Further oxidation of 3.2 g. of the intermediate in 10 cc. of solvent was conducted with 1 g. of chromic anhydride at 60–90°, and the collected product (2 g.), being part liquid and part solid, was oxidized again as before, when an exothermic reaction was noted at 80–90°. The material recovered by ether extraction as a yellow oil crystallized readily from alcohol after obtaining seed and formed long, rectangular, bright yellow plates, m. p. 28–29.5°, identical with the sample described above.

Anal. Calcd. for $C_{16}H_{14}O_2$: C, 80.65; H, 5.92. Found: C, 80.53; H, 6.02.

Summary

Details are given of observations summarized in two recent Communications. The chemical properties and absorption spectra of synthetic model compounds and the marked antihemorrhagic activity of at least one of these substances (2,3-dimethyl-1,4-naphthoquinone, assayed by the Almquist procedure) lends support to the formulation of vitamin K_1 as 2-methyl- (or ethyl)-3-phytyl-1,4-naphthoquinone and of vitamin K_2 as 2,3-difarnesyl-1,4-naphthoquinone.

A theoretical interpretation is given of the purple-blue color reaction of β-unsaturated alkyl naphthoquinones with sodium ethylate and it is shown that the reaction involves the replacement of the unsaturated side chain by hydroxyl. This accounts for the formation of a phthiocol-like pigment as the end-product of the color reaction with vitamin K concentrates, and the pigment probably is phthiocol or the ethyl homolog. The phthiocol isolated from human tubercle bacilli may have arisen from the alkaline cleavage of a K-type vitamin.

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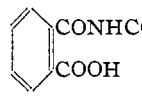
[CONTRIBUTION FROM THE DEPARTMENT OF PHYSIOLOGICAL CHEMISTRY, COLLEGE OF MEDICINE THE OHIO STATE UNIVERSITY]

A Study of Some Derivatives of Phthalyl Urea

BY CLAYTON S. SMITH AND CHESTER J. CAVALLITO

Many types of derivatives of urea have been prepared, most of them in the search for compounds possessing hypnotic activity. A large number of substituted ureas possess some degree of hypnotic activity.[1] The most active hypnotics are found among the cyclic ureas, especially the ureides of substituted malonic acid.

The object of the present work was to synthesize derivatives of phthalyl urea, the cyclic ureide of phthalic acid, and to study them for hypnotic activity.

Phthalyl urea was first described by A. Piutti.[2] He prepared the compound by heating equimolecular quantities of phthalic anhydride and urea to 120–125°, forming first the intermediate acyclic ureide of phthalic acid, 

which when treated with phosphorus oxychloride yielded the closed ring structure.

In the present work, the general method of Piutti was used for the synthesis.

(1) A. Hjort, E. J. de Beer and co-workers, *J. Pharmacol.*, **52**, 211 (1934), to *ibid.*, **61**, 175 (1937).

(2) A. Piutti, *Ann.*, **214**, 17 (1882); *Gazz. chim. ital.*, **12**, 169 (1882).

Experimental Procedure

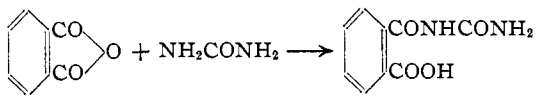
Preparation of Phthalyl Urea and Phthalyl Thiourea.—Equimolecular quantities of phthalic anhydride and urea were ground together in a mortar, then transferred to a round-bottomed flask. The mixture was heated at about 124° on a sulfuric acid bath until the reaction mixture became pasty and finally solidified to a hard mass. The melt should not be heated to such a temperature as to cause the evolution of gas bubbles. The product was pulverized, washed several times with cold water and ether, then recrystallized from hot water (about 35% yield).

Ring closure was brought about by treating the acyclic ureide with enough phosphorus oxychloride to form a thin paste and warming slowly on a water-bath. The reaction was considered complete when hydrogen chloride fumes were no longer liberated. Ether was then added to the reaction mixture, and the product filtered, washed with ether, and recrystallized from hot water.

Phthalyl urea decomposes at 185 to 190° leaving a residue which melts partly at about 230°, and completely above 300° with decomposition. Phthalimide and cyanuric acid $(CONH)_3$ are present as decomposition products.[2]

Phthalyl thiourea was prepared by the same general procedure as described above. The intermediate acyclic ureide did not form as rapidly in the case of thiourea as with urea.

