RESEARCH PAPERS

ANALGESIC PROPERTIES OF 4-ETHOXYCARBONYL-1-(2-HYDROXY-3-PHENOXYPROPYL) 4-PHENYLPIPERIDINE (B.D.H.200) AND SOME RELATED COMPOUNDS

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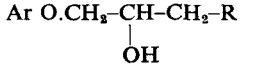
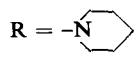
Twenty-one 3-aryloxy or 3-alkoxy-2-hydroxy-n-propyl derivatives of norpethidine were tested subcutaneously for analgesic activity in mice. Many of them are more potent than pethidine. The 2-hydroxy-3-phenoxypropyl derivative (B.D.H. 200) is at least three times more active than morphine and ten times more active than pethidine in this species with a therapeutic index slightly better than morphine and very much better than pethidine. The duration of analgesia is similar to morphine and pethidine and it is less constipating than pethidine. Its effects on respiration and the cardiovascular systems are counteracted by nalorphine. The decrease in activity after oral administration is probably due to a more rapid metabolic breakdown than poor absorption, as this decrease in activity can be modified by pretreatment with iproniazid and BAL while subcutaneous administration is unaffected.

SOME 3-aryloxy-2-hydroxypropylamines with weak analgesic properties were described by Beasley, Petrow and Stephenson[1] and having the general formula (I) where Ar = phenyl or substitued phenyl, and R an alkyl or dialkyl amino group.



(I)



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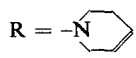
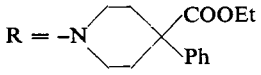


(III)

The analgesic activity was increased by replacing the amino group by cyclic structures such as piperidine (II), pyrrolidine and morpholine. The replacement of the piperidine group by Δ^3-piperideine (III), further enhanced the analgesic activity[2], and led to the synthesis of *N*-(2-hydroxy-3-*o*-toloxypropyl)-Δ^3 piperideine (Tolpronine) whose pharmacological properties were described by David, Leith-Ross and Vallance[3].

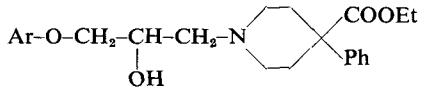
The study of the analgesic properties of structures related to (II) was further extended by preparing the 4-ethoxycarbonyl-4-phenyl derivatives (IV) of *N*-(3-aryloxy-2-hydroxypropyl)piperidine[4].



(IV)

These compounds are analogues of pethidine being substituted derivatifes of norpethidine where the *N*-methyl group has been substituted by the *N*-aryloxyhydroxypropyl group.

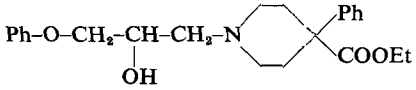
Pethidine



Series (IV)

Since the discovery, by Schaumann[5], of the analgesic properties of the 4-ethoxycarbonyl-4-phenyl-*N*-alkyl piperidines, *N*-methyl substitution has been regarded as essential for the analgesic activity in the pethidine molecule[6–8].

Some doubts on the validity of this conclusion have recently been raised by the discovery that some *N*-aralkyl analogues of pethidine show an analgesic activity superior to that of pethidine itself[9–15], as do the *N*-morpholinoethyl analogue[16,17], and the *N*-2-(2-hydroxyethoxy)ethyl analogue[18]. The results obtained with the *N*-aryloxyhydroxypropyl analogues of pethidine and particularly with 4-ethoxycarbonyl-1-(2-hydroxy-3-phenoxypropyl) 4-phenylpiperidine (B.D.H. 200) provide



further support for the view that *N*-methyl substitution in the pethidine molecule is not the best for analgesic activity. In the following experiments B.D.H. 200 was used as the hydrochloride or hypophosphite.

Physical Properties

The hydrochloride of B.D.H. 200 is a white crystalline compound with a molecular weight of 419·7. It has a melting point of 174·2–175°. It is sparingly soluble in water 0·5 per cent at 25°. A 0·1 per cent solution at 21·5° has a pH of 5·49.

