Highly efficient and concise synthesis of both antipodes of SB204900, clausenamide, neoclausenamide, homoclausenamide and ζ-clausenamide. Implication of biosynthetic pathways of clausena alkaloids†

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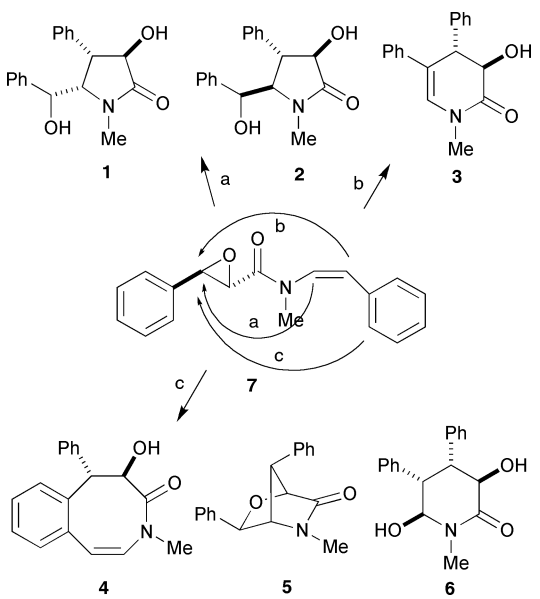
The synthesis of both antipodes of *N*-methyl-*N*-[(*Z*)-styryl]-3-phenyloxirane-2-carboxamide (SB204900), clausenamide, neoclausenamide, homoclausenamide and ζ-clausenamide have been accomplished using (2*S*,3*R*)- and (2*R*,3*S*)-3-phenyloxirane-2-carboxamides as the starting materials, and SB204900 was found to be a common precursor to other *N*-heterocyclic clausena alkaloids. Mediated by Brønsted acids under different conditions, for example, SB204900 underwent efficient and diverse alkene-epoxide cyclization, enamide-epoxide cyclization and arene-epoxide cyclization reactions to produce the five-membered *N*-heterocyclic neoclausenamide, its 6-epimer, the six-membered *N*-heterocyclic homoclausenamide and the eight-membered *N*-heterocyclic ζ-clausenamide, respectively, in good to excellent yields. Regiospecific oxidation of neoclausenamide and its 6-epimer afforded neoclausenamidone. Enolization of neoclausenamidone in the presence of LiOH and the subsequent protonation under kinetic conditions at −78 °C led to the epimerization of neoclausenamidone into clausenamidone. Reduction of clausenamidone using $NaBH_4$ furnished clausenamide in high yield.

Introduction

Rutaceae *Clausena lansium* (Lour.) Skeels is a fruit tree widely distributed in southern China. In folk medicine, its leaves and fruits are used to treat asthma, influenza, gastrointestinal disorders, viral hepatitis and dermatological diseases.[1] Due to the pioneering work of Huang and her co-workers[1] in the mid to late 1980s, a number of clausena alkaloids including clausenamide **1**,[2,3] neoclausenamide **2** (a diastereoisomer of clausenamide **1**),[3,4] homoclausenamide **3**,[5] ζ-clausenamide **4**[5] and cycloclausenamide **5**[3,4] (Scheme 1) were isolated from the hot-water extract of the leaves. Interestingly, while the former four alkaloids were isolated in racemic form, cycloclausenamide was optically active. Clausena alkaloids have been shown to possess efficacious liver-protecting and antiamnesia effects.[1] Later, lansimide-3 **6** (Scheme 1),[6] a hydrated form of homoclausenamide was reported. In 1996, Milner and co-workers[7] isolated the optically active (+)-*N*-methyl-*N*-[(*Z*)-styryl]-3-phenyloxirane-2-carboxamide, SB-204900 (+)-**7** (Scheme 1), from a hexane extract of *Clausena lansium* leaves.

Despite the important pharmacological activity and interesting molecular structures, the synthesis of clausena alkaloids has remained largely unexplored.[8] The same is true for the investigation of biological synthetic pathways[9] for the generation of diverse structures of clausena alkaloids. For years, we have been studying enantioselective nitrile biotransformations for the synthesis of enantiopure carboxylic acids and their amide derivatives.[10] Recently, we established an efficient method for the preparation of highly enantio-enriched oxirane-carboxamides.[11] That study led us to address the biosynthetic pathways and the organic synthesis of clausena alkaloids.



Scheme 1 Structures of alkaloids **1**–**7** from *Clausena lansium* (Lour.) Skeels and hypothetic transformations of **7** into five-, six- and eight-membered lactams.

