Transformation of oxygen-bridged pyrimidines with nitrogen nucleophiles and characterization of resulting products

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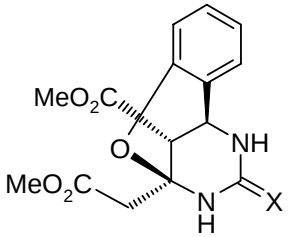
Abstract
The reactivity of oxygen-bridged Biginelli pyrimidines, such as 2,6-methano-1,3,5-benzoxadiazocines, towards nitrogen nucleophiles such as hydrazine hydrate, *N*,*N*-dimethylhydrazine, and benzylamine was studied. While all these reagents caused opening of the oxygen-containing ring, only the reaction with hydrazine furnished the corresponding pyrazolopyrimidine and pyridopyrimidine condensation products. $^{1}H–^{15}N$ HMBC correlation spectroscopy was used for structure confirmation.

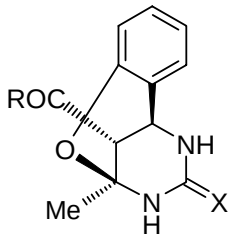
Keywords: Biginelli compounds**,** oxygen-bridged pyrimidines, ring transformation, pyrazolo[3,4-*d*]pyrimidine, pyrido[4,3-*d*]pyrimidine

Introduction

Recently we found that the use of dimethyl acetone-1,3-dicarboxylate in the Biginelli reaction with salicylaldehyde and urea or thiourea resulted in the production of 2,6-methano-1,3,5-benzoxadiazocine **1**.[1] We reported a similar formation of oxygen-bridged pyrimidine **2** for the classical Biginelli condensation under standard conditions.[2]



1a: X=O
1b: X=S



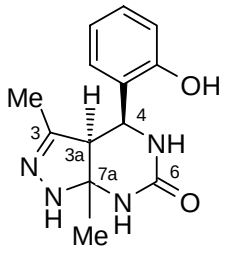
2a: R=OEt, X=O
2b: R=OEt, X=S
3: R=Me, X=O

The presence of two neighbouring ester groups in compounds **1**, provides an opportunity for the elaboration of a further nitrogen heterocycle, fused onto the pyrimidine ring, without altering

the parent bridged system. Here, we describe transformation of oxygen-bridged Biginelli pyrimidines in reactions with various nitrogen-containing reagents.

Results and Discussion

First we decided to test the behaviour of the compound **2a** towards hydrazine. However, a literature search revealed that an analogous reaction of the closely related acetyl derivative **3** had been already studied.[3] The transformation proceeded with the rupture of the oxa-cyclic fragment to give the corresponding pyrazolo[3,4-*d*]pyrimidine **4**.[3] Nevertheless, the authors described the relative stereochemistry only at two of the three chiral centres in the resulting bicyclic skeleton, e.g. at C-3a and C-4. In line with the reported article[3], our ester **2a** underwent a compararable type of conversion to yield the heterocycle **5**.



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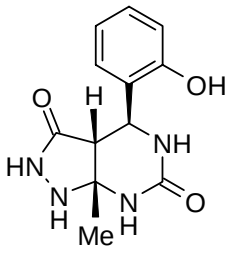


5

Prior to an NMR stereochemical analysis, a full assignment of ^{1}H and ^{13}C signals was unambiguously accomplished using 2D COSY and HMBC techniques (Table 1).

Table 1. NMR Spectroscopic parameters of compound **5** (dimethyl-d_6 sulfoxide)

Position	δ(^{13}C)/ppm	δ(^{1}H)/ppm *multiplicity*	HMBC connectivities (H – Ci,Cj,…)	COSY connectivities
CO-3	174.3	-	-	-
CO-6	155.5	-	-	-
C-2′	153.6	-	-	-
CH-4′	128.0	7.09 t[a]	4′ – 2′, 6′	4′ – 3′, 5′
C-1′	127.8	-	-	-
CH-6′	126.6	7.06 d[a]	6′ – 4, 2′, 4′	6′ – 5′
CH-5′	118.6	6.82 t[a]	5′ – 1′, 3′	5′ – 4′, 6′
CH-3	115.0	6.80 d[a]	3′ – 4, 1′ , 2′ ,5′	3′ – 4′
C-7a	73.5	-	-	-
CH-3a	47.4	2.99 s	3a – 3, 4, 7a, Me, 1′	3a – 4, 5, 7
CH-4	46.5	4.81 d[b]	4 – 3, 3a, 6, 7a, 1′, 2′	4 – 3a, 5
Me	24.5	0.88 s	Me – 3a, 7a	-
OH	-	9.70 br s	OH – 1′ , 2′ , 3′	-
NH-2	-	9.21 br s	2 – 3, 3a, 7a	-

