Br. J. Pharmac. Chemother. (1968), **33**, 266–276.

KINETIC PARAMETERS OF NARCOTIC AGONISTS AND ANTAGONISTS, WITH PARTICULAR REFERENCE TO *N*-ALLYLNOROXYMORPHONE (NALOXONE)

BY

H. W. KOSTERLITZ AND A. J. WATT

From the Department of Physiology, University of Aberdeen

(*Received December* 8, 1967)

Gyang & Kosterlitz (1966) have shown that all narcotic analgesic drugs have dual agonist and antagonist actions, whether they are used clinically as "agonists" or "antagonists." They examined the effects of these drugs on the contractions of the longitudinal muscle of the guinea-pig isolated ileum stimulated by coaxial electrodes and found that the "narcotic antagonists" always exhibited greater antagonist activity than the "narcotic agonists"; however, there was often little difference in the agonist activity of chemically related pairs of "narcotic agonists" and "narcotic antagonists" such as morphine and nalorphine, levorphanol and levallorphan, phenazocine and cyclazocine.

It seemed of interest, therefore, to examine the kinetics of these drugs and for this purpose the following parameters were chosen: the concentration causing 50% inhibition of the contraction of the longitudinal muscle (ID50), to characterize agonist activity; the equilibrium constant (K_e) and the recovery rate constant (K_r), to characterize antagonist activity. The representatives of the morphine, morphinan and benzomorphan series examined by Gyang & Kosterlitz (1966) were included in this investigation and also some other compounds; of particular interest was the morphine derivative, naloxone, a potent "narcotic antagonist" with insignificant agonist activity (Foldes, Lunn, Moore & Brown, 1963; Lasagna, 1965; Blumberg, Dayton & Wolf, 1966; Jasinski, Martin & Haertzen, 1967; McClane & Martin, 1967).

METHODS

Experimental procedure

All experiments were performed on the guinea-pig isolated ileum; the terminal portion was used after the 10 cm nearest to the ileo-caecal junction had been discarded. The depressant action of drugs was tested on the contraction of the longitudinal muscle induced by coaxial electrical stimulation (Paton, 1955). Because of the greater stability of its responses to electrical stimulation, a segment of the intact ileum was used and not a strip of the longitudinal muscle which would have been more suitable for the determination of the recovery rate constant, K_r. The bath fluid, Krebs solution (40 ml.) to which hexamethonium bromide (69 μM) and mepyramine maleate (0.125 μM) were added (Gyang & Kosterlitz, 1966), was bubbled with 95% oxygen and 5% carbon dioxide. The temperature was 36° C. The stimuli were 1.5 times maximal rectangular pulses of 0.5 msec duration, at a frequency of six per minute. The twitch-like contractions of the longitudinal muscle were recorded isometrically by means of a mechanoelectrical transducer (Grass FT.03) and displayed on a pen oscillograph (Devices).

Drugs

The following " narcotic agonists " and " narcotic antagonists " were used: morphine hydrochloride, codeine phosphate, diamorphine (heroin) hydrochloride, dextromoramide ((+)-1-(3-methyl-4-morpholino-2,2-diphenylbutyryl)pyrrolidine) bitartrate, levorphanol ((−)-3-hydroxy-*N*-methylmorphinan) tartrate, nalorphine (*N*-allylnormorphine) hydrochloride, levallorphan ((−)-3-hydroxy-*N*-allylmorphinan) tartrate, pentazocine (2′-hydroxy-2-(3,3-dimethylallyl)5,9-dimethyl-6,7-benzomorphan), cyclazocine (2′-hydroxy-2-cyclopropylmethyl-5,9-dimethyl-6,7-benzomorphan), naloxone (*N*-allylnoroxymorphone) hydrochloride (Endo Laboratories), *N*-methylallylnormorphine hydrochloride, CI 572 (*m*-(1-methyl-3-propyl-3-pyrrolidinyl) phenol) hydrochloride (Parke, Davis and Company), M 5050 (*N*-cyclopropylmethyl-6,16-*endo*ethano-7α-(1-hydroxy-1-methylethyl)-tetrahydronororipavine) (Reckitt & Sons Ltd.).

Other drugs used were: acetylcholine chloride, adrenaline, hexamethonium bromide, mepyramine maleate, phenoxybenzamine hydrochloride and propranolol hydrochloride.

Stock solutions of the drugs were made up in distilled water; M 5050, pentazocine, cyclazocine and adrenaline were dissolved after adding the required amounts of HCl. Concentrations are given as M, μM or nM.

