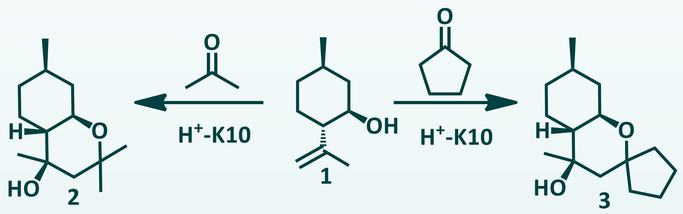
F- And OH-containing Isopulegol-derived Octahydro-2*H*-chromenes as Agents Against Influenza A Virus

Irina V. Ilyina[a], Oksana S. Patrusheva[a], Vladimir V. Zarubaev[b], Konstantin P. Volcho[a], Nariman F. Salakhutdinov[a]

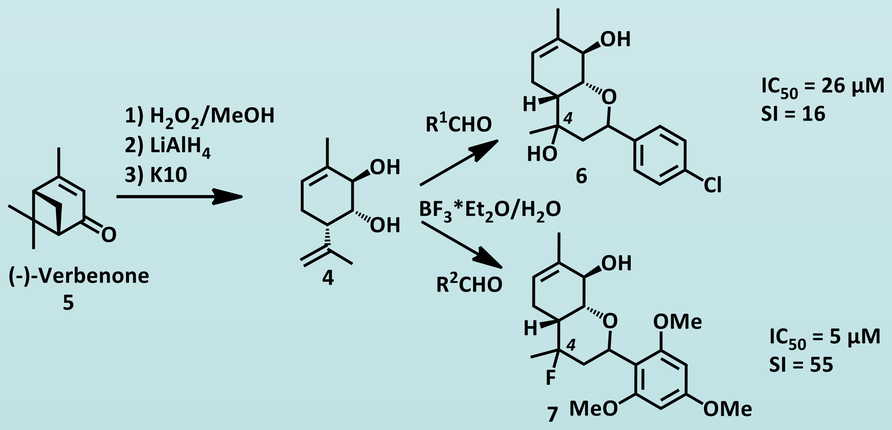
[a]*Novosibirsk Institute of Organic Chemistry, SB RAS, Lavrentjev av.9, 630090, Novosibirsk, Russia*
[b]*Pasteur Institute of Epidemiology and Microbiology, 14 Mira str., 197101 St. Petersburg, Russia*

Monoterpenes, which have a unique diverse structure and are inexpensive, available and often enantiomerically pure, are an attractive renewable raw material for the development of physiologically active agents. One of the important methods to the application of monoterpenes is their interaction with carbonyl compounds resulting in heterocyclic compounds. Often these products exhibit analgesic, antiviral or neuroprotective properties. Earlier, we discovered anti-influenza A (H1N1) virus activity of several compounds with a hydro-2*H*-chromene scaffold, which were synthesized by the Prins reaction using *para*-menthane alcohols and carbonyl compounds; montmorillonite K10 or nanosized halloysite catalyst were used as the reaction catalysts [1]. Chromenols produced from an (–)-isopulegol **1** and aliphatic ketones (acetone and cyclopentanone) demonstrated high activity in combination with low toxicity against the influenza A virus [2].



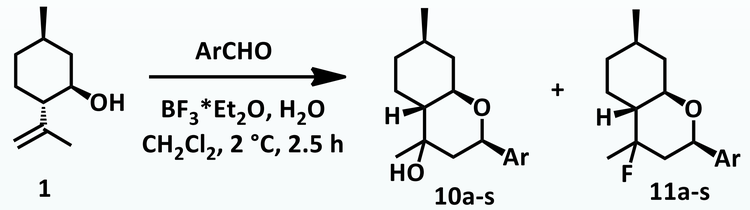
Compound	Strain	Subtype	IC_{50}, μM	SI
2	(A/Puerto Rico/8/34)	H1N1	7.5	**189**
2	(A/California/07/09)	H1N1pdm09	0.9	**1570**
2	(A/Singapore/1/57)	H2N2	0.9	**1570**
2	(B/Florida/04/06)	Yamagata-like	17.0	**83**
3	(A/Puerto Rico/8/34)	H1N1	8.4	**96**

The introduction of the fluorine atom into the molecule is an important strategy in the development of new biologically active compounds, which enables changing lipophilicity and electrostatic interactions and increasing the metabolic stability of compounds and, so affects their physiological activity. Another groups of hydro-2*H*-chromenes exhibiting activity against the influenza virus H1N1 was derived from diol **4**, which is synthesized from monoterpenoid (−)-verbenone **5** in three stages. Thus, by using starting diol **4** and aromatic aldehydes as well as the $BF_3{\cdot}Et_2O/H_2O$ system as a catalyst of the reaction, we synthesized a large set of heterocyclic compounds with a hexahydro-2*H*-chromene scaffold, which contained a hydroxy group at position C-4, as well as compounds fluorinated at this position (Scheme 2). A number of substances thus obtained showed high antiviral activity, for example compound **6** with the hexahydro-2*H*-chromene framework synthesized from **4** and *p*-chlorobenzaldehyde and 2,4,6-trimetoxybenzaldehede-derived fluorine-containing compound **7** [3].



Contact information: ilyina@nioch.nsc.ru
There are no conflicts to declare.
Authors would like to acknowledge the Multi-Access Chemical Research Center SB RAS for spectral and analytical measurements. All calculations were performed on a cluster supercomputer of the Ufa Institute of Chemistry of the Ufa Federal Research Centre of the Russian Academy of Sciences. Molecular docking and quantum chemical calculations were performed under Government Contract AAAA-A17-117011910028-7. The authors are grateful to Russian Ministry of Science and Education for the financial support (grant N 075-15-2020-777).

