



## Novel benzothiophene H<sub>1</sub>-antihistamines for the treatment of insomnia

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### ABSTRACT

SAR of lead benzothiophene H<sub>1</sub>-antihistamine **2** was explored to identify backup candidates with suitable pharmacokinetic profiles for an insomnia program. Several potent and selective H<sub>1</sub>-antihistamines with a range of projected half-lives in humans were identified. Compound **16d** had a suitable human half-life as demonstrated in a human microdose study, but variability in pharmacokinetic profile, attributed to metabolic clearance, prevented further development of this compound. Compound **28b** demonstrated lower predicted clearance in preclinical studies, and may represent a more suitable backup compound.

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Histamine elicits effects on arousal through central action at the H<sub>1</sub> and H<sub>3</sub> receptor subtypes.<sup>1</sup> While first generation H<sub>1</sub>-antihistamines are sedating and are used in over-the-counter sleep aids, these agents are functional antagonists of muscarinic (M) receptors,<sup>2</sup> a property thought to cause undesirable side effects such as dry mouth, blurred vision, constipation, tachycardia, urinary retention, and memory deficits.<sup>3,4</sup> Next-day impairment, presumed to result from protracted CNS exposure and a consequence of long plasma half-life, has also been seen with these compounds.<sup>5,6</sup> Thus, selective H<sub>1</sub>-antihistamines with appropriate duration of CNS exposure may provide an alternative to current medications for the treatment of insomnia.

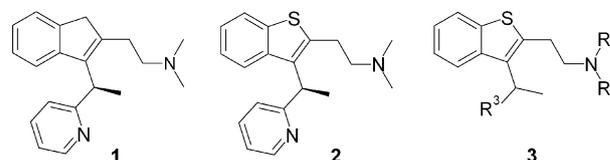
Previously, we described the use of the selective (–)-*R*-dimethindene (**1**)<sup>7</sup> as a starting point to generate novel series of selective and brain penetrating H<sub>1</sub>-antihistamines with pharmacokinetic profiles suitable for use as night time sleep aids.<sup>8</sup> The benzothiophene analog (**2**) was selected as a lead compound with a suitable receptor selectivity profile and, based on modeling of its clearance profile, a projected half-life in a desirable range of 5–18 h (Fig. 1).

In this Letter, we describe the SAR of benzothiophene series **3**. The objective of this study was to identify backup compounds with similar or improved selectivity to **2**, but with differences in clearance profiles and hence projected half-life.<sup>8</sup> Backup compounds

with a range of projected half-lives that model optimal plasma exposure over time as a function of dose were desirable to mitigate any risk of next day residual effects should they be observed in our lead.

