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Diastereoselective synthesis of an argatroban intermediate, ethyl (2*R*,4*R*)-4-methylpipecolate, by means of a Mandyphos/rhodium complex-catalyzed hydrogenation

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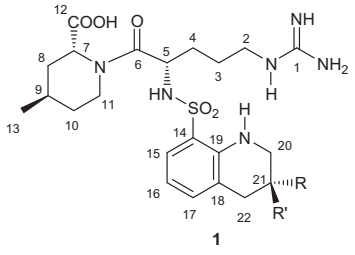
ABSTRACT

The synthetic antithrombotic argatroban is a dipeptide between the nonproteogenic (2*R*,4*R*)-4-methyl-2-piperidine carboxylic acid and L-arginine, in turn bonded to a methyltetrahydroquinoline sulfonyl group. An extensive screening of transition metal-based complexes with different ligands was performed in order to identify the best catalyst for the diastereoselective hydrogenation of a suitable 4,5-dehydropiperidine precursor aimed toward a synthesis of the (2*R*,4*R*)-4-methyl piperidine moiety.

1. Introduction

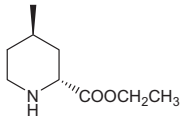
Argatroban **1** is a synthetic inhibitor of thrombin, the serine protease that plays a central role in the initiation and propagation of thrombotic events.[1] The most frequently prescribed anticoagulant with antithrombin activity is heparin but limitations due to its chemical heterogenicity in addition to several adverse events, such as the heparin-induced thrombocytopenia (HIT), led us to perform an extensive search for low molecular weight, selective inhibitors of thrombin. Argatroban is a small molecule (MW 509) approved in the USA, Europe and Japan for prophylaxis or treatment of thrombosis in patients with HIT that binds reversibly to and inhibits thrombin, without generation of antibodies or the degradation of proteases.[2,3]

In the argatroban structure, three moieties can be recognized: the 4-methyl-2-piperidine carboxylic acid bonded to the arginine, which also bears a methyltetrahydroquinoline sulfonyl group on its amino function. Four stereogenic centers are present in the molecule: the stereocenter on the tetrahydroquinoline is introduced *via* hydrogenation, performed in the last step of the synthesis of **1**. This reaction affords a mixture of (21*R*)- and (21*S*)-diastereoisomers, for which a respective ratio of 65/35 ± 2 is acceptable.



1

R = CH_3; R' = H (21*R*)-argatroban
R = H; R' = CH_3 (21*S*)-argatroban



(2*R*,4*R*)-**2**

On the contrary each of the other three stereogenic centers needs to be synthesized with high levels of stereoselectivity. For the arginine moiety it is sufficient to start from the natural L-aminoacid, but more care is required for the synthesis of the suitable diastereoisomer of 4-methylpipecolate, that is, (2*R*,4*R*)-**2**. Following a reported procedure,[4,5] we prepared intermediate **3** in order to investigate the best 4,5-double bond hydrogenation conditions (catalyst, solvent, temperature, pressure) with regard to the stereochemical outcome.

2. Results and discussion

Among the reported syntheses[6] of (2*R*,4*R*)-4-methyl-2-piperidincarboxylate, the good results offered by the reported method of Agami et al.,[7] based on the use of (*R*)-glycidol as a chiral auxiliary,

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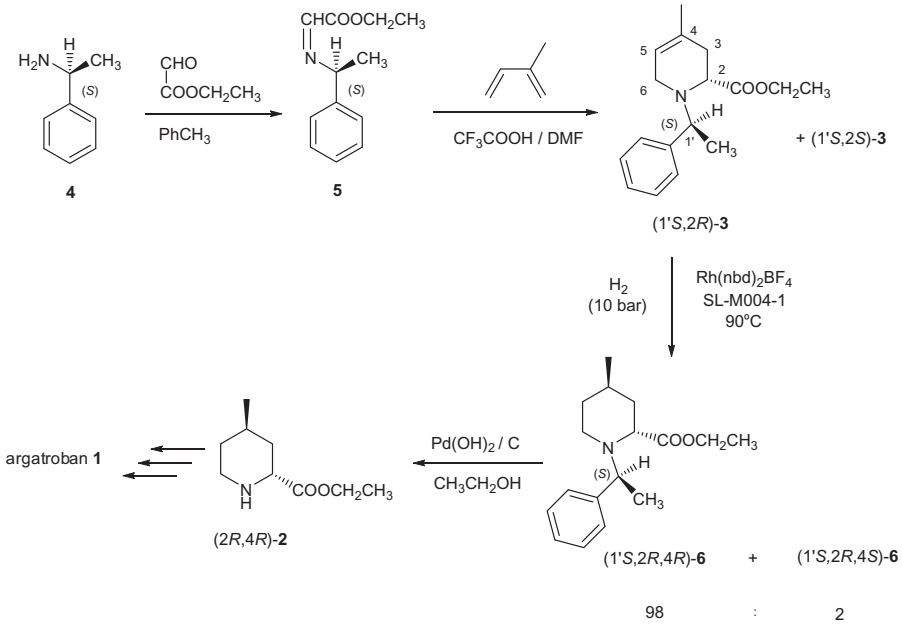

