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Triazoloamides as potent γ-secretase modulators with reduced hERG liability

Christian Fischer *, Susan L. Zultanski, Hua Zhou, Joey L. Methot, Sanjiv Shah, Hugh Nuthall, Bethany L. Hughes, Nadja Smotrov, Armetta Hill, Alexander A. Szewczak, Christopher M. Moxham, Nathan Bays, Richard E. Middleton, Benito Munoz, Mark S. Shearman

Merck Research Laboratories Boston, 33 Avenue Louis Pasteur, Boston, MA 02115, USA

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ABSTRACT

Synthesis and SAR studies of novel aryl triazoles as gamma secretase modulators (GSMs) are presented in this communication. Starting from our aryl triazole leads, optimization studies were continued and the series progressed towards novel amides and lactams. Triazole **57** was identified as the most potent analog in this series, displaying single-digit nanomolar Aβ42 IC_{50} in cell-based assays and reduced affinity for the hERG channel.

Alzheimer's disease (AD) is a progressive neurodegenerative disorder which in 2010 affected over 5 million patients in the US alone.[1] To date, only symptomatic treatments are available, which provide short-term stabilization, but have no effect on overall disease progression. A popular hypothesis for AD describes oligomers of amyloid β (Aβ), and specifically the longer fragment Aβ42, as neurotoxic species that are formed by the γ-secretase complex through proteolytic cleavage of C-99.[2–5] Such oligomers have led to cell death and neurodegeneration in vitro and in vivo, causing symptoms such as cognitive impairment.[6]

In parallel to the development of γ-secretase inhibitors (GSIs), we and others have explored the possibility of modulating γ-cleavage to favor production of shorter fragments, while not affecting total Aβ levels. This was postulated to be a safer approach to a disease-modifying therapy due to the sparing of Notch processing.[7–9]

(*R*)-Flurbiprofen (Tarenflurbil, Flurizan™) (**1**, Fig. 1)[10] is an example of the first generation of carboxylic acid γ-secretase modulators (GSMs), and we have previously reported the discovery and SAR of several exquisitely selective (Aβ IC_{50} 40/42 > 100) classes of GSMs such as piperidine carboxylic acids **2**.[11] In the evolving area of non-acid GSMs, following the early work from Neurogenetics, Eisai disclosed aryl imidazoles, which are represented by E-2012 (**3**).[12] In our own program aimed at identifying novel non-carboxylic acids as modulators of γ-secretase, we previously published our lead optimization strategy for pyrimidine and purine derivatives (**4**) as GSMs.[13,14] Additionally, we detailed the discovery of novel non-acid GSMs such as quinazolinones[15] and, more recently, aryl triazoles (**5**).[16,17]

Following the discovery of triazole GSMs (**5**), we set out to identify novel triazoles, with the goal to improve their modest cell potency while maintaining their salient features, for example, >100-fold Notch selectivity and reduced hERG affinity. We hypothesized that replacing the benzylic substituents with amides would allow for a convergent optimization, while providing a reasonable starting point to optimize hERG binding (Fig. 2).

Initial studies aimed at examining whether the hydrophobic benzyl substituent in **5** could be replaced with amides without loss in potency. In our initial exploration, we focused on simple alkyl and aryl amides, which were prepared according to the general chemistry described in Scheme 1.

Aldehyde **8** was readily prepared from commercially available 4-fluoro-3-methoxybenzaldehyde (**7**), 4-methyl-1*H*-imidazole, and potassium carbonate as the inorganic base, by heating the reaction mixture overnight in DMF. Separation of the undesired regioisomer was achieved through a wash procedure and subsequent purification on silica gel.[18]

Alkynylation of aldehyde **8** with the Bestmann–Ohira reagent gave the desired alkyne **9** in quantitative yield. A click reaction[19] of alkyne **9** with azides **12** in ethanol/water, with copper(II)sulfate and sodium ascorbate as the reductant, furnished triazoles **13** in good to excellent yields. The required α-azido amides **12** were generally prepared from the corresponding α-halo amides **11** and sodium azide in polar aprotic solvents, such as DMF or DMSO, at room temperature. α-Halo amides were either commercially available or readily prepared from α-chloro acetylchloride (**10**) and the corresponding amines.

* Corresponding author.
E-mail address: christian_fischer@merck.com (C. Fischer).