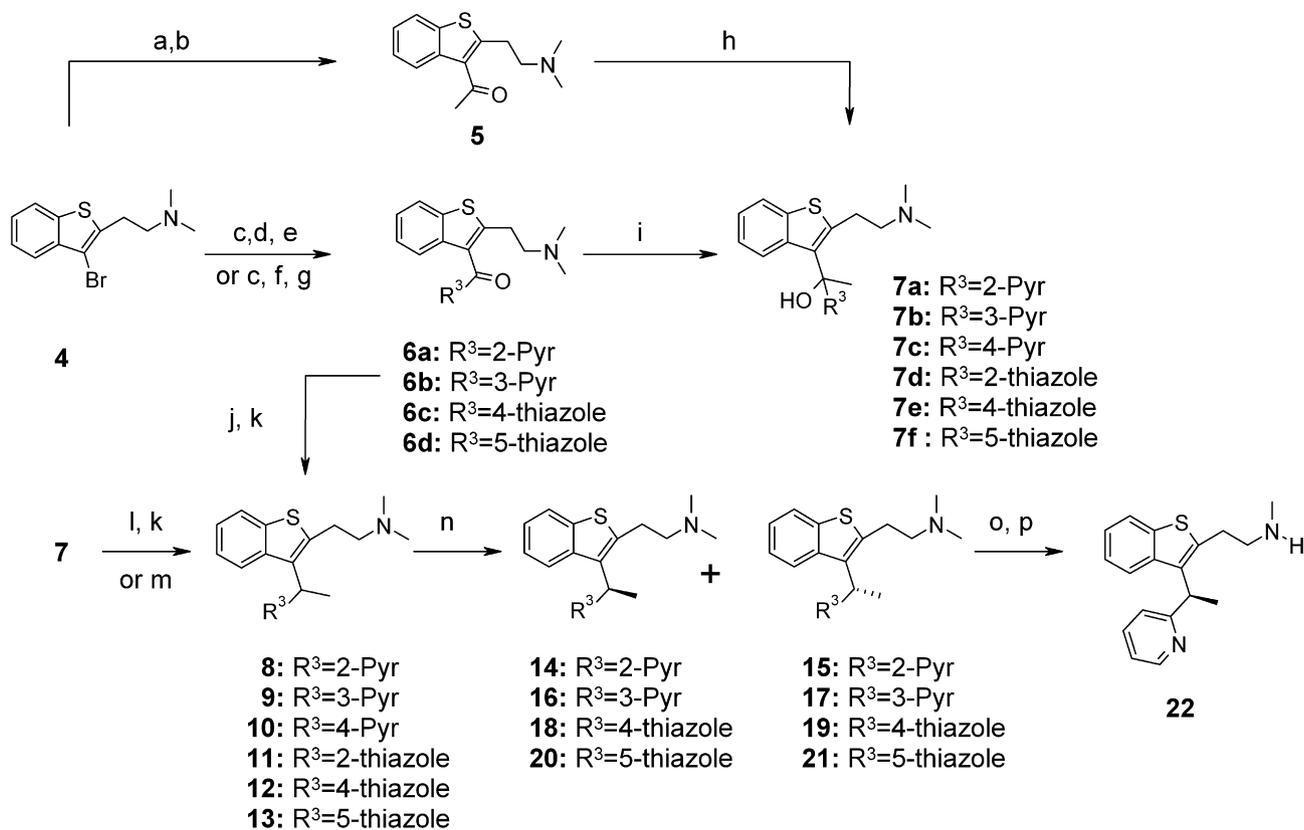
From our previous observations, a pyridyl or other heteroaryl was required for R<sup>3</sup> in **3** to confer selectivity versus cytochrome CYP2D6 inhibition.<sup>8</sup> We also demonstrated that variation of the heteroaryl provided subtle changes to clearance in preclinical models. The importance of the chiral center for binding affinity and selectivity in **3** had also been established.<sup>8,9</sup>

Initial efforts described in this Letter focused on subtle modifications of the pyridyl moiety (R<sup>3</sup>) and amine substituents (R<sup>1</sup>R<sup>2</sup>) in **3**. To achieve a selectivity profile comparable to **2**,<sup>8</sup> candidate compounds were required to demonstrate potent H<sub>1</sub> affinity (K<sub>i</sub> < 10 nM) and acceptable selectivity versus other receptor targets (M<sub>1</sub>, M<sub>3</sub>, H<sub>3</sub>, serotonin 5HT<sub>2A</sub>, >100-fold) and CYP2D6 and CYP3A4



**Figure 1.** (–)-*R*-Dimethindene (**1**), its direct benzothiophene analog (**2**) and series (**3**) described in this communication.

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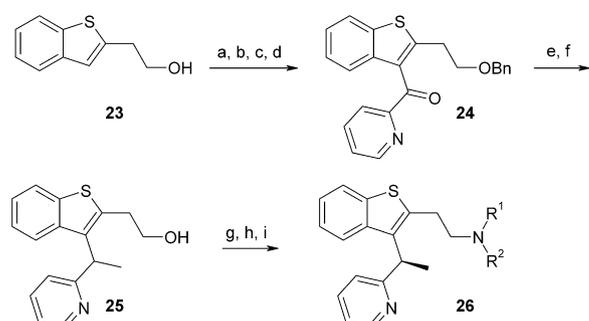


