MORPHINE ANTAGONISM

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A series of compounds which antagonise the depressant activity of morphine in dogs is described. The most active compounds are the previously described 2 : 4-diamino-5-phenylthiazole and tetrahydro-5-aminoacridine. A mixture of both substances is more active than either alone. Certain antihistamine drugs display antimorphine activity.

The action of a series of compounds which antagonised the narcotic property of morphine was described by Shaw and Bentley[1]. One, 2 : 4-diamino-5-phenylthiazole (amiphenazole, Daptazole, DHA.245) has now found clinical application as a partial morphine antagonist in the treatment of intractable pain of terminal carcinoma[2], post-operative pain[3], and pain in childbirth[4].

Amiphenazole is a partial morphine antagonist which counteracts the narcosis of morphine and to a lesser extent the respiratory depression, but has little effect on the nausea and vomiting. It does not affect the analgesia produced by morphine. In animals the drug is a respiratory stimulant[5] but less regularly in man. It has a low toxicity, the LD50 being 309 (oral) and 250 (i.p.) mg./kg. in rats and guinea pigs. The animals die after mild convulsions.

The disadvantage of amiphenazole, clinically, is that whilst the elimination of morphine-induced narcosis is usually complete it is not as efficacious in combating vomiting and respiratory depression. However, up to the present time no death has been reported when a combination of amiphenazole and large doses of morphine has been used. Also amiphenazole in solution in water is not stable for more than a few days. A search has been made now for additional compounds without these disadvantages.

Methods

Healthy dogs of both sexes were used. A mixture of morphine 100 mg. and hyoscine 6 mg. in varying doses produced uniform narcosis in individual animals. The narcosis aimed at was such that the dog would make only a slight movement of the head when stimulated by movement or a sudden sharp noise. Dogs which could not be brought to this state, even with large dosages were rejected. The required dose of morphine varied around 15 mg./kg. The mixture was usually administered subcutaneously, occasionally intramuscularly. The test compound was administered $\frac{3}{4}$–1 hour later. If soluble the compound was usually administered intravenously; if insoluble, a suspension was made in gum tragacanth and given intramuscularly. The antimorphine activity was assessed as follows: good arousal, the animal stood up and walked

spontaneously; mild arousal, the animal sat up spontaneously but only walked when stimulated or called; slight arousal, the animal raised only its head when stimulated.

Some animals were used repeatedly twice a week for several months. No tolerance to the action of morphine appeared when the drugs were given at this interval.

Most of the dogs under morphine-hyoscine had an elevated respiratory rate. Any further respiratory stimulation was noted. If a compound failed to arouse a dog a standard dose of amiphenazole (20 mg./kg.) was usually given. A test compound was not reported to be without activity if this dose of amiphenazole itself failed to rouse the animal.

RESULTS

The compounds of greatest interest are described below; others are mentioned briefly in Table I.

Cyclizine Chloride (*Marzine*). In view of its antiemetic effect against morphine-induced vomiting in man cyclizine was tested and found, surprisingly, to be almost equal to amiphenazole and tetrahydroaminacrin (THA) in producing arousals. Eleven morphinised dogs were given doses (intravenously) ranging from 10–30 mg./kg. The response was an improvement in the level of consciousness in all cases. The effects on respiration were minimal. Muscle tone was strong enough in one dog only to permit it to stand. The dogs seemed to remain analgesic to crude stimuli such as pricking or crushing of the paw or tail. The delayed effect varied from relapse after half an hour to maintenance of the alert state.

Chlorcyclizine (*Perazil*). Chlorcyclizine, a more potent antihistamine drug than cyclizine, was investigated. Given intravenously to nine morphinised dogs in a dose range of 10–30 mg./kg. the response was an immediate return to consciousness and improvement in respiration. Muscle tone was scarcely affected and hypotonia predominated. The animal was able to lift its head and right its body, but was unable to support its weight on its legs and walk about as is usual with amiphenazole and THA. The dog responded to orders and sounds by moving its head and appeared to possess a considerable degree of comprehension.