We envisioned that the oxirane-containing enamide **7** might be a common intermediate or precursor to five-, six- and eight-membered lactam products. As hypothesized in Scheme 1, for

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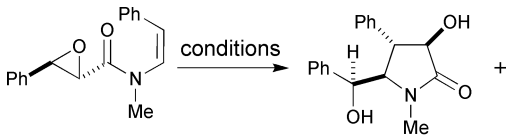
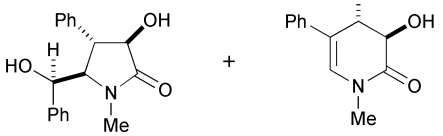
† Electronic supplementary information (ESI) available: Full characterization of products, ^1H and ^{13}C NMR spectra of products, HPLC analysis of all chiral products. See DOI: 10.1039/b901965k

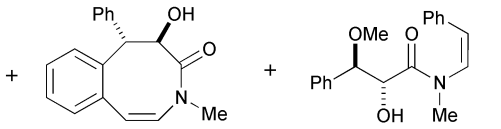
Table 1 Reactions of (±)-**7** under different conditions



(±)-**7** (±)-**2**



(±)-**2'** (±)-**3**



(±)-**4** (±)-**8**

Entry	Conditions	(±)-**2** + (±)-**2′** (%)[a]	(±)-**3** (%)[a]	(±)-**4** (%)[a]	(±)-**8** (%)[a]
1	CH_3CN, reflux, 12 h	—	—	—	—
2	H_2O (2 equiv), CH_3CN, reflux, 12 h	—	—	—	—
3	CH_3CO_2H (2 equiv), H_2O (2 equiv), CH_3CN, reflux, 12 h	—	—	—	—
4	HCl (aq.) (2 equiv), CH_3CN, rt, 2 h	—	—	70	—
5	*p*-$CH_3C_6H_4SO_3H$ (2 equiv), CH_3CN, rt, 2 h	—	—	86	—
6	*p*-$CH_3C_6H_4SO_3H$ (1 equiv), *t*-BuOH, rt, 8 h	—	51	24	—
7	*p*-$CH_3C_6H_4SO_3H$ (1 equiv), *t*-BuOH, reflux, 1 h	–	64	<5	—
8	CF_3CO_2H (2 equiv), *t*-BuOH, reflux, 12 h	–	44	27	—
9	CF_3CO_2H (2 equiv), CH_3OH, rt, 26 h	–	—	37	42
10	H_2O, reflux, 5 h	69[b]	—	27	—
11	CH_3CO_2H (2 equiv), H_2O, reflux, 5h	55[b]	—	36	—
12	Na_2CO_3 (2 equiv), H_2O, reflux, 5 h	75[b]	—	—	—

[a] Isolated yield. [b] The ratio of neocausenamide **2** over its 6-epimer was 3 : 7.

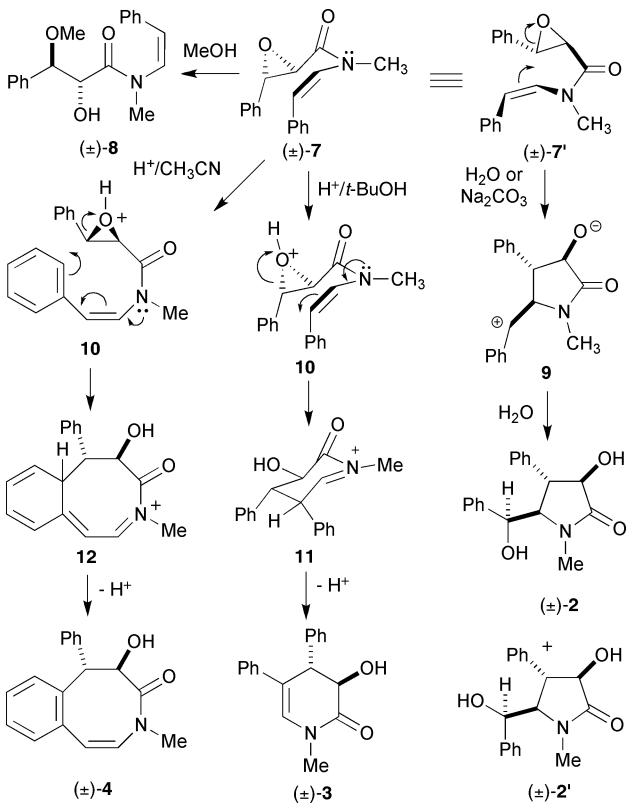
example, intramolecular enamide-epoxide ring opening reaction (pathway b) would lead to the formation of six-membered homoclausenamide while the intramolecular arene-epoxide ring opening reaction (pathway c) would produce eight-membered ζ-clausenamide. The five-membered lactam compound would result from alkene-epoxide ring opening reaction (pathway a). Herein we report a systematic study of the transformations of oxirane-containing enamide **7**,[12] proposing for the first time the (bio)synthetic pathways for diverse clausenamide alkaloids. Based on the (bio)synthetic pathways, we have also accomplished concise and biomimetic synthesis of both antipodes of five-, six- and eight-membered lactam alkaloids.

