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Brief Article

Arylpyrrolizines as Inhibitors of Microsomal Prostaglandin E_2 Synthase-1 (mPGES-1) or as Dual Inhibitors of mPGES-1 and 5-Lipoxygenase (5-LOX)

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We synthesized and evaluated inhibitors for the microsomal prostaglandin E_2 synthase-1 (mPGES-1), based on the arylpyrrolizine scaffold. In a cell free mPGES-1 assay several "sulfonimides" exceeded our lead ML3000 (**3**) in potency. The most promising compound, the tolylsulfonimide **11f**, revealed an IC_{50} of 2.1 μM and is equipotent to the literature reference molecule MK886 (**1**). Selected compounds also potently reduced 5-LOX product formation in intact cells. Inhibition of isolated COX was occasionally remarkably cut down.

Introduction

Prostaglandin (PG[a]) E_2 is one of the most important and powerful prostanoids with diverse biological activity.[1] As a key cyclic lipid mediator derived from arachidonic acid (AA), it is involved in the development and perpetuation of inflammation seen in diseases such as rheumatoid arthritis (RA) and has been implicated in the development of peripheral and central sensitization during nociceptive processing (e.g., hyperalgesia, allodynia) and in tumorigenesis.[2−5] In the eicosanoid pathway (AA metabolism; see Supporting Information), induction of PGE_2 biosynthesis during inflammation requires the enzymatic actions of two cytokine-inducible enzymes: cyclooxygenase (COX) and prostaglandin E synthase (PGES). Inhibition of COX results in a reduced synthesis of prostaglandins (e.g., PGE_2) and thromboxanes (TXA_2) and is the basis for the anti-inflammatory efficacy and probably also for the analgesic activity of nonsteroidal anti-inflammatory drugs (NSAIDs). Owing to undesirable adverse effects of COX inhibitors (NSAIDs) and COX-2-selective inhibitors (COXIBs) in the gastrointestinal, renal, and cardiovascular systems, the selective inhibition of PGE_2-forming enzymes downstream of COX, such as the inducible glutathione-dependent microsomal PGE_2 synthase-1 (mPGES-1), has recently been proposed as a more promising approach for development of drugs for anti-inflammatory and pain therapy.[3−6] mPGES-1 is a member of the MAPEG family, including FLAP, and is the key terminal enzyme in pathology related production of PGE_2 from COX-2 derived PGH_2. The structure of human mPGES-1 has very recently been determined in complex with glutathione (GSH) by electron crystallography from 2D crystals in the presence of phospholipids.[7]

A few synthetic compounds have been reported to act as inhibitors of mPGES-1 activity, but none of these inhibitors showed high activity or specificity of action against mPGES-1 in vivo.[6] The potent FLAP indole inhibitor MK-886[8] (**1**) (Figure 1), an anticancer drug that can more efficiently bind with at least one other protein (FLAP IC_{50} = 26 nM), functions as a moderate inhibitor of rat (IC_{50} = 3.2 μM) and human mPGES-1 (IC_{50} = 1.6 μM) and has been used as a lead structure to develop more effective inhibitors. Some of the indole analogues of **1** have promising higher activity for the inducible human mPGES-1 membrane protein, with the lowest IC_{50} of 3 nM.[8] Optimized compounds in this series are represented by the "biaryl derivatives" exemplified by Merck Frosst compound **23**[8] (**2a**) or **30**[8] (**2b**) and their analogues (Figure 1). However, the latter were less potent in cellular systems and lacked inhibitory properties in whole blood because of strong binding to serum proteins. As of yet, no effective in vivo inhibitor has been reported in the literature.[6] Therefore, the design and discovery of novel mPGES-1 specific inhibitors with different scaffolds are highly desirable.

