Available online at www.sciencedirect.com

ScienceDirect

Mendeleev Commun., 2024, **34**, 706–708

Mendeleev Communications

An alternative synthesis of 7-amino-6-nitro-substituted azolo[1,5-*a*]pyrimidines

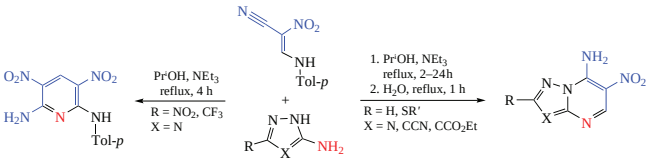
Ilya I. Butorin,*[a] Olga A. Konovalova,[a] Pavel A. Slepukhin,[b] Svetlana K. Kotovskaya[a] and Vladimir L. Rusinov[a]

[a] *Institute of Chemical Engineering, Ural Federal University, 620002 Ekaterinburg, Russian Federation. E-mail: butilig@gmail.com*

[b] *I. Ya. Postovsky Insititute of Organic Synthesis, Ural Branch of the Russian Academy of Sciences, 620108 Ekaterinburg, Russian Federation*

DOI: 10.1016/j.mencom.2024.09.026

New 7-amino-6-nitro-substituted [1,2,4]triazolo- and pyrazolo[1,5-*a*]pyrimidines were synthesized by an alternative strategy based on amino azoles and 2-nitro-3-(*p*-tolylamino)acrylonitrile. Unexpectedly, 3,5-dinitro-*N*-(*p*-tolyl)pyridine-2,6-diamine was formed when the starting 5-amino-1,2,4-triazoles contained NO_2 or CF_3 substituents.



Keywords: [1,2,4]triazolo[1,5-*a*]pyrimidines, pyrazolo[1,5-*a*]pyrimidines, nitro synthons, dinitropyridines, acrylonitrile, DFT.

Fused heterocyclic compounds of azoloazine series with a bridging nitrogen atom relate to one of the current areas of research in medicinal chemistry, largely due to structural similarity with natural purines. Of particular interest are amino- and nitro-substituted azolo[1,5-*a*]pyrimidines and azolo[5,1-*c*]-[1,2,4]triazines which exhibit antitumor,[1] antiplatelet,[2] vasodilatatory,[2] antiviral,[3–5] and some other[6–8] activities (Figure 1). However, few publications have been devoted to azolo[1,5-*a*]pyrimidines containing both functional groups within one molecule. In 2020 Safari[9] proposed a three-component synthesis of substituted 7-alkylamino-5-aryl-6-nitro-substituted azolo[1,5-*a*]pyrimidines possessing cytotoxic activity against tumor cell lines. The same year, Gazizov[10] synthesized 7-amino-6-nitro-containing azolo[1,5-*a*]pyrimidines by nitration of the corresponding 7-amino derivatives. However, those approaches had significant limitations associated with substituents in the azole ring, for example, nitration of 3-alkylthio-substituted derivatives led to the corresponding sulfones due to oxidation properties of the nitrating agent. Hence, it was necessary to use individual conditions and nitrating agents for each particular substrate.

The most promising method for the synthesis of azolo-annulated nitroazines is considered to be the fusion of the azine ring to the azole one using appropriate nitro-containing synthetic equivalents.[11] In this work, the reaction of amino azoles (substituted 5-amino-1,2,4-triazoles and 5-aminopyrazoles) **1a–i** and 2-nitro-3-(*p*-tolylamino)acrylonitrile **2** (synthesized according to a modified procedure[12] from the potassium salt of nitroacetonitrile[13]) under basic conditions has been studied (Scheme 1). Nitroethylene **2** is the synthetic equivalent of 1,3-dielectrophilic synthon analogously to diethyl ethoxy-methylenemalonate or ethoxymethylenemalononitrile that react