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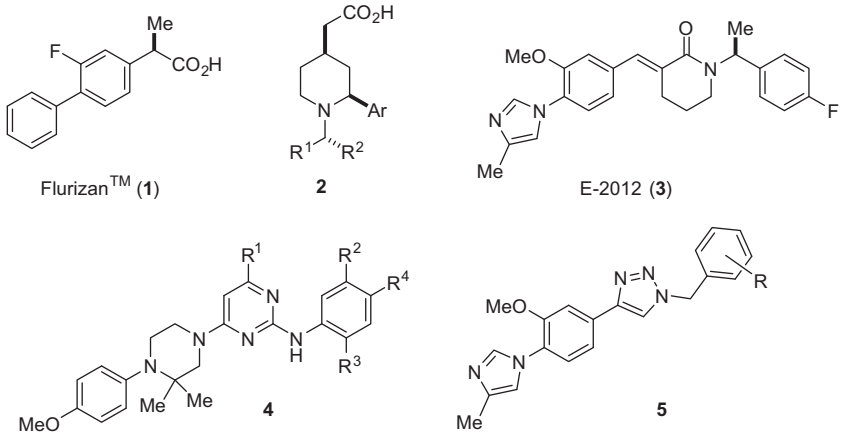


Figure 1. Select gamma secretase modulators.

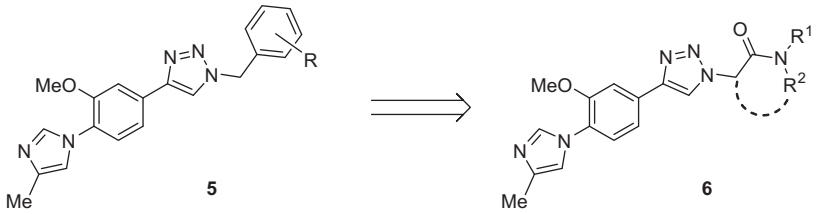
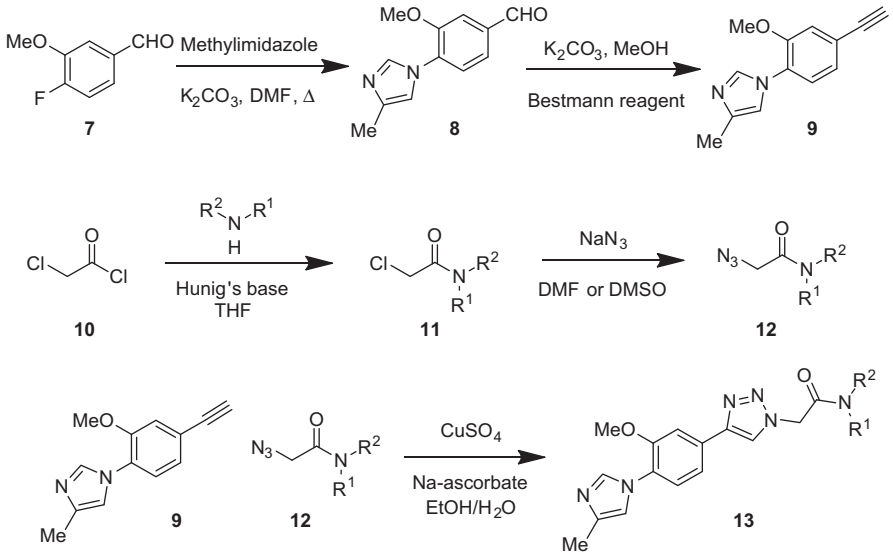


Figure 2. Aryl replacement strategy.



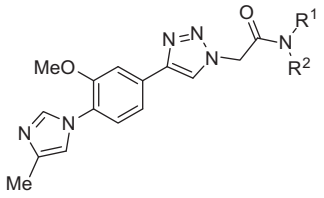
Scheme 1. Synthesis of amide GSMs **13**.

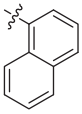
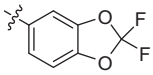
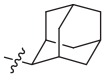
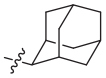
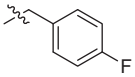
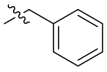
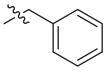
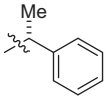
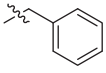
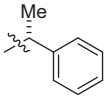
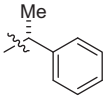
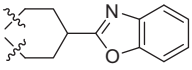
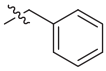
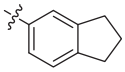
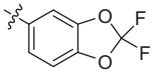
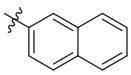
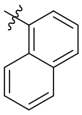
Cell biochemical data for the inhibition of Aβ40/42[20] production and Notch processing[21] as well as binding affinity for the hERG channel[22] for our initial set of amides **13** are reported in Table 1.