We considered the similarities between ML3000 (licofelone)[9] (**3**), a well-known dual COX/5-LOX inhibitor (third generation NSAID, Figure 1), and **1** in terms of structural conformation and mechanism of inhibition of leukotriene synthesis. Werz and co-workers very recently also addressed the mode of action of **3** in the suppression of PGE_2 formation.[1] Their data indicated that **3** appears to suppress inflammatory PGE_2 formation preferentially by inhibiting mPGES-1 at concentrations that do not affect COX-2 (mPGES-1 IC_{50} = 6 μM), implying attractive and thus far unique molecular pharmacological dynamics as an inhibitor of COX-1, 5-LOX, and mPGES-1. Furthermore, **3** is ~60-fold more potent in the cellular assay than in cell-free systems regarding the reduction of PGE_2 formation and thus more than 100-fold more potent than **1**.[1]

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[a]Abbreviations: COX, cyclooxygenase; CDI, carbonyldiimidazole; MAPEG, membrane-associated proteins involved in eicosanoid and glutathione metabolism; FLAP, 5-lipoxygenase activating protein; mPGES, microsomal prostaglandin E_2 synthase; PG, prostaglandin; NSAID, nonsteroidal anti-inflammatory drug; COXIB, COX-2-selective inhibitor; prostanoid, a subclass of eicosanoids consisting of prostaglandins, thromboxanes, and prostacyclins; PMNL, polymorphonuclear leukocytes; 12-HHT, 12(*S*)-hydroxy-5-*cis*-8,10-*trans*-heptadecatrienoic acid; LTB_4, leukotriene B_4; LOX, lipoxygenase.

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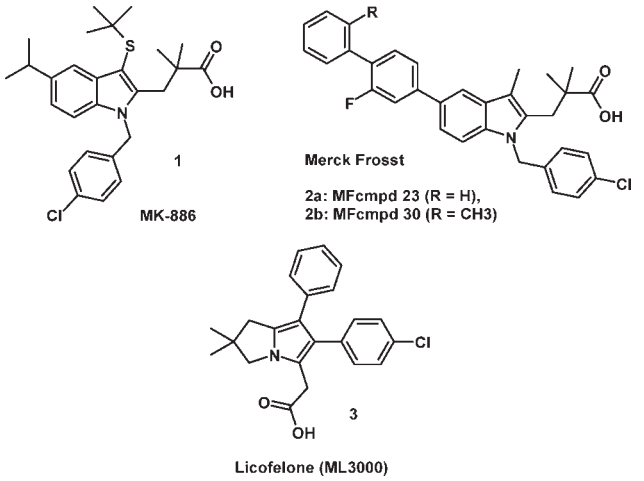
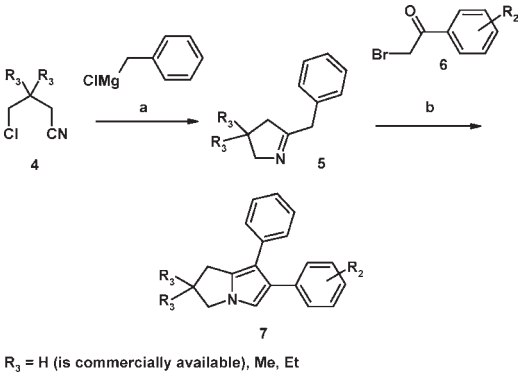


Figure 1. Structures of anti-inflammatory drugs and literature mPEGs-1 inhibitors.

Scheme 1. Construction of the Pyrrolizine Scaffold[a]



[a] Reagents and conditions: (a) benzylmagnesium chloride (Grignard species provided in situ from benzylchloride and Mg 1:1), initially absolute Et_2O, 2 h, reflux, then toluene, 3 h, reflux, 23−30%, depending on the further workup (R_3 = H, 89%);[11,12] (b) ω-bromoacetophenone, absolute EtOH, 24 h, room temp, then addition of saturated $NaHCO_3$, 24 h, room temp, 42−57%.