Scheme 1.

were noteworthy. Nevertheless we still considered an older preparation[4,5] of **2** to be more attractive, not necessarily in terms of the yield of the final product, but due to the facility of obtaining the intermediate product **3**. In this case, the chiral auxiliary is the enantiomerically pure 1-phenylethylamine, the choice between the (*R*)- or (*S*)-isomer being addressed by the desired configuration at the 2-position of the piperidine derivative. We started from commercially available (*S*)-1-phenylethylamine **4**, which induced the formation of the (2*R*)-2-piperidine carboxylate, over the course of a Diels–Alder condensation between 2-methyl butadiene and imine **5** (Scheme 1). In order to establish the diastereoisomeric ratio of the crude Diels Alder products, we developed a GLC method which showed an 82:18 ratio of the (1′*S*,2*R*)- and (1′*S*,2*S*)-isomers. After separation of the diastereoisomers by silica gel column chromatography, a comparison with the reported ^{1}H NMR spectra[†] allowed the assignment desired of the (2*R*)-configuration to the prevalent diastereoisomer, compound **3**.

In the original synthesis[4] the double bond present in (1′*R*,2*S*)-**3** was hydrogenated using 3% Pt/C as a catalyst and afforded an 88:12 (1′*R*,2*S*,4*R*)-*cis*/(1′*R*,2*S*,4*S*)-*trans* isomer mixture with the minor component being the opposite enantiomer of the compound required for the argatroban synthesis. The reduction of (1′*S*,2*R*)-**3** was carried out under the same conditions and afforded (1′*S*,2*R*,4*R*)-**6** with comparable diastereomeric ratio (dr) (established by GLC) and yields (9% after a silica gel column chromatography); this was used as the reference reaction to develop the hydrogenation conditions of the intermediate **3** with improved yields and stereochemical outcome. Since with platinum on charcoal as a catalyst, hydrogen addition to the 4,5-double bond takes place from the less hindered phaseface, the *cis*-isomer is formed predominately. It was, however, reasonable to expect a different, more favorable, result using a different catalyst. After an attempt with rhodium on alumina that furnished a 1:1 (1′*S*,2*R*,4*S*)-*cis*/(1′*S*,2*R*,4*R*)-*trans* diastereoisomeric mixture, we turned our attention to low valent rhodium, ruthenium and iridium complexes, which are known to be very active and versatile catalysts for homogeneous asymmetric hydrogenations.[8] Moreover, the preparation of (2*R*,4*R*)-**2** via the reduction of **3** with rhodium (I)-[1,4-bis(diphenylphosphino)butane]-(1*c*,5*c*-cyclooctadiene) tetrafluoroborate, and the removal of the chiral auxiliary, as reported in a 1997 German patent[9] constituted an encouraging precedent (62% overall yields and 93.6 diastereomeric excess, after column chromatography for each step).

The catalyst screening was performed utilizing P,P- and P,N-coordinating ligands with rhodium, ruthenium, and iridium catalysts from Solvias. Either isolated metal-complexes or free ligand combinations with a neutral or cationic metal precursor were employed.

While iridium complexes bearing P,N ligands afforded low conversions and a 1:1 diastereomeric ratio, use of P,P ligands led to the prevalent formation of the undesired (2*R*,4*S*)-*cis*-isomer. Ruthenium-catalyzed hydrogenations provided low conversions while the high H_2 pressures required to obtain good diastereoselectivities made these results less attractive from an economical standpoint. The best results, shown in Table 1, were observed with rhodium complex-catalyzed hydrogenations (the ligand structures investigated are reported in Fig. 1).

