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Respiratory syncytial virus fusion inhibitors. Part 5: Optimization of benzimidazole substitution patterns towards derivatives with improved activity☆

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Abstract—Extensive SAR studies and optimization of ADME properties of benzimidazol-2-one derivatives led to the identification of BMS-433771 (**3**) as an orally active RSV fusion inhibitor. In order to extend the structure–activity relationships for this compound series, substitution of the benzimidazole ring was examined with a view to establishing additional productive interactions between the inhibitor and functionality present in the proposed binding pocket. Amongst the compounds synthesized, the 5-aminomethyl analogue **10aa** demonstrated potent antiviral activity towards wild-type RSV and retained excellent inhibitory activity towards a virus that had been developed to express resistance to BMS-433771 (**3**), data consistent with an additional productive interaction between the inhibitor and the fusion protein target.

Respiratory syncytial virus (RSV) is a leading cause of lower respiratory tract infection in infants and children.[5] Moreover, the recurrence of RSV infection in later life, a consequence of an immune response of poor durability, poses a serious health threat to the elderly and immunocompromised.[6] Despite the significant morbidity and mortality associated with RSV infections, the only therapeutic option currently available is the nucleoside analogue ribavirin, typically administered as an aerosol.[7,8] However, ribavirin is neither a particularly potent nor specific inhibitor of RSV and its teratogenic properties complicate administration.[8] Consequently, there is an unmet medical need for effective and specific therapies to treat this disease.

We have recently described the discovery of a class of benzimidazole-based inhibitors of RSV fusion, generically represented by **1**,[1] from which evolved a series of more potent RSV inhibitors based on the benzimidazol-2-one template **2**.[2] This chemotype tolerated a broad range of substituents appended to both the benzimidazole and benzimidazol-2-one moieties that encompassed a wide variation of size and functionality.[2] In vivo efficacy towards RSV infection was demonstrated in the cotton rat model with water-soluble derivatives delivered by small particle aerosol.[3] Further optimization for antiviral potency, membrane permeability and metabolic stability in human liver microsomes led to the identification of BMS-433771 (**3**) as a potent RSV inhibitor with oral bioavailability in the mouse, rat, dog and cynomolgus monkey that demonstrated antiviral activity

Keywords: RSV fusion inhibitor; Substitution of the benzimidazole; Binding pocket; 5-Aminomethyl analogue; Retained activity; Resistant virus; Asp200.

☆ For parts 1–4 of this series, see Refs. 1–4.

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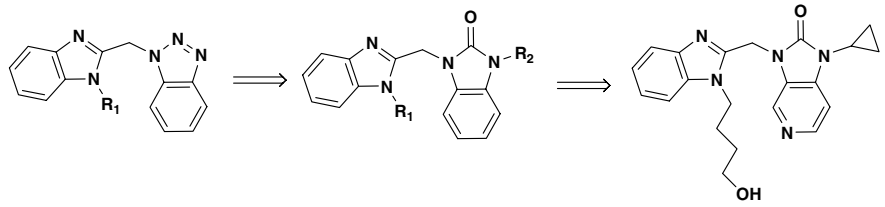


in the BALB/c and cotton rat models of infection following oral administration.[4,9,10]

Mechanistic and photo-affinity labelling studies indicated that this class of RSV inhibitors interfered with the assembly of the F protein 6-helix bundle, a critical step in the fusion of host cell and virus membranes.[11,12] The 6-helix bundle structure is thought to be formed in a stepwise fashion as part of the RSV F_1 protein rearrangement in which the lipophilic amino terminus, the fusion peptide, is inserted into the host cell membrane and a proximal heptad repeat region associates into a trimeric structure. This trimer element subsequently interacts with a complementary heptad repeat present in the carboxy terminal that binds to it in an anti-parallel fashion, creating the 6-helix bundle that is thought to mediate the coalescence of host and viral membranes. Modelling studies suggested that **1**–**3** and related compounds bind into a pocket created by the association of the amino terminus heptad repeat into the trimeric structure.[11,12] The favoured binding pose placed the benzimidazole heterocycle in the cavity occupied by Phe483 of the carboxy terminus heptad repeat in the fusogenic state whilst the benzimidazol-2-one element extended into the pocket accommodating Phe488 and Leu492 of the C-terminus.[11] The proposed binding mode provided an opportunity for structure-based inhibitor design involving an examination of substitution patterns associated with the benzimidazole heterocycle and also suggested potential avenues towards further enhancing the antiviral activity of the chemotype. More specifically, the binding mode hypothesis suggested that substitution at C-4 and C-7 of the benzimidazole would be poorly tolerated, since these sites are in close proximity to elements of the protein, whilst C-5 and C-6 would be more accommodating. More intriguingly, the vectors associated with C-5 and C-6 project towards Asp200, suggesting that the introduction of basic substituents in this region may lead to the establishment of a productive salt bridge or hydrogen bonding interaction (Fig. 1). In this communication, we describe the results of this initiative which led to the identification of potent inhibitors of wild-type RSV in vitro that demonstrated improved activity towards a resistant strain, consistent with the notion that an additional interaction between the inhibitor and the viral fusion protein had been established.[13]

