COMMUNICATION

ChemComm
www.rsc.org/chemcomm

# Synthesis of $^{13}$C-labelled, bicyclic mimetics of natural enediynes

**Parthasarathi Das, Takashi Mita, Martin J. Lear and Masahiro Hirama***

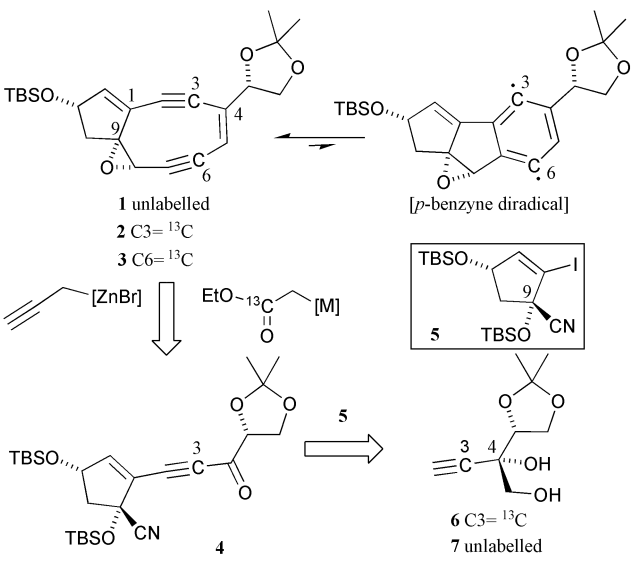
*Department of Chemistry, Graduate School of Science, Tohoku University, and CREST, Japan Science and Technology Corporation (JST), Sendai 980-8578, Japan. E-mail: hirama@ykbsc.chem.tohoku.ac.jp; Fax: 81 22 217 6566; Tel: 81 22 217 6563*

***Received (in Cambridge, UK) 13th September 2002, Accepted 9th October 2002***
***First published as an Advance Article on the web 23rd October 2002***

**Using a versatile synthesis with $^{13}CH_3PPh_3I$ and $CH_3{}^{13}CO_2Et$ as $^{13}$C sources, the first examples of nine-membered chromophores which have been differentially labelled with $^{13}$C in their carbocyclic enediyne cores are described.**

With their complex antitumour behaviours (nucleic acid/protein damage, *p*-diradical/*p*-quinone formation) and complex structures, the chemistry and biology of the enediyne-class of antitumour antibiotics has fascinated and challenged researchers for well over a decade now.[1–3] Yet, despite several mechanistic studies, it can be argued that the reactive diradical intermediate which results from the cycloaromatisation of a natural enediyne has never been proven unambiguously.[3] To address this issue, we have completely re-formulated our former synthesis of the core structure (**1**) of the kedarcidin chromophore (like kedarcidin which exhibits paramagnetic behaviour)[3b,4] and herein provide carbobicyclic models (**2**, **3**) labelled with $^{13}$C at either the 3 or 6 position for the precise characterisation of *p*-benzyne and other radical species through EPR measurements (Scheme 1).

In order to incorporate $^{13}$C into the said strategic positions of the enediyne **1** in an atom-economical fashion, the goal here was to devise a strategy that would not only be flexible to both isotopomers **2** and **3** but also be amenable to the late stage introduction of carbon-13. However, reliable synthetic routes to epoxybicyclo[7.3.0]dodecenediyne frameworks are exceedingly difficult to realise due to product instability.[2,5] For the structural case at hand, we elected to pursue a new approach through the ynone **4** (Scheme 1). Apart from the practical availability of the iodocyclopentene **5**[5] ($\alpha/\beta$ = 5 at C9), two points should be made: (1) the ynone **4** was anticipated to be more stable than those that had been used previously,[5,6] and (2) the coupling of **5** with acetylenic fragments like **6** or **7**[7] was envisaged to give broad scope in carbon-13 labelling studies.