Avil (*p*-amino salicylate of 1-phenyl-1-pyridyl-2′:3-dimethylamino propane) was given to 5 dogs with dosage range 20–30 mg./kg. intravenously. Full consciousness was regained immediately in all dogs; respiration and general muscle tone was slightly improved.

Less satisfactory effects were seen with diphenhydramine (Benadryl) and pyribenzamine.

n-Methyl piperidyl-(3)-methyl phenothiazine deepened the narcosis. Methyl phenidate (Ritalin) in a dosage of 2 mg./kg. to two dogs produced an immediate return to consciousness and some improvement in muscle tone. In one animal there was an improvement in respiratory function.

Amiphenazole and Tetrahydroaminacrin. Shaw and Bentley[1] have described both substances, and preferred the less toxic amiphenazole for clinical use.

TABLE I

SERIES OF COMPOUNDS INVESTIGATED

Compound	Dose in mg./kg.	Arousal	Excitation	Respiratory Effect
Methylacridine	12 i.v.	None	Marked	Resp. very deep, faster. Effect wore off after about 20 min.
	6 i.v.	None	Excitation followed immediately by deepening of coma	Much deeper, faster
9-Amino-5:6:7:8-tetrahydrophenanthridine	5 i.v. in HCl	Transient	Convulsed	Extreme, transient stimulation rate and volume
5-Amino-1-methyl-8:9-benzacridine	4·5 i.v.	None	None	Deepened, became slower; irregular later
5-Amino-6:7-benzacridine	1 in HCl	Mild	Severe convulsion	Deeper, then rate decreased markedly
	0·5 in HCl	Slight, with regression about 1 hour	Twitches developed	Very slow, became very shallow also
	1 in HCl	Good, with total regression after about 20 min.	Excitement twitches	Slowed; then became irregular
5-Amino-2:3:6:7 dibenzacridine	4-5 i.m. in 7 doses at 20 min intervals	Slight	None	Gradually became more forced and irregular
Aminodiazine	10 i.v.	None	Twitches developed	Immediate deepening (irregular)–persisted; very slow and regular later
	17 i.v.	None		Deeper, irregular
Aminomerazine	20 i.v.	None	—	No action
	20 i.v.	None	—	No action
	20 i.v.	None	—	No action
2-Aminopyridine	8·5 i.v.	Slight	Twitches developed	Slower and deeper immediately, then irregular
	2·5 i.v.	None	Great excess of tone in whole body. Convulsion	Deepened, became irregular
	5 i.v.	None	Convulsions	Very deep, then became irregular
	2·5 i.v.	Mild arousal	—	
	7 i.v.	Good arousal with total regression	Extreme excitement with severe twitches	Very deep
2-Aminothiazole	30 i.v.	Slight	—	Deepened, slow and regular
	38 i.v.	None	—	Temporarily deeper
	40 i.v.	None	—	Very deep, then slow and irregular
2-Amino-4:6-dimethylpyrimidine	17 iv.	None	—	Very deep for short while, then shallow and irregular
	20 i.v.	None	Severe tremors and excess tone in hind leg	Deep and regular
3:4-Dimethyl-5-aminoisoxozole	12 iv.	None	—	—
	20 i.v.	None	—	—
	20 i.v.	None	—	—
2-Amino-5-phenylthiazole	20 i.m.	None	None	Irregular
	40 i.m.	None	Developed tremors in hind legs	No effect
2-Amino-4-phenylthiazole	15 i.m.	Slight	—	—
	25 i.m.	Mild, gradual	—	—
	40 i.m.	Mild, gradual	—	Slower, very deep
	30 i.m.	Mild	Slight twitching	Rapid
*iso*Cytosine	8·5 i.v.	—	—	None
	20 i.v.	—	—	—
2-Amino-4-phenylpyrimidine	10 i.v.	—	—	Good volume regular, a few deep resps
	20 i.v.	Good with slight regression	—	—
	25 i.v.	Mild	Mild fit with excitation, then short deep coma followed, by arousal	Good volume
	20 i.v.	None	Injection followed by dry gagging then deep coma	Shallow, fairly fast
5-Amino-1:2:8:9-dibenzacridine	3 i.m.	Slight after ¾ hour	—	—
	6 i.m.	Slight	—	Faster, deeper
	8 i.m.	Slight	—	Irregular
	6 i.m.	Mild, gradual	—	Irregular
	10 i.m.	None	—	Shallower
Aminoacridine pKa 4·4	20 i.v.	None	—	None
	30 i.v.	Slight	—	None
Aminoacridine pKa 5·9	15 i.v.	None	—	None