B.D.H. 200 hypophosphite is a white crystalline compound with a molecular weight of 449·3. It has a melting point of 115·5–116·5°. It is soluble 4·9 per cent in water at 22°. A 1 per cent solution at 20° has a pH of 3·93.

Methods

Analgesic Activity in Mice

The subcutaneous and oral analgesic activities were estimated in male albino mice, weighing between 15 and 20 g., using Haffner's method as described by Bianchi and Franceschini[19]. The animals were fasted overnight before the oral tests but in the subcutaneous experiments were allowed free access to food and water. The sensitivity of each mouse was determined immediately before administration by placing a bulldog artery clip covered with catheter tubing to the base of the tail; only those mice making continuous attempts to remove the clip within 15 seconds were included in the experiments. The hydrochloride and hypophosphite were given in aqueous solutions, the volumes being adjusted to 0·5 ml./20g. weight. At 30, 60 and 90 minutes after administration the clip was applied to each mouse in turn. A positive analgesic response was recorded if no attempt was made to remove the clip at any one of the three observation

times. The ED50 and the activity ratios with fiducial limits (P = 0·05) were calculated using Litchfield and Wilcoxon's method[20].

Duration of Analgesic Action in Mice

This was estimated in sensitive male albino mice after subcutaneous or oral administration of a single submaximal analgesic dose. Tests for analgesia were made as before at 15 or 30 minute intervals for as long as analgesia was present. The duration of analgesic effect, the ET50, and the activity ratio, with fiducial limits (P = 0·05), were calculated according to Litchfield[21]. The compounds were given as aqueous solutions, the volumes being adjusted to 0·5 ml./20 g. weight.

Analgesic Activity in Rats

This was investigated to confirm that the analgesic activity of B.D.H. 200 hydrochloride was not confined to mice. Analgesia was tested using a thermal stimulus[22], 600 W, 165 V, 3·6 A applied from 1 to 1½ inches from the base of the tail, previously blackened with indian ink, for not more than 6 seconds.

The individual reaction time, that is the time taken for each animal to remove its tail from the region of the stimulus, was recorded by means of a stop watch. A period of training was carried out twice a day for 2 days before and once on the day of the experiment. Insensitive or hypersensitive rats were not used. Doses were administered subcutaneously and all volumes adjusted to 0·5 ml./200 g. weight. The thermal stimulus was applied at 30, 60 and 90 minutes following administration and those rats showing an increase of 2 seconds or more over their normal reaction time were regarded as showing analgesia.

Acute Toxicity

The subcutaneous and oral toxicities were estimated in male albino mice weighing between 15 and 20 g. each. The LD50, calculated from the seven days mortalities, the toxicity ratios, and their fiducial limits (P = 0·05), were estimated according to Litchfield and Wilcoxon's method[20].

The compounds were administered in distilled water and all volumes were adjusted to 0·5 ml./20 g. weight.

Effect on Blood Pressure and Respiration

The effects on blood pressure and respiration were determined in male rabbits weighing between 2·4 and 3 kg. and in male cats weighing between 2·4 and 4·3 kg.

The rabbits were anaesthetised with 2 g./kg. of urethane subcutaneously and the cats with 500 mg./kg. of urethane plus 50 mg./kg. of chloralose given intraperitoneally. The carotid blood pressure was recorded by means of a mercury manometer, and respiratory movements by means of a lever attached to a rubber tambour connected by a side arm directly to the trachea.

B.D.H. 200 was given subcutaneously to rabbits in 0·5 ml./kg. of normal saline and in cats through the cannulated femoral vein. The rabbits were

observed for 3 hours after administration and the percentage variation in respiration estimated at intervals. The cats were used chiefly in the experiments designed to estimate the antagonistic effect of nalorphine and B.D.H. 200.

