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Synthesis of uracil–coumarin conjugates as potential inhibitors of virus replication

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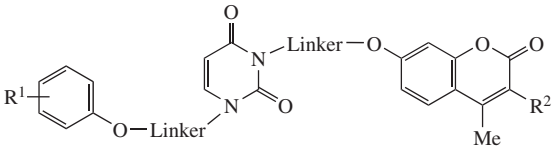
A series of 1-[(bromophenoxy)alkyl]uracil–coumarin conjugates has been obtained through the preparation of starting 1-[(bromophenoxy)alkyl]uracil derivatives, followed by their treatment with 7-(ω-bromoalkoxy)-4-methyl-2*H*-chromen-2-ones. Two of the synthesized uracil–coumarin conjugates demonstrated a pronounced inhibitory activity against HCMV and VZV replication *in vitro*.



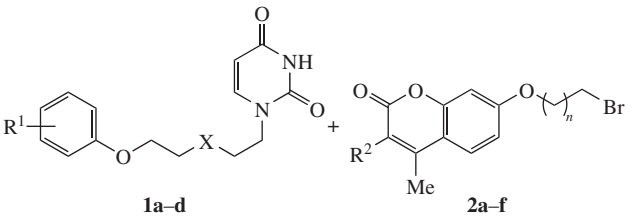
Anti-HCMV and anti-VZV activity

Coumarins are a large family of heterocyclic compounds found in higher plants and localized in their roots, bark and fruits. Natural and synthetic coumarin derivatives are known as antidepressants,[1] antimicrobials,[2] antioxidants,[3] as well as anti-inflammatory,[4] anti-asthmatic[5] and antitumor[6] agents. Coumarins were also found to exhibit inhibitory activity against HIV-1,[7] HCV,[8] and herpesvirus.[9] As well, antiviral effect was shown for the conjugates of coumarin with nucleic bases[10] and nucleosides.[11]

As an example of antiviral nucleic base conjugates, we synthesized 1-[ω-(aryloxy)alkyl]-3-[(4-phenoxyphenyl)aminocarbonylmethyl]uracil derivatives and demonstrated their inhibitory effect towards HCMV, VZV[12,13] and HCV[14] replication. In this work, we synthesized and tested a series of new uracil conjugates bearing coumarin moiety.

The synthesis of starting 1-[5-(4-bromophenoxy)pentyl]uracil **1a** was described.[15] 1-[5-(3-Bromophenoxy)pentyl]uracil **1b** and 1-[5-(2-bromophenoxy)pentyl]uracil **1c** precursors were obtained in 82 and 70% yields, respectively, by condensation of 2,4-bis(trimethylsilyloxy)pyrimidine with 1-bromo-3-[(5-bromopentyl)oxy]-benzene or 1-bromo-2-[(5-bromopentyl)oxy]benzene in equimolar amounts by heating without solvent under dry conditions as described.[15,16] 1-{2-[2-(4-Bromophenoxy)ethoxy]ethyl}uracil **1d** could not be obtained by this way, because bromotrimethylsilane, released during interaction of 2,4-bis(trimethylsilyloxy)pyrimidine with 1-bromo-4-[2-(2-bromoethoxy)ethoxy]benzene, cleaved the dialkyl ether moiety under the conditions employed. For this reason, the synthesis of uracil derivative **1d** was carried out by treatment of the latter with a fourfold molar excess of uracil in DMF in the presence of potassium carbonate. This alternative process led to 1-substituted uracil **1d** in 56% yield. Details of the synthesis and properties for precursors **1b–d** are given in Online Supplementary Materials.

Preparation of 7-(ω-bromoalkoxy)-4-methyl-2*H*-chromen-2-ones **2a–e** is well documented,[17–19] they were obtained by reaction of 7-hydroxy-4-methyl-2*H*-chromen-2-one with an excess of the corresponding α,ω-dibromoalkane in acetone in the presence of K_2CO_3. 7-(2-Bromoethoxy)-3,4-dimethyl-2*H*-chromen-2-one **2f** was synthesized in 72.5% yield by treatment of 7-hydroxy-3,4-dimethyl-2*H*-chromen-2-one with a fourfold molar excess of 1,2-dibromoethane under similar conditions.

