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Design and synthesis of cyclic sulfonamides and sulfamates as new calcium sensing receptor agonists

Lionel Kiefer [a], Tatiana Gorojankina [b], Philippe Dauban [a], Hélène Faure [b], Martial Ruat [b], Robert H. Dodd [a,*]

[a] Institut de Chimie des Substances Naturelles, UPR 2301, Centre National de la Recherche Scientifique, bât. 27, 1 av. de la Terrasse, F-91198 Gif-sur-Yvette, France
[b] Signal Transduction and Developmental Neuropharmacology Team, Laboratoire de Neurobiologie et Développement, Centre National de la Recherche Scientifique, Institut de Neurobiologie Alfred Fessard-IFR 2118, UPR 3294, F-91198 Gif-sur-Yvette, France

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ABSTRACT

The design, synthesis and calcimimetic properties of various cyclic sulfonamides and sulfamates are described. The latter were prepared from the corresponding *o*-alkenylarenesulfonamides via copper- or rhodium-catalyzed intramolecular aziridination. The size of the cyclic sulfonamide rings as well as the position of the crucial (*R*)-naphthylethylamine substituent significantly affected calcimimetic activity. The most active compounds were the six- and seven-membered sulfonamides **30a** and **31a** and sulfamate **34a**.

The extracellular calcium sensing receptor (CaSR), identified a little over a decade ago, is a G-protein coupled receptor (GPCR) which responds to small variations in extracellular calcium levels thereby maintaining calcium homeostasis in the organism.[1,2] Failure to maintain constant levels of extracellular ionized calcium $[Ca^{2+}]_{ex}$, generally in the 1.1–1.3 mM range, has been associated with several disorders such as hyperparathyroidism and osteoporosis.

In addition to the parathyroid glands, the CaSR has been identified in a wide range of tissues including kidney, intestine, and bone which are major contributors to systemic Ca^{2+} homeostasis as well as in cells not directly involved in this regulation process such as neurons and glial cells.[3,4] With respect to the parathyroid glands, low $[Ca^{2+}]_{ex}$ levels result in low activation of the CaSR inducing parathyroid hormone (PTH) secretion which then raises $[Ca^{2+}]_{ex}$.[2] Inversely, in response to high levels of $[Ca^{2+}]_{ex}$, the activated CaSR diminishes the secretion of PTH. Such an action can be mimicked by positive allosteric modulators of the CaSR, referred to as type II calcimimetics, useful for the treatment of primary and secondary hyperparathyroidism (HPT).[5] Only one such compound, cinacalcet **1** (Sensipar® in the USA, Mimpara® in Europe, Fig. 1) is presently commercialized for the treatment of secondary hyperparathyroidism in patients with end-stage renal disease (ESRD) and on maintenance dialysis therapy as well as for the reduction of hypercalcaemia in patients with primary HPT and parathyroid carcinoma.[6] This compound, a structural analogue of NPS-568 (**2**) one of the first synthetic small molecule ligand of the CaSR,[7,8] thus presents an alternative to parathyroidectomy for these disorders. The new treatment options offered by calcimimetics[9] are a strong motivation for the pursuit of new clinically useful drugs of this type.[10,11]

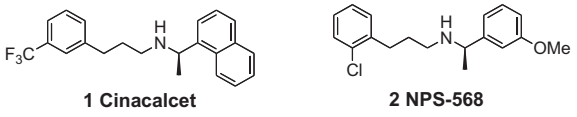

Figure 1. Positive allosteric modulators (calcimimetics) of the CaSR.

We have recently developed calindol (**3**, R = H) as a potent positive allosteric modulator of the CaSR.[12] Structurally, calindol is a rigid analog of the arenesulfonamide **4**, a novel calcimimetic family also first reported by us (Fig. 2).[13] Compound **4** can, in turn, be seen as an arenesulfonamide analogue of cinacalcet (**1**). While the in vivo activity of calindol is currently under investigation, this compound has already proven to be a valuable tool for the study of the mode of function of the CaSR[14] as well as of the GPRC6A receptor, another member of family 3 GPCRs related to the CaSR.[15] Calindol has also been used to demonstrate the presence of the CaSR in arterial endothelial cells and its possible role in the vascular complications associated with type 2 diabetes.[16–18] As part of an exploratory program aimed at the development of new potentially more active or selective calcimimetics, we decided to prepare

* Corresponding author. Tel.: +33 1 69 82 45 94; fax: +33 1 69 07 72 47.
E-mail address: dodd@icsn.cnrs-gif.fr (R.H. Dodd).