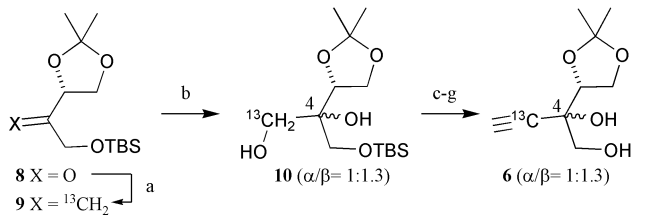



**Scheme 1** Versatile retrosynthesis of **1**, **2** and **3**.

After extensive studies with non-labelled components, a streamlined synthesis of the C3, carbon-13 labelled system **2** was realised as follows. First, the $^{13}$C-labelled component **6** was prepared from **8**[8] through an efficient, gram-scale synthesis via compounds **9** and **10** (Scheme 2). Here, it should be noted that the Wittig reaction between the erythrulose derivative **8** (2 equiv.) and $^{13}CH_3PPh_3I$ (1 equiv.)[9] occurred in quantitative yields and that the stereochemistry of the C4 position was superfluous to our planned synthesis of **2**, since it would be oxidatively destroyed in the formation of the ynone **4** (*cf.* Scheme 1).

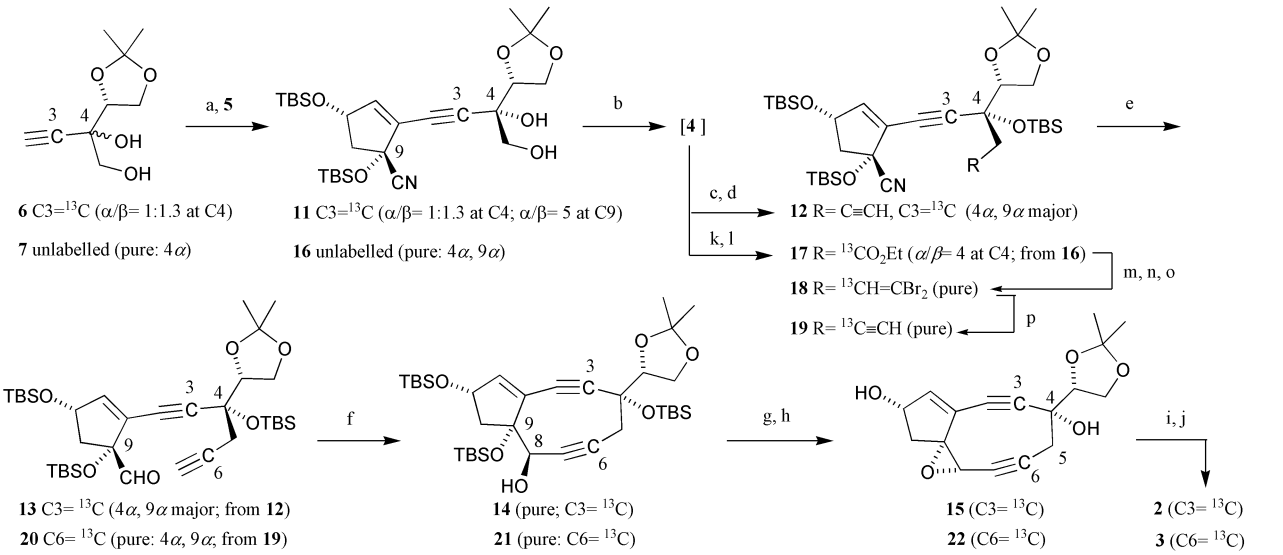
As shown in Scheme 3, using well-established Sonogashira-type conditions on **5** and $^{13}$C-labelled **6**,[2c,5,10] the diol **11** was obtained in 72% yield as a four-component diastereomeric mixture at C4 ($\alpha/\beta$ = 1 : 1.3) and C9 ($\alpha/\beta$ = 5 at C9. Oxidative cleavage of **11** with $NaIO_4$ then cleanly furnished the desired ynone **4** ($\alpha/\beta$ = 5 at C9) which proved to be stable to both silica-gel chromatography and storage. Treatment of **4** with propargyl zinc bromide and TBS ether protection (under anionic conditions)[11] permitted the nitrile **12** to be isolated as its major diastereomer (4α, 9α) in 46% overall yield.† DIBAL reduction of the nitrile **12** afforded the cyclisation precursor **13**† in excellent yield, which was immediately subjected to the typical $CeCl_3$-mediated acetylide cyclisation protocol at −25 °C to generate the *trans*-diol **14** in 45–52% isolated yield.[2c,4,5] Mesylation of **14** followed by global desilylation then afforded the epoxide **15** in 76% yield over two steps. Finally, **15** was silylated selectively at the C11-allylic alcohol and then, in a one-pot reaction, mesylated at 5 °C and treated with DBU at −40 °C to produce the fully-fledged, $^{13}$C-labelled epoxydienediyne **2** in high purities and in yields of about 45–50%.‡

