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Notes

Nitrofurfuryl Heterocycles. 12.† 4-Amino-6-(5-nitro-2-furyl)isoxazolo[5,4-*d*]pyrimidines and 4-Amino-2-(5-nitro-2-furyl)pyrimido[4,5-*d*]pyrimidines

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In two previous papers in this series it was shown that the attachment of a condensed pyrimidine ring system at the 2 position of the nitrofuran ring would give compounds possessing exceptional antibacterial activity. Those papers described the antibacterial activity of numerous 4-amino-2-(5-nitro-2-furyl)quinazoline[2] and 4-amino-6-(5-nitro-2-furyl)-1*H*-pyrazolo[3,4-*d*]pyrimidine[3] analogs. The earlier concept[2] is further exemplified by the present study on isoxazolo[5,4-*d*]pyrimidine and pyrimido[4,5-*d*]pyrimidine analogs.

The 4-amino-3-alkyl-6-(5-nitro-2-furyl)isoxazolo[5,4-*d*]pyrimidine analogs were prepared by the same sequence of reactions used to prepare the 4-amino-6-(5-nitro-2-furyl)-1*H*-pyrazolo[3,4-*d*]pyrimidines[3] except that 5-amino-4-cyanoisoxazoles were substituted for 5-amino-4-cyanopyrazoles. This synthesis is similar to the isoxazolo[5,4-*d*]pyrimidine synthesis reported by Desimoni, *et al.*[4]

The synthesis of Taylor, *et al.*,[5] for the preparation of pyrimido[4,5-*d*]pyrimidines was modified to allow for the introduction of the nitrofuryl group and is summarized in Scheme I.

Pertinent physical data for compounds **7**, **8**, and **13–19** are summarized in Table I. The antibacterial testing data, obtained by standard procedures, for these compounds are summarized in Table II. Although there are too few examples reported here to allow comment on structure–activity relationships within each class, we believe these data give further support to our previously published concept[2] for antibacterial activity.

Experimental Section

All melting points were determined in open capillaries using a Mel-Temp melting point apparatus and are corrected. Ir spectra were determined as Nujol mulls on a Perkin-Elmer Model 135 Infracord. The nmr spectrum was obtained on a Varian A-60A instrument using Me_4Si as an internal standard.

***N*-(4-Cyano-3-methyl-5-isoxazolyl)-2-furamide (1).** To a stirred solution of 940 g (7.65 mol) of 5-amino-4-cyano-3-methylisoxazole[6] in 1 l. of pyridine was added slowly 995 g (7.65 mol) of 2-furoyl chloride. After completing the addition, the solution was heated on a steam bath for 2 hr and poured into 4 l. of ice–H_2O, and the solids were removed by filtration. The solids were suspended in 2 l. of cold H_2O and the mixture was acidified with concentrated HCl. The crude product was filtered and dried to give 300 g (54.2%): mp 227.5–228° (aqueous MeOH). *Anal.* ($C_{11}H_9N_3O_3$) C, H, N.

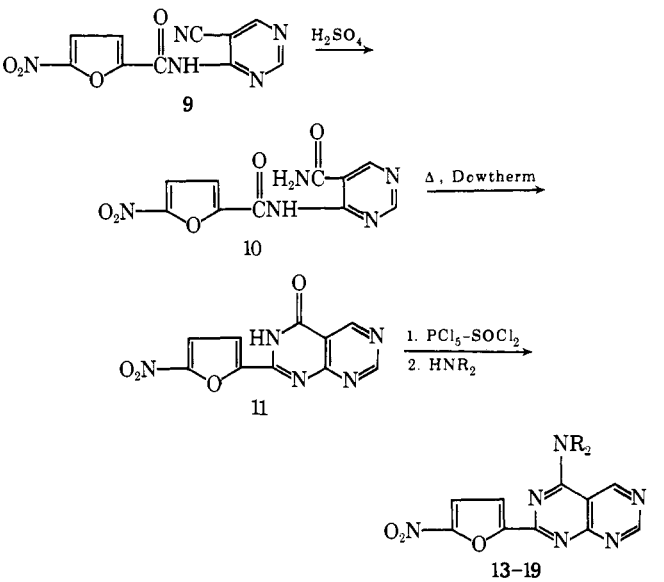
***N*-(4-Cyano-3-ethyl-5-isoxazolyl)-2-furamide (2)** was prepared similarly in 83.7% yield from 2-furoyl chloride and 5-amino-4-cyano-3-ethylisoxazole:[6] mp 171–172.5° (aqueous MeOH). *Anal.* ($C_{11}H_9N_3O_3$) C, H, N.