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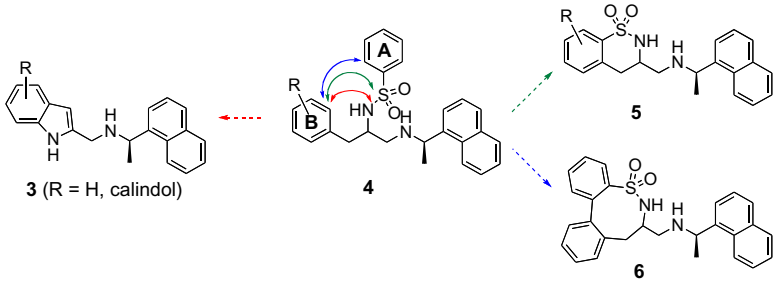


Figure 2. Calcimimetics developed by our groups (**3**, **4**) and targeted in this study (**5**, **6**).

alternative rigid analogs of **4** in which the sulfur atom of the sulfonamide group (instead of the nitrogen atom as in calindol) is now bound to phenyl ring B as in compound **5**. Alternatively, linking of the two A and B phenyl rings to give the biphenyl derivatives of type **6** was also studied. In this Letter, we describe the synthesis and in vitro calcimimetic activity of the prototypic cyclic sulfonamides **5** and **6** and of cyclic sulfamate analogs together with preliminary optimization studies in which, among other structural elements, the size of the sulfonamide (or sulfamate) ring and the position of the naphthylethylamine group were varied.

The key compounds were prepared by way of the one-pot copper(I) catalyzed intramolecular aziridination of unsaturated sulfonamides developed in our laboratory.[19] Thus, using this strategy, iodosylbenzene treatment of an arenesulfonamide bearing an olefinic side-chain of varying length at C2 (**7**) affords, in the presence of a copper(I) salt catalyst, the product of intramolecular aziridination (**8**). The latter should then be subject to nucleophilic attack by (*R*)-1-(1-naphthyl)ethylamine to give the desired cyclic sulfonamides (**9**) (Fig. 3).[20]

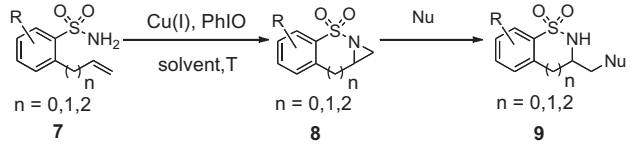
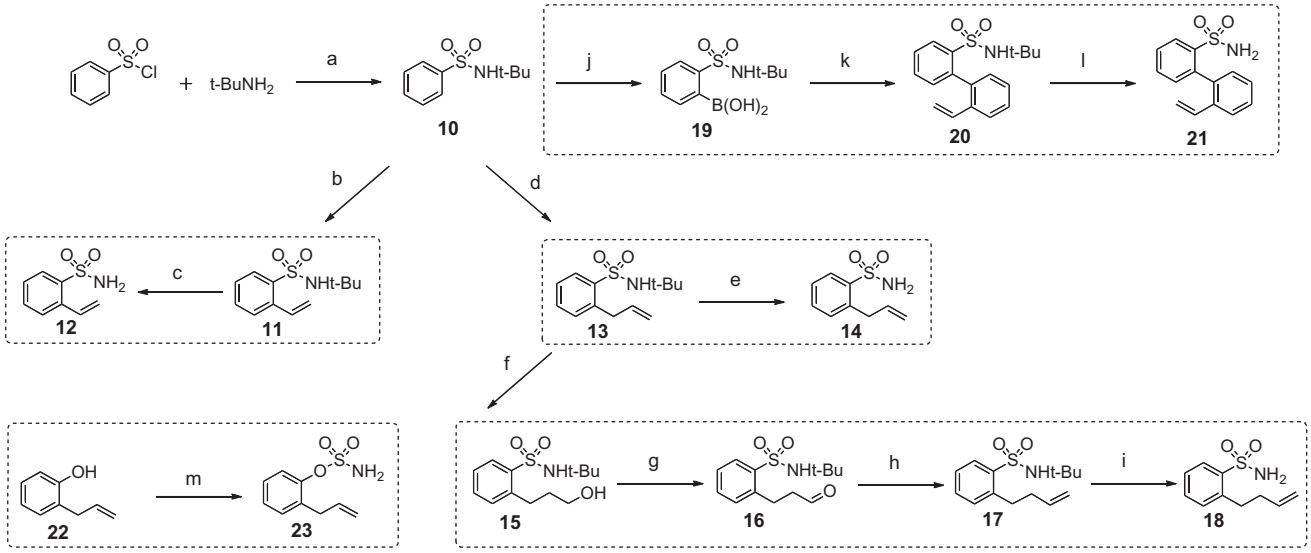