TABLE I—*continued*

Compound	Dose in mg./kg.	Arousal	Excitation	Respiratory Effect
Butylaminoacridine pKa 9·4	5	Good	Convulsed, then cardiac failure and death	—
Diacetylaminoacridine pKa 5·0	17	Mild, delayed	—	None
1-Methyl-5-diacetylaminoacridine	8	None	—	—
	10	None	—	None
1:2:3:4-Tetrahydro-5-diacetylaminoacridine	12	Slight	—	None
2:4-Diamino-5-(*p*-chlorophenyl) thiazole pKa 7·5	50	None	—	None
2:4-Diamino-5-(*o*-chlorophenyl) thiazole	30	Mild, regression later	Convulsed during injection, and again 20 mins later	None
2:4-Diamino-5-(*o*-methoxyphenyl) thiazole	30	Mild	—	None

See also Tables II and III.

Note:—Excitation means fasciculation or other movements if the animal remains unconscious or convulsions, etc., if the animal is first aroused.

Thirty dogs provided a control series with amiphenazole, twenty-two gave a good or mild arousal with 8 to 40 mg./kg., eight, did not show any arousal, even with 60 mg./kg.

In a group of 4 dogs, it was observed that admixtures of amiphenazole and THA enabled the amount of each to be reduced, sometimes to less than half, yet to produce the same degree of arousal. (Table II). The optimum ratio would appear to be amiphenazole: THA, 2:1 by weight.

TABLE II

AROUSAL OF DOGS FROM ADMIXTURES OF AMIPHENAZOLE AND THA

Dog	Amiphenazole mg./kg.	THA mg./kg.	THA + Amiphenazole mg./kg.
I	40 B	10 B 14 A 15 B 20 A	12 + 20 B
II	40 A 60 A	10 A 14 A 14 B 20 A	5 + 10 B 2·5 + 5 B 5 + 10 B 15 + 30 A
III	40 A 60 A	10 B 15 A 17 A 20 A	3 + 15 B 5 + 15 A 5 + 10 B 10 + 20 A 15 + 30 A
IV	40 B	10 A 13 A 20 B 25 A	10 + 7 B 15 + 30 A 6 + 12 B 10 + 20 A

A = Good arousal B = Mild arousal

It was felt that the animals treated with the mixture were more normal in their gait and alertness than those treated with either drug alone. The respiration also appeared to be more normal in depth and rate.

From earlier experiments it was known that THA, was the more convulsive drug. An attempt was now made to see if amiphenazole would lower the motor excitement induced by THA. The dose of THA,

producing mild convulsions was estimated in four dogs. This dose found to be approximately 5 mg./kg., was then given simultaneously with 30 mg./kg. of amiphenazole. The degree of convulsions produced was the same as that given by THA alone. The non-protective action of amiphenazole was confirmed by a similar procedure with 30 rats.

La Barre told us that the barbiturate antagonist bemegride, β-methyl-β-ethyl glutarimide (Megimide) antagonised the respiratory depression in animals anaesthetised with Chlovalox and morphine. Therefore this compound was tested for its ability to arouse morphinised dogs together with three other related glutarimides with barbiturate-antagonistic ability. These were: *N*-methyl-β-methyl-β-ethyl glutarimide and methyl-β-methyl-β-ethyl glutarimide, and β-methyl-β-propyl glutarimide. The results are shown in Table III.

