

# Integrated Batch and Continuous Flow Process for the Synthesis of Goniotalamin

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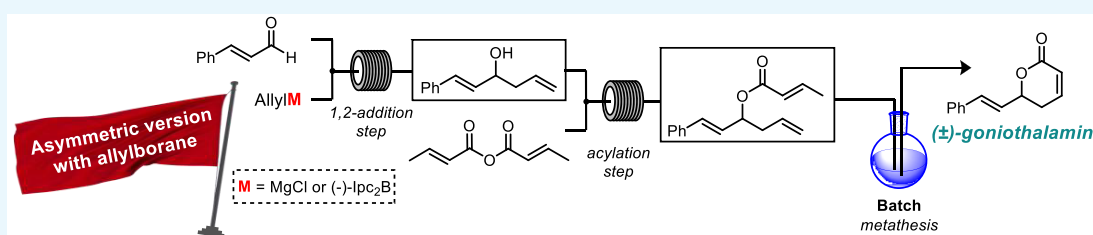
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**ABSTRACT:** An integrated batch and continuous flow process has been developed for the gram-scale synthesis of goniotalamin. The synthetic route hinges upon a telescoped continuous flow Grignard addition followed by an acylation reaction capable of delivering a racemic goniotalamin precursor (**16**) (20.9 g prepared over 3 h), with a productivity of 7 g·h<sup>-1</sup>. An asymmetric Brown allylation protocol was also evaluated under continuous flow conditions. This approach employing (–)-Ipc<sub>2</sub>B(allyl) provided an (S)-goniotalamin intermediate in 98% yield and 91.5% enantiomeric excess (ee) with a productivity of 1.8 g·h<sup>-1</sup>. For the final step, a ring-closing metathesis reaction was explored under several conditions in both batch and flow regimes. In a batch operation, the Grubbs second-generation was shown to be effective and highly selective for the desired ring closure product over those arising from other modes of reactivity, and the reaction was complete in 1.5 h. In a flow operation, reactivity and selectivity were attenuated relative to the batch mode; however, after further optimization, the residence time could be reduced to 16 min with good selectivity and good yield of the target product. A tube-in-tube reactor was investigated for in-situ ethylene removal to favor ring-closing over cross-metathesis, in this context. These results provide further evidence of the utility of flow chemistry for organometallic processing and reaction telescoping. Using the developed integrated batch and flow methods, a total of 7.75 g of goniotalamin (**1**) was synthesized.

## 1. INTRODUCTION

Chemotherapy plays a pivotal role in cancer treatment, improving the quality of life and increasing patient survival. However, the drug resistance of antineoplastic drugs and their high toxicity are some of the limitations of the current chemotherapy treatments. Due to these reasons, drug discovery programs have made efforts to deliver new candidates with high selectivity and low off-target cytotoxicity for cancer treatment. In most cases, such new candidates are inspired by natural compounds due to their limitless chemical diversity and a broad biological profile.<sup>1</sup> To date, more than 50% of all approved small-molecule drugs have been structurally inspired by isolated natural compounds.<sup>2</sup> Moreover, ca. 80% of all antitumor agents used in the clinic are derived from natural products.<sup>3</sup>

Accordingly, the natural product goniotalamin (**1**) has been the object of study of various medicinal chemistry programs for several years.<sup>4</sup> Goniotalamin is a secondary metabolite obtained from the species of the genus *Goniotalamus*.<sup>5</sup> This styryl lactone exhibits important antiproliferative and cytotoxic effects on several tumor cell

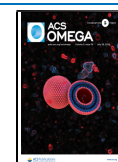
types, such as lung, breast, kidney, prostate, liver, and leukemia.<sup>6</sup> Due to its wide biological profile, many medicinal studies have explored goniotalamin as a chemical platform to the design of new drug candidates and to understand their in vitro mode of action (Scheme 1A).<sup>7</sup>

Using a solid tumor experimental model in vivo assay, our group has demonstrated that both non-natural (R)-**1** and natural (S)-**1**, as well as its racemic-**1** form, present antiedematogenic and inhibitory activity against the proliferation of Ehrlich solid tumor cells.<sup>8</sup> No evidence for toxic effects was found, and similar activity profiles were observed for racemic-**1**, (R)-**1**, and (S)-**1** against NCI-ADR/RES, NCI-460, 786-0, and U251 cancer cells.<sup>8</sup>