Figure 3. General procedure for the aziridination and ring-opening reaction to give cyclic sulfonamides.

We first set out to prepare the different olefinic arenesulfonamide precursors (Scheme 1). Thus, starting from *N*-(*t*-butyl)benzenesulfonamide **10**, prepared by reaction of benzenesulfonyl chloride with *t*-butylamine, the *o*-vinyl derivative **11** was obtained using the modified Schlosser procedure involving sequential additions of *n*-BuLi, DMF, Ph_3PCH_3Br, and *t*-BuONa.[21] The homolog of **11** was efficiently prepared by *ortho*-lithiation using *n*-butyllithium in the presence of TMEDA followed by reaction with allylbromide to give **13**. Further homologation was accomplished by subjecting propene derivative **13** to hydroboration followed by oxidation of the resulting primary alcohol **15** using TPAP to give aldehyde **16**. Wittig reaction between **16** and methyltriphenylphosphonium bromide in the presence of *n*-butyllithium then afforded the 3-butene derivative **17** though in moderate yield.[22] Two alternative structures were also employed for this study, the first being the biphenylsulfonamide **21**. This compound was prepared by treating *N*-*t*-butylphenylsulfonamide **10** with *n*-butyllithium and trimethylborate to give, after acid hydrolysis, the 2-boronic acid derivative **19**. Suzuki coupling between **19** and bromostyrene then furnished the 2-vinylbiphenyl derivative **20**. Trifluoroacetic acid—promoted deprotection of these unsaturated arenesulfonamides in the presence of anisole gave respectively **12**, **14**, **18**, and **21**.[23] Finally, the sulfonamide group of allyl



Scheme 1. Reagents and conditions: (a) (1) $CHCl_3$, 0 °C; (2) HCl, rt (quantitative); (b) (1) *n*-BuLi, THF, 0 °C; (2) DMF, 0 °C; (3) Ph_3PCH_3Br, 0 °C; (4) *t*-BuONa, rt (79%);[21] (c) TFA, anisole, 0 °C to rt (91%); (d) (1) *n*-BuLi, TMEDA, THF, 0 °C; (2) allylbromide, −60 °C to rt (70%); (e) TFA, anisole, 0 °C to rt (89%); (f) (1) 9-BBN, THF, reflux; (2) NaOH, HOOH, 50 °C (85%); (g) TPAP, NMO, MS, CH_3CN, rt (65%); (h) Ph_3PCH_3Br, *n*-BuLi, THF, −20 °C (19%);[22] (i) TFA, anisole, 0 °C to rt (85%); (j) (1) *n*-BuLi, TMEDA, THF, 0 °C; (2) trimethylborate, 0 °C; (3) HCl, rt; (k) $Pd(PPh_3)_4$, $NaHCO_3$, DME/H_2O, 2-bromostyrene, reflux (70% for two steps); (l) TFA, anisole, 0 °C to rt (20%);[23] (m) (1) HCO_2H, $ClSO_2NCO$, DMA, 0 °C; (2) H_2O, rt (80%).[25]