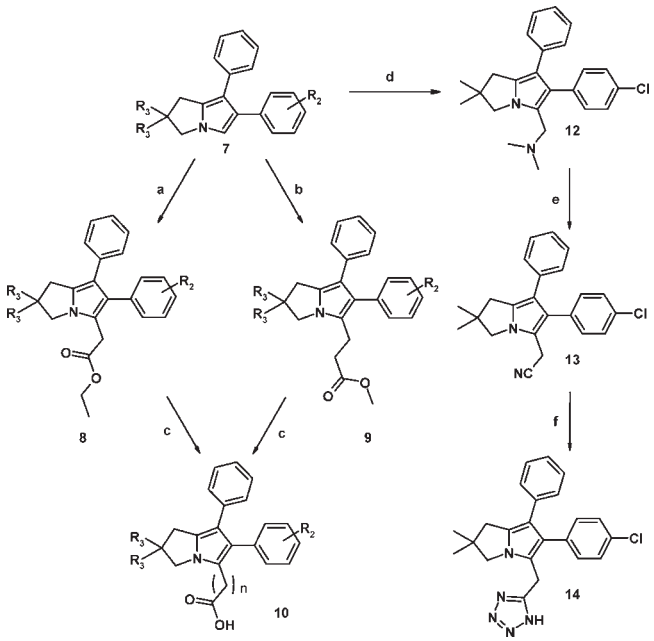
In the present study, to improve the activity profile and at the same time reduce undesirable side effects, we prepared several compounds based on the lead structure of **3** that were modified at the acid group.[10,11] After evaluation of existing compounds and their IC_{50} values against mPGES-1, both isoforms of the COX enzyme, and 5-LOX, we devised a concrete derivatization strategy to optimize the mPGES-1 inhibition potency of these molecules while at the same time reducing or canceling the COX activity. Ideally, the 5-LOX inhibitory activity should also be retained.[9]

Chemistry

Arylpyrrolizines are recognized scaffolds for (dual) COX/LOX inhibitors (e.g., **3**, Figure 1), and this compound class was recently found to comprise putative inhibitors of the mPGES-1 as well.[1] A general synthetic strategy is briefly summarized in Schemes 1−3.

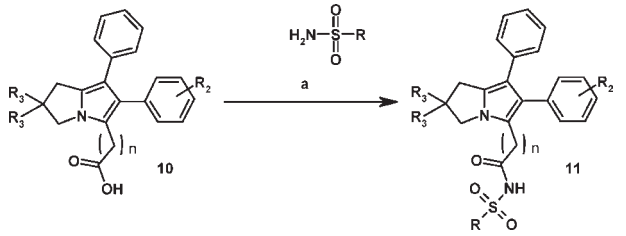
All bioactive target compounds have a pyrrolizine ring system in common whose synthesis typically is based on a differently substituted 4-chlorobutannitrile **4**[11,12] (Scheme 1).

Scheme 2. Synthetic Routes to the Target Compounds[a]



[a] Reagents and conditions: (a) diazoacetic acid ester/Cu^0 (add over 1 h), toluene, reflux (110 °C), 15 min, 39%; (b) acrylic acid ester/BF_3, dichloro ethane, room temp, 1 h, 50−70%; (c) KOH, ethanol, reflux, 5 min to 1 h, 50−75%; (d) $CH_2{=}N^+(CH_3)_2Cl^-$, absolute CH_2Cl_2; (e) CH_3I, NaCN; (f) NaN_3.

Scheme 3. Derivatization of the Acid Functionalities to the Desired Sulfonylimides[a]



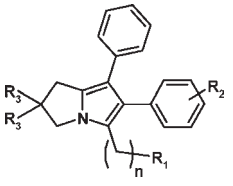
[a] Reagents and conditions: (a) (1) **10** in absolute THF, CDI, 2 h, room temp (mixture A); (2) substituted sulfonylamide in absolute THF, argon, NaH, 1 h, room temp (mixture B); (3) combining mixtures A and B under argon, 40 h, room temp, 39−46%.