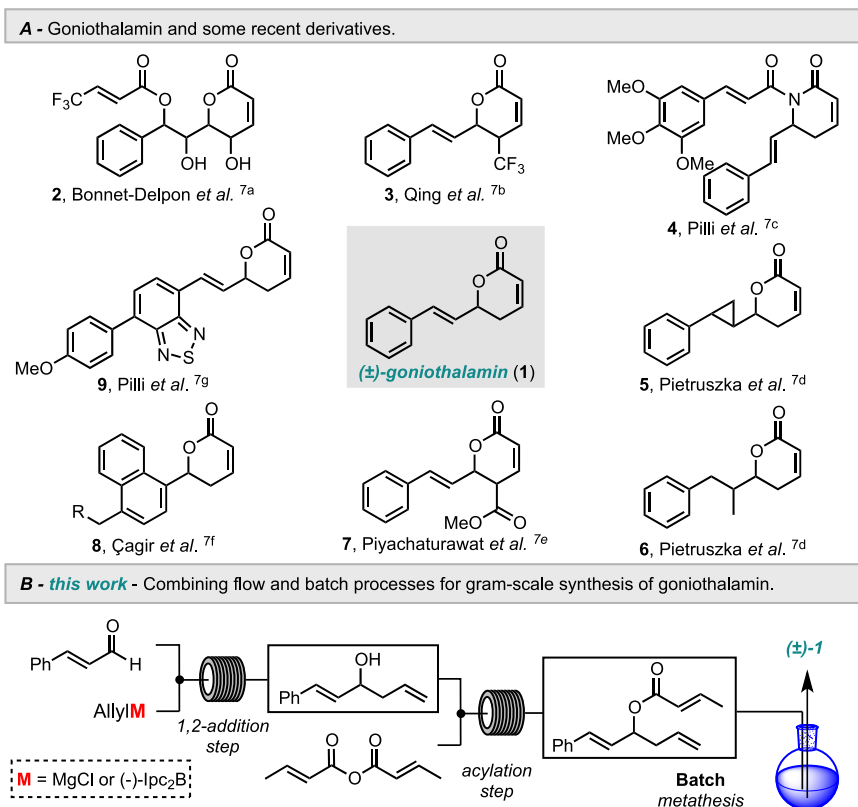
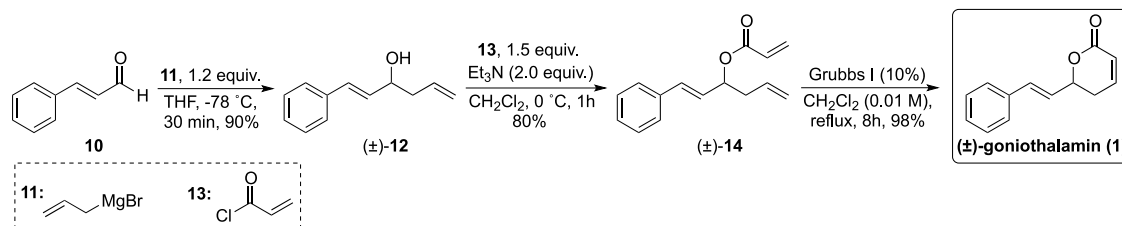
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## Scheme 1. Goniotalamin and Derivatives (A); the Proposed Study (B)

Scheme 2. Previous Synthesis of Racemic Goniotalamin (1) in the Batch Mode<sup>6aa</sup>

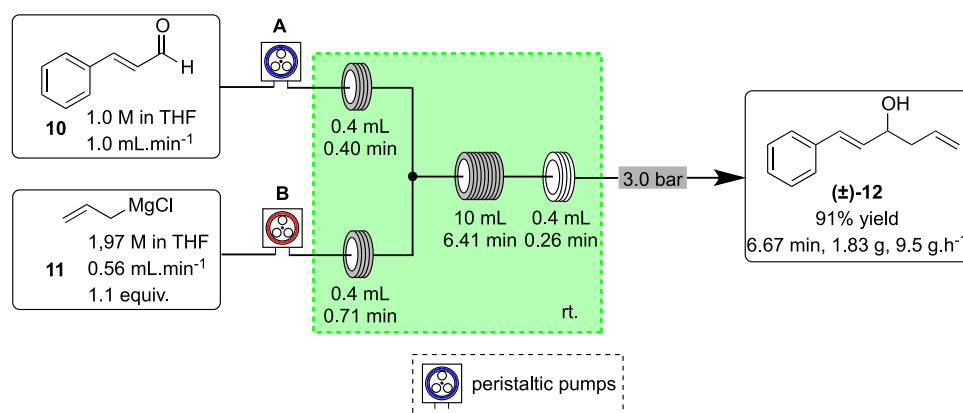
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Concerning the synthesis of (±)-goniotalamin, we have noted the lack of studies focusing on new strategies for the gram-scale synthesis of **1**. In many studies, **1** was used as a starting material for the synthesis of goniotalamin derivatives, or the same reaction sequence was employed for the preparation of new analogues,<sup>6b,7</sup> which shows the importance of a robust and scalable method to accumulate **1**. In addition, gram quantities of **1** are required for more complete and in-depth *in vivo* studies regarding the real potential of **1** as a drug candidate for cancer treatment. Bearing this in mind, continuous flow processing offers a great opportunity for the scaling-up of the synthesis of goniotalamin (**1**).

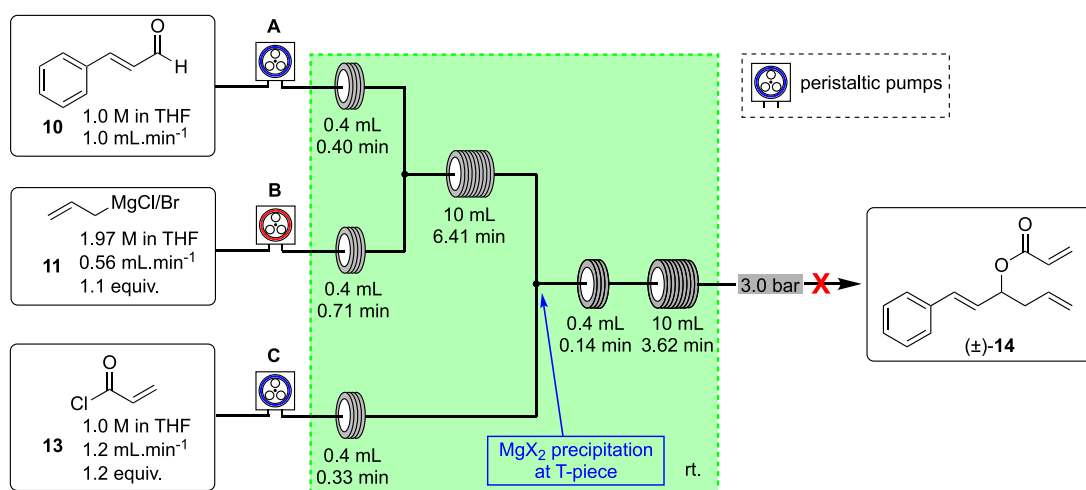
Flow chemistry has emerged in both academia and industry as a promising enabling technology for organic synthesis, providing several advantages in comparison to traditional batch processes for organic and inorganic synthesis.<sup>9</sup> One of the most attractive benefits in a continuous flow approach is the opportunity to integrate several reaction steps (the so-called telescoped synthesis) to rapidly generate focused libraries of bioactive compounds as well as enough material for clinical trials in medicinal chemistry programs.<sup>10</sup> Recently, our group

