skeletal muscle relaxant activity have been presented(2). In general its properties are similar to those of mephenesin except that it is 3 to 6 times longer acting. The influence of styramate on spinal polysynaptic transmission is not asso-

ciated with hypnosis as with pentobarbital. The present report contains findings concerning its effects on polysynaptic reflexes in spinal and in decerebrate cats. In our investigation augmentation of patellar reflex during contralateral sciatic nerve stimulation (SCF), which gives results parallel to electrophysiological determinations(3-5), was selected as method of measure.

Method. Forty-one cats (18 decerebrate and 23 spinal) were anesthetized with ether and tracheotomized. Decerebration was made by sectioning brainstem rostral to superior colliculi. Spinal animals were prepared by transecting the cord at C_1. Respiration was maintained in decerebrate and spinal animals by cut-off valve respiratory pump (18/min at 10 lb/sq in). Pharmacological studies were performed 3 hours after ether anesthesia. Patellar reflex was elicited by mechanically stimulating patellar tendon with percussion hammer driven by 6/min counterclockwise motor. Bipolar platinum shielded electrodes attached to central stump of sectioned contralateral sciatic nerve were used. Stimulation (120 cps, 0.5 msec. and 0.1 to 1.0 V) was induced with Thomas square wave stimulator. Selection of frequency was based upon previous work(6) which showed that low frequencies (1 and 10/sec) produced inhibition, and high frequencies (60 and 120/sec) augmentation of patellar reflex. This augmentation was expressed as percent increase in height of knee jerk response during contralateral sciatic nerve stimulation in comparison to prestimulation response. Voltage required varied from animal to animal, but for each animal it was constant except after drug treatment. For greater constancy, the drug effect was determined by establishing minimal dose necessary to completely contain the patellar reflex response at prestimulation values with intensity of stimulation inducing 100% augmentation. Intravenous injection of small multiple doses (1 to 10 mg/kg) in saline for 30 minutes were used to find proper dose range. Afterwards, single doses were used for accurate measurements.

Results. Styramate in 9 high cervical spinal cats abolished SCF in doses of 15 to 40 mg/kg, average of 21.1 mg/kg. In no instance did a dose lower than 15 mg/kg produce complete depression of SCF. On the other hand, in 5 decerebrate cats, the dose required for same effect was less than 5 mg/kg (avg 2.8 mg/kg) whereas in 3 other cats the doses were 10 to 20 mg/kg (avg 13.3 mg/kg). Thus, total average for the 8 cats was 5.3 mg/kg. To establish whether or not the greater susceptibility of the decerebrate animal to styramate is due to nature of animal preparation, we tested other polysynaptic blocking agents. Meprobamate in 4 spinal cats abolished the SCF at doses of 15 to 40 mg/kg with average of 35 mg/kg. In 6 decerebrate cats, meprobamate was effective in blocking SCF at similar dose; namely, 20 to 40 mg/kg, average of 27.5 mg/kg. Phenaglycadol and chlorpromazine tested on 14 cats showed that their action on the decerebrate animal is greater than on the spinal. Since population distribution of minimal (SCF) dose of styramate in decerebrate cats tended to the left, it may be inferred that the average dose of 5.3 mg/kg represents an upper limit. This makes comparison with average dose of 21.1 mg/kg, required in spinal animal in which the distribution was normal, much more pronounced. In contrast, meprobamate appeared approximately equal in effectiveness in decerebrate and spinal cats.

Height of patellar reflex response *per se* in spinal cats was increased by styramate in doses of 10 mg/kg or less and not modified further by doses sufficient to abolish SCF. Meprobamate, at dose levels required to abolish SCF, did not affect the patellar reflex in spinal cats. In contrast, both styramate and meprobamate in decerebrate cats did produce a transient decrease in height of patellar reflex response when doses needed to abolish SCF were employed. This effect on patellar reflex lasted only a few minutes (1 to 5), whereas the effect on SCF persisted 30 or more minutes.

Discussion. Two drugs, styramate and meprobamate, with central skeletal muscle relaxant properties appear to exert similar pharmacological effects on spinal polysynaptic reflex (SCF) in high cervical spinal cats. In decerebrate cats, styramate is about 4 times more potent than in spinal cats. In contrast,

meprobamate does not exhibit this difference in activity in the 2 animal preparations. This suggests that at the spinal level, both drugs exert similar net effects but at supraspinal levels their actions are different. Mephenesin, another relaxant drug, does act on supraspinal reflex transmissions(7) particularly the reticular facilitatory centers influencing spinal reflexes(8). It is likely that styramate which showed other similarities with mephenesin, may affect such areas. A generalization of our conclusions would lead to the possibility that centrally acting skeletal muscle relaxants may independently affect reticular facilitatory centers without influencing the ascending arousal system. Styramate and mephenesin are examples of this type of drug. On the other hand, there are relaxants *e.g.* pentobarbital, which at same dose level, depress both facilitatory and arousal systems. Meprobamate, another relaxant drug, does not affect the facilitatory system, assuming this is the correct interpretation of our data. Its action on arousal is controversial(9,10) but whether it depresses arousal or not, it still appears to belong to a class different from styramate and mephenesin. At this time, it can only be speculated whether differences encountered in our investigation between styramate and meprobamate are in some way related to their differences in anticonvulsant activity. Styramate in mice is much more effective in preventing extensor tonus spasm of hind leg following electroshock, Metrazol and strychnine induced seizures than meprobamate(1).

Summary. Effects of styramate and meprobamate upon spinal polysynaptic (SCF) and monosynaptic (patellar) reflexes in decerebrate and spinal cats were investigated. Both drugs selectively depressed the spinal polysynaptic reflex, however styramate was much more potent in decerebrate than in spinal cats. The significance of this selective effect was discussed in relation to other actions of muscle relaxants.

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Intracerebral Passage of Sarcoma 180 in Mice and Hamsters.* (24901)

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A number of investigators have transplanted neoplasms, including those of human origin, into brains of laboratory animals. Early literature has been reviewed by Sailer (1) and Vazquez-Lopez(2). Recently other investigators(3-7) have used intracerebral (IC) route for biologic studies of various experimental neoplasms. Sarcoma 180 is an easily transplantable murine tumor, growing well in any strain of mice(8) and has also been adapted to growth in tissue culture (TC) (9). However, despite these facts it has not been previously passed intracerebrally. It seemed probable therefore that

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