Cyclization and simultaneous incorporation of the first aromatic substituent are carried out by Grignard reaction using benzyl chloride. In a final step both, the obtained 5-benzyl-3,3-dialkyl-3,4-dihydro-2*H*-pyrrole and the 5-benzyl-3,4-dihydro-2*H*-pyrrole (**5**)[10] are ring-closed to the favored 2,3-dihydro-1*H*-pyrrolizine derivative **7** with (substituted) ω-bromoacetophenone **6**.[10] The latter reaction step is carried out in an aqueous/ethanolic sodium bicarbonate solution at room temperature (40 h) (Scheme 1). Implementation of the obligatory acid group can be conducted by two different methods depending on the preferred alkyl anchor (spacer) using diazo ethyl acetate and copper in boiling toluol (affording **8**)[12] or acryl acid methyl ester under boron trifluoride etherate catalysis in absolute dichloroethane at ambient temperature (affording **9**).[13] Alternatively, at this stage Friedel−Crafts alkylation may be employed to achieve **9** from **8** and 3-bromopropionic acid ethyl ester using aluminum chloride as catalyst (Scheme 2).

The free carbonic acids **10a**−**g**[10,12−14] (Table 1) arise from these esters by alkaline hydrolysis with 10% potassium hydroxide. Further derivatization of the free alkyl acids to the desired sulfonylimides **11a**−**f**[10,11,15] (Scheme 3) is carried out by the initial activation of the acid function with CDI in dry THF (room temp, 2 h) and subsequent reaction of these intermediates with deprotonated sulfonamides (dry THF, room temp, 40 h).

Biosteric replacement of a carboxyl group by tetrazole could also be a useful method to optimize the lead structure (**3**). Tetrazole **14** was synthesized by converting 6-(4-chlorophenyl)-2,2-dimethyl-7-phenyl-2,3-dihydro-1*H*-pyrrolizine **7** into the dimethylaminomethyl derivative **12**[16] using a mixture of formaldehyde/dimethylaminohydrochloride. Activation by quarternization (methyl iodide) with sodium cyanide finally afforded the corresponding nitrile **13**, which was then converted to the tetrazole **14** using sodium azide.

Table 1. Substitution Patterns of the Synthesized Compounds



test compd	R_1	R_2	R_3	*n*
3	COOH	4-Cl	Me	1
10a	COOH	4-Cl	H	1
10b	COOH	4-Cl	Me	2
10c	COOH	4-Cl	H	2
10d	COOH	4-NO_2	Me	1
10e	COOH	4-CF_3	Me	1
10f	COOH	benzofuran-6-yl[a]	Me	1
10g	COOH	4-*tert*-butyl	Me	1
10h	COOH	4-*tert*-butyl	Me	2
11a	CO-NH-SO_2-CH_3	4-Cl	Me	1
11b	CO-NH-SO_2-CH_3	4-Cl	Et	1
11c	CO-NH-SO_2-Ph	4-Cl	Me	1
11d	CO-NH-SO_2-Tol	4-Cl	Me	1
11e	CO-NH−SO_2−Ph	4-Cl	Me	2
11f	CO-NH−SO_2-Tol	4-Cl	Me	2
14	1*H*-tetrazol-5-yl	4-Cl	Me	1

[a] Instead of the otherwise used phenyl substructure.

Biological Testing

All compounds were screened in a cell free mPGES-1 assay at four distinct concentrations between 10 μM and 1 nM, depending on the estimated intrinsic activity.[1] PGE_2 synthase activity was determined in microsomes of A549-cells. Test results are expressed as a percentage of the PGE_2 level measured in the presence of the vehicle control. Compounds **1** and **2a** (Table 2) were used as internal standards. Furthermore, when mPGES-1 IC_{50} values were $\leq$10 μM, the compounds were investigated for their activity on isolated COX-1/COX-2 and were tested at 30 μM.[1] In these cases, data are presented as the percentage of the concentration of 12-HHT in the vehicle control. Inhibition of 5-LOX was determined in PMNL (and 5-LOX) intact cell assay at inhibitor concentrations between 10 μM and 0.1 μM. LTB_4 was measured by an HPLC method.[17]

Discussion

We recently reported that the arylpyrrolizine carboxylic acid **3** has good intrinsic activity against mPGES-1 and 5-LOX.[1,17] By use of **3** (Figure 1) as a lead structure, structure−activity relationship (SAR) studies were undertaken with the aim of identifying more potent and selective inhibitors of mPGES-1 and/or dual inhibitors of mPGES-1 and 5-LOX. The biological data of the tested compounds and of the standards **1** and **2a** are compiled in Table 2.