**Scheme 1.** Reagents and conditions: (a) CH<sub>3</sub>(CH<sub>2</sub>)<sub>3</sub>OCH=CH<sub>2</sub>, Pd(OAc)<sub>2</sub>, P(*o*-Tol)<sub>3</sub>, Et<sub>3</sub>N, CH<sub>3</sub>CN, 100 °C; (b) 10% HCl in THF; (c) *n*-BuLi, TMEDA, toluene or DCM, –78 °C; (d) R<sup>3</sup>CN; (e) 6 N HCl, MeOH; (f) R<sup>3</sup>CHO; (g) MnO<sub>2</sub>, DCM; (h) R<sup>3</sup>Br or R<sup>3</sup>H/ *n*-BuLi, –78 °C, DCM; (i) MeMgCl or MeLi, toluene, –78 °C; (j) PPh<sub>3</sub>MeBr, *t*-BuOK, THF; (k) Pd/C or Pd(OH)<sub>2</sub> in MeOH, H<sub>2</sub>; (l) H<sub>2</sub>SO<sub>4</sub> in TFA or MsOH in DCM; (m) NaI, TMSCl, CH<sub>3</sub>CN, reflux; (n) fractional crystallization, chiral HPLC or chiral SFC; (o) Cl(CO)OCH(CH<sub>3</sub>)Cl, DIPEA, DCE, 40 °C; (p) MeOH.

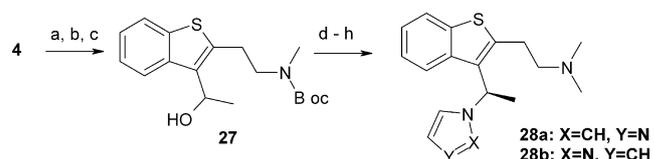
enzymes (>1000-fold). Since **2** lacked an *in vivo* cardiovascular risk,<sup>8</sup> despite weak hERG channel inhibition, we reasoned that compounds with hERG IC<sub>50</sub>/H<sub>1</sub> K<sub>i</sub> selectivity of 400 or greater would be acceptable for lead candidates. To minimize the risk associated with next day impairment, only H<sub>1</sub>-antihistamines with a projected half-life of 18 h or less were considered acceptable.<sup>10</sup>

Various synthetic schemes were employed to generate benzothiophenes **3** (Schemes 1–3) dependent on the availability of relevant starting materials. A Heck reaction of bromide **4**<sup>8</sup> with *n*-butyl vinyl ether, followed by hydrolysis, gave the acetyl intermediate **5**.<sup>11</sup> Addition of a lithium heteroaryl, generated *in situ* from the heteroarene or heteroaryl bromide, yielded alcohol **7**. Alternatively, lithium–halogen exchange of bromide **4**, followed by the addition of a heteroaryl nitrile and subsequent acidic hydrolysis, afforded the heteroaryl ketone **6**, which was converted to the alcohol **7** by addition of MeLi or MeMgBr. An alternative route to ketone **6** was developed involving lithium–halogen exchange of bromide **4**, followed by treatment with a heteroaryl aldehyde and a subsequent oxidation. Compound **7** was either dehydrated with a strong acid followed by hydrogenation, or directly deoxygenated with *in situ* TMSI to yield **8–13**.<sup>12</sup> Alternatively, ketone **6** was subjected to a Wittig reaction, followed by hydrogenation to yield **8, 9, 12** and **13**. Fractional crystallization or chiral chromatography (HPLC or SFC) of racemic **8–13** afforded the enantiomers **14–21**.<sup>13</sup> Demethylation of **14** to generate the secondary amine **22** was accomplished with 1-chloroethyl chloroformate followed by treatment with methanol. (See [Supplementary data](#) for specific synthetic steps to each final compound.)

Benzothiophenes, containing a cyclic amine, were generated through an alternative sequence (Scheme 2). Alcohol **23** was treated with bromine, protected with a benzyl group and subjected to a



**Scheme 2.** Reagents and conditions: (a) Br<sub>2</sub>, CHCl<sub>3</sub>; (b) NaH, BnBr, DMF, 0 °C; (c) *s*-BuLi, TMEDA, PyrCN, toluene, –78 °C; (d) 6 N HCl, MeOH; (e) MeLi or MeMgBr, toluene, –78 °C; (f) TMSCl, NaI, CH<sub>3</sub>CN, reflux; (g) MsCl, Et<sub>3</sub>N, DCM, 0 °C; (h) R<sup>1</sup>R<sup>2</sup>NH, IPA, 85 °C; (i) chiral HPLC.



