ANTIMICROBIAL AGENTS FROM HIGHER PLANTS. SYNTHESIS IN THE CANTHIN-6-ONE (6H-indolo[3,2,1-de][1,5]naphthyridin-6-one) SERIES.[R]

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A practical synthetic route has been developed by which canthin-6-one and a variety of naturally occurring and synthetic analogs have been prepared for evaluation as antimicrobial agents. Less successful and unsuccessful routes are also briefly described.

A crude extract derived from the bark of Zanthoxylum elephantiasis showed significant antibacterial activity against several microorganisms in our laboratory.[1] Fractionation of the crude extract led to the isolation of a known alkaloid, canthin-6-one (1) (6H-indolo[3,2,1-de-[1,5]naphthyridin-6-one) which turned out to be the sole active principle of this bark. Canthin-6-one had been isolated previously from this plant[2] and from a number of other species[3-8] but its antimicrobial activity had not been suspected. A number of alkaloids in the canthine family have been found in nature, e.g.: 4-OMe,[7] 4-SMe,[9] 5-OMe,[2,6] 4,5-diOMe,[10,11] and 4-OMe,5-OH.[11] Canthin-6-one has been synthesized,[9,12,13] although in very modest yields. Additional synthetic work has been reported for the 4-OMe,[7,9] 4-OH,[7,9] 5-OMe,[9,12]

R All new compounds gave satisfactory microanalyses and/or high resolution mass spectra.

5-OH,[9,12] 4-Et,[12,13] 4-Pr,[12] and 4-i-Pr[12] analogs. These syntheses also gave poor yields. Thus all of these alkaloids are rare and a satisfactory synthesis was required before they could be tested and unnatural analogs prepared.

This was accomplished based on the use of harmane-1-carboxaldehyde (4), prepared by modification of the route of Bradsher and Umans.[14] Substitution of the readily available diethylacetal of methoxyacetaldehyde for methoxyacetaldehyde resulted in a considerable enhancement of yield. Hydrolysis of the resulting 1-methoxymethyl-9H-pyrido[3,4-b]indole (2) provided the corresponding hydroxymethyl analog (3) which was oxidized with active MnO_2 to give the desired carboxaldehyde (4), mp 200-201° (EtOH) (reported[14] mp 198-200°); M^+ 196 (92%); etc. On condensation with malonic acid (Doebner reaction)[16] compound 4 gave canthin-6-one directly. The overall yield from tryptophan is 15%. Alternately, compound 4 gave 5-methoxycarbonylcanthin-6-one (5) (5-methoxycarbonyl-6H-indolo[3,2,1-de][1,5]naphthyridin-6-one), mp 185-187° (benzene); M^+ 278 (100%); λ_{max}^{MeOH} 283 nm (log ε 4.10), 308 (3.91), 272 (4.12), 263 (4.08), 234 (4.25), 208 (4.60); ν_{max}^{KBr} 1745, 1680, 1638, 1560 cm^{-1}; δ ($CDCl_3$) 4.02 (3H, s, $CO_2\underline{CH}_3$); etc., in 83% yield when reacted with dimethyl malonate in the presence of NEt_3. This was hydrolyzed to 5-carboxycanthin-6-one (6), mp 271-273°d (water); M^+ 264 (19%); ν_{max}^{KBr} 3100-2400 (br), 1730, 1718 sh, 1618, 1595 cm^{-1} etc., in 95% yield by refluxing 2 hours in 2N aqueous HCl. On heating with copper powder in anhydrous pyridine[17] under N_2 *for 5 hr. at* 80° and allowing to stand overnight, a 49% yield of canthin-6-one was obtained. This sequence gives a 19% overall yield from tryptophan.

Compound 5 has almost equivalent *in vitro* antimicrobial activity to that of canthin-6-one itself and is more conveniently available by synthesis, so a number of analogs were prepared. 5-Carboxycanthin-6-one (6) was con-

verted to its acid chloride using thionyl chloride and then reacted with the appropriate amine to give, i.a., the following amides: (7) R=NH_2, 58%, mp 287-294° (d); (7) R=NEt_2, 54%, mp 153-158°; (7) R=NH-i-Pr, 42%, mp 268-272°; (7) R=$N(CH_2)_5$, 31%, mp 248-251° (d); and (7) R=NH (2,4-dimethoxyphenyl), 33%, mp 258-263°. Unfortunately, none of these amides showed antimicrobial activity in our agar-dilution streak assay[18] so no more amides were made. A number of A-ring substituted analogs of 5 have been made using the requisite tryptophan and their properties will be described in our full paper.

The success of this sequence has made canthine analogs available in considerable quantity for the first time. A less successful synthesis was based upon tetracyclic lactam 8 (1,2,3,5,6,11-b-hexahydro-3-oxo-11H-indole-[3,4-g], mp 250-253°; reported[20] mp 253-255; M^+=226 (100%); ν_{max}^{KBr} 1650, 1618 cm^{-1}, etc.) which was prepared from tryptophan and glutamic acid by a Dakin[19] oxidation of glutamate to the unstable propionic acid β-carboxaldehyde followed by a Mannich condensation, etc. Compound 8 has been prepared in better yield by other workers.[20] Oxidation of 8 by 2 eq. of $Hg(OAc)_2$[21] gave a poor yield of 1. Several interesting mechanisms can be written for these reactions but the pathway is inferior as a method of making canthin-6-one and its analogs and has not been pursued further.

