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Condensation of Triformylmethane with Heteroaromatic Amines, Including Nucleic Acid Bases. A Novel Example of Ring-Chain Tautomerism

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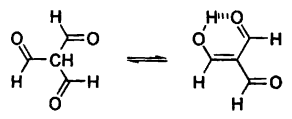
Treatment of several heteroaromatic amines, including the nucleic acid bases cytosine and adenine, with triformylmethane in pyridine at 70 °C afforded 1:1 condensation products (**2–11**) that were characterized by ^{1}H and ^{13}C NMR spectroscopy. All the compounds were assigned as *N*-substituted aminomethylenemalonaldehydes, strongly preferring the amino/keto form over the imino/enol tautomer. With cytosine and 1-methylcytosine, unequivocal evidence for ring-chain tautomerism was additionally obtained: intramolecular nucleophilic attack of cytosine N3 to one of the carbonyl carbons results in reversible cyclization to a 4-hydroxy-4*H*-pyrimido[1,6-*a*]pyrimidin-6(7*H*)-one derivative (**9b**, **10b**). In dry methanol the corresponding *O*-methyl ether (**12**) was obtained.

Triformylmethane (TFM; **1**), first prepared by Arnold and Žemlička,[1] is a versatile reagent for the preparation of heterocyclic compounds.[2–4] It is easily obtained from bromoacetic acid, phosphorus oxychloride and DMF by the Vilsmeier–Haack reaction.[5] TFM may be classified as a derivative of malonaldehyde. Malonaldehyde itself is unstable under aqueous conditions, but it may be prepared *in situ* from malonaldehyde bis(dialkyl acetals). By contrast, malonaldehydes bearing either an electron-withdrawing group, such as halogen[6a] or ethoxycarbonyl,[6b] or an alkyl substituent[6c] at the α-carbon are isolable. TFM belongs to the former category. It is possible that in solution the compound exists as a tautomeric mixture of triformyl and hydroxymethylenediformyl forms (see Scheme 1).[5a]

Reactions of TFM with some selected amino compounds, including simple primary and secondary amines, amino acids, urea and carbamic acid derivatives, have been studied.[7,8] The structure of the resulting condensation products depends on the stoichiometry of the reactants: usually 1:1 conjugates have been obtained, but from *tert*-butylamine a 1:2 conjugate (*tert*-butylaminomethylenemalonaldehyde *tert*-butylimine), and from aniline a 1:3 conjugate (triformylmethane trianil) have been prepared.[7a] The data on reactions of TFM with heterocyclic aromatic amines are scarce. It is known that TFM condenses with 2,6-diamino-4(3*H*)-pyrimidinone to give 2-amino-3,4-dihydro-4-oxopyrido[2,3-*d*]pyrimidine-6-carbaldehyde (6-formyl-5-deazapterin), used as a starting material in preparation of dihydrofolate reductase inhibitors.[9] Analogously, 6-formyl-8-methyl-5-deazapterin hydrochloride is obtained from 2-amino-6-(methylamino)-4(3*H*)-pyrimidinone.[10]



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Scheme 1. Tautomeric forms of triformylmethane **1**.

The present paper is aimed at elucidating more systematically the reactions of TFM with heteroaromatic amines with special emphasis on aminopyridines, aminopyrimidines and aminopurines, including the nucleic acid bases cytosine and adenine. The structural characteristics of the condensation products are discussed on the basis of their ^{1}H and ^{13}C NMR spectra. The data offer a novel example of ring-chain tautomerism related to formation of hypermodified nucleobases.

Results and discussion

The condensation of TFM with primary amines has usually been carried out in inert solvents, typically in dichloromethane, and those with amino acids and urea in water.[7a] The heteroaromatic amines used in the present study are, however, sparingly soluble in inert solvents, and attempted condensation in aqueous solution proved unsuccessful, in particular with cytosine. For these

reasons, pyridine was selected as the reaction medium. Compounds **2–11** were smoothly obtained in this solvent by incubating TFM with the appropriate amine for 1 h at 70 °C. They could not be purified by adsorption chromatography on silica gel, presumably because of sensitivity to adsorbed water. However, correct elemental compositions and sharp melting points indicated reasonable purity. ^{1}H and ^{13}C NMR data for **2–11** are given in Tables 1 and 2, respectively.