**Scheme 3.** Reagents and conditions: (a) (i) Cl(CO)OCH(CH<sub>3</sub>)Cl, DIPEA, DCE, 40 °C; (ii) MeOH; (b) Boc<sub>2</sub>O, DCM; (c) *n*-BuLi, TMEDA, CH<sub>3</sub>C(O)H, toluene, –78 °C; (d) MsCl, DIPEA, DCM, 0 °C; (e) HetAryl/NaH, DMF, rt to 80 °C; (f) 50% TFA in DCM, 0 °C; (g) aq HC(=O)H, NaBH(OEt)<sub>3</sub>, THF; (h) Chiral HPLC or SFC.

lithium–halogen exchange. Reaction with 2-pyridinecarbonitrile followed by hydrolysis afforded the ketone intermediate **24**. After

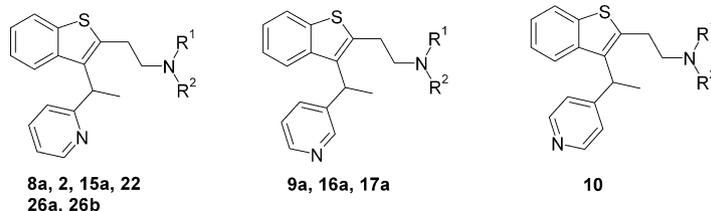
addition of MeLi, treatment with TMSCl/NaI resulted in both deoxygenation and removal of the protecting group to yield alcohol **25**. Conversion of the alcohol to the mesylate, displacement with cyclic amines followed by chiral HPLC, gave the desired (–)-*R* enantiomers **26**.

N-Linked heterocycle substituted benzothiophenes **28** (Scheme 3) were prepared from **4** by a sequence that involved a temporary conversion of –NMe<sub>2</sub> to –NMeBoc via demethylation and Boc protection. Subsequent lithium–halogen exchange and addition of acetaldehyde yielded **27**. This alcohol was converted to the mesylate and reacted with the sodium salts of different heterocycles. Removal of the Boc group and reinstallation of the Me moiety by reductive amination, followed by chiral HPLC gave the (–)-*R* enantiomers **28**. The NMeBoc group was required to prevent intramolecular cyclization of the mesylate intermediate. More efficient syntheses were developed for compounds **28** with promising in vitro profiles.<sup>14</sup>

Compounds were tested in an histamine H<sub>1</sub> receptor binding assay.<sup>8</sup> To confirm initial selectivity, compounds were assessed for M<sub>1</sub> receptor binding affinity<sup>8</sup> and inhibition of CYP2D6 and CYP3A4.<sup>8</sup> Selectivity over the hERG channel was evaluated using the high-throughput dofetilide binding assay.<sup>15</sup> Compounds of interest were subsequently tested in an hERG patch clamp functional assay.<sup>8</sup> (Tables 1–4).

Benzothiophene **2**,<sup>8</sup> was a high affinity H<sub>1</sub>-antihistamine with a good selectivity profile and moderate hERG activity. Similar to dimethindene,<sup>7</sup> the (+)-*S* enantiomer (**15a**) showed significantly lower affinity and selectivity for H<sub>1</sub> than its (–)-*R* enantiomer. This observation was consistent in all the enantiomeric pairs we studied. A tertiary amine was more potent and selective than a secondary amine as exemplified by comparison of **2** to **22**. Incorporation of cyclic amines at NR<sup>1</sup>R<sup>2</sup> position (**26a**, **26b**) yielded compounds with similar affinity to **2** and promising in vitro profiles albeit with a slight increase in hERG inhibition. Examination of the role of the nitrogen position in the pyridine indicated that the 3-pyridine (**16a**) had a very similar profile to **2** with marginally lower selectivity for the hERG channel. The racemic 4-pyridine (**10**) did not appear to offer an advantage with respect to H<sub>1</sub> affinity and selectivity, and therefore the enantiomers were not profiled.

