1,2,3-TRIAZOLE PHARMACOPHORE-BASED DERIVATIVES OF PURINE BIOISOSTERES AND PYRIMIDINES

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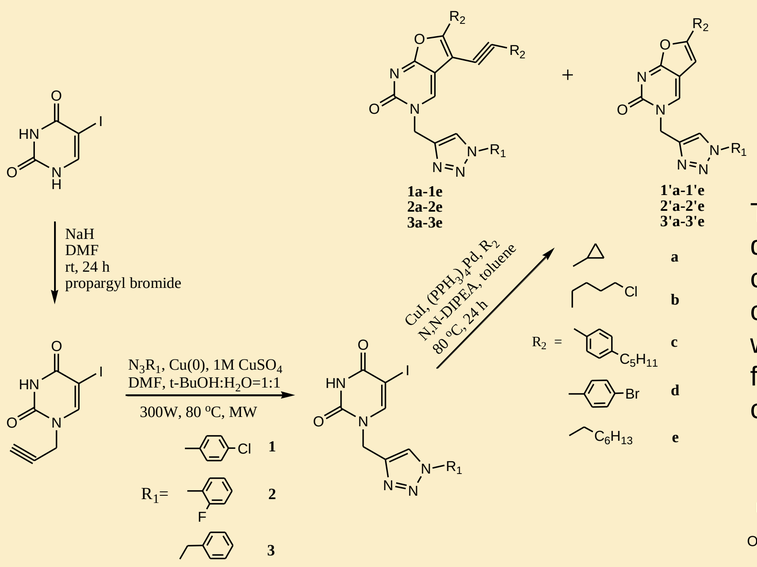
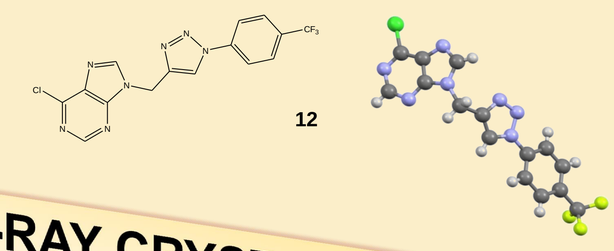
Introduction

Nucleoside analogs have acquired an important role as therapeutic agents in the field of chemotherapy on account of their extensive biological activities. Introduction of a triazole ring into nucleosides to improve their bioactivity for antitumor or antiviral applications has become wide spread in drug design practices.[1,2] The 1,2,3-triazole unit may be considered as a surrogate of the amide group because these moieties have a similar H-bond acceptor capacity, a similar distance between substituents, and a similar dipolar properties.



Scheme 1. Synthesis of mono- and disubstituted furo[2,3-*d*]pyrimidine derivatives with 1,4-disubstituted 1,2,3,- triazoles.



X-RAY CRYSTALLOGRAPHY

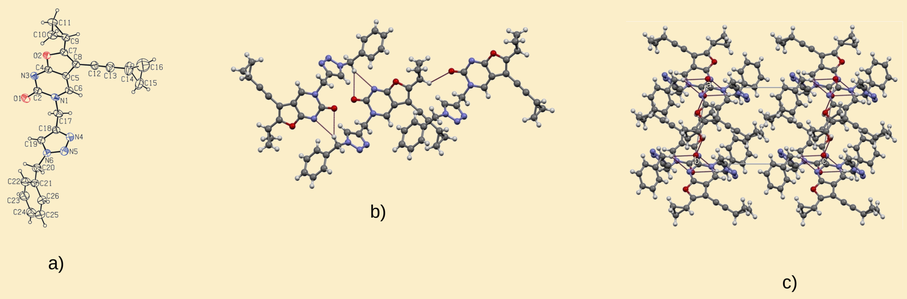


Fig.1. a) Molecular structure of compound **3a**; b) Part of the crystal structure of the compound **3a**, which shows a one-dimensional chain created two CH ••• O and one CH ••• N hydrogen bond; c) crystal packing of **3a** , shown along the crystallographic axis b, which shows the two-dimensional ribbon formed by hydrogen bonds and hydrophobic layers between them.