has demonstrated the telescoped synthesis of highly substituted 3-thio-1,2,4-triazoles, including their gram-scale preparation and application to the continuous flow synthesis of an API.<sup>11</sup> Integration of processing steps through telescoped reactions allows for the expedient preparation of target compounds since there is no need for intermediate isolation, which also leads to reduced waste generation through the reduction in solvent use.<sup>11</sup> Indeed, the synthesis of many natural products and biologically active compounds has been the target of multistep flow chemical processes.<sup>12</sup>

In this context, we demonstrate herein an improved process for the multigram synthesis of (±)-goniotalamin (**1**) in a continuous flow regime (Scheme 1B). Since the racemic mixture of **1** exhibited a similar *in vivo* antitumor profile compared to both enantiomers and (*S*)-goniotalamin (*S*)-**1** showed higher potency *in vitro* than (*R*)-**1** against kidney cancer cell proliferation (786-0) with IC<sub>50</sub> = 4 nM,<sup>6b</sup> both racemic and asymmetric routes (for the *S* enantiomer) were explored under continuous flow conditions.

Scheme 3. Continuous Flow Addition of Allylmagnesium Chloride (11, Pump B) to *trans*-Cinnamaldehyde (10, Pump A)

Scheme 4. Attempt toward Continuous Flow Addition of Allylmagnesium Chloride/Bromide (11, Pump B) to 10 (Pump A) Followed by Acylation of the Alkoxide Intermediate with Acryloyl Chloride (13, Pump C)



## 2. RESULTS AND DISCUSSION

A three-step synthetic route has been used for the total synthesis of goniotalamin in the batch mode in our laboratory (Scheme 2).<sup>6a</sup> This procedure involves the addition of an ethereal solution of allylmagnesium bromide (11) to *trans*-cinnamaldehyde (10), followed by esterification of the secondary alcohol 12 with acryloyl chloride (13) in the presence of triethylamine to prepare the corresponding acrylate ester 14. This is followed by a ring-closing metathesis reaction using the Grubbs first-generation catalyst.

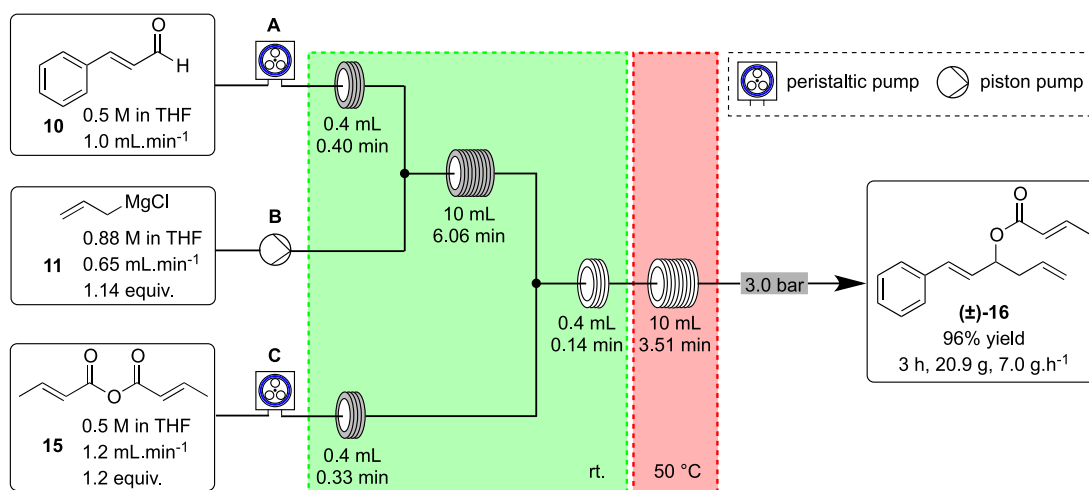
To explore the *in vivo* activity of this family of dihydropyranones, this route was scaled up to provide gram quantities of goniotalamin. Although such methodology was able to secure ca. 25 g of the requisite (racemic) compound, several small-scale batch reactions were required for the ring-closing metathesis step, which did not provide acceptable yields when conducted at a large scale or at an increased concentration.<sup>8</sup> On the practical side, manual work including several chromatographic separations and the use of large quantities of solvents were required to accumulate the requisite amount of goniotalamin for *in vivo* studies.<sup>8</sup> Based on this, such a three-step batch procedure could be potentially adapted to flow conditions to afford a more easily scaled approach. Note that the desired process could not only potentially allow the preparation of goniotalamin on a multigram scale but also

permit rapid access to analogues by changing the aldehyde and Grignard reagent input starting materials.

**2.1. Racemic Allylation Followed by Acylation Reaction.** To initiate experiments under continuous flow conditions, the addition of allylmagnesium chloride (11) to *trans*-cinnamaldehyde (10) was investigated. This step was optimized by careful adjustment of the residence time and stoichiometry toward full conversion to alcohol 12 before the in-line introduction of the acylating agent in the following step. The optimized flow conditions to alcohol (±)-12 are presented in Scheme 3.