Structural characteristics. Compounds **2–11** may, in principle, exist in the two tautomeric forms depicted in Scheme 2. The ^{1}H and ^{13}C NMR spectroscopic data, however, strongly suggest that **2–8**, **9a**, **10a**, and **11** overwhelmingly prefer the amino/keto tautomer (**I**) over the imino/enol form (**II**). The reasoning is as follows. In each case there are two ^{1}H NMR signals in the typical formyl proton range δ 9.3–10. The shift difference between the two signals is 0.3 ppm, except with compounds **2** and **6**, for which only one signal is detected. In structure **I** (Scheme 2) the substituent at C4 makes the formyl protons, H1 and H3, chemically and magnetically non-equivalent. With the exception of compounds **2**, **3** and **6**, the lower-field formyl signal appears as a doublet with a coupling constant of 3 Hz, resulting from coupling to H4 ($^4J_{H1,H4}$). At about 12 ppm, a doublet with a coupling constant of 12 Hz is observed. In all likelihood this resonance refers to an enamino proton, which is coupled to the vicinal H4 ($^3J_{H4,NH}$). In agreement with these assignments, the H4 proton resonance appears as a doublet of doublets. With **2**, **3**, **6**, **9a** and **11a**, the enamino proton signal, and to some extent also that of the H4 proton, appears as a broadened singlet, owing to rapid NH exchange, and the vicinal coupling is hence lost. With compounds **2**, **3** and **6**, even the H1 proton signal is a broadened singlet, resulting in loss of the ^4J coupling information. Probably the planar conformation (possessing a favourable W coupling pathway) depicted for tautomer (**I**) in Scheme 2 no longer prevails. Consistent with this assumption, averaging of the H1 and H3 signals takes places with compounds **2** and **6**, possibly due to the increased acidity of NH protons. Nevertheless, the chemical shifts of H1, H3, H4 and N–H, strongly suggest that the structures given in Scheme 2 are also valid for **2**, **3**, **6**, **9a** and **11a**. Furthermore, the expected downfield effect of the *o*-heteroatom on the H4 shifts is clearly seen (cf., **5**, **7** and **8** vs. **4** and **6**).

The ^{13}C NMR chemical shifts lend further support to the above interpretation. In particular, the appearance of two carbonyl resonances at 190 ppm strongly argues against the imino/enol form. Analogously to the ^{1}H chemical shifts, an upfield γ-effect of the *o*-heteroatom on the C4 shifts is clearly seen with **5**, **7** and **8**. The narrow range of the C2 shifts also suggests similar structures for compounds **2–8**, **9a**, **10a** and **11a**.

Malonaldehyde has been shown to react with cytosine[11a] and adenine[11b,12] nucleosides. The proposed primary condensation products, the 1:1 adducts, exhibit analogous enaminal structures as presented for **2–8**, **9a**, **10a** and **11a**. The ^{1}H NMR shifts in Table 1 agree with those reported for the 1:1 adducts.

Ring-chain tautomerism. The spectra of the condensation products of cytosine (**9**), 1-methylcytosine (**10**) and adenine (**11**) show unequivocal evidence for the presence of two species with the same total number of ^{1}H and ^{13}C resonances. Evidently a ring-chain tautomeric equilibrium as depicted in Scheme 3 occurs. The mole fractions of the ring tautomers **9b**, **10b** and **11b** are, in DMSO-d_6, 0.88, 0.62 and 0.02, respectively. The assignment of the proposed *N*-substituted aminomethylenemalonaldehyde form (**9a**, **9b**) and 4*H*-pyrimido[1,6-*a*]pyrimidin-6(7*H*)-one form (**9b**, **10b**) is based on the following spectral observations. First, in both ^{1}H and ^{13}C NMR spectra an additional formyl signal referring to the ring-tautomer is detected, and the ^{1}H chemical shift of H4 of the ring-tautomer indicates the change in hybridization at HCO from sp^2 (**9a** and **10a**) to sp^3 (**9b** and **10b**) at C4. Second, the magnitude of the $^3J_{H4,HO}$ coupling constants is consistent with this interpretation. The mutual assignment of the H4/HO resonances was verified by a variable temperature NMR experiment with **10**. Above room temperature, the $^3J_{H4,HO}$ coupling gradually diminished, and the HO and H4 resonance signals were broadened. After the disappearance of the $^3J_{H4,HO}$ coupling, the HO signal was further broadened, while the H4 signal began to sharpen. At 60 °C, the HO resonance (centred at about δ 6.73) was hardly detectable, but the H4 signal appeared as a sharp singlet (at δ 6.60 as compared with the value δ 6.57 ppm at 30 °C). Similarly, the ^{1}H NMR spectra of **9** and **10** show a singlet at δ 7.7, arising from H2 of the ring-tautomer, in addition to the doublet of doublets typical for H4 of the chain-tautomer.