The target molecules **10** were assembled by the pathways outlined in Schemes 1 and 2. Substituents were introduced to the benzimidazole ring by employing either a bromo- or cyano-substituted-2-chloro- or 2-fluoro-

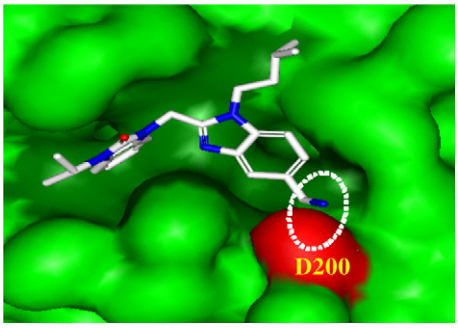
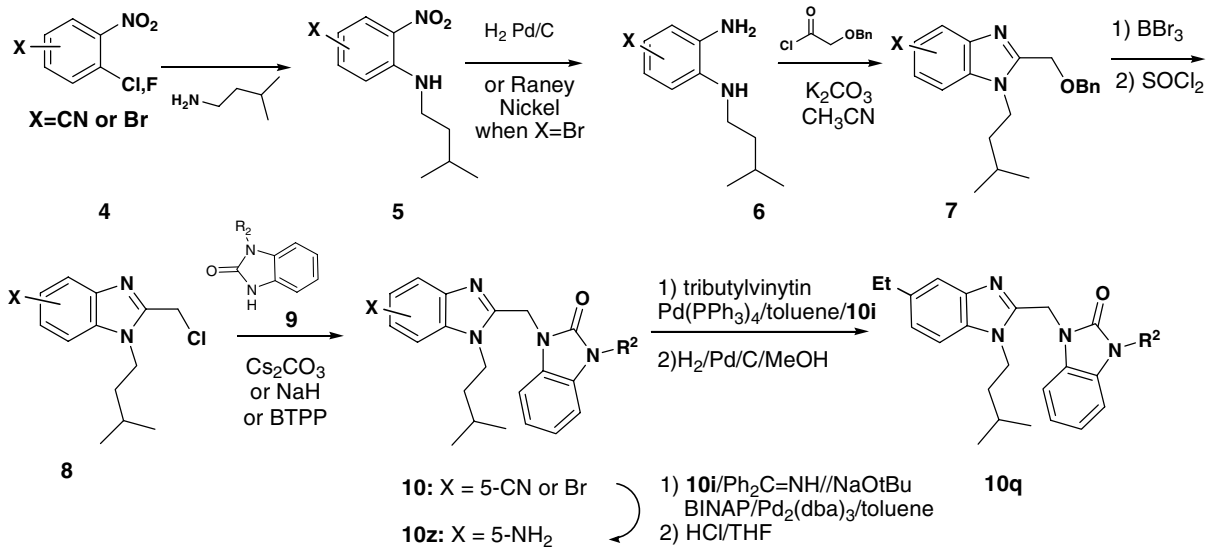
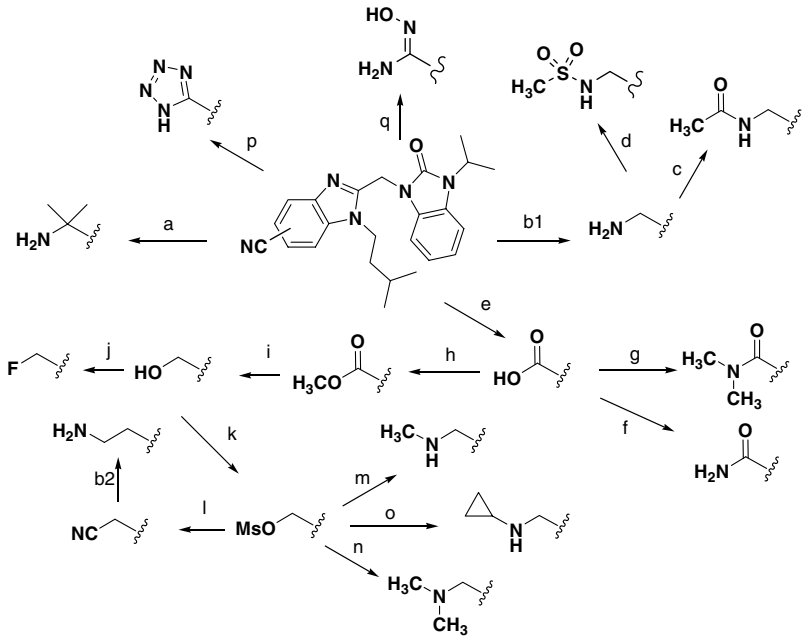