Assessment of agonist and antagonist activity

ID50 as parameter of agonist activity

Tachyphylaxis to agonist activity occurs with all narcotic analgesics whether they are used clinically as " agonists " or " antagonists," so it is difficult to determine the value of ID50 in the conventional manner, from dose-response curves plotted as percentage inhibition of the twitch against the log of molar concentration (" multiple dose " method). Gyang & Kosterlitz (1966) showed that reproducible dose-response curves could be obtained for morphine when doses were added to the bath at intervals of 15–20 min, but that longer intervals were required for some of the other drugs. It will be shown in the RESULTS section that some of the " narcotic antagonists," for example, M 5050, *N*-methylallylnormorphine and cyclazocine, have such low recovery rates that a period of 30–50 min is necessary to free half of the receptors. Because of possible interaction, the interval between doses would have to be at least 2–3 hr, during which time the sensitivity of the ileum might change considerably. Nevertheless, the " multiple dose " method yielded ID50 values which compared satisfactorily with values obtained with an alternative " single dose " method.

When the " single dose " method was used, the preparation was set up and stimulated for 1 hr, as already described; then doses of morphine were added at intervals of 20 min and the depressant effect plotted against log concentration of morphine. Forty-five minutes after the last dose of morphine the ileum was exposed to the compound, the agonist and antagonist activities of which were to be examined; the amount chosen was that which would cause a depression of the twitch by not less than 20% and not more than 60%, but preferably by 30–40%. The ID50 value was then extrapolated by assuming that the slopes of the dose-response curves of morphine and the compound examined were not significantly different.

Because the slopes of the dose-response curves of most " narcotic antagonists " are likely to be less steep than that of morphine, the ID50 values obtained with the " single dose " method may be expected to be too low; on the other hand, the values obtained with the " multiple dose " method may be expected to be too high because of interaction between the multiple doses. In fact, with some " narcotic antagonists "—for example, cyclazocine and *N*-methylallylnormorphine—the ID50 values obtained with the " single dose " method were about half those found with the " multiple dose " method; this was the greatest difference ever found between the two methods. On the other hand, there was little difference between the ID50 values obtained by the two methods for the " narcotic agonists."

While the " single dose " method was used as routine, it was found advisable to examine more fully the dose-relationship for any new drug, in order to exclude the possibility that the maximum depression obtainable was less than 50%.

K_e as parameter of antagonist activity

K_e is the equilibrium constant

$$K_e = \frac{a(1-y)}{y}$$

where a is the molar concentration of the antagonist and y the fraction of receptors occupied. y can be determined from the dose-ratio (DR), that is, the ratio of the concentrations of the agonist, morphine, required to depress the twitch to the same extent in the presence or absence of a given concentration of an antagonist. Since

$$y = \frac{DR-1}{DR}, \quad K_e = \frac{a}{DR-1}$$

(Stephenson, 1956 ; Paton, 1961).

In all estimations of K_e, the agonist was morphine, for which a dose-response curve had been constructed 45 min before exposure of the ileum to the antagonist. The use of morphine as agonist is open to criticism because it has been shown that morphine exhibits dual agonist and antagonist activities (Gyang & Kosterlitz, 1966). Attempts have been made to find an agonist with no or only negligible antagonist activity but this search has so far been unsuccessful. For this reason, the estimation of K_e is somewhat unreliable but this fact does not affect the overall argument as to the relative agonist and antagonist properties of the various compounds.

Because all drugs except naloxone exhibited agonist as well as antagonist activity, the determination of DR had to take into account the agonist component (Fig. 1). The procedure was based on the principles first used by Stephenson (1956). The ileum was exposed to the antagonist as already

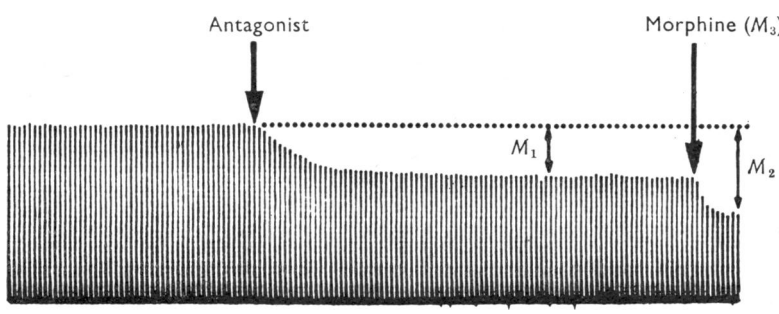

Fig. 1. Measurement of the antagonist activity of a partial agonist by determination of its equilibrium constant, K_e. Guinea-pig isolated ileum. Isometric recording of the contractions of the longitudinal muscle induced by electrical coaxial stimulation. At the arrow marked antagonist, the partial agonist was added and produced a depression of the twitch equal to a depression caused by morphine in concentration M_1. Morphine was added 20 min later to give a concentration M_3 ; the total depression was equal to a reduction in the height of the twitch caused by morphine in a concentration M_2 in the absence of the antagonist. $DR = M_3/(M_2-M_1)$.