A commercially available THF solution of 11 (1.97 M) and a THF solution of 10 were pumped through the flow reactor via two chemically resistant peristaltic pumps containing fluoropolymer tubes compatible with THF solutions, according to our previous work concerning the processing of organometallic reagents in a continuous flow regime.<sup>13</sup> The organometallic reagent solution was pumped from the commercial bottle by inserting argon line through the septum seal, with no requirement for predilution or use of sample loops. Thus, Grignard reagent addition to 10 proceeded quickly at room temperature. In batch, these reactions are typically performed at low temperatures, for example, at 0 °C or below, by dropwise addition, before warming up to room temperature allowing the reaction to go to completion.<sup>6a</sup> In the continuous flow regime, cryogenic conditions were not

**Scheme 5. Continuous Flow Addition of Allylmagnesium Chloride (11, Pump B) to 10 (Pump A) Followed by Acylation of the Alkoxide Intermediate with Crotonic Anhydride (15, Pump C)**



necessary and no uncontrollable exothermic reaction was noticed, leading the reaction to achieve completion at reduced residence times and increased material productivity. Notably, while an uncontrollable exothermic process was not observed, a modest temperature increase of 3–4 °C in the reactor was noted when the cooling device of the flow system was not operating, but neither yield nor selectivity were affected when the results from processing in this way were compared with those obtained from the active temperature control (through cooling). Under the conditions shown in Scheme 3, full conversion could be obtained using only 1.1 equiv of the Grignard reagent 11. After reaching steady-state operation, during a period of ca. 11.5 min, a sample was collected and, after aqueous workup and chromatography purification, alcohol 12 was obtained in 91% yield (1.83 g) and 9.5 g h<sup>-1</sup> productivity, with a residence time of 6.67 min at the indicated flow rates.

After optimizing the racemic allylation, the acylation reaction was adapted from batch to the continuous flow mode. The reaction of the secondary allylic alcohol 12 with acryloyl chloride (13) is usually conducted in the batch mode with triethylamine as base. However, the alkoxide product of the Grignard addition eliminates the need for an additional base in the acylation reaction. The output stream of the first reactor coil was combined with a THF solution of 13, and the combined stream was directed to an additional 10 mL reactor coil kept at room temperature (Scheme 4).

Unfortunately, running the reaction under the conditions depicted in Scheme 4 led to blockage of the system at the T-piece where the alkoxide output and acylating reagent mix; we hypothesized that this would be related to the precipitation of magnesium(II) chloride in the reactor coil. The same behavior was observed when allylmagnesium bromide was used instead of the corresponding chloride. Reducing the reaction concentration was explored but still led to the formation of solids at the T-piece on mixing, resulting in blockage and shutdown of the flow system.

Based on our previous work on the use of organometallic reagents in the flow regime,<sup>13</sup> 13 was replaced by commercially available crotonic anhydride (15) as the acylating agent, leading instead to a more soluble mixed magnesium(II) chloride acetate salt. Indeed, a THF solution of 15 was pumped at 1.2 mL min<sup>-1</sup> by the third peristaltic pump and

combined with the output stream from the first reactor coil, which contained the alkoxide intermediate. The combined stream was then directed to a second 10 mL reactor coil stabilized at 50 °C and pressurized with an appropriate back-pressure regulator. Ester 16 was obtained in full conversion under these conditions.

Although the reaction conditions were optimized for the telescoped preparation of ester 16, compatibility issues with processing this Grignard reagent solution using peristaltic pumps and supplied tubing led to irreproducible results (see Figure S1 for more details), likely due to the presence of remaining allyl chloride in the allylmagnesium chloride solution.

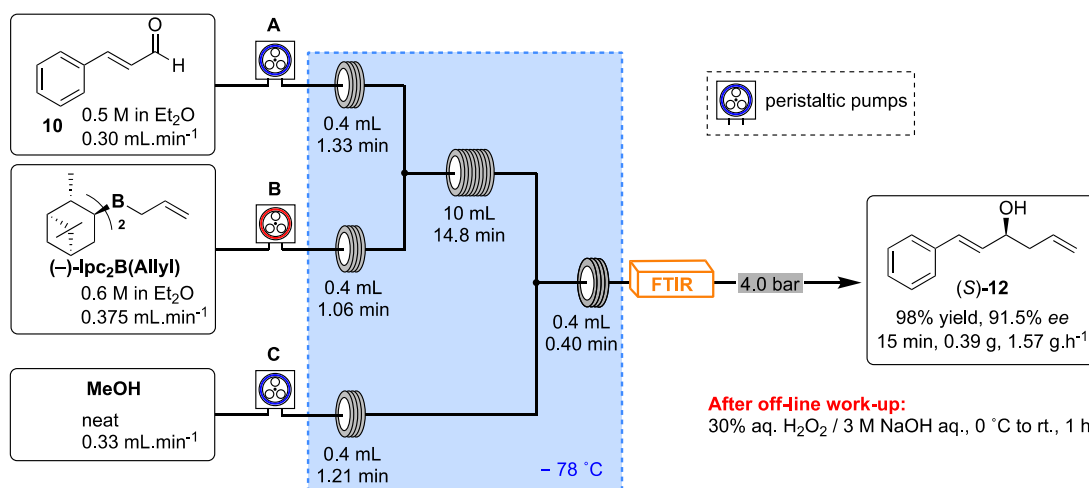
To overcome this issue, an external piston pump (K-120 Knauer pump) was used to deliver the allylmagnesium chloride solution, as shown in Figure S2. In this experiment, the back of the HPLC-type piston pump was purged and connected to an argon line. This measure avoids the exposure of the Grignard solution to the atmosphere and formation of solids that would result in blocking of the pumping system (see the Experimental Section for more details).