We were pleased to find no potency loss relative to our earlier benzylic triazoles,[17] as well as some clear SAR trends favoring bicyclic and cycloalkyl versus simple aryl substituents (**14** vs **16** and **23**). Also consistent with earlier studies, we found hERG binding to be correlated to the lipophilicity of the sidechains. In our earlier study on benzylic triazoles, we found that benzoxazole replacements resulted in attenuated hERG binding[17]; in attempting to translate this finding to the amide series, we identified compound **24**, which indeed displayed a modest improvement in hERG binding, albeit at a loss in potency. We decided to address the hERG liability of this series in a subsequent optimization where we focused on identifying optimal amide substituents. The trifluoroethyl group (compounds **25**–**29**) was identified as a preferred substituent, and we were able to drive the cell potency into the double-digit nanomolar range in combination with previously identified bicyclic pieces. Triazoles **28** and **29** showed the most promising on-target potency, with a >500-fold window over Notch processing. Hence, it was decided to invest further into the optimization of this new triazole series.

To explore the chemical space around the amide moiety and to address liabilities such as the affinity for the hERG channel, we

Table 1
Optimization of amide triazoles

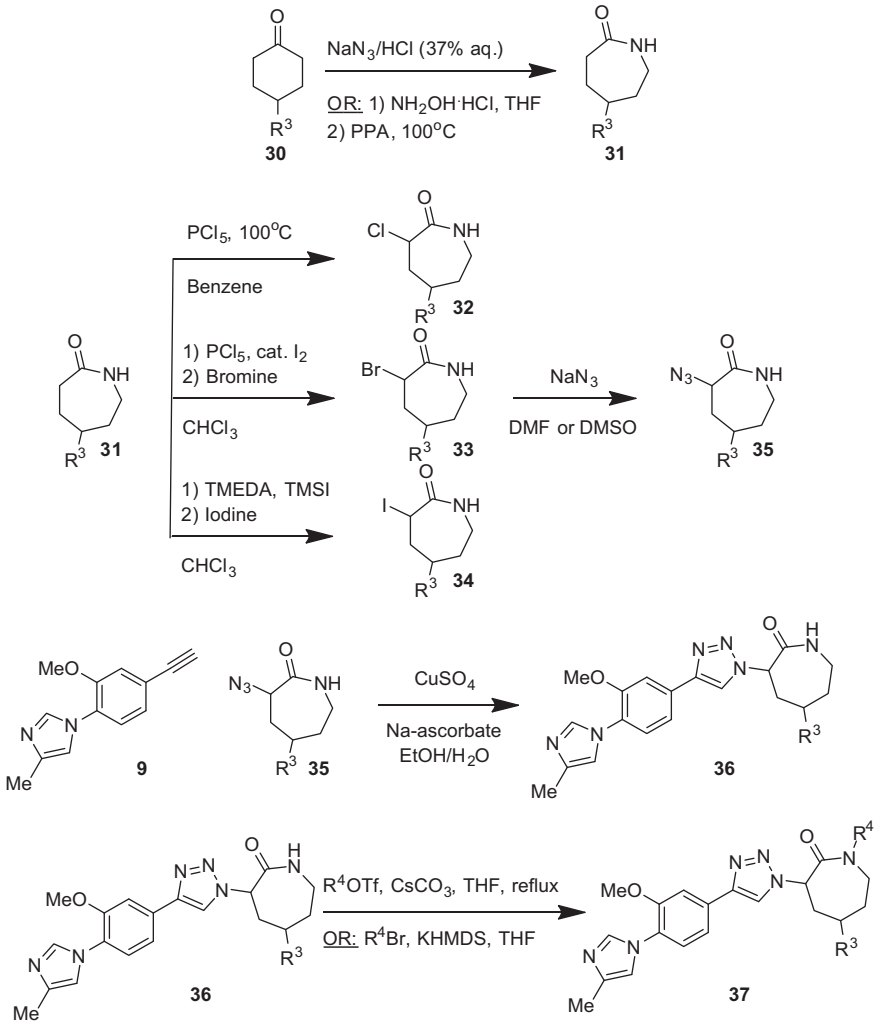


Compound	R^1	R^2	Aβ42 IC_{50}	Aβ40 IC_{50}	Notch IC_{50}	hERG IC_{50}
14			1.63	>10	n.d.	3.05
15			0.62	8.71	n.d.	0.80
16		H	0.61	4.85	26.7	>10
17		H	0.45	2.07	29.6	>10
18		H	0.68	>10	>50	4.03
19			0.24	1.90	>50	0.25
20			0.37	4.42	>50	0.87
21			0.23	3.01	>50	0.43
22			0.43	5.17	>50	0.84
23			0.20	>10	9.00	1.39
24			0.94	>10	>50	10.3
25			0.27	2.43	>40	2.87
26			0.18	1.25	24.6	1.49
27			0.16	1.18	38.2	1.92
28			0.08	0.66	48.5	0.63
29			0.09	0.41	45.1	0.80

All compounds were tested at least twice in independent experiments (at least once for hERG). For a description of assay conditions, see Refs. 21–23. All data is reported in μM.

began to explore constrained amides, and particularly, lactams. Synthesis of triazololactams proceeded as outlined in Scheme 2.

Owing to the facile click chemistry approach, we focused on an efficient synthesis of the corresponding azido lactams **35**, which were prepared from the corresponding chloro-, bromo-, or iodolactams **32**–**34**. α-Halogenation of lactams **31** was accomplished using the standard methods described in Scheme 2; lactams **31** were either commercially available or prepared using



Scheme 2. Synthesis of triazololactams.