described for the " single dose " method for determining ID50. Then the concentration of morphine (M_1) which would have been expected to produce the same depressant effect on the twitch was read off the dose-response curve previously constructed for morphine. After the ileum had been exposed to the antagonist for 20 min, morphine in a concentration M_3 was used to depress the twitch further. The morphine concentration (M_2) which would have a depressant effect equal to the combined

actions of the antagonist and the morphine present in the bath was read off the dose-response curve for morphine. The dose–ratio was then

$$DR = \frac{M_3}{M_2 - M_1}$$

As already stated, the concentration of antagonist was chosen so that the height of the twitch was depressed by 20 to 60% and preferably by 30–40%. The concentration of morphine (M_3) used in the presence of the antagonist was such that the residual contraction was depressed by at least 20% and the total contraction was inhibited by not more than 80%.

After the antagonist had been washed out, it was sometimes observed that the response to morphine had either increased or decreased considerably during the course of the experiment. If the morphine sensitivity changed by more than 25% the experiment was discarded.

Effective antagonist potency

The effective antagonist potency (P_a), which takes into account the agonist activity of partial agonist, was expressed thus

$$P_a = \frac{1}{K_e} \Big/ \frac{1}{ID50} = \frac{ID50}{K_e}$$

$P_a = DR\text{-}1$ when the drug is used in the concentration which has an agonist effect equal to ID50. Since P_a was calculated from the means of ID50 and K_e, the differences between P_a values were not treated statistically.

The method used for the assessment of K_e and P_a is valid only if the antagonism between morphine and the antagonist is competitive ; so far it has been possible to prove that the antagonism by naloxone of the agonist effects of morphine, levorphanol, codeine and nalorphine is competitive.

Although P_a is closely related to efficacy, e (Stephenson, 1956), since $e = 1 + 1/P_a$, the use of P_a is preferred because this investigation is concerned with antagonist rather than agonist activity.

Rate of recovery from the action of an antagonist

K_r was calculated from the equation

$$K_r = \frac{\log_e y_o - \log_e y_t}{t}$$

where y_o and y_t represent receptor occupancy at $t=0$ and at times t (sec) respectively (Paton, 1961).

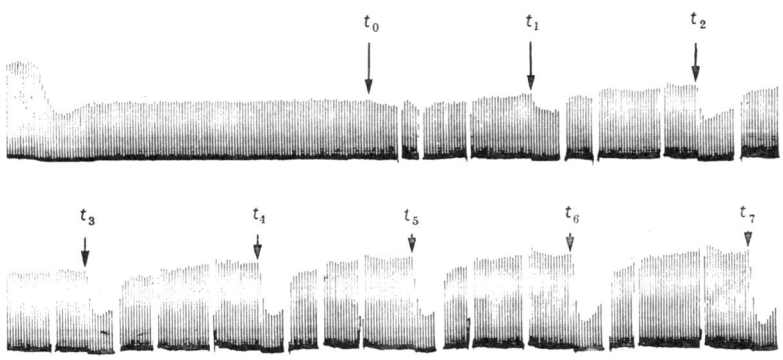

Fig. 2. Measurement of the recovery rate constant, K_r. Record as in Fig. 1. At t_o, the effect of a given concentration of morphine was tested in the presence of the antagonist which had been present in the bath for 20 min. The antagonist was then washed out; each gap in the trace indicates renewal of the organ bath solution. The effect of the same concentration of morphine was tested at 10 min intervals, t_1, t_2, t_3, etc. Receptor occupancy is calculated from the dose ratios determined as in Fig. 1.

