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The Pharmacological Relationship of Isomeric Barbituric Acid Derivatives*

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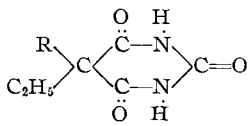
In a previous communication (1), it was observed that there is obvious relationship between the pharmacological action and the chemical structure of certain barbituric acid derivatives. In the primary or secondary alkyl substituted compounds, with an increase in the number of C-atoms in the alkyl group, both the minimal anesthetic dose (M. A. D.) and the minimal lethal dose (M. L. D.) grow relatively smaller, but when the alkyl radical is longer than 5 C-atoms, the amount required to produce anesthesia or death in rats again increases. As the alkyl chain lengthens, the therapeutic index, or ratio between M. L. D. and M. A. D., appears to be gradually greater. The duration of action becomes shorter as the alkyl chain lengthens to 6 C-atoms in the primary, and to 7 C-atoms in the secondary alkyl substituted derivatives, but when the alkyl radical is longer than 6 C-atoms (primary alkyls) or 7 C-atoms (secondary alkyls), the duration of action in rats again increases. More recently, it has been reported that the substitution of a methyl, ethyl or an allyl group on the nitrogen in place of the hydrogen (nitrogen alkyl substituted barbituric acid derivatives) (2), the substitution of an unsaturated alkyl radical (allyl and methallyl [2-methyl-allyl]) or a crotyl (3-methyl-allyl) (3,4) on one of the 5,5-positions, or a sulfur atom (5) in place of the oxygen on the 2 C-atom obviously reduces the duration of action. This shorter duration of action is independent of the quantity of drug administered.

EXPERIMENTAL

The present investigation deals with the study of several series of isomeric substituted barbituric acid derivatives, synthesized by Shonle and his associates of our organic chemical department, with the general formula:



wherein R may be alkyl radicals with 3 to 9 C-atoms.

Albino rats weighing 81 to 120 Gm. (average, 98 Gm.) and New Zealand rabbits weighing 1450 to 2150 Gm. (average, 1750 Gm.) were used in this study. Solutions of the sodium salts of the compounds were injected intraperitoneally in rats and by vein in rabbits. The M. A. D., duration of action and the M. L. D. were determined by using 5 or more animals for each dose level. From these data the median anesthetic dose (A. $D_{50} \pm$ S. E.) and the median lethal dose (L. $D_{50} \pm$ S. E.) were computed according to the formula of Bliss (6). During the tests, the animals were kept in a constant temperature room (29.4° to 32.2° C.).

* Presented before the Scientific Section of the A. PH. A., Atlanta meeting, 1939.

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Table I.—Comparison of the Pharmacological Action of Two Isomeric Propyl Substituted Barbituric Acid Derivatives, Injected Intraperitoneally

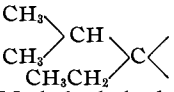
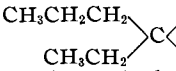
Number	Isomeric Propyl Substituted Barbituric Acid Derivatives	Number of Rats	Observed, mg. per Kg. M.A.D.	Observed, mg. per Kg. M.L.D.	Average Duration of M.A.D., Min.	Computed, mg. per Kg. A. D_{50} ± S. E.	Computed, mg. per Kg. L. D_{50} ± S. E.
1	 1-Methyl-ethyl-ethyl-	27	170	220	1520	161.3 ± 2.4	200.5 ± 12.2
2	 Normal-propyl-ethyl-	49	150	210	1140	141.1 ± 5.3	204.7 ± 3.6

Table II.—Comparison of the Pharmacological Action of Three Isomeric Butyl Substituted Barbituric Acid Derivatives, Injected Intraperitoneally

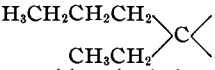
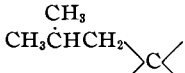
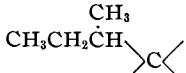
Number	Isomeric Butyl Substituted Barbituric Acid Derivatives	Number of Rats	Observed, mg. per Kg. M.A.D.	Observed, mg. per Kg. M.L.D.	Average Duration of M.A.D., Min.	Computed, mg. per Kg. A. D_{50} ± S. E.	Computed, mg. per Kg. L. D_{50} ± S. E.
1	 Normal-butyl-ethyl	40	80	200	450	74.46 ± 2.92	190.5 ± 3.3
2	 2-Methyl-propyl-ethyl-	35	120	220	540	112.4 ± 3.6	212.6 ± 3.5
3	 1-Methyl-propyl-ethyl-	35	60	130	600	52.7 ± 2.57	122.8 ± 2.7

