ties in Eq. 2 to yield exact functions of the sublattices.

An alternative expression, Eq. 5, for the activities in the F-crystal is suggested by the law of mass action in which the concentration of each species is raised to the power of the stoichiometric coefficient of the sublattice. Thus we are inclined to try

$$a_A^F = (X_A^\alpha)^{1-p}\ (X_A^\beta)^p \qquad (7)$$

and

$$a_B^F = (1 - X_A^\alpha)^{1-p}\ (1 - X_A^\beta)^p \qquad (8)$$

An expression for the equilibrium constant for reaction No. 1 above may be deduced from the data of the heterogeneous system using the expressions Eq. 2 and Eq. 3, if the activity coefficients in these expressions can also be deduced. If, in addition $f_A^G = f_B^G = 1$, we have from Eqs. 1, 3, 7, and 8:

$$K_a = \frac{(X_B^\alpha)^{1-p}\,(X_B^\beta)^p\,(X_A^{\beta'})}{(X_A^\alpha)^{1-p}\,(X_A^\beta)^p\,(X_B^{\beta'})} \qquad (9)$$

If now it can be assumed that the sublattices β and β' are energetically identical with respect to A and B, one may set

$$X_A^{\beta'} = X_A^\beta,\ X_B^{\beta'} = X_B^\beta \qquad (10)$$

since

$$X_A^G = X_A^{\beta'},\ X_B^G = X_B^{\beta'}$$

Whether or not Eq. 10 is a valid assumption will depend to a large extent on the degree of structural homology possessed by β and β'. It would seem that this requirement would be quite strict for ionic crystals because of the relatively long-range forces involved.

In the absence of the requisite strict homology, relations such as the following might be found useful:

$$X_A^{\beta'}\,L_A^{\beta'} = X_A^\beta\,L_A^\beta \qquad (11)$$

L_A^β and $L_A^{\beta'}$ in these expressions being functions of the sublattices analogous to activity coefficients.

If, however, the expressions in Eq. 10 hold, we have by substitution into Eq. 9

$$K_a^{1/(1-p)} = \frac{(1 - X_A^\alpha)\,X_A^\beta}{(1 - X_A^\beta)\,X_A^\alpha} \qquad (12)$$

which is the simplest possible distribution relation for A and B among the sublattices α and β of the F-crystal. The expression No. 1 is thus a simplified alternative expression to the more complex exact expression for the distribution obtainable from Eqs. 2, 3, and 6. The two types of expressions may, of course, be compared to each other and with experimental data such as that derived from x-ray diffraction studies.

A system which has been investigated (*3*, *4*) is comprised of the two naturally coexisting monoclinic amphiboles cummingtonite and actinolite in which Mg^{2+}, Fe^{2+}, and Ca^{2+} correspond to A, B, and C. The nonparticipating framework $Si_8O_{22}(OH)_2$ is virtually identical in both minerals as shown by x-ray diffraction. The formulas are:

$$(Mg,\ Fe)_2^V\ (Mg,\ Fe)_5^W\ Si_8O_{22}(OH)_2$$
(cummingtonite)
$$Ca_2^V\ (Mg,\ Fe)_5^W\ Si_8\ O_{22}\ (OH)_2 \text{ (actinolite)}$$

in which the sublattices are distinguished by the superscripts V and W. Because of differences in ionic radii Ca^{2+} is almost entirely restricted to the V sublattice of actinolite, whereas Mg^{2+} and Fe^{2+} are distributed among the V and W sublattices of cummingtonite and the W sublattice of actinolite. The W sublattice of both minerals consists of Mg^{2+} and Fe^{2+} ions surrounded by six oxygen ions in octahedral coordination. The mean metal-oxygen interionic distance varies only 0.03 A from cummingtonite (*5*) to actinolite (*6*). The V sublattice, on the other hand, consists of a highly distorted polyhedron in which the interionic distances vary as much as 0.30 A and the mean interionic distance as much as 0.15 A between the two minerals.

Because of the close structural homologies of the W sublattices of the two minerals, a consequence largely of the identity of the Si-O framework, a relation such as Eq. 10 was assumed to hold. The observed distribution of Mg^{2+} and Fe^{2+} among the coexisting actinolites and cummingtonites is presented in Fig. 1. The curve which is fitted to these points is also substantiated by additional data (*3*). The derived distribution constant is $K = 1.80$ (*3*). The corresponding calculated distribution among the V and W sublattices is presented in Fig. 2. In this figure the full curve is the more exact one and was derived from relations of the type of Eqs. 2, 3, and 6. The dashed curve shows the simple approximation analogous to Eq. 1, the equilibrium constant being $(1.80)^{7/8}$, as may easily be shown. Comparison of Figs. 1 and 2 shows how Mg^{2+} is concentrated in the actinolite-like W sublattice of cummingtonite. Unfortunately, only one independent determination of this distribution exists for cummingtonite. This distribution, obtained from x-ray diffraction by Ghose (*5*), is shown in Fig. 2 as a cross. The close correspondence with the theoretical curves may be somewhat fortuitous, but the range in which the point falls seems to favor the model.