Substitution of the *p*-chloro atom of the phenyl ring at the C-6 position of the central pyrrolizine core by NO_2 (**10d**), CF_3 (**10e**), or *tert*-butyl (**10g**) resulted in a significant decrease in potency against mPGES-1 (IC_{50} > 10 μM) compared to **3**. However, the substituent at the C-6 phenyl residue appeared to be of marked importance for the COX activity. In the

Table 2. Results of Biological Assays of mPGES-1 Reference Inhibitors and Arylpyrrolizine Test Compounds[a]

			COX residual activity at 30 μM [% 12-HHT of control] (±SEM)		
compd	cell free mPGES-1 residual activity at 10 μM [% PGE_2 of control] (±SEM)	cell free mPGES-1[b] IC_{50} [μM]	COX-1	COX-2	PMNL 5-LOX[c] IC_{50} [μM][14]
1	22.5 ± 2.3***	2.1[1] (lit.: 1.6[6])	19.5 ± 3.2***	56.7 ± 10.5**	nd
2a	11.6 ± 2.9*** at 100 nM	0.012 (lit.: 0.007[8])	nd	nd	nd
3	44.9 ± 2.6***	6.7	12.4 ± 7.6***	53.7 ± 2.4**	0.18[d]
10a	65.3 ± 3.9*	nd	nd	nd	1.6[d]
10b	47.4 ± 2.5***	7.2	82.9 ± 13.8	ni	nd
10c	20.8 ± 18.2***	1.8	nd	nd	1.9[d]
10d	58.8 ± 13.2**	nd	nd	nd	1.5[d]
10e	56.1 ± 4.3**	nd	nd	nd	0.31[d]
10f	42.1 ± 6.2***	6.2	94.0 ± 7.6	75.8 ± 12.8	nd
10g	53.8 ± 6.6***	nd	ni	ni	0.18[d]
11a	63.1 ± 0.9***	nd	nd	nd	0.23[d]
11b	38.5 ± 3.5***	5.9	nd	nd	nd
11c	28.7 ± 3.6***	4.5	25.8 ± 10.8***	43.4 ± 7.6**	0.25[d]
11d	32.2 ± 0.9***	4.8	8.6 ± 7.0***	13.6 ± 2.0***	0.26[d]
11f	23.2 ± 1.9***	2.1	29.2 ± 5.1***	ni	nd
14	27.9 ± 7.2***	3.9	37.4 ± 3.7***	46.5 ± 5.1**	0.5[d]

[a] n = 3−5, (*) p < 0.05, (**) p < 0.01, or (***) p < 0.001 vs vehicle (0.1% DMSO) control, ANOVA + Tukey HSD post hoc tests. nd = not determined. ni = no inhibition. [b] Utilized inhibitor concentrations: 10 μM, 1 μM, 100 nM, 10 nM (1 nM). [c] Utilized inhibitor concentrations: 10, 1, 3.3, 1 μM. [d] Average of at least two determinations.

tert-butyl phenyl derivative **10g**, the inhibitory activity toward both tested COX isozymes was successfully canceled out at an inhibitor concentration of 30 μM, whereas the 5-LOX inhibition remained unaffected ($IC_{50} = 0.18\ \mu M$),[14] which is promising for a possible follow-up investigation toward more selective mPGES-1 inhibitors.