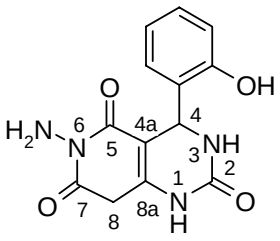
Table 1. Continued

Position	$\delta(^{13}C)$/ppm	$\delta(^{1}H)$/ppm *multiplicity*	HMBC connectivities (H – Ci,Cj,…)	COSY connectivities
NH-5	-	6.66 br s	5 – 3a	5– 4, 7
NH-7	-	6.29 s	7 – 3a	7– 3a, 4
NH-1	-	5.37 s	1– 6, Me	-

[a] The magnitudes of coupling constants for aromatic protons are in the usual ranges.
[b] ^{3}J (H-4,NH-5) = 2.8 Hz.

The relative configuration at all stereogenic atoms was readily be established by 1D NOESY experiments. Selective NOE transfer from the CH_3 resonance at δ_H 0.88 led to significant enhancements of the signal intensities for the adjacent hydrogens NH-1, NH-7 and H-3a. Hence, the methyl group at C-3 is on the same side of the ring as the H-3a methine. Furthermore, a weak NOE interaction with the remote aromatic H-6' was also found, which was indicative of a 1,3-*syn* relationship between the methyl and phenyl moieties. The resulting stereochemical arrangement was further confirmed by the strong interaction between H-3a and the methyl hydrogens and, additionally, by a weak NOE between H-3a and H-4. This showed that the stereochemical outcome of the ring closure resulting in pyrazolopyrimidine **5** was related to the relative stereochemistry of the starting O-bridged pyrimidine **2a**.

With the aforementioned data at hand, we carried out the planned cyclocondensation of diester **1a** with hydrazine under identical conditions (EtOH, reflux 30 h). Although this reaction was accompanied by some decomposition, we were able to isolate a small amount of a product. Considering the nucleophilic nature of both hydrazine nitrogens and also in view of the preceding transformation, we considered two structures with fused ring systems, pyrido[4,3-*d*]pyrimidine **6** and pyrimido[5,4-*d*]diazepine **7**, for the reaction product.



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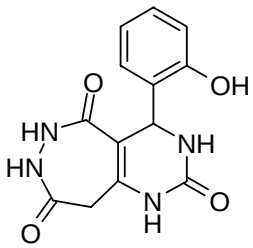

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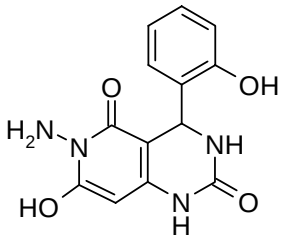
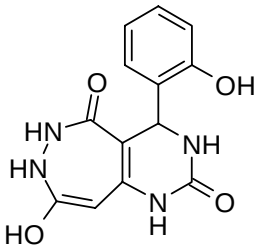
ESI-MS showed an $[M+H]^+$ ion at *m/z* 289 ($C_{13}H_{13}N_4O_4$) which was compatible with both **6** and **7**. However, the absence of a methylene in the ^{1}H and ^{13}C NMR spectra ruled out these two structures. Instead, the spectra indicated the presence of a methine in the product. Additionally, integration of the hydroxyl and amine protons, revealed 6H, i.e. one H atom more than in structures **6** and **7**. The other signals in the NMR spectrum pointed to a 5,6-disubstituted 4-(2-hydroxyphenyl)-pyrimidin-2-one moiety.These findings indicated that the product was another stable isomer related to heterocycles **6** or **7,** e.g., enol tautomers **6A** or **7A**.



6A



7A

Because of the low solubility of the condensation product, the solvent dependence of the tautomeric equilibrium could not be studied by NMR. Nevertheless, the presence of an oxo-form was not observed in DMSO-d_6. To assign a definite structure to the new product, we used ^{1}H–^{15}N heteronuclear chemical shift correlation spectroscopy. ^{1}H–^{15}N HSQC and HMBC experiments are based on direct or multiple bond N–H couplings. In particular, the gradient-enhanced HMBC technique correlating remote ^{1}H and ^{15}N nuclei provides valuable information for molecular structure elucidation and determination of ^{15}N chemical shifts of non-protonated nitrogens.[4] The HSQC spectrum showed only two cross-peaks, between δ_N -270.9 and NH-1 (δ_H 8.77) and between δ_N -288.6 and NH-3 (δ_H 7.13). This suggested structure **6A** rather then structure **7A.** The lack of ^{1}H–^{15}N correlation with the primary amino group is due to a fast amine proton exchange in solution. On the contrary, structure **7A** should exhibit four correlations for all protonated amide-type (not proton exchanging) nitro gen atoms.