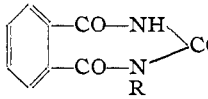
Attempts to prepare the cyclic ureides by treating phthalic anhydride and urea directly with phosphorus oxychloride were unsuccessful. It appears necessary first to bring about the combination



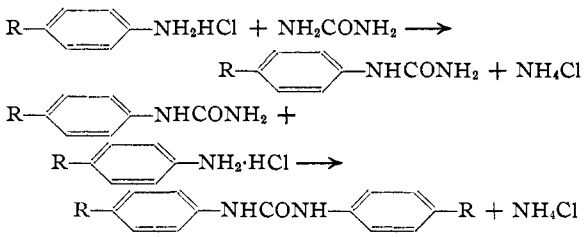
by fusing the two components.

Δ^2-Tetrahydrophthalyl Urea.—This compound was prepared from urea and Δ^2-tetrahydrophthalic anhydride. Tetrahydrophthalic acid was prepared by reducing phthalic acid with sodium amalgam in a strongly alkaline solution.[3] The acid was purified and treated with acetyl chloride to form the anhydride. Δ^2-Tetrahydrophthalic anhydride combined with the urea less readily than did phthalic anhydride, as measured by the time necessary for the fused mixture to solidify. Ring closure was brought about by treatment with phosphorus oxychloride as described above.

N-Substituted Phthalyl Ureas.—This type of derivative was prepared by starting with phthalic anhydride and a monosubstituted urea. Compounds of the type



were prepared, where R consisted of the following groups: methyl, allyl, phenyl, *o*-tolyl, *m*-tolyl, *p*-tolyl, *o*-phenetyl, *p*-phenetyl, *o*-anisyl, and *p*-anisyl. The aromatic substituted ureas used were prepared by heating an aqueous solution of urea with the corresponding aromatic amine hydrochloride.[4]

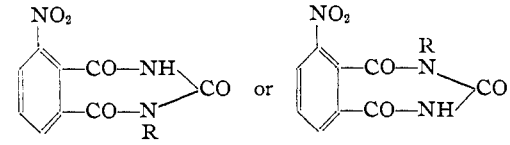


The monosubstituted urea formed by the above reaction in about 50% yield was separated from the disubstituted urea by solution of the former in hot water. The alkyl substituted ureas were purchased from the Eastman Kodak Company.

The monosubstituted ureas combined with phthalic anhydride more slowly than did urea. Attempts to combine phthalic anhydride with N,N¹-disubstituted ureas by the fusion method were unsuccessful. It may be that the high temperature necessary to fuse this mixture is above the decomposition temperature of the expected product.

Nitrophthalyl Ureas. 3-Nitrophthalyl urea and 4-nitrophthalyl urea were prepared from urea and the corresponding 3- and 4-nitrophthalic anhydrides. The nitro acids were prepared by direct nitration of phthalic anhydride[4] (p. 399),[3] (p. 828). The nitro anhydrides were prepared by treating the acids with acetic anhydride. 3-Nitrophthalyl derivatives of *p*-tolyl urea and of *p*-phenetyl urea were also prepared. The nitro anhydrides reacted with the ureas more rapidly upon fusion than did phthalic anhydride.

These last two compounds may possess either of two structures



It has not yet been determined whether both or just one of the above types are formed.

The compounds prepared are listed in Table I, together with some of their physical constants and properties. These compounds generally decompose before melting, and the decomposition points are not sharp. Phthalyl urea when heated slowly decomposes between 185–190° into phthalimide and cyanuric acid, and this mixture softens again at about 230°. If, however, the melting point is determined by lowering the temperature of the melting point apparatus from some high point, to the lowest point which will produce complete liquefaction of freshly added phthalyl urea, it will be found to melt at 207°. The melting points of the compounds were determined in this manner.

The derivatives of phthalyl urea are all relatively insoluble in cold water, but are more soluble in hot water or in hot alcohol. The solubility in hot water is greatly diminished with increase in the molecular weight of the compounds. The above phthalyl ureas are fairly soluble in alkali. This can be explained on the basis of enol formation. The alkali solubility of the nitro phthalyl ureas is greater than that of the phthalyl ureas without a nitro group.