Some preliminary experiments using the achiral ferrocenyl ligand **7** (SL-F201-0) revealed that full conversion and 98:2 dr could be obtained at 100 bar and 80 °C, while at lower pressures and room temperature, the conversion as well as diastereoselectivity greatly decreased (entries a and b). Axially chiral ligands enantiomers, **8** (SL-A101-1) and **9** (SL-A101-2), in combination with a neutral rhodium precursor, (entries c and d) showed opposite diastereoselectivity. A good dr was observed with the latter ligand, but in favor of the undesired isomer. The use of **8** with the cationic precursor $[Rh(cod)_2]BF_4$ (bis(1,5-cyclooctadiene)rhodium tetrafluoroborate) resulted in 85% conversion and a slightly higher diastereoselectivity of 78:22 (entry e). Good diastereoselectivity (entry f), but with low conversion, was observed with ligand **10** (SL-M004-1), a chiral ferrocenyl phosphine belonging to the Mandyphos family,[10–13] which is characterized by a C_2-symmetrical backbone (Fig. 2). Starting from these results, an additional screen was planned while taking into account not only the diastereomeric ratio and the conversion values but also the reaction conditions [most importantly hydrogen pressure, but also the temperature and substrate/catalyst (S/C) ratio]. The cationic complex bis(norbornadiene) rhodium(I) tetrafluorobo-

[†] The respective enantiomers (1′*R*,2*S*) and (1′*R*,2*R*) are describes in the literature.[4]

Table 1
Results of preliminary catalyst screening

Entry	Metal precursor	Ligand (L)	S/C	Solvent	*p* (bar)	*T* (°C)	Conv[b] (%)	dr[b] (%)
a	[Rh(cod)(**7**)]BF_4[a]		25	EtOH	100	80	>99	98.8:1.2
b	[Rh(cod)(**7**)]BF_4[a]		25	EtOH	20	rt	41	81:19
c	$[Rh(nbd)Cl]_2$	**8**	25	$PhCH_3$/DCE	100	80	95	68:32
d	$[Rh(nbd)Cl]_2$	**9**	100	$PhCH_3$/DCE	80	80	77	6:94
e	$[Rh(cod)_2]BF_4$	**8**	25	EtOH	100	80	85	78:22
f	$[Rh(nbd)_2BF_4]$	**10**	100	EtOH	20	rt	23	84:16

[a] Isolated Rh-complex.
[b] Determined by GC.

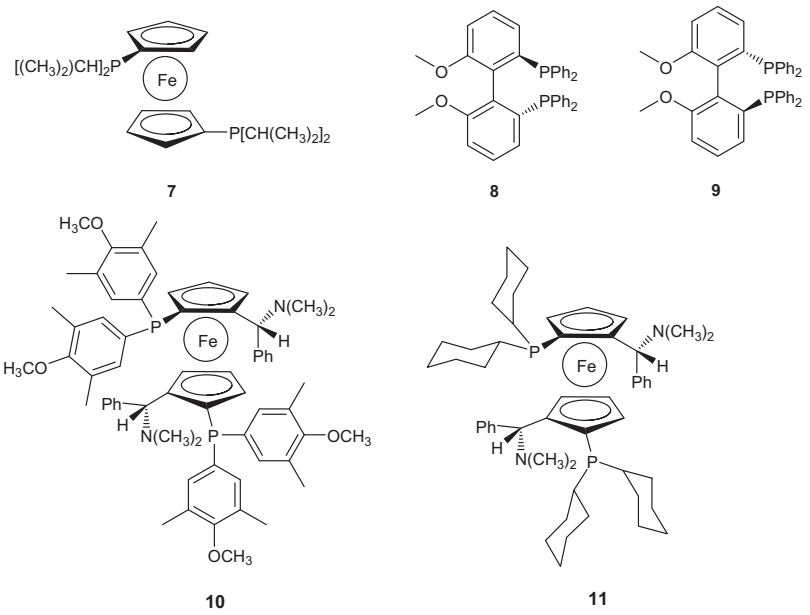

Figure 1. Structures of ligands investigated.

rate ($[Rh(nbd)_2]BF_4$ and the neutral complex (norbornadiene) rhodium(I) chloride dimer ($[Rh(nbd)Cl]_2$) were chosen as rhodium precursors, and tested with a series of our chiral ligands. Since free coordinating groups in the substrate, such as the amine functionality present in compound **3**, can lead to inactivation of the catalyst system, methane sulfonic acid (0.5 equiv) was tested as an additive. Carrying out the reaction at 20 bar and 80 °C with an S/C ratio of 25 for 16 h, more promising results (Table 2) were observed with $[Rh(nbd)_2]BF_4$ as the precursor and two chiral ferrocenyl phosphines of the Mandyphos family, namely ligand **11** (SL-M002-1) and the already tested **10**.