In contrast, total replacement of the *p*-chloro phenyl moiety by benzofuran-6-yl in **10f** had only a minimal effect on potency, showing that such a planar bicyclic moiety is tolerated at this position. However, any advances in more selective mPGES-1 inhibitions were offset by the COX results. Both COX isoforms were only slightly inhibited by **10f**, which still constitutes a pharmacological benefit compared to **3**.

To optimize chemical stability of the ML3000-like compounds (**3**) bearing an acetic acid group in position C-5 (see Chemistry), we attempted to extend the alkyl spacer by one CH_2 unit to obtain the corresponding propionic acid (**10b** and **10c**). This modification had little effect on potency, as the IC_{50} values were comparable to those of the corresponding shorter acetic acid derivatives. However, as proposed, the propionic analogues were chemically more stable and thus better suited for isolation and, ultimately, for further substitution.[12]

With respect to the C-2 unsubstituted derivatives **10a** and **10c** and their mPGES-1 activity in the cell free assay at 10 μM (Table 2), the inhibition values are inverse compared to the C-2 dimethyl substituted compounds (**3** and **10b**). Here, the free propionic acid **10c** (C-2 unsubstituted) tends to be the more potent derivative. In the case of the C-2 substituted compounds, the propionic acid **10b** is almost comparable to the acetic acid **3** (as described above). In contrast, the dialkyl substituent at C-2 appears to be of markedly relevance for the inhibitory potency against 5-LOX, as can be derived from the data of **10a** ($R_3 = H$), which has only a moderate 5-LOX IC_{50} of 1.6 μM compared to its dihomo analogue (**3**). These results once more accent a strong positive relationship between the lipophilicity of the inhibitor molecule and the 5-LOX activity, also correlating with the earlier finding that one of the key structural features of the active site of the 5-lipoxygenase is a hydrophobic domain.[14] As a consequence, all other compounds out of Table 2 having a comparably lipophilic core to **3** and bearing a C-2 dialkyl residue constantly proved to have submicromolar 5-LOX IC_{50} values except for the more hydrophilic **10d** ($R_2 = 4\text{-}NO_2$).

Substitution of sulfonimides at the free acid functionality had a great impact on potency toward mPGES-1. Despite incorporation of rather lipophilic aryl moieties, the acetic character of the side chain is conserved, which appears to be a critical determinant for ligand−enzyme binding.[7] The tolyl sulfonimide **11d** is already 1.4-fold more potent than **3**. Comparable data were achieved with the substitution of a simple phenyl group for the terminal tolyl group in **11c**. Moreover, for both compounds residual activities of COX-1 and COX-2 are relatively low. In each case, the COX-1 isozyme is more affected by the inhibitor than is COX-2, which is consistent with the findings for the lead **3**. Having recognized the importance of the arylsulfonimide substituent, we transferred this portion of the molecule to the above-mentioned more stable propionic acid analogues, here exemplified by **11f**, and achieved a further 2.3-fold increase in potency, as measured by IC_{50} values, compared to **11d**. Accordingly, this compound (**11f**) is 3.2-fold superior to **3** in the mPGES-1 assay and at least equipotent to the literature reference molecule **1**. Fortunately, in comparison to **1** and **3**, the COX activity at 30 μM is clearly less inhibited by **11f**, indicating a better selectivity for mPGES-1 than both of the reference compounds. The data reveal that the propionic acid derivative **11f** also has a better COX inhibition profile (higher residual COX activity) than its nor-derivative **11d.**

To define the minimal requirements for acceptable mPGES-1 activity, we introduced a small and sterically less demanding mesylate onto the acetic acid precursor **3** (realized in **11a**). This modification resulted in increased activity of the isolated mPGES-1 as detected by a 1.4-fold higher PGE_2 level (63.1% at 10 μM compared to control, Table 2). These findings imply that an optimal pattern may be obtained by a bulkier terminal aromatic system (R = Ph, Tol). Finally, in vitro potency could be favorably modulated by variation of the C-2 substitution position of the arylpyrrolizines. For example, when the methylsulfonimide analogue **11b** is compared to **11a**, the observed loss of instrinsic activity could be successfully compensated by replacement of the two methyl groups at C-2 by homologous ethyl residues. **11b** revealed an IC_{50} of 5.9 μM.

