An Evaluation of the Analgetic Activity of the Dromoran Isomers. (19936)

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The relation between optical asymmetry and analgetic action has been investigated utilizing the optical isomers of methadone and some of its analogues. These studies have been reviewed by Chen(1). Denton and Beecher(2) carried out studies on human subjects, and concluded that d-methadone was inactive analgetically and d-isomethadone possessed an anti-analgetic action.

With the availability of the optical isomers of Dromoran, it was of interest to determine the relative analgetic activity of this pair of stereoisomers and the effect of d-Dromoran on morphine analgesia. The analgetic activity and the pharmacology of Dromoran have been described by Randall and Lehman(3), Gross, *et al.*(4), and Fromherz(5). The lack of addictive properties of d-Dromoran has been established by Isbell, *et al.*(6).

Methods. The effects of drug treatments were determined on five trained normal human subjects, using the Hardy-Wolff-Goodell technic. Pain thresholds were determined at 30 minute intervals until a maximal effect had occurred. The minimal intensity increment that the subjects were required to distinguish was equivalent to a 2.2% rise in pain threshold. The drugs dissolved in physiologic saline

TABLE I. Summary of Data.

Treatment	l-Dromoran tartrate	d-Dromoran "	d-Dromoran " and l-Dromoran "	dl-Dromoran hydrobromide	d-Dromoran tartrate	d-Dromoran " 30 min. before l-Dromoran tartrate	d-Dromoran " 30 min. before l-Dromoran tartrate	Saline placebo	Morphine sulfate	d-Dromoran tartrate and morphine sulfate	d-Dromoran tartrate 30 min. before morphine sulfate
mg	.5	.5	.5 .5	.8	1.0	.5 .5	1.0 .5		5	1 5	1 5
	(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)	(11)
n	12	11	10	11	4	10	5	20	10	12	5
$\bar{x}$	10.9	.8	7.7	6.2	5.5	1.5	2.6	.09	8.1	4.2	3.1
$V_{\bar{x}}$	.61	.11	9.33	.25	.40	.54	.19	.06	1.08	.28	.29
$s_{\bar{x}}$	.78	.33	.57	.5	.63	.74	.04	.25	1.04	.53	.54

TABLE II. Statistical Analysis.*

Between treatments	1 vs. 3	2 vs. 5	2 vs. 8	3 vs. 4	3 vs. 6	4 vs. 6	9 vs. 10	10 vs. 11	5 vs. 8	5 vs. 7	5 vs. 10	5 vs. 11
d	3.2	.7	.1	1.5	2.2	.7	3.9	1.1	.6	1.1	2.7	1.6
V_d	.72	.65	.17	.58	.73	.65	1.36	.57	.60	.73	.82	.83
s_d	.85	.81	.41	.76	.85	.81	1.17	.75	.77	.85	.91	.91
"t"	3.76	.86	.24	1.97	2.59	.86	3.33	1.47	.78	1.29	2.97	1.76
N	20	19	29	19	12	13	20	15	28	13	20	13
P	.01–.001	.50–.30	.90–.80	.10–.05	.05–.02	.50–.30	.01–.001	.20–.10	.50–.30	.30–.20	.02–.01	.20–.10

* d = Difference between means. V_d = Variance. s_d = Stand. dev. N = No. of degrees of freedom. P = Probability.

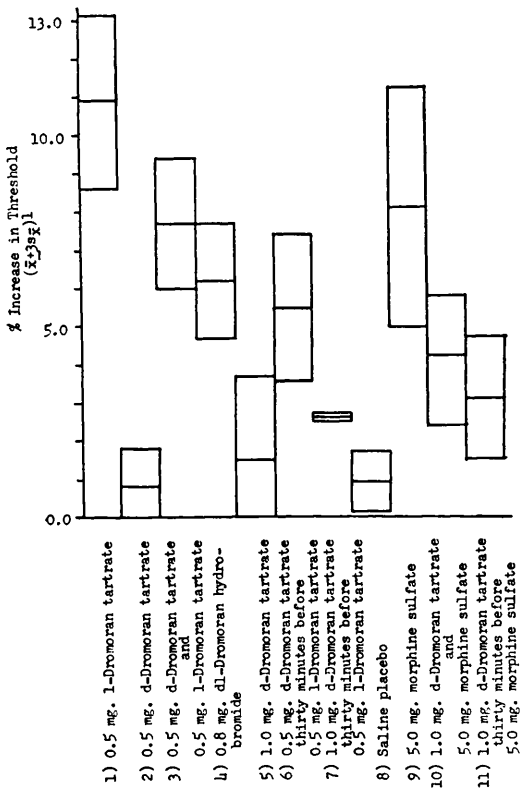

Fig. 1

Comparison of effects of drug treatment on the pain threshold.

[1]The line between each pair of blocks represents the mean ($\bar{x}$) and the distance to the other end of each block is equal to three standard deviations ($3s_{\bar{x}}$).

were administered by subcutaneous injection. Drug treatments were randomized and neither the subject nor operator knew what drug combination each had received.

Results and discussion. The results are summarized in Table I, and a comparison of the effects of the drug combination is made in Fig. 1. Where visual inspection was insufficient to determine the significance of the data, the Null Hypothesis and Student's "t" Test were applied. These comparisons are summarized in Table II.