Once the flow equipment and reaction parameters, such as stoichiometry of reagents, residence time, and temperature, were optimized, the multistep process for the preparation of ester 16 was uninterruptedly run for 3 h to yield 20.9 g of the product, which was obtained in full conversion and 96% isolated yield, for 2 steps (Scheme 5).

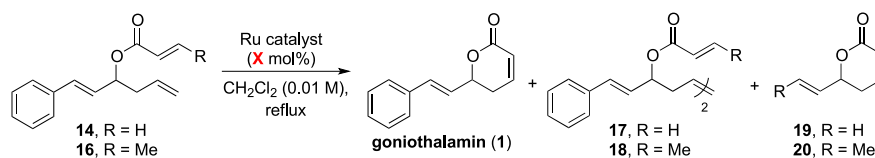
With the racemic synthesis of the goniotalamin intermediate (±)-16 in hand, we investigated the asymmetric allylation to produce the (*R*)-goniotalamin precursor.

**2.2. Asymmetric Allylation Reaction.** Several batch strategies have been proposed for the asymmetric synthesis of goniotalamin (1).<sup>6a,14</sup> For example, Singh and co-workers employed an auxiliary-controlled asymmetric aldol reaction for the construction of an enantiopure homoallylic alcohol intermediate.<sup>14a</sup> Asymmetric allylboration was described by Brown and co-workers, achieving homoallylic alcohol intermediate (12) in 92–97% enantiomeric excess (ee).<sup>14b</sup> Our group has previously employed Ti(O*i*-Pr)<sub>4</sub>/*R*-BINOL-mediated asymmetric allylation, leading to alcohol 12 in 78% yield and 94% ee.<sup>6a,14c</sup> Enzymatic kinetic resolution of alcohol 12 has also been demonstrated.<sup>14d</sup>

**Scheme 6. Continuous Flow Asymmetric Allylation of *trans*-Cinnamaldehyde (**10**, Pump A) with (–)-Ipc<sub>2</sub>B(allyl) (Pump B), Followed by In-Line Quenching with MeOH (Pump C)**



**Table 1. Evaluation of the Ring-Closing Metathesis in the Batch Mode**



| entry | R  | catalyst   | loading (mol %) | t (h) | conversion (%) <sup>a</sup> | 1/17/19 or 1/18/20 <sup>a</sup> | yield (1 + 19 or 1 + 20) (%) <sup>b</sup> | yield 17 or 18 (%) |
|-------|----|------------|-----------------|-------|-----------------------------|---------------------------------|---|--------------------|
| 1     | H  | Grubbs I   | 10              | 7.0   | 100                         | 100:0:0                         | 87  | <5                 |
| 2     | H  | Grubbs II  | 5               | 1.5   | 100                         | 97:0:3                          | 90  | <5                 |
| 3     | H  | Hoveyda II | 5               | 1.5   | 100                         | 94:0:6                          | 95  | <5                 |
| 4     | Me | Grubbs I   | 10              | 14.0  | 96                          | 20:80:0                         | 19  | 75                 |
| 5     | Me | Grubbs II  | 5               | 1.5   | 100                         | 90:0:10                         | 96  | <5                 |
| 6     | Me | Hoveyda II | 5               | 3.0   | 100                         | 92:8:0                          | 92  | 8 <sup>a</sup>     |

<sup>a</sup>Calculated from <sup>1</sup>H NMR of the crude. <sup>b</sup>Isolated yield for the inseparable mixture.

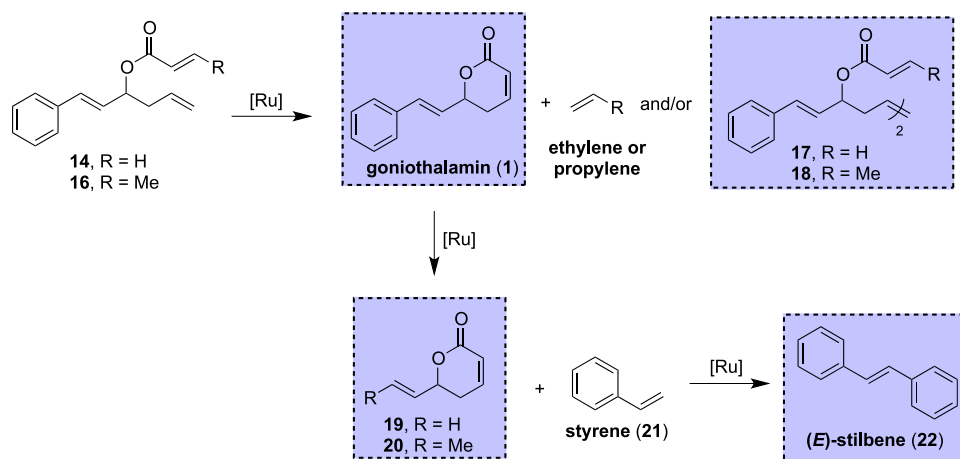
Allylboranes and boronates derived from  $\alpha$ -pinene or diisopropyl tartrates as chiral auxiliaries have been utilized for the enantio- and diastereoselective formation of homoallylic alcohols.<sup>15</sup> Amongst the many reported methods for asymmetric allylation, we chose to investigate these organoborane compounds in this context, in view of their straightforward preparation employing readily available starting materials and because of our continuing interest in exploring the utility of flow chemistry for organometallic synthesis. Moreover, high enantiomeric excess is usually obtained when performing asymmetric allylation using this approach.<sup>15</sup> Based on this, both Brown (allylborane)<sup>15c</sup> and Roush (allylboronates)<sup>15e</sup> reagents were prepared and initially evaluated in batch for the asymmetric allylation of *trans*-cinnamaldehyde (**10**) (Tables S1 and S2 in the SI). The Roush allylation procedure with (+)- and (–)-diisopropyl-tartrate allyl boronate (((+)- and (–)-DIPT)B(Allyl)) afforded homoallylic alcohols (*S*)-**12** and (*R*)-**12** in 69–98% yield and 70.8–72.4% ee (Table S1). On the other hand, the Brown procedure with (–)-*B*-allyldiisopinocampheylborane ((–)-Ipc<sub>2</sub>B(Allyl)) afforded alcohol (*S*)-**12** in 96% yield and 90.8% ee. In light of these results, the Brown approach was taken forward for optimization under continuous flow conditions (Table S3 in the SI). The optimized result is presented in Scheme 6.