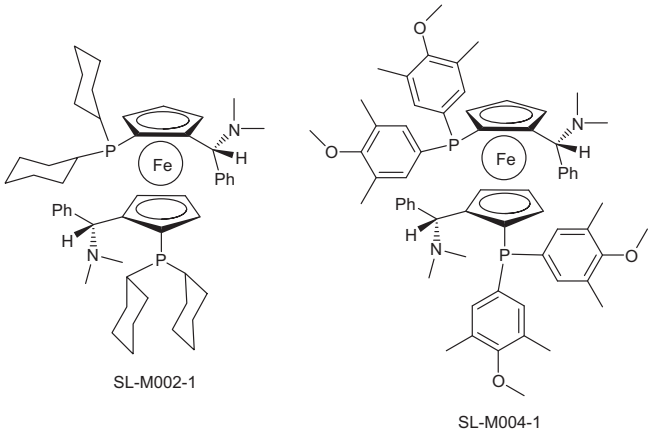

Figure 2.

Due to these excellent results, **10** was chosen for some additional experiments (Table 2) in order to investigate if the hydrogenation conditions could be improved upon. With an increase of the S/C ratio upto 400 and a reduction of the hydrogen pressure to 10 bar, the dr remained excellent (98:2) with only a slight decrease in the conversion (96%). Finally the selected $[Rh(nbd)_2]BF_4$/SL-M004-1 catalyst was used for the preparation of (1′*S*,2*R*,4*R*)-**6** at 90 °C and 10 bar, in the presence of methanesulfonic acid, on a gram-scale (50 ml autoclave). This process was successfully scaled up to a multi-kg scale. The argatroban **1** synthesis was accomplished, according to a reported method,[14] starting from chiral synthon **2**, recovered after removal of the chiral auxiliary from compound **6**, by means of a $Pd(OH)_2$/C-catalyzed hydrogenation.[4,5] Crude argatroban was purified by silica gel column chromatography and subsequent crystallization provided **1** (48% overall yields from **2**) as a mixture of (21*R*/21*S*) 65:35 diastereoisomers as established by HPLC and NMR[15] analyses. The HPLC chromatogram of argatroban **1** showed two other peaks (0.05% each peak) in addition to the signals due to the (21*R*)- and (21*S*)-diastereoisomers. LC–MS analysis revealed that these compounds had the same molecular weight as **1**. In order to verify if these impurities corresponded to the (9*S*,21*R*/*S*)-diastereoisomers (C9 of final product is the C4 of the piperidine moiety), a sample of (9*S*,21*R*/*S*)-argatroban was prepared starting from the ethyl (2*R*,4*S*)-4-methylpipecolate

Table 2
Hydrogenation of (1′*S*,2*R*)-**3** with $[Rh(nbd)_2]BF_4$/L and addition of 0.5 equiv CH_3SO_3H

Entry	Ligand (L)	S/C	Solvent	*p* (bar)	*T* (°C)	Time (h)	Conv[b] (%)	dr[b] (%)
a	**11**	25	EtOH	20	80	16	97	97:3
b	**10**	25	EtOH	20	80	16	>99	98:2
c	**10**	100	EtOH	10	90	18	100	98.5:1.5
d	**10**	400	EtOH	10	90	18	90	98:2
e	**10**[a]	400	EtOH	10	90	16	96	98:2
f	**10**	400	EtOH/$PhCH_3$	10	90	16	95.5	97:3

[a] 1 equiv of CH_3SO_3H instead of 0.5 equiv.
[b] Determined by GC.

and a HPLC analysis spiked with these reference compounds confirmed this hypothesis.

3. Conclusion

Argatroban **1** can be regarded as the dipeptide derivative of the natural L-arginine with the non-proteogenic aminoacid (2*R*,4*R*)-4-methylpipecolic acid, bearing on the arginine amino function a (3*R*/*S*)-3-methyl tetrahydroisoquinoline sulfonyl group. The straightforward preparation of the intermediate (1′*S*,2*R*)-**3** according to a reported method,[4,5] prompted us to study an improvement of its hydrogenation to **6**, described in the same papers with global (*cis* and *trans*) 78% yields and a dr of 88:12, in favor of the undesired diastereoisomer. A careful screening of the iridium, ruthenium, and rhodium complexes with P,P- and P,N-ligands allowed us to identify a hydrogenation catalyst for the intermediate **3** which afforded the desired (2*R*,4*R*)-*trans*-**6** with quantitative conversion and excellent diastereoselectivity (98:2).

