Organic LETTERS

Letter

pubs.acs.org/OrgLett

One-Step Semisynthesis Method of Spirocurcasone and Pyracurcasone from Curcusones A and B

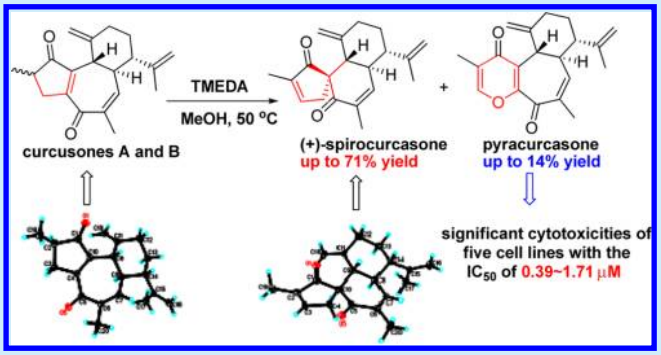
Xu-Yang Li,[†,‡] Yuan-Feng Yang,[†,‡] Xing-Rong Peng,[†,‡] Ming-Ming Li,[†] Liang-Qun Li,[†,‡] Xu Deng,[†] Hong-Bo Qin,*[,†,‡] Jie-Qing Liu,*[,†] and Ming-Hua Qiu*[,†,‡]

[†]State Key Laboratory of Photochemistry and Plant Resources in West China, Kunming Institute of Botany, Chinese Academy of Sciences, Kunming 650201, Yunnan People's Republic of China

[‡]University of Chinese Academy of Sciences, Beijing 100049, People's Republic of China

Supporting Information

ABSTRACT: High contents of curcusones A and B and trace amounts of spirocurcasone exist in the roots of *Jatropha curcas*. Here, a one-step semisynthesis method of spirocurcasone and pyracurcasone was built, not only resulted an increased yield of spirocurcasone but also produced pyracurcasone, which exhibited greater cytotoxicity compared to curcusones A and B. The plausible mechanism of the formation of pyracurcasone was proposed, and the proposed biogenetic origin for spirocurcasone by Taglialatela-Scafati was confirmed.



Jatropha curcas L. is a key raw material for the manufacturing of biodiesel all over the world.[1] Meanwhile, it contains many types of diterpenes, including tigliane, lathyrane, rhamnofolane, and so on, which have attracted a lot of attention.[2−4] Curcusones A and B, belonging to rhamnofolane-type diterpenes, are the main constituents in the roots of *J. curcas*[5] and show significant cytotoxic activity.[6,7]

In 2011, a diterpene with a "spirorhamnofolane" skeleton was first isolated by Taglialatela-Scafati in the root barks of *J. curcas* and was subsequently named spirocurcasone, and its possible biogenetic origin from rhamnofolane diterpene was also proposed.[8] After 2 years, Hisanaka-Ito and co-workers reported the total synthesis of (+)- and (−)-spirocurcasone in nine steps and suggested that the optical rotation value reported by Taglialatela-Scafati was inaccurate.[9] The absolute configuration of spirocurcasone in the two above-mentioned reports was determined by quantum mechanical ECD calculations.[10] Investigations into the 5/6/6 (ABC)-ring system (Figure 1) of the spiro system were rare, and the synthesis of the ABC-spiro system was reported by employing many steps.[11−13] This paper reports a simple, efficient, and stereospecific synthesis method to the ABC-spiro system of (+)-spirocurcasone **1** from curcusones A and B (**2** and **3**). The structures of **1**−**3** are shown in Figure 1. We found that compound **2** or **3** could be converted to **1**[14] under alkali conditions. Interestingly, another byproduct **4**,[15] named pyracurcasone, was produced when methanol was the solvent. In addition, compound **4** exhibited better activities against five tumor cell lines compared to **2** and **3** (see Table 2). To get ketimine derivatives of **2**, as part of our structure−activity relationship (SAR) study, dehydration conditions (Table 1, entry 1) were utilized. Compound **1** was isolated instead, although the yield was only 12%. This phenomenon attracted our attention to improve its yield. We optimized the reaction condition by changing solvents, reaction temperature, base, and catalyst (Table 1). We found that compound **1** could be produced whether the catalyst existed or not (entries 7−13). The yield jumped from 10 (entry 5) to 35% (entry 10), possibly due to better solubility in MeOH. To our satisfaction, the use of 2 equiv of *N*,*N*,*N*′,*N*′-tetramethylethylenediamine (TMEDA) and methanol at 50 °C could improve the yield of **1** (71%) and produce **4** (entry 13). Usually, bidentate amine TMEDA is used to coordinate with metals such as Li[16] or Mg.[17] Due to an absence of metal, the effect of TMEDA in this reaction may be ascribed to its appropriate basicity (lower than DBU, entry 11), and the fact is