conventional ring-expansion chemistry from commercially available cyclic ketones **30**. We found the Schmidt reaction to provide lactams in reproducibly high yields; alternatively a Beckmann rearrangement strategy was employed if the Schmidt reaction proved difficult due to solubility limitations. Regioisomeric mixtures, in the case of unsymmetrical ketones, were readily separable on silica gel. Installation of a second N-substituent on the lactam was generally accomplished through alkylation chemistry at any step in the sequence, but preferably as the final step.

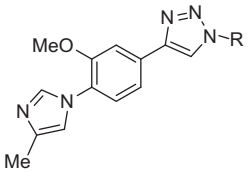
With robust chemistry in hand, we proceeded to explore a variety of lactams to evaluate the effect of ring size and substitution pattern to select the optimal scaffold for further optimization (Table 2).

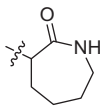
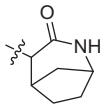
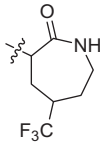
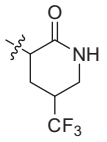
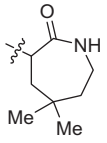
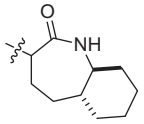
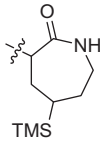
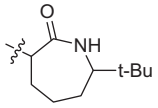
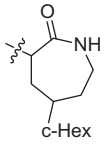
A survey of various ring sizes identified the piperidinone and azepinone rings as optimal (only selected examples are shown). Gratifyingly, we observed no drop in potency relative to the simple amide derivatives in Table 1, and we were delighted to find that binding to the hERG channel was generally attenuated, while excellent Notch selectivity was maintained. Additional substitution in the 5-position of the ring with hydrophobic substituents led to a consistent increase in intrinsic potency, while maintaining good Notch selectivity. Substitution on other positions on the ring was also tolerated, for example, in the 7-position (**45**) or 6,7-fused bicyclic rings (**43**). We proceeded to further optimize triazoloazepinones, but we also continued to test other ring sizes for key compounds. A systematic exploration of the lactam N-substituent as well as additional substitutions around the ring is reported in Table 3.

We initially embarked upon finding the optimal substituent on the lactam nitrogen, and found hydrophobic groups to be preferred with benzyl and trifluoroethyl being optimal. This was consistent with our initial observations with acyclic amides; polar functionality was not tolerated (data not shown).[23] To reassess the effect of substituents in the 5-position and the azepinone ring system relative to smaller ring size lactams, we prepared triazoles **54** and **55** and found them to be significantly less potent. For all compounds in this series, we saw a preference for one enantiomer, with the active enantiomer being 5- to 15-fold more potent than its less potent congener.[24] Ultimately, we identified triazole **57** to be the optimal combination in terms of potency and selectivity over Aβ40, Notch and hERG.