TABLE I

THE SUBCUTANEOUS ED50, LD50, WITH FIDUCIAL LIMITS (P = 0·05), AND THERAPEUTIC INDICES IN MICE OF MORPHINE, PETHIDINE AND B.D.H. 200

Compound	No. of mice	ED 50 mg./kg.	No. of mice	LD 50 mg./kg.	Therapeutic index
Morphine hydrochloride	295	5·80 (4·83–6·96)	160	505·0 (459·0–555·5)	87·0
Pethidine hydrochloride	80	17·00 (11·48–25·16)	50	130·0 (97·7–172·9)	7·6
B.D.H. 200 hydrochloride	300	1·38 (1·16–1·62)	70	145·0 (117·9–178·3)	105·0
B.D.H. 200 hypophosphite	200	1·70 (1·33–2·16)	80	150·0 (127·1–177·0)	88·2

Constipating Effect

This was investigated by Lou's method[23] in unfasted mice weighing approximately 20 g. each. Groups of nine or ten mice were given three doses of B.D.H. 200 or pethidine hydrochloride subcutaneously and

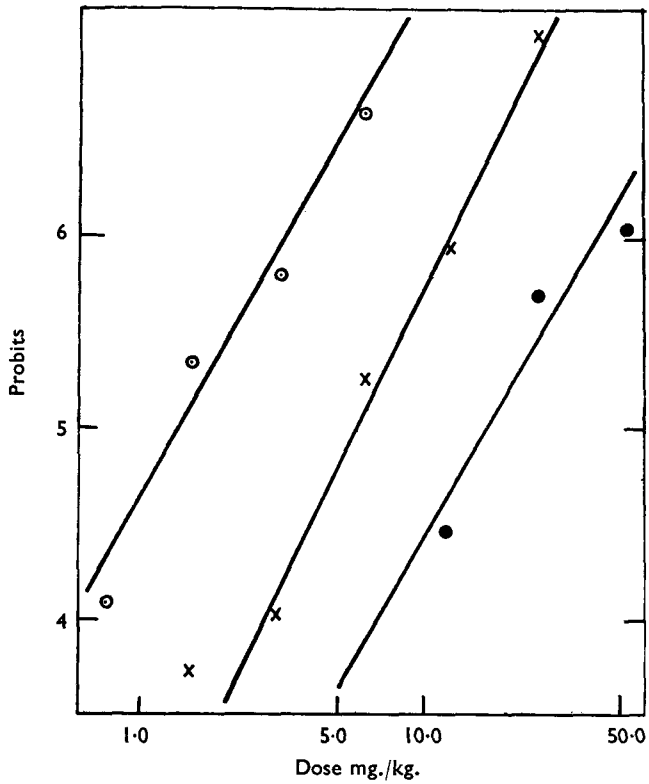


FIG. 1. The subcutaneous analgesic activity of morphine X, pethidine ● and B.D.H. 200 ⊙ in mice.

15 minutes later placed in separate compartments over a wire grid and faeces collected on blotting paper. The number of faecal pellets was counted at 8 and 24 hours and the mean number recorded. The mice were allowed free access to a paste of Rat Diet 41 and water.

RESULTS

Toxicity and Analgesic Properties

The subcutaneous toxicities, analgesic activities and therapeutic indices in male albino mice of morphine hydrochloride, pethidine hydrochloride, and the hydrochloride and hypophosphite of B.D.H. 200 are recorded in Table I.

The percentages of mice insensitive to the pressure stimulus after the administration of morphine, pethidine and B.D.H. 200 were converted

TABLE II

THE SUBCUTANEOUS ANALGESIC AND TOXICITY RATIOS OF B.D.H. 200, PETHIDINE AND MORPHINE IN MICE

Compound	Analgesic ratio (fiducial limits P = 0·05)	Acute toxicity ratio (fiducial limits P = 0·05)
Pethidine : Morphine	0·34 (0·22–0·53)	3·88 (2·87–5·24)
B.D.H. 200 HCl : Morphine	4·20 (3·28–5·37)	3·48 (2·78–4·35)
B.D.H. 200 hypophos : Morphine.	3·41 (2·52–4·60)	3·36 (2·77–4·06)
B.D.H. 200 HCl : Pethidine	12·31 (7·94–19·08)	0·89 (0·62–1·26)
B.D.H. 200 hypophos : Pethidine	10·00 (6·25–16·00)	0·86 (0·61–1·20)
B.D.H. 200 hypophos : B.D.H. 200 HCl	0·81 (0·62–1·04)	0·96 (0·74–1·25)