We determined that $Rh(nbd)_2BF_4$ and Mandyphos ligand **10** afforded the best results when methane sulfonic acid was present in the reaction mixture, at 90 °C and 10 bar, (see entry f, Table 1 and entries b–f, Table 2) showing that a combination of the substrate and the catalyst control is required in order to obtain high selectivity.

The screening was performed employing the Solvias catalysts, which in addition to the observed elevated performances, have the advantage of being commercially available for laboratory scale as well as for industrial preparative purposes. Argatroban **1** was prepared in 48% yield, starting from chiral synthon **2**, and showed excellent purity containing only negligible amounts (0.05%) of the (9*S*,21*R*/*S*)-diastereoisomers.

4. Experimental

All reagents and solvents were purchased from Sigma–Aldrich, Milan (Italy). Iridium, ruthenium and rhodium-based complexes as well as all ligands were obtained from Solvias, Basel (Switzerland). The GLC system consisted of a VARIAN 3900 while the detector for capillary columns FID and acquisition data system was a Varian 3900XL. An AT-5 MS (Alltech) column (60 m × 0.25 mm × 0.25 μm) was employed. Carrier: N_2 (30 psi); injector temperature: 200 °C; detector temperature: 300 °C; oven temperature: 150 °C (3 min), then from 150 to 250 °C (10 °C/min), 3 min at 250 °C and then to 300 °C (30 °C /min). Split ratio: 50:1. The HPLC system consisted of an Agilent 1100-series liquid chromatography, equipped with an auto injector, DAD detector, and a Chemostation software installed on a PC, for data collecting and processing. A Zorbax SB-C18 (Agilent) column (100 × 4.6 mm, 1.8 μm) was employed. Column temperature: 60 °C. Mobile phase: A: ammonium acetate buffer solution pH 5.5; B: acetonitrile/methanol 1:1. Elution gradient from A/B 70:30 (35 min) to 55:45 (50 min). Flow rate: 1.2 mL/min. Detection UV 260 nm.

LC–MS system: HPLC Agilent 1100; Alltima C18 column (150 × 4.6 mm, 3 μm); column temperature 40 °C. Mobile phase: A: ammonium acetate solution 0.01 M; B: acetonitrile. Elution gradient: A/B 90:10 (10 min), A/B 75:25 (35 min), A/B 50:50 (30 min), A/B 90:10 (5 min). Flow rate: 1 mL/min. Spectrometer Esquire 300plus using the ESI source with positive ion polarity. Data acquisition and analysis were accomplished with the Bruker Daltonics DataAnalysis 3.3 software.

Optical rotation values were registered on a Perkin Elmer instrument (Mod 343) at 589 nm and 25 °C. NMR spectra were recorded in $CDCl_3$ with a Bruker AVANCE 500 spectrometer. Chemical shifts are reported on δ (ppm) scale from TMS.

4.1. Ethyl[(*S*)-1-phenylethyl]imino ethanoate 5

To a solution of ethyl glyoxylate (20 mL, 100 mmol) in toluene (100 mL), (*S*)-1-phenylethylamine **4** (12.7 mL, 100 mmol) was added. Water was azeotropically removed by evaporation at atmospheric pressure until the temperature rose to 110 °C. Toluene was evaporated at reduced pressure. The obtained orange oil (quantitative yields) if not immediately used in the next step, must be kept under nitrogen at −20 °C in order to avoid degradation. ^{1}H NMR ($CDCl_3$) δ: 1.38 (t, 3H, *J* = 7.35 Hz, CH_3CH_2); 1.65 (d, 3H, *J* = 6.47 Hz, CH_3CH); 4.36 (q, 2H, *J* = 7.35 Hz, CH_3CH_2); 4.63 (q, 1H, *J* = 6.47 Hz, $CHCH_3$); 7.35 (br s, 5H, Ar); 7.75 (s, 1H, CH=).