Antitumor evaluations

Compounds **4a-4d** and **5a-5d** were evaluated for their cytostatic activities against human malignant tumor cell lines: lung carcinoma (A549), hepatocellular carcinoma (HepG2), pancreatic carcinoma (CFPAC-1) cervical carcinoma (HeLa), colorectal adenocarcinoma (SW620), pancreatic carcinoma (MiaPaCa-2) as well as normal mouse embryonic fibroblast (3T3) cell lines. Compounds **4c**, **5a**, **5c** and **5d** showed marked cytostatic activities. Cytostatic evaluations for other synthesized compounds are currently underway.

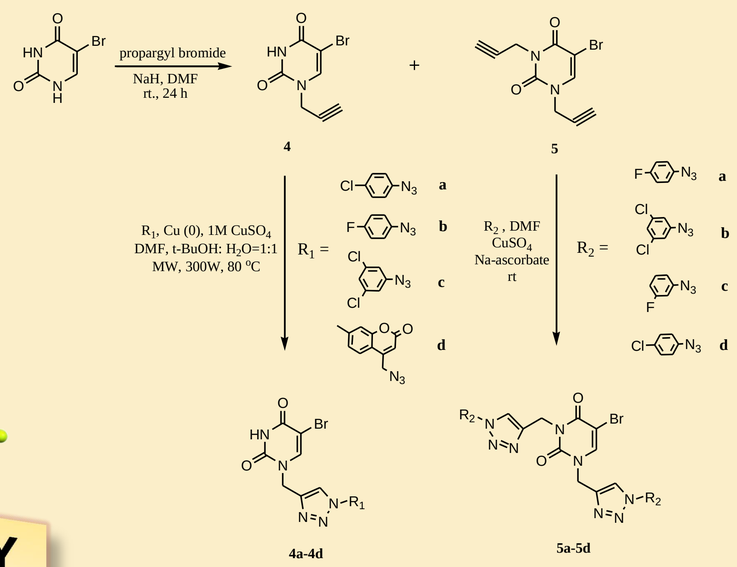
Table 1. Antitumor activitie of compounds **4a-4d** and **5a-5d**.

	IC_{50} (μM)					
Cmpd.	**A549**	**Hep-G2**	**CFPAC-1**	**HeLa**	**SW620**	**3T3**
4a	40.93	19.14	39.54	33.28	>100	6.7
4b	64	68.93	>100	72.9	>100	50.79
4c	80.68	**7.66**	**5.51**	**8.42**	**9.87**	1.16
4d	70.27	40.75	62.74	56.36	89.58	<0.01
5a	25.49	**4.47**	**6.87**	**12.58**	**9.82**	0.07
5b	95.79	42,.73	>100	>100	>100	4.3
5c	**5.33**	**7.11**	**4.53**	**1.64**	**7.86**	0.59
5d	**7.41**	**3.62**	**5.92**	**6.13**	**5.63**	0.37

