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Respiratory syncytial virus fusion inhibitors. Part 6: An examination of the effect of structural variation of the benzimidazol-2-one heterocycle moiety☆

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Abstract—The effect of structural variation of the benzimidazol-2-one ring of RSV fusion inhibitors related to BMS-433771 (**1**) was examined in conjunction with side chain modifications and the introduction of an aminomethyl substituent at the 5-position of the core benzimidazole moiety. Replacement of the benzimidazol-2-one moiety with benzoxazole, oxindole, quinoline-2-one, quinazolin-2,4-dione and benzothiazine derivatives provided a series of potent RSV fusion inhibitors **4**. However, the intrinsic potency of 6,6-fused ring systems was generally less than that of comparably substituted 5,6-fused heterocycles of the type found in BMS-433771 (**1**). The introduction of an aminomethyl substituent to the benzimidazole ring enhanced antiviral activity in the 6,6-fused ring systems.

The discovery of a class of benzimidazole-based inhibitors of respiratory syncytial virus (RSV) fusion using a virus replication assay and the development of this chemotype into compounds that demonstrate antiviral activity in two rodent models of infection has been described.[1–5] BMS-433771 (**1**) was identified from this initiative as a compound that combines potent in vitro RSV inhibitory activity with oral bioavailability in four species and efficacy in mouse and cotton rat infection models following oral drug administration.[6–9] Mechanistic studies using a photoaffinity probe have established that these compounds interfere with the formation of the 6-helix bundle of the RSV F_1 protein, an essential step in the fusion of virus and host cell membranes that occurs in the latter stages of the entry process.[10] A model of the binding of this class of RSV inhibitor to the assembled trimer of the N-terminal heptad repeat element of the RSV F_1 protein provided an opportunity to explore the introduction of functionality designed to establish additional contact with the viral protein.[10] This exercise proved to be successful to the extent that basic substituents introduced at C-5 of the benzimidazole heterocycle led to compounds with enhanced potency, most effectively demonstrated in the context of viruses resistant to the unsubstituted parent.[5] In this article we describe the effects of variation of the azabenzimidazol-2-one moiety of BMS-433771 (**1**) to a range of structurally analogous and homologous

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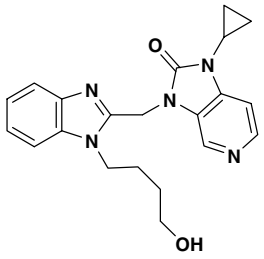
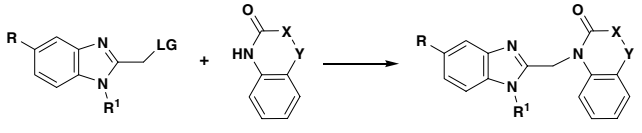
heterocycles, a study designed to develop a deeper understanding of the silhouette associated with the pharmacophore. Moreover, it was anticipated that by judiciously selecting the heterocycle moiety, both novel substituent vectors and the effect of introducing polar structural elements within the heterocycle ring could be evaluated. Of particular interest, this survey was viewed as a potentially useful approach to examining the effects of reducing electron density in the fused phenyl ring, an element of the pharmacophore of critical importance to potency but which, in the prototypical benzimidazol-2-one series, is also subject to metabolic hydroxylation to phenol derivatives with reduced antiviral potency.



1 (BMS-433771)

Target compounds represented generically by **4** were prepared in a straightforward fashion that relied upon variations of previously described chemistries in which a heterocycle **3** was alkylated with an N-substituted benzimidazole derivative **2** incorporating a leaving group (**LG**), typically chlorine, bromine or a mesylate, on the 2-methyl moiety.[1–5] The side chains defined by R^1 were selected based on established structure-activity relationships (SAR) for antiviral activity, pharmacokinetic properties (as predicted by in vitro microsomal stability and Caco-2 cell permeability) and physical properties.[1–5] This facilitated the optimal combination of substitution patterns in which the polarity of R^1 could be modulated to complement the nature of the heterocycle or the appended substituents, also selected based on prior experience. The compounds prepared as part of this survey are compiled in Table 1.