An attempt to produce the ring system of 1 via an AB-D-C construct failed due to poor yields primarily caused by the extreme lability of the indole-N-acyl bond in various intermediates. Some interesting new compounds were, however, generated so this route will be described briefly.

Indole-3-acetic acid (9) was treated with 1 eq. of NaH in DMF followed by addition of succinic anhydride to give 62% of 3-(2-methoxy-2-oxoethyl)-γ-oxo-1H-indole-1-butanoic acid (10), mp 151-153°, M^+ 289 (14%); ν_{max}^{KBr} 3300-2500 (br), 1740, 1708, 1692, 1608 cm^{-1}; δ (d_6-DMSO) 2.70 (2H, m, CH_2CO_2H),

3.20 (2H, m, $NCOCH_2$), 7.83 (1H, s, H_2), 8.20-8.42 (1H, m, H_7)*; etc. Cyclization to ketolactam 11 occurred with great difficulty, probably because the reactivity of C_2 is greatly diminished by acylation at N_1.[22] Ultimately, the acid chloride of 10, prepared by using oxalyl chloride, was cyclized successfully using anhydrous $AlCl_3$ to give a 21% yield of methyl 6,7,8,9-tetrahydro-6,9-dioxopyrido[1,2-a]indole 10-acetate (11), mp 132-133° (benzene-hexane); ν_{max}^{KBr} 1740, 1705,** 1670, 1575, 1560 cm^{-1}, etc., δ ($CDCl_3$) 3.05 (4H, m, -N-CO-CH_2CH_2-CO-); M^+ 271 (44%); λ_{max}^{MeOH} 308 (log ε 4.36), 237 (4.24) and 211 (4.30). The uv is especially characteristic of 2-acylindoles unsubstituted in the benzene ring.[22,23] Some of the byproducts of these reactions have interesting structures and chemistry and will be the subject of future papers. Cyclodehydration of carboxylic acid 10 with polyphosphoric acid succeeded in giving 11 also, but in exceedingly small yield.

Attempted ring closure to a modified canthine system using a variety of amines generally failed. The usual result was cleavage of the lactam ring to produce open chain amides (e.g., 15) instead. Catalysis[24] of the reaction with $TiCl_4$ stabilized the lactam ring to scission, but enamine formation did not occur (starting material was removed). The method of Kutney *et al.*[25] also gave ring cleavage. A hoped-for Bishler-Napieralski cyclodehydration reaction of methyl 1-[1,4-dioxo-4-[(phenylmethyl)amino]butyl]-1H-indole-3-acetate (16), mp 113-114°, ν_{max}^{KBr} 3240, 1728, 1702, 1630, 1570 cm^{-1}; δ ($CDCl_3$) 3.2 (2H, m, $NCOCH_2$), 4.38 (2H, d, J = 5.5 Hz, $NHCH_2\emptyset$); M^+ 378.1583 (1.6%); λ_{max}^{MeOH} 299 nm (log ε 3.90), 282 (3.86), 238 (4.29); etc., readily

* The peak for H_7 in derivatives bearing a lactam carbonyl at position 6 is quite characteristic because of the anisotropy of the latter group.

