A concise total synthesis of phytotoxic radulanin A facilitated by the photochemical ring expansion of a 2,2-dimethylchromene in flow

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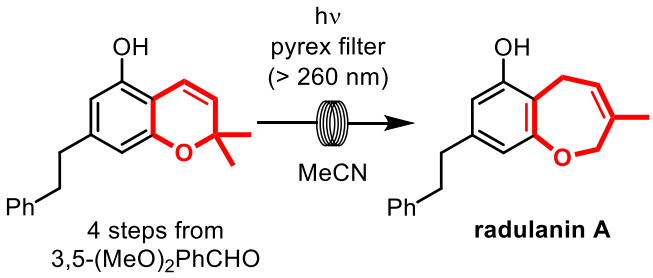

ABSTRACT: The radulanins are biologically active bibenzyl natural products featuring a synthetically challenging 2,5-dihydro-1-benzoxepine core. In contrast with previous reports exhibiting lengthy strategies, we demonstrate the shortest synthesis of radulanin A to date, featuring a largely unexplored photochemical ring expansion reaction of a 2,2-dimethylchromene precursor. This work was adapted to a continuous flow setup for larger scale preparation, in view of biological investigations into the herbicidal properties of this natural product.

The radulanins are plant-derived natural products extracted from the leafy *Radula* genus of liverworts – a taxonomic subdivision of bryophytes.[1–7] They constitute a large group of bibenzyl compounds (*e.g.* **1**-**3**) involving fused oxacycles, which might be biosynthetically formed from the oxidative cyclization of *o*-prenylphenols like **4** (Figure 1a). Recently, based on structural similarities, we postulated that radulanin A (**1**) may exhibit similar biological properties to lunularic acid,[8] a bibenzyl phenolic bearing plant-growth inhibition and allelopathic properties.[9,10] This theory was later corroborated when an active dose of radulanin A (at 30-50 µM) was shown to cause wilting of *Arabidopsis thaliana* seedlings (a plant commonly considered as a weed in agriculture). As a number of current agrochemical products face scrutiny,[11–13] we believe radulanin A may constitute a promising phytopharmaceutical alternative.

It is clear from all reported syntheses that the greatest challenge associated with the synthesis of the radulanins is the formation of the 3-methyl-2,5-dihydro-1-benzoxepin ring (Figure 1b). In the first reported synthesis of **1**, Stefinovic and Snieckus[14] installed the heterocycle *via* a key ring closing olefin metathesis step on diene **5**. This approach was later adopted by Yoshida and co-workers[15] for the synthesis of radulanin H. Yamaguchi and co-workers[16] were able to form the dihydrobenzoxepine ring of radulanin A *via* an intramolecular Mitsunobu reaction of a 2-prenylphenol intermediate (**6**). In 2019, we reported the synthesis of bicyclic 3,5-dihydrooxepines *via* retro-Claisen rearrangements of 2-vinylcyclopropane-1-carbonyl intermediates (**7**), and applied this methodology to the synthesis of **1**.[8,17] This rearrangement competed with a Cloke-Wilson rearrangement, possibly furnishing benzofuran side-products related to the structure of perrottetin D (**2**).