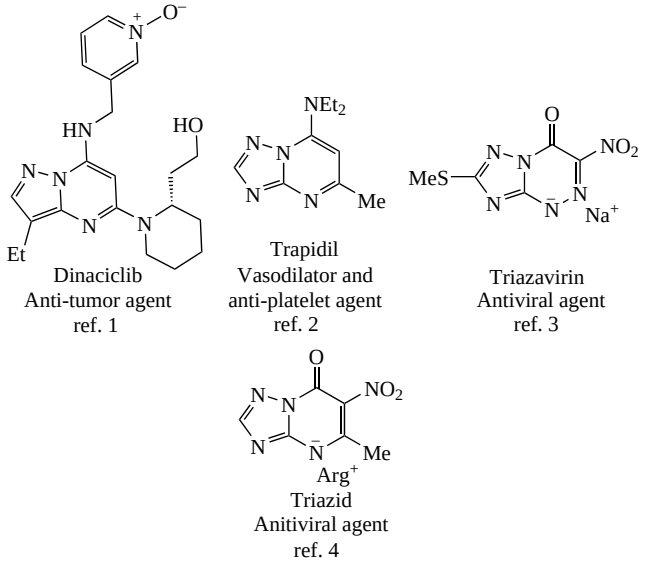
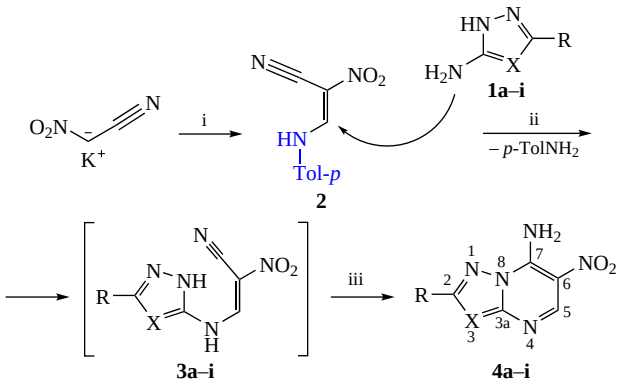


Figure 1 Amino- and nitro-containing bioactive azoloazines.



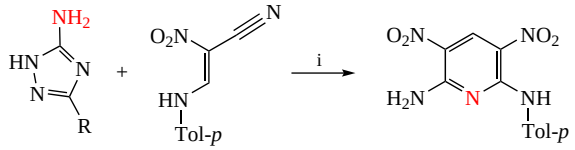
a R = MeS, X = N, 20%
b R = EtS, X = N, 28%
c R = PrS, X = N, 11%
d R = 2-thienyl, X = N, 26%
e R = 2-furyl, X = N, 55%
f R = BnS, X = N, 31%
g R = HC≡CCH$_2$S, X = N, 26%
h R = H, X = CCN, 13%
i R = H, X = CCO$_2$Et, 28%

Scheme 1 *Reagents and conditions*: i, Pr^{i}OH, CF_3CO_2H (1.2 equiv.), *p*-$MeC_6H_4NH_2$ (1 equiv.), $HC(OEt)_3$ (1 equiv.); ii, Pr^{i}OH, NEt_3 (1 equiv.), reflux, 2–24 h; iii, H_2O, reflux, 1 h.

with amino azoles to give the corresponding 6,7-substituted azolo[1,5-*a*] pyrimidines.[14–17] In this way, we synthesized a series of novel 7-amino-6-nitro-substituted azolo[1,5-*a*]-pyrimidines **4a**–**h**,† while known pyrazolo[1,5-*a*]pyrimidine **4i** was obtained with comparable yield.[10]

It is assumed that at the first stage of the reaction, the nucleophilic exocyclic nitrogen atom of amino azole **1** attacks the electrophilic center of acrylonitrile **2**, accompanied by the elimination of *p*-toluidine (see Scheme 1). The subsequent cyclization of intermediate **3** proceeds upon further refluxing of the reaction mixture in water. Intermediates **3** were isolated from the reaction mixture and used in the cyclization step without purification. The structure of **3** was assumed according to IR spectra containing bands in the region of ~2200–2300 cm^{-1} for the stretching vibrations of the cyano group.

