Journal of Medicinal Chemistry

Article

Synthesis and Evaluation of N-phenyl-3-sulfamoyl-benzamide Derivatives as Capsid Assembly Modulators inhibiting Hepatitis B Virus (HBV).

Koen Vandyck, Geert Rombouts, Bart Stoops, Abdellah Tahri, Ann Vos, Wim Verschueren, Yiming Wu, Jingmei Yang, Fuliang Hou, Bing Huang, Karen Vergauwen, Pascale Dehertogh, Jan-Martin Berke, and Pierre Jean Marie Bernard Raboisson

J. Med. Chem., **Just Accepted Manuscript** • DOI: 10.1021/acs.jmedchem.8b00654 • Publication Date (Web): 15 Jun 2018

ACS Publications

is published by the American Chemical Society. 1155 Sixteenth Street N.W., Washington, DC 20036

Synthesis and Evaluation of *N*-phenyl-3-sulfamoyl-benzamide Derivatives as Capsid Assembly Modulators inhibiting Hepatitis B Virus (HBV).

Koen Vandyck,[†,*] Geert Rombouts,[†] Bart Stoops,[†] Abdellah Tahri,[†] Ann Vos,[†] Wim Verschueren,[†] Yiming Wu,[‡] Jingmei Yang,[‡] Fuliang Hou,[‡] Bing Huang,[‡] Karen Vergauwen,[†] Pascale Dehertogh,[†] Jan Martin Berke, [†] Pierre Raboisson[†]

†Janssen Pharmaceutica NV, Janssen Pharmaceutical Companies of Johnson & Johnson, Turnhoutseweg 30, 2340, Beerse, Belgium

[‡]WuXi AppTec, 288 Fute Zhong Road, China (Shanghai) Pilot Free Trade Zone; Shanghai 200131, PR China

KEYWORDS

HBV, CAM, Capsid Assembly Modulators, JNJ-6379, JNJ-632

ABSTRACT

Small molecule induced Hepatitis B virus (HBV) capsid assembly modulation is considered an attractive approach for new antiviral therapies against HBV. Here we describe efforts towards the discovery of a HBV capsid assembly modulator in a hit-to-lead optimization, resulting in

JNJ-632, a tool compound used to further profile the mode of action. Administration of JNJ-632 (**54**) in HBV genotype D infected chimeric mice, resulted in a 2.77 log reduction of the HBV DNA viral load.

Introduction

Despite the availability of a safe and effective prophylactic HBV vaccine, it is estimated that about 240 million people worldwide are chronically infected by hepatitis B virus.[1] Chronic HBV infection puts patients at high risk for developing cirrhosis and liver cancer, with estimation of more than 686 000 people dying every year due to complications of hepatitis B. Besides (Pegylated) interferon-alpha ((PEG)-IFNα), multiple nucleos(t)ide analogs have been approved for treatment of hepatitis B. This current treatment can slow the progression of cirrhosis, reduce incidence of liver cancer and improve long term survival, but the majority of patients will not be cured from HBV infection. Therefore, chronic HBV infection presents a major global health concern.

HBV capsid assembly is a critical step in virus production and it is considered an attractive target for new antiviral therapies against HBV. Capsid assembly modulators (CAMs), compounds that modulate the kinetics/thermodynamics of HBV core protein aggregation, can block encapsidation of pregenomic RNA (pgRNA) and will consequently inhibit the synthesis of HBV DNA and infectious virion production. Recently, it has been shown that capsid assembly modulators, for example JNJ-632[2] described in this paper, and JNJ-6379, currently in phase 2 clinical trial,[3, 4] are also able to block *de novo* HBV infection *in vitro*.

Prototypical examples of two distinct types of capsid assembly modulators are AT-130[5, 6] and Bay 41-4109[7] (Fig 1). Both are inducing aggregation of the HBV core protein, but the first

induces the formation of empty capsids (CAM-I class of compounds), lacking the pgRNA-polymerase, while the latter induces the formation of aberrant structures (CAM-II class of compounds).

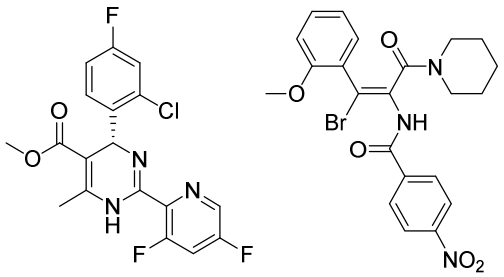
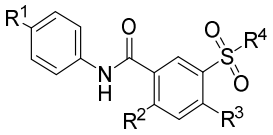
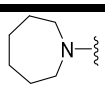
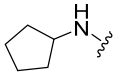
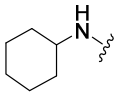


Figure 1. Two prototypical capsid assembly modulators Bay 41-4109 (CAM-II) and AT-130 (CAM-I)

Originating from our compound library, a series of *N*-phenyl-3-sulfamoyl-benzamide derivatives[8] (Table 1) was picked up, represented by compounds **1** to **5**, that inhibit the hepatitis B virus (HBV), evidenced by dose dependent reduction of HBV DNA in the HepG2.2.15 cell line. In addition, compounds **1** to **4** induce HBV core protein aggregation, resulting in the formation of particles of regular capsid size, but not larger aberrant structures, as indicated by size exclusion chromatography (data not shown). A preliminary compound optimization was initiated, exploring initial SAR while generating a suitable tool compound to assess the mode of action in a relevant *in vivo* HBV mouse model, either by oral or subcutaneous administration, as at the time of this work, *in vivo* data was only available for the heteroaryldihydropyrimidine (HAP) CAM-II class of compounds (known to aggregate the core protein in aberrant structures). At this time, with the publication of both our own and other patent applications,[8-10] Campagna et al.[11] (Sulfamoylbenzamide; SBA) Klumpp et al.[12] (Sulfamoyl Carboxamide (SOC)) and others[13] reported on *N*-phenyl-3-sulfamoyl-benzamide derivatives for HBV.

Table 1. Initial *N*-phenyl-3-sulfamoyl-benzamide derivatives.



ID	R^1	R^2	R^3	R^4	EC_{50}* (μM)	CC_{50}# (μM)	Sol$ (μM)	HLM/ MLM (%)	SEC ratio Cpd/ctrl
1	F	H	Cl		0.66	17.0	1	98/100	0.01/11.5
2	H	Cl	Cl		0.50	>25	2	99/100	0.2/13.7
3	H	Cl	H		1.04	>25	-	80/ND	0.7/9.9
4	H	H	H		0.60	27.2	91	94/98	0.3/15.2
5	H	H	F		2.23	16.4	-	100/ND	ND

*HBV DNA HepG2.2.15;# HepG2, 6 days; $ Kinetic Solubility pH 7.4 (see supporting information for details)

HLM/MLM: Human/Mouse liver microsome stability, expressed as percentage metabolisation after 15 minutes of incubation (see supporting information for details).SEC: Size exclusion chromatography. The aggregation of core protein, expressed as a ratio between the area under the curve for the observed amount of core dimer and area under the curve for the aggregated core protein, as determined in size exclusion chromatography after 24 hours of aggregation. Determined for both the compound at 20 μM (cpd) and a no-compound-control (ctrl), (see supporting information for details).

ND not determined.

Results and discussion

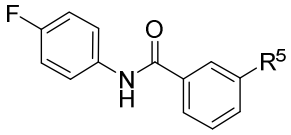
The formation of a capsid containing pre-genomic RNA (pgRNA) and the viral polymerase, is the first, cytoplasmic, step in the formation of an infectious HBV virion. Only within the viral nucleocapsid, the pgRNA is reverse transcribed to produce relaxed-circular DNA (rcDNA). As CAMs accelerate the kinetics of capsid assembly, preventing encapsidation of the Pol-pgRNA complex, they block the reverse transcription of the pregenomic RNA and the formation of rcDNA. It is hypothesized that CAMs should nucleate misassembly faster than normal HBV virion assembly.[14] Therefore, it is expected that, in case of CAMs, the kinetics of the compound induced assembly will be indicative for its anti-HBV activity (HBV DNA). In this SAR-exploration, in addition to measuring HBV DNA inhibition (EC_{50}) in stable HepG2.117 (and HepG2.2.15) cell lines, also the core protein assembly kinetics were monitored in the fluorescence quenching assay[15] (See Supporting Information).

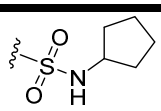
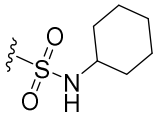
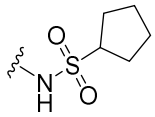
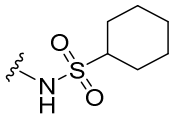
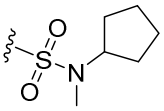
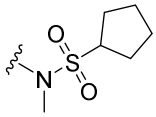
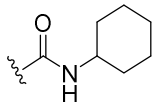
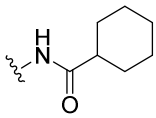
However, it is expected that the intrinsic capability of a compound to accelerate assembly, in this assay, might be clouded by a low kinetic solubility of a compound. To assess if a compound targets the capsid assembly, even if it only induces a slow assembly, either due to intrinsic capability or low kinetic solubility, the assembly was also monitored in a 24-hours assay. Here, the assembly domain (amino acids 1-149) of a recombinant HBV core protein (Cp149) was incubated with the compounds for 24 hours and the compounds tendency to assemble the Cp149, is expressed as a ratio between the area under the curve for the observed amount of Cp149 dimer and area under the curve for the aggregated Cp149, as determined by size exclusion chromatography. This is compared to the same ratio obtained without addition of compound, as in this case, also a minor amount of empty assembled capsids is formed.

As the eventual goal of this study was to assess the *in vivo* effect of this class of compounds in a humanized mouse model and the initial hits suffered from low metabolic stability, both human (HLM) and mouse liver microsome (MLM) stability were assessed. Stability in human microsomes was considered relevant for the humanized mouse model, whereas mouse liver microsome stability would allow generation of PK data in a relevant non-humanized mouse strain, but also in the context of residual presence of mouse hepatocytes in the humanized mice.

Initial SAR-exploration included the synthesis of bioisosteric derivatives of the sulphamoyl and *N*-phenyl amide moieties as depicted in table 2 and table 3 respectively. Whereas the kinetic solubility of a compound **1** and close analogues was low, possibly hampering further SAR studies, compound **4** demonstrated higher solubility. As an example, as compound **1** and **2** where clearly impacting capsid assembly as indicated by the SEC ratio (table 1), they however had limited impact in a shorter timeframe as indicated by the quenching curves (supporting information), likely explained by a low kinetic solubility of those two compounds. The higher solubility of compound **4** however, together with its high potency, resulted in faster kinetics in the quenching assay (supporting information). Compound **6**, the p-Fluoro- analogue of compound **4**, showed slightly increased metabolic stability versus compound **4**, and was used, together with its cyclohexyl analogue compound **7**, as a starting point for an initial SAR-study. Inversion of the sulfamoyl moiety, as for compound **8** and **9**, retained some, although slightly lower activity. Interestingly, this specifically improved HLM stability (compound **8** versus **6**), but not MLM stability.

Table 2. Variations and replacement of the eastern sulphamoyl part.

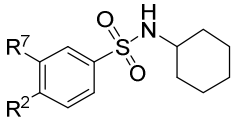


ID	R^5	EC_{50}* (μM)	EC_{50}° (μM)	CC_{50}# (μM)	HLM/ MLM (%)	Sol$ (μM)	SEC Cpd/ ctrl
6		0.47	0.6	32.7	50/91	83	ND
7		ND	0.58	>25	69/93	43	0.2/12.0
8		1.2	1.2	>50	4/93	92	ND
9		1.1	2.2	33.6	46/91	86	0.3/6.8
10		2.6	11.1	23.8	98/100	19	ND
11		7.5	16.3	43.2	86/99	53	2.0/6.6
12		>100	>25	>100	56/93	<0.4	6.4/7.2
13		62.2	>25	57.7	55/87	7	7.2/7.7

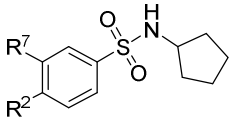
*HBV DNA HepG2.2.15; ° HBV DNA HepG2.117;# HepG2, 4 days; $ Kinetic Solubility pH 7.4

The methylated derivatives **10** and **11** showed reduced potency versus there NH-analogues, **6** and **8** respectively. Replacing the sulfamoyl moiety by an amide, as in **12** and **13**, rendered the compound inactive, without any CAM property indicated by the SEC ratio.

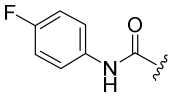
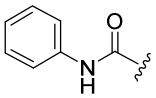
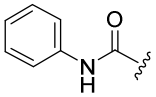
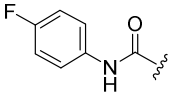
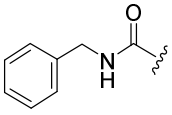
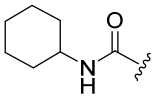
Table 3. Variation and replacement of the western *N*-phenyl amide part

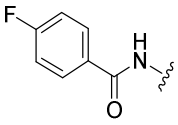
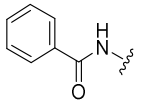
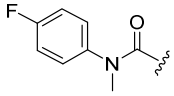
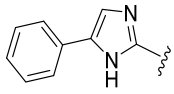
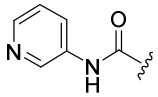
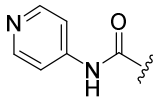
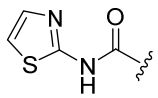
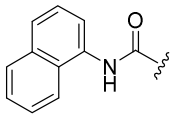
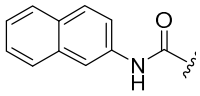
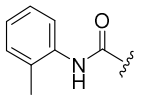
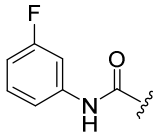
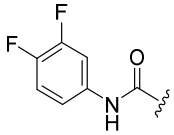


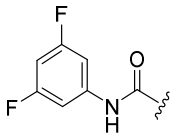
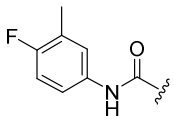
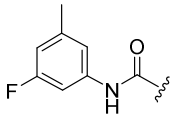
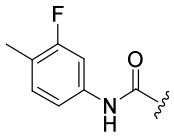
A



B

ID	A or B	R^7	R^2	EC_{50}* (μM)	EC_{50}° (μM)	CC_{50}# (μM)	HLM/ MLM (%)	Sol$ (μM)	SEC Cpd/ctrl
14	A		F	0.78	1.30	>50	61/79	58	0.3/9.2
15	A		F	0.66	0.60	>25	63/100	21	0.1/11.5
4	B		H	0.60	0.76	>25	94/98	91	0.3/15.2
16	B		F	0.36	0.66	>25	36/77	82	ND
17	A		F	>100	-	93.9	74/-	29	8.4/11.0
18	A		F	28.8	>25	>25	-/-	-	ND

19	A		H	8.0	>25	>25	91/100	22	ND
20	A		H	9.6	>25	>25	99/100	10	7.0/7.2
21	B		H	89.6	>25	>25	100/100	>100	ND
22	A		F	1.7	23.3	19.2	75/99	3	10.2/9.9
23	A		F	ND	24.8	>25	57/99	>100	9.8/11.8
24	A		F	5.3	16.29	>25	68/75	>100	6.0/10.0
25	A		F	29.3	>25	>25	85/100	>100	9.7/10.0
26	A		F	15.6	>25	>25	94/100	12	ND
27	A		F	12.7	>25	13.0	36/56	<0.2	7.5/7.7
28	A		H	4.18	-	>25^	86/ND	ND	ND
29	B		H	0.27	0.43	>25	88/100	57	ND
30	B		H	0.093	0.36	>25	45/86	46	ND