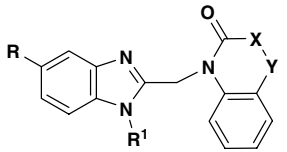
2 **3** **4**

The antiviral activity of synthetic compounds was established using an in vitro tissue cell culture screen in which prevention of RSV long strain-induced cytopathy towards the human HEp-2 epithelial carcinoma lung cell line was determined in concert with an essentially identical experiment that omitted virus as a means of assessing the cytotoxicity of the compounds towards the host cells.[1–7] Experiments were typically performed on consecutive weeks to provide an average EC_{50} and CC_{50} and repeated a third time if data between the first two experiments were considered to be significantly discordant. The data are compiled in Table 1, where results of individual experiments are included to provide an indication of the variability associated with the assay. Long strain virus expressing a K394R mutation in the F_1 protein that confers resistance to BMS-433771 (**1**) was generated in HEp-2 cells by passage of virus in the presence of increasing concentrations of an earlier representative of this chemotype, 2-(2-((3-iodo-1*H*-indazol-1-yl)methyl)-1*H*-benzo[*d*]imidazol-1-yl)-*N*,*N*-dimethylethanamine (BMS-243458), as described previously.[6]

All of the compounds presented in Table 1 retain the amide element present in the azabenzimidazol-2-one of BMS-433771 (**1**) and its predecessors with the exception of the benzotriazole series with which RSV inhibitory activity was originally discovered.[1] Whilst the binding mode proposed to explain the activity of these RSV inhibitors[10] does not anticipate the carbonyl moiety functioning as a hydrogen bond acceptor and can, indeed, be dispensed with,[1] we have observed that compounds with this structural element generally demonstrate favourable antiviral and physical chemical properties in a more consistent fashion. The data presented in Table 1 reveal that a wide range of heterocycles are compatible with in vitro RSV inhibitory activity, although there is a clear variation in potency that appears to be attributable, in part, to the unique combinations of substituents appended to the invariant benzimidazole moiety and the heterocycle, with polar moieties at both of these sites generally providing poorer antiviral agents.

The initial survey focused on a short series of fused 5–6 ring systems with the benzoxazoles **4a** and **4b** maintaining the basic silhouette of BMS-433771 (**1**), whilst the oxindoles **4c**–**k** restore the opportunity for substitution afforded by the benzimidazol-2-one by functionalization of the oxindole carbon. This is a critical site of the pharmacophore and the oxindole series, uniquely within the fused azole topology, allows an evaluation of the effect of probing the stereochemical dimension orthogonal to the plane of the heterocycle. Whilst the benzoxazole **4a** is a potent antiviral agent, the introduction of the more polar nitrile-containing side chain found in **4b** markedly reduces potency, a result confirmed using an assay that relied upon the quantitative assessment of viral protein synthesis.[6] This result may reflect the trend observed earlier in which the combination of polar moieties at both of these sites of the pharmacophore leads to reduced potency.[1–5] Within the oxindole series, the prototype **4c** is a potent RSV inhibitor comparable to the unsubstituted benzimidazol-2-one parent[2] and the benzoxazole **4a**. The activity of **4c** is marginally improved by double methylation of the C-3 carbon atom (**4g**), but progressively eroded by increasing polarity at this site (**4d**–**f**) or when polar moieties are incorporated into the side chain, as exemplified by compounds **4h**–**k** (note that compounds **4e**–**f** and **4h**–**k** were examined as racemates). These results largely recapitulate the SAR observed with the benzimidazol-2-one series where

