ANALGESIC AND ANTIPYRETIC PROPERTIES OF SOME ASPIRIN DERIVATIVES

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Several derivatives of acetylsalicylic acid have been tested for analgesic and antipyretic properties in mice and rats. *o*-Diphenylacetoxy benzoic acid (*O*-(diphenylacetyl)salicylic acid; DPA) is a superior analgesic but an inferior antipyretic to aspirin. DPA has a low toxicity: it failed to show significant anti-inflammatory or tranquillising properties. In dogs it produced only slight and transient falls in blood pressure and there was no appreciable effect on the direct or reflex muscle preparation.

THE first synthetic ester of salicylic acid to be introduced into medicine was phenyl salicylate (Nencki, 1886). Later acetylsalicylic acid was prepared by Dreser (1899). The compound was named aspirin and has been perhaps the most successful synthetic drug in the whole field of medicinal chemistry. Since aspirin is one of the oldest remedies still occupying an important place in modern therapeutics, some of its analogues (Raczynski, 1943) were examined (see Table I).

METHODS

Male albino mice of the Swiss-Webster strain, male albino rats of the Harlan-Wistar strain, and adult mongrel dogs, of both sex, were used. Animals had food and water except during the period of testing. All drugs were given either as aqueous solutions or as suspension in 1 per cent cellulose (CMC-120, high viscosity, Hercules Powder Co.) gum solution.

Analgesic Experiments

A thermal method was used for determining analgesic activity in mice (Chen and Beckman, 1951). All drugs for analgesic testing were administered intragastrically (i.g.) except morphine sulphate, which was given intraperitoneally (i.p.). Controls of distilled water (i.p.) and morphine were run simultaneously. Six animals were used at each dose level. The ability of drugs to block hydrochloric acid-induced writhing in mice was also determined (Vander, Wende and Margolin, 1956, and Eckhardt, Cheplovitz, Lipo and Govier, 1958). Simultaneous controls with hydrochloric acid are necessary.

Antipyretic Experiments

The effect of compounds given i.g. on normal body temperature and yeast-induced fever (Maren, 1951) was evaluated in rats. For temperature determinations, thermistor probes were inserted rectally into male rats confined in wire mesh tubes. Two control temperature readings at 30 min. intervals were made before drug injection and served as controls for each group of four animals; saline controls were run simultaneously.

Temperature readings were made at hourly intervals. For experiments with fevered rats, control readings were taken before yeast injection (3 ml. of a 15 per cent brewer's yeast suspension in saline subcutaneously) and again before drug administration. The temperature of the room was $24 \pm 1°$.

Acute Toxicity

The i.p. and i.g. lethal doses for 50 per cent of mice were determined (Litchfield and Wilcoxon, 1949) for *o*-diphenylacetoxybenzoic acid (*O*-(diphenylacetyl) salicylic acid; DPA).

TABLE I

TOXICITIES AND BLOCKADE OF IRRITANT-INDUCED WRITHING IN MICE

Compound	Writhing in mice	
	Dose mg./kg.	No. writhing/No. tested
Hydrochloric acid	10	84/90
Acetylsalicylic acid (*o*-hydroxybenzoic acid) (Aspirin)	50	24/30
	100	11/20
	200	1/10
2-Acetoxy-4,5, dimethylbenzoic acid (I)	50	20/20
	100	19/20
2-diphenylacetoxybenzoic acid (*O*-(diphenylacetyl)salicylic acid; DPA)	50	9/10
	100	19/20
	200	13/20
Methyl 2-diphenylacetoxybenzoate (II)	200	9/10
Ethyl 2-diphenylacetoxybenzoate (III)	200	10/10
Pentyl 2-diphenylacetoxybenzoate (IV)	200	8/10
Isopentyl 2-diphenylacetoxybenzoate (V)	200	10/10
Phenyl 2-diphenylacetoxybenzoate (VI)	200	9/10
Benzyl 2-diphenylacetoxybenzoate (VII)	200	10/10
Methyl 2-dibenzylacetoxybenzoate (VIII)	200	9/10
Ethyl 2-dibenzylacetoxybenzoate (IX)	200	10/10

Other Studies

Tests for antigranulomatous activity were made in male rats (Meier, Schuler and Desaulles, 1950). Tranquillising activity was assessed in rats by the method of Hughes and Kopmann (1960). Mongrel dogs were used to evaluate the effects of DPA on cardiovascular and neuromuscular systems.

RESULTS

Only two compounds, aspirin and possibly DPA at the highest dose, blocked irritant-induced writhing (Table I); aspirin was the most effective compound by this test. When groups of animals were challenged at

various intervals after drug administration of a dose of 200 mg./kg. i.g., DPA had a peak activity of 30 min. whereas aspirin was equally effective over a period of 2 hr. Of the salicylate derivatives listed in Table I only DPA showed significant analgesic effect by the thermal method. It can be seen in Fig. 1 that DPA at 200 mg./kg. i.g. was approximately equivalent to morphine sulphate at 5 mg./kg. i.p. All reaction times greater than 5 sec. were significantly greater ($P < 0.05$) than control.