31	B		H	0.28	0.78	>25	79/100	28	ND
32	B		H	0.10	0.42	48.2	66/97	33	0.02/9.3
33	B		H	0.11	1.51	>23	74/100	11	ND
34	B		H	1.99	15.3	>25	67/99	12	ND

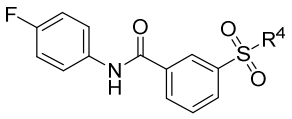
[*]HBV DNA HepG2.2.15; ° HBV DNA HepG2.117; [#] HepG2, 4 days; [^] HepG2, 6 days; $ Kinetic Solubility pH 7.4

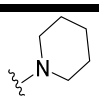
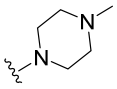
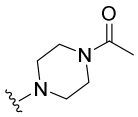
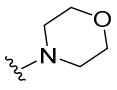
Assessment of replacements of the western *N*-phenyl amide part was performed relative to compounds **4**, **14**, **15** and **16** (Table 3). First, when looking at the impact of R_2=F on the central scaffold (R^2, Table 3) in **14** and **16** versus hydrogen substituted compound **7** and **6** respectively, a limited impact was noticed. For isosteric replacements, both R^2: F and H (Table 3) were used. Replacement of the F-Phenyl of **14**, or phenyl of **15**, by either a benzyl (**17**) or a cyclohexyl (**18**) deteriorated the potency of the compound (as indicated by HBV DNA EC_{50}, SEC ratio and/or quenching). In addition, inverting the amide of **7**, resulting in **19**, or the related compound **20**, diminished the activity significantly, without any indication of remaining CAM properties (table 3, SEC ratio for **20**). *N*-Methylation of the amide of **6**, as in **21** also resulted in significantly reduced activity (lack of quenching and significantly increased HBV DNA EC_{50}), altogether indicating the essential role of the -NH-phenyl amide. Additionally, replacement of the -*N*-phenyl amide by a phenyl-imidazole, as in **22** (compared to **15**), did not results in a CAM compound as indicated by the high SEC ratio versus the control. When for this scaffold, the N-

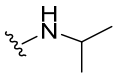
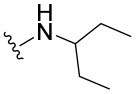
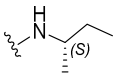
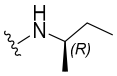
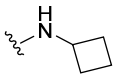
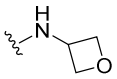
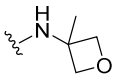
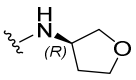
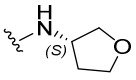
phenyl was replaced by either a -*N*-Heteroaryl (Table 3, **23**, **24**, **25**) or a *N*-naphthyl (compounds **26**, **27**), no relevant CAM property could be picked up according to SEC and/or quenching.

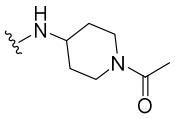
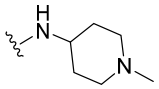
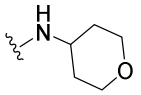
An early assessment of the substitution pattern on the *N*-phenyl, indicated that switching the *p*-Fluoro of **6** to the *ortho* position of the *N*-phenyl, as in **29**, retained activity. Reintroduction of a *p*-Fluoro (**30**) or a *m*-Fluoro (**31**) both led to potent CAM's. Similarly, replacing one of the *m*-Fluoro's in **30** and **31** by a *m*-Methyl, as in **32** and **33** respectively maintained the high potency, both on HBV reduction as on the SEC and quenching assays. Nevertheless, when replacing the *p*-Fluoro by a *p*-Methyl, as in **30** versus **34**, or introducing an *o*-Methyl, as in **28** versus **4** or **15**), potency is significantly reduced (EC_{50} HBV DNA).

Table 4. SAR of the sulphonamide substitutent R^4.



ID	R^4	EC_{50}* (μM)	EC_{50}° (μM)	CC_{50}# (μM)	Sol$ (μM)	HLM/ MLM (%)	SEC Cpd/ctrl
35		4.50	2.7	70.4	9	65/99	0.4/7.2
36		26.5	11.1	>100	>100	53/84	6.3/10.8
37		7.10	6.3	>100	86	19/52	0.9/7.7
38		8.49	11.0	>100	>100	27/69	0.6/9.2

39		4.53	3.03	96.9	48	44/96	ND
40		5.02	2.99	>100	43	50/89	1.4/9.2
41		0.74	0.96	57.5	>100	16/61	ND
42		0.75	3.63	40.3	15	53/99	ND
43		0.75	8.63	39.6	92	18/82	ND
44		0.34	0.89	46.0	59	21/88	ND
45		1.24	2.28	52.5	1	15/31	0.9/10.8
46		0.83	0.90	57.7	84	51/99	ND
47		2.10	3.05	>100	>100	14/21	0.8/7.7
48		0.96	0.93	>100	>100	3/29	0.5/11.9
49		1.18	2.03	>100	>100	21/24	ND
50		0.54	1.36	>100	>100	32/29	ND

51		6.77	4.68	>100	>100	0/23	2.9/10.9
52		5.3	16.0	>100	94	10/13	4.2/9.6
53		0.93	1.67	>100	>100	14/48	ND

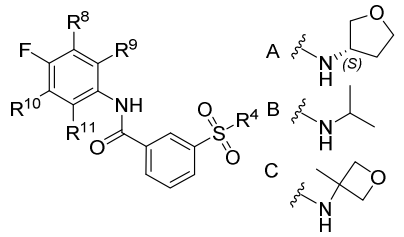
*HBV DNA HepG2.2.15; ° HBV DNA HepG2.117; # HepG2, 4 days; $ Kinetic Solubility pH 7.4

In table 4, the alteration of the sulphone amide substitution was investigated. Introduction of secondary amines on R^4 (**35**-**40**) rendered compounds that were still showing CAM properties, in the case of **35**, **37**-**40** (SEC ratio and/or quenching), but nevertheless with reduced activity versus **6** and **7** on HBV DNA EC_{50}. Introduction of the basic piperazine as in **36**, significantly impacted the CAM properties according to quenching and SEC ratio, while maintaining some potency on HBV DNA reduction. Interestingly, branched NH-alkyls like -iPr (**41**), 1-ethylpropyl (**42**) or *sec*Butyl, with a preference for the *R*-isomer (**44** versus **43**), displayed good potency, and for example in the case of **41** versus compounds **6** and **7** also improved metabolic stability on both HLM and MLM was observed. Slightly decreased potency (HBV DNA), nevertheless improved metabolic stability, was observed for the *t*Bu-derivative **45**. Introduction of a NH-*c*Bu showed similar stability (and slightly lower potency) versus compound **6**, while the corresponding oxetane (**47**) loses potency (HBV DNA), and gains in metabolic stability, both on MLM and HLM. Introduction of an extra methyl on the oxetane (**48**), returned potency (HBV DNA) while retaining metabolic stability (versus compound **47**). The introduction of a heteroatom also

improved metabolic stability for other ring sizes, as observed for tetrahydrofuryl (**49** and **50**) versus cyclopentyl and tetrahydro-2H-pyranyl (**53**) versus cyclohexyl (**7**), in addition, an improved kinetic solubility was observed. Although introduction of a nitrogen in the cyclohexyl of **7** also increased metabolic stability (compound **51** and **52**), a decreased potency was observed (HBV DNA).

Using some of the optimal substituents from table 4, taking into account potency, solubility and metabolic stability, R^4 substituents A, B and C (Table 5) were selected for a further combination with the optimal substituent patterns from table 3.

Table 5. SAR of *N*-Phenyl Substitution.



ID	R^4	R^8	R^9	R^{10}	R^{11}	EC_{50}* (μM)	EC_{50}° (μM)	$CC_{50}$$ (μM)	Sol (μM)	HLM/ MLM (%)
54	A	Me	H	H	H	0.12	0.43	>50	82	26/51
55	A	Me	F	H	H	0.29	0.78	>25	>100	25/66
56	A	Me	H	H	F	2.22	3.30	>25	>100	17/69
57	A	F	Me	H	H	3.63	1.91	>25	>100	37/68
58	A	F	H	H	H	0.38	0.51	>50	>100	5/42
59	A	CN	H	H	H	0.81	0.98	>50	>100	13/27
60	A	OCH_3	H	H	H	2.33	2.66	>25	>100	20/35
61	B	Me	H	H	H	0.07	0.15	47.5	41	26/80
62	B	Et	H	H	H	0.31	2.59	28.5	15	31/88

63	B	F	H	H	H	0.21	0.40	25.2	>100	18/60
64	B	iPr	H	H	H	1.40	2.79	29.8	13	31/96
65	B	cPr	H	H	H	0.32	0.91	35.2	31	27/61
66	C	Me	H	H	H	0.12	0.30	47.1	>100	7/65
67	C	Me	Me	H	H	2.74	1.65	>25	>100	0/77
68	C	Me	H	H	Me	3.06	1.91	>25	>100	1/78

[*]HBV DNA HepG2.2.15; ° HBV DNA HepG2.117; [#] HepG2, 4 days; [$] Kinetic Solubility pH 7.4

Combination of substituents A, B or C with either the *N*-(4-fluoro-3-methylphenyl)amide (Table 5: **54**, **61** or **66** respectively) or *N*-(3,4-difluorophenyl)amide (Table 5: **58**, **63** respectively) led to compounds with improved potency and/or metabolic stability versus **6** (and very significant effect on quenching, see supporting information). Further introduction of a Fluor on R^9 (**55**) or R^{11} (**56**) versus compound **54** did not results in improved potency or significant stability (in contrast the potency of compound **56** is significantly reduced). In addition, introduction of a methyl on R^9 (**57** versus **58**, or **67** versus **66**) or R^{11} (**68** versus **66**), reduces potency (HBV DNA) significantly. Variation of the *N*-Phenyl *m*-alkyl, as in compounds **62** (Et), **64** (iPr) and **65** (cPr), reduced potency (HBV DNA) versus the methyl analogue **61.** Replacing the *m*-Methyl (R^8) in **54** by *m*-OMethyl (**60**) reduced potency (HBV DNA), while variation to *m*-cyano (**59**), resulted in decreased potency, metabolic stability was further improved.

Among several suitable candidates (compounds **54**, **58**, **61**, **63** and **66**) that were further profiled *in vitro,* **54** (JNJ-632) was selected based on it's suitable *in vitro* profile, for further *in vivo* characterization. In the high-content multiparameter cytotoxicity (HepG2), JNJ-632 showed EC_{20}'s in the 10-30 μM range (considered weakly cytotoxic. Notably, in contrast, **63** was considered cytotoxic, with and EC_{20} ranging down to 1 μM). [16, 17]

While JNJ-632 was clean in kinase panel (1 μM, n=240), a receptor, ion channel and transporter assay profiling panel (n=80), resulted in one hit at 10 μM (5-HT2B (71%)). The compound didn't show agonist activity of 5-HT2B, but was an antagonist of 5-HT2B with IC_{50} of 5.6 μM. Activity on the serotonin receptor 2B was observed across several analogues in this series, with **12** and **63** as more potent examples (IC_{50} 5HT2B: 0.09 μM and 0.23 μM respectively versus 4.0 μM for JNJ-632). JNJ-632 did not significantly bind to NaCH, CaCH or hERG with IC_{50}> 10 μM and an automated hERG patch clamp system indicated that **54** showed 31% of IK_r inhibition at 3 μM (versus 14% of IK_r inhibition by the solvent). Moreover, JNJ-632 exhibits low CYP inhibition with IC_{50} > 10 μM against six major CYP enzymes (CYP1A2, CYP2C8, CYP2C9, CYP2C19, CYP2D6 and CYP3A4). **54** was found positive in GSH (glutathione) trapping assay, (oxidative defluorination followed by GSH addition) in human liver microsomes. No cleavage of the *N*-phenylamide was observed in primary hepatocytes, in contrast to related analogues like compound **66** (data not shown). Finally, JNJ-632 tested negative in Ames II and the *in vitro* micronucleus test.

The permeability of JNJ-632 was measured in the LLC-PK1-MDR1 cell line in A to B direction in absence and presence of the P-gp inhibitor GF-120,918. The apparent permeability value Papp (A-B) (-GF) was 7.3 x 10^{-6} cm/s and the apparent permeability value Papp (A-B) (+GF) was 16.2 x 10^{-6} cm/s. The ratio Papp (A-B) (+GF)/Papp(A-B) (-GF) indicated that the compound was a weak P-gp substrate. The unbound fraction of JNJ-632 was determined to be 12.6% and 8.1% in mouse and human plasma, respectively. Notably, when determining plasma protein binding, for certain compounds, not for JNJ-632, a low recovery was noticed specifically in the case of mouse plasma, not in human plasma, possibly indicating instability in the former

medium. Finally, JNJ-632 gave 51% turnover after 15 min incubation in mouse liver microsomes and 26% turnover after 15 min incubation in human liver microsomes.

The single dose PK profile of JNJ-632 was evaluated in C57BL/6 mice following intravenous (i.v.) and oral (p.o.) administration. JNJ-632 had a moderate plasma clearance of 34 mL/min/kg and a moderate volume of distribution of 1.3 L/kg. The oral bioavailability was 40% following oral administration of 10 mg/kg and 66% following oral administration of 50 mg/kg. To circumvent the first pass metabolism, compound was also dosed subcutaneously at 50 mg/kg in C57BL/6 mice and this resulted in a concentration in plasma after 24 h of dosing of 102 ng/mL and concentration in liver after 24 h of dosing of 1297 ng/g (Table 6). Finally, the PK of **54** was assessed in uninfected humanized mice (Phoenix Bio®) following subcutaneous administration of 50 and 200 mg/kg as a methocel suspension. Compound JNJ-632 was dosed for 8 days and PK was evaluated on days 1 and 8. Repeated subcutaneous dosing of a suspension resulted in higher exposure at day 8 versus day 1 (Table 7).

Table 6. PK parameters C57BL/6 mice for **54**

Dose (mg/kg)	CL (mL/min/kg)	V_{ss} (L/kg)	$t_{1/2}$ (h)	C_{max} (ng/mL)	T_{max} (h)	AUC $_{0\text{-last}}$ (ng.h/mL)	F (%)
2.5 (i.v.)	34 ±9.2	1.3 ±0.20	0.42 ±0.06	-	-	1240 ±303	-
10 (p.o)	-	-	1.1 ±0.67	1940 ±296	0.5	1990 ±159	40 ±3.3
50 (p.o)	-	-	2.4 ±2.3	9440 ±1420	0.7 ±0.3	16400 ±2350	66 ±9.5
50 (s.c)	-	-	5.3 ±0.1	3367 ±342	1.0	16877 ±2959	68 ±12

Table 7. Exposure of JNJ-632 in uninfected humanized mice after subcutaneous administration of 50 and 200 mg/kg.

QD Dose (mg/kg)	Dosing day	C_{max} (ng/mL)	T_{max} (h)	AUC_{0-24} (ng.h/mL)	$AUC_{0-\infty}$ (ng.h/mL)	$t_{1/2}$ (h)	$C_{plasma,24h}$ (ng/mL)	$C_{liver,24h}$ (ng/g)
50	Day 1	2190 ±246	1	4750* ±326	4780 ±313	1.1 ±0.2		
	Day 8	4380 ±1280	0.8 ±0.4	19200 ±2180	20700 ±2850	6.2 ±1.1	167 ±70.2	570 ±168
200	Day 1	2110 ±941	0.8 ±0.4	12000 ±1060	12700 ±561	5.6 ±1.7		
	Day 8	7700 ±2490	1	86900 ±6520	119000 ±4910	12.1 ±3.2	1820 ±179	4880 ±1060

*$AUC_{(0-8h)}$ was used instead of $AUC_{(0-24h)}$

Antiviral activity of JNJ-632 in HBV-infected chimeric mice

The anti-HBV activity of JNJ-632 was tested in HBV infected primary human hepatocytes (PHHs) derived from naive chimeric mice. qPCR analysis of the HBV DNA in the cell culture supernatant showed anti-HBV activity with an EC_{50} of 200 nM for genotype D, which is similar to the EC_{50} obtained in HepG2.2.15 cells (genotype D).[2] The anti-HBV activity of JNJ-632 was tested in an *in vivo* proof of mechanism study in genotype D HBV-infected chimeric mice with humanized liver. HBV-infected chimeric mice were used extensively for HBV research in the past due to authentic HBV infection of human hepatocytes and persistent formation of cccDNA *in vivo*.[18] All mice used in this study had a human hepatocyte replacement index of 70% or more as assessed by the level of human albumin in the serum (data not shown).