K_r was determined in the same experiment in which the values of ID50 and K_e had been obtained. The fraction of receptors occupied at the time of the estimation of K_e was y_0; y_{600}, y_{1200}, etc., were determined from $y=(DR-1)/DR$ by exposing the ileum to the test dose of morphine 600, 1200, etc., seconds after washing out the antagonist (Fig. 2); DR was determined as described for the estimation of K_e, M_1 being calculated from the height of the twitch at the moment of testing with morphine. The values of log y were plotted against time and, if a straight line was obtained, K_r was calculated (Fig. 3).

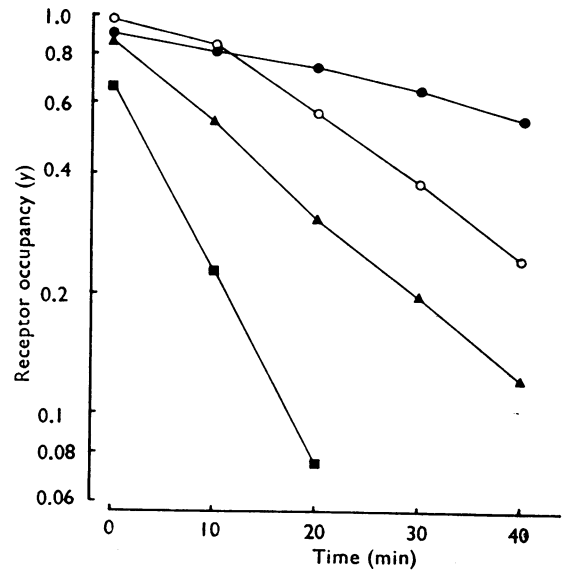

Fig. 3. Recovery rates of levorphanol (■), nalorphine (▲), naloxone (○) and cyclazocine (●).

The rate of recovery was also expressed as the "half time" (T_r) of dissociation

$$T_r\,(\text{min}) = \frac{\log_e 0.5}{60\,K_r} = \frac{0.0116}{K_r}$$

RESULTS

Three of the parameters, ID50, K_e and P_a, of thirteen drugs are listed in Table 1, which has been arranged to give the values of effective antagonist activity, P_a, in ascending order. The first two compounds, heroin and morphine, have the weakest antagonist activity, with P_a values of 0.6 and 0.8, while at the other end of the table naloxone, with a P_a value of more than 56,000, is an antagonist with negligible agonist activity in man (Foldes *et al.*, 1963; Jasinski *et al.*, 1967).

The values of K_e and P_a do not vary in the same direction; this is because the agonist potency of a compound influences P_a but not K_e. Thus K_e equals the concentration producing a dose-ratio of 2, whereas P_a+1 equals the dose-ratio caused by the compound when present in a concentration which has an agonist effect equal to its ID50. For instance, the "narcotic antagonist" M 5050 (Blane, 1967), a member of the very potent oripavine series, has a K_e as low as 0.13; but, because it also has agonist activity, its P_a is only 5.2. On the other hand, *N*-methylallynormorphine has a K_e as

TABLE 1

KINETIC PARAMETERS OF "NARCOTIC AGONISTS" AND "NARCOTIC ANTAGONISTS"
Means and S.E. of means of agonist activity (ID50) and antagonist activity (K_e) obtained from six observations (seven for pentazocine). The values of effective antagonist potency, P, were calculated from the means of ID50 and K_e. * This concentration caused an inhibition of only 2–3%.

Drug	ID50 (nM)	K_e (nM)	P_a ($ID50/K_e$)
Heroin	35·1±4·4	60·6±26·5	0·6
Morphine	68·2±15·0	87·5±18·1	0·8
Codeine	10,300±3,560	8,840±2,540	1·2
Levorphanol	9·18±1·05	7·04±1·48	1·3
CI 572	420±40·3	305±41·0	1·4
Dextromoramide	6·68±1·15	4·33±1·78	1·5
Pentazocine	250±36	151±21	1·7
Cyclazocine	3·60±1·15	1·48±0·37	2·4
Levallorphan	4·28±1·64	1·12±0·23	3·8
M 5050	0·68±0·07	0·13±0·02	5·2
Nalorphine	24·3±1·3	4·47±0·59	5·4
N-methylallylnormorphine	640±96	43·5±8·0	14·7
Naloxone	>68,000*	1·22±0·03	>56,000

high as 43.5 but, since it is a very poor agonist, its P_a of 14.7 is considerably higher than that of M 5050.