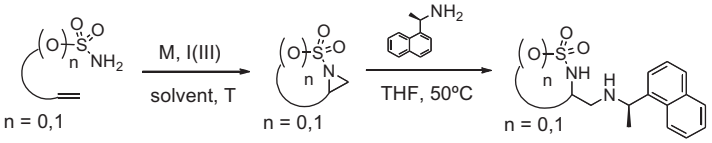
derivative **14** was replaced by a sulfamate group (i.e., compound **23**)[24] by treating 2-allylphenol **22** with chlorosulfonylisocyanate.[25]

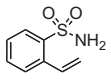
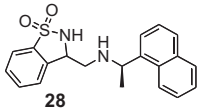
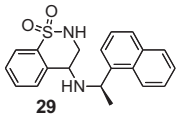
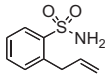
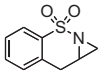
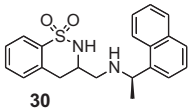
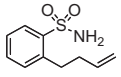
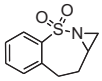
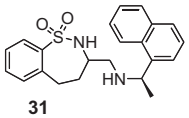
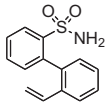
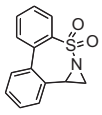
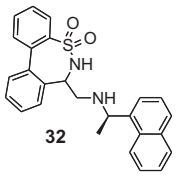
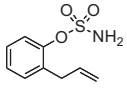
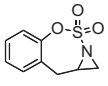
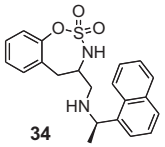
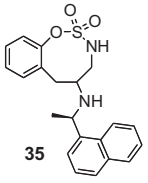
Transformation of the olefinic arenesulfonamides **12**, **14**, **18**, **21** and of sulfamate **23** to their corresponding aziridines was then effected (Table 1). Using our one-pot procedure (iodosylbenzene and catalytic $Cu(CH_3CN)_4PF_6$ in acetonitrile), compounds **14**, **18** and **21** gave the expected aziridines **25**, **26** and **27** in 65%, 31% and 70% yields, respectively. The relatively low yield of compound **26** is due to competitive formation of the product of allylic insertion.[20,26] Application of these reaction conditions to the vinyl derivative **12** proved unsuccessful. However, use of dirhodium tetraacetate as the catalyst together with iodosylbenzene diacetate afforded the desired aziridine **24** in 90% yield.[27] These same reaction conditions were used to promote aziridination of the sulfamate substrate **23**, affording compound **33** in 83% yield.

Previous SAR studies have shown that the biological activity of NPS-568 and of calindol is intimately related to the configuration of their methyl-bearing chiral centers. Thus, both (*R*)-NPS-568 and calindol are approximately 100 times more potent than their (*S*) counterparts.[12,8] The aziridines **24**–**27** and **33** were thus opened using (*R*)-naphthylethylamine as the nucleophile.

These ring-opening reactions, conducted in THF at 50 °C, were completely regioselective in the case of **25**, **26**, and **27**: the nucleophile attacks the less hindered carbon of the aziridine affording the corresponding ring-opened products **30**, **31**, and **32**, respectively, in good yields (Table 1). In the case of compound **24**,

Table 1
Intramolecular aziridination and nucleophilic ring-opening reactions



Sulfonamide	Aziridine	Yield[a] (%)	Product	Yield[a,b] (%)
 **12**	 **24**	90[c]	 **28**	50
			 **29**	26
 **14**	 **25**	65[d]	 **30**	85
 **18**	 **26**	31[d]	 **31**	68
 **21**	 **27**	70[d]	 **32**	86[f]
 **23**	 **33**	83[e]	 **34**	35[g]
			 **35**	35[g]

[a] Isolated yield after flash chromatography based on starting sulfonamides.
[b] 1:1 mixture of two diastereoisomers.
[c] Rhodium-catalyzed aziridination using $Rh_2(OAc)_4$, $PhI(OAc)_2$, Al_2O_3, CH_2Cl_2, 40 °C.[27]
[d] Copper-catalyzed aziridination using $Cu(CH_3CN)_4PF_6$, PhIO, CH_3CN, MS, rt.[16]
[e] Rhodium-catalyzed aziridination using $Rh_2(OAc)_4$, $PhI(OAc)_2$, MgO, CH_2Cl_2, −28 °C.[24]
[f] Reaction performed at 80 °C.
[g] Reaction performed at rt.

opening of the aziridine at the more substituted site was also observed to a minor degree, leading to a 2:1 mixture of five- and six-membered ring products **28** and **29**, respectively. While the two diastereomers of products **28**, **30**, and **31**, formed in equimolar amounts, could be easily separated by chromatography on silica gel, HPLC was required to separate the diastereomers of products **29** and **32**.