Figure 1. Engaging Asp200 in the proposed binding site.

nitrobenzene **4** as the starting material, as depicted in Scheme 1. The introduction of the side chain ultimately attached to the benzimidazole ring was accomplished by displacement of the chlorine or fluorine atom of **4** by *iso*-amylamine to give the nitroaniline **5** in good yield. Hydrogenation of the nitro group in **5**, catalyzed either by Pd/C (when X = CN) or Raney nickel (when X = Br), afforded the diamine **6** quantitatively. Acylation and ring closure to the benzimidazole **7** was effected by reaction with 2-(benzyloxy)acetyl chloride. The benzyl protecting group of **7** was removed by treating with BBr_3 and the resultant alcohol converted to the chloride **8** using $SOCl_2$. Coupling of the chloride **8** with a monosubstituted benzimidazol-2-one **9**, readily obtained by several complementary pathways,[14–19] was accomplished using either Cs_2CO_3/acetone, NAH/DMF, or BTPP/THF[5] to afford bromo- or cyano-substituted derivatives **10**. The bromide moiety of **10i** was converted to the 5-ethyl analogue **10q** by a Pd-catalyzed Stille[20] coupling with tributylvinyltin, to afford the vinyl substituted compound **10p**, followed by hydrogenation. Amination of **10i** with diphenylmethanimine under Buchwald–Hartwig conditions,[21] followed by acid hydrolysis, provided the aniline **10z**. The nitrile-substituted derivatives of **10** were particularly versatile intermediates, readily elaborated into a series of substituted analogs, as summarized in Scheme 2. Hydrogenation of the nitrile moiety of **10j** and **10aq** provided the aminomethyl derivatives **10aa** and **10ar**, respectively (Scheme 2, step b1), which were readily acylated or sulfonylated (Scheme 2, steps c and d). Heating **10j** with NaN_3 in DMF provided the tetrazole **10v** (Scheme 2, step p) whilst reaction with NH_2OH in EtOH at reflux provided hydroxyamidines **10al**–**an** (Scheme 2, step q). The disubstituted aminomethyl compound **10ah** was obtained by the reaction of methylcerium with the nitrile **10j** (Scheme 2, step a).[22] Hydrolysis of the nitrile moiety provided the structurally analogous carboxylic acid derivatives



Scheme 1.

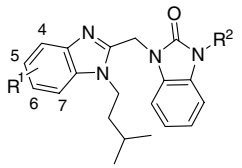


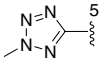
Scheme 2. Reagents and conditions: (a) $CeCl_3$, MeLi, THF; (b1 and b2) H_2, Pd/C, MeOH; (c) acetyl chloride, TEA, THF; (d) MsCl, TEA, THF; (e) 6 M NaOH, refluxing; (f) ammonia, EDC, DCM; (g) dimethylamine, EDC, DCM; (h) MeOH, H_2SO_4; (i) LAH, THF; (j) DAST, DCM; (k) MsCl, TEA, DCM; (l) KCN, DMF; (m) methylamine, THF; (n) dimethylamine, THF; (o) cyclopropylamine, THF; (p) NaN_3, DMF; (q) NH_2OH, refluxing EtOH.