Quantitative applications of the method require rather close homologies between the structures involved. However, it seems probable that the theory might provide semiquantitative or qualitative predictions about distributions in a variety of crystals. Such predictions may be of considerable utility in the interpretation of x-ray diffraction and other data. I am reporting one such semiquantitative application, in this case to the pyroxene minerals (*4*).

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Narcotic Antagonists as Analgesics

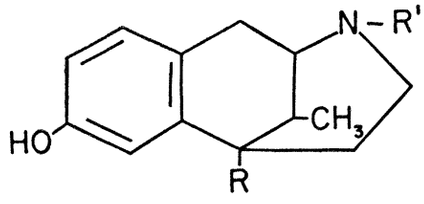
Abstract. 2-Dimethylallyl-5,9-dimethyl-2′-hydroxybenzomorphan (Win 20,228) was found to be a weak antagonist of morphine and meperidine, whereas 2-allyl-5-ethyl-2′-hydroxy-9-methyl-6,7-benzomorphan (Win 19,362) and 2-allyl-2′-hydroxy-5,9-dimethyl-6,7-benzomorphan (Win 19,631) were about three times as potent as nalorphine. Preliminary clinical trials indicate that Win 19,362 is about twice as potent as morphine as an analgesic but, like nalorphine, is capable of producing severe psychic side effects. Win 20,228 is estimated to be about half as active as morphine as an analgesic, but no bizarre psychic effects were reported at any dose tested. Studies of addiction liability with Win 20,228 in monkeys suggest that this compound will not support morphine addiction.

Lasagna and Beecher (*1*) and Keats and Telford (*2*) showed that nalorphine (N-allylnormorphine), which is a potent antagonist of morphine clinically and in experimental animals, is, in itself, a potent analgesic in man. The subsequent report by Isbell (*3*) that this drug was unable to support morphine addiction or induce primary addiction in man broke the bond which previously linked strong analgesia with addiction liability. Nalorphine produced serious side effects, particularly of a psychic nature, and thus proved to be unacceptable as an analgesic agent. Further studies by Keats and his group (*4*) on a series of antagonists de-

rived from the morphine and morphinan ring systems revealed that many of these antagonists were effective analgesics in man but that none met the criteria of a clinically useful drug.

The report (*5*) that Phenazocine (I) was a potent analgesic in man, albeit capable of replacing morphine in stabilized addicts, prompted us to turn our attention to the investigation of antagonists derived from the benzomorphan nucleus in the hope that a clinically acceptable strong analgesic devoid of addiction liability could be found in such a series.

To this end a number of benzomorphan derivatives were prepared and subjected to pharmacological evaluation.



I R = CH_3-, R' = $C_6H_5CH_2CH_2$-
(Phenazocine)

II R = CH_3-, R' = $CH_2=CHCH_2$-
(Win 19,631)

III R = CH_3-, R' = $(CH_3)_2C=CHCH_2$-
(Win 20,228)

IV R = C_2H_5-, R' = $CH_2=CHCH_2$-
(Win 19,362)

The parent nucleus was prepared by a modification of the method previously described (*5*). This secondary base was in turn treated with the appropriate halide to furnish the desired compounds. In this report we wish to confine our attention to three of these: II (m.p. 141.2 to 143.8°C), III (m.p. 145.4 to 147.2°C), and IV (m.p. 159.8 to 162.8°C) (*6*). During the course of this work Gordon *et al.* (*7*) independently reported the synthesis and antagonist activity of II. Our chemical and biological results are in essential agreement. The interest of Gordon *et al.* was directed toward combinations of Phenazocine (I) with II based on evidence in animals that such mixtures reduced the addiction liability of I without affecting its analgesic potency.

The reversal of morphine- and meperidine-induced analgesia proved to be the most convenient way of quantitatively comparing the antagonist activity in this series. A modification of the rat tail-flick method of D'Amour and Smith and Bass and VanderBrook (*8*) was used. Groups of animals were medicated (subcutaneously) with logarithmically increasing doses of the antagonist 10 minutes prior to receiving a standard dose of morphine or meperidine. The dose of analgesic chosen was one that consistently produced an 80-percent analgesic effect which was taken as the maximum possible effect or 100 percent. The difference between this maximum possible effect and the percentage effect of the combination of the analgesic and the antagonist was considered to be due to the effect of the antagonist. This percentage effect was plotted against the dose of the antagonist on log-probit paper to obtain an AD_{50}, that is, the dose which caused a 50-percent antagonism of the maximum possible effect of the analgesic employed. Win 19,362 and Win 19,631 were approximately three times as potent as nalorphine as antagonists of the analgesic effects of meperidine. Win 20,228 was considerably weaker, being about 1/50th as active as nalorphine as a meperidine antagonist. These antagonists by themselves were not capable of blocking the tail-flick response and therefore were not analgesics by this criterion.