Table 1. Structure of RSV inhibitors **4**



Compound	R	R^1	X	Y	Average EC_{50}[a] (μM)	EC_{50} (μM) from individual experiments or number of repetitions if variant from 2	Average CC_{50}[a] (μM)	CC_{50} (μM) from individual experiments or number of repetitions if variant from 2
4a	H	$(CH_2)_2CH(CH_3)_2$	O	—	0.044	0.050, 0.038	30.7	36.5, 24.9
4b	H	$(CH_2)_3CN$	O	—	2.51	2.73, 2.30	>300	>300, >300
4c	H	$(CH_2)_2CH(CH_3)_2$	CH_2	—	0.032	0.026, 0.039	8.58	5.50, 11.66
4d	H	$(CH_2)_2CH(CH_3)_2$	C=O	—	0.23	0.236, 0.221	69.80	6.33, 133.26
4e	H	$(CH_2)_2CH(CH_3)_2$	CH(OH)	—	0.164	0.085, 0.243	3.31	4.33, 2.28
4f	H	$(CH_2)_2CH(CH_3)_2$	$C(CH_3)OH$	—	0.097	0.043, 0.152	101.19	65.56, 136.81
4g	H	$(CH_2)_3CN$	$C(CH_3)_2$	—	0.019	0.014, 0.025	>100.44	100.44, >278.98
4h	H	$(CH_2)_2CH(CH_3)_2$	$CHCH_2CO_2H$	—	0.705 ± 0.912	$n = 3$	16.51 ± 14.28	$n = 3$
4i	H	$(CH_2)_2CH(CH_3)_2$	$CHCH_2CON(CH_3)_2$	—	0.205	0.085, 0.325	4.67	4.11, 5.23
4j	H	$(CH_2)_2CH(CH_3)_2$	$CHCH_2CONHCH_2Ph$	—	0.110	0.086, 0.134	0.72	1.12, 0.31
4k	H	$(CH_2)_2CH(CH_3)_2$	$CHCH_2CONHCH_2CH_2Ph$	—	0.379	0.303, 0.455	0.94	0.14, 1.75
4l	H	$(CH_2)_4F$	CH	CH	2.305	3.77, 0.84	103.4	160.7, 46.1
4m	H	$(CH_2)_4F$	CEt	CH	0.902	$n = 1$	85.06	$n = 1$
4n	H	$(CH_2)_4F$	CH	$C(OCH_3)$	0.211	0.293, 0.130	59.49	56.45, 62.53
4o	NH_2CH_2	$(CH_2)_4F$	CH	$C(OCH_3)$	0.009	0.016, 0.003	24.05	31.39, 16.71
4p	NH_2CH_2	$(CH_2)_4OH$	CH	$C(OCH_3)$	0.008	0.013, 0.003	81.46	80.11, 82.82
4q	H	$(CH_2)_4F$	$C(OCH_3)$	$C(CO_2Et)$	0.147	0.213, 0.081	61.61	68.45, 54.78
4r	H	$(CH_2)_4F$	C(OH)	$C(CO_2H)$	25.92	16.70, 35.14	>38.63	>244.25, 38.63
4s	H	$(CH_2)_4F$	C(Br)	CH	0.788	$n = 1$	26.21	$n = 1$
4t	H	$(CH_2)_4F$	C(OH)	CH	>273.67	>273.67, >273.67	20.16	10.39, 29.93
4u	H	$(CH_2)_4CN$	CH_2	CH_2	1.107	1.577, 0.637	27.10	42.60, 11.60
4v	H	$(CH_2)_2N(CH_3)_2$	CH_2	CH_2	0.456	0.172, 0.740	153.37	229.55, 77.18
4w	H	$(CH_2)_4F$	NCH_3	C=O	0.036	0.049, 0.023	>25.88	>262.86, 25.88
4x	H	$(CH_2)_4OH$	NCH_3	C=O	0.082	0.125, 0.040	48.64	45.57, 51.71
4y	NH_2CH_2	$(CH_2)_2CH(CH_3)_2$	NcC_3H_5	C=O	0.011	0.018, 0.005	8.58	4.38, 14.78
4z	CH_3NHCH_2	$(CH_2)_2CH(CH_3)_2$	NcC_3H_5	C=O	0.788	1.178, 0.399	17.90	23.52, 12.29
4aa	$nC_4H_9NHCH_2$	$(CH_2)_2CH(CH_3)_2$	NcC_3H_5	C=O	>190.8	>190.8, >190.8	4.15	1.66, 6.65
4ab	CO_2CH_3	$(CH_2)_2CH(CH_3)_2$	NcC_3H_5	C=O	>217.14	>217.14, >217.14	0.23	0.34, 0.12
4ac	CO_2H	$(CH_2)_2CH(CH_3)_2$	NcC_3H_5	C=O	>68.71	68.71, >207.23	102.21	138.42, 66.00
4ad	CH_2OH	$(CH_2)_2CH(CH_3)_2$	NcC_3H_5	C=O	0.314	0.423, 0.205	62.12	67.69, 56.56
4ae	H	$(CH_2)_4F$	NcC_3H_5	C=O	0.050	0.044, 0.057	38.15	27.71, 48.59
4af	NH_2CH_2	$(CH_2)_4F$	NcC_3H_5	C=O	0.009	0.016, 0.003	40.56	53.41, 27.52