JNJ-632 was dosed subcutaneously once a day over 28 days at a dose of 200 mg/kg, whereas saline and ETV was dosed once a day orally and served as negative and positive control

respectively. None of the compounds reduced human albumin levels in the serum over time (data not shown), indicating that the treatment did not have cytotoxic effects on the human hepatocytes in the liver of the mice. A mean -2.77 log reduction of HBV DNA in the serum was observed for JNJ-632, demonstrating that the compound inhibited HBV replication in vivo (Figure 3). No reduction of HBe- and HBsAg levels was observed during this 28-days study (data not shown).

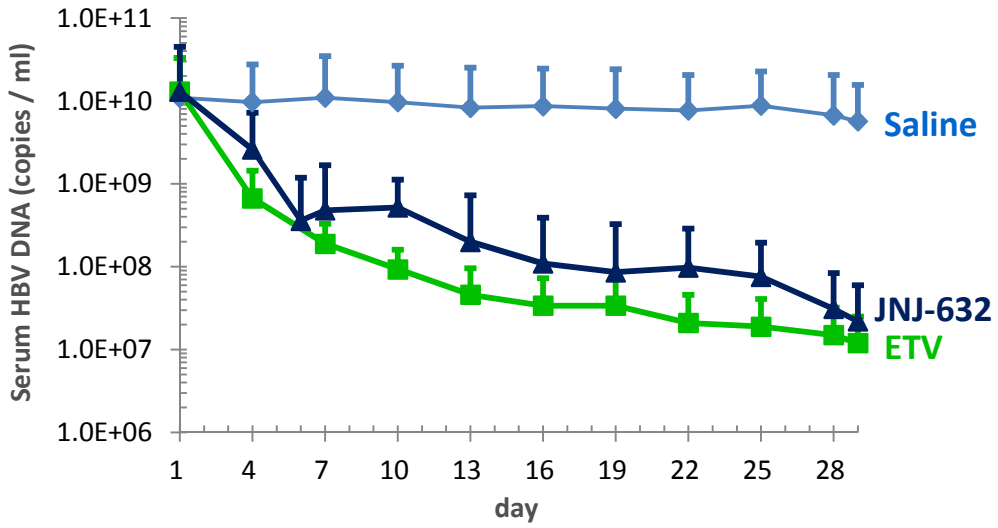


Figure 3: Antiviral activity in HBV genotype D infected chimeric mice. Mean HBV DNA change in serum after daily treatment with saline, ETV and JNJ-632 for 28 days.

Table 8. Mean HBV DNA change of ETV and JNJ-632 versus the saline control in HBV genotype D infected chimeric mice.

	Mean HBV DNA change (SD)*
Saline	-0.29 (0.14)
ETV 0.03mg/kg QD, p.o.	-3.03 (0.19)
JNJ-632 200mg/kg QD, s.c.	-2.77 (0.10)

*from baseline at day 29

HBV genotype D infected chimeric mice with a human hepatocyte replacement index of 70% or more were treated once a day for 28 days with saline (oral), 0.03mg/kg ETV (oral) and 200 mg/kg JNJ-632 (subcutan; s.c.). Values in the table represent the mean HBV DNA change from baseline at day 29 and the standard deviation (SD) from 4 (ETV) and 5 (saline and JNJ-632) mice per group respectively.

Docking of JNJ-632

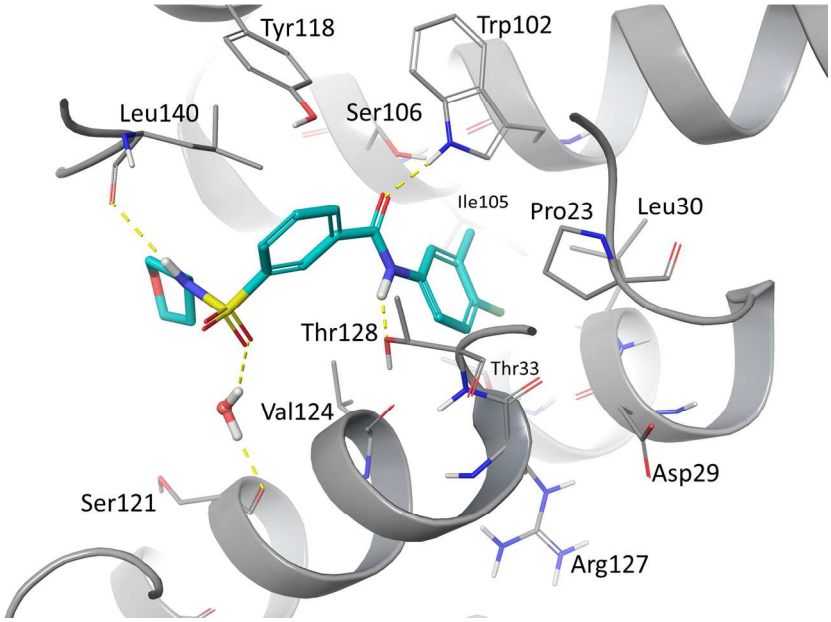


Figure 2. Docked binding mode of JNJ-632.

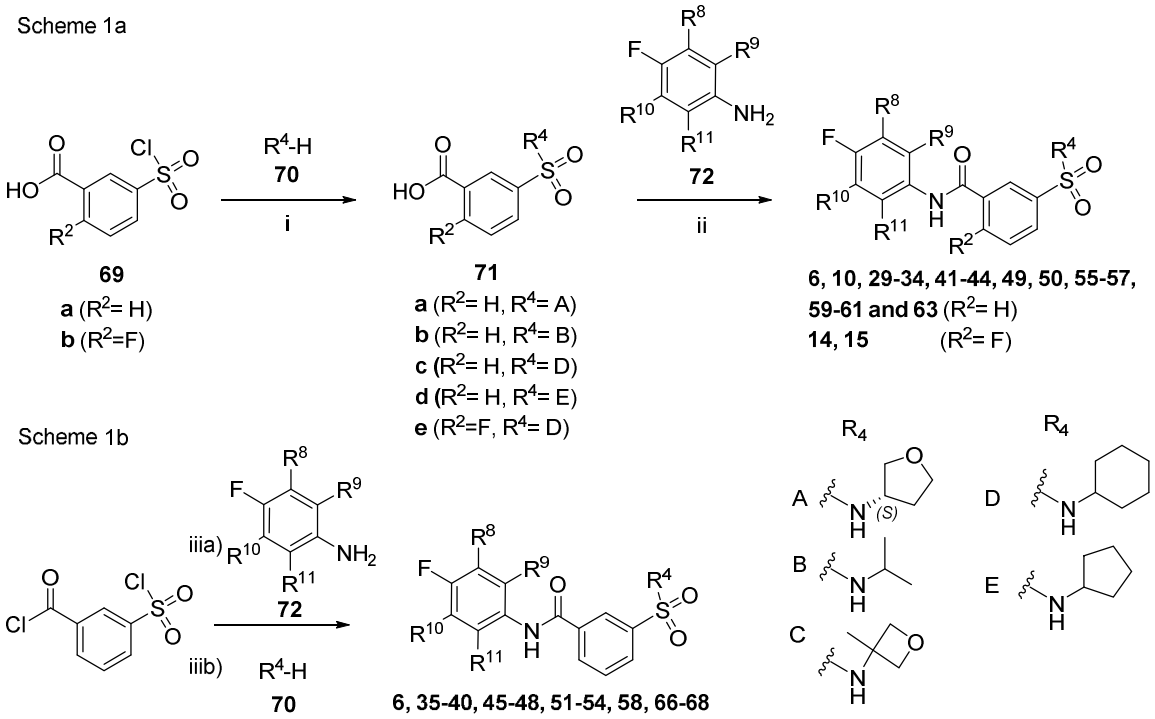
JNJ-632 was docked to adopt a binding mode as depicted in figure 2. The Methyl-Fluor-phenyl group binds in a hydrophobic pocket surrounded by residues Pro25, Asp29, Leu30, Thr33, Trp102, Ile105, Ser106, Val124, Arg127 and Thr128. The amide group makes hydrogen bonds with Trp102 and Thr128, while the sulfonamide group picks up a hydrogen bond with Leu140 and a water mediated hydrogen bond with Ser121. The tetrahydrofuryl group resides in a solvent-accessible area. It is possible that this part of the inhibitor helps in stabilizing the arginine-rich domain of the C-terminus, which is not part of the crystal structure. [19]

Chemistry

New 3-sulfamoyl-*N*-phenylbenzamide compounds were prepared according to the general scheme 1 starting from either 3-(chlorosulfonyl)benzoic acid derivatives (Scheme 1a) or 3-(chlorosulfonyl)benzoyl chloride (scheme 1b).[8] As depicted in scheme 1a, starting from 3-(chlorosulfonyl)benzoic acid **69a**, for compounds **7**, **10**, **29-34, 41-44, 49, 50, 55-57, 59-61** and **63** and from 5-(chlorosulfonyl)-2-fluorobenzoic acid **69b** for compounds **14** and **15**, the

chlorosulfone was condensed with the corresponding amine in the presence of a base, followed by amide formation under influence of HATU, or by activation of the carboxylic acid to the corresponding acid chloride followed by amide formation. Alternatively, for the synthesis of compounds **6, 35-40, 45-48, 51-54, 58, 66-68** applying the well described difference in reactivity of carboxy acid chlorides and sulfonyl chlorides, [20,21] 3-(chlorosulfonyl)benzoyl chloride is first reacted with an aniline **72**, resulting in the formation of an amide, followed by the reaction of another amine **70**, resulting in the formation of the sulfonamide. Synthesis of compounds **8**, **9**, **11-13**, **17-27, 62**, **64** and **65** is described in the supporting information.

Scheme 1. Synthesis new *N*-phenyl-3-sulfamoyl-benzamide derivatives.



Reagents and conditions: i) **69a**, **70**, CH_2Cl_2, NEt_3, room temperature, overnight or **69a**, **70**, NaOH, THF, 20-25°C, 2-3 hours, **71a-d**: 33-93% yield; **71b**: EtOAc, $iPrNH_2$, 25°C, 3 hours, 79 % yield, **71e**: **69b**, cyclohexamine, room temperature, EtOAc, 10 minutes, 86 % yield ii) HATU, CH_2Cl_2, DIPEA, 0°C to room temperature, 2 h to overnight, 9-66 % yield; or 13-39 % yield over

two steps from **69a** (**7**, **10**, **42**, **44**); **14** and **15**: **71e**, **72**, HATU, CH_2Cl_2, DIPEA or NEt_3, 2-3 hours, 28-52 % yield; **55**, **56**, **59** and **60**: **71a**, **72**, DMF/CH_2Cl_2, $(COCl)_2$, 2 hours, 0 to 20°C, 2 hours then NEt_3, CH_2Cl_2, 0°C to 20°C, 2 hours, 14-61 % yield; **57**: **71a**, 3,4-difluoro-2-methyl aniline, DIPEA, Pybrop, DMF, 18°C, 23 % yield. iiia) **72**, CH_2Cl_2, NEt_3, 0°C then 20°C, 1 h or toluene, reflux, 30 min iiib) **70**, NEt_3 0°C then 20°C, 1h; 10-50 % yield over two steps, or (**54**) **70**, toluene/CH_2Cl_2, DIPEA, 4 h room temperature, 39 % two steps.

Conclusions

In summary, the authors described a hit-to-lead optimization, resulting in the discovery of JNJ-632 (**54**), a *N*-phenyl-3-sulfamoyl-benzamide derivative and HBV capsid assembly modulator that can inhibit HBV *in vitro* (HepG2.117, HepG2.2.15 and HBV infected primary human hepatocytes) and *in vivo (*HBV infected humanized mice*)*. Further optimization towards JNJ-6379, a capsid assembly modulator currently in phase 2 clinical trials,[3] will be published in due course.

Experimental section

Chemistry

Reagents and solvents were purchased from commercial sources and used without purification. Purity of commercial, tested compounds was assessed by at least one LC-MS method and that of all synthetized and tested compounds by two independent LC-MS methods. Compounds exhibited greater than 95% purity, except for compound **26** which was tested at 94-95 % purity. Purities and LC methods are listed in the supporting information.

NMR spectra were recorded on Bruker DPX-400, Bruker Avance I-400, Bruker Avance III-400 and Bruker Avance III-H- 600 spectrometers, operating at the frequencies specified. High resolution mass spectrometry was performed on a Waters Acquity IClass UPLC-DAD and Xevo G2-S QTOF. The samples were run on a Waters BEH C18 (1.7 μm, 2.1 mm × 50 mm, at 50 °C) column using reversed-phase chromatography with a gradient from 95% A to 5% A in 4.6 min, held for 0.4 min (A, 95% 6.5 mM CH_3COONH_4/5% CH_3CN; B, CH_3CN).

3-(cyclopentylsulfamoyl)-*N*-(4-fluorophenyl)benzamide (6). Synthesis according to the representative procedure of scheme 1b as described for compound **53.** 21% yield. Method B; Rt: 4.27 min. m/z: 363.1 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 363.1184 (calcd for $C_{18}H_{19}FN_2O_3S$: 362.1100). ^{1}H NMR (600 MHz, DMSO-d_6) δ 10.50-10.62 (m, 1H), 8.36 (t, *J*=1.70 Hz, 1H), 8.19 (ddd, *J*=1.00, 1.70, 7.80 Hz, 1H), 8.00 (ddd, *J*=1.00, 1.70, 7.80 Hz, 1H), 7.74-7.82 (m, 4H), 7.19-7.25 (m, 2H), 3.41-3.48 (m, 1H), 1.49-1.64 (m, 4H), 1.33-1.43 (m, 1H), 1.24-1.32 (m, 2H). ^{13}C NMR (151 MHz, DMSO-d_6) δ 164.2, 158.5, 142.1, 135.6, 135.2, 131.1, 129.5, 129.3, 125.9, 122.4, 115.3, 54.5, 32.4, 22.8.

3-(cyclohexylsulfamoyl)-*N*-(4-fluorophenyl)benzamide (7). To a solution of 3-(chlorosulfonyl)benzoic acid (1 g, 4.53 mmol) in CH_2Cl_2 (20 mL) at 5°C, cyclohexanamine (0.899 g, 9.06 mmol) and triethylamine (1.38 g, 13.60 mmol) were successively added drop wise. The solution was stirred at room temperature overnight. The mixture was washed with 1N HCl (50 mL). The organic phase was dried over $MgSO_4$ and concentrated resulting in 3-(*N*-cyclohexylsulfamoyl)benzoic acid as a white solid (1.2 g, 93%), which was used in the next step without purification. To a solution of 3-(*N*-cyclohexylsulfamoyl)benzoic acid (1.2 g, 4.24 mmol) in DMF (15 mL) at 5°C, 4-fluoroaniline (0.52 g, 4.66 mmol) and DIPEA (1.64 g, 12.71 mmol) were successively added.. The mixture was stirred for 20 minutes and then HATU (1.93 g, 5.08

mmol) was added. The solution was stirred at room temperature overnight. To the reaction mixture aqueous $NaHCO_3$ (50 mL) was added followed by EtOAc (50 mL). The organic layer was washed with HCl (5%; 50 mL) and brine. The organic layer was dried with $MgSO_4$ and concentrated, resulting in a residue. The obtained residue was purified by a silica gel chromatography column (Petroleum ether:EtOAc=2:1) resulting in compound **7** as a white solid (850 mg, 25 % yield over two steps). Method B; Rt: 4.50 min. m/z: 377.2 $(M+H)^+$ ESI-HRMS (TOF) m/z: [M + H]+ 377.1339 (calcd for $C_{19}H_{21}FN_2O_3S$: 376.1257). ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.55 (s, 1H), 8.36-8.39 (m, 1H), 8.16-8.22 (m, *J*=8.10 Hz, 1H), 7.99-8.05 (m, *J*=8.10 Hz, 1H), 7.74-7.83 (m, 4H), 7.19-7.25 (m, 2H), 2.94-3.04 (m, 1H), 1.51-1.63 (m, 4H), 1.40-1.51 (m, 1H), 1.00-1.23 (m, 5H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.2, 158.4, 142.8, 135.5, 135.2, 131.0, 129.4, 129.0, 125.6, 122.4, 115.2, 52.1, 33.2, 24.8, 24.3

3-[cyclopentyl(methyl)sulfamoyl]-*N*-(4-fluorophenyl)benzamide (10).