TABLE III

THE ORAL ED50, LD50 AND RELATIVE ACTIVITIES WITH FIDUCIAL LIMITS (P = 0·05) OF PETHIDINE AND B.D.H. 200 HYDROCHLORIDE IN MICE

Compound	No. of mice	ED 50 mg./kg.	No. of mice	LD 50 mg./kg.	Relative activity
Pethidine hydrochloride	80	27·00 (20·00–36·45)	30	230·0 (178·1–296·7)	1
B.D.H. 200 hydrochloride	100	48·00 (30·96–74·40)	40	419·0 (335·2–523·7)	0·56 (0·32–0·95)
Pethidine hydrochloride	50	28·00 (19·31–40·60)	—	—	1
B.D.H. 200 hydrochloride	50	53·00 (36·55–76·85)	—	—	0·52 (0·30–0·87)

into probits and plotted against log dose in Figure 1. The responses to the hydrochloride and hypophosphite were similar and therefore the latter was not plotted. The regression lines do not deviate significantly from parallelism and the activity ratios were estimated. The subcutaneous activity and toxicity ratios are recorded in Table II.

In this species, B.D.H. 200 is at least three times more active than morphine hydrochloride and ten times more active than pethidine hydrochloride. It is approximately three times more acutely toxic than

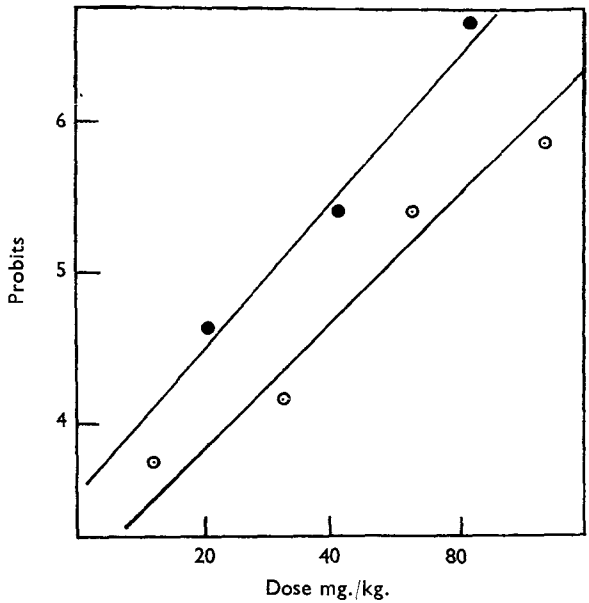


FIG. 2. The oral analgesic activity of pethidine ● and B.D.H. 200 ⊙ in mice.

morphine and has a similar toxicity to pethidine. Its therapeutic index is slightly better than morphine and much better than pethidine.

B.D.H. 200 also possesses oral analgesic properties in mice but there is a reduction in activity compared to subcutaneous administration. Table III and Figure 2 record the oral toxicity, the analgesic activity and ratio with pethidine hydrochloride.

TABLE IV

THE DURATION OF ANALGESIA (ET50) IN MICE AFTER SUBCUTANEOUS OR ORAL ADMINISTRATION OF MORPHINE, PETHIDINE AND B.D.H. 200

Compound	Route	Dose mg./kg.	No. showing analgesia at intervals (min.) 15	30	60	90	120	150	180	ET 50* min. (limits P = 0·05)	Significance of difference P = 0·05
Morphine hydrochloride	Subcut.	12·5		22/25	21/25	15/25	6/25	4/25	0/25	100·0 (86·9–115·0)	None
B.D.H. 200 hydrochloride	Subcut.	2·75		20/25	17/25	10/25	7/25	5/25	0/25	95·0 (81·2–111·1)	
Pethidine hydrochloride	Subcut.	35·0		21/25	10/25	3/25	0/25	0/25	0/25	59·0 (50·0–69·6)	None
B.D.H. 200 hydrochloride	Subcut.	3·5		23/25	16/25	7/25	3/25	1/25	0/25	73 (61·8–86·1)	
			15	30	45	60	75	90	120		
Pethidine hydrochloride	Orally	40·0	10/10	7/10	5/10	2/10	1/10	1/10	0/10	37·0 (26·4–51·8)	None
B.D.H. 200 hydrochloride	Orally	80·0	10/10	8/10	5/10	4/10	2/10	1/10	0/10	47·0 (34·0–64·8)	

* Calculated only from those showing analgesia.