**Table 1**  
Primary profile of unsubstituted pyridyl-benzothiophenes



| Compd      | Isomer                     | R <sup>1</sup> | R <sup>2</sup>                     | H <sub>1</sub> <sup>b</sup> K <sub>i</sub> (nM) | M <sub>1</sub> <sup>c</sup> K <sub>i</sub> (μM) | CYP2D6 <sup>d</sup> IC <sub>50</sub> (μM) | hERG <sup>e</sup> K <sub>i</sub> (μM) | hERG <sup>f</sup> IC <sub>50</sub> (μM) |
|------------|----------------------------|----------------|------------------------------------|---|---|---|---------------------------------------|---|
| <b>8a</b>  | <i>rac</i>                 | Me             | Me                                 | 12 ± 1.5  | 1.4   | 6.9                                       | >5                                    | NT <sup>g</sup>                         |
| <b>2</b>   | (–)- <i>R</i>              | Me             | Me                                 | 4.0 ± 0.5                                       | 5.3   | 28  | >5                                    | 1.4                                     |
| <b>15a</b> | (+)- <i>S</i>              | Me             | Me                                 | 256 ± 16  | 0.99  | 3.3                                       | >5                                    | NT                                      |
| <b>22</b>  | (–)- <i>R</i>              | H              | Me                                 | 32 ± 6  | 3.7   | 9.8                                       | >5                                    | 6.5                                     |
| <b>26a</b> | (–)- <i>R</i>              |                | –(CH <sub>2</sub> ) <sub>4</sub> – | 6.1 ± 1.2                                       | 5.8   | 2.7                                       | 1.1                                   | NT                                      |
| <b>26b</b> | (–)- <i>R</i>              |                | –(CH <sub>2</sub> ) <sub>3</sub> – | 3.9 ± 0.9                                       | >10   | >10                                       | 4.2                                   | 0.73                                    |
| <b>9a</b>  | <i>rac</i>                 | Me             | Me                                 | 10 ± 2  | 4.7   | 7.3                                       | >5                                    | NT                                      |
| <b>16a</b> | (–)- <i>S</i> <sup>a</sup> | Me             | Me                                 | 3.5 ± 0.6                                       | 28  | 5.2                                       | >5                                    | 0.49                                    |
| <b>17a</b> | (+)- <i>R</i> <sup>a</sup> | Me             | Me                                 | 79 ± 4  | >10   | 2.0                                       | >5                                    | NT                                      |
| <b>10</b>  | <i>rac</i>                 | Me             | Me                                 | 23 ± 2  | 5.6   | 0.85                                      | 1.8                                   | NT                                      |

<sup>a</sup> *R/S* nomenclature inverted.

<sup>b</sup> SEM for K<sub>i</sub> values derived from dose–response curves generated from triplicate or more data points.

<sup>c</sup> K<sub>i</sub> values derived from dose–response curves generated from duplicate data points.

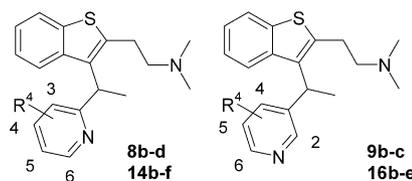
<sup>d</sup> CYP3A4 IC<sub>50</sub> >10 μM except for **16a** (IC<sub>50</sub> = 4.0 μM) and **10** (IC<sub>50</sub> = 2.8 μM).

<sup>e</sup> Dofetilide binding assay.

<sup>f</sup> Patch clamp analysis.

<sup>g</sup> NT = not tested.