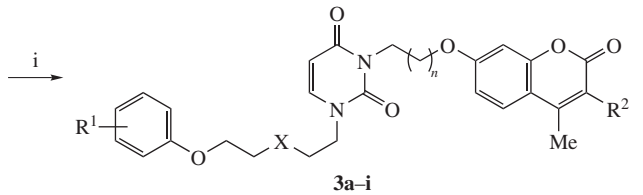


1a–d

a R^1 = 4-Br, X = CH_2
b R^1 = 3-Br, X = CH_2
c R^1 = 2-Br, X = CH_2
d R^1 = 4-Br, X = O

2a–f

a R^2 = H, n = 1
b R^2 = H, n = 2
c R^2 = H, n = 3
d R^2 = H, n = 4
e R^2 = H, n = 5
f R^2 = Me, n = 1



3a–i

a R^1 = 4-Br, R^2 = H, X = CH_2, n = 1
b R^1 = 4-Br, R^2 = H, X = CH_2, n = 2
c R^1 = 4-Br, R^2 = H, X = CH_2, n = 3
d R^1 = 4-Br, R^2 = H, X = CH_2, n = 4
e R^1 = 4-Br, R^2 = H, X = CH_2, n = 5
f R^1 = 3-Br, R^2 = Me, X = CH_2, n = 1
g R^1 = 2-Br, R^2 = Me, X = CH_2, n = 1
h R^1 = 4-Br, R^2 = Me, X = CH_2, n = 1
i R^1 = 4-Br, R^2 = H, X = O, n = 1

Scheme 1 *Reagents and conditions*: i, DMF, K_2CO_3, 80 °C, 18 h.

Table 1 Anti-HCMV and anti-VZV activity of uracil–coumarin conjugates **3a–i** in HEL cell culture.

Compound	Antiviral activity, EC_{50}/μM[a]				Cytotoxicity	
	HCMV AD-169	HCMV Davis	VZV Oka (TK^+)	VZV 07-1 (TK^-)	Cell morphology MCC/μM[b]	Cell growth CC_{50}/μM[c]
3a	>20	>20	>20	>20	100	–[d]
3b	0.39	0.51	0.52	0.6	4	–
3c	>0.8	>0.8	1.37	>0.8	4	–
3d	>4	>4	>4	>4	20	–
3e	0.44	0.44	0.16	0.47	4	4.95
3f	>100	>100	>100	>100	>100	–
3g	>4	4	>100	>100	20	–
3h	>20	>20	>100	>100	100	–
3i	>0.8	>0.8	11.7	5.72	<0.8	–
Ganciclovir	2.4	2.01	–	–	350	196.41
Cidofovir	0.38	0.38	–	–	300	129.43
Acyclovir	–	–	0.44	2.89	>100	>100
Brivudine	–	–	0.022	12.01	100	>100

[a] Concentration required to reduce virus plaque formation by 50%; virus load was 100 plaque forming units (PFU). [b] Concentration that caused a microscopically detectable alteration of cell morphology. [c] Concentration required to reduce cell growth by 50%. [d] Not determined.

The synthesis of the target uracil–coumarin conjugates was carried out in 74–86% yields by the treatment of 1-substituted uracil derivatives **1a–d** with an equimolar amount of bromides **2a–f** in DMF in the presence of K_2CO_3 (Scheme 1). Details of the synthesis and properties for the resulting compounds **3a–i** are given in Online Supplementary Materials.

Activity of the obtained uracil–coumarin conjugates against HCMV strains AD-169 and Davis as well as against VZV strains Oka and 07-1 was tested in HEL cell culture (Table 1). Compounds **3b** and **3e** have been found to inhibit HCMV replication with EC_{50} 0.39–0.51 μM, which is more effective than ganciclovir and comparable with cidofovir, though the test compounds are much more cytotoxic than both ganciclovir and cidofovir. The uracil–coumarin conjugates **3b** and **3e** have also been found to effectively block VZV replication with EC_{50} 0.16–0.52 μM. Note that, unlike acyclovir or brivudine, these two compounds demonstrate a similar inhibitory effect on both VZV strains tested, namely the wild type Oka strain and the thymidine kinase-deficient (TK^-) mutant 07-1 strain. This allows us to suggest, that conjugates **3b** and **3e** do not interact with viral thymidine kinase, and the mechanism of their anti-VZV effect differs from that for acyclovir or brivudine.

In summary, we have synthesized a number of new uracil–coumarin conjugates, and some of them have demonstrated activity against HCMV and VZV *in vitro*. These results open a possibility for a further search for potent antiviral agents based on 1-[ω-(aryloxy)alkyl]uracil structure.

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Online Supplementary Materials

Supplementary data associated with this article can be found in the online version at doi: 10.1016/j.mencom.2019.11.010.