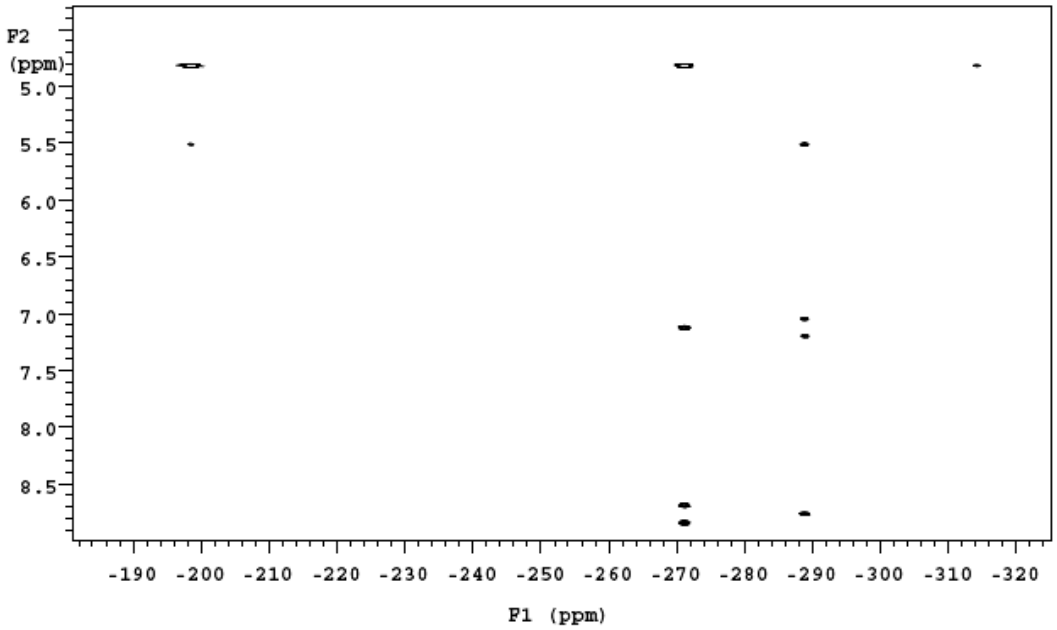


Figure 1. ^{1}H–^{15}N HMBC spectrum of compound **6A**.

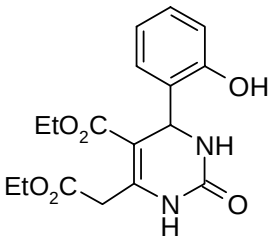
The assignment of the urea ^{15}N chemical shifts from HSQC measurements was supported by the long-range coupling pathways detected in the HMBC spectrum (Figure 1). Since the signal at

δ_N -270.9 correlates with the olefinic methine, it must be N-1. On the other hand, because of two-bond coupling to the benzylic methine, the other ^{15}N resonance at δ_N -288.6 corresponds to N-3. Moreover, the mutual ^3J(H–N–CO–N) interactions were consistent with this assignment. The third, non-protonated, nitrogen at δ_N -198.4 is significantly coupled to the olefinic proton (H-8) and displays a weak four-bond correlation with benzylic H-4 and thus can be attributed to the nitrogen ring atom N-6. Finally, the last ^{15}N nucleus of the exocyclic amino group was assigned to δ_N -314.3 through its $^4J_{NH}$ with H-8. The chemical shift value conforms with previous data[5] reported for hydrazine derivatives (~ -320 ppm). To summarize, data from ^1H–^{15}N correlation spectroscopy allowed us to establish the new product as 6-amino-7-hydroxy-4-(2-hydroxyphenyl)-4,6,dihydro[4,3-*d*]pyrimidine-2,5-(1*H*,3*H*)-dione (**6A**) and to exclude alternative structures such as pyrimidine[5,4-*d*]diazepine derivative **7A**.