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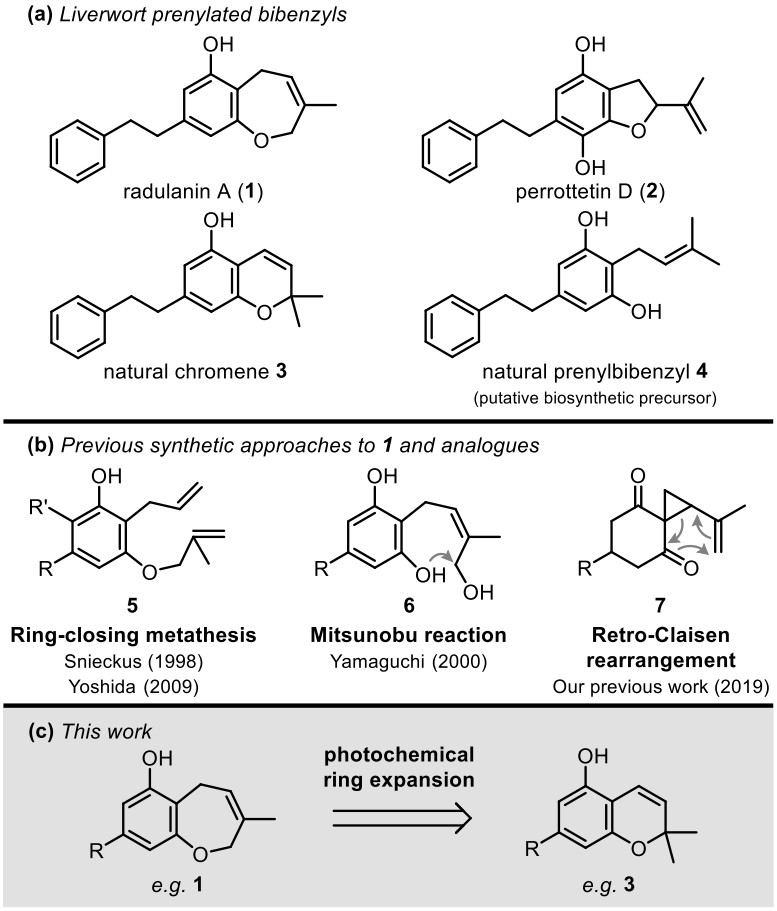
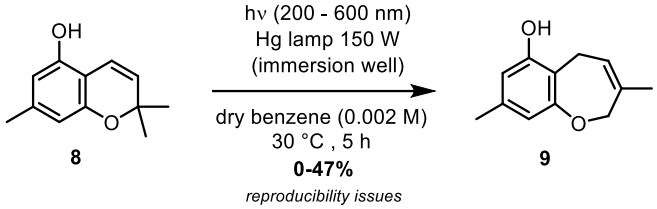


Figure 1. Structure of liverwort bibenzyl natural products (a) and comparision of past synthetic strategies for 2,5-hydrobenzoxepines (b) and this work (c).

Despite their merit, all these strategies suffer from long reaction sequences, and involve sensitive steps which are not easily adaptable to larger scale synthesis. In 1988, Chakrabarti and Chakraborty reported a photochemical ring-expansion of the chromene moiety of a carbazole alkaloid (girinimbine) and other derivatives to afford a 2,5-dihydrobenzoxepine in moderate to good yields.[18] This reaction was however never exploited for synthetic purpose. Inspired by this work, we envisaged a deployment of this methodology as a late-stage molecular edit to afford radulanin A (**1**) from readily accessible 2,2-dimethylchromenes (**3**) and in flow (Figure 1c), which has resulted in the shortest and highest yielding total synthesis of **1** to date. Since radulanin A and the analogous 2,2-dimethylchromene are both natural products present in the liverworts, we suspect that this rearrangement may be of some biosynthetic significance.[5,6] Furthermore, this work allowed a larger scale preparation of **1** for extended biological assays.

To investigate the feasibility of the photochemical ring expansion of 2,2-dimethylchromenes, preliminary studies were carried out on the model substrate **8**, which revealed a major UV/Vis absorption band at 278 nm, much similar to the analogous chromene **9**, our expected precursor to radulanin A. The chromene substrates were synthesised by the condensation of a 1,3-diphenol with prenal, catalyzed by ethylenediamine diacetate (EDDA) according to Lee *et al*.[19] To perform the ring expansion, several light sources, including sunlight, with various emission spectra were tested, with and without added photosensitiser, and in various solvents (supporting information, Table S1). In particular, we deemed a light source emitting in the UV wavelength range near the absorption band of the substrate. In their previous study, Chakrabarti and Chakraborty described the chromene rearrangement of carbazole-chromene

alkaloids at 254 nm (16 W) or at 365 nm (400 W) in benzene, with improved yields at 365 nm.[18] Gratifyingly in our case, the irradiation of **8** by a 150 W medium pressure Hg lamp (λ = 200–600 nm) in a pyrex immersion well apparatus afforded 2,5-benzoxepine **9** in 47% yield after 5 hours (Scheme 1). Attempts to better the yield (prolonged degassing, increased irradiation time) were not possible due to the irreproducibility of this method. Despite nearly quantitative conversions, most of the reaction mixture was seen to have decomposed.