6-(2-Furyl)-3-methylisoxazolo[5,4-*d*]pyrimidin-4(5*H*)-one (3). Compound 1 (217 g, 1.0 mol) was added in small portions during about 1 hr to a stirred, warm (50–60°) solution of 510 g of NaOH pellets and 850 ml of 30% H_2O_2 in 2 l. of H_2O. Considerable effervescence occurred which was controlled by the periodic addition of a few milliliters of EtOAc. After the exothermic reaction had ceased (0.5–1 hr), the mixture was heated on a steam bath for 2–3 hr. The clear solution was chilled, acidified slowly with glacial AcOH, and filtered. The crude product was washed thoroughly with H_2O and dried to give 188 g (86.6%): mp 306–308° ($MeNO_2$). *Anal.* ($C_{10}H_7N_3O_3$) C, H, N.

3-Ethyl-6-(2-furyl)isoxazolo[5,4-*d*]pyrimidin-4(5*H*)-one (4) was prepared similarly in 76% yield from **2**: mp 269–269.5° ($MeNO_2$). *Anal.* ($C_{11}H_9N_3O_3$) C, H, N.

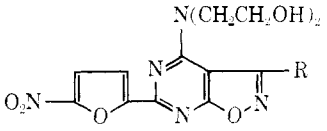
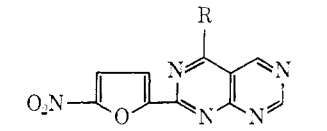
3-Methyl-6-(5-nitro-2-furyl)isoxazolo[5,4-*d*]pyrimidin-4(5*H*)-one (5). To 200 ml of concentrated H_2SO_4 was added in portions with stirring at 25–30° (external cooling needed) 72.5 g (0.33 mol) of **3**. A solution of 33 ml of concentrated HNO_3 in 66 ml of concentrated H_2SO_4 was added dropwise at 25–30° with cooling during 15 min. The temperature was kept at 25° for 10 min following the addition and then lowered to <10° for 1 hr. After pouring the mixture cautiously into 2 l. of ice–H_2O, the

Scheme I



†For the preceding paper in this series, see ref 1.

Table I

No.	R	Mp, °C	Yield, %[a]	Recrystn solvent	Formula[b]
					
7	CH_3	204–205	48	Aq DMF	$C_{14}H_{15}N_5O_6$
8	C_2H_5	185.5–187	53	$MeNO_2$	$C_{15}H_{17}N_5O_6$
					
13	$N(Me)CH_2CH_2OH$	206–207	91	$MeNO_2$	$C_{13}H_{12}N_6O_4$
14	$N(CH_2)CH_2CH_2OH$	200–201	95	DMF–MeOH	$C_{12}H_{11}N_7O_4$
15	$NHCH_2CH_2OH$	278–279	94	DMF	$C_{12}H_{10}N_5O_4$
16	$NHCH_2CH_2OCH_3$	275	79	$MeNO_2$	$C_{13}H_{12}N_6O_4$
17	c-NC_4H_8	279–280	86	$MeNO_2$	$C_{14}H_{12}N_6O_3$
18	$NHCH_2CHOHCH_3$	276–277	84	$EtOCH_2CH_2OH$	$C_{13}H_{12}N_6O_4$
19	$N(CH_2CHOHCH_3)_2$	215.5–216.5	78	$MeOCH_2CH_2OH$	$C_{16}H_{18}N_6O_5$

[a]Yields calculated before recrystallization. [b]All compounds analyzed for C, H, and N within ±0.40% of the theoretical values.