Results and discussion

To test our hypothesis of biosynthetic pathways of clausena alkaloids, we investigated the transformations of racemic oxirane enamide (±)-**7** under various conditions. We first examined the reaction of enamide in organic solvents. No reactions were observed when compound (±)-**7** was heated in acetonitrile for 12 h (entry 1, Table 1). Addition of water (2 equiv) and a weak acid such as acetic acid (2 equiv) did not effect the transformation either (entries 2 and 3, Table 1). Hydrochloric acid, a strong Brønsted acid, was then found to promote efficiently the reaction of (±)-**7**, affording ζ-clausenamide (±)-**4** in 70% yield (entry 4, Table 1). The use of *p*-toluenesulfonic acid further improved the conversion of (±)-**7** and ζ-clausenamide (±)-**4** was obtained in 86% yield (entry 5, Table 1). Interestingly, when the *p*-toluenesulfonic acid-mediated reaction of (±)-**7** was performed in *t*-butyl alcohol for 8 h, six-membered homoclausenamide (±)-**3** was obtained in addition to the formation of ζ-clausenamide (±)-**4** (entry 6, Table 1). Shortening reaction time to 1 h led to the formation of homoclausenamide (±)-**3** in an improved chemical yield (entry 7, Table 1). It should be noted that trifluoroacetic acid appeared effective to promote the transformation of (±)-**7** into (±)-**4** and (±)-**3** albeit in low chemical yields (entry 8, Table 1). Methanol as a solvent was found, however, to have a detrimental effect, as it attacked the epoxide ring to give simply the epoxide-ring opening product (±)-**8** as the major product (entry 9, Table 1). We finally tested the transformations of oxirane enamide (±)-**7** in water. While it was quite stable at room temperature in water, oxirane enamide (±)-**7** underwent efficient transformations in boiling water. To our delight, a mixture of five-membered lactam products (±)-**2** and (±)-**2′** was obtained in 69% yield in addition to ζ-clausenamide (±)-**4** in 27% yield (entry 10, Table 1). It is noteworthy that the presence of acetic acid (2 equiv) was found to favour slightly the formation of eight-membered lactam (±)-**4** (entry 11, Table 1). Basic reaction conditions using two equivalents of sodium carbonate gave rise

to the exclusive formation of five-membered heterocyclic products (±)-**2** and (±)-**2′** in 75% yield (entry 12, Table 1). The ratio of neoclausenamide (±)-**2** over its 6-epimer (±)-**2′**, as determined by measuring the intensity of 6-H signals of (±)-**2** and (±)-**2′** in the ^{1}H NMR spectrum, was roughly 3 : 7 (entries 10–12, Table 1).

The transformations of enamide (±)-**7** into five-, six- and eight-membered lactams under different reaction conditions were intriguing. It was apparent that the reaction pathways were strongly influenced by both Brønsted acid and, more remarkably, solvent used. While the detailed mechanisms await further study, we proposed in Scheme 2 the plausible chemical conversions of oxirane-enamide SB204900 (±)-**7** into the five-, six- and eight-membered lactam products (±)-**2**, (±)-**2′**, (±)-**3** and (±)-**4**.

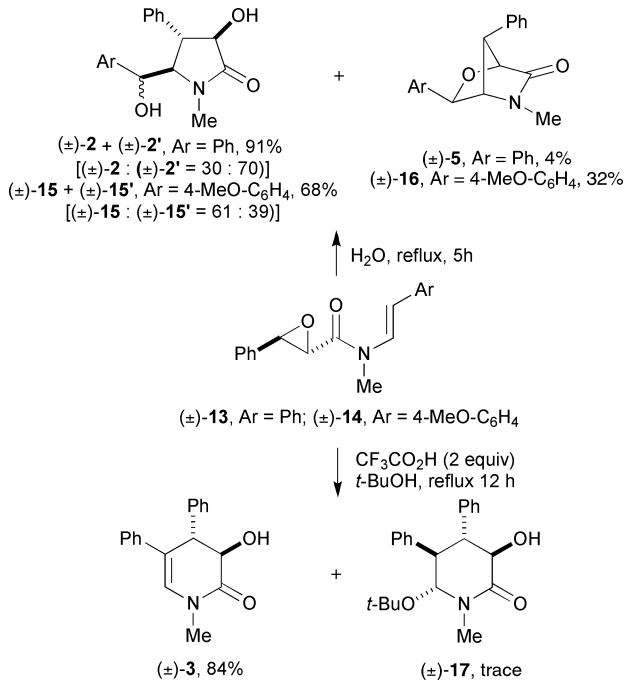


Scheme 2 Plausible mechanisms of the transformations of (±)-**7**.

The formation of ζ-clausenamide would most probably arise from an intramolecular Friedel–Crafts reaction.[13] As illustrated in Scheme 2, protonation of epoxide (±)-**7** gives rise to oxonium intermediate **10** which undergoes an electrophilic reaction with the benzene moiety to afford 5,6-dihydrobenzo[*d*]azocin-4(3*H*)-one structure (±)-**4**. It should be noted that the facile intramolecular aryl-epoxide cyclization of (±)-**7** in acetonitrile is determined by its intrinsic favourable electronic effect and folded conformational structure that was revealed by X-ray single crystal diffraction analysis.[7] Both the delocalization of enamide electrons into the benzene ring and the perfectly predisposed conformational structure dictate the 8-*endo*-epoxy-arene cyclization. When reacted in *t*-butyl alcohol, a more polar and protonic solvent, the protonated intermediate adopts most likely a chair conformation **10**. Enaminic attack of enamide on the oxonium results in the formation of iminium intermediate **11**, which undergoes deprotonation to furnish homoclausenamide product (±)-**3**. The transformation of (±)-**7** into the γ-lactam products (±)-**2** and (±)-**2′** appeared most surprising. A plausible reaction pathway might involve the attack of the α-carbon of the enamide moiety of (±)-**7** to the benzylic carbon of the epoxide.[13] The resulting zwitterion intermediate **9** reacts with water to yield a mixture of epimers (±)-**2** and (±)-**2′**. The function of the α-carbon rather than the β-carbon (enaminic carbon) of the enamide as the nucleophilic site to attack the epoxide ring would suggest that the lone pair electrons on the nitrogen delocalize into the carbonyl rather than the carbon–carbon double bond. In other words, under the reaction conditions such as in pure water or under basic conditions, the oxirane-enamide molecule (±)-**7** might adopt the conformation (±)-**7′** in which the amido plane might be perpendicular to the plane of the carbon–carbon double bond. This conformation inhibits delocalization of the lone pair electrons of nitrogen into the carbon–carbon double bond, alleviating therefore the character of enamide and the nucleophilicity of the "enaminic β-carbon" of enamide. The conformation (±)-**7′** also brings the proximity of the *p*-orbital of the α-carbon with the benzylic carbon of the epoxide ring, facilitating the 5-*endo*-enamide-epoxide cyclization reaction. In addition, the stabilization gained from the formation of benzylic cation **9** might also be a factor in the 5-*endo*-enamide-epoxide cyclization.