The analgesic ratio of oral to subcutaneous administration is 1·6 for pethidine and 36·6 for B.D.H. 200. The toxicity ratio of oral and subcutaneous administration is 1·8 for pethidine and 2·9 for B.D.H. 200. The mean durations of analgesia of subcutaneous equi-effective doses of B.D.H. 200, morphine and pethidine are similar, but the dose of B.D.H.

TABLE V

THE NUMBER OF RATS DEVELOPING ANALGESIA AT INTERVALS FOLLOWING SUBCUTANEOUS ADMINISTRATION OF B.D.H. 200 HYDROCHLORIDE AND PETHIDINE HYDROCHLORIDE

Compound	Dose mg./kg.	No. developing analgesia at intervals (minutes) 30	60	90
B.D.H. 200 hydrochloride	0·5	2/4	1/4	0/4
	1·0	3/4	2/4	0/4
	2·0	4/4	4/4	0/4
Pethidine hydrochloride	5·0	0/4	0/4	1/4
	10·0	0/4	0/4	0/4
	20·0	0/4	0/4	1/4
Controls	—	0/4	0/4	0/4

200 is only one-quarter that of morphine and one-tenth that of pethidine. After oral administration of equi-effective doses of pethidine and B.D.H. 200 the durations of analgesia are similar but the dose of B.D.H. 200 was twice that of pethidine. Table IV records the results.

Analgesic Effect in Rats

The analgesic response in rats after subcutaneous injection of B.D.H. 200 and pethidine hydrochloride are recorded in Table V. These results demonstrate that B.D.H. 200 possesses analgesic properties in rats and is much more active than pethidine hydrochloride by the method used.

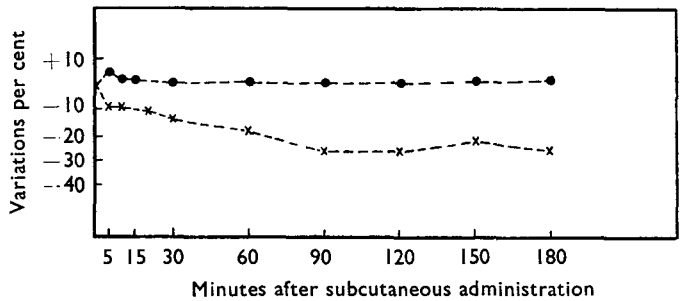


FIG. 3. The variations per cent in respiration of rabbits given 0·5 mg./kg. of B.D.H. 200 or 0·5 ml./kg. of normal saline.

Effect on Blood Pressure and Respiration

B.D.H. 200 causes respiratory depression in rabbits after subcutaneous administration with no apparent effect on blood pressure compared to controls. Figure 3 records the mean per cent variation in respiration in two groups of three rabbits after subcutaneous injection of 0·5 mg./kg. of B.D.H. 200 and control rabbits given a similar volume of normal saline.

In cats the intravenous administration of 0·5 mg./kg. causes a rapid respiratory depression with a concomitant fall in blood pressure. These effects were, however, reversed by intravenous injection of 3 mg./kg. of nalorphine. Figure 4 records the results.

Constipating Effect

At analgesic doses B.D.H. 200 has no constipating effect in mice, for example at 4 mg./kg., which is three times its subcutaneous ED50, there is

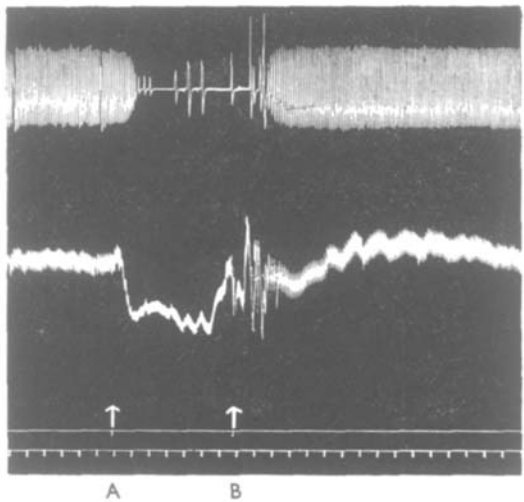


FIG. 4. Cat. Urethane and chloralose anaesthesia. Respiration and carotid blood pressure record. Time = 30 sec. Line above, zero of mercury manometer. At A, B.D.H. 200, 0·5 mg./kg. i.v.; at B, nalorphine, 3·0 mg./kg. i.v.