Table 2. NMR Spectroscopic parameters[a] of compound **6A** (dimethyl-d_6 sulfoxide)

Position	$\delta(^{13}C)$/ppm	$\delta(^1H)$/ppm multiplicity	HMBC connectivities (H – Ci,Cj,…)	Pertinent COSY connectivities
C-7	158.4	-	-	-
CO-2	157.0	-	-	-
CO-5	154.7	-	-	-
C-2′	154.5	-	-	-
C-8a	144.2	-	-	-
C-1′	134.7	-	-	-
CH-4′	127.3	7.00 t[b]	4′ – 6′, 2′	-
CH-6′	125.6	7.04 d[b]	6′ – 4, 4′, 2′	-
CH-5′	119.0	6.74 t[b]	5′ – 3′, 6′, 1′	-
CH-3′	118.2	6.69 d[b]	3′ – 5′, 1′, 2′	-
C-4a	86.2	-	-	-
CH-8	79.8	4.81 s	8 – 4a, 8a, 7, 4	-
CH-4	47.4	5.50 d[c]	4 – 4a, 6′, 1′, 8a, 5, 2	4 – NH-3
OH, NH_2	-	6.40-7.40	-	-
NH-3	-	7.13 br s	3 – 4a	3 – NH-1
NH-1	-	8.77 s	1 – 4a, 8, 8a, 2	-

[a] $\delta(^{15}N)$/ppm: N-6, -198.4; NH-1, -270.9; NH-3, -288.6; NH_2, -314.3.
[b] magnitudes of coupling constants for aromatic protons are in usual ranges.
[c] ^3J (H-4,NH-3) = 3.0 Hz.



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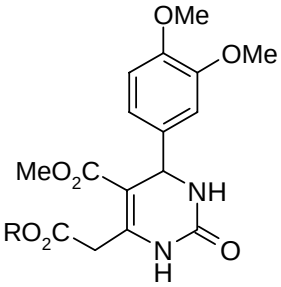

9: R=Me **10**: R=Et

In order to further investigate the formation of structure of **6A**, we used the reaction of *N*,*N*-dimethylhydrazine with **1a**. This hydrazine reagent which has only one condensable nitrogen would presumably force the formation of a 6-6 fused system. Unfortunately, the desired cyclocondensation failed and we were able to isolate only a ring-open dihydropyrimidine[1,8] with a free phenolic moiety **8**. The same product **8** was also obtained in the reaction of **1a** with benzylamine as a result of transesterification and breaking of the oxygen-bridge. Interestingly, when Biginelli compound **9** was treated with $PhCH_2NH_2$, only acetate transesterification was observed forming compound **10**.

Experimental Section

General Procedures. The melting points (uncorrected) were determined with a Kofler hot stage microscope. Elemental analyses were performed on a Carlo-Erba Elemental Analyzer Model 1012 apparatus. The IR spectra were recorded on a Nicolet Impact 400D spectrophotometer. The MS (ESI+) spectrum was obtained on an Agilent HP 1100LC-MSD Trap (VL) instrument with acetonitrile as a solvent.

For NMR measurements saturated 0.6 ml of solution of 5 or 6A in DMSO-d_6 was placed in a 5 mm NMR tube. All measurements were run on Varian Unity-Inova 600 MHz spectrometer at 25 °C. In all correlation methods, gradient-enhanced versions of experiments were used to achieve superior suppression of unwanted coherence transfer pathways. ^{1}H–^{1}H through-bond correlations were extracted from 2D g-COSY spectra, acquired with the following conditions: optimized spectral width in both domain; number of increments in t1-domain 256; number of scans for each increment 1; number of data points in t2 domain 1024; experimental time 5 min. Prior 2D FT data were appodized with sine-bell weighting functions.

In 1D NOESY experiments the DPFGSE excitation[6] of the selected proton was achieved using selective Gaussian 180° pulses with selectivity optimized for each proton depending on the width of the multiplet associated with particular proton. Mixing period was set to 1s.

2D ^{1}H–^{15}N g-HMBC and HSQC spectra were acquired with optimized spectral width for proton dimension and the spectral window ranging from -100 ppm to -330 ppm for ^{15}N dimension. HSQC experiment was optimized for $^1J(^1\text{H}–^{15}\text{N}) = 86$. Two of HMBC experiments optimized for two $^nJ(^1\text{H}-^{15}\text{N}) = 8$ and 3 Hz respectively. Other important parameters in HSQC experiment:

number of increments 256; number of scans per increment 4 ; experimental time 41m. Window functions in t1 domain: gaussian (0.0108s), in t2 domain: gaussian (0.069s); Fourier number in F1 domain 1k; F2 domain 2k. HMBC experiment: number of increments 256; number of scans per increment 32 ; experimental time 6h. Window functions in t2 domain: sinebell-shifted (-0.107s) , in t2 domain: gaussian (0.008s). Fourier number in F1 domain 1k; F2 domain 2k. ^{15}N chemical shifts in both HSQC and HMBC spectra were referenced via the ^{1}H solvent resonance (DMSO), using the absolute scale method.[7]

General procedure for the preparation of pyrimidines 5, 6A, 8, and 10

A suspension of starting compounds **1a**, **2a** or **9** (1.81 mmol) in dry ethanol (50 ml) was refluxed with hydrazine hydrate, *N*,*N*-dimethylhydrazine or benzylamine (4.16 mmol) for 31 h. While compound **6A** precipitated during the reaction, the other products crystallized after removal of the solvent and trituration of an oily residue with a small volume of methanol.