During optimization, an FTIR spectrometer was placed after the coil reactor in the continuous flow system for the real-time

monitoring of the conversion of **10** into (*S*)-**12**. Such an approach facilitates the reaction optimization, and (*S*)-**12** was obtained in 98% yield and 91.5 ee with a productivity of 1.57 g·h<sup>–1</sup>. Gratifyingly, the chemical yield and the enantiomeric excess obtained under continuous flow conditions at reduced reaction times are slightly higher than those obtained in batch. Probably, more efficient mixing of a 1:1 stoichiometric mixture and improved temperature control in view of the high surface-to-volume ratio of the flow reactor could account for this superior outcome, as opposed to dropwise addition of one to the other in batch. Note that compound (*S*)-**12** could be converted to the ester intermediate **14** or **16** and then to (*S*)-**1** in batch using the conditions presented in Scheme 2. The need for off-line work using hydrogen peroxide under basic conditions would require an in-depth study to telescope into the acylation under these conditions, probably making use of the continuous membrane separation technology before integration of the steps.

Finally, in the last step of the synthesis, the ring-closing metathesis reaction to afford the  $\alpha,\beta$ -unsaturated  $\delta$ -lactone present in the structure of goniotalamin (**1**) was evaluated.

**2.3. Ring-Closing Metathesis Reaction.** Numerous immobilized Grubbs–Hoveyda catalysts for the ring-closing metathesis reaction are reported in the literature,<sup>16</sup> including their investigation under continuous flow conditions.<sup>17</sup> For example, Zednic and co-workers demonstrated the metathesis

Scheme 7. Possible Reaction Pathways and Products for the Olefin Metathesis Reaction<sup>a</sup>

<sup>a</sup>Compounds enclosed in the blue dotted box were isolated and characterized; see the SI.

of cardanol using Hoveyda–Grubbs type catalysts supported on mesoporous silica in the continuous flow regime.<sup>17a</sup> In this study, the cumulative TON in flow was significantly improved relative to the maximum TONs in a batch reactor (2500 versus 1600). The product contamination by metal leaching was also verified, and only 1.0–1.5% of ruthenium was detected in the final product. However, cardanol conversion fell over time due to catalyst deactivation. Chmielewski and co-workers showed the immobilization of ruthenium alkylidene complexes by noncovalent interactions within metal–organic frameworks (MOFs).<sup>17b</sup> Ruthenium leaching was not observed, but much lower TONs were obtained in flow compared to batch procedures (4700 versus 8900). A monolithic metathesis catalyst was prepared by Kirschning and co-workers for further use in a flow microreactor; however, it was affected by partial leaching under continuous flow conditions.<sup>17c</sup> Other examples include immobilization of Grubbs/Hoveyda catalysts on commercial silica,<sup>17d</sup> microcapillaries coated with a film of dimethylpolysiloxane soaked with the Grubbs second-generation catalyst,<sup>17e</sup> and the polystyrene-supported Grubbs second-generation catalyst.<sup>17f</sup>

Even though the use of columns with packed immobilized catalysts could be a convenient approach to minimize postreaction manipulations, concerns such as leaching, recycling, and anchoring of immobilized ruthenium catalysts led us to focus our study on homogeneous conditions using Grubbs first- and second-generation as well as Hoveyda–Grubbs second-generation catalysts. Initially, we performed the reactions in batch to evaluate the concentration and catalyst selectivity (Table 1).

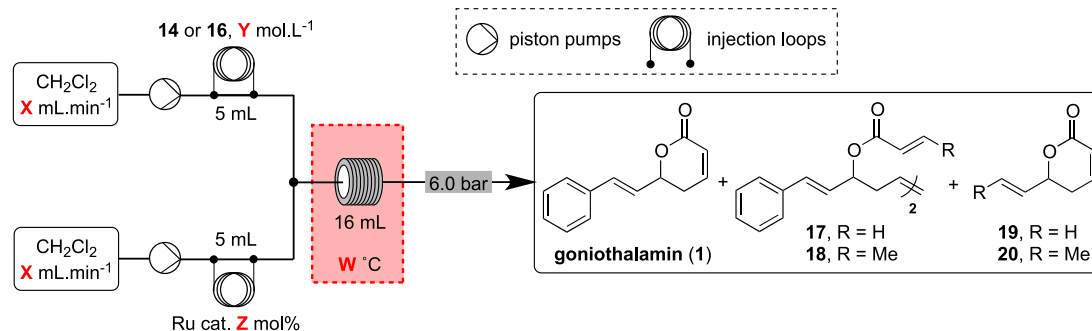
The ring-closing metathesis reaction for acrylate ester 14 is typically performed using the Grubbs first-generation catalyst, in refluxing dichloromethane and at low concentrations to circumvent the formation of dimer 17 in view of the intermolecular metathesis reaction.<sup>6a</sup> Indeed, under these conditions (Table 1, entry 1), goniiothalamin (1) was obtained in good yield and selectivity from ester 14. On the other hand, the use of either Grubbs or Hoveyda–Grubbs second-generation catalysts led to the formation of goniiothalamin (1) along with the vinyl lactone 19 (Table 1, entries 2 and 3), resulting from a cross-metathesis between the styryl unit of goniiothalamin (1) and ethylene gas, which is generated during the ring-closing metathesis reaction (Scheme 7).