(Scheme 2, step e) which were coupled with ammonia or dimethylamine, to afford the corresponding amides (Scheme 2, steps f and g), or heated in acidic MeOH to provide the methyl ester derivatives (Scheme 2, step h). Reduction of the methyl ester using LAH in THF afforded the corresponding benzyl alcohol (Scheme 2, step i), which was converted to a fluoride by exposure to DAST (Scheme 2, step j). Alternatively, mesylation of the benzyl alcohol (Scheme 2, step k) and subsequent displacement with KCN yielded the benzyl cyanide (Scheme 2, step l) which, in turn, was hydrogenated to give the aminoethyl homologue (Scheme 2, step b2). Reaction of the mesylate with methylamine, dimethylamine and cyclopropylamine afforded a series of benzylamines in good yield (Scheme 2, steps m–o). The compounds prepared as part of this survey are compiled in Table 1.

Inhibition of the Long A strain of RSV replicating in the HEp-2 human lung epithelial carcinoma cell line was used to evaluate the antiviral activity of target compounds by monitoring the RSV-induced cytopathic effect (CPE).[9,10] Long strain virus resistant to BMS-433771 (**3**) that expressed a K394R mutation in the F_1 protein was generated in HEp-2 cells by passage of virus in the presence of increasing concentrations of 2-(2-((3-

Table 1. Structure, RSV inhibitory activity and cytotoxicity associated with a series of 5-substituted benzimidazole derivatives