Table III.—Comparison of the Pharmacological Action of Five Isomeric Amyl or Pentyl Substituted Barbituric Acid Derivatives, Injected Intraperitoneally

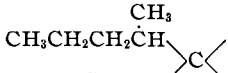
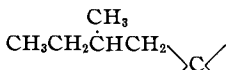
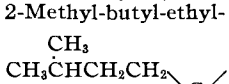
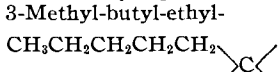
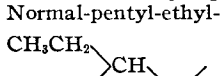
Number	Isomeric Pentyl Substituted Barbituric Acid Derivatives	Number of Rats	Observed, mg. per Kg. M.A.D.	Observed, mg. per Kg. M.L.D.	Average Duration of M.A.D., Min.	Computed, mg. per Kg. A. D_{50} ± S. E.	Computed, mg. per Kg. L. D_{50} ± S. E.
1	 1-Methyl-butyl-ethyl-	75	50	120	180	42.52 ± 2.64	119.1 ± 3.7
2	 2-Methyl-butyl-ethyl-	48	80	230	190	74.54 ± 3.02	222.7 ± 3.5
3	 3-Methyl-butyl-ethyl-	55	85	210	200	80.26 ± 1.58	204.9 ± 3.2
4	 Normal-pentyl-ethyl-	50	70	210	220	62.75 ± 2.62	202.6 ± 3.6
5	 1-Ethyl-propyl-ethyl-	54	60	100	300	56.53 ± 2.21	94.91 ± 4.69

Table IV.—Comparison of the Pharmacological Action of Four Isomeric Pentyl Substituted Thiobarbituric Acid Derivatives, Injected Intravenously

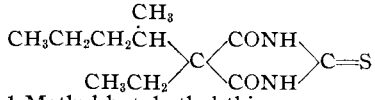
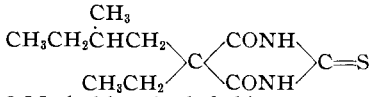
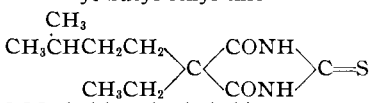
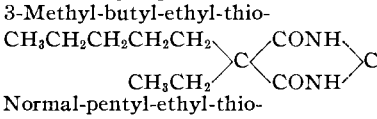
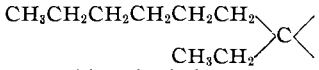
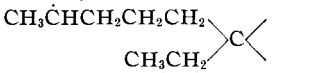
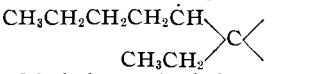
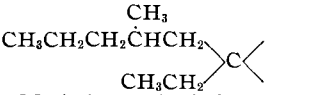
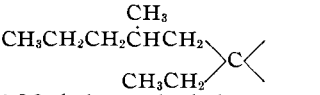
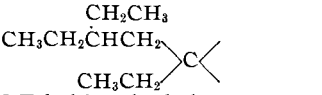
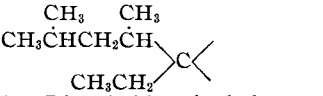
Number	Isomeric Pentyl Substituted Thiobarbituric Acid Derivatives	Number of Rabbits	Observed, mg. per Kg. M.A.D.	M.L.D.	Average Duration of M.A.D., Min.	Computed, mg. per Kg. A. D_{50} ± S. E.	L. D_{50} ± S. E.
1	 1-Methyl-butyl-ethyl-thio-	63	20	35	25	18.59 ± 0.68	28.41 ± 2.34
2	 2-Methyl-butyl-ethyl-thio-	43	30	55	32	26.32 ± 1.31	51.40 ± 1.99
3	 3-Methyl-butyl-ethyl-thio-	45	35	65	38	32.24 ± 1.48	61.22 ± 1.79
4	 Normal-pentyl-ethyl-thio-	76	40	70	42	34.96 ± 1.81	65.24 ± 1.59

Table V.—Comparison of the Pharmacological Actiou of Seven Isomeric Hexyl Substituted Barbituric Acid Derivatives, Injected Intraperitoneally