To prove the tautomeric nature of the cyclization process, the effect of solvent (DMSO-d_6, DMF-d_7 or CD_3OD) on the spectral properties of **10** was studied. In DMSO-d_6 (mole fraction of **10b** 0.62) and DMF-d_7 (mole fraction of **10b** 0.46) only the ring- and chain-tautomers appeared to be present. In these solvents, the position of the ring-chain tautomeric equilibrium was rapidly established and remained unaltered during repeated ^{1}H NMR measurements. Probably **10b** is stabilized by formation of an intramolecular hydrogen bond between the hydrogen of the pseudoequatorial HO group and the C=O oxygen. Through this hydrogen bond a nearly planar six-membered ring is formed. In CD_3OD, the situation seems to be more complicated. The first recorded ^{1}H NMR spectrum (total time 20 min from dissolution) exhibited only the tautomeric products. However, the initial condensation products seemed to be unstable, and a new reaction product appeared. After 2 h, only a minor amount of the initial products was detected, and after 24 h they virtually disappeared. The assignment of the thermodynamically stable solvolysis product (**12**) as the *O*-methyl derivative of **10b**, formed as depicted in

Table 1. ^{1}H NMR parameters for the condensation products **2–11** in DMSO-d_6 (ppm from Me_4Si).

Compound	R	H1	$^4J_{H1,H4}$	H3	H4	$^4J_{H4,NH}$	N–H	H2′	H3′	H4′	H5′	H6′	1′-CH_3
2	H	9.65		9.65	7.77		9.3						
3	CH_2Ph	9.64		9.35	8.11		10.8						
4	Ph	9.82	3.1	9.50	8.50	13.9	11.99	7.59	7.47	7.29	7.47	7.59	
5	2-Pyridyl	9.87	3.0	9.59	8.87	12.9	11.96		7.63	7.90	7.29	8.44	
6	4-Pyridyl	9.70		9.70	8.61		11.9	8.58	7.64		7.64	8.58	
7	2-Pyrimidinyl	9.93	3.7	9.64	8.85	13.1	11.87			8.81	7.42	8.81	
8	4-Pyrimidinyl	9.93	3.0	9.65	8.88	9.5	11.82	9.02			7.72	8.77	
9a	2-Oxo-1,2-dihydropyrimidin-4-yl	9.94	2.8	9.65	8.71		11.6				6.68	7.98	
10a	1-Methyl-2-oxo-1,2-dihydropyrimidin-4-yl	9.91	3.0	9.63	8.68	11.9	11.48				6.69	8.20	3.40
10a[a]		9.98	3.2	9.75	8.83	11.3	11.60				6.78	8.32	3.52
10a[b]		9.98	3.3	9.58	8.73						6.52	8.10	3.54
		H1		H3	H4		N–H	H2′	H8′	H9′			
11a	6-Purinyl	9.94	3.5	9.67	9.17		12.3	8.71[c]	8.60[c]	13.8			
		HCO		H4	H–O	$^3J_{H4,OH}$	H2	H8	H9				7-CH_3
9b		9.46		6.56	6.89	6.8	7.69	7.54	6.02				
10b		9.44		6.57	6.84	7.3	7.65	7.68	6.07				3.37
10b[a]		9.57		6.78	6.92	7.0	7.71	7.76	6.09				3.47
10b[b]		9.48		6.84			7.65	7.60	6.07				3.46
12[d]		9.50		6.50			7.78	7.73	6.11				3.39
12[b,d]		9.51		6.66			7.70	7.60	6.08				3.47
		HCO		H7	H–O		H2	H5	H9				
11b		9.44		6.69	7.14		8.55[c]	8.70[c]	7.81				

[a] Solvent DMF-d_7. [b] Solvent CD_3OD. [c] The asssignment in question is tentative. [d] O–Me: 3.20 ppm (DMSO-d_6); 3.36 ppm (CD_3OD).