**Table 2**  
SAR profiles of substituted 2- and 3-pyridyl-benzothiophenes



| Compd      | Isomer        | R <sup>4</sup> | H <sub>1</sub> <sup>a,b</sup> K <sub>i</sub> (nM) | CYP2D6 <sup>c</sup> IC <sub>50</sub> (μM) | hERG <sup>e</sup> IC <sub>50</sub> (μM) |
|------------|---------------|----------------|---|---|---|
| <b>8b</b>  | <i>rac</i>    | 4-Me           | 390 ± 15  | NT <sup>d</sup>                           | NT                                      |
| <b>8c</b>  | <i>rac</i>    | 6-OMe          | 59 ± 10   | NT  | NT                                      |
| <b>8d</b>  | <i>rac</i>    | 6-F            | 38 ± 4  | NT  | NT                                      |
| <b>14b</b> | (–)- <i>R</i> | 3-Me           | 9.3 ± 0.8   | 5.5                                       | 0.91                                    |
| <b>14c</b> | (–)- <i>R</i> | 3-OMe          | 5.0 ± 2.0   | 9.5                                       | 3.0                                     |
| <b>14d</b> | (–)- <i>R</i> | 3-F            | 2.7 ± 0.3   | 4.0                                       | 0.33                                    |
| <b>14e</b> | (–)- <i>R</i> | 3-Cl           | 8.0 ± 1.3   | 1.5                                       | NT                                      |
| <b>14f</b> | (–)- <i>R</i> | 5-F            | 13 ± 2  | >10                                       | 0.24                                    |
| <b>9b</b>  | <i>rac</i>    | 6-Me           | 69 ± 8  | NT  | NT                                      |
| <b>9c</b>  | <i>rac</i>    | 6-OMe          | 120 ± 15  | NT  | NT                                      |
| <b>16b</b> | (–)- <i>S</i> | 2-Me           | 17 ± 1  | >10                                       | NT                                      |
| <b>16c</b> | (–)- <i>S</i> | 2-OMe          | 2.9 ± 0.2   | 9.6                                       | 1.8                                     |
| <b>16d</b> | (–)- <i>S</i> | 2-F            | 1.7 ± 0.2   | 7.5                                       | 2.3                                     |
| <b>16e</b> | (–)- <i>S</i> | 2-Cl           | 2.9 ± 0.5   | 9.5                                       | 1.5                                     |

<sup>a</sup> SEM for K<sub>i</sub> values derived from dose–response curves generated from triplicate or more data points.

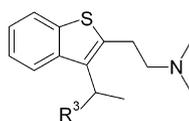
<sup>b</sup> M<sub>1</sub> K<sub>i</sub> > 10 μM for all compounds except for **14d** (K<sub>i</sub> = 2.2 μM).

<sup>c</sup> CYP3A4 IC<sub>50</sub> > 10 μM.

<sup>d</sup> NT = not tested.

<sup>e</sup> In dofetilide binding assay: hERG K<sub>i</sub> > 5 M for all compounds except for **14f** (K<sub>i</sub> = 2.5 μM).

Subsequent SAR focused on the substituent effect of Me, OMe, F, and Cl in both the 2-pyridyl and 3-pyridyl moieties. Initially, the racemates were assessed for H<sub>1</sub> binding and when affinity was sufficient (K<sub>i</sub> < 25 nM), the (–) enantiomers were isolated and profiled against H<sub>1</sub>, M<sub>1</sub>, CYP2D6, CYP3A4 and hERG (Table 2). These data indicated that substitution in the 4- (**8b**) or 6-position (**8c**, **8d**) of the 2-pyridyl moiety reduced H<sub>1</sub> binding. In contrast, substitution at the 3-position of the 2-pyridine was tolerated (K<sub>i</sub> of racemates < 20 nM). Of the enantiomers, the 3-F analog (**14d**) had highest H<sub>1</sub>

**Table 3**  
SAR profiles of pyridine isosteres

| Compd      | Isomer | R <sup>3</sup> | H <sub>1</sub> <sup>a</sup> K <sub>i</sub> (nM) | M <sub>1</sub> <sup>b</sup> K <sub>i</sub> (nM) | CYP2D6 <sup>c</sup> IC <sub>50</sub> (nM) | hERG <sup>d</sup> IC <sub>50</sub> (μM) |
|------------|--------|----------------|---|---|---|---|
| <b>11</b>  | rac    |                | 35  | 2072  | 856                                       | NT <sup>e</sup>                         |
| <b>18</b>  | (-)-R  |                | 4.6 ± 4   | 4759  | 4926                                      | 1.7                                     |
| <b>20</b>  | (-)-R  |                | 5.2 ± 2   | >10,000   | >10,000                                   | NT                                      |
| <b>28a</b> | rac    |                | 122   | NT  | 8012                                      | NT                                      |
| <b>28b</b> | (-)-R  |                | 4.4 ± 3   | >10,000   | >10,000                                   | 2.0                                     |