Table II. Antibacterial Testing of 4-Amino-6-(5-nitro-2-furyl)isoxazolo[5,4-*d*]pyrimidines and 4-Amino-2-(5-nitro-2-furyl)pyrimido[4,5-*d*]pyrimidines

	Minimal inhibitory concentration, μg/ml[a]											
No.	Mi-12[b]	Di-10	Er-4	StA-1	StB-12	StD-7	Es-2	Es-L	SaD-13	Ae-6	Pr-12	Ps-44
7	1.5	0.38	0.048	1.5	50	1.5	0.75	3.1	1.5	12.5	>50	>50
8	1.5	1.5	3.1	6.25	50	3.1	1.5	25	3.1	>50	>50	>50
13	6.25	1.5	0.38	3.1	6.25	12.5	0.75	6.25	3.1	25	>50	50
14	1.5	0.38	0.19	0.75	3.1	1.5	0.75	12.5	3.1	25	>50	>50
15	25	3.1	0.38	3.1	6.25	25	3.1	>50	25	>50	>50	>50
16	0.75	1.5	0.048	1.5	12.5	3.1	1.5	6.25	3.1	50	>50	>50
17	0.38	0.75	0.048	0.19	1.5	0.38	0.75	3.1	0.75	6.25	>50	>50
18	12.5	3.1	0.38	3.1	25		3.1	>50	25	>50	>50	>50
19	6.25	3.1	0.75	6.25	50		12.5	>50	25	>50	>50	>50
Nitrofurazone[c]	12.5	3.1	12.5	6	12.5	50	3	12.5	3	100	100	>50

[a]Minimal inhibitory concentration is the lowest concentration of compound that prevents visible growth after 24 hr of incubation. [b]The Norwich Pharmacal Co. strain number: Mi-12 = *Staphylococcus aureus*, Di-10 = *Diplococcus pneumoniae*, Er-4 = *Erysipelothrix insidiosa*, StA-1 = *Streptococcus pyogenes*, StB-12 = *Streptococcus agalactiae*, StD-7 = *Streptococcus faecalis*, Es-2 and Es-L = *Escherichia coli*, SaD-13 = *Salmonella typhosa*, Ae-6 = *Aerobacter aerogenes*, Pr-12 = *Proteus vulgaris*, Ps-44 = *Pseudomonas aeruginosa*. [c]Furacin, for comparison.

crude yellow product was filtered, washed thoroughly with H_2O, and dried to give 30.0 g (34.3%): mp 329–329.5° dec (aqueous DMF); γ_{max} 5.9 and 6.5 (CO), 6.65 and 7.43 (NO_2), 9.78 and 10.4 μ (furan); nmr (DMSO) peaks at τ 7.48 (CH_3), quartet centered at 2.08 (J = 4.5 Hz) (furan 3 H and 4 H). *Anal.* ($C_{10}H_6N_4O_5$) C, H, N.

3-Ethyl-6-(5-nitro-2-furyl)isoxazolo[5,4-*d*]pyrimidin-4(5*H*)-one (6) was prepared similarly in 44.2% yield from **4**: mp 282–283.5° dec (aqueous DMF). *Anal.* ($C_{11}H_8N_4O_5$) C, H, N.

4-Bis(2-hydroxyethyl)amino-3-methyl-6-(5-nitro-2-furyl)-isoxazolo[5,4-*d*]pyrimidine (7). A solution of 90 g (0.35 mol) of **5** and 71.5 g of PCl_5 in 1 l. of $SOCl_2$ was refluxed for 5 hr and concentrated nearly to dryness under reduced pressure, and the cooled residue was shaken with 500 ml of Et_2O. The crude 4-chloro compound was filtered, washed with Et_2O, and air-dried to give 45 g (47%): mp 188.5–189.5°. This material (40 g, 0.15 mol) was dissolved in 250 ml of DMF containing 33 g (0.3 mol) of diethanolamine. The resulting solution was heated with stirring on a steam bath for 6 hr, diluted with 50 ml of H_2O, and chilled. The crude yellow product was filtered, washed thoroughly with H_2O, and dried.

3-Ethyl-4-bis(2-hydroxyethyl)amino-6-(5-nitro-2-furyl)isoxazolo[5,4-*d*]pyrimidine (8) was prepared similarly from **6**.