TABLE VI

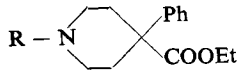
THE EFFECT OF B.D.H. 200 AND PETHIDINE GIVEN SUBCUTANEOUSLY TO MICE ON THE NUMBER OF FAECAL PELLETS PASSED IN 24 HOURS

Compound	Dose mg./kg.	Mean no. of faecal pellets	
		0–8 Hours	8–24 Hours
B.D.H. 200 hydrochloride	4	14	66
	13	8	58
	40	3	39
	120	1	19
Pethidine hydrochloride	4	13	64
	13	17	59
	40	10	54
	120	4	51
Controls	—	15	66

no difference between the treated and control animals. Table VI records the results after various doses of B.D.H. 200 and pethidine. On a weight for weight basis B.D.H. 200 is more constipating than pethidine but in terms of equi-effective analgesic doses it is probably less constipating.

TABLE VII

THE RELATIVE SUBCUTANEOUS ANALGESIC ACTIVITIES IN MICE OF TWENTY-ONE ANALOGUES OF PETHIDINE



Compound	R	Analgesic activity
Pethidine	$-CH_3$	1
B.D.H. 200	$-CH_2-CH(OH)-CH_2-O-Ph$	12·31
1638	$-CH_2-CH(OH)-CH_2-O-C_6H_4-o-Me$	5·1
1947	$-CH_2-CH(OH)-CH_2-O-C_6H_4-m-Me$	2·0
1944	$-CH_2-CH(OH)-CH_2-O-C_6H_4-p-Me$	0·25
1959	$-CH_2-CH(OH)-CH_2-O-C_6H_4-o-O.Me$	5·8
1899	$-CH_2-CH(OH)-CH_2-O-C_6H_4-o-Cl$	5·0
1969	$-CH_2-CH(OH)-CH_2-O-C_6H_4-m-Cl$	0·93
1982	$-CH_2-CH(OH)-CH_2-O-C_6H_4-p-Cl$	0·32
1954	$-CH_2-CH(OH)-CH_2-O-C_6H_4-o-F$	4·9
1957	$-CH_2-CH(OH)-CH_2-O-C_6H_4-o-O.CH_2.CH=CH_2$	1·3
2006	$-CH_2-CH(OH)-CH_2-O-Ph$	6·1
1932	$-CH_2-CH(OAc)-CH_2-O-C_6H_4-o-Me$	0·5
1951	$-CH_2-CH(O.COEt)-CH_2-O-C_6H_4-p-COOEt$	0·1
1985	$-CH_2-CH(OH)-CH_2-N$ (piperidine)	0·60
2042	$-CH_2-CH(OH)-CH_2-N$ (morpholine)	0·43
1945	$-CH_2-CH(OH)-CH_2-OH$	0·1
1993	$-CH_2-CH(OH)-CH_2-OEt$	0·71
3132	$-CH_2-CH(OH)-CH_2-O-CMe_3$	3·3
2040	$-CH_2-CH(OH)-CH_2-O-Bu$	2·0
1965	$-CH_2-CH(OH)-CH_2-O-CH(Ph)_2$	0·1
1949	$-CH_2-C(OH)(Me)-CH_2-O-C_6H_4-o-Me$	0·1

Analogues of B.D.H. 200

In addition, a number of analogues of B.D.H. 200 were examined and are recorded in Table VII.

Discussion

The analgesic activities of several *N*-(2-hydroxy-3-phenoxypropyl) derivatives of norpethidine confirm recent studies that a methyl substitution on the *N*-group of norpethidine does not necessarily give the best analgesic activity in animals. Table VII shows that many derivatives are more potent than pethidine in mice; for example, B.D.H. 200 is twelve times more active than pethidine.