As indicated in our retrosynthetic analysis (Scheme 1), the incorporation of carbon-13 into the C6-position would necessitate a simple modification of the aforementioned route. Instead of using propargylic zinc, carbon-13 incorporation was thus achieved by the anionic-addition of labeled ethyl acetate ($CH_3{}^{13}CO_2Et$) onto the enantiopure ynone **4**, that was derived from the readily available synthon **7**[7] (Scheme 3). After *O*-TBS silylation, **17** was subsequently obtained as a 4 : 1 $\alpha/\beta$ mixture at C4 in a 66% yield over two steps. Following the chemoselective reduction of the ester functionality in the presence of the nitrile group and re-oxidation, the dibromoalkene **18** was generated




**Scheme 2** *Reagents and conditions*: (a) $^{13}CH_3PPh_3I$, $NaN(TMS)_2$, THF, −78 °C to rt, 1 h, quantitative; (b) $OsO_4$ (0.01 eq.), NMO (2.5 eq.), acetone–$H_2O$ (4 : 1), 12 h, 86%; (c) $SO_3$·Py (3 eq.), $Et_3N$ (15 eq.), DMSO, 10 min, rt; (d) TBSOTf (3 eq.), 2,6-lutidine (4 eq.), $CH_2Cl_2$, 1 h, rt; (e) $CBr_4$ (3 eq.), $Ph_3P$ (6 eq.), $CH_2Cl_2$, 1 h rt; (f) *n*-BuLi (2.2 eq.), THF, −78 °C, 1 h; (g) TBAF (3 eq.), THF, 1 h, rt, 63% for five steps.

DOI: 10.1039/b208939d




**Scheme 3** *Reagents and conditions*: (a) **5** (1 eq.), $Pd_2(dba)_3{\cdot}CHCl_3$ (0.05 eq.), CuI (0.1 eq.), DMF–*i*-$Pr_2NEt$ (2:1), rt, 1 h, 72% for **11**, 40% for **16** (after crystallisation); (b) $NaIO_4$ (5 eq.), $CH_3OH$–$H_2O$ (4:1), rt, 1 h, 84%; (c) Zn (3 eq.), propargyl bromide (3 eq.), THF, −15 °C, 1 h; (d) TBSCl (1.2 eq.), KH (3 eq.), 18-crown-6 (0.05 eq.), THF, 0 °C, 5 min, 46% for two steps; (e) DIBAL (1.5 eq.), $CH_2Cl_2$, −78 °C, 1 h; (f) $CeCl_3$ (25 eq.), $LiN(TMS)_2$ (26 eq.), THF, −25 °C to rt, 1 h, 45–52% for **14**, or 68–72% for **21**, for two steps; (g) MsCl (3 eq.), $Et_3N$ (6 eq.), $CH_2Cl_2$, rt, 1 h; (h) TBAF (5 eq.), THF, rt, 10 min, 70–76% for **15** or **22**, for two steps; (i) TBSOTf (3 eq.), 2,6-lutidine (6 eq.), $CH_2Cl_2$, −78 °C, 10 min; (j) MsCl (5 eq.), $Et_3N$ (10 eq.), $CH_2Cl_2$, 5 °C, 30 min, then DBU (2 eq.), $CD_2Cl_2$, −40 °C, then rt, 30 min, 45–50% for **2** or **3**, for two steps; (k) $CH_3{}^{13}CO_2Et$, $LiN(SiMe_3)_2$ (1.1 eq.), THF, −78 °C, 1 h, 78%; (l) TBSOTf (3 eq.), 2,6-lutidine (6 eq.), $CH_2Cl_2$, rt, 10 h, 85%; (m) DIBAL (4 eq.), THF, −78 to −25 °C, 2 h, 70%; (n) Dess–Martin periodinane (1.5 eq.), $CH_2Cl_2$, 1 h, 80%; (o) $CBr_4$ (3 eq.), $Ph_3P$ (6 eq.), $CH_2Cl_2$, 0 °C, 30 min, then $SiO_2$ separation; (p) *n*-BuLi (2 eq.), THF, −78 °C, 58% for two steps.