In biological systems, there are microenvironments of varied acidity and lipophilicity or hydrophility. It is most likely that, in these different acidic and hydrophilic biological microenvironments, oxirane-epoxide (±)-**7** undergoes different intramolecular reactions to give naturally occurring neoclausenamide (±)-**2**, homoclausenamide (±)-**3** and ζ-clausenamide (±)-**4**. At this point, however, we cannot exclude an artifact, *viz*. the possibility of conversions of (±)-**7** into clausena alkaloids (±)-**2**, (±)-**2′**, (±)-**3** and (±)-**4** during the isolation process.

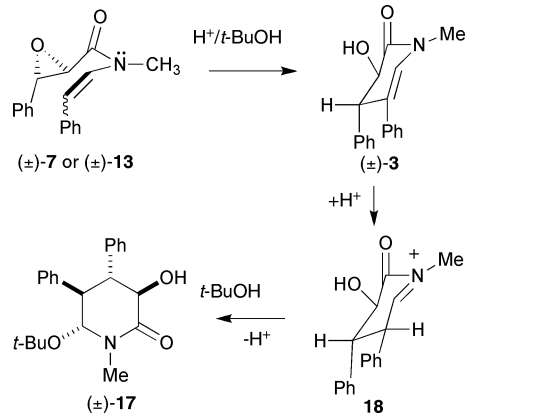
Although racemic SB204900 (±)-**7** was shown convincingly to be the common precursor to neoclusenamide (±)-**2**, homocalusenamide (±)-**3** and ζ-clausenamide (±)-**4**, unfortunately, no clausenamide (±)-**1** was obtained from (±)-**7** under any of the reaction conditions tested. We then investigated the reaction of racemic *N*-(*E*-styryl)-3-phenyloxirane-2-carboxamide (±)-**13**, an isomer of (±)-**7**, envisaging the configuration of enamide double bond might play a subtle role in intramolecular cyclization reactions.[12b] When refluxed in pure water for 5 h in a large-scale reaction (10 mmol), (±)-**13** was efficiently transformed into a mixture of neoclausenamide (±)-**2** and its 6-epimer (±)-**2′** in 91% yield (Scheme 3). In contrast to the reaction of (±)-**7**, the *E*-configured enamide (±)-**13** did not undergo intramolecular arene-epoxide cyclization to produce ζ-clausenamide (±)-**4** at all, because of the unfavourable molecular geometry. No clausenamide product was formed from (±)-**13** either. Interestingly, however, cycloclausenamide (±)-**5** was isolated in 4% yield. Using racemic *N*-(*E*-4-methoxyphenylvinyl)-3-phenyloxirane-2-carboxamide (±)-**14** as reactant, the identical reaction afforded a mixture of neoclausenamide derivatives (±)-**15** and (±)-**15′** in 68% and cycloclausenamide derivative (±)-**16** in 32% (Scheme 3). These reaction outcomes indicated that the reaction of (±)-**14** in pure water proceeded *via* the same zwitterion intermediate **9** as the reaction of (±)-**7**. Cyclization of **9** led to the formation of cycloclausenamide (±)-**5**. In the case of (±)-**15**, the introduction of a *para*-methoxy group at the benzene ring resulted in the stabilization of the benzyl cation structure of intermediate **9**,



Scheme 3 Reaction of *N*-(*E*-styryl)-3-phenyloxirane-2-carboxamide (±)-**13**.

leading thus to an improved chemical yield of cycloclausenamide derivative (±)-**16**.