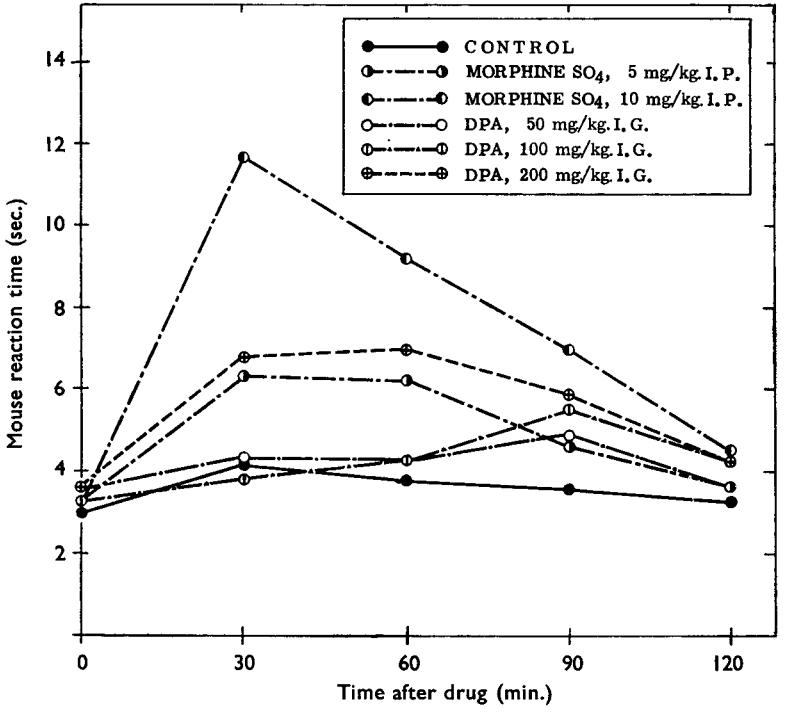

FIG. 1. Analgesic effects of morphine and DPA in mice.

Aspirin failed to show analgesia even at a dose of 400 mg./kg. i.g. DPA appeared to enhance the effects of morphine (Fig. 2) either when given in combination simultaneously or given separately 1 hr. apart. Aspirin used in combination with morphine did not show similar activity.

Antipyretic Activity

None of the compounds tested (DPA, II, VI, VIII) at 400 mg./kg. i.g. produced a decrease in normal body temperature of rats. However, DPA and aspirin were both effective in lowering the rectal temperature of rats with yeast-induced fever (Fig. 3).

Acute Toxicities

In mice the LD50 for DPA was 316(277·2–360·2) mg./kg. i.p. and 1,010 (878–1,162) mg./kg. i.g. Toxic signs appeared within 7 min.

by either route and consisted of tremors or shaking with arched back which proceeded to clonic convulsions and death within 30 min. to 24 hr. There was some moderate depression remaining at 24 hr.

Other Studies

DPA in doses of 50 and 200 mg./kg./day i.g. for 7 days failed to reduce the weights of granulomas in rats below those of controls.

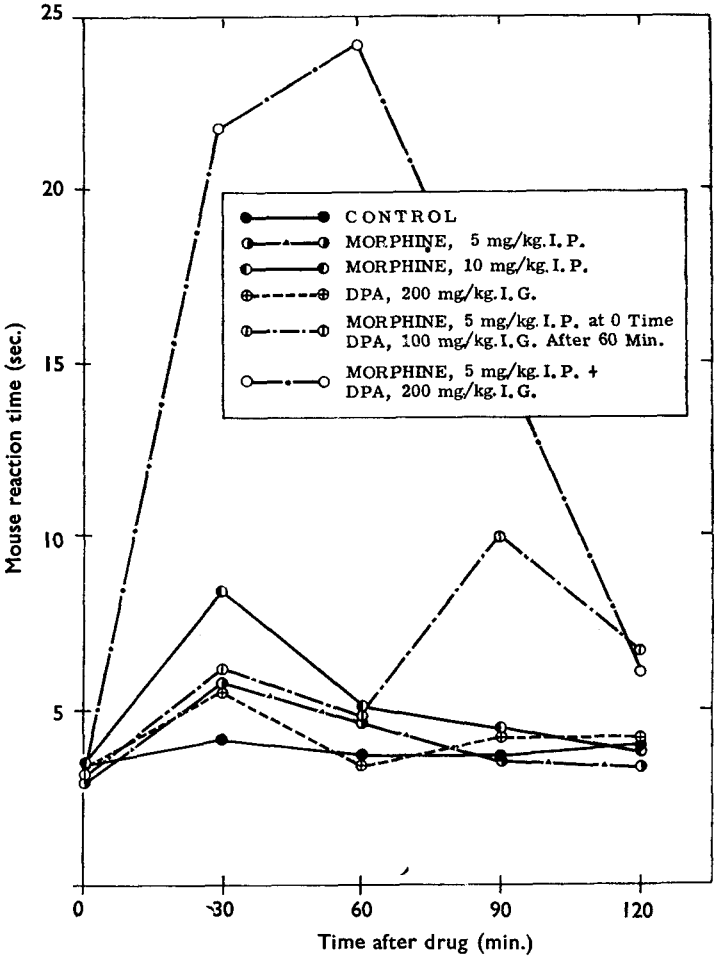

FIG. 2. Analgesic effects of morphine and DPA alone and in combination in mice.