4ag	H	$(CH_2)_4OH$	NcC_3H_5	C=O	0.025	0.033, 0.016	42.62	49.45, 35.79
4ah	NH_2CH_2	$(CH_2)_4OH$	NcC_3H_5	C=O	0.018 ± 0.008	*n* = 3	>181	>181, >181, >181
4ai	H	$(CH_2)_3CN$	NcC_3H_5	C=O	0.171	0.217, 0.126	9.05	11.80, 6.30
4aj	NH_2CH_2	$(CH_2)_4F$	NCH_2CF_3	C=O	0.008	0.010, 0.005	11.77	8.62, 14.92
4ak	NH_2CH_2	$(CH_2)_4OH$	NCH_2CF_3	C=O	0.008 ± 0.009	*n* = 3	52.68 ± 40.61	*n* = 3
4al	NH_2CH_2	$(CH_2)_3SO_2CH_3$	NCH_2CF_3	C=O	0.021	0.017, 0.025	>122. 68	122. 68, >156.85
4am	H	$(CH_2)_2CH(CH_3)_2$	NCH_2CO_2Et	C=O	1.855	2.405, 1.306	16.58	24.44, 8.72
4an	H	$(CH_2)_2CH(CH_3)_2$	NCH_2CO_2H	C=O	1.016	1.529, 0.503	>220.7	*n* = 1
4ao	H	$(CH_2)_2$(N-1-N_4CH)	NCH_2CO_2Et	C=O	0.288	0.372, 0.204	8.06	12.77, 3.35
4ap	H	$(CH_2)_2$(N-1-N_4CH)	NCH_2CO_2H	C=O	5.239	6.650, 3.629	213.49	213.49, 213.49
4aq	H	$(CH_2)_2CH(CH_3)_2$	CH_2Ph	C=O	1.807 ± 1.566	*n* = 3	20.09 ± 23.19	*n* = 3
4ar	H	$(CH_2)_2CH(CH_3)_2$	NCH_2-4-$C_6H_4CO_2Me$	C=O	0.260	0.413, 0.107	11.19	10.66, 11.72
4as	H	$(CH_2)_2CH(CH_3)_2$	NCH_2-4-$C_6H_4CO_2H$	C=O	0.009	0.005, 0.014	19.32	19.33, 19.31
4at	H	$(CH_2)_2$(N-1-N_4CH)	NCH_2-4-$C_6H_4CO_2Me$	C=O	0.170	0.176, 0.165	25.32	12.47, 37.28
4au	H	$(CH_2)_2$(N-1-N_4CH)	NCH_2-4-$C_6H_4CO_2H$	C=O	0.550	0.832, 0.269	54.70	85.10, 24.30