To judge from the P_a values, there seem to be two major groups of narcotic drugs. Compounds which have P_a values less than 2 comprise the " narcotic agonists " including the less potent, non-addicting analgesics, pentazocine (Telford, Papadopoulos & Keats, 1961), CI 572 (Winder, Welford, Wax & Kaump, 1966) and codeine. On the other hand, compounds with P_a values of more than 2 belong to the group of " narcotic antagonists." In this latter series of compounds, the relative agonist potency decreases with increasing P_a value ; the maximum value of P_a is reached with naloxone, which, in whatever concentration used, never reduces the twitch by more than 2–3%.

The values of the recovery rate constant, K_r, and of the half time of recovery, T_r, are shown in Table 2. There is considerable variation but the values can be roughly divided into two groups. The " narcotic agonists " have half time values of less than 8 min, while the " narcotic antagonists " have values between 12 min (nalorphine) and 50 min (cyclazocine). The value found for pentazocine (8 min) is on the border line between the " narcotic agonists " and " narcotic antagonists."

TABLE 2

RECOVERY RATE CONSTANT, K_r, AND HALF TIME OF RECOVERY, T_r, OF "NARCOTIC AGONISTS" AND "NARCOTIC ANTAGONISTS"

Drug	No. of observations	K_r ($\times 10^{-4}$/sec) + S.E. of mean	T (min)
Dextromoramide	6	>18	<6
CI 572	6	>18	<6
Levorphanol	7	>16	<7
Heroin	6	>14	<8
Codeine	6	>14	<8
Pentazocine	6	14·6±2·6	8
Nalorphine	6	10·1±1·31	12
Levallorphan	4	8·88±1·15	13
Naloxone	4	6·12±0·72	19
M 5050	4	4·16±0·36	28
N-methylallylnormorphine	5	3·89±0·36	30
Cyclazocine	3	2·11±0·07	50

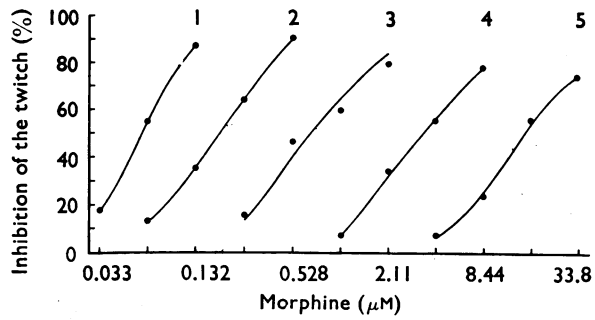

Fig. 4. Antagonism of morphine by naloxone. The dose-response curves were obtained, from left to right: 1, in the absence of naloxone; 2–5, in the presence of naloxone, 3.4, 13.7, 55 and 220 nM, respectively.

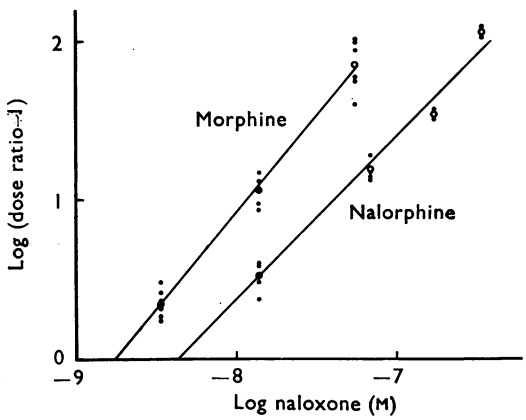

Fig. 5. Competitive nature of the antagonism by naloxone of the depressant effects of morphine, levorphanol and nalorphine. Individual observations are given by the filled circles and mean values by the open circles. The lines are drawn from the regression equations, log $(DR-1)=1.25$ (8.73 + log naloxone) for morphine and log $(DR-1)=1.06$ (8.31 + log naloxone) for nalorphine. The individual observations and the regression line for levorphanol, log $(DR-1)=1.10$ (8.94 + log naloxone), are not shown because the regression line almost coincides with that of morphine.

Competitive antagonism by naloxone

As naloxone is almost devoid of agonist activity, it could be used in high concentrations to examine the nature of its antagonist action. It was found that naloxone caused a parallel displacement of the dose-response curve of morphine to the right (Fig. 4). When log $(DR-1)$ was plotted against log naloxone concentration, straight lines were obtained with either morphine or nalorphine as agonist (Fig. 5). The slopes of the regression lines were 1.25 for morphine and 1.06 for nalorphine; none of these values differed significantly from unity. The K_e values obtained from the intersection of the regression lines with the abscissae in these and other experiments, were 1.18 nM for morphine, 1.11 nM for levorphanol, 1.16 nM for codeine and 1.20 nM for CI 572. On the other hand, a considerably higher value, 1.49 nM, was found for nalorphine. These observations agree with the view that naloxone is a competitive antagonist of the agonist action of these compounds.