Reactions using crotonate ester 16, with the methyl group at the acrylic unit, proved to be more complex. First, as one could expect, the use of the Grubbs first-generation catalyst gave dimer 18 as the major product with goniiothalamin (1) being isolated in only 19% yield (Table 1, entry 4). Note that both goniiothalamin (1) and 18 were also formed using the Hoveyda–Grubbs second-generation catalyst; nevertheless, the selectivity was reversed toward the formation of the desired product goniiothalamin (1) (entry 6). Finally, the ring-closing metathesis of ester 16 carried out in the presence of the Grubbs second-generation catalyst led to the formation of goniiothalamin (1) and lactone 20 in excellent combined yield (96%), obtained as a 90:10 inseparable mixture (entry 5). All additional experiments such as sparging with nitrogen gas to force the removal of ethylene from the reaction mixture, slow addition of the substrate and catalyst solutions (“infinite dilution approach”) to reduce the amount of dimer 17 or 18 formation, reflux in toluene as solvent, higher reaction concentration (up to 0.1 M), and using 1-heptene as a sacrifice olefin (to avoid lactone 19/20 formation by reaction with ethylene/propylene) proved to be unsuccessful (for more details, see Table S4).

The very low concentrations usually employed for the ring-closing metathesis reaction present an acute limitation of material productivity when operating at large/pilot scales.<sup>18</sup> In batch, when the reaction with ester 16 was evaluated at higher reaction concentrations (0.05 and 0.1 M), a more complex mixture of dimers was observed.<sup>19</sup> Bearing in mind the aforementioned advantages of flow chemistry over batch procedures, we sought to investigate the ring-closing metathesis reaction in a continuous flow regime. Considering that the monomeric  $\alpha,\beta$ -unsaturated  $\delta$ -lactone should be more thermodynamically favorable than the dimer, pushing the reaction in flow conditions to its limits could potentially convert the observed dimer into goniiothalamin (1), which could enable the use of a higher concentration for this transformation. The use of a back-pressure regulator to pressurize the system allows the heating of a solvent above its boiling point, and this fact was advantageously employed in this synthesis.<sup>9</sup>

The flow setup used for the preliminary optimization experiments is shown in Figure S3. Initially, the Grubbs first-generation catalyst was evaluated in the reaction with esters 14

Table 2. Evaluation of the Ring-Closing Metathesis toward Goniothalamin (1) under Flow Conditions



| entry | R  | conc. (M) | catalyst   | loading (mol %) | T (°C) | t (min) | conv. (%) <sup>a</sup> | 1/17/19 or 1/18/20 <sup>a</sup> | yield (1 + 19 or 1 + 20) (%) <sup>b</sup> |
|-------|----|-----------|------------|-----------------|--------|---------|------------------------|---------------------------------|---|
| 1     | H  | 0.05      | Grubbs I   | 10              | 100    | 16.0    | 23                     | 100:0:0                         | ND <sup>c</sup>                           |
| 2     | H  | 0.01      | Grubbs II  | 5               | 100    | 16.0    | 90                     | 79:0:21                         | 90  |
| 3     | H  | 0.05      | Grubbs II  | 5               | 50     | 16.0    | 100                    | 38:44:18                        | 55  |
| 4     | H  | 0.05      | Grubbs II  | 5               | 100    | 16.0    | 100                    | 57:0:43                         | 78  |
| 5     | H  | 0.05      | Grubbs II  | 1               | 100    | 16.0    | 55                     | 83:0:17                         | ND <sup>c</sup>                           |
| 6     | H  | 0.05      | Grubbs II  | 5               | 100    | 8.0     | 97                     | 54:0:46                         | 79  |
| 7     | H  | 0.01      | Hoveyda II | 5               | 100    | 16.0    | 100                    | 44:0:56                         | 97  |
| 8     | H  | 0.05      | Hoveyda II | 5               | 100    | 16.0    | 100                    | 41:0:59                         | 58  |
| 9     | Me | 0.05      | Grubbs I   | 10              | 100    | 16.0    | 0                      |                                 |   |
| 10    | Me | 0.01      | Grubbs II  | 5               | 100    | 16.0    | 80                     | 100:0:0                         | 75 (1 only)                               |
| 11    | Me | 0.01      | Grubbs II  | 5               | 100    | 32.0    | 75                     | 100:0:0                         | 65 (1 only)                               |
| 12    | Me | 0.05      | Grubbs II  | 5               | 100    | 16.0    | 100                    | 66:0:34                         | 94  |
| 13    | Me | 0.01      | Hoveyda II | 5               | 100    | 16.0    | 100                    | 83:0:17                         | 91  |
| 14    | Me | 0.05      | Hoveyda II | 5               | 100    | 16.0    | 100                    | 63:0:37                         | 77  |

<sup>a</sup>Calculated from <sup>1</sup>H NMR of the crude. <sup>b</sup>Isolated yield for the inseparable mixture. <sup>c</sup>ND = not determined.