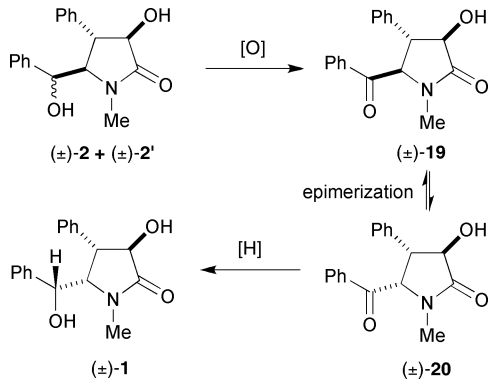
It is also worth noting that no clausenamide (±)-**1** was obtained either from the reaction of (±)-**13** under acidic conditions. Careful scrutiny of a large scale reaction (10 mmol) of oxirane-containing enamide (±)-**13** in the presence of two equivalents of trifluoroacetic acid, however, allowed us to isolate a trace amount of a new compound (±)-**17** in addition to homoclausenamide (±)-**3** (Scheme 3). On the basis of spectroscopic evidence, especially the NOE data, the compound was assigned as δ-lactam (±)-**17** with four substituents on the six-membered *N*-heterocycle being all trans-configured to each other. It is a derivative of the stereoisomer of lansamide-3 **6**. The formation of (±)-**17** is most probably due to the protonation of homoclausenamide, giving an all trans-configured iminium intermediate **18** (Scheme 4). Nucleophilic attack of *t*-butyl alcohol at iminium intermediate **18** from the less sterically hindered side of the six-membered ring afforded



Scheme 4 Proposed mechanism for the formation of (±)-**17**.

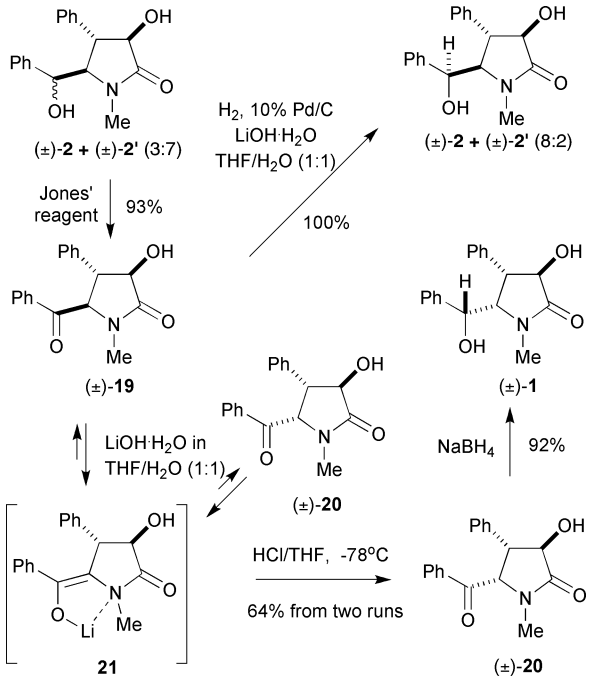
product (±)-**17** (Scheme 4). To account for the formation of naturally occurring lansamide-3 **6**, we proposed a rapid trap of the intermediate **11** by a water molecule.

Since chemical transformation of (±)-**7** did not straightforwardly afford clausenamide (±)-**1**, the naturally occurring clausenamide (±)-**1** would probably be generated from (±)-**7** by the action of an enzyme. Alternatively clausenamide (±)-**1** would be formed from its stereoisomer neoclausenamide (±)-**2**; *viz.*, a redox process might be responsible for the conversion of neoclausenamide (±)-**2** into clausenamide (±)-**1** in the biological system. As depicted in Scheme 5, selective oxidation of the hydroxyl group at the 6-position would result in the ketone derivative. The consecutive epimerization at the 5-position due to the acidity of the 5-position hydrogen, and the reduction of the carbonyl group would finally furnish the clausenamide product.



Scheme 5 Proposed biosynthetic route from neoclausenamide (±)-**2** to clausenamide (±)-**1**.