and isolated in enantiopure form after silica-gel chromotography. Completion of this oxidation-homologation sequence then gave the desired C6–carbon-13 labelled alkyne **19** in 22% yield over six steps from **4**. Lastly, under a parallel reaction sequence to that described for **2**, the targeted epoxyenediyne **3** was generated from **19** (*via* the C6-labelled intermediates **20**–**22**) in a 22% overall yield over six steps.‡

In closing, we should first point out that for all isotopomers **2** and **3** the nine-membered cyclization steps to **14** or **21** have been reliably performed in yields that reflect the diastereomeric purity of the cyclization precursors **13** or **20**, *i.e*. 45 to 72%. Second, the routes described herein are highly expeditious and practical, particulary considering the high lability of the compounds involved. For example, the unlabelled enediyne **1**[4] can now be prepared in a 10–15% overall yield from enantiopure **7**[11] over the shortest 10-step sequence. Comprehensive EPR characterisation studies on the radical cycloaromatised states of **1**–**3** are currently being finalised and will be detailed elsewhere.

A postdoctoral CREST fellowship (to P. D.) from the Japan Science and Technology Corporation (JST) and a fellowship (to T. M.) from the Japanese Society for the Promotion of Science for Young Japanese Scientists are gratefully acknowledged.

## Notes and references

† **12** and **13** were isolated and used as (4α, 9α):(4β, 9β) ⩾ 10:1 mixtures.

‡ *NMR data* for **2** and **3** (500 MHz, $CD_2Cl_2$, ref. 5.32 ppm for $^1H$ and 53.10 ppm for $^{13}C$).

**2**: $^1H$ NMR: $\delta$ 0.11 (s, 3H), 0.11 (s, 3H), 0.90 (s, 9H), 1.37 (s, 3H), 1.46 (s, 3H), 1.86 (dd, 1H, *J* 15.0, 2.5 Hz), 2.71 (dd, 1H, *J* 15.0, 7.3 Hz), 3.75 (d, 1H, *J* 1.5 Hz), 3.80 (dd, 1H, *J* 8.3, 6.7 Hz), 4.17 (dd, 1H, *J* 8.3, 6.7 Hz), 4.63 (ddt, *J* 6.7, 1.5 Hz, $^3J_{C-H}$ 5.0 Hz), 5.00 (ddd, *J* 7.3, 2.5 Hz, $^5J_{C-H}$ 1.5 Hz), 5.99 (dt, *J* 1.5 Hz, $^3J_{C-H}$ 12.5 Hz), 6.40 (br d, *J* 2.0 Hz).

$^{13}C$ NMR: $\delta$ −5.41, −5.34, 17.65, 25.10, 25.17, 25.74, 38.60, 49.81, 68.82 (d, $J_{C-C}$ 1.4 Hz), 69.77 (d, $J_{C-C}$ 2.8 Hz), 72.07, 75.07 (d, $J_{C-C}$ 3.9 Hz), 88.38 (d, $J_{C-C}$ 2.9 Hz), 97.18 ($^{13}C$ label), 102.42 (d, $J_{C-C}$ 182.6 Hz), 104.52, 110.02, 119.70 (d, $J_{C-C}$ 1.9 Hz), 124.63 (d, $J_{C-C}$ 11.4 Hz), 143.00 (d, $J_{C-C}$ 89.6 Hz), 145.47 (d, $J_{C-C}$ 4.3 Hz).

**3**: $^1H$ NMR: $\delta$ 0.11 (s, 3H), 0.11 (s, 3H), 0.90 (s, 9H), 1.37 (s, 3H), 1.46 (s, 3H), 1,86 (dd, 1H, *J* 15.0, 2.5 Hz), 2.72 (dd, 1H, *J* 15.0, 7,3 Hz), 3.75 (dd, 1H, *J* 1.5 Hz, $^2J_{C-H}$ 3.7 Hz), 3.80 (dd, 1H, *J* 8.5, 6.7 Hz), 4.17 (dd, 1H, *J* 8.5, 6.5 Hz), 4.63 (bt, 1H, *J* 6.5 Hz), 5.00 (dt, *J* 7.3, 2.5 Hz), 5.99 (br d, *J* 1.5 Hz), 6.40 (d, *J* 2.5 Hz).