(3a*R,4*S**,7a*R**)-(±)-4-(2-Hydroxyphenyl)-7a-methyl-4,5,7,7a-tetrahydro-1*H*-pyrazolo[3,4-*d*]pyrimidine-3,6-(2*H*,3a*H*)-dione (5).** Yield: 0.276 g (58%); m.p.: 225-227 ºC (MeOH); Anal. Calcd for $C_{12}H_{14}N_4O_3$ (262.27): C, 54.96; H, 5.38; N, 21.36%; Found: C, 55.24; H, 5.13; N, 21.19%; IR (KBr): 3338 (NH), 3213 (OH), 1688 (NCON), 1669 (CON) cm^{-1}.

6-Amino-7-hydroxy-4-(2-hydroxyphenyl)-4,6-dihydropyrido[4,3-*d*]pyrimidine-2,5-(1*H*,3*H*)-dione (6A). Yield: 0.278 g (53%); m.p.: 217-219 ºC (EtOH); Anal. Calcd for $C_{13}H_{12}N_4O_4$ (288.26): C, 54.17; H, 4.20; N, 19.44%; Found: C, 53.89 H, 4.41; N, 19.70%; IR (KBr): 3408 (NH_2), 3326 (NH), 3207 (OH), 1702 (NCON), 1635 (CON) cm^{-1}; MS-(ESI+): 289 $[M+H]^+$, 311 $[M+Na]^+$.

Ethyl 6-ethoxycarbonylmethyl-4-(2-hydroxyphenyl)-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carboxylate (8). Yield 0.145 g (23%) for reaction with *N*,*N*-dimethylhydrazine and 0.196 g (29%) for benzylamine; m.p. 214-216 ºC (MeOH), lit.[8] 215-217 ºC; ^{1}H NMR (DMSO-d_6) δ (ppm): 1.01 (t, 3H, *J*=6.9 Hz, CH_3 ester-5), 1.20 (t, 3H, *J*=6.9 Hz, CH_3 ester-6), 3.63 (d, 1H, *J*=16.8 Hz, CH_2), 3.83 (d, 1H, *J*=16.8 Hz, CH_2), 3.91 (q, 2H, *J*=6.9 Hz, CH_2 ester-5), 4.12 (q, 2H, *J*=6.9 Hz, CH_2 ester-6), 5.53 (d, 1H, *J*=3.0 Hz, H-4), 6.71 (t, 1H, *J*=7.3 Hz, H-5′), 6.81 (d, 1H, *J*=7.8 Hz, H-3′), 7.06 (t, 1H, *J*=7.0 Hz, H-4′), 7.17 (br s, 1H, OH), 7.23 (d, 1H, *J*=7.3 Hz, H-6′), 9.17 (br s, 1H, NH-3), 9.63 (s, 1H, NH-1).

Methyl 6-ethoxycarbonylmethyl-4-(3,4-dimethoxyphenyl)-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carboxylate (10). Yield 0.510 g (74%); m.p. 187-189 ºC (MeOH); Anal. Calcd for $C_{18}H_{22}N_2O_7$ (378.38): C, 57.14; H, 5.86; N, 7.40%; Found: C, 56.81; H, 5.59; N, 7.71%; IR (KBr): 3320 (NH), 1737 (COO), 1681 (COO + NCON), 1638 (C=C) cm^{-1} ; ^{1}H NMR (DMSO-d_6) δ (ppm): 1.20 (t, 3H, *J*=6.9 Hz**,** CH_3 ester-6), 3.49 (s, 3H, CH_3 ester-5), 3.56 (d, 1H, *J*=16.8 Hz**,** CH_2), 3.72 (s, 3H, OCH_3), 3. 74 (s, 3H, OCH_3), 3.88 (d, 1H, *J*=16.8 Hz**,** CH_2), 4.11 (q, 2H, *J*=6.9 Hz**,** CH_2 ester-6), 5.11 (d, 1H, *J*=3.3 Hz. H-4), 6.83-6.89 (m, 2H, H-5′ + H-6′), 6.97 (d, 1H, *J*=1.8 Hz**,** H-2′), 7.74 (br s, 1H, NH-3), 9.27 (s, 1H, NH-1).

Acknowledgements

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