We have no direct evidence of the importance of the carbon chain length or of the ether link in the *N*-arylalkyl chain for the activity of B.D.H. 200. It is also difficult to draw valid comparisons from the results of other

TABLE VIII

THE RELATIVE ANALGESIC ACTIVITY OF B.D.H. 200 AND OF THE *N*-PHENOXYPROPYL ANALOGUE OF PETHIDINE (B.D.H. 3022) IN MICE AFTER SUBCUTANEOUS ADMINISTRATION

Compound	Number of mice	Relative analgesic activity (fiducial limits P = 0·05)
Pethidine	90	1·00
B.D.H. 200	90	15·71 (9·88–24·97)
B.D.H. 3022	90	6·87 (4·40–10·71)

TABLE IX

ORAL AND SUBCUTANEOUS ANALGESIC ACTIVITY OF B.D.H. 200 AND PETHIDINE IN MICE PRETREATED WITH IPRONIAZID 100 MG./KG. I.P. OR BAL 40 MG./KG. I.P. INJECTED 60 MINUTES BEFORE ADMINISTRATION

Compound	Pretreatment	Oral ED50 mg./kg.	Subcutaneous ED50 mg./kg.
B.D.H. 200 ..	—	49·51	1·46
B.D.H. 200 ..	Iproniazid	14·21	1·43
B.D.H. 200 ..	BAL	75·00	1·96
Pethidine ..	—	29·75	18·38
Pethidine ..	Iproniazid	27·10	16·51
Pethidine ..	BAL	74·36	9·19

workers with other series using different techniques. However, Elpern, Gardner and Grumbach[12] and Grumbach[13] found in rats that the *N*-phenylpropyl derivative of norpethidine was 10 times more active than the *N*-phenylethyl derivative, and that the *N*-phenoxypropyl derivative was slightly less active than the corresponding *N*-phenylpropyl derivative. Moreover the *N*-phenoxyethyl derivative of norpethidine is only three times more active than the *N*-phenylethyl derivative. Winter, Orahovats and Lehman[24], who examined in rats many *N*-analogues of morphine synthetised by Clark, Pessolano, Weyland and Pfister[25], found that the *N*-phenylethyl derivative of normorphine is six times more active than morphine and that the *N*-phenoxyethyl derivative is inactive.

These studies indicate that the length of the carbon chain may be more important for analgesic activity than the presence of an oxygen ether linkage whether in a ring system or not.

We know that the presence of the 2-hydroxy group in the B.D.H. 200 molecule is important for its analgesic properties. The relative subcutaneous activities of B.D.H. 200 and B.D.H. 3022, the phenoxypropyl analogue, to pethidine are recorded in Table VIII where B.D.H. 200 is more than twice as active as B.D.H. 3022.

The thirty-six fold fall in analgesic activity of B.D.H. 200 from subcutaneous to oral administration in mice cannot be explained entirely by poor alimentary absorption as the corresponding fall in toxicity is less than three. In contrast the decrease in toxicity and analgesic activity of pethidine from subcutaneous to oral administration is similar, the oral LD50 and ED50 values being approximately two-thirds of the subcutaneous values.

A marked decrease in analgesic activity after oral compared to subcutaneous administration[10,15] has been reported by others. With the *N*-phenylethyl and the *N*-2-hydroxy-2-phenylethyl analogues of norpethidine, the decrease in oral activity is about the same as B.D.H. 200[10].

The marked decrease in activity of B.D.H. 200 on oral administration compared to pethidine can possibly be explained by differences in metabolism. Table IX demonstrates an increase in oral analgesic activity of B.D.H. 200 in mice after pre-treatment with iproniazid and a decrease after pre-treatment with BAL. Neither of these compounds modified the responses to subcutaneous administration.

After this work had been completed the following papers dealing with the analgesic activity of norpethidine derivatives were published. Janssen and Eddy, *J. med. pharm. Chem.*, 1960, 2, 31. Blair and Stephenson, *Brit. J. Pharmacol.*, 1960, 15, 247. Lister, *ibid.*, 1960, **15**, 254.

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