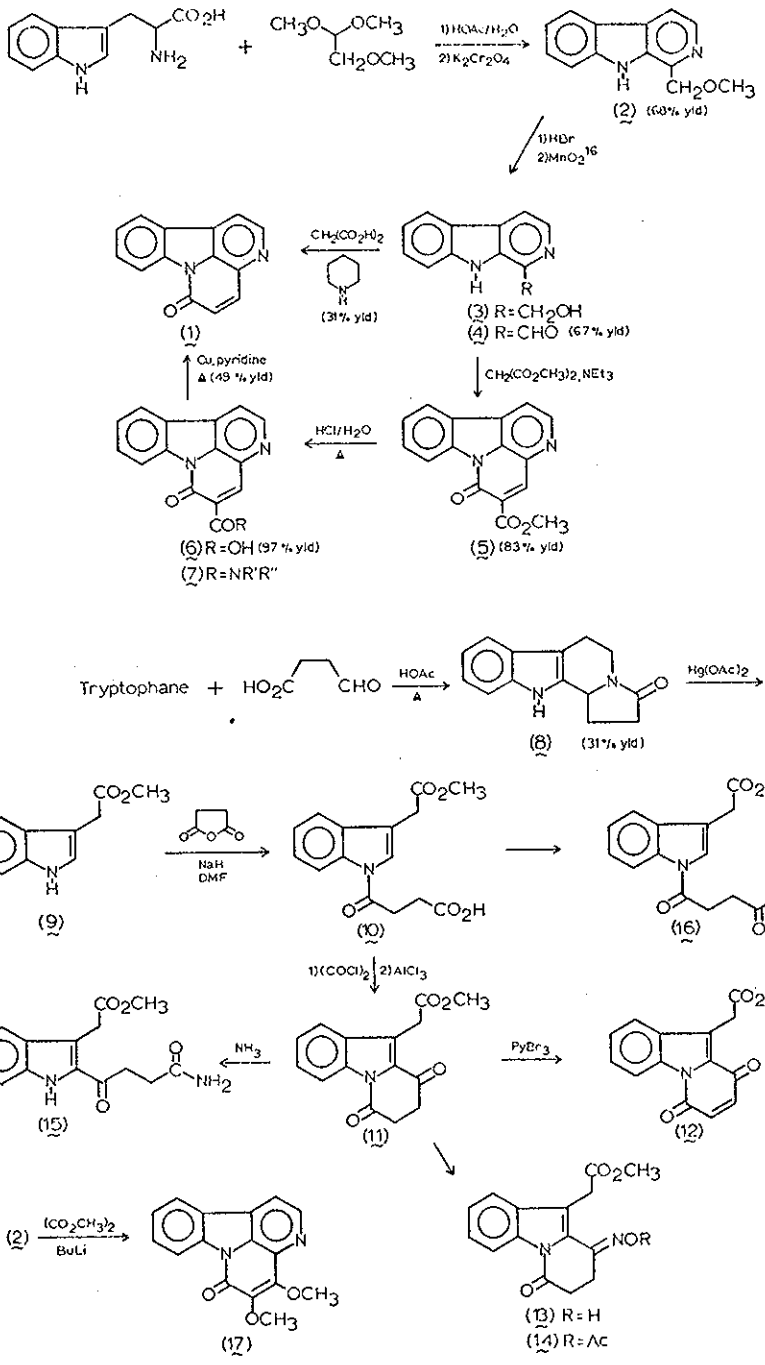
** This absorption is at higher frequency than usual for tertiary amides, suggesting ketonic character and susceptibility to nucleophilic attack.

prepared (44% yield) by reaction of the acid chloride of 10 with benzylamine at -77°, gave oily products lacking the anticipated spectroscopic properties of cyclized materials.

Ketolactam 11 readily gave an oxime on refluxing with hydroxylamine and pyridine in EtOH. The product, methyl 6,7,8,9-tetrahydro-(9-hydroxyimino)-6-oxopyrido[1,2-a]indole 10-acetate), mp 186-187° (benzene - hexane); ν_{max}^{KBr} 3460, 1726, 1685 cm^{-1}; M^+ 286.0958 (100%); etc., was formed in 80% yield and further characterized as its acetate (14), 87% yield, mp 116-117°; M^+ 328.1064; λ_{max}^{MeOH} 310 nm (log ε 4.47), 231 (4.37) and 217 (4.43); ν_{max}^{KBr} 1765, 1705 (br), 1160 cm^{-1}; δ ($CDCl_3$) 2.25 ($COCH_3$); etc. Attempts to reduce the oxime or its acetate with diborane resulted in lactam scission. Dissolving metal reductions with Zn/HOAc or Hg/Na and catalytic reductions also gave intractable oils lacking the desired spectrochemical properties, or recovered starting material. The lactam ir frequency at 1690-1710 cm^{-1} is generally lacking in these mixtures.

Dehydrogenation of 11 with pyridinium hydrobromide perbromide in glacial HOAc at 25° gave methyl 6,9-dihydro-6,9-dioxopyrido[1,2-a]indole 10-acetate (12) in 50% yield, mp 198.5-199.5° ($CHCl_3$-EtOAc); ν_{max}^{KBr} 1745, 1695, 1648, 1562 cm^{-1}; δ (TFA) 6.72 (2H, s, $COCH{=}CHCO$); M^+ 269 (31%); λ_{max}^{MeOH} 399 nm (log ε 3.86), 257 (4.16), 251 (4.15) and 217 (4.62). The shifts to lower wave length in the ir and the bathochromic shifts in the uv agree with the proposed structure and indicate that the desired oxidation to the level of canthin-6-one was achieved. No further chemistry was performed on this compound.

Finally, 4,5-dimethoxycanthin-6-one (17) was prepared in 9% yield by the reaction of 2 with n-butyl lithium and dimethyloxalate, followed by treatment of the intermediate nigakinone[11] with diazomethane. The product required extensive chromatography for purification, mp 143-146° (reported[10,11]


(2) (63% yld)
(1)
(31% yld)
(3) R=CH2OH
(4) R=CHO (67% yld)
Cu, pyridine
Δ (49 % yld)
(6) R=OH (97 % yld)
(7) R=NR'R''
(5) (83 % yld)
Tryptophane
(8) (31% yld)
(1)
(8% yld)
(9)
(10)
(16)
(15)
(11)
(12)
(13) R=H
(14) R=Ac
(17)

mp 147°) alone and admixed with an authentic sample and identical tlc behavior, nmr and ir spectra.

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