The spectral characteristics of product **4i** coincide with those reported previously.[10] The structure of other compounds was initially proposed based on the known reactivity of amino-pyrazoles and aminotriazoles in the reactions with similar dielectrophiles.[14,18–20] Additional heteronuclear NMR experiments were performed to prove the regioiselectivity. Based on the ^{1}H–^{13}C HMBC NMR spectrum of compound **4g** (Figure S7), it was shown that the hydrogen atom from the pyrimidine ring correlated only with the nearest C^{5}, C^{6} and C^{7} carbon atoms. The absence of other correlations (except for interactions in prop-2-yn-1-yl moiety) does not allow us to assign the structure with one of the four possible regioisomers (see Online Supplementary Materials, Table S1). Thus, based on the works,[21–23] an additional ^{1}H–^{15}N HMBC NMR experiment was conducted using another compound **4c**. Nitrogen atoms of four possible regioisomers were associated with real chemical shifts according to the calculated isotropic shieldings in Orca v5.0.3.[24–26] Further evaluation of ^{1}H–^{15}N spectrum allowed us to exclude two of four possible isomers (triazolo[4,3-*a*]pyrimidine derivatives) based on the unreliable signals with assigned nitrogen atoms (see Table S1, Figure S3). 5-Amino-6-nitro regioisomer **4′c** also seems less comparable with experimental spectrum due to the lower correlation between chemical shifts and calculated shielding effects and the absence of three-bond cross peak including N^{1} and C^{7}H nuclei detected for other azolo[1,5-*a*]pyrimidines.[21] We showed that optimized conditions for the synthesis of 7-amino-6-nitro-substituted azolo[1,5-*a*]pyrimidines **4** include triethylamine as a base and prolonged refluxing time (Table S2).



1j,**k**
j R = CF_3
k R = NO_2

2
(2 equiv.)

5
33% from **1j**
20% form **1k**

Scheme 2 *Reagents and conditions*: i, Pr^{i}OH, NEt_3 (1 equiv.), reflux, 4 h.

In the search for the scope of the developed transformation we found that unexpected product **5** was formed in the cases of 3-trifluoromethyl- and 3-nitro-5-amino-1,2,4-triazoles **1j**,**k**. The structure of product **5** was assigned as 5-dinitro-*N*-(*p*-tolyl)-pyridine-2,6-diamine (Scheme 2).‡ The anticipated product in the reaction involving aminoazole **1j**, 7-amino-6-nitro-2-trifluoromethyl[1,2,4]triazolo[1,5-*a*]pyrimidine, was not isolated in pure form, but compound having [M]$^{+}$ = 248 was detected in the course of GC-MS analysis of the crude material.

A mechanism for this reaction was proposed according to results of GC-MS of the reaction mixture containing compounds with [M]$^{+}$ 135 and 269. It was assumed that amino azole acted as a ‘donor’ of the amino group in this reaction (Scheme 3) proceeding through intermediates **A**–**F**. Apparently, pyridine **5** can be formed only in the reaction of acrylonitrile **2** with aminotriazoles **1j**,**k** containing strong electron-withdrawing groups in position 3. Importantly, compound **5** was never detected in the reaction mixture in the cases of other amino azoles **1a**–**i**. The higher yield of pyridine **5** was associated with a molar excess of 3-trifluoromethyl-5-amino-1,2,4-triazole **1j** in reaction with ethylene **2**, while stoichiometric ratio led to better results in the case of 3-nitro-5-amino-1,2,4-triazole **1k** (Table S3).

The structure of pyridine **5** was confirmed by ^{1}H, ^{13}C NMR data, IR, mass spectra and single-crystal X-ray diffraction analysis (Figure 2).§ According to the X-ray diffraction data, the compound crystallizes in the centrosymmetric space group of the triclinic system. The crystal unit cell includes two crystallographically independent molecules of compound **5** and one pyridine molecule. The molecular geometry is close to the expected one. The molecules are flat (except for the hydrogen atoms of the methyl group) and form a layered structure in the crystal. The tolylamino group forms an intramolecular hydrogen