<sup>a</sup> SEM for K<sub>i</sub> values derived from dose–response curves generated from triplicate or more data points.

<sup>b</sup> K<sub>i</sub> values derived from dose response curves generated from duplicate data points.

<sup>c</sup> CYP3A4 IC<sub>50</sub> values >10 μM for all compounds, except for **28a** CYP3A4 IC<sub>50</sub> = 5.2 μM.

<sup>d</sup> In dofetilide binding assay: hERG K<sub>i</sub> >5 μM for all compounds.

<sup>e</sup> NT not tested.

**Table 4**  
hERG Selectivity and pharmacokinetic parameters for key compounds<sup>a</sup>

| Compd      | hERG IC <sub>50</sub> /H <sub>1</sub> K <sub>i</sub> | Pred hClS <sub>ys</sub> (ml/min/kg) | [P] <sup>b</sup> 4 h (ng/mL) | [B] <sup>b</sup> 4 h (ng/g) | B/P <sup>b</sup> 4 h | hClS <sub>ys</sub> <sup>c,e</sup> (ml/min/kg) | Human <sup>c</sup> AUC <sub>0-t</sub> (h ng/mL) |
|------------|--|-------------------------------------|------------------------------|-----------------------------|----------------------|---|---|
| <b>2</b>   | 339  | 9.7                                 | 2.8                          | 23                          | 8                    | 6.2 (5.8–7.2)                                 | 2.78 (1.90–4.23)                                |
| <b>16c</b> | 629  | 14.8                                | 37.5                         | 347                         | 9                    | NT <sup>d</sup>                               | NT  |
| <b>16d</b> | 1370   | 13.5                                | 3.0                          | 83                          | 26                   | 12.1 (4.6–18.5)                               | 0.64 (0.21–1.86)                                |
| <b>28b</b> | 443  | 6.0                                 | 0.6                          | 11                          | 20                   | NT  | NT  |

<sup>a</sup> All compounds highly permeable (Caco-2: P<sub>app</sub> > 21.7 × 10<sup>-6</sup> cm/s; A > B/B > A > 0.7), no evidence for pGP substrate.

<sup>b</sup> Rat 30 mpk oral.

<sup>c</sup> Microdose of 0.1 mg/kg; data are median values of four individual subjects; data in parentheses are the range of four subjects (for full details see Ref. 17).

<sup>d</sup> NT not tested.

<sup>e</sup> Calculated for an average weight of 60 kg.

affinity. However, hERG inhibition for this and the 5-F analog **14f** was increased compared to **2**. The 3-OMe analog (**14c**) had comparable H<sub>1</sub> affinity to **2** with a marginally improved selectivity versus hERG. These data along with the profile of methyl analog (**14b**) suggested an electronic effect at the 3-position on hERG affinity. In the case of the 3-pyridyl moiety, substitutions at the 6-position were not tolerated (**9b**, **9c**) whereas substitutions at the 2-position met the defined criteria for affinity. Interestingly, in the 3-pyridyl subseries the 2-OMe, 2-F and 2-Cl compounds (**16c**, **16d**, **16e**) all demonstrated improved in vitro profiles. H<sub>1</sub> affinities were comparable to **16a**, while selectivities versus hERG were significantly improved. In contrast, the 2-Me substituent (**16b**) resulted in an H<sub>1</sub>-antihistamine with reduced binding. Electronic effects were not apparent for this subseries, resulting in improved selectivity profiles for these compounds compared to the 2-pyridyl subseries.