The drug treatments* tested were (1) 0.5 mg l-Dromoran† tartrate, (2) 0.5 mg d-Dromoran tartrate,† (3) 0.5 mg d-Dromoran tartrate and 0.5 mg l-Dromoran tartrate, (4) 0.8 mg dl-Dromoran hydrobromide,† (5) 1.0 mg d-Dromoran tartrate, (6) 0.5 mg d-Dromoran tartrate 30 minutes before 0.5 mg l-Dromoran tartrate, (7) 1.0 mg d-Dromoran tartrate 30 minutes before 0.5 mg l-Dromoran tartrate, (8) saline placebo, (9) 5.0 mg morphine sulfate, (10) 1.0 mg d-Dromoran tartrate and 5.0 mg morphine sulfate, (11) 1.0 mg d-Dromoran tartrate thirty minutes before 5.0 mg morphine sulfate.

The results of treatments 1, 2, and 5 show that the analgetic activity of Dromoran lies in the l-isomer. The d-isomer is able to diminish the analgetic activity of the l-isomer (compare treatments 1, 3, 4, 6, and 7). The results of treatments 9, 10, and 11 show that d-Dromoran is an effective inhibitor of morphine analgesia. A 30 minute interval between administration of the d-Dromoran and l-Dromoran or morphine does not appear to be highly significant (compare treatments 3 and 6 and 10 and 11)

The marked inhibition of the analgesia produced by l-Dromoran and morphine by d-Dromoran is evidence in support of the contention that these agents possess a similar mechanism of action. An extension of this type of "crossover" comparison of the effects of inactive or less potent optical isomers of analgetic compounds on the analgetic activity of its own stereoisomer and the active or more potent stereoisomer of members of other analgetic series would be an effective test of the following accepted although unproven contention: the mechanisms of action of the so-called "morphine-type" analgetics are similar.

Summary. 1. The relative analgetic potencies of the optical isomers and racemate of Dromoran in normal human subjects were determined. 2. The inhibitory effect of d-Dromoran on l-Dromoran analgesia and morphine analgesia was noted.

* The doses of dl-, d-, and l-Dromoran represent equimolecular doses on the basis of each of the stereoisomers.

† The supplies of Dromoran and its stereoisomers were supplied by Hoffmann-LaRoche, Nutley, N. J.

1. Denton, J. E., and Beecher, H. K., *J. Am. Med. Assn.*, 1949, v141, 1146.

2. Chen, K. K., *Annals N. Y. Acad. Sci.*, 1948, v51, 83.

3. Randall, L. O., and Lehmann, G., *J. Pharm. and Exp. Therap.*, 1950, v99, 163.

4. Gross, E. G., Fraser, H. F., Brotman, M. A,

Nagyfy, S. F., Sawtelle, W. W., and Zager, L. L., *Fed. Proc.*, 1949, v8, 297.

5. Fromherz, K., *Arch. Int. de Pharm. et de Therap.*, 1951, vLXXXV, Fasc. III-IV, 387.

6. Isbell, H., and Fraser, H. F., *J. Pharm. and Exp. Therap.*, 1951, v103, 348.

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Size of the Spores of *Histoplasma capsulatum*. (19937)

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Cozad and Furcolow(1) recently reported the results of their measurements of a large number of spores from several strains of *Histoplasma capsulatum*. These authors reported that most of the spores were small and nontuberculated. Since these findings were contrary to the usual concepts, it appeared worthwhile to make an independent check of their observations.

Materials and methods. Cultures of 5 of the isolates of *Histoplasma* (Traub, Spitzli, Guest, Perdue, and White) studied by Cozad and Furcolow were obtained, and spore counts of each strain were made. The isolates were grown in screw-top test tubes on potato dextrose agar slants. They were incubated at room temperature for approximately 6 months. In each instance the entire culture was scraped from the slant and was macerated by grinding in a sterile tissue grinder. Microslide preparations were made from each of the suspensions using lactophenol without the dye. Semi-permanent mounts were made by ringing the coverslips with a preparation of paraffin and vaseline. Microscopical observations were made with the high power oil immersion lens. Three traverse sweeps across each slide preparation were followed with no attempt to select areas, but all of the spores were counted as they appeared in the fields of observation. Therefore, differences in technics of Cozad and Furcolow's studies as compared to ours were as follows: 1) The cultures were approximately 6 months old when studied in our laboratory; their measurements were made on 1 and 5 months old cultures. 2) Our measurements were made with a 95x high power oil immersion lens (approximate magnification 1200 diameters); theirs were made with a 44x high dry lens (approximate magnification 500 diameters). 3) In our studies the entire culture was macerated by grinding in a sterile glass tissue grinder and the mounts then prepared from the suspension; theirs were made from a selected area of the culture. We hoped to secure, in this manner, a more representative sampling of the entire colony than could be obtained by the method of Cozad and Furcolow. We measured 1000 spores from each strain; they measured 250. Aside from the specific differences numerated above, technics and methods of observations were the same.

Results and interpretations. Fig. 1 presents a comparison of the results obtained in our laboratory to those obtained for the same strains of *Histoplasma* by Cozad and Furcolow(1). Although the curves shown in Fig. 1 are similar, our measurements show a greater frequency of small spores and fewer numbers of tuberculated spores. Only 6.8% of the 5000 spores measured by us exceeded 4.8 μ in diameter, and only 3.4% were tuberculated. This compares with 31.1% larger than 4.8 μ by their measurement and 11.2% tuberculated. It is also interesting to note that the largest percentage of tuberculated spores observed by us was 8.7% in strain 5, whereas strains 14 and 1 showed more than 20% tuberculated spores according to their measurements.

The greatest difference in measurements of the same strain occurred with strain 14 where

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