4av	H	$(CH_2)_2N(CH_3)_2$	NCH_2-4-$C_6H_4CO_2Me$	C=O	0.418	0.210, 0.626	3.43	1.413, 5.438
4aw	H	$(CH_2)_2N(CH_3)_2$	NCH_2-4-$C_6H_4CO_2H$	C=O	0.046 ± 0.044	*n* = 3	19.85 ± 22.40	*n* = 3
4ax	H	$(CH_2)_2CH(CH_3)_2$	CH_2	S	0.274	0.411, 0.137	6.78	9.81, 3.75
4ay	H	$(CH_2)_2CH(CH_3)_2$	CH_2	SO_2	0.059	0.082, 0.036	63.00	53.43, 72.58
4az	H	$(CH_2)_4CN$	CH_2	SO_2	1.340	1.629, 1.052	>270.0	270.0, >275.89
4aaa	H	$(CH_2)_4F$	CH_2	S	1.022	0.758, 1.287	>96.39	96.39, >270.66
4aab	H	$(CH_2)_4F$	CH_2	SO_2	0.229	0.284, 0.175	211.39	249.09, 173.69
4aac	H	$(CH_2)_4F$	$C(CH_3)_2$	S	>251.56	>251.56, >251.56	32.60	47.38, 17.820
4aad	H	$(CH_2)_4F$	$C(CH_3)_2$	SO_2	7.67 ± 12.15	*n* = 3	157.65 ± 110.48	*n* = 3
4aae	H	$(CH_2)_4OH$	NCH_3	SO_2	0.019 ± 0.011	*n* = 3	218.94 ± 20.40	*n* = 3
4aaf	H	$(CH_2)_3SO_2CH_3$	NCH_3	SO_2	5.73 ± 0.72	*n* = 3	34.51 ± 10.22	*n* = 3
4aag	H	$(CH_2)_4F$	NCH_3	SO_2	0.037 ± 0.056	*n* = 3	87.68 ± 50.73	*n* = 3
4aah	H	$(CH_2)_4F$	NcC_3H_5	SO_2	0.432 ± 0.340	*n* = 3	111.16 ± 60.30	*n* = 3
4aai	H	$(CH_2)_3CN$	NcC_3H_5	SO_2	0.055	*n* = 1	>244.22	*n* = 1
4aaj	NH_2CH_2	$(CH_2)_2CH(CH_3)_2$	NcC_3H_5	SO_2	0.003	0.003, 0.003	7.03	10.93, 3.14
4aak	H	$(CH_2)_2CH(CH_3)_2$	NCH_2CO_2t-tBu	SO_2	0.083	0.108, 0.059	18.36	6.12, 30.60
4aal	H	$(CH_2)_2CH(CH_3)_2$	NCH_2CO_2H	SO_2	0.263	0.150, 0.376	60.18	70.21, 150.15