Compound	R^1	R^2	EC_{50}[a] (μM)	CC_{50} (μM)	Therapeutic index[b]
2a	H	Isopropenyl	0.004 (0.005, 0.003)	24.6 (37.6, 11.7)	6162
10a	4-CH_2NH_2	*i*-Pr	0.715 (0.395, 1.03)	7.00 ± 2.50 (n = 3)	9.78
10b	4-$CH_2CH_2NH_2$	*i*-Pr	0.230 (0.375, 0.084)	3.92 (3.71, 4.12)	17.0
10c	4-CH_2OH	*i*-Pr	0.82 (1.34, 0.30)	22.0 (14.1, 29.8)	26.8
10d	4-CH_2OCH_3	*i*-Pr	7.50 (7.50, 7.51)	26.7 (20.5, 32.9)	3.56
10e	4-CO_2CH_3	*i*-Pr	230 (230, 230)	49.2 (7.39, 91)	0.213
10f	4-CH_2CN	*i*-Pr	241 (241, 241)	116 (182, 49.4)	0.482
10g	4-CH_2CH_2CN	*i*-Pr	234 (234, 234)	2.24 (1.38, 3.10)	0.096
10h	4-$CH_2CH_2CH_2NH_2$	*i*-Pr	213 (213, 213)	2.34 (1.54, 3.13)	0.011
10i	5-Br	*i*-Pr	0.018 (0.012, 0.025)	18.1 (15.4, 20.8)	1006
10j	5-CN	*i*-Pr	0.476 (0.298, 0.655)	7.93 (10.9, 4.96)	16.6
10k	5-CN	Isopropenyl	0.057 (0.026, 0.088)	1.48 (1.04, 1.96)	26.0
10l	5-CN	$CH_2CO_2{}^t$Bu	2.55 ± 2.22 (n = 3)	32.2 ± 17.8 (n = 3)	12.6
10m	5-CH_2CN	*i*-Pr	0.117 (0.153, 0.081)	13.3 (19.8, 6.81)	114
10n	5-$CH_3C(O)$	Isopropenyl	0.179 (0.264, 0.094)	0.74 (0.50, 0.98)	4.1
10o	5-$CH_3C(O)$	Me	0.193 ± 0.178 (n = 3)	5.20 ± 2.00 (n = 3)	39.8
10p	5-CH_2=CH	*i*-Pr	0.630 (0.490, 0.770)	6.52 (4.45, 8.58)	10.3
10q	5-CH_3CH_2	*i*-Pr	0.692 (0.420, 0.963)	18.5 (33.7, 6.37)	26.8
10r		*i*-Pr	21.6 ± 20.2 (n = 3)	223 ± 2.9 (n = 3)	10.3
10s		Me	239 (n = 1)	2.97 (n = 1)	0.012
10t	5-CO_2H	Isopropenyl	264 (229, 299)	30.0 (26.8, 33.2)	0.11
10u	5-CO_2H	*i*-Pr	238 (n = 1)	17.3 (n = 1)	0.07
10v		*i*-Pr	225 (225, 225)	2.00 (1.60, 2.41)	0.009
10w	5-CO_2Me	*i*-Pr	230 (230, 230)	33.7 (15.5, 51.9)	0.15
10x	5-$C(O)NH_2$	*i*-Pr	0.166 (0.146, 0.187)	23.4 (21.1, 25.6)	141
10y	5-$C(O)N(CH_3)_2$	*i*-Pr	24.8 (20.4, 29.1)	25.6 (12.5, 38.6)	1.03
10z	5-NH_2	*i*-Pr	0.125 (0.206, 0.044)	32.2 (23.2, 41.3)	258
10aa	5-CH_2NH_2	*i*-Pr	0.002 (0.002, 0.002)	3.92 (3.23, 4.62)	1962
10ab	5-$CH_2CH_2NH_2$	*i*-Pr	4.33 (1.16, 7.5)	11.0 (10.8, 11.2)	2.54
10ac	5-$CH_2NH.CH_3$	*i*-Pr	0.057 (0.026, 0.088)	1.48 (1.04, 1.96)	26.0
10ad	5-CH_2NH.c-Pr	*i*-Pr	0.363 (0.313, 0.414)	17.2 (26.7, 7.62)	47.3
10ae	5-$CH_2N(CH_3)_2$	*i*-Pr	183 (183, 183)	5.65 (3.30, 8.00)	0.031
10af	5-$CH_2NHC(O)CH_3$	*i*-Pr	0.631 (0.357, 0.905)	178 (178, 178)	282
10ag	5-$CH_2NHS(O)_2Me$	*i*-Pr	0.725	30.8 (21.4, 40.1)	42.4
10ah	5-$C(CH_3)_2NH_2$	*i*-Pr	0.694 (0.689, 0.698)	3.27 (0.50, 0.98)	4.71
10ai	5-$C(NH)NH_2$	Isopropenyl	0.004 (0.004, 0.004)	10.8 (10.2, 11.4)	2700
10aj	5-$C(NH)NH_2$	$CH_2CO_2{}^t$Bu	0.009 (0.006, 0.012)	7.90 (7.40, 8.40)	878
10ak	5-$C(NH)NH_2$	CH_2CO_2H	0.031± 0.011 (n = 3)	173 ± 5.2 (n = 3)	5574
10al	5-$C(NOH)NH_2$	Isopropenyl	0.005 (0.006, 0.005)	3.47 (2.22, 4.72)	694
10am	5-$C(NOH)NH_2$	*i*-Pr	0.111 (0.194, 0.028)	9.14 (11.2, 7.07)	82.2
10an	5-$C(NOH)NH_2$	$CH_2CO_2{}^t$Bu	0.016 (0.014, 0.018)	19.8 (10.6, 29.1)	1240
10ao	5-CH_2OH	*i*-Pr	0.050 ± 0.044 (n = 3)	31.6 ± 17.7 (n = 3)	632
10ap	5-CH_2F	*i*-Pr	0.047 (0.037, 0.058)	37.3 (29.2, 80.6)	793
10aq	6-CN	*i*-Pr	0.044 (0.026, 0.063)	10.0 (17.5, 2.53)	228
10ar	6-CH_2NH_2	*i*-Pr	0.095 (0.118, 0.071)	28.1 (16.6, 40.2)	296
10as	6-CH_2OH	*i*-Pr	0.134 (0.220, 0.047)	81.6 (119, 44.3)	609
10at	6-CH_2NHCH_3	*i*-Pr	187	4.81 ± 3.41 (n = 3)	0.026
10au	6-$CH_2N(CH_3)_2$	*i*-Pr	182	26.7 ± 15.4 (n = 3)	0.026
10av	6-$CH_2CH_2NH_2$	*i*-Pr	7.11 ± 6.89 (n = 3)	57.0 ± 87.4 (n = 3)	8.02
10aw	6-CO_2H	*i*-Pr	238	125	0.525
10ax	6-$C(O)NH_2$	*i*-Pr	123 (238, 8.04)	49.9 (42.6, 57.2)	0.406
10ay	7-CH_2NH_2	*i*-Pr	192 (192, 192)	11.0 (13.6, 8.50)	0.058