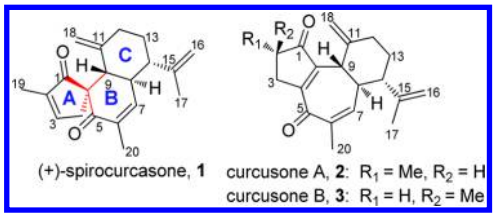
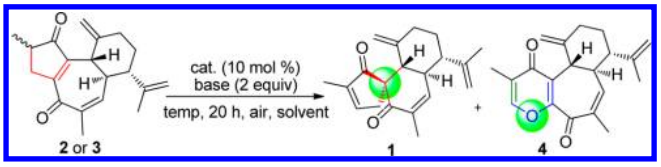


Figure 1. Structures of (+)-spirocurcasone (**1**) and curcusones A and B (**2** and **3**).

Received: March 5, 2014
Published: April 3, 2014

   dx.doi.org/10.1021/ol500692j | *Org. Lett.* 2014, 16, 2196−2199

Table 1. Screening of Reaction Conditions[a]



entry	material	solvent	temp (°C)	base (2 equiv)	catalyst	yield[b] of **1** (%)	yield[b] of **4** (%)
1	**2**	toluene	reflux	DEAEA	pTSA	12	NR
2	**2**	toluene	reflux	benzylamine	pTSA	NR	NR
3	**2**	toluene	reflux	cyclohexylamine	DMAP	<5	NR
4	**2**	DCM	reflux	DEAEA	DMAP	<5	NR
5	**3**	toluene	reflux	K_2CO_3	TBAB	10	NR
6	**3**	toluene	reflux	NaOH	TBAB	NR	NR
7	**3**	toluene	reflux	Et_3N		8	NR
8	**3**	toluene	reflux	*t*-BuOK		10	NR
9	**3**	toluene	90	piperidine		NR	NR
10	**3**	MeOH	50	K_2CO_3		35	NR
11	**3**	MeOH	50	DBU		38	NR
12	**3**	MeOH	50	4-DMAP		20	NR
13	**3**	MeOH	50	TMEDA		71	14

[a]Reaction conditions: **2** or **3** (1 equiv), base (2 equiv), solvent (0.3 M), under air atmosphere, 20 h, heated. [b]Isolated yield. NR = no reaction.

that it has two nitrogen atoms in one molecule, which means two-fold base is used.

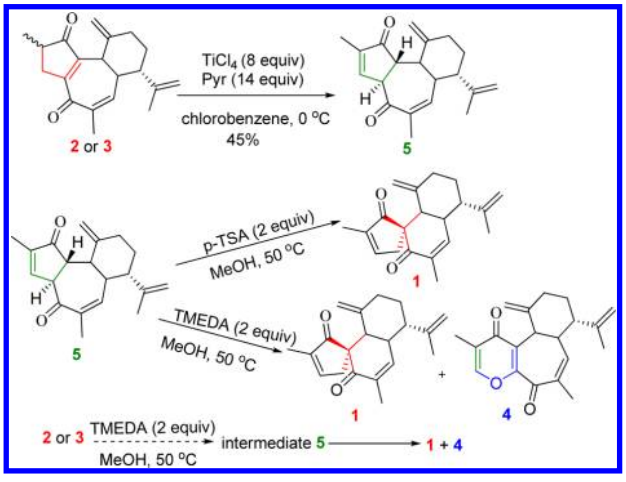
Due to the high yield of **1**, we obtained the X-ray crystal structure of **1** for the first time (Figure 2). The optical rotation

Figure 2. X-ray crystal structures of **1** and **3**.

of **1** was $[\alpha]_D^{23}$ +155.3, which was consistent with the synthetic compound reported by Hisanaka-Ito and confirmed the inaccuracy of the optical rotation value reported in 2011 again. Due to the absolute configuration of **1** and the X-ray crystal structure (Figure 2) as well as the optical rotation value (see Supporting Information) of compound **3**, we conclude that the absolute configuration of compounds **2** and **3** reported by Clardy et al. is inaccurate.[18]