[a] Values are means of two or more experiments performed on consecutive weeks with the data from individual experiments shown in the adjacent column.

antiviral activity was frequently negatively impacted by polar elements deployed in close proximity to the heterocycle.[2,3]

The series of quinolin-2-one derivatives surveyed by **4l**–**n** and **4q**–**v** are generally modest to poor RSV inhibitors, with activity largely insensitive to the oxidation state of the C–C bond or the pattern of substitution of the heterocycle ring. However, the introduction of an aminomethyl moiety at C-5 of the benzimidazole ring produced the highly potent RSV inhibitors **4o** and **4p**, with **4o** being 23-fold more potent than its unsubstituted congener **4n**. This result is consistent with an additional important and productive interaction between the basic amine and the RSV F_1 protein, findings aligned with earlier results in the benzimidazol-2-one series where this enhancing effect was attributed to the formation of a salt bridge with Asp200 in the N-terminal heptad repeat trimer pocket to which these compounds are believed to bind.[5,10] The antiviral potency associated with **4o** and **4p** clearly enhances the attractiveness of compounds in a series that in the absence of a basic C-5 substituent is rather unimpressive.

The quinazolin-2,4-dione series represented by **4w**–**aw** were explored in some detail based on the facility with which substituents could be introduced at the ring N-3 atom and because the C-4 carbonyl moiety more directly influences electron density in the fused phenyl ring than in the quinolin-2-one series **4l**–**m**. The methyl, cyclopropyl and trifluoroethyl quinazolinedione N-3 substituents were selected for examination based on the enhanced metabolic stability in human liver microsomes observed when these elements were introduced in the azabenzimidazol-2-one series.[4] Compounds **4w**–**x**, **4ae**, **4ag** and **4ai** are potent RSV inhibitors, with half-maximal inhibition of virus replication observed at concentrations below 100 nM. The acetic ester and acid derivatives **4am**–**ap** are considerably weaker antiviral agents, as might be anticipated based on prior SAR, but the simple *N*-benzyl compound **4aq** is a somewhat surprisingly poor antiviral agent. Interestingly, *para*-substitution with a carboxylic ester or acid moiety, to give **4ar** and **4as**, respectively, leads to enhanced potency, particularly marked for the acid **4as**. This trend shows some dependence on the polarity associated with the benzimidazole side chain since the tetrazoles **4at** and **4au** and dimethylamines **4av** and **4aw** are less impressive antiviral agents. Within the quinazolin-2,4-dione series, the incorporation of an aminomethyl substituent at C-5 of the benzimidazole heterocycle is uniformly associated with potent antiviral activity, as demonstrated by examples **4y**, **4af**, **4ah** and **4aj**–**l**. These results are consistent with an intimate interaction with Asp200 of the RSV F_1 protein, a hypothesis further supported by the poor activity associated with the substituted amines **4z** and **4aa** and the ester **4ab**, carboxylic acid **4ac** and alcohol **4ad**.[5,10]

The more powerful electron withdrawing sulfone moiety[11] was also examined in the context of benzothiazin-3-ones **4ax**–**aad** and benzothiadiazin-3-ones **4aae**–**aal**. The benzothiazine series provides a unique opportunity to directly compare the effect of an electron donating sulfide with an electron-withdrawing sulfone substituent within the context of an almost identical topology. The three sulfones examined, **4ay**, **4aab** and **4aad**, are at least fivefold more potent than the corresponding sulfides **4ax**, **4aaa** and **4aac**, respectively, suggestive of a positive effect of reducing electron density in the fused phenyl ring, a trend discernible in the comparison between the quinolin-2-ones **4l**–**v** and quinazolin-2,4-diones **4w**–**aw**. In the benzothiadiazine series, excellent antiviral activity is observed with the less polar benzimidazole side chains whilst the single C-5 aminomethyl derivative examined, **4aaj**, is the most potent representative of this chemotype. Antiviral activity is typically similar for identically substituted quinazoline diones and thiadiazine dioxides with a maximal variation of eightfold observed between examples **4af** and **4aah**.

A comparison of the SAR data for the fused 6,6 heterocyclic ring systems presented in Table 1 with that of similarly-substituted benzimidazol-2-one derivatives indicates that the silhouette presented by the fused 5,6-ring chemotype generally provides the more potent RSV inhibitors.[2–4] It is not immediately obvious from an inspection of models of representative examples of the 6,6-fused heterocycles docked in the proposed binding pocket why this structural variation should produce intrinsically less potent antiviral agents. A model of BMS-433771 (**1**) bound to the trimeric assembly of the RSV F_1 protein heptad repeats is depicted in Figure 1 and a proposed model of the binding of the quinazolin-2,4-dione **4ag** to the hydrophobic pocket is presented in Figure 2. The phenyl rings of the 5,6- and 6,6-fused systems can be superimposed with accommodations made by the heterocyclic moiety such that the substituent vectors project at different angles within the plane of the ring system. In the preferred binding pose, the heterocycle substituents distal to the site of attachment of the benzimidazolyl-2-methyl moiety project away from the protein, providing no apparently obvious explanation for the reduced potency. However, binding of these inhibitors to the trimeric RSV F_1 protein N terminal

