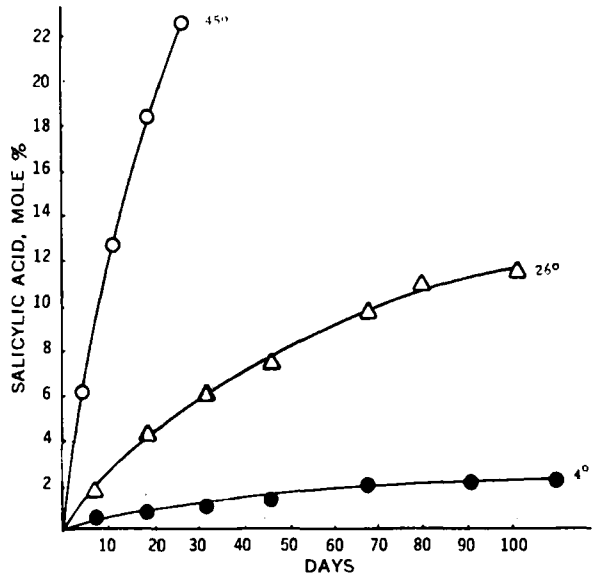

Figure 2—*Mole percent decomposition of aspirin with time in a polyethylene glycol base containing 1% citric acid.*

The effect of 10% tartaric acid on aspirin decomposition in this base was studied for a limited period, and the results were similar to those obtained with 5 and 10% citric acid for a comparable time. The percent decomposition at 26° was slightly lower with tartaric acid, but little difference was noted between these similar acids at 45 and 4°.

CONCLUSIONS

Certain substances can be added to a polyethylene glycol-type aspirin suppository mixture without appreciably changing its properties. Of the several additives studied, citric and tartaric acids inhibited decomposition. A 5% concentration of citric acid appeared to be the optimum concentration necessary to hinder decomposition. The importance of refrigeration at 4° on decomposition of aspirin in polyethylene glycol was evident.

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Biological Activity of Pentobarbital Metabolites

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Abstract □ The (1′*S*,3′*R*)-, (1′*S*,3′*S*)-, (1′*R*,3′*R*)-, and (1′*R*,3′*S*)-5-ethyl-5-(3′-hydroxy-1′-methylbutyl)barbituric acid metabolites of pentobarbital are relatively inactive and, therefore, cannot account for the differences in potency found between *R*(+)- and *S*(−)-pentobarbital. The (1′*S*,3′*R*)- and (1′*S*,3′*S*)-hydroxypentobarbital metabolites have very weak sedative activity when administered to CF-1 mice intravenously at an AD_{90} anesthetic dose of *RS*-pentobarbital.

Keyphrases □ Pentobarbital metabolites—anesthetic and behavioral activity in mice □ Barbituric acid metabolites, pentobarbital—anesthetic and behavioral activity in mice □ Anesthetic activity—pentobarbital metabolites, mice

The diastereoisomeric pairs of 5-ethyl-5-(3′-hydroxy-1′-methylbutyl)barbituric acid were first shown by Maynert and Dawson (1) to be biotransformation products of pentobarbital. More recently, the four optical isomers of 3′-hydroxypentobarbital were isolated in this laboratory from metabolism studies of optically pure *R*(+)- and *S*(−)-pentobarbital (2, 3). *R*(+)-Pentobarbital is metabolized to approximately equal amounts of the two hydroxylated metabolites in contrast to (*S*)-pentobarbital in which the ratio is 9:1. The *S*(−)-isomer is more toxic and potent as an anesthetic agent in mice than is the *R*(+)-isomer or the racemate (4). Maynert and Dawson (1) reported that the diastereoisomeric metabolites were inactive. Dickert *et al.* (5) found that (1′*RS*,3′*SR*)-3′-hydroxypentobarbital, prepared by synthetic methods, had very weak anticonvulsant activity and no anesthetic properties. Pharmacological differences in the enantiomers of pentobarbital and the respective ratios of their 3′-hydroxy enantiomers indicated the desirability of determining if some 3′-hydroxypentobarbital isomers were active and contributing to the differences in the activity of the pentobarbital stereoisomers.