***N*-(5-Cyano-4-pyrimidinyl)-5-nitro-2-furamide (9).** Molten 5-nitro-2-furoyl chloride (880 g, 5 mol) was added at a moderate rate to a vigorously stirred mixture of 600 g (5 mol) of 4-amino-5-cyanopyrimidine[7] and 3.2 l. of pyridine. Ice bath cooling was used when the temperature reached 55°. The mixture thickened and turned brown. When the addition was completed, the ice bath was removed and stirring continued for 0.5 hr before the mixture was cooled to 20° and diluted with 3.2 l. of ice–H_2O. The dark solid was filtered, washed successively with H_2O, *i*-PrOH, and Et_2O, and air-dried. The crude product was slurried for 5 min at 25° with 1.8 l. of DMF, filtered, and washed with 500 ml of DMF followed by Et_2O. The yield of light gray product melting at 283–284° was 837 g (64%). *Anal.* ($C_{10}H_5N_5O_4$) C, H, N.

***N*-(5-Carbamoyl-4-pyrimidinyl)-5-nitro-2-furamide (10).** Powdered **9** (268 g, 1.03 mol) was added in portions below 30° with stirring to 1070 ml of concentrated H_2SO_4. Stirring was continued for 2 hr following the addition. The mixture was poured over ice and diluted to 8 l. with H_2O. The crude tan product was filtered, washed thoroughly with H_2O, *i*-PrOH, and Et_2O, and dried to give 248 g (87%): mp 335–337° dec (DMF). *Anal.* ($C_{10}H_7N_5O_5$) C, H, N.

2-(5-Nitro-2-furyl)pyrimido[4,5-*d*]pyrimidin-4(3*H*)-one (11). A stirred mixture of 200 g (0.72 mol) of **10** in 1 l. of Dowtherm was boiled for 10 min. The mixture was cooled, diluted with MeOH, and filtered, and the product was washed with MeOH and dried to give 176 g (92%): mp 334–335° (methyl cellosolve). *Anal.* ($C_{10}H_5N_5O_4$) C, H, N.

4-Chloro-2-(5-nitro-2-furyl)pyrimido[4,5-*d*]pyrimidine (12). A mixture of 318 g (1.22 mol) of **11**, 383 g (1.84 mol) of PCl_5, and 1590 ml of $SOCl_2$ was refluxed for 15 hr. The mixture was cooled and filtered, and the residue was washed thoroughly with Et_2O to give the crude product as tan crystals melting *ca.* 210° in a yield of 260 g (77%). Since further purification of this hygroscopic solid could not be achieved, the crude solid was used in the next step as soon as possible.

4-Substituted Amino-2-(5-nitro-2-furyl)pyrimido[4,5-*d*]pyrimidine (13–19). A mixture of 50 g (0.18 mol) of crude 12 and 0.36 mol of the appropriately substituted amine in 500 ml of MeOH was refluxed with stirring for 10 min. The mixture was chilled and filtered, and the crude product was washed with H_2O, *i*-PrOH, and Et_2O, followed by drying and recrystallization from an appropriate solvent. The formation of a precipitate when 14 is treated with 5-nitro-2-furaldehyde in DMF supports the structure assignment of 14 as does γ_{max} 2.95, 3.05, and 6.1 μ (NH_2).

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Structure–Activity Studies on Narcotic Antagonists. 1. N-Substituted Ethyl 3-Phenylpyrrolidine-3-carboxylates and Ethyl 3-Phenylnipecotates†

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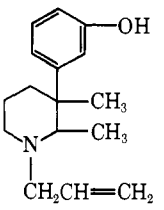
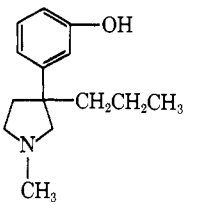
It is well established that replacement of the *N*-methyl group in potent opioid analgetics by certain small alkyl groups usually produces a compound possessing narcotic or opioid antagonist activity. Maximum antagonistic properties are found when the *N*-alkyl group is allyl, *n*-propyl, or cyclopropylmethyl, although the relative order of potency among these is dependent upon the analgesiophore.[2,3] That this type of group is not essential for opioid antagonism has been demonstrated by several (−) isomers of *N*-methylbenzomorphans.[4] These and other observations raise several questions in regard to the exact structural features and/or physicochemical properties necessary for potent opioid antagonism.