As a tetrazole may serve as a biosteric replacement for carboxylic acids, we also synthesized **14**. The biological data in Table 2 support this concept, with the IC_{50} of **14** (3.9 μM) being similar to the those of arylsulfonimides **11c** and **11d** (4.5 and 4.8 μM, respectively). Furthermore, the COX inhibition profile of **14** is comparable to that of **11c**, whereas the COX activity at 30 μM is markedly better suppressed by **11d**.

In addition, all tested sulfonimides (**11a**, **11c**, and **11d**) revealed good 5-LOX inhibitory results in the submicromolar range (IC_{50} = 0.23−0.26 μM); the tetrazole **14**, however, slightly declines (IC_{50} = 0.5 $_m$M). Thus, for these molecule portions the active site of the 5-LOX seem to be relatively insensitive for sterical factors, as even bulkier residues like phenyl or tolyl were tolerated. Also, definite fluctuations in the acidity (a certain acidity is essential as an inevitable structural property for ligand−protein binding) of the substituents at the pyrrolizin C-5 position appear to have no significant influence on the 5-LOX inhibition.

On the basis of the biological data for mPGES-1 of our lead **3** (see above) in a cell-based test matrix,[1] we presume that the potencies of the newly synthesized compounds **10b**/**f**, **11b**−**f**, and **14** in the whole cell essay should correlate in order of magnitude with their inhibitory power in the cell free mPGES-1 test system, because of a presumably moderate or low degree of protein binding (see Supporting Information, table of clogP values). Thus, we predict only a small shift in potency caused by the presence of plasma proteins for these compounds, even if they revealed an inhibitory activity on PGE_2 in the single-digit micromolar range in the cell free test assay.

With regard to the future oral bioavailability of these potential drug candidates, we also intended to maintain an opportune hydrophilic/lipophilic balance, favoring an expected absorption or permeation behavior in vivo. The arylpyrrolizine compounds described here, i.e., the sulfimides, generally appear to better meet with the Lipinski rule-of-five criteria with respect to the molecular weight, their calculated logP values, and/or the number of H-bond donor/acceptor positions (see Supporting Information). These characteristics are in strong contrast to most of the other published inhibitor candidates such as the reference **1** and, in particular, its highly potent but less selective indole analogues.

To conclude, the concise synthetic method described in Schemes 1−3 allowed us to prepare a selection of diverse substituted small arylpyrrolizine derivatives from similar

starting materials (Table 1). In a cell free mPGES-1 assay, many of our test compounds exceeded **3** in potency (Table 2). The incorporation of arylsulfonimides led to a 3.2-fold increase in in vitro potency against mPGES-1 compared to **3**. Moreover, the good submicromolar 5-LOX inhibitory potency remains unaffected for those sulfonimides, whereas the COX inhibition could be casually reduced compared to our lead **3**.

These primary results reveal that the structure of **3** can be successfully modified to yield potent in vitro inhibitors of mPGES-1. Additional investigations will provide in-depth information regarding the effect of further structural modifications of our lead compound and of selective mPGES-1 inhibition in vivo. In this context, mPGES-1 appears to be a promising future target for the development of anti-inflammatory drugs.

Experimental Section

General. All reagents and solvents were of commercial quality and used without further purification. HPLC analyses (see Supporting Information for details) were employed for establishing the grade of purity of each test compound. The purity of all tested compounds is ≥95%, if not denoted otherwise.