TABLE III

EFFECT OF BEMEGRIDE AND RELATED GLUTARIMIDES ON THE AROUSAL OF DOGS ANAESTHETISED WITH MORPHINE AND CHLOVALOX

Compound	Dose mg./kg.	Arousal	Excitation	Respiration
Bemegride	10	Good	Extremely violent convulsions, ending in death	Marked stimulation
	1·5	Moderate, regression later	Convulsions	Marked stimulation
	0·5	Moderate, regression later	Twitching	Moderate stimulation
N-Methyl-β-methyl-β-ethyl glutarimide	1·6	Mild, regression later	Convulsions	Moderate stimulation
β-Methyl-β-propyl glutarimide	5	None	—	Brief moderate stimulation
α-Methyl-β-methyl-β-ethyl glutarimide	5	None	—	Brief moderate stimulation

DISCUSSION

It is not difficult to obtain substances which antagonise the narcotic action of morphine in animals[1]. This is in distinction to the difficulty of obtaining an antagonist to the barbiturates, where only a few out of over 100 compounds tested possessed this property[7,8]. Furthermore, in man, the barbiturate antagonist bemegride counteracts the depression of vital reflexes rather than eliciting a return to consciousness[9]. Amiphenazole is particularly useful in restoring consciousness to morphinised dogs and man[2]. There seems to be only slight structural specificity required for a morphine antagonist. In the grossest sense any compound with one or more *N*-containing rings (5 or 6 membered) and one or more amino side chains would appear to be a potential morphine antagonist. What is surprising is the discovery that certain antihistamine drugs also possess this property. Cyclizine and chlorcyclizine are as effective as amiphenazole and THA in dogs (and cyclizine in man). Three other antihistamine substances investigated were also very effective. Amiphenazole and THA possess only slight antihistaminic activity.

In a previous paper[1] it had been pointed out that all the highly active morphine antagonists in this series were basic compounds with acid dissociation constants of 8 or higher. However, until recently, very few compounds of low basicity had been available. This deficiency

has now been partly removed, through unfortunately a number of the compounds listed in Table I were in such short supply that it was not possible to measure their dissociation constants. Table I gives a list of some of the compounds with their dissociation constants. By adding these figures to those already presented[1], it can be seen that morphine antagonism is conspiciously absent from the weakly basic compounds in this series. However, it must be emphasised again that all compounds with a high basicity are not antagonistic towards morphine.

Proflavine (pK 10) and acriflavine (pK 11) are notable examples of this phenomenon. It is also interesting to note that the three derivatives of 2:4-diamino-5-phenylthiazole (pK 8) have almost negligible activity. Their dissociation constants are only slightly less than that of the parent compound, and it would be surprising if this small reduction in basicity was the sole cause of the reduced biological activity. Other factors, such as lipid solubility would also play their part.

The moderate activity of the poorly ionised compound 5-diacetylaminoacridine was unexpected. However, since its activity appeared some 5–10 minutes after injection, it seems possible that it is hydrolysed in the body to the active parent compound 5-aminoacridine.

The four glutarimide derivatives must be considered separately. They are all very weakly acidic compounds, with dissociation constants above 10, i.e., at biological pH they are completely unionised.

A surprising aspect of this work is the marked antimorphine effect of certain compounds with antihistaminic activity. This prompted us to test the antihistaminic activity of amiphenazole and THA. The action is very weak indeed. On the guinea pig intestine amiphenazole at a concentration of 10^{-5} abolishes the action of histamine for 5 minutes, the corresponding figures for THA, are $10^{-5\cdot5}$ and 6. These drugs do not influence the action of histamine on the cat's blood pressure. *p*-Chlor-2:4-diamino-5-phenylthiazole is also a weak antihistamine drug. Whilst the antihistamine drugs appear to antagonise the narcotic action of morphine in dogs they do not usually bring about a complete return of tone and voluntary movement in the animal.

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