In dogs, Win 19,362 was able to reverse the cardiovascular, respiratory, and behavioral depression induced by morphine. In monkeys, low doses of Win 19,362 produced mild sedation while high doses caused tremors, salivation, prostration and convulsions. This syndrome closely resembles that seen with nalorphine. Win 20,228 behaved similarly in the monkey at about twice the dose. In dogs this compound poorly but definitely reversed the respiratory, cardiovascular, and behavioral effects induced by morphine (*9*).

Since it had been shown previously that there is no real correlation between antagonistic potency in animals and analgesia in man (*4*), a weak (Win 20,228) and strong (Win 19,362) antagonist were selected for human trials. These were carried out according to the procedure of Keats and Telford (*4*, *10*). It was estimated that the analgesic effect of 5 mg/70 kg of Win 19,362 was approximately equivalent to 10 mg/70 kg of morphine in postoperative patients. However, in preoperative patients doses of 5 mg/70 kg and 10 mg/70 kg produced severe side effects including marked sedation, nausea, and psychic symptoms similar to those produced by nalorphine. The psychic symptoms were also noted in postoperative patients at higher doses.

A total of 59 postoperative patients received 127 paired doses of Win 20,228 and placebo. The doses used were 5, 10, 20, and 40 mg/70 kg. All doses of Win 20,228 showed analgesia greater than that of a placebo. In an additional group of 18 patients in whom Win 20,228 was compared with morphine, the analgesia of 20 mg/70 kg of Win 20,228 was equal to or slightly greater than that of 10 mg/70 kg of morphine. Only 3 of the 77 patients who received Win 20,228 had side actions of sufficient magnitude to be recorded. One patient had marked sedation, another had nausea and vomiting, and a third was restless, probably secondary to unrelieved pain. The subjective effects produced by Win 20,228 in preoperative patients were much less marked than those noted with Win 19,362 in similar patients. None of the 44 preoperative patients who received Win 20,228 in doses of 10, 20, or 40 mg/70 kg reported the nalorphine-like psychic effects of Win 19,362. In these patients Win 20,228 produced primarily sedation, which increased with dose, and dizziness. Nonspecific sedative effects such as difficulty in concentration, grogginess, and difficulty in focusing eyes were also noted.

Preliminary studies in healthy subjects indicate that 20 mg/70 kg of Win 20,228 produces respiratory depression, similar to that produced by 10 mg/70 kg of morphine (*4*). Preliminary studies also indicate that 20 mg/70 kg of Win 20,228 partially antagonizes the respiratory depression of 1 mg/kg of morphine given intravenously to anesthetized patients. The drug appears to be a weak morphine antagonist in man, confirming the results seen in animals.

Deneau and Seevers (*11*) estimated the physical dependence capacity of Win 20,228 in morphine-dependent monkeys to be nil. At present the addictiveness of Win 20,228 is being studied at the Addiction Research Center at Lexington. If the findings in monkeys are borne out by subsequent studies in man, Win 20,228 shows promise of being a clinically acceptable strong analgesic comparable to morphine, without the addiction liability associated with the latter drug.

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Recognition and Quantitation of Herpesvirus Particles in Human Vesicular Lesions

Abstract. Herpesvirus particles from crude vesicular fluid of a patient were stained with uranyl acetate and potassium phosphotungstate, and then were identified and counted by electron microscopy. Virus was seen and quantitated in all the samples taken from five vesicles. Specimens from human lesions can be prepared and examined within 3 hours, permitting rapid presumptive identification of herpesvirus.

Van Rooyen and Scott (*1*) observed elementary bodies of smallpox, taken from pustular material, directly in the electron microscope and suggested the use of such examination as an aid in diagnosis. Evans and Melnick (*2*) described the appearance of herpes simplex, varicella and herpes zoster particles obtained from vesicular and spinal fluids. Melnick *et al.* (*3*) found characteristic particles in purified extracts of human papillomas (warts) and molluscum contagiosum lesions. All of these viruses were prepared for microscopy by metal shadowing. Almeida *et al.* (*4*) recently described the use of negative staining for the recognition of varicella virus particles from vesicle fluid. Human papova virus can be identified and quantitated directly in crude tissue homogenates (*5*). Smith and Melnick (*6*) described a method for identifying the nucleic acid type of viruses in a variety of crude materials by staining and electron microscopy. Stains which reveal identifying structures of viruses are very helpful in examining crude materials which nearly always contain substantial amounts of tissue debris. The report presented here illustrates the combined advantages of staining, to facilitate particle recognition, and counting of these particles in the study of crude unpurified vesicle fluids from a patient with a herpetic infection.

