Sonidegib bioisosteres: activity and ADME properties

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Introduction and Aim

Sonidegib is a patented drug used for skin cancer therapy, which is characterized by low solubility and high LogD value. Therefore, the search for new bioisosteric analogs that retain bioactivity and have improved ADME properties is of interest.

In this work, we synthesized spiro[3.3]heptane, 7-oxa-azaspiro[3.5]nonane, bicyclo[2.1.1]hexane, and oxabicyclo[2.1.1]hexane analogs of Sonidegib (Fig. 1) and investigated how this affects its physicochemical properties and biological activity.

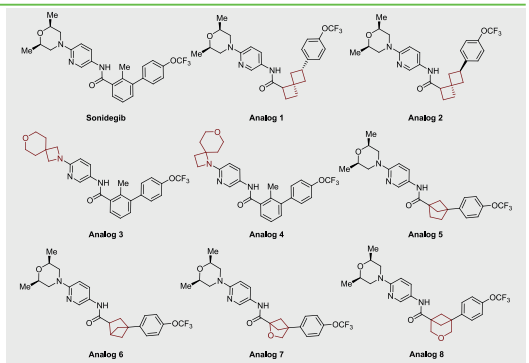

Figure 1. Structure of Sonidegib and bioisosteric analogs 1-8.

Methods

Kinetic solubility of the compounds in phosphate-buffered saline (PBS, pH=7.4; 2% final DMSO), and $LogD_{7.4}$ in *n*-octanol/PBS were experimentally measured. Metabolic stability of the test articles followed by metabolite identification (for analogs 1-4) was performed in human liver microsomes (HLM) using HPLC-MS/MS: half-life ($T_{1/2}$), and intrinsic clearance (Clint) were calculated. Plasma protein binding was tested by equilibrium dialysis with 100% and 10% human plasma.

To compare the biological activity, the experimental inhibition (IC50) of the Hedgehog signaling pathway in Gli-Luc reporter NIH3T3 cell line was tested using the ONE-Step™ Luciferase Assay System (BPS Bioscience, #60690-1).

Results

Table 1. ADME parameters and biological activity of Sonidegib analogs

Name	Sonidegib	Analog 1	Analog 2	Analog 3	Analog 4	Analog 5	Analog 6	Analog 7	Analog 8
Solubility in PBS (pH=7.4), μM	4.4	<1	<1	9	26	8	8	8	34
LogD (pH=7.4)	>4.5	3.61	3.55	3.9	>4.5	3.9	>4.5	4.0	>4.5
Clint, μL/min/mg (HLM)	16	36	156	23	106	20	19	27	28
T1/2, min (HLM)	104	46,7	11	72	16	82	86	61	61
PPB (100% human plasma), %	99	99,7	99,7	99,7	99,8	99,7	99,5	99	99.5
PPB (10% human plasma), %	99	99,5	99	96	98	96	99	99	98
IC50, μM (Hedgehog signaling inhibition in Gli Reporter NIH3T3 cells)	0.007	0.476	0.242	0.116	0.021	2.051	1.983	0.179	0.096
Cytotoxicity in Gli Reporter NIH3T3 cells, μM	70.41	10.9	27.97	>100	>100	11.45	16.38	>100	17.81

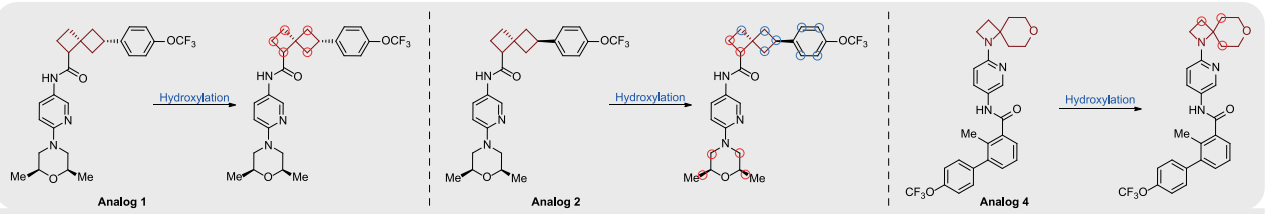

Figure 2. Main metabolites of Sonidegib analogs 1-4 in HLM.

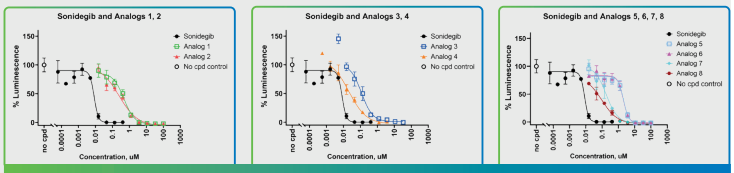

Figure 3. Evaluation of the Hedgehog signaling pathway inhibition by Sonidegib and analogs. The intensity of luminescence correlates with the pathway activation. The activation was considered 100% without the presence of inhibiting substances (no cpd).

Replacing the *meta*-substituted phenyl ring with spiro[3.3]heptane (analogs **1** and **2**) in the structure of Sonidegib had minimal impact on logD, plasma protein binding, and solubility in PBS. However, the metabolic stability of both analogs was notably lower, with significant differences observed between *trans*- and *cis*-isomers. Spiro[3.3]heptane analogs were less active inhibitors of Hedgehog signaling pathway but more cytotoxic on NIH3T3 cells than Sonidegib.

Replacement of the 2,6-dimethylmorpholine with 7-oxa-2-azaspiro[3.5]nonane and 7-oxa-1-azaspiro[3.5]nonane (analogs **3** and **4**) resulted in slightly increased solubility but decreased metabolic stability for analog **4**. Both analogs were less cytotoxic on NIH3T3 cells and analog **3** retained nanomolar inhibition of the Hedgehog signaling (Table 1). The main metabolite detected for analogs **1-4** was a hydroxylated product. In addition, for analog **2**, further hydroxylation occurs in HLM (Figure 2).

Analogs **5-8** were less active but more cytotoxic (**5, 6, 8**) than Sonidegib. ADME parameters do not differ significantly, only analog **8** exhibited slightly higher solubility.

Conclusions

In this study, we showed that spiro[3.3]heptane, 7-oxa-azaspiro[3.5]nonane, bicyclo[2.1.1]hexane and oxabicyclo[2.1.1]hexane can be useful structural elements in drug discovery projects

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References

1.Prysiazhniuk K, Datsenko O, *et al.* Spiro[3.3]heptane as a Non-collinear Benzene Bioisostere. *ChemRxiv*. Cambridge: Cambridge Open Engage; **2023**.