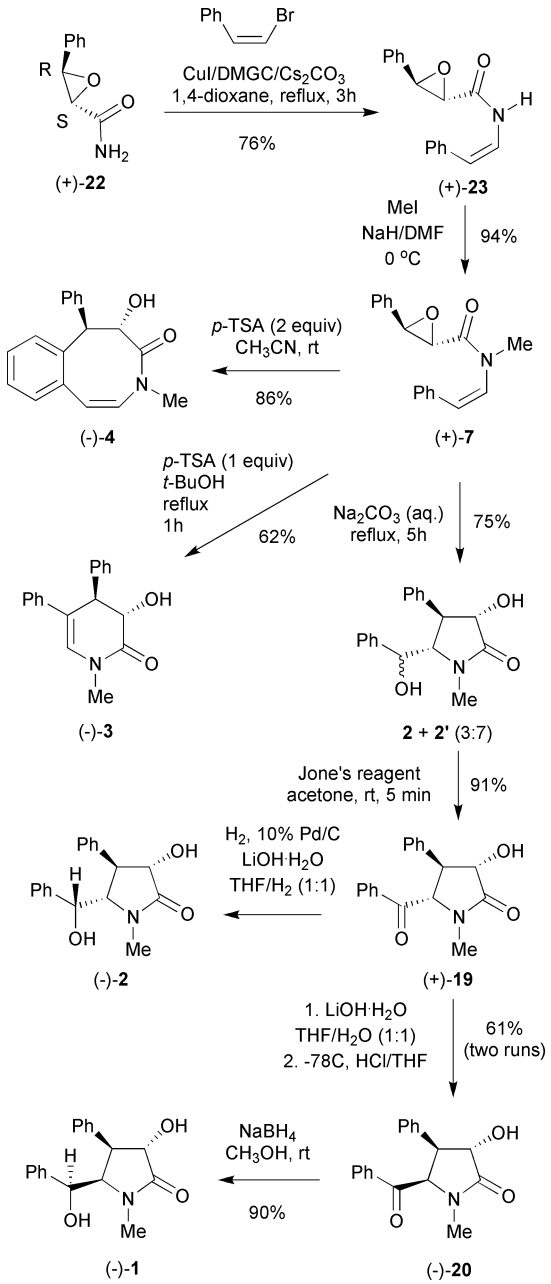
Following our hypothetical route, we studied the selective oxidation of neoclausenamide (±)-**2** and its 6-epimer (±)-**2′** [(±)-**2** : (±)-**2′** = 3 : 7], a mixture obtained directly from (±)-**7**. Treatment of the mixture with $KMnO_4$ gave rise to the oxidation of both hydroxyl groups at 3- and 6-positions, while the use of MnO_2 as an oxidant did not effect any reaction of neoclausenamide or its 6-epimer. Finally we found that the efficient oxidation reaction using Jones' reagent occurred highly selectively at the 6-position of compounds (±)-**2** and (±)-**2′**, affording the neoclausenamidone product (±)-**19** in 93% yield (Scheme 6). Having ketone (±)-**19** at hand, we thought the enolization of neoclausenamidone (±)-**19** and the consecutive reduction of enol in a one-pot reaction fashion might give rise to the desired clausenamide (±)-**1**. Hydrogenation of neoclausenamidone (±)-**19** with palladium on carbon (10%) as the catalyst, however, gave only a mixture of neoclausenamide (±)-**2** and its 6-epimer (±)-**2′**. Nevertheless, the ratio of neoclausenamide (±)-**2** over its 6-epimer (±)-**2′** was increased to 8 : 2 (Scheme 6) in comparison with that (3 : 7) of the mixture obtained directly from the intramolecular alkene-epoxide cyclization of (±)-**7**. When treated with LiOH in $CDCl_3$–D_2O (v/v 10 : 1) solution, the enolization of ketone (±)-**19** was evidenced by the ^1H NMR spectrum in which a new set of proton signals at 4.91 (d, $J = 9.7$ Hz, 1H), 3.86 (d, $J = 9.8$ Hz, 1H) and 2.87 (s, 3H) were observed (see ESI†). The quench of enolate **21** with hydrochloric acid at room temperature gave mainly the starting neoclausenamidone (±)-**19**, with only a small amount of epimer clausenamidone (±)-**20** being generated [(±)-**19** : (±)-**20** = 98 : 2]. This indicated clearly that



Scheme 6 Conversion of a mixture of (±)-**2** and (±)-**2′** into clausenamide (±)-**1** and neoclausenamide (±)-**2**.

neoclausenamidone (±)-**19** is thermodynamically more stable than clausenamidone (±)-**20**. Under kinetic reaction conditions such as at a temperature of −78 °C, protonation of enolate **21** led to an improved chemical yield of epimer (±)-**20** [(±)-**19** : (±)-**20** = 60 : 18]. Thus, clausenamidone (±)-**20** was obtained in 64% yield from neoclausenamidone (±)-**19** after two runs of protonation of enolate **21** at −78 °C (Scheme 6). The reduction of clausenamidone (±)-**20** by $NaBH_4$ in methanol furnished the desired clausenamide (±)-**1** in 92% yield (Scheme 6).

Although clausena alkaloids **1–4** were isolated from nature in racemic form, pharmacological studies indicate that different enantiopure stereoisomers exhibit varied biological activity.[1] Having established the reaction pathways for the conversion of racemic SB204900 (±)-**7** into other clausena alkaloids, we attempted the synthesis of both antipodes of five-, six- and eight-membered lactam alkaloids (Scheme 7). Starting with (2*S*,3*R*)-3-phenyloxirane-2-carboxamide (+)-**22**, which was prepared from Sharpless epoxidation of cinnamyl alcohol followed by oxidation, esterification and ammonolysis,[14] the CuI-catalyzed *N*-styrylation of amide (+)-**22** with *Z*-styryl bromide followed by *N*-methylation afforded naturally occurring (+)-SB204900 (+)-**7** in good yield. Employing the aforementioned optimized conditions, we readily converted (+)-**7** into eight-membered (−)-ζ-clausenamide (−)-**4** and (−)-homoclausenamide (−)-**3** in 86% and 62% yields, respectively. Under basic conditions, enamide (+)-**7** underwent 5-*endo* cyclization to produce a mixture of neoclausenamide and its 6-epimer in a total yield of 75%. Oxidation using Jones' reagent led to (+)-neoclausenamidone (+)-**19** in 91% yield. Hydrogenation of (+)-**19** afforded a quantitative yield of (−)-neoclausenamide (−)-**2** and its 6-epimer **2′** with the ratio being 8 : 2. Epimerization of (+)-**19** under kinetic condition through enolate intermediate gave (−)-clausenamidone (−)-**20**. $NaBH_4$ reduction of (−)-clausenamidone (−)-**20** under mild conditions yielded (−)-clausenamide



Scheme 7 Synthesis of enantiopure clausena alkaloids.

(−)-**1** in 90% yield. In all chemical transformations, no racemization of the compounds was observed. When (2*R*,3*S*)-3-phenyloxirane-2-carboxamide (−)-**22**, an enantiopure reactant prepared from nitrile biotransformations[11a] was used, the same chemical manipulations allowed us to synthesize (−)-SB204900[12a] and (+)-homoclausenamide, (+)-ζ-clausenamide,[12a] (+)-neoclausenamide and (+)-clausenamide (see ESI†).