Number	Isomeric Hexyl Substituted Barbituric Acid Derivatives	Number of Rats	Observed, mg. per Kg. M.A.D.	M.L.D.	Average Duration of M.A.D., Min.	Computed, mg. per Kg. A. D_{50} ± S. E.	L. D_{50} ± S. E.
1	 Normal-hexyl-ethyl-	66	100	250	45	88.51 ± 3.21	242.9 ± 6.7
2	 4-Methyl-pentyl-ethyl-	45	120	400	60	109.6 ± 6.80	381.4 ± 8.9
3	 1-Methyl-pentyl-ethyl-	54	60	130	90	54.44 ± 3.58	127.2 ± 3.9
4 Low m. p.	 2-Methyl-pentyl-ethyl-	40	70	180	105	64.52 ± 3.03	167.0 ± 3.9
5 High m. p.	 2-Methyl-pentyl-ethyl	40	70	180	210	62.73 ± 2.64	161.5 ± 6.2
6	 2-Ethyl-butyl-ethyl-	54	100	170	300	70.18 ± 3.20	162.9 ± 2.7
7	 1, 3-Dimethyl-butyl-ethyl-	95	*	20	*	.....	....

* Convulsions.

Table VI.—Comparison of the Pharmacological Action of Five Isomeric Heptyl Substituted Barbituric Acid Derivatives, Injected Intraperitoneally

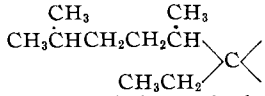
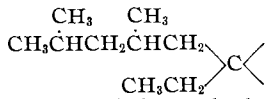
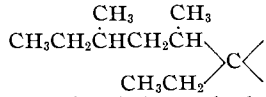
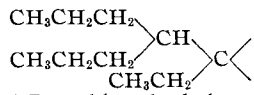
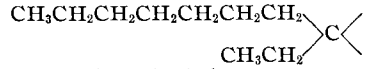
Number	Isomeric Heptyl Substituted Barbituric Acid Derivatives	Number of Rats	Observed, mg. per Kg. M.A.D.	M.L.D.	Average Duration of M.A.D., Min.	Computed, mg. per Kg. A. D_{50} ± S. E.	L. D_{50} ± S. E.
1	 1, 4-Dimethyl-pentyl-ethyl-	46	80	240	50	72.09 ± 3.62	235.4 ± 4.60
2	 2, 4-Dimethyl-pentyl-ethyl-	45	70	140	54	64.46 ± 2.98	138.7 ± 3.20
3	 1, 3-Dimethyl-pentyl-ethyl-	105	70	200	74	66.70 ± 3.92	190.6 ± 3.20
4	 1-Propyl-butyl-ethyl-	107	30	65	81	26.31 ± 1.31	60.54 ± 1.98
5	 Normal-heptyl-ethyl-	65	120	300	120	114.7 ± 3.0	290.4 ± 9.2

Table VII.—Comparison of the Pharmacological Action of Five Isomeric Octyl Substituted Barbituric Acid Derivatives, Injected Intraperitoneally

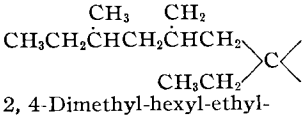
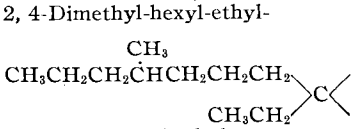
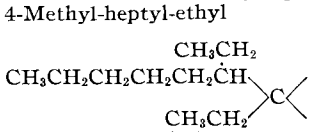
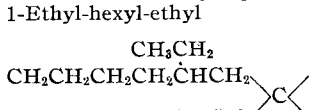
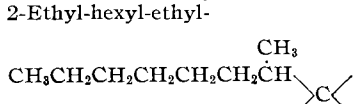
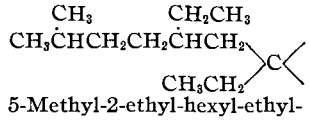
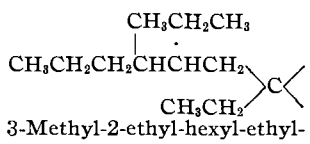
Number	Isomeric Octyl Substituted Barbituric Acid Derivatives	Number of Rats	Observed, mg. per Kg. M.A.D.	M.L.D.	Average Duration of M.A.D., Min.	Computed, mg. per Kg. A. D_{50} ± S. E.	L. D_{50} ± S. E.
1	 2, 4-Dimethyl-hexyl-ethyl-	54	80	230	60	71.99 ± 3.92	222.7 ± 3.60
2	 4-Methyl-heptyl-ethyl	60	80	230	60	72.80 ± 2.62	225.3 ± 4.60
3	 1-Ethyl-hexyl-ethyl	55	80	230	60	74.71 ± 4.74	226.7 ± 2.70
4	 2-Ethyl-hexyl-ethyl-	42	80	220	75	72.76 ± 2.63	222.7 ± 3.60
5	 1-Methyl-heptyl-ethyl-	39	120	220	150	116.5 ± 2.60	210.5 ± 3.20