In summary, we have reported the discovery and SAR of novel amides and lactams as modulators of γ-secretase. Starting from simple amides as replacements of the benzylic groups in triazoles **5**, we were able to optimize potency while attenuating binding to the hERG channel. A novel series of potent and selective lactam GSMs was subsequently discovered, and through SAR studies within this series, we identified lactam **57**, which is one of the most potent GSMs we have discovered during our program. To the best of our knowledge, this is one of the first structural disclosures of GSMs with single-digit nanomolar cell activity. Although the

Table 2
Modulation of Aβ40/42 processing by lactams **38**–**46**

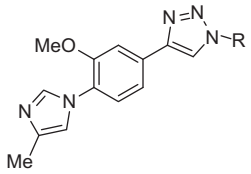


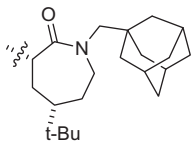
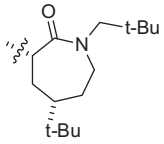
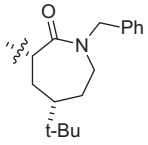
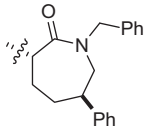
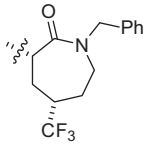
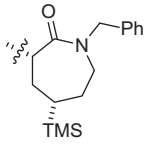
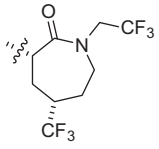
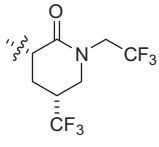
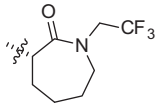
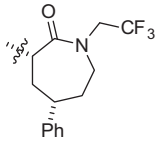
Compound	R	Aβ42 IC_{50}	Aβ40 IC_{50}	Notch IC_{50}	hERG IC_{50}
38		4.42	>10	>50	43.9
39		1.09	9.87	>50	5.35
40		1.27	9.10	>50	>30
41		1.11	6.16	>50	>30
42		1.31	7.42	>50	20.8
43		0.62	3.86	>50	20.8
44		0.33	1.23	45.3	6.27
45		0.48	5.12	>50	44.2
46		0.16	0.66	17.0	3.68

All compounds are racemic and were tested at least twice in independent experiments (at least once for hERG). For a description of assay conditions, see Refs. 20–22. All data is reported in μM.

selectivity of Aβ42 over Aβ40 was moderate (~5- to 25-fold), the compounds in this series behaved as GSMs, and the selectivity over Notch processing was generally very high (>100- to 1000-fold). Interestingly, the selectivity over Notch processing appears to be independent of Aβ 42/40 ratios and compound potencies; even for our most potent compounds, the selectivity over Notch processing was not diminished. Selected compounds in this series were profiled in vivo, and while we found the PK to be acceptable, the brain to plasma ratio was generally poor.[25] Future directions in this series that focus on improving central exposure while maintaining the intrinsic potency will be reported in due course.

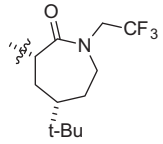
Table 3
Modulation of Aβ40/42 processing by triazoles **47**–**57**



Compound	R	Aβ42 IC_{50}	Aβ40 IC_{50}	Notch IC_{50}	hERG IC_{50}
47		0.24	1.33	>50	0.87
48		0.13	0.84	20.0	2.10
49		0.08	0.42	40.1	n.d.
50		0.08	0.51	11.1	0.20
51		0.08	0.42	40.8	5.48
52		0.07	0.44	10.2	40.8
53		0.02	0.21	>50	31.9
54		0.29	1.91	>50	13.1
55		0.30	2.21	>50	21.9
56		0.02	0.15	>50	2.22

(*continued on next page*)

Table 3 (*continued*)

Compound	R	Aβ42 IC_{50}	Aβ40 IC_{50}	Notch IC_{50}	hERG IC_{50}
57		0.009	0.17	24.3	5.4

All compounds are enantiopure (the more active enantiomer is shown) and were tested at least twice in independent experiments (at least once for hERG). For a description of assay conditions, see Refs. 20–22. All data is reported in μM.

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23. The fluoroethyl and difluoroethyl groups were found to be less potent (data not shown) while affording no improvements in terms of selectivity.
24. We were unable to obtain a single crystal X-ray for compounds in Table 3; however, for a structurally related compound which will be the subject of a forthcoming publication, the absolute stereochemistry was established as 3*S*.
25. Upon oral dosing of 50 mpk in SD rats or APP YAC mice, compounds in this series generally displayed brain to plasma ratios of <0.1, and consequently did not show significant brain exposure upon oral dosing. Owing to the low brain exposures, we were unable to observe consistent Aβ lowering at 50 mpk.