To confirm the possible precursor of **1** proposed by Taglialatela-Scafati, we tried to achieve the proposed precursor **5**. As shown in Scheme 1, the transference of the double bond of compound **2** or **3** was accomplished by using 14 equiv of pyridine and 8 equiv of titanium tetrachloride to yield **5** (45%). Compound **5** was treated with *p*-toluenesulfonic acid in methanol to produce **1**. As we expected, a solution of **5** in methanol was treated with the optimized reaction conditions (Table 1, entry 13) to give **1** and **4**. Interestingly, when compounds **2** and **3** reacted under the same reaction conditions, we found intermediate **5** by TLC (see Supporting Information). As a result, we proved the proposed biosynthetic pathway of compound **1**. Natural product **2** or **3** can be converted to intermediate **5**, then **5** turned into natural compound **1** by a process of [1,5]-sigmatropic alkyl shift. The stereospecific formation of **1** could be rationalized by the conformation analysis of **5**, which has α-H on C-4 and β-H on

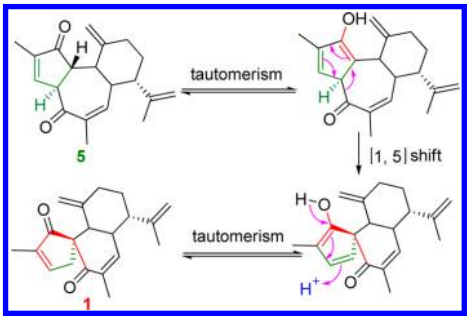
Scheme 1. Synthesis of Compounds 1 and 4 from 5



C-10 (see Supporting Information). The [1,5]-shift occurred thermodynamically to get **1** (Scheme 2).

After making clear the formation of **1**, concomitantly producing the non-normal compound **4** raised our interest to investigate the mechanism of its formation. When compound **2**, **3**, or **5** were treated with the optimized reaction conditions to

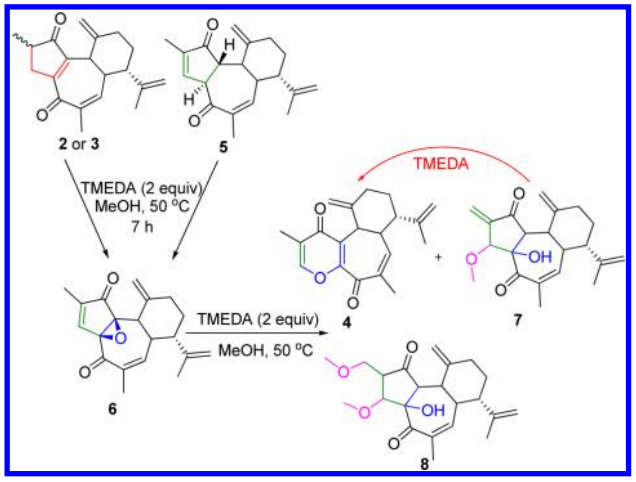
Scheme 2. Stereospecific Rearrangement of the Rhamnofolane 5



dx.doi.org/10.1021/ol500692j | Org. Lett. 2014, 16, 2196–2199

produce **4**, we found intermediate **6** after reacting for 1 h, but it disappeared after reacting for 20 h. On the basis of this phenomenon, we controlled the reaction time to achieve some intermediates (Scheme 3). Intermediate **6** was isolated after

Scheme 3. Intermediates of the Formation of Compound 4



reacting for 7 h. The absolute configuration of the epoxy group in **6** was deduced as β-epoxy by the optical rotation (OR) computational methods by using the density functional theory (DFT) studies[19] in the Gaussian 09 program package (see Supporting Information). When **6** was treated with the same reaction conditions, compounds **4**, **7**, and **8** were isolated. As far as we know, **8** would be derived from **7** by Michael addition with methanol. Compound **7** could convert to **4** under the same conditions, which confirmed that compounds **6** and **7** are important intermediates in the formation of **4**. Based on these elements, the plausible reaction mechanism of the formation of compound **4** is proposed in Scheme 4.