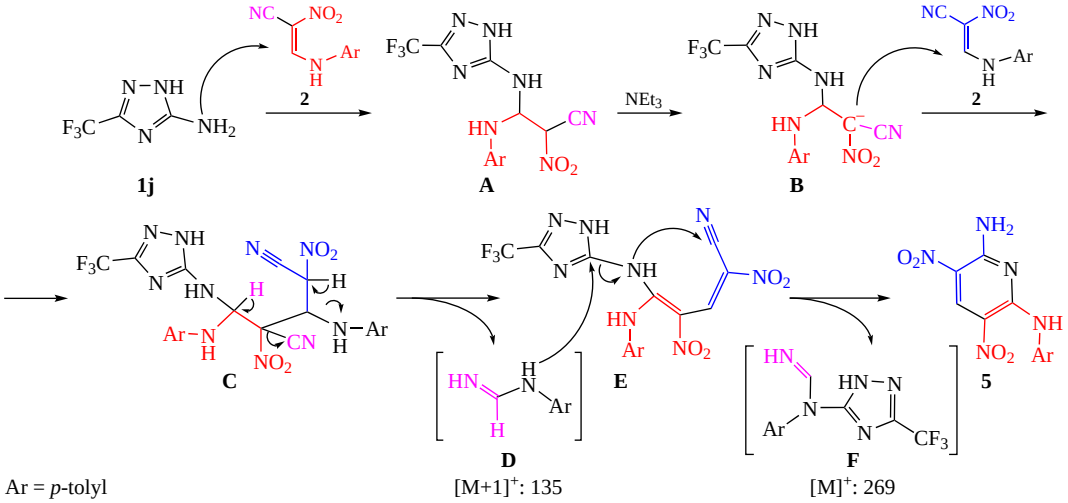
† *2-Methylthio-6-nitro[1,2,4]triazolo[1,5-*a*]pyrimidin-7-amine* **4a**. To a suspension of **1a** (1 mmol) in Pr^{i}OH (3 ml), triethylamine (1 mmol) and dry 2-nitro-3-(*p*-tolylamino)acrylonitrile **2** (1 mmol) were added sequentially. The mixture was refluxed for 2 h. The resulting suspension was filtered, and the solid was washed with a minimum amount of Pr^{i}OH. The precipitate was transferred to a new flask, suspended in water (5 ml) and refluxed for another 1 h. Then the suspension was cooled to 20 °C, filtered, washed with water and dried in air. Yellow solid. Yield 20%.

‡ *3,5-Dinitro-*N^{2}*-(*p*-tolyl)pyridine-2,6-diamine* **5**. *Method A.* 5-Amino-3-trifluoromethyl-1,2,4-triazole **1j** (5 mmol) was suspended in Pr^{i}OH (4 ml), then triethylamine (5 mmol) and 2-nitro-3-(*p*-tolylamino)-acrylonitrile **2** (5 mmol) were added sequentially. The mixture was refluxed for 4 h, then filtered and the solid was washed with Pr^{i}OH. The yellow-orange precipitate was dried in air. Yield 33%.

Method B. 5-Amino-3-nitro-1,2,4-triazole **1k** (5 mmol) was suspended in Pr^{i}OH (4 ml), then triethylamine (5 mmol) and 2-nitro-3-(*p*-tolyl-amino)acrylonitrile **2** (10 mmol) were added sequentually. The mixture was refluxed for 4 h, then the brown precipitate was filtered and the solid was washed with Pr^{i}OH. The target product was isolated from this precipitate by flash column chromatography (eluent: chloroform). The fraction with R_f = 0.56 ($CHCl_3$) was collected, evaporated under reduced pressure and air dried. Yellow-orange powder. Yield 20%.