To a solution of 3-(chlorosulfonyl)benzoic acid (1.10 g, 4.97 mmol) in THF (60 mL), sodium hydroxide was added (aq., 2 mL, 5N) while cooled in an ice bath, followed by adding *N*-methyl-cyclopentanamine (0.50 g, 4.97 mmol). After stirring at 25°C for 3 hours, the reaction mixture was diluted with H_2O (50mL) and extracted with EtOAc (50 mL). The aqueous layer was adjusted to pH=3 by HCl (2N) and extracted with EtOAc (3 x 50 mL). The combined organic layer was washed with brine, dried over anhydrous $MgSO_4$ and concentrated in vacuo resulting in 3-(*N*-cyclopentyl-*N*-methylsulfamoyl)benzoic acid (0.8 g). To a solution of 3-(*N*-cyclopentyl-*N*-methylsulfamoyl)benzoic acid (0.80 g, 2.82 mmol), 4-fluoroaniline (0.31 g, 2.82 mmol), and HATU (1.61 g, 4.24 mmol) in CH_2Cl_2 (10 mL), cooled in an icebath, DIPEA (1.09 g, 8.47mmol) was added under N_2 atmosphere. The resulting mixture was diluted with CH_2Cl_2 (15 mL) and washed with saturated aqueous $NaHCO_3$ (15 mL) and brine (10 mL), dried over anhydrous

$MgSO_4$ and the solvent was removed in vacuo. The obtained residue was purified by preparative high performance liquid chromatography over RP-18 (eluent: CH_3CN in H_2O from 30% to 80%, v/v; 0.05% TFA as addition). The pure fractions were collected and the volatiles were removed in vacuo. The aqueous layer was adjusted to Ph=7 with Amberlite IRA-900 ion exchange resin (OH form), filtrated and lyophilized to dryness resulting in compound **10** (0.73 g, 39 % two steps). Method B; Rt: 4.43 min. m/z: 377.2 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 377.1337 (calcd for $C_{19}H_{21}FN_2O_3S$: 376.1257). ^1H NMR (400 MHz, DMSO-d_6) δ 10.55 (s, 1H), 8.31-8.35 (m, 1H), 8.22-8.27 (m, *J*=7.70 Hz, 1H), 7.97-8.02 (m, *J*=8.10 Hz, 1H), 7.74-7.82 (m, 3H), 7.18-7.25 (m, 2H), 4.29 (quin, *J*=7.97 Hz, 1H), 2.68 (s, 3H), 1.26-1.54 (m, 8H); ^{13}C NMR (101 MHz, DMSO-d_6) δ 163.9, 158.5, 139.1, 135.6, 135.1, 131.7, 129.8, 129.7, 125.9, 122.5, 115.2, 57.8, 28.5, 27.3, 23.6

5-(cyclohexylsulfamoyl)-2-fluoro-*N*-(4-fluorophenyl)benzamide (14).

5-(*N*-cyclohexylsulfamoyl)-2-fluorobenzoic acid (2.0 g, 6.637 mmol), 4-fluoroaniline (0.740 g, 6.660 mmol) and HATU (3.78 g, 9.941 mmol) were dissolved in dichloromethane (40 ml) at 20°C. Triethylamine (1.34 g, 13.3 mmol) was added to the reaction mixture. The reaction mixture was stirred for 2 hours at 20°C. The mixture was was washed with saturated aqueous K_2CO_3 (20 ml), brine and dried over Na_2SO_4. The solvent was removed under vacuum. The crude product was purified by preparative high performance liquid chromatography (column: Phenomenex Synergi C18 250*50mm*10um, mobile phase: CH_3CN in water (TFA 0.1%) from 20% to 65%, flow rate: 60 ml/min). The pure fractions were collected and washed with aqueous Na_2CO_3, extracted with DCM (40 mL). The organic layer was concentrated in vacuum to give compound **14** (1080 mg, 52 %). Method C; Rt: 4.21 min. m/z: 395.1 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 395.1244 (calcd for $C_{19}H_{20}F_2N_2O_3S$: 394.1163). ^1H NMR (400 MHz, DMSO-d_6)

δ ppm 10.68 (1 H, br. s), 8.08 (1 H, dd, *J*=6.0, 2.5 Hz), 8.01 (1 H, ddd, *J*= 8.5, 4.5, 2.5 Hz), 7.83 (1 H, br. s), 7.70 - 7.77 (2 H, m), 7.60 (1 H, app. t, *J*= 9.0 Hz), 7.18 - 7.27 (2H, m), 2.90 - 3.07 (1 H, m), 1.53 - 1.67 (4 H, m), 1.40 - 1.53 (1 H, m), 0.96 - 1.25 (5 H, m)**.** ^{13}C NMR (101 MHz, DMSO-d_6) δ 161.2, 160.5, 158.5, 138.9, 134.9, 130.7, 128.4, 125.3, 121.7, 117.4, 115.4, 52.1, 33.2, 24.8, 24.3.

5-(cyclohexylsulfamoyl)-2-fluoro-*N*-phenyl-benzamide (15).

To an ice cooled mixture of 5-(*N*-cyclohexylsulfamoyl)-2-fluorobenzoic acid (602 mg, 2 mmol, 1.0 eq.), aniline (223 mg, 2.4mmol, 1.2 eq.), HATU (1.14 g, 3.06 mmol, 1.5 eq.) in CH_2Cl_2 (15 mL) was added DIPEA (0.774 g, 6 mmol, 3.0 eq.) under N_2 atmosphere. The resulting mixture was stirred at room temperature for 3 h. The reaction mixture was diluted with CH_2Cl_2 (30 mL) and washed with saturated aq.$NaHCO_3$ solution (10 mL), brine (10 mL) and dried over Na_2SO_4. The organic layer was concentrated under vacuum. The crude product was purified by preparative high performance liquid chromatography preparative high-performance liquid chromatography (column: C18, eluent: CH3CN/H2O from 35/65 to 60/40, 0.1% CF_3COOH). The desired fraction was collected and basified by saturated $NaHCO_3$ (aq.). The mixture was concentrated and extracted with CH_2Cl_2 (30 mL* 3). The organic layer was separated, dried over Na_2SO_4 and the solvent was removed under vacuum. The product was dried under vacuum to result in compound **15** (210 mg, 28%). Method C; Rt: 4.17 min. m/z: 377.1 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 377.1334 (calcd for $C_{19}H_{21}FN_2O_3S$: 376.1257). ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.59 (s, 1H), 8.06-8.11 (m, 1H), 7.97-8.05 (m, 1H), 7.81 (br s, 1H), 7.68-7.76 (m, 2H), 7.59 (t, *J*=9.15 Hz, 1H), 7.38 (t, *J*=7.53 Hz, 2H), 7.09-7.19 (m, 1H), 2.99 (br s, 1H), 1.60 (br d, *J*=7.73 Hz, 4H), 1.46 (br d, *J*=11.80 Hz, 1H), 0.96-1.28 (m, 6H); ^{13}C NMR (101 MHz, DMSO-d_6) δ 161.3, 160.4, 138.8, 138.5, 130.6, 128.8, 128.4, 125.5, 124.1, 119.8, 117.4, 52.1,

33.2, 24.8, 24.3;

3-(cyclopentylsulfamoyl)-*N*-(3-fluorophenyl)benzamide (29).

66% yield from 3-(*N*-cyclopentylsulfamoyl)benzoic acid according to the representative procedure of scheme 1a as described for compound **32**. Method B; Rt: 5.45 min. m/z: 363.2 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 363.1178 (calcd for $C_{18}H_{19}FN_2O_3S$: 362.1100). ^1H NMR (400 MHz, DMSO-d_6) δ ppm 1.23 - 1.43 (m, 4 H) 1.47 - 1.64 (m, 4 H) 3.38 - 3.51 (m, 1 H) 6.97 (td, *J*=8.3, 2.5 Hz, 1 H) 7.41 (td, *J*=8.3, 7.0 Hz, 1 H) 7.54 - 7.59 (m, 1 H) 7.71 - 7.85 (m, 3 H) 8.02 (ddd, *J*=7.8, 1.5, 1.0 Hz, 1 H) 8.19 (ddd, *J*=7.8, 1.5, 1.0 Hz, 1 H) 8.35 (t, *J*=1.5 Hz, 1 H) 10.68 (br s, 1 H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.5, 162.0, 142.1, 140.6, 135.4, 131.2, 130.3, 129.5 (2xC), 125.9, 116.2, 110.4, 107.2, 54.5, 32.4, 22.8

3-(cyclopentylsulfamoyl)-*N*-(3,4-difluorophenyl)benzamide (30).

47 % yield from 3-(*N*-cyclopentylsulfamoyl)benzoic acid according to the representative procedure of scheme 1a as described for compound **32**. Method B; Rt: 4.34 min. m/z: 381.2 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 381.1085 (calcd for $C_{18}H_{18}F_2N_2O_3S$: 380.1006). ^1H NMR (400 MHz, DMSO-d_6) δ ppm 1.20 - 1.44 (m, 4 H), 1.44 - 1.68 (m, 4 H), 3.44 (sxt, *J*=6.8 Hz, 1 H), 7.45 (dt, *J*=10.6, 9.0 Hz, 1 H), 7.51 - 7.60 (m, 1 H), 7.77 (t, *J*=7.8 Hz, 1 H), 7.80 (d, *J*=7.2 Hz, 1 H), 7.93 (ddd, *J*=13.2, 7.5, 2.5 Hz, 1 H), 8.02 (d, *J*=7.8 Hz, 1 H), 8.19 (d, *J*=7.7 Hz, 1 H), 8.35 (t, *J*=1.7 Hz, 1 H), 10.70 (s, 1 H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.4, 148.8, 145.7, 142.1, 135.8, 135.2, 131.1, 129.5 (2xC), 125.8, 117.3, 116.8, 109.5, 54.5, 32.4, 22.8

3-(cyclopentylsulfamoyl)-*N*-(3,5-difluorophenyl)benzamide (31).

9 % yield from 3-(*N*-cyclopentylsulfamoyl)benzoic acid according to the representative procedure of scheme 1a as described for compound **32**. Method B; Rt: 4.43 min. m/z: 381.2 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 381.1087 (calcd for $C_{18}H_{18}F_2N_2O_3S$: 380.1006). ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.86 (br s, 1H), 8.37-8.40 (m, 1H), 8.19-8.24 (m, *J*=7.70 Hz, 1H), 8.00-8.06 (m, *J*=8.50 Hz, 1H), 7.71-7.88 (m, 2H), 7.54-7.62 (m, 2H), 6.99 (tt, *J*=2.42, 9.28 Hz, 1H), 3.38-3.50 (m, 1H), 1.48-1.65 (m, 4H), 1.23-1.43 (m, 4H); ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.7, 162.3, 142.2, 141.4, 135.0, 131.3, 129.7, 129.5, 125.9, 103.2, 99.0, 54.5, 32.4, 22.8

3-(cyclopentylsulfamoyl)-*N*-(4-fluoro-3-methyl-phenyl)benzamide (32)

Representative procedure route scheme 1a: To an iced-cooled mixture of cyclopentanamine (1.93 g, 22.66 mmol) and a solution of NaOH (1.81 g, 45.32 mmol) in H_2O (25 mL) and THF (25 mL) was added 3-(chloro-sulfonyl)benzoic acid (5.0 g, 22.66 mmol) in portions. The reaction mixture was stirred at 20°C for 2 hours. The resulting mixture was diluted with H_2O (20 mL) and extracted with ethyl acetate (30 mL). The aqueous layer was separated and adjusted pH =2 by 4 N HCl and extracted with dichloromethane (3 x 30 mL). The combined organic layers were washed with brine (15 mL), dried over anhydrous Na_2SO_4 and concentrated under reduced pressure to afford 3-(*N*-cyclopentylsulfamoyl)benzoic acid (**71d**, 4.5 g, 73 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.30 - 8.34 (m, 1H), 8.10 - 8.20 (m, 1H), 8.00 - 8.05 (m, 1H), 7.80 (d, *J* = 7.2 Hz, 1H), 7.65 - 7.75 (m, 1H), 1.40 - 1.60 (m, 4H), 1.30 - 1.38 (m, 2H), 1.20 - 1.28 (m, 2H), one CH signal under residual water peak. To an ice cooled mixture of 3-(*N*-cyclopentylsulfamoyl)benzoic acid (250 mg, 0.928 mmol), 4-fluoro-3-methylaniline (116.2 mg, 0.928 mmol) and HATU (388.2 mg, 1.021 mmol) in CH_2Cl_2 (15 mL) DIPEA (359.8 mg, 2.784 mmol) was added under a N_2 atmosphere. The resulting mixture was stirred at 20°C for 16 hours. The solvent was removed in vacuo. The mixture was washed with saturated aqueous critic acid

(10 mL), brine and dried over Na_2SO_4. The solvent was removed in vacuo. The residue was purified by column chromatography over silica gel (gradient eluent: petroleum ether/ethyl acetate from 100/0 to 10/90). The pure fractions were collected and the solvent was removed in vacuo. The residue was further purified by preparative high performance liquid chromatography over RP-18 (eluent: CH_3CN in H_2O from 45% to 75%, v/v; 0.01% HCl as addition). The pure fractions were collected and the volatiles were removed in vacuo. The aqueous layer was adjusted to pH=7 with Amberlite IRA-900 ion exchange resin (OH form), filtrated and lyophilized to dryness to afford compound **32** (170.0 mg, 49 %). Method B; Rt: 4.31 min. m/z: 377.2 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 377.1338 (calcd for $C_{19}H_{21}FN_2O_3S$: 376.1257); ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 10.47 (1 H, br. s), 8.33-8.35 (1 H, m), 8.17 (1 H, dm, *J*=8.0), 7.98 (1 H, dm, *J*=8.0), 7.78 (1 H, d, *J*=7.0 Hz), 7.74 (1 H, t, *J*=8.0 Hz), 7.62 - 7.68 (1 H, m), 7.53 - 7.61 (1 H, m), 7.13 (1 H, t, *J*=9.0 Hz), 3.37 - 3.48 (1 H, m), 2.23 (3 H, d, *J*=1.8 Hz), 1.44 - 1.69 (4 H, m), 1.12 - 1.45 (4 H, m). ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.1, 157.1, 142.1, 135.6, 134.8, 131.1, 129.4, 129.3, 125.8, 124.1, 123.7, 119.9, 114.8, 54.5, 32.4, 22.8, 14.4

3-(cyclopentylsulfamoyl)-*N*-(3-fluoro-5-methyl-phenyl)benzamide (33).