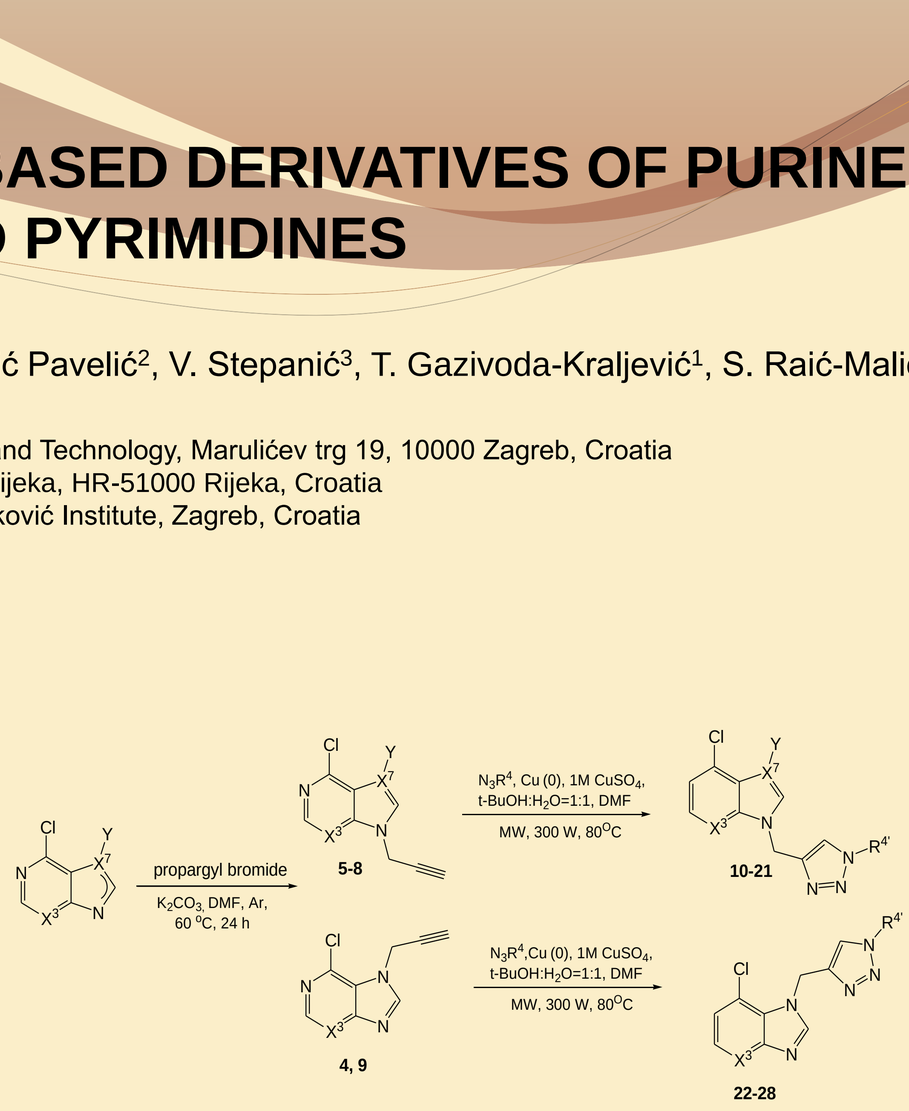
IC_{50}: 50% inhibitory concentration or concentration of the compound required to inhibit tumor cell proliferation by 50%. The cell growth rate was evaluated by performing the MTT assay.

Synthesis

The target regioselective 1,4-disubstituted 1,2,3-triazole derivatives were prepared by the microwave assisted Cu-catalyzed alkyne-azide cycloaddition (CuAAC). Linear alkyl chain, substituted aromatic moiety and cyclopropyl ring were introduced as alkynyl substituents at C-5 and C-6 of furo[2,3-*d*]pyrimidine core using modified palladium catalyzed cross-coupling Sonogashira reaction.



Scheme 2. Synthesis of N-1 and N-1, N-3-subsituted pyrimidine derivatives with 1,2,3-triazoles.



Compd.	X^3	X^7	Y	R^4	Yield (%)	ClogP[a]
6	N	NH	-	-	15	0.97
7	N	N	-	-	50	0.97
8	N	C	-	-	92	1.74
9	N	C	Br	-	70	2.56
10	CH	N	-	-	36	0.83
11	CH	NH	-	-	67	0.83
12	N	N	-		64	2.68
11	N	N	-		36	8.68
12	N	N	-		26	3.44
13	N	N	-		34	1.68
14	N	C	-		61	3.84
15	N	C	-		74	3.44