With the exception of phthalyl allyl urea, the compounds prepared appear quite stable. The allyl derivative was observed to liberate formaldehyde in the presence of air and moisture at room temperatures, and its melting point also dropped about 10° in a few months. It is fairly stable in the absence of moisture.

The yields in the synthesis of these compounds are rather low. Practically all the loss of material takes place in the first step of the reaction. The ring closure takes place with little loss.

In order to purify some of the more insoluble substituted phthalyl ureas, several washings were made with water containing a few drops of ammonium hydroxide, and then with several portions of warm water. This procedure, when carried out after the ether washings, removed any phthalimide or non-cyclic acid ureide present.

Pharmacological Procedure

Preliminary tests for pharmacological activity of the phthalyl ureas were conducted on rats.

(3) Beilstein, 4th ed., Vol. IX, p. 770.
(4) "Organic Syntheses," Coll. Vol. I, p. 442.

TABLE I

Most of these compounds crystallize from hot water as small white needles or rods. Those indicated with an asterisk* appear as plates. The colored derivatives are: [1] light yellow, [2] orange-yellow, [3] light greenish-yellow.

Phthalyl urea deriv.	Mol. wt.	% yield	M. p., °C.	Analyses, % Calcd.	Found	Calcd.	Found
Phthalyl urea	190.06	35	207–207.5	C, 56.9	57.1	H, 3.18	3.38
Phthalyl thiourea*	206.12	49	181–181.5	S, 15.55	15.35		
Δ^2-Tetrahydro	194.10	19	270	C, 55.7	56.0	H, 5.20	5.12
3-Nitro[1]	235.10	57	190 (dec.)	C, 45.9	46.1	H, 2.14	2.30
4-Nitro[1]	235.10	61	206–207	C, 45.9	46.2	H, 2.14	2.25
N-Methyl	204.08	40	190–192	C, 58.5	58.8	H, 3.95	4.11
N-Allyl	230.09	30	135–?	C, 62.6	62.9	H, 4.38	4.53
N-Phenyl*	266.09	48	164–165 residue—194	C, 67.6	67.7	H, 3.79	3.78
N-*o*-Tolyl	280.11	18	190 (dec.) heat slow 200.5	C, 68.5	68.7	H, 4.32	4.45
N-*m*-Tolyl	280.11	38	139	C, 68.5	68.9	H, 4.32	4.51
N-*p*-Tolyl	280.11	21	155–160	C, 68.5	68.5	H, 4.32	4.47
3-Nitro, N-*p*-tolyl[1]	325.11	38	189–190	C, 59.1	59.3	H, 3.41	3.48
3-Nitro, N-*p*-phenetyl[1]	355.13	35	191–195	C, 57.5	57.5	H, 3.66	3.78
N-*o*-Phenetyl	310.13	17	220	C, 65.8	66.0	H, 4.55	4.50
N-*p*-Phenetyl[2]	310.13	42	196–198	C, 65.8	65.7	H, 4.55	4.75
N-*o*-Anisyl	296.11	20	218	C, 64.8	65.0	H, 4.08	3.94
N-*p*-Anisyl[3]	296.11	12 (low)	199	C, 64.8	64.8	H, 4.08	4.00

In order to facilitate rapid absorption of the compounds, it was decided to inject them by the intraperitoneal route. Since the *p*H necessary to dissolve the compounds in the concentrations desired was too high for animal administration, the compounds were pulverized to pass through a 100-mesh sieve, then suspended in an aqueous solution of glycerol and glucose (20 g. glucose, 20 g. glycerol per 100 cc. solution). About 2 cc. of fluid was injected into each rat, with the amount of compound to be tested in suspension. The glycerol–glucose solution alone produced a little irritation for about five minutes as evidenced by the behavior of the rat, but if the rat were killed the next day and examined, the internal organs appeared normal.