49 % yield from 3-(*N*-cyclopentylsulfamoyl)benzoic acid according to the representative procedure of scheme 1a as described for compound **32**. Method B; Rt: 4.41 min. m/z: 377.2 $(M+H)^+$ Exact mass: 376.1. ESI-HRMS (TOF) m/z: $[M + H]^+$ 377.1340 (calcd for $C_{19}H_{21}FN_2O_3S$: 376.1257); ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 10.60 (1 H, bs), 8.36 (1 H, t, *J*=1.5 Hz), 8.19 (1 H, dm, *J*=7.5 Hz), 8.02 (1 H, dm, *J*=7.5 Hz), 7.81 (1 H, d, *J*=7.5 Hz), 7.78 (1 H, t, *J*=7.5 Hz), 7.55 (1 H, dm, *J*=11.0 Hz), 7.38 - 7.46 (1 H, m), 6.82 (1 H, dm, *J*=9.5 Hz), 3.41 - 3.54 (1 H, m), 2.34 (3 H, s), 1.45 - 1.70 (4 H, m), 1.19 - 1.45 (4 H, m); ^{13}C NMR (101 MHz,

DMSO-d_6) δ 164.4, 161.9, 142.1, 140.2, 140.2, 135.4, 131.2, 129.5, 129.4, 125.8, 116.6, 111.0, 104.4, 54.5, 32.4, 22.8, 21.1

3-(cyclopentylsulfamoyl)-*N*-(3-fluoro-4-methyl-phenyl)benzamide (34).

63 % yield from 3-(*N*-cyclopentylsulfamoyl)benzoic acid according to the representative procedure of scheme 1a as described for compound **32**. The residue was purified by column chromatography over silica gel (gradient eluent: petroleum ether/ethyl acetate from 100/0 to 40/60).. Method B; Rt: 4.41 min. m/z: 377.2 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 377.1339 (calcd for $C_{19}H_{21}FN_2O_3S$: 376.1257). ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.58 (br s, 1H), 8.35 (t, *J*=1.60 Hz, 1H), 8.19 (td, *J*=1.32, 7.70 Hz, 1H), 8.01 (d, *J*=7.70 Hz, 1H), 7.73-7.82 (m, 2H), 7.68 (dd, *J*=2.03, 12.21 Hz, 1H), 7.46 (dd, *J*=2.03, 8.14 Hz, 1H), 7.26 (t, *J*=8.54 Hz, 1H), 3.45 (br s, 1H), 2.21 (d, *J*=1.63 Hz, 3H), 1.45-1.64 (m, 4H), 1.25-1.43 (m, 4H)^{13}C NMR (101 MHz, DMSO-d_6) δ 164.3, 160.1, 142.1, 138.2, 135.5, 131.3, 131.1, 129.4, 129.4, 125.9, 119.3, 116.0, 107.1, 54.5, 32.4, 22.8, 13.7

***N*-(4-fluorophenyl)-3-(4-methylpiperazin-1-yl)sulfonyl-benzamide (36).**

Synthesis according to the representative procedure of scheme 1b as described for compound **53.** 31 % yield,. Method A; Rt: 4.14 min. m/z: 378.2 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 378.1304 (calcd for $C_{18}H_{20}FN_3O_3S$: 377.1209). ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.57 (s, 1H), 8.28-8.34 (m, 1H), 8.23-8.27 (m, 1H), 7.92-7.98 (m, 1H), 7.75-7.86 (m, 3H), 7.17-7.26 (m, 2H), 2.88-3.06 (m, 4H), 2.36-2.48 (m, 4H), 2.18 (s, 3H); ^{13}C NMR (101 MHz, DMSO-d_6) δ 163.9, 158.5, 135.8, 135.4, 135.0, 132.2, 130.3, 129.7, 126.4, 122.5, 115.2, 53.3, 45.5, 45.0

3-(4-acetylpiperazin-1-yl)sulfonyl-*N*-(4-fluorophenyl)benzamide (37).

Synthesis according to the representative procedure of scheme 1b as described for compound **53**. 13 % yield. Method A; Rt: 4.82 min. m/z: 406.2 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 406.1239 (calcd for $C1_9H_{20}FN_3O_4S$: 405.1159). ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.56 (s, 1H), 8.28-8.33 (m, 1H), 8.25-8.28 (m, 1H), 7.92-7.98 (m, 1H), 7.74-7.86 (m, 3H), 7.17-7.28 (m, 2H), 3.47-3.57 (m, 4H), 2.94-3.01 (m, 2H), 2.85-2.94 (m, 2H), 1.93 (s, 3H); ^{13}C NMR (101 MHz, DMSO-d_6) δ 168.3, 163.8, 158.5, 135.8, 135.3, 135.0, 132.3, 130.3, 129.8, 126.4, 122.5, 115.2, 46.0, 45.7, 44.9, 21.0

***N*-(4-fluorophenyl)-3-morpholinosulfonyl-benzamide (38).**

Synthesis according to the representative procedure of scheme 1b as described for compound **53**. 42 % yield. Method A; Rt: 5.07 min. m/z: 365.1 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 365.0972 (calcd for $C_{17}H_{17}FN_2O_4S$: 364.0893). ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.57 (s, 1H), 8.28-8.34 (m, 1H), 8.23-8.28 (m, 1H), 7.93-7.97 (m, 1H), 7.84 (t, *J*=7.73 Hz, 1H), 7.76-7.81 (m, 2H), 7.17-7.27 (m, 2H), 3.60-3.69 (m, 4H), 2.86-2.98 (m, 4H); ^{13}C NMR (101 MHz, DMSO-d_6) δ 163.9, 158.5, 135.9, 135.1, 134.9, 132.4, 130.4, 129.8, 126.6, 122.5, 115.2, 65.2, 45.9

_N_-(4-fluorophenyl)-3-pyrrolidin-1-ylsulfonyl-benzamide (39).

Synthesis according to the representative procedure of scheme 1b as described for compound **53**. 35 % yield. Method A; Rt: 5.42 min. m/z: 349.1 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 349.1025 (calcd for $C_{17}H_{17}FN_2O_3S$: 348.0944). ^1H NMR (400 MHz, DMSO-d_6) δ 10.56 (s, 1H), 8.30-8.37 (m, 1H), 8.25-8.30 (m, 1H), 7.99-8.05 (m, 1H), 7.72-7.85 (m, 3H), 7.17-7.26 (m, *J*=8.90, 8.90 Hz, 2H), 3.19 (s, 4H), 1.60-1.75 (m, 4H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 163.9, 158.5, 136.7, 135.6, 135.1, 132.0, 130.1, 129.8, 126.2, 122.6, 115.2, 47.9, 24.7

3-(dimethylsulfamoyl)-_N_-(4-fluorophenyl)benzamide (40).

Synthesis according to the representative procedure of scheme 1b as described for compound **53**. 50 % yield. Method A; Rt: 5.09 min. m/z: 323.1 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 323.0870 (calcd for $C_{15}H_{15}FN_2O_3S$: 322.0787). ^1H NMR (400 MHz, DMSO-d_6) δ 10.56 (s, 1H), 8.25-8.31 (m, 2H), 7.92-7.98 (m, 1H), 7.75-7.85 (m, 3H), 7.18-7.25 (m, 2H), 2.66 (s, 6H); ^{13}C NMR (101 MHz, DMSO-d_6) δ 163.9, 158.5, 135.7, 135.3, 135.0, 132.0, 130.3, 129.7, 126.4, 122.6, 115.2, 37.5

_N_-(4-fluorophenyl)-3-(isopropylsulfamoyl)benzamide (41).

49 % yield from 3-(*N*-isopropylsulfamoyl)benzoic acid (**71b**), prepared similarly as described for compound **61**. Method A; Rt: 5.01 min. m/z: 337.2 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 337.1025 (calcd for $C_{16}H_{17}FN_2O_3S$: 336.0944); ^1H NMR (400 MHz, DMSO-d_6) δ 10.54 (br s, 1H), 8.36 (t, *J*=2.03 Hz, 1H), 8.17-8.21 (m, 1H), 7.98-8.03 (m, 1H), 7.76 (s, 4H), 7.21 (s, 2H), 3.23-3.32 (m, 1H), 0.96 (d, *J*=6.51 Hz, 6H); ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.1, 158.4, 142.3, 135.5, 135.1, 131.1, 129.4, 129.1, 125.7, 122.4, 115.2, 45.3, 23.2

3-(1-ethylpropylsulfamoyl)-*N*-(4-fluorophenyl)benzamide (42).

22 % yield over two steps from 3-(chlorosulfonyl)benzoic acid similarly as described for compound **50**. Method B; Rt: 4.24 min. m/z: 365.2 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 365.1342 (calcd for $C_{18}H_{21}FN_2O_3S$: 364.1257). ^{13}C NMR (151 MHz, DMSO-d_6) δ 164.6, 158.9, 143.3, 135.8, 135.6, 131.4, 129.8, 129.6, 126.1, 122.9, 115.7, 56.8, 27.3, 10.3; ^1H NMR (600 MHz, DMSO-d_6) δ 10.55 (s, 1H), 8.36 (t, *J*=1.80 Hz, 1H), 8.18 (ddd, *J*=1.00, 1.70, 7.80 Hz, 1H), 8.00 (ddd, *J*=1.00, 1.70, 7.80 Hz, 1H), 7.76-7.83 (m, 2H), 7.74 (t, *J*=7.80 Hz, 1H), 7.65 (d, *J*=7.76 Hz, 1H), 7.19-7.24 (m, 2H), 2.95-3.03 (m, 1H), 1.30-1.39 (m, 2H), 1.19-1.27 (m, 2H), 0.65 (t, *J*=7.43 Hz, 6H)

***N*-(4-fluorophenyl)-3-[[(1*S*)-1-methylpropyl]sulfamoyl]benzamide (43).**

Enantiomer of compound **44**. Prepared similar as described for compound **44** starting from (*S*)-butan-2-amine instead of (*R*)-butan-2-amine.Method B; Rt: 4.03 min. m/z: 351.2 $(M+H)^+$ Exact mass: 350.1 ($[\alpha]_D^{20}$ = + (*c* = 0.2, MeOH). $[\alpha]_D^{20}$ = + 9.49 (c 0.611 w/v %, DMF), Column: Chiralpak AD-3 150×4.6mm I.D., 3um; Mobile phase: methanol (0.05% diethylamine) in CO_2 from 5% to 40%; Flow rate: 2.5 mL/min; Rt: 7.73 min. $[\alpha]_{589}^{20}$ +9.49 ° (c 0.61 w/v %, MeOH). ESI-HRMS (TOF) m/z: $[M + H]^+$ 351.1184 (calcd for $C_{17}H_{19}FN_2O_3S$: 350.1100).

***N*-(4-fluorophenyl)-3-[[(1*R*)-1-methylpropyl]sulfamoyl]benzamide (44).**

To an iced-cooled mixture of (*R*)-butan-2-amine (0.500 g, 6.837 mmol) and NaOH (0.547 g, 13.67 mmol) in H_2O (15 mL) and THF (15 mL), 3-(chlorosulfonyl)benzoic acid was added (1.508 g, 6.84 mmol) in portions. The reaction mixture was stirred at 20°C for 2 hours. The resulting mixture was diluted with H_2O (15 mL) and extracted with ethyl acetate (15 mL). The aqueous layer was separated and pH was adjusted to 3 by 1 N HCl and extracted with ethyl

acetate (3 x 10 mL). The combined organic layers were washed with brine (10 mL), dried over anhydrous Na_2SO_4 and concentrated under reduced pressure resulting in (*R*)-3-(*N*-sec-butylsulfamoyl)benzoic acid (0.73 g, 35 %). To an ice cooled mixture of (*R*)-3-(*N*-sec-butylsulfamoyl)benzoic acid (730 mg), 4-fluoroaniline (347 mg, 3.121mmol), HATU (1.294 g, 3.404 mmol) in DMF (10 mL) DIPEA (1.48 mL, 8.51 mmol) was added under N_2 atmosphere. The resulting mixture was stirred at 20°C for 2 hours. The solvent was removed in vacuo. The mixture was washed with saturated aqueous critic acid (10 mL), brine and dried over Na_2SO_4. The solvent was removed in vacuo. The residue was purified by column chromatography over silica gel (gradient eluent: petroleum ether/ethyl acetate from 100/0 to 55/45). The pure fractions were collected and the solvent was removed in vacuo. The residue was purified by SFC separation (Chiralcel OJ, 20 μm; Supercritical CO_2: MeOH (0.2% diethylamine)). The pure fractions were collected and the solvent was removed in vacuo, resulting in compound **44** (300 mg, 13% over two steps). Method A; Rt: 5.25 min. m/z: 351.2 $(M+H)^+$ Exact mass: 350.1**.** ESI-HRMS (TOF) m/z: $[M + H]^+$ 351.1183 (calcd for: $C_{17}H_{19}FN_2O_3S$: 350.1100) $[\alpha]_D^{20}$ = -9.9 (c 0.435 w/v %, DMF); Column: Chiralpak AD-3 150×4.6mm I.D., 3um; Mobile phase: methanol (0.05% diethylamine) in CO_2 from 5% to 40%; Flow rate: 2.5 mL/min; Rt: 7.58 min; ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 0.70 (t, *J*=7.4 Hz, 3 H), 0.88 (d, *J*=6.5 Hz, 3 H), 1.30 (quin, *J*=7.2 Hz, 2 H), 3.01 - 3.18 (m, 1 H), 7.21 (t, *J*=8.8 Hz, 2 H), 7.67 (br. d, *J*=5.5 Hz, 1 H), 7.75 (t, *J*=7.8 Hz, 1 H), 7.78 (dd, *J*=8.8, 5.1 Hz, 2 H), 8.00 (d, *J*=7.8 Hz, 1 H), 8.19 (d, *J*=7.8 Hz, 1 H), 8.36 (s, 1 H), 10.55 (s, 1 H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.1, 158.5, 142.5, 135.4, 135.1, 131.0, 129.4, 129.2, 125.7, 122.4, 115.2, 50.8, 29.5, 20.6, 10.0

3-(tert-butylsulfamoyl)-*N*-(4-fluorophenyl)benzamide (45).

Synthesis according to the representative procedure of scheme 1b as described for compound **53**. 10 % yield. Method B; Rt: 4.24 min. m/z: 351.1($M+H)^+$ ESI-HRMS (TOF) m/z: [M + H]$^+$ 351.1181 (calcd for $C_{17}H_{19}FN_2O_3S$: 350.1100). ^{1}H NMR (600 MHz, DMSO-d_6) δ 10.55 (s, 1H), 8.38 (t, *J*=1.70 Hz, 1H), 8.17 (ddd, *J*=1.00, 1.70, 7.80 Hz, 1H), 8.03 (ddd, *J*=1.00, 1.70, 7.80 Hz, 1H), 7.77-7.81 (m, 2H), 7.74 (t, *J*=7.80 Hz, 1H), 7.70 (br s, 1H), 7.18-7.25 (m, 2H), 1.10 (s, 9H); ^{13}C NMR (151 MHz, DMSO-d_6) δ 164.2, 158.5, 144.8, 135.5, 135.2, 130.8, 129.3, 129.2, 125.6, 122.4, 115.3, 53.5, 29.8

3-(cyclobutylsulfamoyl)-*N*-(4-fluorophenyl)benzamide (46).