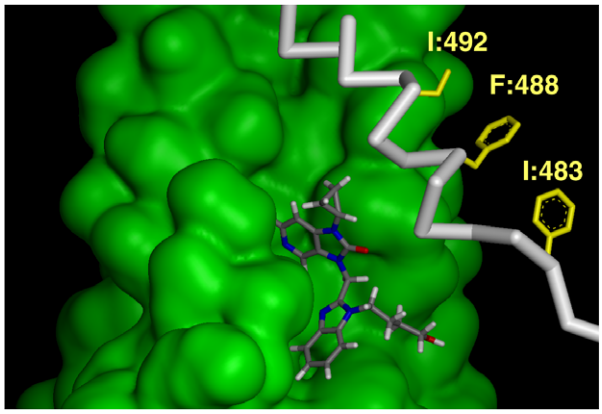


Figure 1. BMS-433771 (**1**) bound in the hydrophobic pocket created by the trimerization of the N-terminal heptad repeats of the RSV F_1 protein. Nitrogen atoms are depicted in blue, oxygen atoms in red, hydrogen atoms in white and carbon atoms in grey. The portion of the C-terminal heptad repeat that binds to the proposed small molecule binding pocket is also depicted.

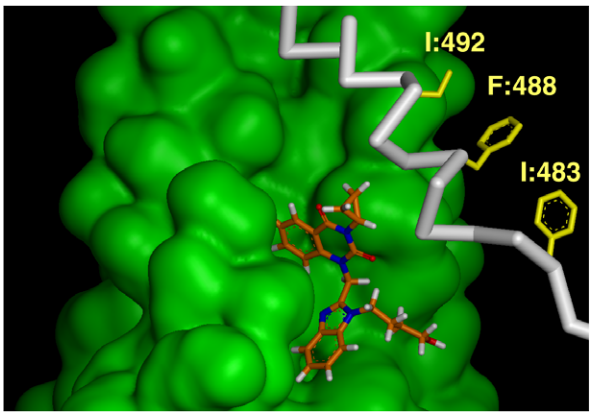


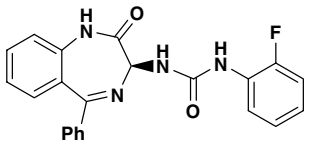
Figure 2. A depiction of compound **4ag** bound in the hydrophobic pocket created by the trimerization of the N-terminal heptad repeats of the RSV F_1 protein. Nitrogen, carbon and oxygen atoms are coloured as in Figure 1, whilst carbon atoms are depicted in bronze.

heptad repeat assembly may be a necessary but insufficient event for the expression of antiviral activity. Whilst there exists a reasonable understanding of the events leading to six-helix bundle formation in viruses that rely on a Type 1 fusion protein mechanism, the subsequent events leading to viral and host cell membrane coalescence remain enigmatic. There has been the suggestion that oligomerization of the 6-helix bundles occurs as part of the process leading to the formation of a fusion pore. Similarly, knowledge of the precise mode of action of this class of RSV fusion inhibitors is incomplete, particularly with respect to the interaction of the bound inhibitors with the C-terminus heptad repeat strand from which Phe483, Phe488 and Leu492 project. Effective RSV fusion inhibitors like BMS-433771 (**1**) may form a pseudo ternary complex with the amino and carboxy termini heptad repeats in a fashion that leads to a disruption of subsequent events that are critical to the execution of the membrane fusion process. Although subtle in nature, the altered substituent vector associated with the fused 6,6 ring systems may be such that interactions between the amino and carboxy termini are less effectively compromised, leading to the observed reduction in potency.