4.2. (2*R*/*S*)-1-[(1′*S*)-1′-Phenylethyl]-2-ethoxycarbonyl-4-methyl-4,5-dehydro piperidine 3

To a solution of imine **5** (20.11 g, 98 mmol) in *N*,*N*-dimethylformamide (70 mL), being stirred at room temperature under a nitrogen atmosphere, freshly distilled isoprene was added dropwise (13.3 g, 195 mmol) over 10 min. The reaction temperature was brought to 0–5 °C and trifluoroacetic acid (11.25 g, 98.7 mmol) was added dropwise (60 min) keeping the temperature at 0–5 °C. The reaction mixture was allowed to reach the room temperature and stirred for 18–20 h, monitoring the reaction progress by TLC (*n*-hexane/ethyl acetate 8:2). The solvent was evaporated under reduced pressure and the oily residue was dissolved in toluene (80 mL). The mixture obtained was sequentially washed with a saturated sodium hydrogen carbonate aqueous solution (80 mL × 2) until pH 8 and then with brine (80 mL). The organic phase was dried over sodium sulfate, filtered, and evaporated under reduced pressure to obtain a dark oil that was purified by silica gel column chromatography (30:1). The desired (1′*S*,2*R*)-isomer was obtained by elution with *n*-hexane/ethyl acetate 98:2 with 33% yields. ^{1}H NMR ($CDCl_3$) δ: 1.33 (t, 3H, *J* = 6.99 Hz, CH_3CH_2); 1.40 (d, 3H, *J* = 6.5 Hz, CH_3CH); 1.73 (s, 3H, CH_3C=); 2.38 (d, 1H, *J* = 16.9 Hz, H-3a); 2.60 (d, 1H, *J* = 16.2 Hz, H-3b); 2.98 (d, 1H, *J* = 16.7 Hz, H-6a); 3.21 (d, 1H, *J* = 16.7 Hz, H-6b); 4.00 (q, 1H, *J* = 6.5 Hz, CHN); 4.10 (dd, 1H, *J* = 5.4, 1.9 Hz, *CH*COOEt); 4,22 (q, 2H, *J* = 6.99 Hz, CH_2CH_3); 5.31(br s, 1H, CH=); 7.22–7.45 (m, 5H, Ar). GC R_t 14.23; for (1′*S*,2*S*)-isomer R_t 14.05.

4.3. Ethyl (2*R*,4*R*/*S*)-1-[(1′*S*)-1′-phenylethyl]-4-methylpipecolate 6

4.3.1. 5% Pt/C as catalyst

To a solution of (1′*S*,2*R*)-**3** (1.13 g, 4.13 mmol) in ethyl acetate (90 mL) was added 3% Pt/C (0.170 g). The hydrogenation was performed at room temperature and at an atmospheric pressure (48 h) following the reaction progress by TLC (hexane/diethyl ether 8:2) and GC. The catalyst was removed by filtration through a Celite pad; evaporation of the solvent afforded an oil (1.05 g), which was purified by silica gel column chromatography (10 g). By elution with hexane/diethyl ether 97:3, pure (1′*S*,2*R*,4*R*)-**6** (0.101 g, 9%) was recovered. ^{1}H NMR ($CDCl_3$) δ 0.92 (d, 3H, J = 5.90 Hz, CH_3-4); 1.08 (m, 1H, H-5a); 1.27 (d, 3H, J = 6.6 Hz, CH_3CHN); 1.33 (t, 3H, J = 7.1 Hz, CH_3CH_2); 1.45–1.57 (m, 3H, H-3a, H-4, H-5b); 2.08 (m, 1H, H-3b); 2.50 (m, 1H, H-6a); 2.86 (dt, 1H, J = 12.6 and 2.8 Hz, H 6b); 3.95–4.02 (m, 2H, H-2 and NC*H*CH_3); 4.20 (m, 2H, CH_2CH_3): 7.23–7.52 (m, 5H, Ar). GC R_t 13.76. Elution with hexane/diethyl ether 96:4 afforded the (1′*S*,2*R*,4*S*)-isomer **6** (0.34 g, 30%). ^{1}H NMR ($CDCl_3$) δ 0.94 (d, 3H, J = 6.10 Hz, CH_3-4); 1.19 (qd, 1H, J = 11.9 and 3.5 Hz, H-5a), 1.32 (t, 3H, J = 7.1 Hz, CH_3CH_2); 1.35(d, 3H, J = 6.9 Hz, CH_3CHN); 1.40–1.55 (m, 3H, H-3a, H-4, H-5b); 1.89 (m, 1H, H-3b); 2.17 (td, 1H, J = 11.2 and 2.3 Hz, H-6a); 2.51 (dt, 1H, J = 11.2 and 3.2 Hz, H-6b); 3.39 (dd, 1H, 11.2 and 3.2 Hz, H-2); 3.97 (q, 1H, J = 6.9 Hz, NC*H*CH_3); 4.23 (m, 2H, CH_2CH_3): 7.16–7.44 (m, 3H, Ar); 7.44–7.68 (m, 2H, Ar). GC R_t 14.44.