Table VIII.—Comparison of the Pharmacological Action of Two Isomeric Nonyl Substituted Barbituric Acid Derivatives, Injected Intraperitoneally

Number	Isomeric Nonyl Substituted Barbituric Acid Derivatives	Number of Rats	Observed, mg. per Kg. M.A.D.	Observed, mg. per Kg. M.L.D.	Average Duration of M.A.D., Min.	Computed, mg. per Kg. A. D_{50} ± S. E.	Computed, mg. per Kg. L. D_{50} ± S. E.
1	 5-Methyl-2-ethyl-hexyl-ethyl-	54	140	340	120	134.6 ± 5.6	321.3 ± 6.40
2	 3-Methyl-2-ethyl-hexyl-ethyl-	50	160	400	240	151.1 ± 3.90	375.8 ± 10.60

In the propyl substituted isomers (Table I), the isopropyl derivative (1-methyl-ethyl-ethyl barbituric acid) has a longer duration of action than the normal-propyl-ethyl barbituric acid. The M. A. D. is slightly larger, but the therapeutic index is approximately the same for both isomers. Normal-butyl-ethyl barbituric acid (butyl isomers, Table II) with a M. A. D. of 80 mg. per Kg. has a duration of action of 450 minutes. The M. A. D. of 2-methyl-propyl-ethyl barbituric acid is 120 mg. per Kg. and the duration of action 540 minutes. For the secondary butyl (1-methyl-propyl-ethyl barbituric acid), the duration of action is 600 minutes and the M. A. D. 60 mg. per Kg. Thus, in the propyl and butyl isomeric barbituric acid derivatives, the secondary alkyl substituted compounds have a longer duration of action than their corresponding isomeric normal alkyl substituted derivatives. This variation in duration of action is independent of the amount necessary to produce anesthesia.

As summarized in Tables III and IV, the amyl or pentyl substituted isomeric compounds and the pentyl substituted isomeric thiobarbituric acid derivatives show a more definite physiological relationship to their chemical structure. The secondary pentyl (1-methylbutyl) substituted derivatives (compound No. 1 in Tables III and IV) have the smallest M. A. D. and the shortest duration of action of any of the pentyl isomers. As the methyl radical in the alkyl chain transfers from the 1-position (1-methyl-butyl) to the 2-position (2-methyl-butyl), or to the 3-position (3-methyl-butyl), and finally to the normal pentyl, the dosage or number of mg. per Kg. to produce anesthesia (M. A. D.) increases and the duration of action for each compound gradually becomes longer. Compound No. 5 in Table III, 1-ethyl-propyl-ethyl barbituric acid, has the longest duration of action of any of the pentyl isomers.

Of the 6 hexyl isomers (Table V), the normal hexyl derivative has the shortest duration of action. Derivatives Nos. 4 and 5 (2-methyl-pentyl) are optical isomers. Both compounds have the same M. A. D. (70 mg. per Kg.), but the duration of action of No. 5 is twice as long as that of No. 4. Isomer No. 6 (2-ethyl-butyl-ethyl) with a M. A. D. of 100 mg. per Kg. (100 mg. per Kg. is also the M. A. D. for isomer No. 1, normal hexyl) has a duration of action approximately six times longer than the normal hexyl substituted isomer. Thus, the duration of action of the hexyl isomers is independent of the size of the dose required to produce anesthesia. Curiously enough and unpredictable from a chemical structure point of view, as previously reported by Swanson (1), Knoefel (7) and Swanson and Chen (8), compound No. 7 (1,3-dimethyl-butyl-ethyl barbituric acid) is a powerful convulsant to warm-blooded animals but a true hypnotic to amphibians frogs and toads).