Differentiation between adrenaline and morphine receptors

Schaumann (1958) suggested that morphine might exert its inhibitory action by releasing catecholamines. Adrenaline and noradrenaline are known to depress the twitch induced by coaxial stimulation. However, the inhibition produced by adrenaline was antagonized by a mixture of the α- and β-blocking agents, phenoxybenzamine and

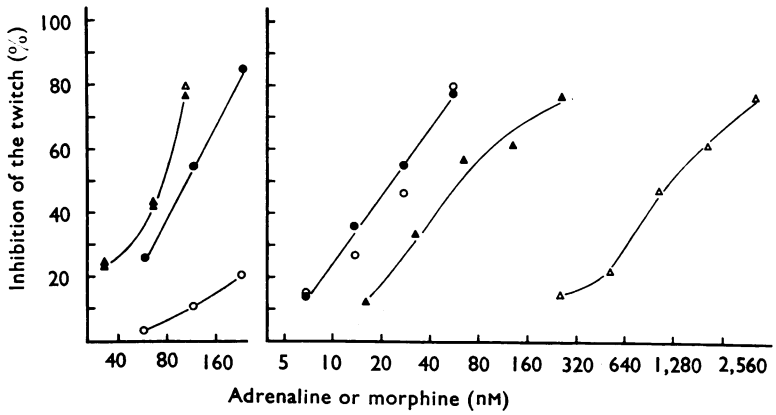

Fig. 6. Antagonist effects of a mixture of phenoxybenzamine and propranolol and of naloxone on the depressant actions of adrenaline and morphine. Left panel, effects of phenoxybenzamine 15nM) and propranolol (850 nM); right panel, effects of naloxone (13.7 nM). Adrenaline alone (●) and in the presence of antagonists (○); morphine alone (▲) and in the presence of antagonists (△).

propranolol, while the inhibitory effect of morphine was unaffected; on the other hand, naloxone antagonized the inhibitory effect of morphine but not that of adrenaline (Fig. 6). Thus the receptors for morphine and adrenaline are separate and independent of each other.

DISCUSSION

The conclusion reached by Gyang & Kosterlitz (1966), that both "narcotic agonists" and "narcotic antagonists" are partial agonists, has been confirmed for an increased number of compounds. It has been shown that the equilibrium constant, K_e, does not adequately describe the suitability of a partial agonist for use as an antagonist drug. The ratio ID50/K_e, which has been named effective antagonist potency, or P_a, together with the recovery rate constant, K_r, would seem to give a satisfactory description of the kinetic properties of "narcotic agonists" and "narcotic antagonists."

A compound of interest is naloxone, which is the only true antagonist because it is almost devoid of agonist activity, at least in the guinea-pig ileum; it promises to be useful in the elucidation of the mode of action of morphine-like drugs. Naloxone has been shown to be a competitive inhibitor of the agonist activity not only of "narcotic agonists" such as morphine and levorphanol, but also of the agonist activity of the "narcotic antagonist," nalorphine. Further, by the use of naloxone it has been possible to confirm an earlier observation (Gyang & Kosterlitz, 1966) that the receptors of the two inhibitory drugs, morphine and adrenaline, are quite separate and independent.

While it is possible to use naloxone in high concentration and thus obtain dose ratios of well over 200, the use of "narcotic antagonists" with high agonist activity is limited by their agonist action. The dose-ratios which can be obtained with these latter compounds are therefore much lower than those possible with naloxone. A measure of the usefulness of a compound as an antagonist can be obtained from the value of $P_a + 1$,

which indicates the dose-ratio produced by a concentration having an agonist effect equal to ID50.

In the assessment of a drug as an antagonist in isolated tissues, effective antagonist potency and rate of recovery from the antagonist action have to be taken into account. A compound with a relatively low effective antagonist potency and a low rate of recovery, for example cyclazocine, may be more useful than a compound with a higher effective antagonist potency and a higher rate of recovery, for example nalorphine. The most useful compound will be one with little or no agonist activity and a low rate of dissociation. When used in the whole animal or in man, there will be additional considerations such as rate of absorption, distribution of the drug and rate of metabolic inactivation.