4.3.2. 5% Rh/Al_2O_3 as catalyst

To a solution of (1′*S*,2*R*)-**3** (0.64 g, 2.34 mmol) in ethyl acetate (50 mL), 5% Rh/Al_2O_3 (0.120 g) was added. The hydrogenation was performed at room temperature and atmospheric pressure. After 24 h, the catalyst was removed by filtration and a new amount of Rh/Al_2O_3 (0.120 g) was added. Work-up after an additional 24 h and purification as above afforded the desired (1′*S*,2*R*,4*R*)-**6** in 42% yield.

4.3.3. Rh(nbd)$_2$$BF_4$/10 as catalyst

A solution of (1′*S*,2*R*)-**3** (1.45 g, 5.3 mmol) in toluene (10 mL) was placed in a flask. The solvent was removed under reduced pressure and the residue dried under high-vacuum. After three vacuum-argon cycles, the equipment was set under argon. Ethanol (10 mL), stored over molecular sieves and degassed by bubbling argon through the solvent, was added with a needle. Methanesulfonic acid (190 μL, 2.65 mmol, 0.5 eq) was added. In a second flask, the metal precursor Rh(nbd)$_2$$BF_4$ (4.97 mg, 0.0133 mmol, S/C 400) was placed, followed by **10** (15.39 mg, 0.0146 mmol). The flask was set under argon by three vacuum-argon cycles. Degassed ethanol (6 mL) was added and the solution obtained was stirred at room temperature for 20 min. The stainless steel 50 mL autoclave was set under argon. Under a constant flow of argon, the solution from the first flask was transferred into the autoclave with a needle, followed by the solution from the second flask. The autoclave was closed, purged with argon (3×, 10 bar) and then with hydrogen (3×, 10 bar). The pressure was adjusted to 7 bar and heating was started. When the reaction temperature reached 90 °C, the pressure was re-adjusted to 10 bar and stirring was started. The reaction mixture was hydrogenated overnight (16–18 h). Afterward, heating was stopped, the autoclave was cooled, and vented and the solution transferred into a flask. Ethanol was evaporated at reduced pressure; to the residue, toluene (10 mL) and a 10% sodium hydrogen carbonate aqueous solution (5 mL) were added. The organic phase was separated, washed with water and dried by treatment with sodium sulfate. The toluene solution was filtered through a silica gel pad; after evaporation of the solvent, the title compound (1′*S*,2*R*,4*R*)-**6** (1.27 g, 87%, 98:2 dr by GC) was recovered and used in the next step without further purification.

4.4. Ethyl (2*R*,4*R*)-4-methyl-2-pipecolate 2

To a solution of (1′*S*,2*R*,4*R*)-**6** (2.5 g, 9 mmol) in ethanol (120 mL) 10% Pd(OH)$_2$/C (0.27 g) was added. The hydrogenation was performed at room temperature and at atmospheric pressure. The reaction progress was monitored by TLC (chloroform/methanol 9:1). After 4 h, the catalyst was removed by filtration through a Celite pad. Solvent evaporation at reduced pressure afforded the crude mixture that was purified by silica gel (1:20) column chromatography; pure title compound **2** (0.7 g, 45%) was recovered by elution with dichloromethane/methanol 98:2. ^{1}H NMR ($CDCl_3$) δ 0.96 (d, 3H, J = 6.50 Hz, CH_3-4); 1.15 (m, 1H, H-5a), 1.32 (t, 3H, J = 7.2 Hz, CH_3CH_2); 1.42–1.54 (m, 1H, H-3); 1.54–1.69 (m, 2H, H-4, H-5b); 2.00–2.11 (m, 2H, H-3b and NH); 2.87 (m, 2H, H-6); 3.65 (m, 1H, H-2); 4.21 (q, 2H, J = 7.2 Hz, CH_2CH_3). $[\alpha]_D^{20} = -22$ (c 5, ethanol) (lit.[4] +24 for the enantiomer).

Compound (2*R*,4*S*)-**2** was prepared in the same way starting from (1′*S*,2*R*,4*S*)-**6**. ^{1}H NMR ($CDCl_3$) δ 0.97 (d, 3H, J = 6.50 Hz, CH_3-4); 1.07 (m, 1H, H-5a), 1.29 (m+t, 4H, J = 7.1 Hz, CH_3CH_2, H-4); 1.48–1.66 (m, 2H, H-3a, H-5b); 1.78 (br s, 1H, NH); 2.02 (m, 1H, H-3b); 2.46 (td, 1H, J = 2.4 and 12.4 Hz, H-6a); 3.17 (ddd, 1H, J = 1.9, 3.8, 12.4 Hz, H-6b); 3.31 (dd, 1H, J = 3.0 and 11.9 Hz, H-2); 4.20 (q, 2H, J = 7.1 Hz, CH_2CH_3).