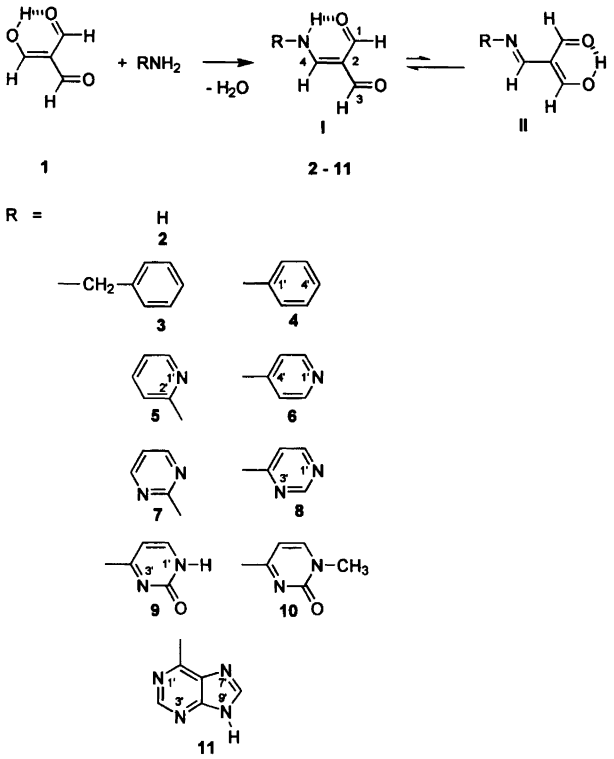
Table 2. ^{13}C NMR parameters for the condenstation products **2–11** in DMSO-d_6 (ppm from Me_4Si).

Compound	R	C1	C2	C3	C4	C1′	C2′	C3′	C4′	C5′	C6′
2	H	188.67	111.74	188.67	162.18						
3	CH_2Ph	188.01	111.38	188.01	160.15	127.79	128.67	127.79	137.04	127.79	128.67
4	Ph	189.35	112.71	189.03	154.72	138.47	129.60	118.69	126.19	118.69	129.60
5	2-Pyridyl	189.89	113.15	189.68	151.86		149.79	113.71	139.29	121.50	148.42
6	4-Pyridyl	189.67	113.77	189.67	154.44		150.85	112.83	145.54	112.83	150.85
7	2-Pyrimidinyl	191.15	113.83	189.82	152.14		159.00		159.28	118.72	159.28
8	4-Pyrimidinyl	190.26	114.50	190.26	150.58		158.85[a]		156.47	110.49	158.14[a]
9a	2-Oxo-1,2-dihydropyrimidin-4-yl	190.59	115.03	190.36	150.36		161.45		155.25	94.73	148.26
10a[b]	1-Methyl-2-oxo-1,2-dihydropyrimidin-4-yl	190.59	115.00	190.38	150.42		160.51		154.86	94.79	152.95

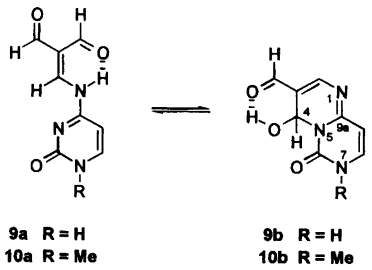
		C1	C2	C3	C4	C2′	C4′	C5′	C6′	C8′
11a	6-Purinyl	191.67	114.68	189.85	151.72	151.72	146.23	121.19	153.48	144.60

	C2	C3	C4	C6	C8	C9	C9a	HCO	7–CH_3
9b	140.44	120.33	66.27	153.42	152.19	101.25	149.51	189.06	
10b	143.22	120.34	67.04	153.04	151.93	101.92	149.48	189.08	36.89
12[c]	143.65	117.68	74.22	153.48	153.82	101.74	149.77	189.31	37.11

[a] The asssignment in question is tentative. [b] 1′-Me: 37.68 ppm. [c] O-Me: 55.19 ppm.

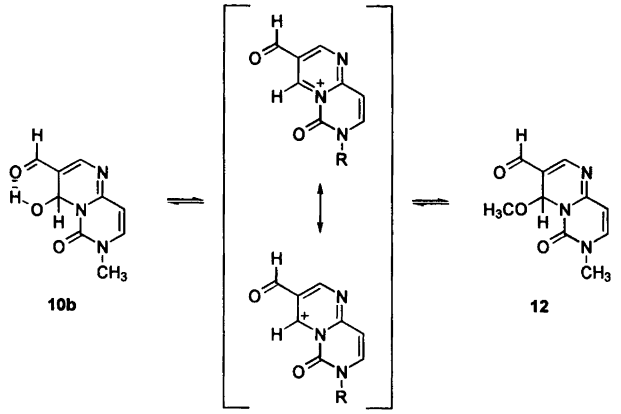


Scheme 2. Preparation of aminomethylenemalonaldehydes **2–11**.