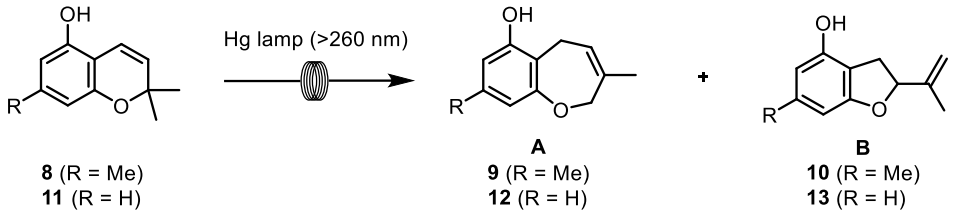
Scheme 1. Batch photochemical ring expansion of **8** in an immersion well, characterized by poor reproducibility.

In order to improve this reaction and develop a reliable preparative method for radulanin A, foreseeing extensive biological testing, a photochemical flow process was next investigated.[20–22] Utilising a flow chemistry system fitted with a 10 mL photochemical reactor equipped with a pyrex filter (Vapourtec E-series UV-150, Filter #3), we first sought to determine whether our preliminary batch conditions could be translated to a flow setup by retaining all possible reaction parameters: solvent, concentration, temperature and lamp power (the light source was also tested by coiling a PFA tubing around the cooling system of our previous 150 W medium pressure mercury lamp, but with no improvement). Promisingly, at a flow rate of 1 mL·min^{-1} in benzene at a concentration of 0.002 M, a small aliquot of collected reaction mixture at steady state output revealed only 10% remaining starting material by ^{1}H NMR, and afforded dihydrobenzoxepine **9** and its inseparable 5-membered isomer **10** in 24% yield in a respective 88:12 ratio (Table 1, entry 1). Prior to further optimization of this process, various wavelength filters were screened (supporting information, Table S2), which ultimately showed the pyrex filter (Vapourtec #3 cuts at 260 nm) to render superior selectivity for **9** at the highest conversion. Despite the apparent cleanliness of crude ^{1}H NMR spectrum (supporting information, Figure S1), the final yield suggested extensive decomposition must have occurred.

Further optimization of this process began by examining a more practical reaction concentration of 10 mM (entry 2). Although the reaction had not reached full conversion at this flow rate, several unknown signals appeared in the aromatic region of the crude NMR spectra alongside the product, likely formed from intermolecular reactions between reactive intermediates. Reducing the concentration to 1 mM proved favourable, rendering a combined yield of 35% at a flow rate of 2 mL·min^{-1} (entry 3). Reducing the lamp power by 50% (entry 4) led to improved selectivity but a lower yield of rearranged products. In an effort to improve safety, other solvents were screened. Toluene (entries 5-6) was shown to facilitate a higher conversion rate, but with inferior yields. Ethyl acetate was briefly trialed (entry 6), but resulted in poor selectivity. We last examined acetonitrile (entries 8-11), and observed a marked decrease in rates of conversion compared to benzene. However a slight improvement in yield (entry 10) was observed by working at 1 mM at a higher flow rate. It is noteworthy that when increasing residence time in the reactor (lower flow rates) in order to achieve full conversion, selectivity worsened and simultaneously impacted yield (entry 11). To test the influence of the methyl substituent on this difficult rearrangement, we applied the best conditions (entry 10) to chromene **11** (entry 12). We were pleased to observed that dihydrobenzoxepine **12** was obtained in a 55% yield at 100% conversion, with only 5% of isomer **13** (92:8 ratio). This result shows the crucial influence of substituents in this photochemical

rearrangement. To complete this study, we explored the scope of this rearrangement to more complex 2,2-dimethylchromenes with various substitutions, possibly applicable to the synthesis of other natural products (see supporting information, Figure S2).[23] These attemps resulted in degradation (or no reaction when an extended chromophores was present), showing a limited scope for this reaction. However, owing to the structural similarity between radulanin chromene **3** and compound **8**, we were confident we could apply this method to a preparation of radulanin A.