Conclusion

In summary, we have demonstrated that *N*-methyl-*N*-[(*Z*)-styryl]-3-phenyloxirane-2-carboxamide, namely, racemic SB204900 (±)-**7**, was a common precursor to other *N*-heterocyclic alkaloids in Rutaceae *Clausena lansium* (Lour.) Skeels. Under different

conditions, (±)-**7** underwent efficient and diverse cyclization reactions to form the five-membered *N*-heterocyclic neoclausenamide and its 6-epimer, the six-membered *N*-heterocyclic homoclausenamide and the eight-membered *N*-heterocyclic ζ-clausenamide, respectively, in good to excellent yields. We have also shown that neoclausenamide and its 6-epimer were readily oxidized regiospecifically to afford neoclausenamidone. Enolization of neoclausenamidone in the presence of LiOH and the subsequent protonation under kinetic conditions at −78 °C led to the epimerization of neoclausenamidone into clausenamidone. Reduction of neoclausenamidone and clausenamidone furnished neoclausenamide and clausenamide, respectively, in excellent yields. The facile interconversions of racemic SB204900 into other clausena alkaloids and the transformation of neoclausenamide into clausenamide most probably imply the synthetic pathways of all clausena alkaloids from SB204900 in biological systems. Following the biosynthetic pathways, the concise synthesis of both antipodes of SB204900, clausenamide, neoclausenamide, homoclausenamide and ζ-clausenamide have been accomplished using (2*S*,3*R*)- and (2*R*,3*S*)-3-phenyloxirane-2-carboxamides as the starting materials. The easy availability of a wide variety of *N*-(2-arylvinyl)-3-aryloxirane-2-carboxamides should render efficient and diverse reactions powerful for the construction of natural product-like libraries for biological studies.

Experimental section

Synthesis of (+)-SB204900 (+)-7

The synthesis of (+)-SB204900 (+)-7 was based on our previous method for its enantiomer (−)-**7**.[12a] Under argon protection, a mixture of enantiopure (2*S*,3*R*)-3-phenyloxirane-2-carboxamide[14] (1 mmol, 163 mg), CuI (0.4 mmol, 76 mg), *N*,*N*-dimethylglycine hydrochloride (0.8 mmol, 112 mg), Cs_2CO_3 (2 mmol, 650 mg) and *Z*-styryl bromide (3 mmol, 549 mg) in dry 1,4-dioxane (34 mL) was refluxed for 3 h. After cooling to room temperature, ethyl acetate (100 mL) was added and the resulting mixture was filtered through a short silica gel (100–200 mesh) pad. The filtrate was concentrated and the residue was subjected to a silica gel (200–300 mesh) column eluted with a mixture of petroleum ether and ethyl acetate (10 : 1) to give (+)-**23** (201 mg, 76%) as the pure product.

To an ice-bath cooled solution of (+)-**23** (0.5 mmol, 133 mg) in dry DMF (3 mL) under argon protection was added NaH (2 mmol, 48 mg). After stirring for 0.5 h, methyl iodide (6 mmol, 0.375 mL) was added and the resulting mixture was kept stirring for another 2 h. Water (50 mL) was then added and the mixture was extracted with ethyl acetate (3 × 15 mL). The organic layer was washed with brine and dried with anhydrous Na_2SO_4, filtered and concentrated. Pure (+)-SB204900 (+)-**7** was obtained as a pale yellow oil (124 mg, 94%) from silica gel (200–300 mesh) column chromatography using a mixture of petroleum ether and ethyl acetate (3 : 1) as the mobile phase.

Synthesis of (−)-ζ-clausenamide (−)-4

The synthesis of ζ-clausenamide (−)-**4** was based on our previous method for its enantiomer (+)-**4**.[12a] A solution of (+)-**7** (0.5 mmol, 140 mg) and *p*-TSA (1 mmol, 172 mg) in dry acetonitrile (15 mL) was stirred at room temperature until the starting material was completely consumed. Water (100 mL) was added and the mixture was extracted with ethyl acetate (3 × 50 mL). The organic layer was dried with anhydrous Na_2SO_4, filtered and concentrated. The residue was subjected to a silica gel (200–300 mesh) column eluted with a mixture of petroleum ether and ethyl acetate (1 : 1) to afford pure (−)-ζ-clausenamide (−)-**4** (122mg, 86%) as a yellow oil.

Synthesis of (−)-homoclausenamide (−)-3

Our previous method for the reaction of racemic *N*-(*E*-styryl)-3-phenyloxirane-2-carboxamide (±)-**13** was adopted.[12b] A mixture of (+)-**7** (1 mmol, 279 mg) in dry *t*-BuOH (30 mL) under argon protection was heated to reflux. *p*-TSA (1 mmol, 172 mg) was then added, and the mixture was refluxed for another 1 h. After cooling to room temperature, a saturated aqueous solution of $NaHCO_3$ (30 mL) was added, and the resulting mixture was extracted with ethyl acetate (3 × 20 mL). The organic layer was dried with anhydrous Na_2SO_4, filtered and concentrated under vacuum. Pure (−)-homoclausenamide (−)-**3** (173 mg, 62%) was obtained after silica gel (200–300 mesh) column chromatography using a mixture of petroleum ether and ethyl acetate (1 : 1) as an eluant.