Synthesis of the Dihydropyrrolizinyl Acetic and Propionic Acid Derivatives. The three-step general synthetic procedure for the derivatization of the free acid position with varied sulfonimides is specified in the Supporting Information.[15]

Preparation and Analytical Characterization of Exemplified Test Compounds. 2-(6-(4-Chlorophenyl)-2,2-dimethyl-7-phenyl-2,3-dihydro-1*H*-pyrrolizin-5-yl)-*N*-tosylacetamide (11d). According to general procedure, **11d** was obtained from [6-(4-chlorophenyl)-2,2-dimethyl-7-phenyl-2,3-dihydro-1*H*-pyrrolizin-5-yl]acetic acid (**3**) (0.5 g, 1.3 mmol) and toluene-4-sulfonamide (0.26 g, 1.5 mmol) after 40 h at room temperature. The crude product was recrystallized from isopropanol to yield 0.28 g (40%) of **11d**: $C_{30}H_{29}ClN_2O_3S$ (M_r = 533.08); ^{1}H NMR (DMSO-d_6) δ (ppm) 1.21 (s, 6H, $(-CH_3)_2$), 2.40 (s, 3H, tosyl-CH_3), 2.70 (s, 2H, C1-H_2), 3.40 (s, 2H, CH_2-CON<), 3.47 (s, 2H, C3-H_2), 6.84−7.28 (m, 9H, aryl-H), 7.43−7.47 (d, 2H, ^3J = 8.1 Hz, tosyl-C3-/C5-H), 7.79−7.83 (d, 2H, ^3J = 8.3 Hz, tosyl-C2-/C6-H); IR (ATR) 3300, 2960, 1725, 1599, 1531, 1489, 1416, 1394, 1354, 1316, 1213, 1189, 1175, 1119, 1087, 1043, 1014, 978, 878, 837, 821, 812, 767, 735, 722, 701, 686, 668, 656 cm^{-1}; HRMS (FT-ICR-MS) m/z [M + H]$^+$ calcd for $C_{30}H_{30}ClN_2O_3S$ 533.166 02, found 533.166 15.

2-(6-(4-Chlorophenyl)-2,2-dimethyl-7-phenyl-2,3-dihydro-1*H*-pyrrolizin-5-yl)-*N*-tosylpropanamide (11f). According to general procedure, **11f** was obtained from 3-(6-(4-chlorphenyl)-2,2-dimethyl-7-phenyl-2,3-dihydro-1*H*-pyrrolizin-5-yl)propionic acid (**10b**) (0.5 g, 1.3 mmol) and toluene-4-sulfonamide (0.26 g, 1.5 mmol) after 40 h at room temperature. The crude product was recrystallized from diisopropyl ether to yield 0.3 g (39%) of **11f**: $C_{31}H_{31}ClN_2O_3S$ (M_r = 547.11); ^{1}H NMR (DMSO-d_6) δ (ppm) 1.18 (s, 6H, $(-CH_3)_2$), 2.36 (m, 5H, $-CH_2$-CON<, tosyl-CH_3), 2.71 (m, 4H, C5-CH_2/C1-H_2), 3.61 (s, 2H, C3-H_2), 6.89−7.33 (m, 9H, aryl-H), 7.36−7.40 (d, 2H, ^3J = 8.2 Hz, tosyl-C3-/C5-H), 7.79−7.83 (d, 2H, ^3J = 8.2 Hz, tosyl-C2-/C6-H); IR (ATR) 3234, 3149, 2956, 2843, 1723, 1597, 1530, 1486, 1428, 1409, 1330, 1186, 1171, 1121, 1121, 1085, 1013, 956, 861, 828, 810, 758, 694, 660, 631, 579, 556, 505 cm^{-1}; MS (FAB) m/z (%) 546.1 (M^+, 45); HPLC (HP1090) 11.3 min, 89.8%; HRMS (FT-ICR-MS) m/z [M + H]$^+$ calcd for $C_{31}H_{32}ClN_2O_3S$ 547.181 67, found 547.181 78.

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Supporting Information Available: Eicosanoid pathway, comparison of the Lipinski rule criteria, synthetic procedures, routine spectroscopic data, HRMS and HPLC data, biological testing methods. This material is available free of charge via the Internet at http://pubs.acs.org.

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