Compounds **3** with pyridine isosteres (R<sup>3</sup>) were also investigated (Table 3). The racemates of either a 4- or 5-thiazolyl moiety were tolerated in the H<sub>1</sub> pharmacophore (K<sub>i</sub> <20 nM), whereas the 2-thiazolyl analog (**11**) had reduced binding. While introduction of a N-1 pyrazole moiety was acceptable, the corresponding imidazolyl modification (**28a**) negatively impacted H<sub>1</sub> binding. The primary profiles of the enantiomers for the active racemates (**18**, **20**, **28b**)

were comparable to **2**, with the pyrazole **28b** demonstrating acceptable selectivity versus hERG (hERG/ H<sub>1</sub> K<sub>i</sub> = 443).

Compounds with the best primary profiles (**16c**, **16d** and **28b**) were subsequently screened against other receptor off-targets (M<sub>3</sub>, H<sub>3</sub> and 5HT<sub>2A</sub>) and assessed for pharmacokinetic parameters (Table 4) to determine how they compared to **2**.<sup>8</sup> Compounds exhibited >1000-fold selectivity over these three off-target receptors, except **16c** (400-fold selective over 5HT<sub>2A</sub>) and **28b** (200-fold selective over H<sub>3</sub>). Predicted microsomal stability in humans (HLM assay<sup>8</sup>) indicated that both **16c** and **16d** showed higher clearance, whereas **28b** appeared more stable relative to **2**. Like benzothio-phenone **2**,<sup>8</sup> compounds **16c**, **16d** and **28b** were unstable in rat liver microsomes with projected clearances approaching hepatic blood flow rate (Pred. systemic clearance >67 ml/min/kg). Discrete oral PK studies of these compounds in rat at a relatively high dose (30 mpk) indicated that each of these compounds was brain-penetrating as judged by B/P ratios (4 h post dose) with brain levels sufficient to produce efficacy in a rat EEG/EMG model.<sup>16</sup> Subsequently, efficacy was demonstrated for **16c** and **16d** and found to be comparable to **2** (See Supplementary data).<sup>8</sup>

As a higher clearance alternative with lower projected half-life, **16d** was selected for comparison of pharmacokinetic profile to **2** in

a human microdose study.<sup>17</sup> While clearance projections from allometric scaling were reasonably predictive (predicted clearance: 13.5 mL/min/kg, measured mean clearance: 12.1 mL/min/kg), the measured human half-life for **16d** was longer than anticipated (13 h vs the estimate of 1.6 h predicted from allometry) and longer than for **2** (6.8 h). This discrepancy in half-life prediction was attributed to both an underestimate of the volume of distribution of **16d** ( $V_{ss}$  measured as 5.6 L/kg in humans<sup>16</sup> versus 1.7 L/kg from allometric scaling) and an overestimate of clearance. Even though a small sample size was used in the microdose study, **16d** showed an increasing trend to high variability in exposure compared to **2** following both iv (hClSys, Table 4) and oral dosing (AUC<sub>0-t</sub>, Table 4). PK variability in our leads was considered a significant issue because of the potential for varying exposures to adversely impact duration of action. Although a specific mechanism for the higher variability remains unclear, the higher clearance of **16d** compared to **2**, coupled with lower bioavailability (23% vs 47% for **2**) is consistent with mechanistic expectations that reduced metabolic stability generally contributes to increased variation in PK profile.<sup>17</sup> Based on these human PK data, neither compound **16d** nor **2** was pursued further. The more stable **28b** may therefore represent a more suitable backup to **2** and warrants further assessment.

In summary, discrete modifications around a lead benzothio-*phene* **2** provided several compounds with a comparable in vitro profile and a small improvement in hERG selectivity. From rat PK studies, compounds with a range of projected human half-lives were identified. Compound **16d** was chosen as a representative high clearance compound and assessed in a human microdose study. In contrast to allometric projections, this compound had a longer half-life in humans compared to **2**. Unfortunately, both compounds **2** and **16d** showed a trend to high PK variability indicating neither analog was suitable for further development. Compound **28b**, with improved metabolic stability, may represent the more promising backup candidate.

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### Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.bmcl.2010.01.134.

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