Intramolecular alkene-epoxide cyclization of (+)-7

Refluxing a suspension of (+)-**7** (1 mmol, 279 mg) in an aqueous Na_2CO_3 solution (2 mmol, 212 mg in 30 mL of pure water) for 5 h under argon protection gave rise to a homogeneous solution. After addition of brine (30 mL), the mixture was extracted with ethyl acetate (3 × 20 mL). The organic layer was dried with anhydrous Na_2SO_4, filtered and concentrated under vacuum. Chromatography on a silica gel (200–300 mesh) column eluting with a mixture of ethyl acetate and methanol (95 : 5) gave a mixture of enantiopure products **2** and **2′** (224 mg, 75%). The ratio of **2** over **2′**, which was determined roughly by ^{1}H NMR integration, was 3 : 7. The same mixture of racemic products was obtained from the reaction of racemic *N*-(*E*-styryl)-3-phenyloxirane-2-carboxamide (±)-**13** in boiling water.[12b]

Preparation of (+)-neoclausenamidone (+)-19

A mixture of enantiopure **2** and **2′** (**2** : **2′** = 3 : 7) (0.5 mmol, 149 mg) was dissolved in acetone (20 mL). Jones' reagent (0.5 mmol, 2.672 M, 0.187 mL) was then added while stirring. After 5 min, water (100 mL) was added and the mixture was extracted with ethyl acetate (3 × 50 mL). The combined organic layer was dried with anhydrous Na_2SO_4, filtered and concentrated. The residue was subjected to a silica gel (200–300 mesh) column eluted with a mixture of CH_2Cl_2 and ethyl acetate (1 : 1) to afford pure (+)-neoclausenamidone (+)-**19** (134 mg, 91%) as a white solid.

Synthesis of (−)-neoclausenamide (−)-2

(+)-Neoclausenamidone (+)-**19** (0.5 mmol, 148 mg) and $LiOH{\cdot}H_2O$ (1 mmol, 42 mg) were dissolved in a mixture of THF and H_2O (v/v = 1 : 1, 15 mL). After stirring at room temperature

for 30 min, Pd/C (15 mg, 10%) was added and the resulting mixture was stirred at room temperature overnight under hydrogen atmosphere using a hydrogen balloon. Water (50 mL) was added and the mixture was extracted with ethyl acetate (3 × 50 mL). The combined organic layer was dried with anhydrous Na_2SO_4, filtered and concentrated under vacuum. The residue was subjected to a silica gel (200–300 mesh) column eluted with a mixture of CH_2Cl_2 and ethyl acetate (1 : 2) to afford a mixture of (−)-neoclausenamide (−)-**2** and its 6-epimer **2′** (149 mg, 100%) as a white solid. The ratio of **2** over **2′**, which was determined by ^{1}H NMR, was 8 : 2. Recrystallization of the mixture in petroleum ether and ethyl acetate afforded pure (−)-neoclausenamide (−)-**2**.

Conversion of (+)-neoclausenamidone (+)-19 into (−)-clausenamidone (−)-20

(+)-Neoclausenamidone (+)-**19** (0.5 mmol, 148 mg) and $LiOH{\cdot}H_2O$ (2 mmol, 84 mg) were mixed in THF and H_2O (v/v = 1 : 1, 10 mL). After stirring at room temperature for 12 h, the mixture was cooled to −78 °C and stirred at −78 °C for another 12 h. Then, a solution of HCl in THF (2 M, 1 mL) was added using a syringe at −78 °C. The temperature of the reaction mixture was then raised gradually to room temperature within about 2 h. Water (50 mL) was added and the mixture was extracted with ethyl acetate (3 × 50 mL). The combined organic layer was dried with anhydrous Na_2SO_4, filtered and concentrated under vacuum. The residue was subjected to a silica gel (200–300 mesh) column eluted with a mixture of CH_2Cl_2 and ethyl acetate (1 : 1) to afford pure (−)-clausenamidone (46 mg) as a white solid. The recovered (+)-neoclausenamidone (102 mg) was subjected to epimerization again to give another 44 mg of pure (−)-clausenamidone (−)-**20**. The combined yield was 61%.

Synthesis of (−)-clausenamide (−)-1

The reduction of (−)-clausenamidone (−)-**20** followed a literature method.[8g] To a solution of (−)-clausenamidone (−)-**20** (0.5 mmol, 148 mg) in methanol (10 mL) was added $NaBH_4$ (0.5 mmol, 20 mg) while stirring at room temperature. Monitored by TLC, the reactant was completely consumed in 2 h. Water (10 mL) and 2 mL of hydrochloric acid (2 M) were added consecutively to decompose reducing agent $NaBH_4$. The mixture was neutralized by aqueous NaOH solution (1 M) and extracted with ethyl acetate (3 × 50 mL). The combined organic layer was dried with anhydrous Na_2SO_4, filtered and concentrated. The residue was subjected to a silica gel (200–300 mesh) column eluted with a mixture of methanol and ethyl acetate (5 : 95) to afford pure (−)-clausenamide (−)-**1** (133 mg, 90%) as a white solid.

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