In earlier studies, the introduction of an aminomethyl substituent at C-5 of the benzimidazole ring provided potent antiviral agents that retained activity towards RSV expressing a K394R mutation in the F_1 protein, a virus that was isolated to be resistant to BMS-433771.[6] To probe this aspect of the antiviral profile, compounds **4o**, **4p**, **4af**, **4ah**, **4ai**–**al** and **4aaj** were evaluated against the K349R mutant virus. Only **4aaj** demonstrated potent inhibition, $EC_{50} = 20$ nM, whilst the EC_{50}s for the remainder of the compounds ranged from 0.5 μM (**4al**) to >5 μM (**4o**). These data reinforce the observation that the intrinsic potency of the fused 6,6-heterocyclic systems is less than that of the fused 5,6 ring silhouette probed extensively with the earlier compounds.[1–5]

Whilst the pharmacokinetic properties of this series of compounds were not subjected to the detailed scrutiny used to identify BMS-433771 (**1**) with a view to careful optimization for in vivo activity, several representatives were evaluated in the BALB/c mouse model of RSV infection.[7] Compounds **4af**, **4ah** and **4aj** all demonstrated excellent metabolic stability in human liver microsomes with half lives of >100 min, superior to the $T_{1/2}$ of 36 min reported for BMS-433771 (**1**).[4] In the mouse model, these 3 compounds, administered as single doses of 50 mpk 1 h prior to inoculation with virus, reduced RSV RNA in the lungs at 4 days post-inoculation by over 50 cycles using the RT-PCR methodology.[7] Virus titres measured by the $TCID_{50}$ method were reduced by 1.4 $\log_{10}$ after administration of **4af**, by 0.45 $\log_{10}$ for **4ah** and by 1.6 $\log_{10}$ for **4aj**.

Winter outbreaks of RSV in the United States continue to be a significant source of morbidity and mortality with 17,000 deaths annually attributed to this virus.[12,13] Individuals for whom an RSV infection poses a particularly difficult problem are infants with underlying cardio-pulmonary problems, those that are immunosuppressed and the elderly population.[14–17] However, there is an emerging appreciation that the morbidity and mortality associated with RSV infections may be underestimated, since infections are frequently misdiagnosed as influenza.[13,17,18] In order to prevent RSV infections, a humanized monoclonal antibody that is administered as a series of monthly intramuscular injections was licensed in 1998. However, this agent is expensive and is recommended only for those at high risk for complications. For those individuals with an established RSV infection, the non-specific antiviral agent ribavirin is the only available therapeutic option. This compound is administered by aerosol, presenting a hazard to nursing staff because of its teratogenic properties.[12–18] Whilst there has been considerable recent interest in identifying inhibitors of RSV as potential prophylactic and therapeutic agents, few of the compounds described in the literature possess properties suitable for oral dosing.[19] In addition to BMS-433771 (**1**), only the RSV N protein inhibitor A-60444 (**5**), which interacts synergistically with **1** in vitro, demonstrates oral bioavailability.[20,21]



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In summary, we have demonstrated that substitution of the azabenzimidazol-2-one heterocycle of BMS-433771 (**1**) by a range of alternative heterocyclic ring systems largely retains RSV fusion inhibitory activity, although the intrinsic potency associated with the 6,6-fused ring systems appears to be less than that for the fused 5,6-heterocycles. The introduction of an aminomethyl substituent at C-5 of the core benzimidazole ring leads, in all cases examined, to an increase in potency towards wild-type virus, confirming the value of this structural modification for potent RSV fusion inhibition.[5] Three of the compounds prepared as part of this survey, the quinazolin-2,4-diones **4af**, **4ah** and **4aj**, demonstrated antiviral activity in a mouse model of RSV infection.

References and notes

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