Reaction of the tricyclic *N*-sulfamate **33** with (*R*)-naphthylethylamine was also non-regioselective though in contrast to the reaction of the same nucleophile with **24**, a 1:1 ratio of regioisomers, that is, **34** and **35**, was obtained, HPLC being required to separate the diastereomers of **35**.

The hydrochloride salts of compounds **28a**,**b**–**32a**,**b**, **34a**,**b** and **35a**,**b** (where 'a' and 'b' designate the more and less lipophilic diastereomers, respectively) were then evaluated for agonist activity in Chinese hamster ovarian (CHO) cells stably expressing cloned calcium sensing receptor from rat brain (CHO(CaSR)) as previously described.[28] In these cells, Ca^{2+} as well as positive allosteric modulators (agonists) stimulate PLC activity resulting in accumulation of inositol phosphates (IP). The IP accumulation produced by the test compounds (10 μM) in the presence of 3 mM $[Ca^{2+}]_e$ was measured and compared to the effect produced by calindol.

As shown in Table 2, all arenesulfonamide and arenesulfamate derivatives demonstrated calcimimetic activity at 10 μM ranging from weak (**29**, **32**) to strong (**30**, **34**). The more lipophilic 'a' isomers were consistently more active than their less lipophilic 'b' counterparts and in some cases (e.g., **31a** vs **31b**; **35a** vs **35b**), this difference in activity was quite large. Since the absolute configurations of only compounds **29** and **32** have been determined,[29] no correlations can be made at this point regarding activity and stereochemistry. However, it may be noted that for compounds **29** and **32**, the ring substituents of the more active 'a' isomers both show the same spatial arrangement (Fig. 4).

Both diastereomers of the six-membered sulfonamide **30** were more active than those of the five-membered sulfonamide **28** (71% and 69% IP accumulation for **30a** and **30b** vs 57% and 39% for **28a** and **28b**) though less active than calindol (95% accumulation). Interestingly, the six-membered sulfonamides **29a**,**b**, in which the amine function of the side-chain is directly attached to the sulfonamide ring, both showed diminished activities (30% and 13% accumulation) compared to **28a**,**b** and **30a**,**b** in which the amine function is now linked to the sulfonamide ring via a methylene group. Finally, in this series, the seven-membered sulfonamide **31a** displayed almost identically high agonist activity (75% accumulation) as the six-membered sulfonamide **30a** (71% accumulation), its diastereomer **31b** being again significantly less active (37% accumulation). While the biphenyl cyclic sulfonamide derivatives **32a**,**b** were revealed to be among the least active CaSR agonists prepared (29% and 19% accumulation), the seven-membered sulfamate derivatives **34a** and **34b** were the most active, the former stimulating IP accumulation by 85% which compares favorably with the agonist effect of calindol (95% accumulation) at the same concentration. Finally, the eight-membered sulfamate derivative **35a** was also observed to be very active (67% accumulation) while its diastereomer **35b** was considerably less active (17% accumulation). Compound **35** thus displays the largest stereochemically determined activity difference of all the compounds studied.

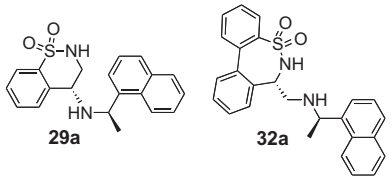


Figure 4. Absolute configurations of **29a** and **32a** as determined by X-ray crystallography.[29]

All the synthesized compounds except the less active ones **29** and **32** were also assayed for agonist activity on human CaSR (hCaSR) expressed in HEK293 cells.[30] As shown in Table 2 (column 2), the measured activities were in all cases remarkably similar to those determined in rat CaSR (rCaSR).