The findings obtained on the guinea-pig isolated ileum agree well with observations made on man and animals *in vivo*. Thus while nalorphine and cyclazocine exhibit agonist activity, for example, produce analgesia and respiratory depression in man (Martin, Fraser, Gorodetzy & Rosenberg, 1965; Martin & Gorodetzky, 1965), naloxone has no such agonist actions (Foldes *et al.*, 1963; Jasinski *et al.*, 1967). Similarly, morphine, nalorphine and cyclazocine depress the flexor reflex in the spinal dog, whereas naloxone has no inhibitory effect but sometimes potentiates this reflex (McClane & Martin, 1967). Discrepancies between findings on the guinea-pig ileum and observations on the whole animal are found only occasonally; one interesting example is the very potent antagonist, M 5050, which exhibits agonist activity but has, as Blane (1967) has shown, no antinociceptive activity in the phenylbenzoquinone test in mice.

It is tempting to speculate about a possible correlation between the agonist and antagonist potencies of these compounds and their liability to cause addiction. In man, physical dependence of some kind seems to be caused by all narcotic drugs which have agonist properties; this has recently been shown for the "narcotic antagonists," nalorphine and cyclazocine (Martin *et al.*, 1965; Martin & Gorodetzky, 1965). Of the compounds listed in Table 1 the following are not likely to be used as drugs of addiction: (1) the mild analgesics, codeine (Krueger, Eddy & Sumwalt, 1943), pentazocine (Fraser & Rosenberg, 1964) and CI 572 (Winder *et al.*, 1966) and (2) the "narcotic antagonists," nalorphine (Eddy, Halbach & Braenden, 1956, 1957), *N*-methylallylnormorphine (Telford *et al.*, 1961), levallorphan (Eddy *et al.*, 1956), cyclazocine (Martin *et al.*, 1965) and naloxone (Jasinski *et al.*, 1967). Thus the impression is gained that compounds will have only a small liability to cause addiction when any of the following conditions is fulfilled:

1. Absence of agonist activity, for example naloxone.

2. Low agonist activity even when combined with low effective antagonist potency, for example weak analgesics, codeine, pentazocine and CI 572.

3. High agonist potency when associated with high effective antagonist potency, for example nalorphine and cyclazocine.

SUMMARY

1. The agonist and antagonist properties of "narcotic agonists" and "narcotic antagonists" were characterized by their action on the contractions of the longitudinal muscle of the guinea-pig isolated ileum. The agonist activity was measured by the

concentration which causes 50% depression of the twitch induced by coaxial stimulation (ID50) and the antagonist activity by the equilibrium constant (K_e) and the recovery rate constant (K_r).

2. Since all " narcotic agonists " and most " narcotic antagonists " exhibit both agonist and antagonist actions, the effective antagonist potency ($P_a = ID50/K_e$) was calculated to obtain a measure of the antagonist activity (dose ratio - 1) at a concentration equal to ID50. " Narcotic agonists " have P_a values <2 and " narcotic antagonists " have $P_a > 2$.

3. The usefulness of a narcotic drug as an antagonist is enhanced by a high effective antagonist potency and a low recovery rate.

4. The morphine derivative, naloxone, is the only drug examined so far which has little or no agonist activity and can therefore be used as a pure antagonist. The antagonism by naloxone of the agonist actions of the " narcotic agonists," morphine, levorphanol, codeine and CI 572, and of the agonist action of the " narcotic antagonist," nalorphine, has been shown to be of a competitive nature.

5. The receptors for the depressant actions of adrenaline and morphine are separate and independent of each other.

This investigation was supported in part by U.S. Public Health Service Grant NB 03026, National Institute of Neurological Diseases and Blindness. Acknowledgement is made of generous gifts of drugs: dextromoramide (M.C.P. Pure Drugs Ltd.), levorphanol and levallorphan (Roche Products Ltd.), phentazocine, NIH 7958 (Dr. N. B. Eddy, National Institutes of Health, Bethesda and Bayer Products Ltd.), cyclazocine, NIH 7981 (Dr. N. B. Eddy), naloxone (Endo Laboratories Inc.), *N*-methylallylnormorphine, NIH 5706 (Dr. N. B. Eddy and Merck, Sharp & Dohme Research Laboratories), CI 572 (Parke, Davis & Co. Ltd.), M 5050 (Reckitt & Sons Ltd.), hexamethonium bromide (May & Baker Ltd.) and propranolol (Imperial Chemical Industries Ltd.).

REFERENCES

BLANE, G. F. (1967). Blockade of bradykinin-induced nociception in the rat as a test for analgesic drugs with particular reference to morphine antagonists. *J. Pharm. Pharmac.*, **19**, 367–373.