Scheme 3. Ring-chain tautomerism of **9** and **10**.

Scheme 4, is based on the following spectral observations: (*i*) one ^1H formyl resonance signal is clearly retained, (*ii*) the AB system typical for the HO-C(H4) fragment is replaced by a singlet, and (*iii*) a ^1H resonance signal typical for a CH_3O group is observed. In addition, the spectrum of **12** shows some informative specific long-range ^1H–^1H couplings, while such couplings could not be observed for **10b** while it was still present. Accordingly, the H2, H9 and CHO proton resonances of **12** appear as doublets, the coupling constants being 0.61, 0.30 and 0.46 Hz, respectively. The H4 proton resonance appears as a somewhat broadened quartet with an approximate coupling of 0.46 Hz, suggesting the appearance of both $^4J_{H4,H2}$ and $^4J_{H4,HCO}$ couplings. In particular, a coupling pathway close to a W system seems to be reflected in the magnitude of the $^4J_{H4,H2}$ coupling. The appearance of the long-range ^1H–^1H couplings for **12** may refer to a change from a pseudoequatorial orientation at C4 (HO



Scheme 4. Mechanism for *O*-methylation of **10** in methanol.

in **10b**) to a pseudoaxial one (CH_3O in **12**), or to blocking of the OH exchange reaction.

The ^{13}C NMR chemical shifts lend further support to the preceding interpretation. A downfield β-effect on C4 and a typical upfield γ-effect on C3 apart, the ^{13}C chemical shifts of **10b** and **12** are practically equal. The structural assignment of the *O*-methyl ether **12** was further verified by an independent synthesis from **10**. *O*-Methylation of **10b** is a strong proof for the proposed assignment of **9b** and **10b** as the ring-tautomers of **9a** and **10a**. Moreover, **12** equilibrates (2 days, mole fraction of **12** 0.39) on prolonged standing in DMSO-d_6 into a mixture of **12**, **10a** and **10b**, in all likelihood due to a trace amount of water in the commercial DMSO-d_6.

Experimental

The starting materials employed were commercial products of Sigma or Aldrich, and were used without further purification. Elemental analysis was carried out on a Perkin Elmer Series II CHNS/O Analyzer 2400. The melting points reported are uncorrected values determined on a Büchi 510 apparatus. ^1H and ^{13}C NMR spectra were recorded at 30 °C (if not otherwise stated) on a JEOL JNM-A 500 FT NMR spectrometer operating at 500.16 and 125.77 MHz for ^1H and ^{13}C, respectively. Usually 0.1 M solutions in DMSO-d_6 were used (with less soluble derivatives as saturated solutions). Chemical shift values refer to internal Me_4Si. The deuterium of the solvent was used as a lock signal. ^{13}C NMR spectra were measured with both proton broad-band decoupling and gated decoupling with nuclear Overhauser enhancement techniques. Standard spectral parameters for ^{13}C NMR spectra were sweep width 30000 Hz, pulse width 45°, pulse delay 3 s. The typical digital resolution for ^1H NMR spectra was 0.15 Hz per data point and that for ^{13}C NMR spectra either 0.92 Hz (broad-band decoupling) or 0.46 Hz (gated decoupling) per data point.

Crude TFM[5b] was purified by sublimation (Büchi GKR-50 glass tube oven; 45 °C, 1 mmHg; on average the yield after sublimation was 88% and an amount of