DPA at 400 and 600 mg./kg. i.g. and morphine at 5 mg./kg. i.p. did not exhibit desirable tranquillising properties.

DPA in doses of 32 mg./kg. i.v. in dogs produced only slight and transient falls in blood pressure. Similarly there was no appreciable effect on the direct or reflex muscle preparation.

Discussion

Burger (1951) has noted the analgesic and antipyretic properties of several derivatives of salicylic acid were usually not as active as the parent compound. Similarly, the compounds reported in this study seem to follow this trend; the exception being DPA.

Most investigators agree that present day analgesic methods are inadequate for assessing the analgesic properties of aspirin; the writhing test being an exception to this general conclusion. However, the type of "pain" being measured by the writhing test is not completely understood.

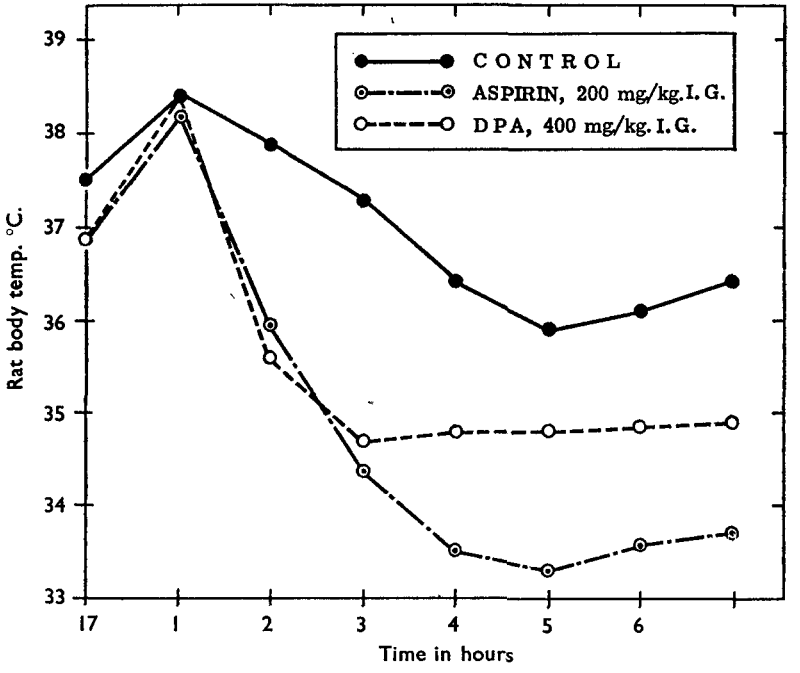

FIG. 3. Effect of aspirin and DPA on yeast-induced fever in rats.

It would seem logical, then, that when a derivative of aspirin is shown to possess analgesic properties by a thermal method, that compound might be a more potent analgesic than aspirin. DPA exhibited this ability in a dose of 200 mg./kg. i.g.; the analgesic effects were approximately equal to morphine at 5 mg./kg. i.p. (Fig. 1). Further, DPA added to, or enhanced, the analgesic properties of morphine sulphate when given simultaneously or separated by a 1-hr. interval (Fig. 2). Nevertheless, DPA was inferior to aspirin in its ability to counteract fevers in rats. Finally, DPA exhibited relatively little antigranuloma, or tranquillising activity in rats and, with dogs, there were negligible effects on blood pressure and direct and reflex muscle preparations The low toxicity exhibited suggests that it should be evaluated further.

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REFERENCES

Burger, A. (1951). *Medicinal Chemistry*, vol. 1, p. 201, New York: Interscience.
Chen, J. Y. P., and Beckman, H. (1951). *Science*, **113**, 631.
Dreser, H. (1899). *Arch. ges. Physiol.*, **76**, 306–318.
Eckhardt, E. T., Cheplovitz, F., Lipo, M., and Govier, W. M. (1958). *Proc. Soc. exp. Biol., N.Y.*, **98**, 186–188.
Hughes, F. W., and Kopmann, E. (1960). *Arch. int. Pharmacodyn.*, **126**, 158–170.
Litchfield, J. T., Jun., and Wilcoxon, F. (1949). *J. Pharmacol.*, **96**, 99–113.
Maren, T. H. (1951). *Ibid.*, **101**, 313–316.
Meier, R., Schuler, W., and Desaulles, P. (1950). *Experientia*, **6**, 469–471.
Nencki, M. (1886). *Arch. exp. Path. Pharmacol.*, **20**, 396.
Raczynski, W. (1943). Doctoral Thesis, Indiana University, Bloomington, Ind.
Vander Wende, C., and Margolin, S. (1956). *Fed. Proc.*, **15**, 494.