We have previously shown using point mutations of the hCaSR expressed in HEK293 cells that glutamate residue 837 (E837) was essential for efficient stimulation of IP accumulation by positive allosteric ligands such as NPS-568 and calindol.[31,32] Thus, as shown in Table 2 (column 3), mutation of this glutamate residue to alanine (E836A) results in only 41% IP accumulation produced by calindol (10 μM) compared to 95% accumulation in the WT receptor. Again, except for the less potent compounds **29** and **32**, the sulfonamides and sulfamates were evaluated for agonist activity on E837A mutated human CaSR. Interestingly, all tested compounds showed a major loss of activity as a result of its interaction with the mutated receptor. This suggests that the activity of all of the compounds studied depends on E837 and bind to the CaSR in a similar manner.

In conclusion, this report describes exploratory studies aimed at the discovery of new calcimimetic lead structures. The strategy used was based on imposing conformational restrictions to the first calcimimetic family discovered in our groups, the sulfonamides **4**, an approach previously used successfully by us for the development of calindol.[12] Among the cyclic sulfonamides prepared, the six- and seven-membered derivatives **30a** and **31a**, respectively, were shown to be the most potent calcimimetics. Conservation of the seven-membered ring of the latter but replacement of the sulfonamide function by a sulfamate led to an even more active compound, **34a**. Although the calcimimetic activities of **30a**, **31a** and **34a** are less robust than that of calindol, a wide variety of easily accessible structural modifications of these lead candidates can be envisioned which can be expected to result in higher activities. This study is currently underway. It has been recently proposed that NPS-568 is also a positive allosteric modulator of the GPRC6A, a receptor closely related to the CaSR, stimulated by L-α-amino

Table 2
Accumulation of IP induced by the test compounds in CHO cells expressing wild-type (WT) rat CaSR (rCaSR) and in HEK293 cells expressing WT human CaSR (hCaSR) or mutant (E837A) hCaSR

Compound[a] (10 μM)	IP accumulation[b] (% of Ca^{2+} 10 mM) Mean ± S.E.M.		
	rCaSR (WT)	hCaSR (WT)	hCaSR (E837A)
10 mM $[Ca^{2+}]_e$	100 ± 4	100 ± 3	100 ± 4
Calindol	95 ± 5	94 ± 4	41 ± 4
28a	57 ± 5	58 ± 3	11 ± 1
28b	39 ± 4	51 ± 4	11 ± 3
29a	30 ± 4	ND	ND
29b	13 ± 3	ND	ND
30a	71 ± 4	73 ± 3	22 ± 2
30b	69 ± 4	64 ± 2	10 ± 3
31a	75 ± 2	70 ± 3	3 ± 2
31b	37 ± 1	40 ± 3	1 ± 4
32a	29 ± 4	ND	ND
32b	19 ± 4	ND	ND
34a	85 ± 3	75 ± 3	51 ± 2
34b	76 ± 5	65 ± 3	27 ± 3
35a	67 ± 3	67 ± 3	1 ± 4
35b	17 ± 5	2 ± 2	1 ± 3

[a] 'a' and 'b' refer to the more and less lipophilic diastereomers, respectively.

[b] The IP accumulation produced by the test compounds in the presence of 3 mM $[Ca^{2+}]_e$ was measured and compared to the effect produced by calindol and by 10 mM $[Ca^{2+}]_e$ (a concentration resulting in maximal CaSR activation). Results are expressed as % of stimulation by Ca^{2+} 10 mM taken as 100%. See Ref. 30 for experimental details.

acids and modulated by calcium.[33] These data might suggest that other CaSR ligands could also modulate GPRC6A activation. Further work is needed for evaluating the specificity of new calcimimetics towards the CaSR and the GPRC6A, including those described in the present work.

Acknowledgments

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References and notes

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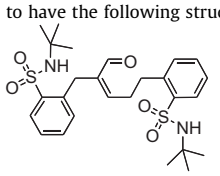
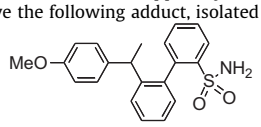
22. Under the strongly basic conditions of the Wittig reaction, compound **16** led to a 40% yield of the product of aldol condensation which, isolated and characterized, was shown to have the following structure:



23. The strongly acidic conditions required to cleave the *t*-butyl group of **20** also generated a vinyl cation which was trapped by the excess anisole in the reaction mixture to give the following adduct, isolated in 65% yield:



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