As part of a study into these questions, we prepared several *N*-alkyl derivatives of ethyl 3-phenylpyrrolidine-3-carboxylate (**8–13**) and ethyl 3-phenylnipecotate (**14–19**). These represent two series of analgetic analogs that possess a β-phenethylamine moiety, a structural feature that is present in almost all opioid antagonists.[2] Our attention to these series was drawn by the lack of antagonistic activity in the *N*-allyl[5,6] and *N*-dimethylallyl[6] derivatives of normeperidine. Archer and Harris[2] have suggested this may be due to the absence of the β-phenylethylamine moiety. Additional support for the β-phenethylamine hypothesis comes from the observations that the *N*-allyl compound **1** was found to be an antagonist devoid of analgetic activity[7] and that the *N*-methyl compound **2** was found to possess both analgetic and antagonist properties.[8] Both of these compounds contain a β-phenethylamine moiety.

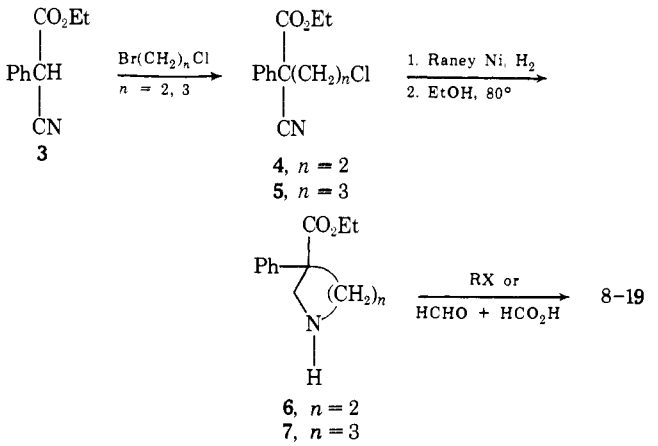


1



2

Chemistry. Of the published methods available for the synthesis of esters **6** and **7**, that used by Avison and Morrison[9] was selected over the earlier approaches of Bergel, *et al.*[10] Thus, ethyl phenylcyanoacetate (**3**) was alkylated with either 1-bromo-2-chloroethane or 1-bromo-3-chloropropane to yield the corresponding chloronitriles **4** and **5**, which were reduced to the corresponding primary amines. The primary amines were not isolated but were cyclized by refluxing in ethanol to give ethyl 3-phenylpyrrolidine-3-carboxylate (**6**) or ethyl 3-phenylnipecotate (**7**). It was found necessary to employ more rigorous hydrogenation conditions than Pd/C, as used earlier.[9] Satisfactory yields (45–60%) were obtained after 8 hr with Raney nickel in ammonia–ethanol solution. Addition of a catalyst promoter, platinic chloride, did not improve yields but reduced hydrogenation time to 1–2 hr. The *N*-methyl derivatives **8** and **14** were prepared by reductive methylation with formaldehyde and formic acid. The other N-substituted compounds (**9–13** and **15–19**) were prepared by treating the free base in ethanol in the presence of sodium bicarbonate or carbonate with the appropriate alkyl halide (Table I).



Of interest in the nmr spectra of these compounds was the nonequivalence of the protons on the 2- and 4-methylenes of the pyrrolidine and piperidine rings. In the spectra of unsubstituted esters **6** and **7**, only the C-2 protons are sufficiently resolved to allow assignment of coupling constants. The low field α-H in the pyrrolidine was analyzed as an AB system giving ^2J = 11.5 Hz. The corresponding α-H in the piperidine gave ^2J = 13 Hz, with further long-range coupling with the C-6 proton, ^4J = 2.5 Hz. The coupling was unchanged either after D_2O treatment or in the *N*-methyl **14**. This is indicative of diaxial coupling[11] and provides tentative evidence that the axial C-2 proton, which is almost 1 ppm downfield, is cis to an equatorial phenyl. Further experiments using decoupling techniques are in progress to confirm these observations and to provide additional conformational information.

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