The cytotoxicity of **1**−**4** was assessed against HL-60, SMMC-7721, A-549, MCF-7, and SW480 human cancer cells. Cisplatin (DDP) was used as a positive control in this experiment. The IC_{50} values of individual compounds against five cell lines are presented in Table 2. As a result, compound **1** did not show

Scheme 4. Plausible Reaction Mechanism of the Formation of Compound 4

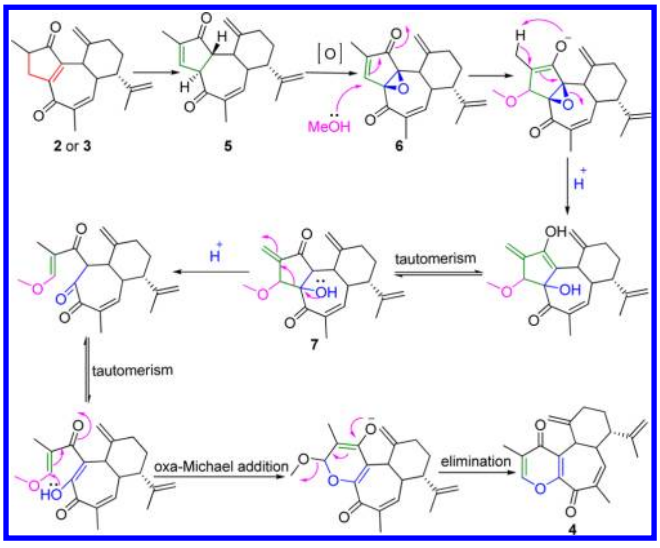


Table 2. In Vitro Cytotoxicity of Compounds 1−4 with IC_{50} Values (μM)

	HL-60	SMMC-7721	A-549	MCF-7	SW480
1	>40	>40	>40	>40	>40
2	1.63	3.10	3.35	2.47	2.10
3	2.64	3.30	3.88	3.14	2.91
4	1.71	1.08	0.82	0.68	0.39
DDP	1.14	14.51	12.76	19.61	17.54

any activity against test cell lines, while compound **4** had a great increase in the activity against SMMC-7721, A-549, MCF-7, and SW480 compared to **2** and **3**. Especially, the activity against SW480 of **4** was 7 times more potent than that of **3**.

In conclusion, we have found a one-step and efficient approach to form stereospecific **1** and cytotoxic **4** and demonstrated the biosynthetic pathway of **1**. Currently, we are focusing on developing this method into a general approach to construct a spiro-ring system. Meanwhile, the application of our method to the total synthesis of bioactive natural compounds is underway in our laboratory. The accurate absolute configuration of compounds **1**−**3** is also confirmed, and the X-ray crystal structure of **1** is reported for the first time. In addition, we have also proposed the plausible reaction mechanism of the formation of **4**. Furthermore, the significant improvement of antitumor activity of compound **4** makes it a good lead compound for further SAR investigation.

■ ASSOCIATED CONTENT

Supporting Information

General experimental procedures, X-ray crystal structure of **1** and **3**, detection of intermediate **5** by TLC, absolute configuration of the epoxy group of **6**, and spectroscopic data of compounds **1**−**8**. This material is available free of charge via the Internet at http://pubs.acs.org.

■ AUTHOR INFORMATION

Corresponding Authors

*E-mail: mhchiu@mail.kib.ac.cn (M.-H. Qiu).
*E-mail: liujieqing@mail.kib.ac.cn (J.-Q. Liu).
*E-mail: qinhongbo@mail.kib.ac.cn (H.-B. Qin).

Notes

The authors declare no competing financial interest.

■ ACKNOWLEDGMENTS

The project was financially supported by the General Program of NSFC (No. 81202437) and Joint Fund of NSFC and NSFY (No. U1132604), Pillar Program of MOST (No. 2007BAD32B03), as well as Foundation of State Key Laboratory of Phytochemistry and Plant Resources in West China (P2010-ZZ14). The authors are also grateful to Jonathan Teichroew and Yu Zhang from the Kunming Institute of Botany, Chinese Academy of Sciences, for editing the manuscript for English and the calculation study, respectively.

■ REFERENCES

(1) Achten, W. M. J.; Nielsen, L. R.; Aerts, R.; Lengkeek, A. G.; Kjaer, E. D.; Trabucco, A.; Hansen, J. K.; Maes, W. H.; Graudal, L.; Akinnifesi, F. K.; Muys, B. *Biofuels* **2010**, *1*, 91−107.
(2) Liu, J. Q.; Yang, Y. F.; Wang, C. F.; Li, Y.; Qiu, M. H. *Tetrahedron* **2012**, *68*, 972−976.