Table 1. Optimisation of the photochemical ring expansion in flow



8 (R = Me)
11 (R = H)

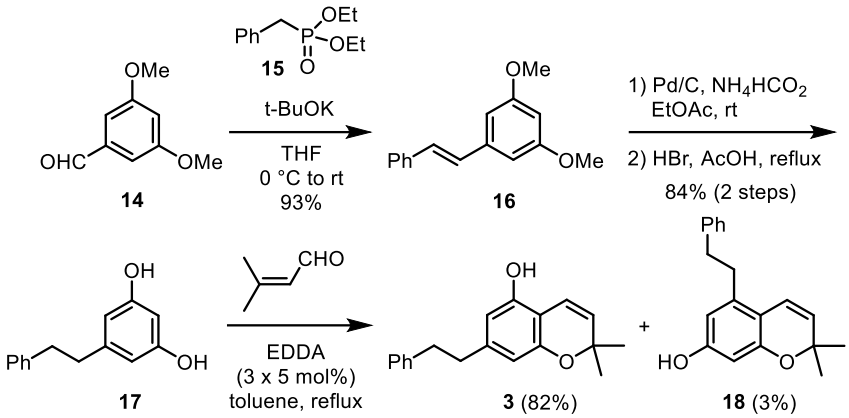
A
9 (R = Me)
12 (R = H)

B
10 (R = Me)
13 (R = H)

Entry	R	Solvent	Concentration (mM)	Lamp power (%)[a]	Flow rate (mL·min^{-1})	Conversion[b]	Yield A (%)[b]	Yield B (%)[b]	Ratio A:B
1	Me	PhH	2	50	1.0	90	21	3	88:12
2	Me	PhH	10	100	1.0	67	11	0	100:0
3	Me	PhH	1	100	2.0	98	28	7	80:20
4	Me	PhH	1	50	2.0	94	18	2	90:10
5	Me	PhMe	10	100	1.0	77	9	0	100:0
6	Me	PhMe	1	100	4.0	90	17	2	89:11
7	Me	EtOAc	1	100	1.0	100	21	12	64:36
8	Me	MeCN	10	100	0.2	92	22	7	76:24
9	Me	MeCN	5	100	0.5	96	18	4	82:18
10	Me	MeCN	1	100	1.4	95	36	11	77:23
11	Me	MeCN	1	100	1.2	100	21	11	66:34
12	H	MeCN	1	100	1.4	100	55	5	92:8

[a] Power refers to electrical power output of lamp, 100% being 150 W. [b]Conversion and yields determined by ^{1}H NMR with respect to 1,3,5-trimethoxybenzene as internal standard.

Chromene intermediate **3**, a natural product first isolated from another *Radula* moss,[2,6] was synthesised in four steps from 3,5-dimethoxybenzaldehyde **14**. The Horner-Wadsworth-Emmons reaction of **14** with diethyl benzylphosphonate **15** first afforded pinosylvin dimethyl ether **16** in 93% yield. Subsequent hydrogenation of the olefin moiety and deprotection of the phenols in presence of HBr gave dihydropinosylvin **17** in 94% yield. This nucleophilic diphenol was engaged in the formation of chromene **3** in the presence of prenal and the Brønsted acid catalyst EDDA (Scheme 2).[19,24] On a practical point of view, adding EDDA in three portions of 5 mol% over three hours furnished the best yields of **3** (82%), which was accompanied by a small amount of regioisomer **18** (3%). Compound **18** is also a natural product found in *Radula* species.[5]

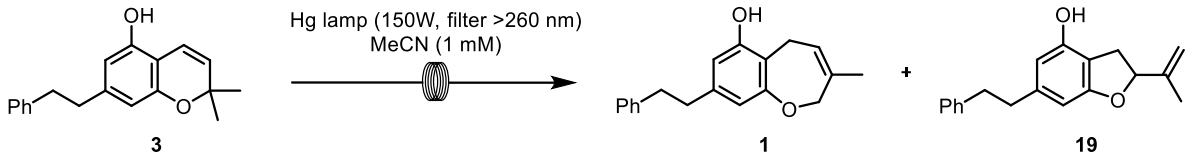


Scheme 2. Optimised total synthesis of natural chromene **3**

Chromene **3** was submitted to the same photochemical flow conditions as **8** (Table 1, entries 9 and 10). Gratifyingly, it was converted into radulanin A (**1**) in a higher NMR yield of 41% (Table 2, entry 1), accompanied by 15% of five-membered isomer **19** (73:27 ratio). Despite the good conversion of 92% and after exhaustive efforts, compound **1** was found to be inseparable from the chromene substrate **3**. As a result, we were compelled to reduce the flow rate to achieve full consumption of the chromene, which inevitably impacted selectivity and yield (entry 2). These new conditions allowed to isolate **1** in 26% yield (with 17% yield of benzofuran **19**, also a natural product). With a productivity of 127 mg/24 h, we were able to afford more than 300 mg of **1** for biological investigations.