A 43-year-old male suffering from Hodgkins disease became seriously ill with a disease resembling generalized herpes. Numerous large vesicular lesions appeared over his body. Vesicular fluid was aspirated from five different lesions with fine needles and syringes. Very small volumes were obtained, about 0.01 to 0.02 ml per lesion. Each fluid was washed from the needle by withdrawing and expelling 2.0 ml of tissue culture fluid. Tissue cultures (human lung fibroblast and cercopithecus monkey kidney epithelial cells) were inoculated and were observed to develop cytopathogenic effects typical of herpesvirus within 24 hours (microplaques). Specific antiherpes serum inhibited the development of this effect.

The diluted fluids were each treated with a trypsin-chymotrypsin mixture to digest cell debris, then were diluted further and sedimented on agar in the ultracentrifuge as described previously (*6*). The collodion pseudoreplicas from these agar blocks were stained with 0.2 percent uranyl acetate or 0.2 percent potassium phosphotungstate, or both (*7*). The preparations were then examined with an RCA-EMU-3-F microscope.

Numerous particles resembling herpesvirus (*7*, *8*) were seen in each of the five specimens. Figure 1 (top) shows the morphology of these particles following the treatment with uranyl acetate. The particles are somewhat angular in outline, and they display a lightly stained capsid and a densely stained central core. Some structural detail can be seen around the less densely stained periphery of each particle. Over 80 percent of the particles stained in the core area and were therefore presumed to contain a nucleic acid core. This conclusion was supported by the observation that only a small fraction were penetrated and stained in the core following phosphotungstate treatment.

Two virus particles stained by phosphotungstate are shown in Fig. 1 (middle). These particles show the surface structure much more clearly than do the particles stained with uranyl acetate (Fig. 1, top). There is some evidence of collapse of particles treated with phosphotungstate, as manifested by the rather ragged periphery. Figure 1 (bottom) shows a particle stained first

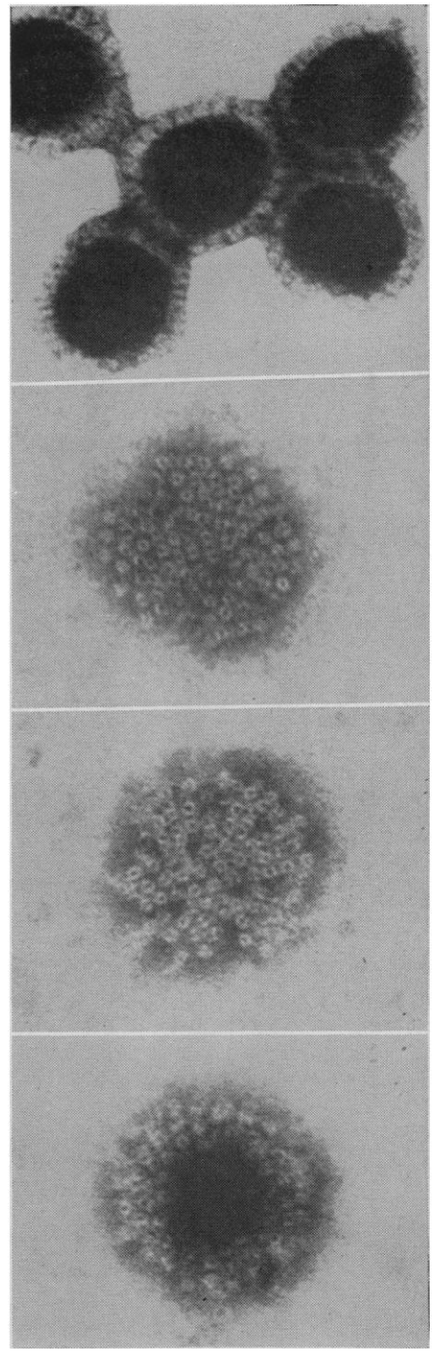

Fig. 1. Herpesvirus particles obtained directly from human vesicular fluid. (Top) Group of five virus particles stained with uranyl acetate (× 230,000). (Middle) Two virus particles stained with potassium phosphotungstate at *p*H 7.0 (× 230,000). (Bottom) Virus particle stained with uranyl acetate, then counterstained with potassium phosphotungstate (× 260,000).