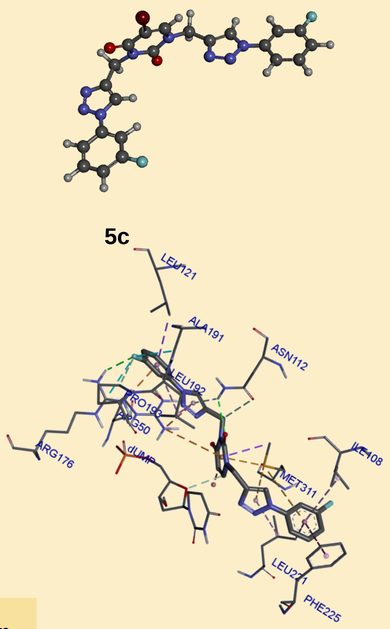
Compd.	X^3	X^7	Y	R^4	Yield (%)	ClogP[a]
16	N	C	-		49	3.44
17	N	C	-		41	4.20
18	N	C	-		55	1.68
19	N	C	Br		34	4.27
20	N	C	Br		35	5.00
21	N	C	Br		37	3.27
22	CH	N	-		80	3.29
23	CH	N	-		74	1.53
24	N	N	-		38	2.14
25	CH	N	-		51	2.93
26	CH	N	-		72	2.53
27	CH	N	-		80	3.29
28	CH	N	-		66	1.53

[a] Values of n-octanol/water partition coefficients logP were calculated using ChemAxon algorithm (MarvinView Ver. 6.2.2.).

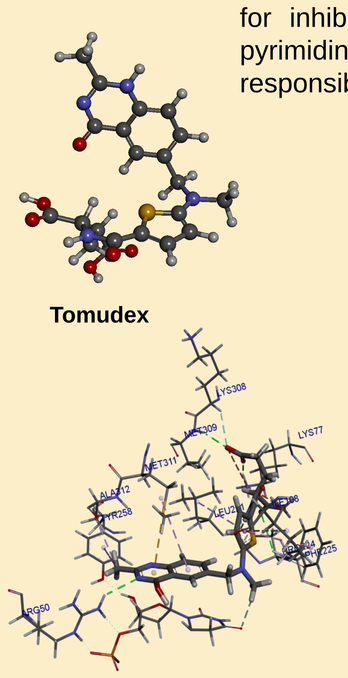
Scheme 3. Hybrids of purines, pseudopurines and 1,2,3-triazoles.

MOLECULAR DOCKING

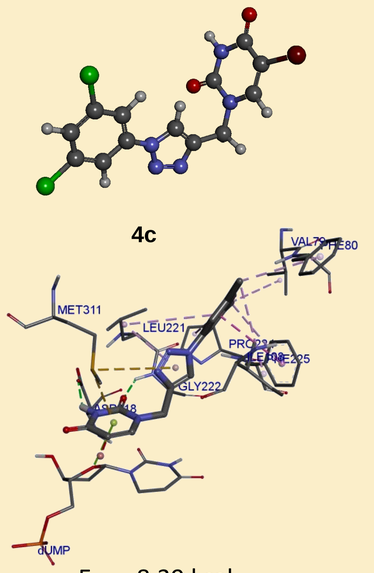
Molecular docking of compounds **4a-4d** and **5a-5d** in the active site of thymidilate synthase was performed and compared with known inhibitors. It was found that interactions with dUMP is of great importance for inhibitory effect of compounds. Naimly, pyrimidine ring of synthesized compounds is responsible for the interaction with dUMP.



E_{5a}= -10,02 kcal



E_{ref}= -11,19 kcal



E_{4c}= -8,39 kcal

	IC_{50} (μM)					
	Cell lines					
Cmpd	**A549**	**Hep-G2**	**CFPAC-1**	**HeLa**	**SW620**	**3T3**
5c	5.33	7.11	4.53	1.64	7.86	0.59
4c	80.68	7.66	5.51	8.42	9.87	1.16

Fig.3 Comparison of binding energy and binding site between known inhibitor tomudex and compounds **5a** and **4c** in the active site of the enzyme thymidylate synthase and their antitumor activities.

References

[1] S. Raić-Malić, A. Meščić, Curr. Med. Chem. **2015**, *22*, 1462
[2] S. Krištafor, A. Bistrović, J. Plavec, D. Makuc, T. Martinović, S. Kraljević Pavelić, S. Raić-Malić, Tetrahedron Lett. **2015**, *56*, 1222

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