§ *Crystal data for* **5** (solvate with pyridine). Single crystal was formed upon slow evaporation from pyridine solution at room temperature. $C_{14.5}H_{13.5}N_{5.5}O_4$ (M = 328.81), triclinic, space group $P\bar{1}$, a = 6.6352(6), b = 12.7380(17) and c = 18.3280(16) Å, α = 86.951(9), β = 89.244(7) and γ = 89.381(9)°, V = 1546.7(3) Å^{3}, μ(MoKa) = 0.107 mm^{-1}. X-ray diffraction analysis was carried out using the equipment of the Center for Collective Use ‘Spectroscopy and Analysis of Organic Compounds’ of the Institute of Organic Synthesis, Ural Branch of the Russian Academy of Sciences. The experiment was carried out on an automatic X-ray diffractometer Xcalibur 3 with a CCD detector according to the standard procedure [MoKα radiation, graphite monochromator, ω-scanning with a step of 1° at T = 295(2) K]. An empirical correction for absorption has been introduced. The solution and refinement of the structure were carried out in the Olex2 software shell.[27] The structure was solved using the ShelXT program[28] by the internal phase method and refined in the ShelXL program[29] by full-matrix least squares on F^2 in the anisotropic approximation for non-hydrogen atoms. The hydrogen atoms of C–H bonds were placed in calculated positions, the protons of NH groups were localized along the peaks of the spatial electron density and refined independently in the isotropic approximation. GOOF = 1.000, final R values: R_1 = 0.0856, wR_2 = 0.1873 [$I > 2\sigma(I)$]; R_1 = 0.2471, wR_2 = 0.2692 (all data). Residual electronic density max/min was 0.47/–0.22 eÅ^{-3}.

CCDC 2345245 contains the supplementary crystallographic data for this paper. These data can be obtained free of charge from The Cambridge Crystallographic Data Centre *via* http://www.ccdc.cam.ac.uk.



Scheme 3

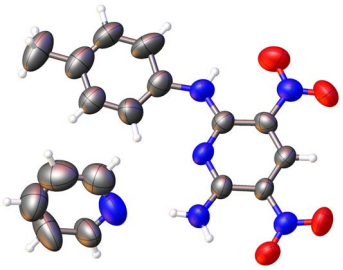


Figure 2 Results of X-ray diffraction analysis of compound **5** (solvate with pyridine). Thermal ellipsoids are given with 50% probability.

bond with the NO_2 group. The pyridine molecule is involved in an intermolecular hydrogen bond with the amino group of one of the molecules of the compound.

In conclusion, we have optimized the reaction conditions for the synthesis of novel 2-R-7-amino-6-nitro-substituted azolo[1,5-*a*]pyrimidines. Unexpected 3,5-dinitro-*N*-(*p*-tolyl)-pyridine-2,6-diamine was formed in cases of aminotriazoles containing electron-withdrawing groups.

Funding from Ministry of Science and Higher Education of the Russian Federation, project FEUZ-2023-0021, is acknowledged.

Online Supplementary Materials

Supplementary data associated with this article can be found in the online version at doi: 10.1016/j.mencom.2024.09.026.