Synthesis according to the representative procedure of scheme 1b as described for compound **53**. 43 % yield. Method A; Rt: 5.32 min. m/z: 349.1 ($M+H)^+$ ESI-HRMS (TOF) m/z: [M + H]$^+$ 349.1025 (calcd for: $C_{17}H_{17}FN_2O_3S$: 348.0944). ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.54 (s, 1H), 8.34 (t, *J*=1.70 Hz, 1H), 8.20 (ddd, *J*=1.00, 1.70, 7.80 Hz, 1H), 8.09 (br d, *J*=8.00 Hz, 1H), 7.98 (ddd, *J*=1.00, 1.70, 7.80 Hz, 1H), 7.69-7.83 (m, 3H), 7.14-7.27 (m, 2H), 3.67 (sxt, *J*=8.22 Hz, 1H), 1.83-1.97 (m, 2H), 1.66-1.80 (m, 2H), 1.38-1.55 (m, 2H); ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.1, 158.5, 142.1, 135.5, 135.2, 131.2, 129.4, 129.2, 125.8, 122.4, 115.2, 47.5, 30.5, 14.5

_N_-(4-fluorophenyl)-3-(oxetan-3-ylsulfamoyl)benzamide (47).

Synthesis according to the representative procedure of scheme 1b as described for compound **53**. 29 % yield. Method A; Rt: 4.64 min. m/z: 351.1 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 351.0818 (calcd for $C_{16}H_{15}FN_2O_4S$: 350.0737) ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.56 (s, 1H), 8.70 (br s, 1H), 8.33 (t, *J*=1.60 Hz, 1H), 8.22 (ddd, *J*=1.00, 1.70, 7.90 Hz, 1H), 7.98 (ddd, *J*=1.00, 1.70, 7.90 Hz, 1H), 7.74-7.82 (m, 3H), 7.19-7.25 (m, 2H), 4.38-4.55 (m, 3H), 4.24-4.30 (m, 2H); ^{13}C NMR (101 MHz, DMSO-d_6) δ ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.0, 158.5, 141.2, 135.7, 135.1, 131.6, 129.7, 129.1, 125.6, 122.5, 115.3, 77.0, 47.0

_N_-(4-fluorophenyl)-3-[(3-methyloxetan-3-yl)sulfamoyl]benzamide (48).

Synthesis according to the representative procedure of scheme 1b as described for compound **53**. 39 % yield. Method A; Rt: 4.87 min. m/z: 365.1 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 365.0972 (calcd for: $C_{17}H_{17}FN_2O_4S$: 364.0893). ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.56 (s, 1H), 8.50 (br s, 1H), 8.36-8.40 (m, 1H), 8.19-8.25 (m, *J*=7.70 Hz, 1H), 8.03 (d, *J*=7.80 Hz, 1H), 7.74-7.83 (m, 3H), 7.22 (t, *J*=8.38 Hz, 2H), 4.56 (d, *J*=6.06 Hz, 2H), 4.14 (d, *J*=6.46 Hz, 2H), 1.42 (s, 3H); ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.0, 158.5, 143.5, 135.7, 135.1, 131.3, 129.6, 128.9, 125.4, 122.4, 115.2, 81.2, 54.7, 24.2

_N_-(4-fluorophenyl)-3-[[(3*R*)-tetrahydrofuran-3-yl]sulfamoyl]benzamide (49).

To a solution of (*R*)-tetrahydrofuran-3-amine (0.87 g, 9.97 mmol) in THF (20 mL) aqueous sodium hydroxide was added (4 mL, 5 N) in ice bath followed by 3-(chlorosulfonyl)benzoic acid (2.2 g, 9.97 mmol). After stirring at 25°C for 3 hours, the reaction mixture was diluted with H_2O (20 mL) and extracted with EtOAc (20 mL). The aqueous layer was adjusted to pH=3 by aq. HCl

(2 N) and then the resulting mixture was extracted with EtOAc (3 x 20 mL). The combined organic layers were washed with brine, dried over anhydrous $MgSO_4$ and concentrated in vacuo resulting in compound (*R*)-3-(*N*-(tetrahydrofuran-3-yl)sulfamoyl)benzoic acid (900 mg, 33%). To a solution of compound (*R*)-3-(*N*-(tetrahydrofuran-3-yl)sulfamoyl)benzoic acid (0.80 g, 2.95 mmol), 4-fluoroaniline (0.39g, 3.54 mmol), and HATU (3.36 g, 8.85 mmol) in CH_2Cl_2 (10 mL) cooled in an ice bath under N_2 atmosphere,DIPEA (0.57g, 0.44 mmol) was added. The resulting mixture was diluted with CH_2Cl_2 (15 mL) and washed with saturated aqueous $NaHCO_3$ (15 mL) and brine (10 mL). After drying over anhydrous $MgSO_4$ the solvent was removed in vacuo. The obtained residue was purified by preparative high performance liquid chromatography over RP-18 (eluent: CH_3CN in H_2O: from 40% to 80%, v/v; 0.05% TFA as addition). The pure fractions were collected and the volatiles were removed in vacuo. The aqueous layer was adjusted to PH=7 with Amberlite IRA-900 ion exchange resin (OH form), filtrated and lyophilized. The obtained residue was further purified by prep. SFC (Column:Chiralpak AD-3 150×4.6mm I.D., 3um Mobile phase: 40% of methanol (0.05% diethylamine) in CO_2. Flow rate: 2.5 mL/min) resulting in compound 49 (370 mg, 34 %) Method A; Rt: 4.6 min. m/z: 365.2 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 365.0970 (calcd for $C_{17}H_{17}FN_2O_4S$: 364.0893). $[\alpha]_D^{20} = -13.60$ (*c*=0.11, MeOH) ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 10.57 (1 H, br. s), 8.34 - 8.40 (1 H, m), 8.18 - 8.27 (1 H, m), 8.09 (1 H, br. s), 7.99 - 8.06 (1 H, m), 7.74 - 7.84 (3 H, m), 7.13 - 7.33 (2 H, m), 3.64 - 3.83 (2 H, m), 3.50 - 3.64 (2 H, m), 3.35 - 3.39 (1 H, m), 1.80 - 1.99 (1 H, m), 1.51 - 1.68 (1 H, m). ^{13}C NMR (151 MHz, DMSO-d_6) δ 164.1, 158.5, 141.4, 135.7, 135.2, 131.5, 129.7, 129.4, 125.9, 122.5, 115.3, 72.0, 66.1, 53.2, 32.1

_N_-(4-fluorophenyl)-3-[[(3S)-tetrahydrofuran-3-yl]sulfamoyl]benzamide (50).

To an iced-cooled mixture of (*S*)-tetrahydrofuran-3-amine hydrochloride (0.500 g, 4.41 mmol) and NaOH (0.485 g, 12.138 mmol) in H_2O (5 mL) and THF (5 mL) 3-(chlorosulfonyl)benzoic acid (0.893 g, 4.406 mmol) was added in several portions. Then, the reaction mixture was stirred at 20°C for 2 hours. The resulting mixture was diluted with H_2O (10 mL) and extracted with ethyl acetate (10 mL). The pH value of aqueous layer was adjusted to 3 by adding 1N HCl and then the mixture was extracted with ethyl acetate (3 x 10 mL). The combined organic layer was washed by brine (10 mL), dried over anhydrous Na_2SO_4 and concentrated under reduced pressure resulting in (*S*)-3-(*N*-(tetrahydrofuran-3-yl)sulfamoyl)benzoic acid (**71a**, 0.60 g, 45 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.36 (s, 1H), 8.19 (d, J = 8.0 Hz, 1H), 8.12 (d, J = 6.8 Hz, 1H), 8.06 (d, J = 7.6 Hz, 1H), 7.70 - 7.80 (m, 1H), 3.65 - 3.75 (m, 2H), 3.50 - 3.64 (m, 3H), 1.80 - 1.95 (m, 1H), 1.50 – 1.65 (m, 1H). To an ice cooled mixture of (*S*)-3-(*N*-(tetrahydrofuran-3-yl)sulfamoyl)benzoic acid (600 mg, 2.212 mmol), 4-fluoroaniline (270 mg, 2.433mmol) and HATU (1.01 g, 2.654 mmol) in DMF (5 mL) DIPEA (1.15 mL, 6.636 mmol) was added under N_2 atmosphere. The resulting mixture was stirred at 20°C for 2 hours. The solvent was removed in vacuo. The mixture was washed with saturated aqueous critic acid (10 mL), brine and dried over Na_2SO_4. The solvent was removed in vacuo. The residue was purified by column chromatography over silica gel (gradient eluent: petroleum ether/ethyl acetate from 100/0 to 10/90). The pure fractions were collected and the solvent was removed in vacuo. The residue was further purified by preparative high performance liquid chromatography over RP-18 (eluent: CH_3CN in H_2O from 40% to 80%, v/v; 0.06% NH_4HCO_3 as addition). The pure fractions were collected and the volatiles were removed in vacuo. The aqueous layer was lyophilized to dryness resulting in compound **50** (0.48 g, 59 %) Method A; Rt: 4.6 min. m/z: 365.2 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ (calcd for 365.0972 $C_{17}H_{17}FN_2O_4S$: 364.0893). ;$[\alpha]_D^{20}$ = +15.56 (*c* 0.10,

MeOH); ^{1}H NMR (400 MHz, 80°C, DMSO-d_6) δ ppm 10.35 (1 H, br. s), 8.32 - 8.48 (1 H, m), 8.15 - 8.32 (1 H, m), 8.03 (1 H, br. s), 7.83 - 7.94 (1 H, m), 7.68 - 7.83 (3 H, m), 7.06 - 7.31 (2 H, m), 3.70 - 3.87 (2 H, m), 3.51 - 3.70 (2 H, m), 3.32 - 3.48 (1 H, m), 1.85 - 2.04 (1 H, m), 1.59 - 1.78 (1 H, m) ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.1, 158.5, 141.4, 135.7, 135.1, 131.4, 129.6, 129.4, 125.9, 122.5, 115.3, 72.0, 66.1, 53.1, 32.1

3-[(1-acetyl-4-piperidyl)sulfamoyl]-*N*-(4-fluorophenyl)benzamide (51).

Synthesis according to the representative procedure of scheme 1b as described for compound **53**. 10 % yield. Method A; Rt: 4.75 min. m/z: 420.2 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 420.1402(calcd for $C_{20}H_{22}FN_3O_4S$: 419.1315). ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.55 (s, 1H), 8.36-8.42 (m, 1H), 8.18-8.25 (m, 1H), 8.01-8.07 (m, 1H), 7.95 (br s, 1H), 7.72-7.85 (m, 3H), 7.16-7.28 (m, 2H), 3.98-4.12 (m, 1H), 3.58-3.70 (m, 1H), 3.21-3.30 (m, 1H), 2.95-3.07 (m, 1H), 2.60-2.74 (m, 1H), 1.93 (s, 3H), 1.50-1.64 (m, 2H), 1.11-1.36 (m, 3H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 167.9, 164.1, 158.5, 142.4, 135.6, 135.1, 131.2, 129.5, 129.1, 125.6, 122.4, 115.2, 50.0, 44.0, 32.8, 32.0, 21.1

***N*-(4-fluorophenyl)-3-[(1-methyl-4-piperidyl)sulfamoyl]benzamide (52).**

Synthesis according to the representative procedure of scheme 1b as described for compound **53**. 18 % yield. Method A; Rt: 4.05 min. m/z: 392.2 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 392.1440 (calcd for $C_{19}H_{22}FN_3O_3S$: 391.1366). ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.55 (s, 1H), 8.38 (s, 1H), 8.15-8.24 (m, 1H), 7.98-8.06 (m, 1H), 7.67-7.89 (m, 4H), 7.11-7.29 (m, 2H), 2.87-3.05 (m, 1H), 2.54-2.69 (m, 2H), 2.10 (s, 3H), 1.77-1.96 (m, 2H), 1.46-1.60 (m, 2H), 1.29-1.46 (m, 2H) ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.1, 158.5, 142.5, 135.6, 135.1, 131.1, 129.5, 129.1, 125.6, 122.4, 115.2, 53.6, 50.0, 45.5, 32.1

_N_-(4-fluorophenyl)-3-(tetrahydropyran-4-ylsulfamoyl)benzamide (53).

Representative procedure route scheme 1b: 3-(chlorosulfonyl)benzoyl chloride (207 mg, 1 mmol) was dissolved in dichloromethane (3 mL) and 4-fluoroaniline (111 mg, 1.0 mmol) and triethylamine (112 mg, 1.0 mmol) in dichloromethane (2 mL) were added to the mixture at 0°C. The mixture was next stirred at 20°C for 1 hour. To this reaction mixture containing 3-(4-fluoro-phenylcarbamoyl)benzene-1-sulfonyl chloride at 0°C, a solution of triethylamine (121 mg, 1.2 mmol) and 4-aminotetrahydropyran (88 mg, 0.861 mmol) in dichloromethane (3 mL) was added. The mixture was stirred at 20°C for 1 hour. The solvent was removed in vacuo. The residue was purified by high performance liquid chromatography (Column: Phenomenex Synergi C18 150*20mm*5um. A: H_2O+0.1%TFA; B: MeCN). The product fractions were collected and the organic solvent was evaporated. The fraction was neutralized by saturated $NaHCO_3$. The mixture was extracted with dichloromethane. The organic layer was dried over Na_2SO_4 and concentrated resulting in compound **53** (85.4 mg, 23 % yield) Method A; Rt: 4.88 min. m/z: 379.2 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 379.1125 (calcd for $C_{18}H_{19}FN_2O_4S$: 378.1050). ^1H NMR (400 MHz, CHLOROFORM-d) δ 8.38 (t, *J*=1.51 Hz, 1H), 8.11-8.17 (m, *J*=7.80 Hz, 1H), 8.06 (d, *J*=8.00 Hz, 1H), 7.99-8.04 (m, 1H), 7.68 (t, *J*=7.78 Hz, 1H), 7.58-7.65 (m, 2H), 7.11 (d, *J*=8.53 Hz, 2H), 4.82 (d, *J*=7.28 Hz, 1H), 3.83-3.93 (m, 2H), 3.30-3.47 (m, 3H), 1.70-1.84 (m, 2H), 1.41-1.55 (m, 2H)

_N_-(4-fluoro-3-methyl-phenyl)-3-[[(3_S_)-tetrahydrofuran-3-yl]sulfamoyl]benzamide (54).