about 0.6 g crude TFM could be conveniently handled). The white crystalline solid obtained melted at 105 °C (lit.[5b] 104–106 °C). Compound **2** was prepared as described in the literature.[8] The following methods were used to obtain the condensation products of TFM with the amines studied. Equal amounts (1–3 mmol) of TFM and the appropriate amine were stirred in dry pyridine (5 to 12 ml) for 1 h under nitrogen at 70 °C. Removal of the solvent under reduced pressure yielded a whitish crystalline residue, which was further washed with pyridine or dichloromethane and dried in a vacuum desiccator. The *O*-methyl derivative (**12**) of **10** was prepared as follows. **10** (0.1 mmol) was stirred in dry methanol (15 ml) for 8 h, during which period the slightly soluble starting material dissolved giving a light yellow solution. Removal of the solvent under reduced pressure yielded a glassy residue, which was dried in a vacuum desiccator. The following compounds were obtained. **1**: M.p. 105 °C. Anal. Found: C 47.40; H 3.95. Calc. for $C_4H_4O_3$: C 48.01; H 4.03. **3**: M.p. 114–115 °C. Anal. Found: C 69.70; H 5.70; N 7.21. Calc. for $C_{11}H_{11}NO_2$: C 69.83; H 5.86; N 7.40. **4:** M.p. 106 °C. Anal. Found: C 68.70; H 5.16; N 7.88. Calc. for. $C_{10}H_9NO_2$: C 68.56; H 5.18; N 8.00. **5**: M.p. 133–135 °C. Anal. Found: C 61.25; H 4.55; N 15.64. Calc. for $C_9H_8N_2O_2$: C 61.36; H 4.58; N 15.90. **6**: M.p. 164–165 °C. Anal. Found: C 60.16; H 4.62; N 15.95. Calc. for $C_9H_8N_2O_2$: C 61.36; H 4.58; N 15.90. **7**: M.p. 144–146 °C. Anal. Found: C 54.08; H 3.92; N 23.58. Calc. for $C_8H_7N_3O_2$: C 54.24; H 3.98; N 23.72. **8**: M.p. 143–145 °C. Anal. Found: C 53.36; H 4.01; N 23.42. Calc. for $C_8H_7N_3O_2$: C 54.24; H 3.98; N 23.72. **9**: M.p. 190 °C (decomp.). Anal. Found: C 50.27; H 3.61; N 21.97. Calc. for $C_8H_7N_3O_3$: C 49.74; H 3.65; N 21.75. **10**: M.p. 207–209 °C. Anal. Found: C 52.01; H 4.33; N 20.45. Calc. for $C_9H_9N_3O_3$: C 52.17; H 4.38; N 20.28. **11**: M.p. 192 °C (decomp.). Anal. Found: C 49.40; H 3.20; N 32.56. Calc. for $C_9H_7N_5O_2$: C 49.77; H 3.25; N 32.25.

References

1. Arnold, Z. and Žemlička, J. *Collect. Czech. Chem. Commun. 25* (1960) 1318.
2. Tietze, L.-F., Glüsenkamp, K.-H., Harms, K., Remberg, G. and Sheldrick, G. M. *Tetrahedron Lett. 23* (1982) 1147.
3. Angus, R. O., Jr., Bryce, M. R., Keshavarz-K., M. and Wudl, F. *Synthesis* (1988) 746.
4. Takagi, K., Bajnati, A. and Hubert-Habart, M. *Bull. Soc. Chim. Fr. 127* (1990) 660.
5. (a) Keshavarz-K. M., Cox, S. D., Angus, R. O., Jr. and Wudl, F. *Synthesis* (1988) 641; (b) Buděšínský, M., Fiedler, P. and Arnold, Z. *Synthesis* (1989) 858.
6. (a) Torii, S., Inokuchi, T. and Kubota, M. *Synthesis* (1986) 400; (b) for a review see: Reichardt, C. and Halbritter, K. *Angew. Chem., Int. Ed. Engl. 14* (1975) 86; (c) Reichardt, C. and Würthwein, E.-U. *Chem. Ber. 107* (1974) 3454 and references therein.
7. (a) Arnold, Z., Buděšínský, M. and Pánková, M. *Collect. Czech. Chem. Commun. 56* (1991) 1019; (b) Burkhard, J. and Arnold, Z. *Collect. Czech. Chem. Commun. 57* (1992) 1947.
8. Takagi, K., Bajnati, A. and Hubert-Habart, M. *J. Heterocyclic Chem. 31* (1994) 973.
9. (a) Temple, C., Jr., Elliott, R. D. and Montgomery, J. A. *J. Org. Chem. 47* (1982) 761; (b) Taylor, E. C., Palmer, D. C., George, T. J., Fletcher, S. R., Tseng, C. P. and Harrington, P. J. *J. Org. Chem. 48* (1983) 4852.
10. Koen, M. J. and Gready, J. E. *J. Org. Chem. 58* (1993) 1104.
11. (a) Stone, K., Uzieblo, A. and Marnett, L. J. *Chem. Res. Toxicol. 3* (1990) 467; (b) Stone, K., Ksebati, M. B. and Marnett, L. J. *Chem. Res. Toxicol. 3* (1990) 33.
12. Nair, V., Turner, G. A. and Offerman, R. J. *J. Am. Chem. Soc. 106* (1984) 3370.

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