(*continued on next page*)

Table 1 (*continued*)

Compound	R^1	R^2	EC_{50}[a] (μM)	CC_{50} (μM)	Therapeutic index[b]
10az	7-CO_2CH_3	*i*-Pr	230 (230, 230)	25.3 (15.5, 35.1)	0.11
10aaa	7-CH_2OH	*i*-Pr	246 (246, 246)	246	1.00
10aab	7-N_3	*i*-Pr	3.70 (6.45, 0.94)	57.9 (52.9, 62.9)	15.6

[a] Values are means of two or more experiments performed on consecutive weeks with the data from individual experiments shown in parentheses.
[b] Therapeutic index = CC_{50}/EC_{50}.

iodo-1*H*-indazol-1-yl)methyl)-1*H*-benzo[*d*]imidazol-1-yl)-*N*,*N*-dimethylethanamine (BMS-243458), as described previously.[9] The EC_{50} values presented in Table 1 represent the concentration of test compound preserving 50% of infected cells whilst the CC_{50} data are the concentration of drug at which cytotoxicity towards 50% of uninfected HEp-2 cells was observed. The therapeutic index reported in Table 1 is the ratio of CC_{50} to EC_{50}. Compounds were typically tested in two experiments conducted on consecutive weeks with additional evaluations performed if there was a significant discrepancy between the initial data sets. Where the data reported are the average of two experiments, the individual results are provided as a measure of assay variability.

The proposed model of the binding of this class of RSV fusion inhibitors to the assembled trimer of the N-terminal heptad repeat elements suggests that the 4- and 7-positions of the benzimidazole ring are proximal to the wall of the binding pocket. Consequently, antiviral activity would be anticipated to be quite sensitive to substitution at either of these sites of the core heterocycle. It is clearly apparent from the data presented in Table 1 for the series of 4-substituted analogues **10a**–**h** and the 7-substituted derivatives **10ay**–**aab** that the introduction of substituents at these positions of the heterocyclic nucleus results in a significant reduction in potency compared to the parent compound **2a**.[2] Both an aminomethyl (**10a**) and a hydroxymethyl (**10c**) substituent at C-4 result in over a 150-fold abrogation of potency, a circumstance marginally improved by the aminoethyl homologue **10b** but considerably degraded by the larger or more rigid polar substituents exemplified by **10d**–**h**. The 7-position is particularly sensitive to modification and, with the exception of azide **10aab**, all of the compounds examined in this series, **10ay**–**aaa**, are at least 30,000-fold less potent than the unsubstituted prototype **2a**. Only the azide **10aab** demonstrates measurable RSV inhibitory activity but this compound is, nevertheless, still 900-fold weaker than **2a**.

The C-6 position proved to be more tolerant of substitution with cyanide (**10aq**), aminomethyl (**10ar**) and hydroxymethyl (**10as**) derivatives potent inhibitors of RSV, although they are inferior to the parent compound **2a** by an order of magnitude. Homologation of the aminomethyl derivative **10ar** to the aminoethyl substituted compound **10av** erodes potency by 75-fold whilst the mono- and di-methylated amines **10at** and **10au** are inactive. The final compound in this series, the carboxylic acid **10aw**, is cytotoxic without demonstrating significant antiviral activity.