3-(chlorosulfonyl)benzoyl chloride (1200 mg, 5.0 mmol) was dissolved in dichloromethane (15 mL). A solution of 4-fluoro-3-methylaniline (625 mg, 5.0 mmol) and triethylamine (606 mg, 6.0 mmol) in dichloromethane (15 mL) was added to the mixture at 0°C. The mixture was stirred at

25°C for 1 hour. The reaction mixture was used to the next step without further purification. To the above reaction mixture a solution of triethylamine (606 mg, 6.0 mmol) and (*S*)-tetrahydrofuran-3-amine (460.0 mg, 5.3 mmol) in dichloromethane (15 mL) was added at 0°C. The mixture was stirred at 25°C for 1 hour. The solvent was removed in vacuo. The residue was purified by reversed phase high performance liquid chromatography (eluent: CH_3CN in water (0.1% TFA) from 25 to 55, v/v). The pure fractions were collected and the organic solvent was evaporated. The aqueous layer was neutralized with saturated aqueous $NaHCO_3$ to pH=7-8. The mixture was extracted with dichloromethane (3 x 15 mL). The combined organic layers were dried over Na_2SO_4 and concentrated in vacuo resulting in compound **54** (620 mg, 33 %). Method A; Rt: 4.88 min. m/z: 379.2 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 379.1130 (calcd for $C_{18}H_{19}FN_2O_4S$: 378.1050). ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 1.56 - 1.65 (m, 1 H), 1.85 - 1.94 (m, 1 H), 2.22 - 2.28 (m, 3 H), 3.33 - 3.39 (m, 1 H), 3.52 - 3.65 (m, 2 H), 3.65 - 3.73 (m, 1 H), 3.73 - 3.79 (m, 1 H), 7.14 (t, *J*=9.2 Hz, 1 H), 7.56 - 7.62 (m, 1 H), 7.67 (dd, *J*=7.0, 2.3 Hz, 1 H), 7.78 (t, *J*=7.8 Hz, 1 H), 8.02 (d, *J*=7.8 Hz, 1 H), 8.10 (d, *J*=4.5 Hz, 1 H), 8.21 (d, *J*=7.8 Hz, 1 H), 8.37 (s, 1 H), 10.49 (s, 1 H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.0, 157.1, 141.4, 135.7, 134.7, 131.3, 129.5, 129.3, 125.8, 124.1, 123.7, 119.9, 114.8, 71.9, 66.1, 53.1, 32.1, 14.3. Alternative synthesis of compound **54**: A mixture of 3-(chlorosulfonyl)benzoyl chloride (4.61 g ,19.28mmol) in toluene (45 mL) was refluxed under a gentle flow of nitrogen. 4-fluoro-3-methylaniline (2.19 g, 17.53 mmol) in toluene (15 mL) was added drop wise to the refluxing solution. After addition, the mixture was refluxed for another 30 minutes. The mixture was next cooled to room temperature, and a mixture of (S)-3-aminotetrahydrofuran tosylate (5 g, 19.28 mmol) and diisopropylethylamine (15 mL) in toluene (15 mL) and CH_2Cl_2 (10 mL) was added drop wise. After addition, the mixture was stirred for 4 hours at room temperature. The

resulting mixture was washed with HCl (2 x 100 mL, 1M aq), water (2 x 100 mL) and $NaHCO_3$ (2 x 100 mL, sat. aq). The organic layer was dried on $MgSO_4$, filtered and concentrated under reduced pressure. The obtained residue was purified using silica gel chromatography (CH_2Cl_2-MeOH 100:0 to 95:5) yielding 3-(4-fluoro-3-methylphenylcarbamoyl)benzene-1-sulfonyl chloride (1.07 g; Method J; Rt: 1.11 min. m/z: 326.0 $(M-H)^-$ exact mass: 327.0) during CH_2Cl_2 elution, followed by compound **54** (2.85 g, 39 %) as a white solid after removal of the solvent (dried in a vacuum oven at 55°C for 20 hours). ($[\alpha]_D^{20}$ = - 5.21 (c 0.67 w/v %, MeOH), Method J; Rt: 0.88 min. m/z: 379.1 $(M+H)^+$ Exact mass: 378.1. The compound was crystallized from CH_2Cl_2: DSC (From 30 to 300 °C at 10°C/min): 149°C. $[\alpha]_D^{20}$ = + 3.21 (c 0.65 w/v %, DMF).

***N*-(2,4-difluoro-3-methyl-phenyl)-3-[[(3*S*)-tetrahydrofuran-3-yl]sulfamoyl]benzamide (55).** 14 % yield, prepared similarly as described for compound **60**. Method A; Rt: 5.17 min. m/z: 397.3 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 397.1031 (calcd for $C_{18}H_{18}F_2N_2O_4S$: 396.0955). ^{1}H NMR (600 MHz, DMSO-d_6) δ 10.40 (br s, 1H), 8.35-8.42 (m, 1H), 8.20-8.27 (m, 1H), 8.05-8.16 (m, 1H), 8.00-8.05 (m, 1H), 7.78 (t, *J*=7.76 Hz, 1H), 7.38-7.47 (m, 1H), 7.06-7.14 (m, 1H), 3.72-3.79 (m, 1H), 3.66-3.72 (m, 1H), 3.60-3.66 (m, 1H), 3.54-3.60 (m, 1H), 3.33-3.39 (m, 1H), 2.20 (s, 3H), 1.85-1.98 (m, 1H), 1.53-1.67 (m, 1H); ^{13}C NMR (151 MHz, DMSO-d_6) δ 164.3, 158.4, 154.7, 141.5, 134.7, 131.5, 129.8, 129.6, 126.0, 125.1, 121.6, 113.1, 110.5, 72.0, 66.1, 53.1, 32.1, 7.2

***N*-(2,4-difluoro-5-methyl-phenyl)-3-[[(3*S*)-tetrahydrofuran-3-yl]sulfamoyl]benzamide (56).** 61 % yield, prepared similarly as described for compound **60**. Method A; Rt: 5.18 min. m/z: 397.2 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 397.1034 (calcd for $C_{18}H_{18}F_2N_2O_4S$: 396.0955). ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 1.54 - 1.69 (m, 1 H) 1.82 - 1.98 (m, 1 H) 2.24 (s, 3 H) 3.35 - 3.40 (m, 1 H) 3.52 - 3.66 (m, 2 H) 3.66 - 3.83 (m, 2 H) 7.32 (t, *J*=10.0 Hz, 1 H) 7.49 (t,

J=8.5 Hz, 1 H) 7.79 (t, *J*=7.8 Hz, 1 H) 8.04 (d, *J*=8.0 Hz, 1 H) 8.07 - 8.18 (m, 1 H) 8.23 (d, *J*=7.8 Hz, 1 H) 8.39 (s, 1 H) 10.40 (br. s, 1 H); ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.2, 158.0, 154.2, 141.5, 134.7, 131.5, 129.7, 129.6, 129.5, 125.9, 121.2, 120.1, 103.9, 72.0, 66.1, 53.1, 32.1, 13.5.

***N*-(3,4-difluoro-2-methyl-phenyl)-3-[[(3*S*)-tetrahydrofuran-3-yl]sulfamoyl]benzamide (57).** To a stirred solution of 3,4-difluoro-2-methyl-aniline (369 mg, 2.6 mmol), 3-[[(3*S*)-tetrahydrofuran-3-yl]sulfamoyl]benzoic acid (700 mg, 2.58 mmol) and *N*,*N*-diisopropylethylamine(1.35 ml, 7.74 mmol) in DMF (10 mL), Pybrop (132705-51-2, 1.82 g, 3.9 mmol) was added at 0° C. The resulting mixture was stirred overnight at 18 °C. The mixture was concentrated in vacuo, ethyl acetate (15 mL) was added and the organic layer was washed with 1N HCl (15 ml) and saturated aqueous $NaHCO_3$ (15 mL). After drying over Na_2SO_4 and concentration in vacuo, the crude residue was purified by reversed phase preparative high-performance liquid chromatography (eluent: CH_3CN in H_2O (0.05% $NH_3.H_2O$) from 37% to 37%, v/v). The pure fractions were collected and the volatiles were removed in vacuo. The aqueous layer was lyophilized to dryness, resulting in compound **57** (238 mg, 23 %). Method D; Rt: 5.01 min. m/z: 396.9 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ *397.1037* (calcd for *$C_{18}H_{18}F_2N_2O_4S$: 396.0955*). ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.37 (br s, 1H), 8.38-8.43 (m, 1H), 8.22-8.28 (m, *J*=7.70 Hz, 1H), 8.10 (br s, 1H), 8.01-8.07 (m, *J*=7.70 Hz, 1H), 7.79 (t, *J*=7.67 Hz, 1H), 7.26-7.35 (m, 1H), 7.16-7.24 (m, 1H), 3.67-3.80 (m, 2H), 3.56-3.67 (m, 2H), 3.35-3.39 (m, 1H), 2.18 (d, *J*=2.02 Hz, 3H), 1.87-1.96 (m, 1H), 1.59-1.67 (m, 1H); ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.4, 148.0, 148.1, 141.5, 135.0, 133.2, 131.4, 129.7, 129.5, 126.0, 124.1, 122.9, 113.9, 72.0, 66.1, 53.1, 32.1, 10.3.

***N*-(3,4-difluorophenyl)-3-[[(3*S*)-tetrahydrofuran-3-yl]sulfamoyl]benzamide (58).**

3-(chlorosulfonyl)benzoyl chloride (1200 mg, 5.0 mmol) was dissolved in dichloromethane (15 mL). A solution of 3,4-difluoroaniline (650 mg, 5.0 mmol) and triethylamine (606 mg, 6.0mmol) in dichloromethane (15 mL) was added to the mixture at 0°C. The mixture was stirred at 25°C for 1 hour. To the obtained reaction mixture, a solution of triethylamine (606 mg, 6.0 mmol) and (S)-tetrahydrofuran-3-amine (460.0 mg, 5.3mmol) in dichloromethane (15 mL) was added at 0°C. The mixture was stirred at 25°C for 1 hour. The solvent was removed in vacuo. The obtained residue was purified by high performance liquid chromatography over RP-18 (eluent: CH_3CN in water (0.1%TFA) from 30 to 60, v/v). The pure fractions were collected and the organic solvent was evaporated. The aqueous layer was neutralized with saturated aqueous $NaHCO_3$ to pH=7-8. The mixture was extracted with dichloromethane (3 x 15 mL). The combined organic layers were dried over Na_2SO_4 and concentrated in vacuo resulting in compound **58** (710 mg, 37 %) Method A; Rt: 4.16 min. m/z: 383.0 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 383.0875 (calcd for $C_{17}H_{16}F_2N_2O_4S$: 382.0799); ^1H NMR (400 MHz, DMSO-d_6) δ ppm 1.54 - 1.63 (m, 1 H), 1.83 - 1.93 (m, 1 H), 3.32 - 3.38 (m, 1 H), 3.52 - 3.63 (m, 2 H), 3.63 - 3.77 (m, 2 H), 7.45 (dt, *J*=10.5, 9.0 Hz, 1 H), 7.51 - 7.57 (m, 1 H), 7.78 (t, *J*=7.8 Hz, 1 H), 7.92 (ddd, *J*=13.3, 7.5, 2.5 Hz, 1 H), 8.02 (d, *J*=7.8Hz, 1 H), 8.09 (d, *J*=6.5 Hz, 1 H), 8.20 (d, *J*=7.8 Hz, 1 H), 8.35 (s, 1 H), 10.70 (s, 1 H); ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.4, 148.8, 145.7, 141.4, 135.8, 135.4, 131.5, 129.7, 129.6, 125.9, 117.4, 116.9, 109.6, 72.0, 66.1, 53.1, 32.1; SFC: Column: Chiralcel OJ-H 250×4.6mm I.D., 5um; Flow: 2.35 mL/min; Mobile phase: methanol (0.05% diethylamine) in CO_2 from 5% to 40%; Rt: 5.61 Min.$[\alpha]_D^{20}$ = + 3.21 (c 0.624 w/v %, DMF)

_N_-(3-cyano-4-fluoro-phenyl)-3-[[(3S)-tetrahydrofuran-3-yl]sulfamoyl]benzamide (59).

25 % yield, prepared similarly as described for compound **60**. Method A; Rt: 5.10 min. m/z: 390.2 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 390.0923 (calcd for $C_{18}H_{16}FN_3O_4S$: 389.0846). ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.44 (br s, 1H), 8.40 (t, *J*=1.70 Hz, 1H), 8.28 (dd, *J*=2.60, 5.90 Hz, 1H), 8.24 (ddd, *J*=1.00, 1.70, 7.90 Hz, 1H), 8.08 (ddd, *J*=2.60, 4.90, 9.19 Hz, 1H), 8.04 (br ddd, *J*=1.00, 1.70, 7.80 Hz, 1H), 7.79 (t, *J*=7.80 Hz, 1H), 7.56 (t, *J*=9.20 Hz, 1H), 3.66-3.81 (m, 2H), 3.53-3.65 (m, 2H), 3.32-3.39 (m, 1H), 1.84-1.96 (m, 1H), 1.56-1.66 (m, 1H); ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.5, 158.5, 141.6, 136.2, 135.2, 131.4, 129.6, 129.6, 128.0, 125.9, 124.6, 116.9, 113.9, 99.8, 72.0, 66.1, 53.1, 32.1.

***N*-(4-fluoro-3-methoxy-phenyl)-3-[[(3*S*)-tetrahydrofuran-3-yl]sulfamoyl]benzamide (60).**

3-[[(3*S*)-tetrahydrofuran-3-yl]sulfamoyl]benzoic acid (400 mg, 1.47 mmol) was dissolved in DMF (0.5 mL) and CH_2Cl_2 (10 mL). $(COCl)_2$ (223 mg, 1.76 mmol) was added at 0°C. The mixture was stirred at 20°C for 2 hours. The solvent was removed in vacuo and the obtained residue was co-evaporated with toluene (2 x 10 mL) resulting in crude 3-[[(3*S*)-tetrahydrofuran-3-yl]sulfamoyl]benzoyl chloride (400 mg).The crude product was used in the next step without purification. 3-[[(3*S*)-tetrahydrofuran-3-yl]sulfamoyl]benzoyl chloride (200 mg) was dissolved in dichloromethane (5 mL). 4-fluoro-3-methoxy-aniline (78 mg, 0.552 mmol) and triethylamine (167 mg, 165 mmol) were added at 0°C. The mixture was stirred at 20°C for 2 hours, washed with H_2O (5 mL) and the water layer extracted with dichloromethane (3 x 10 mL). The combined organic layers were concentrated in vacuo. The obtained residue was purified by reversed phase high performance liquid chromatography (mobile phase: CH_3CN in water (0.1% TFA) from 30% to 60%). The pure fractions were collected and neutralized with solid $NaHCO_3$. The organic solvent was removed in vacuo. The obtained precipitate was filtered, washed with H_2O (5 mL) and dried under high vacuum. The residue was suspended in water (5 mL) lyophilized to dryness

resulting in compound **60** (140 mg, 24 % yield). Method A; Rt: 4.98 min. m/z: 395.2 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 395.1077 (calcd for $C_{18}H_{19}FN_2O_5S$: 394.0999). ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.52 (br s, 1H), 8.34-8.42 (m, 1H), 8.18-8.26 (m, *J*=7.70 Hz, 1H), 8.08 (br s, 1H), 7.99-8.05 (m, *J*=7.70 Hz, 1H), 7.79 (t, *J*=7.87 Hz, 1H), 7.65 (dd, *J*=1.82, 7.87 Hz, 1H), 7.31-7.42 (m, 1H), 7.21 (dd, *J*=8.88, 10.90 Hz, 1H), 3.85 (s, 3H), 3.66-3.78 (m, 2H), 3.55-3.66 (m, 2H), 3.34-3.39 (m, 1H), 1.85-1.96 (m, 1H), 1.57-1.66 (m, 1H); ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.1, 146.8, 147.9, 141.4, 135.7, 135.5, 131.4, 129.6, 129.3, 125.8, 115.6, 112.5, 106.6, 71.9, 66.1, 55.8, 53.1, 32.1

***N*-(4-fluoro-3-methyl-phenyl)-3-(isopropylsulfamoyl)benzamide (61).**

To an iced-cooled solution of 3-(chlorosulfonyl)benzoic acid (50.0 g, 226.6 mmol) in ethylacetate (1000 mL) was added isopropylamine (67.0 g, 1.13 mol) in one portion. The reaction mixture was stirred at 25°C for 3 hours. The resulting mixture was diluted with 1N HCl (500 mL) and extracted with ethyl acetate (2 x 500 mL). The combined organic layers were washed with brine (400 mL), dried over anhydrous Na_2SO_4 and concentrated under reduced pressure resulting 3-(*N*-isopropylsulfamoyl)benzoic acid (**71b**, 46 g, 79 %). ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.33 (s, 1H), 8.14 (d, *J* = 7.6 Hz, 1H), 7.84 (d, *J* = 8.0 Hz, 1H), 7.65 - 7.80 (m, 2H), 3.20 – 3.30 (m, 1H), 0.92 (d, *J* = 6.4 Hz, 6H). To an ice-cooled mixture of 3-(*N*-isopropylsulfamoyl)benzoic acid (7.0 g, 28.77 mmol), 4-fluoro-3-methylaniline (3.6 g, 28.77 mmol) and DIPEA (18.6 g, 143.91 mmol) in CH_2Cl_2 (70 mL) HATU (12.0 g, 31.56 mmol) was added under N_2 atmosphere. The resulting mixture was stirred at 20° for 16 hours. The solvent was removed in vacuo. The mixture was washed with saturated aqueous critic acid (30 mL), brine (20 mL) and dried over Na_2SO_4. The solvent was removed in vacuo. The residue was purified by preparative high performance liquid chromatography on SYNERGI 250*50 10um