References

1 D. Parry, T. Guzi, F. Shanahan, N. Davis, D. Prabhavalkar, D. Wiswell, W. Seghezzi, K. Paruch, M. P. Dwyer, R. Doll, A. Nomeir, W. Windsor, T. Fischmann, Y. Wang, M. Oft, T. Chen, P. Kirschmeier and E. M. Lees, *Mol. Cancer Ther.*, 2010, **9**, 2344.
2 E. Tenor and R. Ludwig, *Pharmazie*, 1971, **26**, 534.
3 O. N. Chupakhin, V. L. Rusinov, M. V. Varaksin, E. N. Ulomskiy, K. V. Savateev, I. I. Butorin, W. Du, Z. Sun and V. N. Charushin, *Int. J. Mol. Sci.*, 2022, **23**, 14537.
4 V. L. Rusinov, V. N. Charushin and O. N. Chupakhin, *Russ. Chem. Bull.*, 2018, **67**, 573.
5 E. G. Deeva, Yu. I. Shevchik, A. A. Shaldghan, K. A. Zagorodnikova, A. A. Tumashov, A. V. Baklykov, S. K. Kotovskaya, O. N. Chupahin, V. N. Charushin, V. L. Rusinov and D. S. Kopchuk, *Razrab. Regist. Lek. Sredstv*, 2018, no. 3, 172 (in Russian).
6 K. Oukoloff, B. Lucero, K. R. Francisco, K. R. Brunden and C. Ballatore, *Eur. J. Med. Chem.*, 2019, **165**, 332.
7 S. Pinheiro, E. M. C. Pinheiro, E. M. F. Muri, J. C. Pessôa, M. A. Cadorini and S. J. Greco, *Med. Chem. Res.*, 2020, **29**, 1751.
8 S. Federico, A. Ciancetta, N. Porta, S. Redenti, G. Pastorin, B. Cacciari, K. N. Klotz, S. Moro and G. Spalluto, *Eur. J. Med. Chem.*, 2016, **108**, 529.
9 F. Safari, M. Bayat, S. Nasri and S. Karami, *Bioorg. Med. Chem. Lett.*, 2020, **30**, 127111.
10 D. A. Gazizov, E. B. Gorbunov, G. L. Rusinov, E. N. Ulomskiy and V. N. Charushin, *ACS Omega*, 2020, **5**, 18226.
11 D. N. Lyapustin, V. V. Fedotov, E. N. Ulomsky, V. L. Rusinov and O. N. Chupakhin, *Russ. Chem. Rev.*, 2023, **92**, RCR5077.
12 O. S. Wolfbeis, *Chem. Ber.*, 1977, **110**, 2480.
13 E. K. Voinkov, E. N. Ulomskiy, V. L. Rusinov, K. V. Savateev, V. V. Fedotov, E. B. Gorbunov, M. L. Isenov and O. S. Eltsov, *Mendeleev Commun.*, 2016, **26**, 172.
14 G. V. Urakov, K. V. Savateev and V. L. Rusinov, *Dokl. Chem.*, 2022, **505**, 168.
15 Kh. S. Shikhaliev, D. V. Krylski, A. Yu. Potapov, S. E. Nefedov and O. E. Sidorenko, *Russ. Chem. Bull.*, 2008, **57**, 1268.
16 M. V. Goryaeva, Y. V. Burgart, M. A. Ezhikova, M. I. Kodess and V. I. Saloutin, *Beilstein J. Org. Chem.*, 2015, **11**, 385.
17 R. H. Springer, M. B. Scholten, D. E. O'Brien, T. Novinson, J. P. Miller and R. K. Robins, *J. Med. Chem.*, 1982, **25**, 235.
18 M. D. Wendt, A. Kunzer, R. F. Henry, J. Cross and T. G. Pagano, *Tetrahedron Lett.*, 2007, **48**, 6360.
19 G. V. Urakov, K. V. Savateev, V. V. Melekhin, S. K. Kotovskaya and V. L. Rusinov, *Russ. Chem. Bull.*, 2023, **72**, 3022.
20 A. E. M. Mekky and S. M. H. Sanad, *Mendeleev Commun.*, 2023, **33**, 331.
21 A. Salgado, C. Varela, A. M. García Collazo and P. Pevarello, *Magn. Reson. Chem.*, 2010, **48**, 614.
22 S. L. Deev, I. A. Khalymbadzha, T. S. Shestakova, V. N. Charushin and O. N. Chupakhin, *RSC Adv.*, 2019, **9**, 26856.
23 D. Sanz, R. M. Claramunt, A. Saini, V. Kumar, R. Aggarwal, S. P. Singh, I. Alkorta and J. Elguero, *Magn. Reson. Chem.*, 2007, **45**, 513.
24 F. Neese, F. Wennmohs, U. Becker and C. Riplinger, *J. Chem. Phys.*, 2020, **152**, 224108.
25 F. Neese, *WIREs Comput. Mol. Sci.*, 2011, **2**, 73.
26 S. Grimme, S. Ehrlich and L. Goerigk, *J. Comput. Chem.*, 2011, **32**, 1456.
27 O. V. Dolomanov, L. J. Bourhis, R. J. Gildea, J. A. K. Howard and H. Puschmann, *J. Appl. Crystallogr.*, 2009, **42**, 339.

Received: 12th April 2024; Com. 24/7463