$^{13}C$ NMR: $\delta$ −5.41, −5.34, 17.65, 25.09, 25.17, 25.74, 38.60, 49.80 (d, $J_{C-C}$ 14.4 Hz), 68.82, 69.76 (d, $J_{C-C}$ 2.8 Hz), 72.07, 75.07 (d, $J_{C-C}$ 6.1 Hz), 88.38 ($^{13}C$ label), 97.17 (d, $J_{C-C}$ 6.3 Hz), 102.36, 104.55 (d, $J_{C-C}$ 193.9 Hz), 110.02, 118.95 (d, $J_{C-C}$ 95.8 Hz), 124.62, 143.01 (d, $J_{C-C}$ 3.4 Hz), 145.46.


1 Recent reviews: (*a*) J. W. Grissom, G. U. Gunawardena, D. Klingberg and D. Huang, *Tetrahedron*, 1996, **52**, 6453; (*b*) Z. Xi and I. H. Goldberg, *Compr. Nat. Prod. Chem.*, 1999, **7**, 553: enediyne-induced cleavage of RNA (*c*) J.-M. A. Battigello, M. Cui, S. Roshong and B. J. Carter, *Bioorg. Med. Chem.*, 1995, **6**, 839; inhibition of DNA-repair enzymes (*d*) W. Solomon, K. L. Colson and D. R. Schroeder, *Biochemistry*, 1995, **34**, 11591; M. Hashimoto, M. M. Greenberg, Y. W. Kow, J.-T. Hwang and R. P. Cunningham, *J. Am. Chem. Soc.*, 2001, **123**, 3161; cytotoxic action of an enediyne-derived quinone species (*e*) L. H. Jones, C. W. Harwig, P. Wentworth Jr., A. Simeonov, A. D. Wentworth, S. Py, J. A. Ashley, R. A. Lerner and K. D. Janda, *J. Am Chem. Soc.*, 2001, **123**, 3607.

2 Latest advances in the synthesis of fully-fledged, nine-membered enediyne chromophores: (*a*) A. G. Myers, R. Glatthar, M. Hammond, P. M. Harrington, E. Y. Kuo, J. Liang, S. E. Schaus, Y. Wu and J.-N. Xiang, *J. Am. Chem. Soc.*, 2002, **124**, 5380; (*b*) A. G. Myers, P. C. Hogan, A. R. Hurd and S. D. Goldberg, *Angew. Chem., Int. Ed.*, 2002, **41**, 1062; (*c*) S. Kobayashi, S. Ashizawa, Y. Takahashi, Y. Sugiura, M. Nagaoka, M. J. Lear and M. Hirama, *J. Am. Chem. Soc.*, 2001, **123**, 11294.

3 Recent study of a specialised *p*-benzyne diradical: (*a*) H. H. Wenk A. Baster, W. Sander, D. A. Hrovat and W. T. Borden, *Angew. Chem., Int. Ed.*, 2001, **40**, 2295; for direct evidence that C-1027 and kedarcidin form paramagnetic radical species, see (*b*) M. Hirama, K. Akiyama, T. Tanaka, T. Noda, K.-I. Iida, I. Sato, R. Hanaishi, S. Fukuda-Ishisaka, M. Ishiguro, T. Otani and J. E. Leet, *J. Am. Chem. Soc.*, 2000, **122**, 720.

4 K.-I. Iida and M. Hirama, *J. Am. Chem. Soc.*, 1995, **117**, 8875.

5 For routes to partly-completed, nine-membered enediynes, see: K. Toyama, S. Iguchi, H. Sakazaki, T. Oishi and M. Hirama, *Bull. Chem. Soc. Jpn.*, 2001, **74**, 997 and references therein.

6 G. X. Wang, S. Iguchi and M. Hirama, *J. Org. Chem.*, 2001, **66**, 2146.

7 S. Kobayashi, P. Das, X. Wang, T. Mita, M. J. Lear and M. Hirama, *Chem. Lett.*, 2002, 300.

8 K. Nakatani, K. Arai, N. Hirayama, F. Matsuda and S. Terashima, *Tetrahedron*, 1992, **48**, 633.

9 C. D. Poulter, M. Muehlbacher and D. R. Davis, *J. Am. Chem. Soc.*, 1989, **111**, 3740.

10 K. Sonogashira, Y. Tohda and N. Hagihara, *Tetrahedron Lett.*, 1975, **50**, 4467.

11 T. F. Braish and P. L. Fuchs, *Synth. Commun.*, 1986, **16**, 111.