The introduction of substituents at C-5 proved to be a considerably more productive exercise, with the consequence that the survey conducted at this position of the benzimidazole heterocycle is more extensive, examined in the context of the series of derivatives **10i**–**ap**. The structure–activity relationships enumerated by **10i**–**ap** are both interesting and informative, providing critical insights into aspects of the pharmacophore and the proposed mode of binding of this series of fusion inhibitors to the RSV F_1 protein trimer. Small aliphatic elements, bromine, cyanide, cyanomethyl, and acetyl substituents afford compounds **10i**–**q** that are quite potent antiviral agents when compared to the progenitor **2a**, with the weaker activity associated with **10l** attributable to the acetic ester moiety appended to the benzimidazol-2-one.[2] The poor activity associated with oxadiazole **10r** is suggestive of limited tolerance for steric bulk at C-5, an observation further explored with the methylated tetrazole **10s**, which is not only poorly active, but also cytotoxic. The two C-5 carboxylates **10t** and **10u** and the isosteric[23] tetrazole **10v** are also cytotoxic at micromolar concentrations. The ester **10w** demonstrates cytotoxicity comparable to the acid **10u**, possibly a consequence of esterolytic cleavage under the assay conditions. Stabilization of this moiety by conversion to the primary amide **10x** revealed the inherent potency of a non-negatively charged element at this site but increasing the steric bulk by dimethylation led to a 100-fold reduction in potency. The C-5 amino derivative **10z** proved to be modestly potent but was substantially improved by homologation to the more basic aminomethyl derivative **10aa**, a compound with antiviral activity comparable to the parent **2a**. Informatively, extension of the amino moiety of **10aa** by a single carbon atom, to give **10ab**, led to a precipitous 2000-fold decline in potency, indicative of a precise disposition for the basic amine relative to the heterocyclic nucleus. Whilst **10aa** and **2a** demonstrate comparable activity towards wild-type virus, a more effective indicator of the intrinsic antiviral properties of the aminomethyl derivative **10aa** is revealed by the EC_{50} for inhibition of the K394R resistant virus.[9] The K394R mutation in the RSV F_1 protein confers resistance ranging from 35- to >1250-fold towards representatives of this fusion inhibitor chemotype but remains highly sensitive to inhibition by **10aa**, which displays an EC_{50} of 20 nM, as assessed by the reduction in RSV-specific protein synthesis.[9] In contrast, the EC_{50} for BMS-433771 (**3**) and related compounds towards the K394R-containing virus is >20 μM. These data suggest the presence of an additional positive and specific interaction between **10aa** and the target protein and are consistent with the formation of a salt bridge interaction between the amine and Asp200 in

the active site model proposed for these molecules. A series of derivatives of **10aa** prepared to further probe the requirements of this interaction lend support to this concept. Alkylation of **10aa** leads to reduced potency, with the methyl analogue **10ac** and the less basic[24] cyclopropylamine **10ad** significantly weaker antiviral agents, whilst the dimethyl derivative **10ae** is essentially inactive. Similarly, acylation (**10af**) and sulfonylation (**10ag**) lead to substantially diminished antiviral activity whilst gem-dimethylation of the benzylic carbon (**10ah**), a modification that adds steric bulk and is anticipated to affect conformational preference,[25] also erodes activity. Substitution at the benzylic methylene atom in the context of an amidine, compounds **10ai–k**, provided a symmetrical and more basic element that conferred potent RSV inhibition. However, in this series, antiviral activity is not absolutely dependent on basicity since the hydroxyamidines[26] **10al–n** retain activity, possibly a reflection of productive hydrogen bonding interactions with Asp200. The survey was completed with the evaluation of the hydroxymethyl (**10ao**) and fluoromethyl (**10ap**) analogues, both of which are more potent than the 5-ethyl derivative **10q**.

The pattern of structure–activity relationships summarized by the data presented in Table 1 are compatible with the basic tenets of the model of the binding of RSV fusion inhibitors to the pocket originally identified by Zhao and colleagues as a potential target for small molecule drugs.[12] The generally poor activity associated with the introduction of substituents at C-4 and C-7 of the benzimidazole nucleus and the greater, but still limited, tolerance for substituents at C-6 are predicted quite effectively by the proposed binding mode.[11] Most gratifyingly based on the original premise, the introduction of basic functionality at C-5 provided compounds retaining full activity towards wild-type virus that also demonstrated potent antiviral activity towards virus developed to be resistant to representatives of this class of fusion inhibitors. Virus expressing the K394R mutation is potently inhibited by the benzylamine **10aa** with an EC_{50} comparable to that observed towards wild-type virus. A model of **10aa** bound in the proposed binding site and engaging Asp200 is depicted in Figure 2, where the vector for projecting the basic amine or amidine moiety is optimal. However, modest alterations in the binding pose may allow access to Asp200 from C-4 or C-6, providing a potential explanation for the significant antiviral activity retained by the aminomethyl derivatives **10a** and **10ar**, respectively.