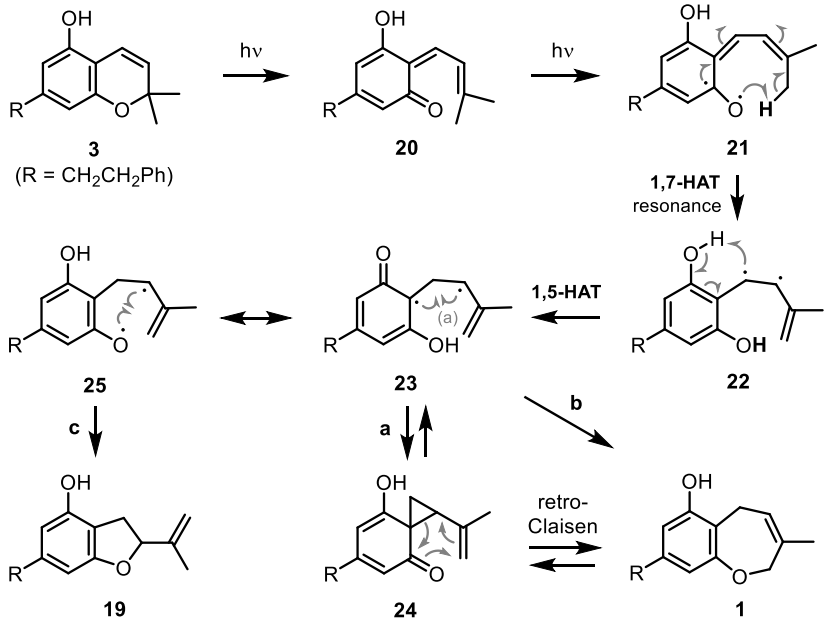
Despite the simplicity of this transformation, no obvious rearrangement mechanism can be drawn at first sight. A key intermediate could be quinone methide isomer **20** (Scheme 3). Under UV excitation, we supposed that it could generate a reactive oxy radical species (**21**), substrate of a 1,7-hydrogen atom transfer (1,7-HAT) leading to intermediate **22**. From this symmetrical diphenol, the benzylic radical would be reduced through 1,5-HAT leading to **23**, which could evolve through several routes. First, the new 1,3-diradical could recombine into 2-vinylcyclopropanecarbonyl intermediate **24** (route a), a good substrate for spontaneous but reversible retro-Claisen rearrangement according to our previous study,[8] leading to kinetically favoured 2,5-dihydrobenzoxepine **1**. Second, resonance and radical recombination could also directly lead to **1** (route b). Finally, resonance form **25** could be an intermediate toward benzofuran product **19** (route c), which tends to be the thermodynamic end-product when increasing irradiation times.

Table 2. Photochemical rearrangement of chromene 3 into radulanin A (**1**)



Entry	Flow rate (mL min^{-1})	Conversion (%)[a]	1 (%)	19 (%)	Ratio 1:19
1	1.4	92	41[a]	15[a]	73:27
2	1.2	100	26[b]	17[b]	60:40

[a] Conversions and yields determined by ^{1}H NMR with respect to trimethoxybenzene as internal standard. [b] Isolated yield.



Scheme 3. Proposed mechanism for 2,2-dimethylchromene reorganizations.

In conclusion, the photochemical ring extension of 2,2-dimethylchromene substrates has been studied, showing the advantage of using a flow process to improve the reproducibility and scale up the reaction. It was applied to the total synthesis of radulanin A, obtained in 19% yield over five steps from aldehyde **14**. As far as we know it is the shortest preparation reported to date. Although it would need additional improvements to scale up the process, the method allowed us to prepare substantial amounts (320 mg) of radulanin A for extended biological studies as a weedkiller. Finally, to explain this rearrangement, we supposed that a quinone methide intermediate would lead to highly reactive radical species, involving a key 1,7-hydrogen atom transfer and a possible retro-Claisen rearrangement. Considering that both the chromene substrate (**3**) and radulanin A (**1**) are natural products present in the same liverwort genus,[2,6] it is questionable whether this rearrangement could occur in Nature.

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