Here we synthesized fluoro- and hydroxy-containing octahydro-2*H*-chromenes by the Prins reaction starting from an (–)-isopulegol **1** and a wide range of aromatic aldehydes in the presence of the $BF_3{\cdot}Et_2O/H_2O$ system acting as both an acid catalyst and a fluorine source.

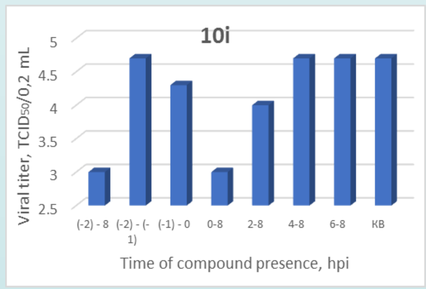


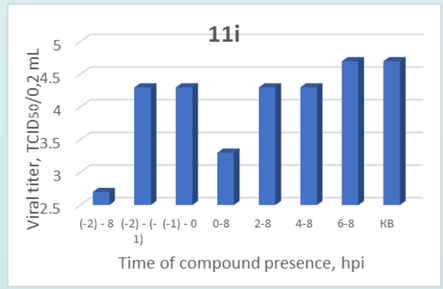
10a, **11a** Ar = C_6H_5; **10b**, **11b** Ar = 4-MeO-C_6H_4; **10c**, **11c** Ar = 2,3-MeO-C_6H_3; **10d**, **11d** Ar = 2,4-MeO-C_6H_3; **10e**, **11e** Ar = 2,5-MeO-C_6H_3; **10f**, **11f** Ar = 3,4-MeO-C_6H_3; **10g**, **11g** Ar = 2,3,4-MeO-C_6H_2; **10h**, **11h** Ar = 2,4,5-MeO-C_6H_2; **10i**, **11i** Ar = 2,4,6-MeO-C_6H_2; **10j**, **11j** Ar = 3,4,5-MeO-C_6H_2; **10k**, **11k** Ar = 2-OH-C_6H_4; **10l**, **11l** Ar = 4-OH-C_6H_4; **10m**, **11m** Ar = 3-OH,4-MeO-C_6H_3; **10n**, **11n** Ar = 3-MeO,4-OH-C_6H_3; **10o**, **11o** Ar = 4-OH,3,5-MeO-C_6H_2; **10p**, **11p** Ar = 4-F-C_6H_4; **10q**, **11q** Ar = 4-Cl-C_6H_4; **10r**, **11r** Ar = 4-Br-C_6H_4; **10s**, **11s** Ar = 4-NO_2-C_6H_4

Ar	10i			11i		
	CC_{50}, μM	IC_{50}, μM	SI[c]	CC_{50}, μM	IC_{50}, μM	SI
2,4,6-MeO-C_6H_2	856±49	19±2	**45**	851±62	24±3	**35**

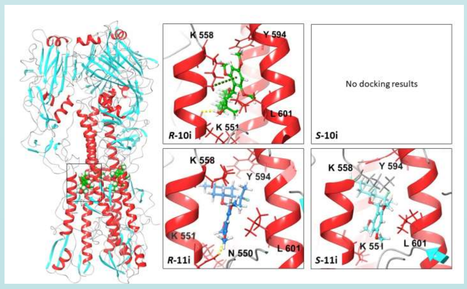
The total yield of octahydro-2*H*-chromenes **10** and **11** in the reactions was 70–90%, with the predominant formation of fluorides **11** (see [4]). For biological testing, we used reference laboratory strain of H1N1 subtype (A/Puerto Rico/8/34). Determination of the antiviral activity and toxicity studies described in detail in [4].

The highest activity was demonstrated by fluoro- (**11i**) and hydroxy-containing (**10i**) derivatives of 2,4,6-trimethoxybenzaldehyde. Compounds **10i** and **11i** were tested for antiviral activity depending on the time of addition to infected cells. In both cases, compounds **10i** and **11i** exhibited the most pronounced virus-inhibiting activity at an early stage of infection. These compounds were supposed to be capable of binding to viral hemagglutinin, which is an agreement with data on the effect of **10i** and **11i** on the viral fusogenic activity test, as well as by molecular docking studies [4].





Activity of **10i** and **11i** against influenza virus A/Puerto Rico/8/34 (H1N1) according to time-of-addition experiment.



Hemagglutinin and the binding site of potential inhibitors: π-cation stacking interactions are shown by the green dashed line, hydrogen bridges by the yellow line.

In general, affinity of the ligands is comparable. The compounds are located between two α-helices, forming a series of non-covalent interactions with the surrounding amino acids. This binding of potential HA inhibitors complicates separation of α-helices and may impede further conformational transition from pre- to post-fusion

[1] Salakhutdinov N, Volcho K, Yarovaya O, *Pure Appl. Chem.* 2017; 89:1105-1117.
[2] Ilyina IV, Zarubaev VV, Lavrentieva IN, Shtro AA, Esaulkova IL, Korchagina DV, Borisevich SS, Volcho KP, Salakhutdinov NF, *Bioorg Med Chem Lett.* 2018; 28:2061–2067.
[3] Patrusheva OS, Zarubaev VV, Shtro AA, Orshanskaya YaR, Boldyrev SA, Ilyina IV, Kurbakova SYu, Korchagina DV, Volcho KP, Salakhutdinov NF, *Bioorg Med Chem.* 2016; 24:5158–5161.
[4] Ilyina IV, Patrusheva OS, Zarubaev VV, Misiurina MA, Slita AV, Esaulkova IL, Korchagina DV, Gatilov YuV, Borisevich SS, Volcho KP, Salakhutdinov NF, *Bioorg. Med. Chem. Lett.* 2021, 31:127677