(eluent: CH_3CN in H_2O (0.05% TFA) from 35% to 65%, v/v). The pure fractions were collected and adjusted to pH=7 with Amberlite IRA-900(OH) anionic exchange resin. The resin was filtered off. The filtrate was lyophilized to dryness resulting in compound **61** (7.5 g, 74 % yield). Method B; Rt: 3.44 min. m/z: 351.1 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 351.1185 (calcd for $C_{17}H_{19}FN_2O_3S$: 350.1100). ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 10.49 (1 H, br. s), 8.36 (1 H, t, *J*=1.5 Hz), 8.19 (1 H, ddd, *J*=7.8, 1.5, 1.0 Hz), 8.01 (1 H, ddd, *J*=7.8, 1.5, 1.0 Hz), 7.76 (1 H, t, *J*=7.8 Hz), 7.68 (1 H, dd, *J*=7.0, 3.0 Hz), 7.75 (1 H, bs), 7.59 (1 H, ddd, *J*=9.0, 4.5, 3.0 Hz), 7.15 (1 H, t, *J*=9.0 Hz), 3.14 - 3.33 (1 H, m), 2.25 (3 H, d, *J*=1.5 Hz), 0.96 (6 H, d, *J*=6.5 Hz). ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.1, 157.1, 142.4, 135.6, 134.8, 131.0, 129.4, 129.1, 125.7, 124.1, 123.7, 119.9, 114.8, 45.3, 23.2, 14.3

***N*-(3,4-difluorophenyl)-3-(isopropylsulfamoyl)benzamide (63).**

65 % yield from 3-(*N*-isopropylsulfamoyl)benzoic acid (**71b**), similarly as described for compound **61**. Method E; Rt: 5.31 min. m/z: 355.1 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 355.0933 (calcd for $C_{16}H_{16}F_2N_2O_3S$: 354.0850); ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 10.71 (s, 1 H), 8.36 (t, *J*=1.5 Hz, 1 H), 8.19 (d, *J*=7.8 Hz, 1 H), 7.98 - 8.08 (m, 1 H), 7.94 (ddd, *J*=13.2, 7.5, 2.4 Hz, 1 H), 7.71 - 7.83 (m, 2 H), 7.53 - 7.59 (m, 1 H), 7.42 - 7.51 (m, 1 H), 3.21 - 3.29 (m, 1 H), 0.96 (d, *J*=6.5 Hz, 6 H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.4, 148.8, 145.7, 142.4, 135.8, 135.2, 131.1, 129.5, 129.4, 125.7, 117.4, 116.9, 109.5, 45.3, 23.2

***N*-(4-fluoro-3-methyl-phenyl)-3-[(3-methyloxetan-3-yl)sulfamoyl]benzamide (66).**

To 3-(4-fluoro-3-methylphenylcarbamoyl)benzene-1-sulfonyl chloride (500 mg, 1.53 mmol) in toluene (10 mL) at room temperature, a solution of diisopropylethylamine (0.657 mL, 141.6 mmol) and 3-methyl-3-oxetanamine hydrochloride (207 mg, 1.68 mmol) in toluene (5 mL) and dichloromethane (10 mL) was added drop wise. After 2 hours, the reaction mixture was washed

with 1M hydrochloric acid (2 x 10 mL, saturated $NaHCO_3$ (2 x 10 mL) and brine (2 x 10 mL). The organic layer was dried on $MgSO_4$, filtered and concentrated under reduced pressure until only toluene remained. The formed white precipitate was filtered and recrystallised out of diisopropylether and acetonitrile. The crystals were dried in a vacuum oven at 55°C for 20 hours yielding compound **66** (361 mg, 63 %) as a white solid. Method J; Rt: 0.89 min. m/z: 379.0 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 379.1129 (calcd for $C_{18}H_{19}FN_2O_4S$: 378.1050); ^1H NMR (400 MHz, DMSO-d_6) δ ppm 1.41 (s, 3 H), 2.25 (d, *J*=1.5 Hz, 3 H), 4.14 (d, *J*=6.3 Hz, 2 H), 4.56 (d, *J*=6.3 Hz, 2 H), 7.14 (t, *J*=9.0 Hz, 1 H), 7.52 - 7.64 (m, 1 H), 7.68 (dd, *J*=7.0, 2.2 Hz, 1 H), 7.77 (t, *J*=8.0 Hz, 1 H), 7.99 - 8.06 (m, 1 H), 8.20 (d, *J*=8.0 Hz, 1 H), 8.37 (t, *J*=1.5 Hz, 1 H), 8.50 (br. s., 1 H), 10.48 (s, 1 H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 163.9, 157.1, 143.5, 135.7, 134.7, 131.2, 129.6, 128.9, 125.4, 124.1, 123.7, 119.9, 114.8, 81.2, 54.7, 24.2, 14.3

_N_-(4-fluoro-2,3-dimethyl-phenyl)-3-[(3-methyloxetan-3-yl)sulfamoyl]benzamide (67).

Synthesis according to the representative procedure of scheme 1b as described for compound **53**. 27% yield. Method A; Rt: 4.98 min. m/z: 393.3 $(M+H)^+$ ESI-HRMS (TOF) m/z: $[M + H]^+$ 393.1287 (calcd for $C_{19}H_{21}FN_2O_4S$: 392.1206). ^1H NMR (400 MHz, DMSO-d_6) δ 10.22 (br s, 1H), 8.49 (br s, 1H), 8.40 (s, 1H), 8.25 (br d, *J*=8.07 Hz, 1H), 8.02 (d, *J*=8.07 Hz, 1H), 7.77 (t, *J*=7.87 Hz, 1H), 7.16 (dd, *J*=5.65, 8.48 Hz, 1H), 7.04 (t, *J*=8.88 Hz, 1H), 4.57 (d, *J*=6.06 Hz, 2H), 4.15 (d, *J*=6.06 Hz, 2H), 2.20 (d, *J*=1.61 Hz, 3H), 2.14 (s, 3H), 1.43 (s, 3H); ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.2, 158.7, 143.5, 135.7, 135.3, 131.9, 131.2, 129.6, 128.9, 125.8, 125.5, 123.4, 112.1, 81.2, 54.7, 24.2, 14.6, 11.2

_N_-(4-fluoro-2,5-dimethyl-phenyl)-3-[(3-methyloxetan-3-yl)sulfamoyl]benzamide (68).

Synthesis according to the representative procedure of scheme 1b as described for compound **53**. 34 % yield. Method A; Rt: 5.27 min. m/z: 393.3 $(M+H)^+$ Exact mass: 392.1. ESI-HRMS (TOF) m/z: $[M + H]^+$ 393.1290 (calcd for $C_{19}H_{21}FN_2O_4S$: 392.1206). ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.14 (br s, 1H), 8.53 (br s, 1H), 8.39 (s, 1H), 8.23 (d, *J*=7.26 Hz, 1H), 7.99-8.05 (m, *J*=7.30 Hz, 1H), 7.76 (t, *J*=7.81 Hz, 1H), 7.23 (d, *J*=7.48 Hz, 1H), 7.08 (d, *J*=10.34 Hz, 1H), 4.57 (d, *J*=6.16 Hz, 2H), 4.15 (d, *J*=6.60 Hz, 2H), 2.21 (s, 3H), 2.19 (s, 3H), 1.43 (s, 3H); ^{13}C NMR (101 MHz, DMSO-d_6) δ 164.1, 158.6, 143.5, 135.3, 133.8, 131.8, 131.1, 129.7, 129.6, 128.8, 125.4, 121.4, 116.2, 81.2, 54.7, 24.2, 17.4, 13.7

ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at http://pubs.acs.org. Experimental procedures for the synthesis of compound **8**, **9**, **11**-**13**, **17**-**27**, **62**, **64** and **65**; LC-MS methods, list of retention times and purities of tested compounds; details on JNJ-632 modeling; experimental details on the anti HBV cellular assay; experimental details on the size exclusion chromatography; experimental details on the biochemical fluorescence quenching; fluorescence quenching curve for representative compounds; experimental details on determination of metabolic stability in liver microsomes, kinetic solubility and pharmacokinetic assessment; experimental details on the determination of the anti-HBV activity of JNJ-632 in chimeric mice; List of molecular formula strings and associated biological data (CSV).

AUTHOR INFORMATION

Corresponding Author

*Koen Vandyck; Email: kvandyck@its.jnj.com

ACKNOWLEDGMENT

We thank Michel Carpentier, Alberto Fontana and Alex De Groot for analysis. We also thank Kenny Simmen for his support.

ABBREVIATIONS USED

HBV, Hepatitis B virus; (PEG)-IFNα (pegylated) interferon-alpha; CAM, Capsid assembly modulator; SBA, sulfamoylbenzamide; SOC, sulfamoyl carboxamide; PHH, primary human hepatocytes; HLM/MLM, human/mouse liver microsome stability; SEC, size exclusion chromatography; ND not determined, pgRNA, pre-genomic RNA; rcDNA, relaxed circular DNA; QD, quaque die or once a day.

REFERENCES

1. Ott, J. J.; Stevens, G. A.; Groeger, J.; Wiersma, S. T. Global Epidemiology of Hepatitis B Virus Infection: New Estimates of Age-Specific HBsAg Seroprevalence and Endemicity. *Vaccine* **2012,** 30, 2212-2219.

2. Berke, J. M.; Dehertogh, P.; Vergauwen, K.; Van Damme, E.; Mostmans, W.; Vandyck, K.; Pauwels, F. Capsid Assembly Modulators Have a Dual Mechanism of Action in Primary Human Hepatocytes Infected with Hepatitis B Virus. *Antimicrob Agents Chemother* **2017,** 61, e00560-17.

3. An Efficacy, Safety, and Pharmacokinetics Study of JNJ-56136379 in Participants With Chronic Hepatitis B Virus Infection. In https://ClinicalTrials.gov/show/NCT03361956 (accessed June 4, 2018).

4. Berke, J. M.; Dehertogh, P.; Vergauwen, K.; Van Damme, E.; Raboisson, P.; Pauwels, F.; Vandyck, K. In Capsid Assembly Modulator JNJ-56136379 Prevents de Novo Infection of Primary Human Hepatocytes With Hepatitis B Virus, AASLD The Liver Meeting, Boston, 2016; Boston, 2016.

5. Perni, R. B.; Conway, S. C.; Ladner, S. K.; Zaifert, K.; Otto, M. J.; King, R. W. Phenylpropenamide Derivatives as Inhibitors of Hepatitis B Virus Replication. *Bioorg Med Chem Lett* **2000,** 10, 2687-2690.

6. Feld, J. J.; Colledge, D.; Sozzi, V.; Edwards, R.; Littlejohn, M.; Locarnini, S. A. The Phenylpropenamide Derivative AT-130 Blocks HBV Replication at the Level of Viral RNA Packaging. *Antiviral Res* **2007,** 76, 168-177.

7. Deres, K.; Schroder, C. H.; Paessens, A.; Goldmann, S.; Hacker, H. J.; Weber, O.; Kramer, T.; Niewohner, U.; Pleiss, U.; Stoltefuss, J.; Graef, E.; Koletzki, D.; Masantschek, R. N.; Reimann, A.; Jaeger, R.; Gross, R.; Beckermann, B.; Schlemmer, K. H.; Haebich, D.; Rubsamen-Waigmann, H. Inhibition of Hepatitis B Virus Replication by Drug-Induced Depletion of Nucleocapsids. *Science* **2003,** 299, 893-896.

8. Last, S. J.; Raboisson, P. J.-M. B.; Rombouts, G.; Vandyck, K.; Verschueren, W. G. Sulfamoyl-Arylamides and the Use Thereof as Medicaments for the Treatment of Hepatitis B. WO2014033176, 2014.

9. Guo, J.-T.; Xu, X.; Block Timothy, M. Sulfamoylbenzamide Derivatives as Antiviral Agents Against HBV Infection. WO 2013/006394 A1, 2013.

10. Hartman George, D.; Flores Osvaldo, A. Hepatitis B Antiviral Agents. WO 2013/096744 A1, 2013.

11. Campagna, M. R.; Liu, F.; Mao, R.; Mills, C.; Cai, D.; Guo, F.; Zhao, X.; Ye, H.; Cuconati, A.; Guo, H.; Chang, J.; Xu, X.; Block, T. M.; Guo, J. T. Sulfamoylbenzamide Derivatives Inhibit the Assembly of Hepatitis B Virus Nucleocapsids. *J Virol* **2013,** 87, 6931-6942.

12. Klumpp, K.; Shimada, T.; Allweiss, L.; Volz, T.; Lütgehetmann, M.; Hartman, G.; Flores, O. A.; Lam, A. M.; Dandri, M. Efficacy of NVR 3-778, Alone and in Combination with Pegylated Interferon, vs Entecavir In uPA/SCID Mice with Humanized Livers and HBV Infection. *Gastroenterology* **2018,** 154, 652-662.

13. Sari, O.; Boucle, S.; Cox, B. D.; Ozturk, T.; Russell, O. O.; Bassit, L.; Amblard, F.; Schinazi, R. F. Synthesis of Sulfamoylbenzamide Derivatives as HBV Capsid Assembly Effector. *European Journal of Medicinal Chemistry* **2017,** 138, 407-421.

14. Bourne, C.; Lee, S.; Venkataiah, B.; Lee, A.; Korba, B.; Finn, M. G.; Zlotnick, A. Small-Molecule Effectors of Hepatitis B Virus Capsid Assembly Give Insight into Virus Life Cycle. *J Virol* **2008,** 82, 10262-10270.

15. Stray, S. J.; Johnson, J. M.; Kopek, B. G.; Zlotnick, A. An *In Vitro* Fluorescence Screen to Identify Antivirals that Disrupt Hepatitis B Virus Capsid Assembly. *Nat Biotechnol* **2006,** 24, 358-362.

16. Abraham, V. C.; Towne, D. L.; Waring, J. F.; Warrior, U.; Burns, D. J. Application of a High-Content Multiparameter Cytotoxicity Assay to Prioritize Compounds Based on Toxicity Potential in Humans. *J Biomol Screen* **2008,** 13, 527-537.

17. O'Brien, P. J.; Irwin, W.; Diaz, D.; Howard-Cofield, E.; Krejsa, C. M.; Slaughter, M. R.; Gao, B.; Kaludercic, N.; Angeline, A.; Bernardi, P.; Brain, P.; Hougham, C. High Concordance

of Drug-Induced Human Hepatotoxicity with In Vitro Cytotoxicity Measured in a Novel Cell-Based Model Using High Content Screening. *Arch Toxicol* **2006,** 80, 580-604.

18. Dandri, M.; Petersen, J. Chimeric Mouse Model of Hepatitis B Virus Infection. *J Hepatol* **2012,** 56, 493-495.

19. Rath, S. L.; Liu, H.; Okazaki, S.; Shinoda, W. Identification of Factors Promoting HBV Capsid Self-Assembly by Assembly-Promoting Antivirals. *Journal of Chemical Information and Modeling* **2018,** 58, 328-337.

20. Barr, C. R.; Salminen, I. F.; Weissberger, A. The Reaction of 3-Chlorosulfonylbenzoyl Chloride with Amines. *Journal of the American Chemical Society* **1951,** 73, 4131-4133.

21. Yang, Y.-L.; Rajagopal, B.; Liang, C.-F.; Chen, C.-C.; Lai, H.-P.; Chou, C.-H.; Lee, Y.-P.; Yang, Y.-L.; Zeng, J.-W.; Ou, C.-L.; Lin, P.-C. Chemoselective Synthesis of Aryl Carboxamido Sulfonic Acid Derivatives. *Tetrahedron* **2013,** 69, 2640-2646.

Table of content graphic