References

1 K. V. Sashidhara, A. Kumar, M. Chatterjee, K. B. Rao, S. Singh, A. K. Verma and G. Palit, *Bioorg. Med. Chem. Lett.*, 2011, **21**, 1937.
2 B. Li, R. Pai, M. Di, D. Aiello, M. H. Barnes, M. M. Butler, T. F. Tashjian, N. P. Peet, T. L. Bowlin and D. T. Moir, *J. Med. Chem.*, 2012, **55**, 10896.
3 G.-L. Xi and Z.-Q. Liu, *J. Agric. Food Chem.*, 2015, **63**, 3516.
4 S. Bua, L. Di Cesare Mannelli, D. Vullo, C. Ghelardini, G. Bartolucci, A. Scozzafava, C. T. Supuran and F. Carta, *J. Med. Chem.*, 2017, **60**, 1159.
5 A. Sánchez-Recillas, G. Navarrete-Vázquez, S. Hidalgo-Figueroa, M. Y. Rios, M. Ibarra-Barajas and S. Estrada-Soto, *Eur. J. Med. Chem.*, 2014, **77**, 400.
6 M.-M. Liu, X.-Y. Chen, Y.-Q. Huang, P. Feng, Y.-L. Guo, G. Yang and Y. Chen, *J. Med. Chem.*, 2014, **57**, 9343.
7 H. Xue, X. Lu, P. Zheng, L. Liu, C. Han, J. Hu, Z. Liu, T. Ma, Y. Li, L. Wang, Z. Chen and G. Liu, *J. Med. Chem.*, 2010, **53**, 1397.
8 H.-K. Peng, W.-C. Chen, J.-C. Lee, S.-Y. Yang, C.-C. Tzeng, Y.-T. Lin and S.-C. Yang, *Org. Biomol. Chem.*, 2013, **11**, 1858.
9 B. Xu, L. Wang, L. González-Molleda, Y. Wang, J. Xu and Y. Yuan, *Antimicrob. Agents Chemother.*, 2014, **58**, 563.
10 J. R. Hwu, M. Kapoor, S.-C. Tsay, C.-C. Lin, K. C. Hwang, J.-C. Horng, I.-C. Chen, F.-K. Shieh, P. Leyssen and J. Neyts, *Antiviral Res.*, 2015, **118**, 103.
11 J. R. Hwu, S.-Y. Lin, S.-C. Tsay, E. De Clercq, P. Leyssen and J. Neyts, *J. Med. Chem.*, 2011, **54**, 2114.
12 D. A. Babkov, A. L. Khandazhinskaya, A. O. Chizhov, G. Andrei, R. Snoeck, K. L. Seley-Radtke and M. S. Novikov, *Bioorg. Med. Chem.*, 2015, **23**, 7035.
13 M. P. Paramonova, A. L. Khandazhinskaya, K. L. Seley-Radtke and M. S. Novikov, *Mendeleev Commun.*, 2017, **27**, 85.
14 A. Magri, A. A. Ozerov, V. L. Tunitskaya, V. T. Valuev-Elliston, A. Wahid, M. Pirisi, P. Simmonds, A. V. Ivanov, M. S. Novikov and A. H. Patel, *Sci. Rep.*, 2016, **6**, 29487.
15 M. S. Novikov, D. A. Babkov, M. P. Paramonova, A. L. Khandazhinskaya, A. A. Ozerov, A. O. Chizhov, G. Andrei, R. Snoeck, J. Balzarini and K. L. Seley-Radtke, *Bioorg. Med. Chem.*, 2013, **21**, 4151.
16 M. S. Novikov and A. A. Ozerov, *Chem. Heterocycl. Compd.*, 2005, **41**, 905 (*Khim. Geterotsikl. Soedin.*, 2005, 1071).
17 S.-S. Xie, X.-B. Wang, J.-Y. Li, L. Yang and L.-Y. Kong, *Eur. J. Med. Chem.*, 2013, **64**, 540.
18 R. Farina, L. Pisani, M. Catto, O. Nicolotti, D. Gadaleta, N. Denora, R. Soto-Otero, E. Mendez-Alvarez, C. S. Passos, G. Muncipinto, C. D. Altomare, A. Nurisso, P.-A. Carrupt and A. Carotti, *J. Med. Chem.*, 2015, **58**, 5561.
19 S.-S. Xie, X. Wang, N. Jiang, W. Yu, K. D. G. Wang, J.-S. Lan, Z.-R. Li and L.-Y. Kong, *Eur. J. Med. Chem.*, 2015, **95**, 153.

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