The important effect of a basic functional group at C-5 of these RSV fusion inhibitors provides an interesting convergence of the silhouette, particularly for the amidine derivatives **10ai–h**, with bis(5-amidino-2-benzimidazolyl)methane (BABIM, **11**), a compound with broad spectrum, trypsin-like serine protease inhibition that has also been reported to be an inhibitor of RSV fusion.[27–31] In our hands, BABIM (**11**) displays somewhat modest potency with an EC_{50} of 100 nM towards the Long A strain of virus. More importantly, serial passage of RSV in the presence of BABIM (**11**) selected for resistant virus incorporating a F140I mutation in the F_1 protein that showed cross-resistance to BMS-433771 (**3**), strongly suggesting that these two compounds share a similar mode of action.[9,32] Whilst BABIM (**11**) is a topologically symmetrical molecule in contrast to the asymmetric silhouette presented by BMS-433771 (**3**) and closely related compounds, the structure–activity relationships associated with the benzotriazole series **1** with which activity was originally discovered indicated that a pseudo-symmetrical silhouette for the two heterocycle elements is compatible with antiviral activity.[1] In the benzotriazole series, those matched pairs of N-1 and N-2 isomers examined proved to be essentially equipotent.[1] However, BABIM (**11**) clearly lacks the hydroxybutyl side chain found in BMS-433771 (**3**), an element of the pharmacophore tolerant of structural variation but of critical importance for potency within this series of fusion inhibitors.[1–4] Taken together, these data suggest that the absence of a side chain in BABIM (**11**) may be offset by the positive effect of the amidine moiety which, presumably, can effectively engage Asp200.

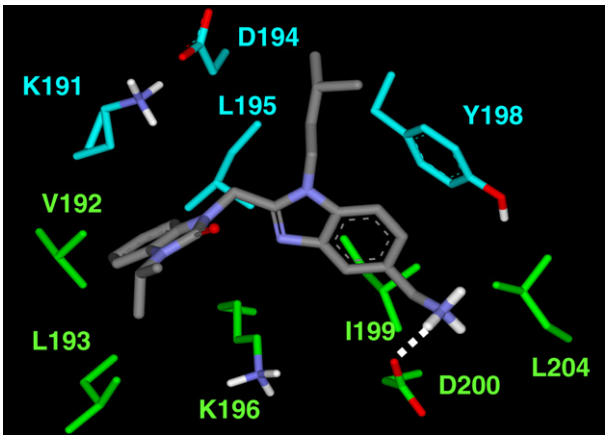
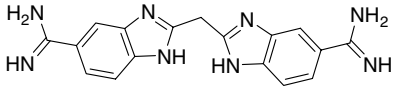


Figure 2. A model of **10aa** in the binding site of the RSV F_1 protein amino terminus heptad repeats showing the proposed intermolecular interactions with Asp200. The model depicts **10aa** bound into a pocket created by two of the α-helixes, designated A (green) and B (light blue). All N atoms are depicted in blue, O atoms in red and H atoms in white with only polar H atoms depicted for simplicity. This model is based on the 1G2C.pdb file.[12]



11 (BABIM)

In summary, the structure–activity relationships summarized in Table 1 are consistent with the binding pose that was proposed based on an analysis of photo-affinity labelling experiments. This model suggests limited tolerance for substitution at C-4 and C-7 of the benzimidazole moiety and the potential to establish a salt bridge interaction with Asp200 in the pocket created by assembly of the N-terminal heptad repeat into a trimer. The installation of basic moieties at C-5, particularly aminomethyl and amidine, resulted in potent RSV inhibitors that fully retain activity towards virus demonstrating over 1000-fold resistance to molecules devoid of substitution.

References and notes

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