The pharmacological tests were made to determine whether the compounds possessed hypnotic activity, and to study the effect of various chemical groups upon the activity. For the sake of comparison, all the compounds were tested in doses of 500 mg. per kilogram body weight. It was observed that if the compounds possessed any activity it would be evident with this dose. Larger doses did not produce greater degrees of depression, and the absorption of larger quantities was not always complete. This was probably due to the limited solubility of these compounds. Most of the compounds did not produce complete anesthesia; therefore, some criterion for judging the degree of depression was necessary. It was observed that a rat deeply depressed but not completely anesthetized would remain on his back when so placed, and would not turn over ($^{++++}$ in Table II). A rat showing somewhat less depression would turn over if placed on his back but would remain lying on his side if so placed ($^{+++}$ in Table II). One still less depressed would not remain lying on his side but would not struggle if held in the hand in the inverted position ($^{++}$ in Table II). A normal rat (0 in Table II) or one but very slightly depressed ($^{+}$ in Table II) would struggle if held in this fashion. The above criteria merely served to compare the hypnotic activities of the compounds with one another. Table II shows the results obtained.

TABLE II

Phthalyl urea deriv.	Relative order of hypnotic activity
3-Nitro	+ + + +
N-*p*-Tolyl	+ + +
3-Nitro-, N-*p*-Tolyl	+ + +
N-*p*-Anisyl	+ +
N-*p*-Phenetyl	+ +
N-*m*-Tolyl	+
N-*o*-Anisyl	+
N-Phenyl	+
N-*o*-Tolyl	+
N-*o*-Phenetyl	+
N-Methyl	0
Δ^2-Tetrahydro	0
Phthalyl urea	0

The induction period for depression ranged from five to fifteen minutes, after which the effects of the more active compounds lasted for three to four hours.

Degrees of depression simulating natural sleep are very often observed in rats under the influence

of these compounds. Doses of 150–200 mg./kg. of the more active compounds induce sleep, from which, however, the animals may be aroused rather easily.

The compounds 4-nitrophthalyl urea, phthalyl allyl urea, and phthalyl thiourea are not included in the above table, as they are toxic with the doses required to produce depression. The minimum fatal dose for the 4-nitrophthalyl urea is between 100 and 250 mg./kg.; phthalyl allyl urea and phthalyl thiourea are fatal with a dose of about 500 mg./kg. The 3-nitrophthalyl urea is toxic with a dose of about 1000 mg./kg.

It may be observed from the above table that phthalyl urea itself is practically inactive as a hypnotic. The introduction of a nitro group in the 3-position of the aromatic nucleus increases the activity without increasing the toxicity, while a 4-nitro group increases the toxicity considerably. The introduction of a thio group in the urea molecule also increases toxicity. The partial hydrogenation of the aromatic nucleus has practically no effect in increasing hypnotic activity of phthalyl urea. The introduction of a methyl group on a nitrogen does not enhance the activity. However, if an allyl group is substituted, the activity increases. A phenyl group on the nitrogen produces greater activity than does a methyl, but less than does the allyl. A *p*-tolyl group on a nitrogen increases the activity almost as much as does the allyl group, and the former is much less toxic. If a *p*-phenetyl or *p*-anisyl group is introduced on a nitrogen, the activity is increased but not as much as by the *p*-tolyl group. The ortho compounds and the meta tolyl derivative are relatively inert.

Since 3-nitrophthalyl urea and phthalyl *p*-tolyl urea were found to produce the greatest depression, together with the least toxicity, the compound 3-nitro phthalyl *p*-tolyl urea was prepared. It proved to be, if anything, slightly less active than the phthalyl *p*-tolyl urea. 3-Nitrophthalyl *p*-phenetyl urea was also prepared and found to differ little in activity from phthalyl *p*-phenetyl urea. If chemical structure were the only deciding factor in hypnotic activity, the nitro substituted phthalyl *p*-tolyl and *p*-phenetyl ureas should be very much more active. This apparently is not the case.

Summary and Conclusions

The synthesis and properties of a series of seventeen phthalyl urea derivatives have been described.

The hypnotic activity of the compounds was tested on rats by intraperitoneal injections of suspensions of the compounds in an aqueous solution of glucose and glycerol. The derivatives of phthalyl urea prepared do not appear to be very active as hypnotics. This is of theoretical interest, since several other types of cyclic ureides such as the barbiturates are quite active as hypnotics. The low solubility of the phthalyl ureas may be a limiting factor preventing the compounds from reaching their site of action in effective concentrations.

The combination in a molecule of two groups, each of which separately increases activity, does not necessarily lead to a compound more potent than one with either of the active groups alone, as in the case of 3-nitrophthalyl urea, phthalyl *p*-tolyl urea, and 3-nitrophthalyl *p*-tolyl urea.

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