BLUMBERG, H., DAYTON, H. B. & WOLF, P. S. (1966). Counteraction of narcotic antagonist analgesics by the narcotic antagonist naloxone. *Proc. Soc. exp. Biol. N.Y.*, **123**, 755–758.

EDDY, N. B., HALBACH, H. & BRAENDEN, O. J. (1956). Synthetic substances with morphine-like effect. Relationship between analgesic action and addiction liability, with a discussion of the chemical structure of addiction-producing substances. *Bull. Wld Hlth Org.*, **14**, 353–402.

EDDY, N. B., HALBACH, H. & BRAENDEN, O. J. (1957). Synthetic substances with morphine-like effect. Clinical experience: Potency, side-effects, addiction liability. *Bull. Wld Hlth Org.*, **17**, 569–863.

FOLDES, F. F., LUNN, J. N., MOORE, J. & BROWN, I. M. (1963). N-allylnoroxymorphone: a new potent narcotic antagonist. *Am. J. med. Sci.*, **245**, 23–30.

FRASER, H. F. & ROSENBERG, D. E. (1964). Studies on the human addiction liability of 2'-hydroxy-5,9-dimethyl-2-(3,3-dimethylallyl)-6,7-benzomorphan (Win 20228), a weak narcotic antagonist. *J. Pharmac. exp. Ther.*, **143**, 149–156.

GYANG, E. A. & KOSTERLITZ, H. W. (1966). Agonist and antagonist actions of morphine-like drugs on the guinea-pig isolated ileum. *Br. J. Pharmac. Chemother.*, **27**, 514–527.

JASINSKI, D. R., MARTIN, W. R. & HAERTZEN, C. A. (1967). The human pharmacology and abuse potential of N-allylnoroxymorphone (naloxone). *J. Pharmac. exp. Ther.*, **157**, 420–426.

KRUEGER, H., EDDY, N. B. & SUMWALT, M. (1943). Codeine. In *The Pharmacology of the Opium Alkaloids*, part 2. U.S. Public Health Reports, suppl. No. 165, p. 817–852. Washington: U.S. Government Printing Office.

LASAGNA, L. (1965). Drug interaction in the field of analgesic drugs. *Proc. R. Soc. Med.*, **58**, 978–983.

MCCLANE, T. K. & MARTIN, W. R. (1967). Effects of morphine, nalorphine, cyclazocine and naloxone on the flexor reflex. *Int. J. Neuropharmac.*, **6**, 89–98.

MARTIN, W. R., FRASER, H. F., GORODETZKY, C. W. & ROSENBERG, D. E. (1965). Studies on the dependence-producing potential of the narcotic antagonist 2-cyclopropylmethyl-2'-hydroxy-5,9-dimethyl-6,7-benzomorphan (cyclazocine, Win-20,740, ARC II-C-3). *J. Pharmac. exp. Ther.*, **150**, 426–436.

MARTIN, W. R. & GORODETZKY, C. W. (1965). Demonstration of tolerance to and physical dependence on N-allylnormorphine (nalorphine). *J. Pharmac. exp. Ther.*, **150**, 437–442.

PATON, W. D. M. (1955). The response of the guinea-pig ileum to electrical stimulation by coaxial electrodes. *J. Physiol., Lond.*, **127**, 40–41*P*.

PATON, W. D. M. (1961). A theory of drug action based on drug-receptor combination. *Proc. R. Soc.* B, **154**, 21–69.

SCHAUMANN, W. (1958). Zusammenhänge zwischen der Wirkung der Analgetica und Sympathicomimetica auf den Meerschweinchen-Dünndarm. *Naunyn-Schmiedeberg's Arch exp. Path. Pharmak.*, **233**, 112–124.

STEPHENSON, R. P. (1956). A modification of receptor theory. *Br. J. Pharmac. Chemother.*, **11**, 379–393.

TELFORD, J., PAPADOPOULOS, C. N. & KEATS, A. S. (1961). Studies of analgesic drugs. VII. Morphine antagonists as analgesics. *J. Pharmac. exp. Ther.*, **133**, 106–116.

WINDER, C. V., WELFORD, M., WAX, J. & KAUMP, D. H. (1966). Pharmacologic and toxicologic studies of *m*-(1-methyl-3-propyl-3-pyrrolidinyl)phenol (CI-572), an analgetic and antitussive agent. *J. Pharmac. exp. Ther.*, **154**, 161–175.