4.5. Argatroban 1

Crude **1** prepared from (2*R*,4*R*)-**2** according to a reported method[14] was purified by silica gel (1:25) column chromatography (elution with dichloromethane/methanol 8:2), followed by crystallization (acetone/water 1:1). Pure **1**, recovered in 48% yield from **2**, showed chemical and physical properties in agreement with the reported ones.[14] For complete ^{1}H and ^{13}C NMR assignments see the literature.[15] HPLC: (21*R*)-**1**, R_t 26.87; (21*S*)-**1**, R_t 27.44.

Starting from (2*R*,4*S*)-**2**, diastereomeric (5*S*,7*R*,9*S*,21*R*/*S*) argatroban was prepared and analyzed by HPLC and LC–MS using the methods developed for compound **1**: two peaks with R_t 20.76 and 22.04, respectively, were observed. ^{1}H NMR ($CDCl_3$) (selected signals) δ 0.95−1.02 (d+d, 3H, CH_3-21); 1.07 (d, 3H, J = 6.6 Hz, CH_3-7); 3.43 (ddd, 0.4H, J = 11.5, 1.6 and J = 3.3 Hz, H-20a); 3.47 (ddd, J = 0.6H, 11.3, 1.9 and J = 3.5 Hz, H-20a); 6.59 (m, 1H, Ar); 7.10 (d, 1H, Ar); 7.48 (d, 1H, Ar). $[\alpha]_D^{20} = +42$ (c 1.02 M, HCl) (lit.[16] +43).

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References

1. McKeage, K.; Plosker, G. L. *Drugs* **2001**, *61*, 515–522.
2. Yeh, R. W.; Jang, I. K. *Am. Heart. J.* **2006**, *151*, 1131–1138.
3. Jeske, W. P.; Fareed, J.; Hoppensteadt, D. A.; Lewis, B.; Walenga, J. M. *Expert Rev. Hematol.* **2010**, *3*, 527–539.
4. Bailey, P. D.; Brown, G. R.; Korber, F.; Reed, A.; Wilson, R. D. *Tetrahedron: Asymmetry* **1991**, *2*, 1263–1282.
5. Bailey, P. D.; Wilson, R. D.; Brown, G. R. *J. Chem. Soc., Perkin Trans. 1* **1991**, 1337–1340.
6. Kadouri-Puchot, C.; Comesse, S. *Amino Acids* **2005**, *29*, 101–130.
7. Agami, C.; Bisaro, F.; Comesse, S.; Guesnè, S.; Kadouri-Puchot, C.; Morgentin, R. *Eur. J. Org. Chem.* **2001**, 2385–2389.
8. *Comprehensive Asymmetric Catalysis*; Jacobsen, E. N., Pfaltz, A., Yamamoto, H., Eds.; Springer: Berlin, Heidelberg, 1999; Vol. 1,.
9. Grell, W.; DE 19616049 A1, 1997.
10. Almena Perea, J. J.; Börner, A.; Knochel, P. *Tetrahedron Lett.* **1998**, *39*, 8073–8076.
11. Almena Perea, J. J.; Lotz, M.; Knochel, P. *Tetrahedron: Asymmetry* **1999**, *10*, 375–384.

12. Lotz, M.; Ireland, T.; Almena Perea, J. J.; Knochel, P. *Tetrahedron: Asymmetry* **1999**, *10*, 1839–1842.
13. Spindler, F.; Malan, C.; Lotz, M.; Kesselgruber, M.; Pittelkow, U.; Rivas-Nass, A.; Briel, O.; Blaser, H.-U. *Tetrahedron: Asymmetry* **2004**, *15*, 2299–2306.
14. Cossy, J.; Belotti, D. *Bioorg. Med. Chem. Lett.* **2001**, *11*, 1989–1992.
15. Colombo, D.; Ferraboschi, P.; Grisenti, P.; Legnani, L. *Magn. Reson. Chem.* **2008**, *46*, 99–102.
16. Kikumoto, R.; Tamao, Y.; Tonomura, S.; Hara, H.; Ninimyia, K.; Hijikata, A.; Okamoto, S. *Biochemistry* **1984**, *23*, 85–90.