As shown in Tables VI and VII, the 5 heptyl and the 5 octyl isomers show distinct differences in their duration of action. In the heptyl series of isomers, the normal heptyl derivative has the longest duration of action, but in the octyl series the secondary octyl (1-methyl-heptyl) has the longest duration of action. Thus, in these two series of isomers, it is difficult to observe any pharmacological relationship with the chemical structure.

In the 2 nonyl isomers (Table VIII), the M. A. D. is approximately the same for both compounds, but there is a distinct difference in their duration of action. The 3-methyl-2-ethyl-hexyl derivative has a duration of action that is twice as long as the 5-methyl-2-ethyl-hexyl substituted compound. Thus, the chemical structural difference of the methyl radical from the 3-position in the alkyl chain (3-methyl-5-ethyl-hexyl) to the 5-position (5-methyl-2-ethyl-hexyl) will double the length of duration of action without much change in the dosage (M. A. D.).

SUMMARY

1. Several series of isomeric barbituric acid derivatives have been studied.

2. The minimal anesthetic dose, minimal lethal dose and the duration of action of the minimal anesthetic dose vary considerably among isomers. The duration of action is

independent of the quantity of the drug administered.

REFERENCES

(1) Swanson, *Proc. Soc. Exptl. Biol. Med.*, 31 (1933–1934), 961 and 963.

(2) Swanson, JOUR. A. PH. A., 25 (1936), 858.

(3) Swanson and Fry, *Ibid.*, 26 (1937), 317.

(4) Swanson and Fry, *Ibid.*, 27 (1938), 776.

(5) Tabern and Volwiller, *J. Am. Chem. Soc.*, 57 (1935), 1961.

(6) Bliss, *Quart. J. Pharm. Pharmacol.*, 11 (1938), 192.

(7) Knoefel, *J. Pharmacol.*, 57 (1936), 130.

(8) Swanson and Chen, *Quart. J. Pharm. Pharmacol.*, 12 (1939), 657.

The Preparation and Study of Silver Antiseptics with and without Ephedrine*

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The therapeutic effect of silver has been known for many years. Silver in protein combinations finds wide use in modern therapy in the treatment of infections of the eye, nose and urinary tract. These commercial preparations of silver are made in protein combination in order to obtain stable preparations with the desired disinfecting properties of the silver salts but without their irritant action. Most of the preparations are inferior to silver nitrate so far as germicidal activity is concerned.

The mild and strong protein silver preparations of the U. S. P. XI are examples of typical protein silver combinations. However, they, along with practically all other similar preparations, possess certain characteristics which are undesirable. The most important of these are: (1) their physical appearance, (2) their staining effect on the skin and clothing, (3) their instability and (4) their incompatibility with the alkaloid, ephedrine.

Solutions of mild and strong protein silver should be freshly prepared before use because, according to certain investigators, they lose some of their antiseptic activity and sometimes become irritant on standing.

At times physicians desire to prescribe for use in the nasal passages a mixture containing both silver and ephedrine, the former for its antiseptic effect and the latter for its shrinking action on the mucous lining of the nose. Combinations of alkaloids with silver in solution are unstable, particularly if the solution tends toward alkalinity, the silver being reduced. Hence the protein silver preparations, which are slightly alkaline in reaction, cannot be dispensed in combination with ephedrine.

It was for the purpose of preparing an effective silver antiseptic which would be free from the objectionable features of the protein silver medicaments and of the nitrate that this investigation was undertaken.

A portion of the research was concerned with the testing of the antiseptic potency of the silver medicaments prepared. The test employed was one used by the Food and Drug Administration, namely, the agar plate penetration method, in which the zone of inhibition of growth of organisms (from a standard culture of *Staphylococcus aureus*) immediately surrounding the antiseptic on a seeded agar plate is measured. The sodium chloride normally added to the nutrient agar medium was omitted when tests were performed on the organic silver preparations which yielded ionic silver in solution, in order to prevent the formation of slightly ionized silver chloride.

EXPERIMENTAL

PART ONE

Hydrosols of silver were prepared by four different methods and penetration tests performed. The results were not particularly significant because of variance in size of the particles.

Fine gelatin-protected dispersions of the following silver salts were prepared and incorporated into ointment form:

1. Silver chloride
2. Silver chromate
3. Silver thiocyanate
4. Silver iodide

* An abstract of a thesis submitted to the Faculty of Purdue University by Abraham Slesser in partial fulfilment of the requirements for the degree of Doctor of Philosophy, August 1939. An extensive bibliography accompanies the original thesis.

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