EXPERIMENTAL

The structure and absolute stereochemical assignment of *R*- and *S*-pentobarbital as well as the four optically active 3′-hydroxypentobarbital metabolites were reported previously (6–9). The optical rotations and melting-point values for pentobarbital stereoisomers are: (*RS*), m.p. 129–130°; (*R*), $[\alpha]_D$ +13.1°, m.p. 121–122°; and (*S*), $[\alpha]_D$ −13.2°, m.p. 121–122°. For 3′-hydroxypentobarbital the values are: (1′*RS*,3′*RS*), m.p. 145–147°; (1′*RS*,3′*SR*), m.p. 191–192°; (1′*R*,3′*R*), $[\alpha]_D$ +12.8°, m.p. 170–176°; (1′*R*,3′*S*), $[\alpha]_D$ +30.5°, m.p. 202–205°; (1′*S*,3′*S*), $[\alpha]_D$ −15.3°, m.p. 181–183°; and (1′*S*,3′*R*), $[\alpha]_D$ −31.5°, m.p. 208–210°.

Biological activity was assessed by administering each epimer at the AD_{90} anesthetic dose of *RS*-pentobarbital to 10 CF-1 male mice.

Table I—Comparison of Activities of 3′-Hydroxy Metabolites to Pentobarbital

Compound	Number of Animals	Number Anesthesized	Average Duration of Anesthesia, min. ± *SD*	Time to Onset of Anesthesia, min.	Behavioral Alterations[a]
Pentobarbital					
RS	20	19	110 ± 24	3.1	+++
R(+)	10	1	35 ± —	10.0	+++
S(−)	10	10	114 ± 32	2.0	+++
3′-Hydroxypentobarbital					
1′*RS*,3′*RS*	10	0	— —	—	+
1′*RS*,3′*SR*	10	0	— —	—	+
1′*R*,3′*R*	10	0	— —	—	−
1′*R*,3′*S*	10	0	— —	—	−
1′*S*,3′*S*	10	0	— —	—	+
1′*S*,3′*R*	7	0	— —	—	+

[a] Indicates the magnitude: + = positive and − = none.

The behavior and time of onset and duration of anesthesia, measured as sleeping time, were observed. Each 3′-hydroxypentobarbital metabolite was further evaluated in five mice with a behavioral examination (10). This consisted of a systematic observation method for assessing the behavioral and physiological state of the mouse and its response to drugs. The pattern profile of various classes of pharmacological agents can be identified and differentiated, and the relative potency of members of the same class of drugs can be established. The dose volume used, 10 ml./kg. i.v., was administered *via* the tail vein.

The free acid was dissolved in 0.1 *M* aqueous sodium hydroxide to give the monosodium salt and diluted to appropriate volume with physiological saline.

RESULTS

The comparison of activity for *R*(+)- and *S*(−)-pentobarbital and the six 3′-hydroxypentobarbital derivatives of *RS*-pentobarbital is shown in Table I. Each compound was administered at 40 mg./kg. i.v.

The potency difference between the *R*(+)- and *S*(−)-pentobarbital is shown by the relative number of mice exhibiting sleeping by loss of the righting reflex. There is a slightly faster onset of action with the *S*(−)-form than with racemic pentobarbital. All *R*(+)-treated mice were very sedated; two of these mice just failed to meet the sleeping criterion of complete loss of the righting reflex for at least 1 min. None of the mice administered a 3′-hydroxypentobarbital metabolite was close to the sleeping criterion. However, the animals treated with the two racemic isomers as well as the two optically active metabolites having the 1′*S* configuration were sedated for approximately 1 hr. By using the screening test of the mouse's behavioral and physiological state, mice treated with 1′*RS*, 3′*RS*, 3′*SR*; and 1′*S*,3′*S* exhibited behavioral alterations of decreased awareness, increased positional passivity, and sedation. The onset of action is between 15 20 min. and lasts for approximately 1 hr. This screen is subjective, but the slight difference observed between the 3′-hydroxy metabolites indicates some variation in potency. The order of potency is: (1′*S*,3′*R*) > (1′*RS*,3′*SR*) > (1′*S*,3′*S*) > (1′*RS*,3′*RS*). The (1′*R*,3′*R*)- and (1′*R*,3′*S*)-isomers produced no behavioral or physiological alteration at 40 mg./kg.

DISCUSSION

The 3′-hydroxy metabolites are not pharmacologically inert as reported (11), but their activity is much less than that of the parent compound. When they are administered intravenously, some sedation occurs. Dickert *et al.* (5) reported that the (1′*RS*,3′*SR*)-isomer, given orally to mice at 1 g./kg., did not produce anesthesia or ataxia. They did, however, observe weak anticonvulsant activity. The ED_{50} in the maximal electroshock test was observed at 310 mg./kg. Their results (5) and the data reported here indicate that the activity profile of the 3′-hydroxybarbiturates is not identical to that of their parent compound. The difference found in the potency of *R*(+)- and *S*(−)-pentobarbital cannot be ascribed to differences in accumulations of these metabolites. Their potency difference could be due to differences in the rate of metabolism, binding properties, or relative penetration capacity to the receptor sites as well as other factors.

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