(3) Yang, Y. F.; Liu, J. Q.; Shi, L.; Li, Z. R.; Qiu, M. H. *Nat. Prod. Bioprospect.* **2013**, *3*, 99−102.
(4) Devappa, R. K.; Makkar, H. P. S.; Becker, K. *J. Am. Oil Chem. Soc.* **2011**, *88*, 301−322.
(5) Zhang, X.; Wang, M.; Zhao, Z.; Liu, X. *Huaxue Yanjiu Yu Yingyong* **2011**, *23*, 87−91.
(6) Aiyelaagbe, O. O.; Hamid, A. A.; Fattorusso, E.; Taglialatela-Scafati, O.; Schroder, H. C.; Muller, W. E. G. *J. Evidence-Based Complementary Altern. Med.* **2011**, 134954.
(7) Liu, J. Q.; Yang, Y. F.; Li, X. Y.; Liu, E. Q.; Li, Z. R.; Zhou, L.; Li, Y.; Qiu, M. H. *Phytochemistry* **2013**, *96*, 265−272.
(8) Chianese, G.; Fattorusso, E.; Aiyelaagbe, O. O.; Luciano, P.; Schroder, H. C.; Muller, W. E. G.; Taglialatela-Scafati, O. *Org. Lett.* **2011**, *13*, 316−319.
(9) Abe, H.; Sato, A.; Kobayashi, T.; Ito, H. *Org. Lett.* **2013**, *15*, 1298−1301.
(10) Stephens, P. J.; Pan, J. J.; Devlin, F. J.; Krohn, K.; Kurtan, T. *J. Org. Chem.* **2007**, *72*, 3521.
(11) Paquette, L. A.; Hong, F. T. *J. Org. Chem.* **2003**, *68*, 6905−6918.
(12) Paquette, L. A.; Hu, Y.; Luxenburger, A.; Bishop, R. L. *J. Org. Chem.* **2007**, *72*, 209−222.
(13) Srikrishna, A.; Pardeshi, V. H.; Thriveni, P. *Tetrahedron: Asymmetry* **2008**, *19*, 1392−1396.
(14) (+)-Spirocurcasone (**1**): $C_{20}H_{24}O_2$, colorless crystal; $[\alpha]_D^{23}$ +155.3 (*c* 2.5 MeOH); UV (MeOH) λ_{max} (log ε) 233 (4.14) nm; IR (KBr) ν_{max} 2938, 2864, 1715, 1667, 1639, 1444, 1378, 1327, 1291, 1104, 1026, 903 cm^{-1}; HREIMS *m*/*z* 296.1773 (calcd for $C_{20}H_{24}O_2$, 296.1776); CD (Figure S1); for ^1H and ^{13}C NMR spectra, see Supporting Information.
(15) Pyracucasone (**4**): $C_{20}H_{22}O_3$, yellow oil; $[\alpha]_D^{24}$ −209.6 (*c* 2.1 MeOH); UV (MeOH) λ_{max} (log ε) 231(3.78), 201 (3.92) nm; IR (KBr) ν_{max} 3433, 2928, 1686, 1644, 1450, 1414, 1377, 1333, 1273, 1190, 1122, 892 cm^{-1}; HREIMS *m*/*z* 310.1560 (calcd for $C_{20}H_{22}O_3$ 310.1569); for ^1H and ^{13}C NMR spectra, see Supporting Information.
(16) Gao, L.; Lu, J.; Song, Z. L.; Lin, X. L.; Xu, Y. J.; Yin, Z. P. *Chem. Commun.* **2013**, *49*, 8961−8963.
(17) Bugarin, A.; Connell, B. T. *J. Org. Chem.* **2009**, *74*, 4638−4641.
(18) Naengchomnong, W.; Thebtaranonth, Y.; Wiriyachitra, P.; Okamoto, K. T.; Clardy, J. *Tetrahedron Lett.* **1986**, *27*, 2439−2442.
(19) (a) Zhang, Y.; Di, Y. T.; Zhang, Q.; Mu, S. Z.; Tan, C. J.; Fang, X.; He, H. P.; Li, S. L.; Hao, X. *J. Org. Lett.* **2009**, *11*, 5414−5417. (b) Tang, G. H.; He, H. P.; Gu, Y. C.; Di, Y. T.; Wang, Y. H.; Li, S. F.; Li, S. L.; Zhang, Y.; Hao, X. J. *Tetrahedron* **2012**, *68*, 9679−9684.