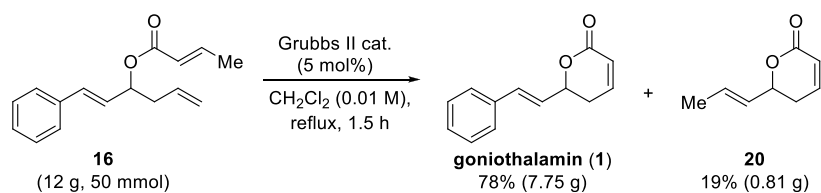
and 16 at 100 °C (Table 2, entries 1 and 9). Low conversion was obtained for ester 14 in 16 min of residence time. When the more active and thermally stable Grubbs second-generation catalyst was employed, full conversion was obtained even at 50 °C, but a mixture of goniothalamin (1), dimer 17, and lactone 19 was formed (entry 3). As we anticipated, increasing the temperature led to the consumption of the dimer and a mixture of 1 and lactone 19 was obtained in almost 1:1 ratio (entry 4), being isolated in 78% combined yield. The use of toluene as solvent at 150 °C (results not shown) gave essentially the same results.

The formation of lactone by-product 19 in a greater extent in the flow regime is not surprising, considering that the reaction under flow conditions is performed in a closed system, thus keeping the ethylene gas in solution, which favors the olefin cross-metathesis reaction with the desired goniothalamin (1) product (entries 2–5). Hypothesizing that such reaction occurs after the ring-closing reaction, the residence time was halved (entry 6). In this instance, for shorter residence time, we observed a slightly lower conversion and the selectivity remained poor (1:1 ratio of 1 and 19). The selectivity was essentially the same when the Hoveyda–Grubbs second-generation catalyst was investigated in the RCM reaction (entries 7 and 8). Compound 19 could be converted back to goniothalamin (1) using an olefin cross-metathesis reaction with styrene. However, considering that both olefins should present the same reactivity,<sup>20</sup> a large excess of 19 or styrene would be required in view of this nonselective cross-metathesis reaction. Indeed, Das et al. performed the synthesis of goniothalamin (1) using this strategy; however, a large excess of 19 (20 equiv) was employed.<sup>20</sup>

The reaction with ester 16 in the presence of Grubbs and Hoveyda–Grubbs second-generation catalysts yielded better selectivities (entries 10 and 13), which could be explained by the lower reactivity of the propylene by-product in the cross-metathesis reaction. Surprisingly, the reaction carried out under identical conditions, but at a lower molar concentration (entry 10, 0.01 M), afforded goniothalamin (1) as a single product, although in a lower conversion (80%). Such similar results were not observed using ester 14 (entries 2 and 4). The higher dilution combined with the lower reactivity of propylene completely suppressed the intermolecular cross-metathesis reaction. However, this reaction performed under essentially identical conditions but with a longer residence time gave comparable results, indicating that the reaction reached equilibrium.<sup>21</sup> Finally, the selectivity was found to decrease at a higher reaction concentration of ester 16 in both Grubbs and Hoveyda–Grubbs second-generation catalysts (entries 12 and 14).

Additional olefin metathesis experiments were performed using a gas–liquid tube-in-tube flow reactor. The tube-in-tube reactor relies on the use of a semipermeable membrane (Teflon AF-2400), which allows the loading of the desired gas into the liquid stream in view of its permeation through the membrane. In the last decade, several reactions have been explored using this membrane technology.<sup>22</sup> For example, Knapkiewicz and co-workers demonstrated homo- and heterogeneous olefin metathesis using a tube-in-tube reactor of this type.<sup>23</sup> In this configuration, ethylene was continuously removed by connecting the reactor to a vacuum pump, and substantial improvement was perceived for macrocyclization metathesis reactions. Based on this precedent, the same strategy was evaluated for this study (Table S5). However, no

## Scheme 8. Preparative-Scale Synthesis of Goniotalamin (1) in the Batch Mode



improvement was observed using a tube-in-tube reactor (90 cm; 1.25 mL·min<sup>-1</sup> flow rate) for ester 14, i.e., the same selectivity and yield compared to entry 4 in Table 2 was found using this reactor. In our case, the cyclization step is likely to be fast, meaning that the catalyst turnover is quick enough to compete with ethylene removal. Conversely, in the macrocyclization reaction, the resting state would likely be substrate bound to catalyst before cyclization, meaning ethylene has more time to be purged.

The use of sacrifice olefin (1-heptene) in the flow regime was also explored but led to no improvements (Table S5). Although all attempts to improve the selectivity of this RCM reaction at higher concentrations failed, it is worth mentioning that continuous flow processing could at least deliver significant conversions in tighter residence times than batch protocols using the same concentration.

In view of the limitation imposed by the formation of the lactone by-products 19 and 20 and the high dilution issues under different flow strategies, which in turn presents a constraint for throughput and productivity, we decided to perform the final step in the synthesis of goniotalamin (1) on a preparative scale in the batch mode (Scheme 8). For this reaction, a 12 g sample of ester 16 was employed in the ring-closing metathesis reaction using the Grubbs second-generation catalyst. The reaction was performed with sparging of nitrogen gas and heated to reflux in dichloromethane. Full conversion of the substrate was observed after 1.5 h of reaction, and the target product was isolated in 78% yield (7.75 g). However, lactone 20 was also isolated in 19% yield.<sup>24</sup> In comparison to the same reaction performed at 1 mmol scale (Table 1, entry 5), the overall chemical yield was similar; however, the selectivity was affected by the less efficient elimination of the propylene gas from the reaction mixture, in view of the poorer surface area-to-volume ratio that occurs in a larger batch vessel (10 L) under conventional magnetic stirring. Although good quantities of the desired compound could be obtained, this result highlights the current limitations of our synthetic approach and equipment capabilities as far as scale-up is concerned.

As new reactor technologies emerge, the synthesis and preparation of chemicals of interest should be re-evaluated to look for more streamlined approaches;<sup>25</sup> herein, our current best route to goniotalamin (1) consists of two flow steps followed by a batch process.

### 3. CONCLUSIONS

In summary, the synthesis of the styryl lactone goniotalamin (1) was investigated in a continuous flow regime. The 1,2-addition of the Grignard reagent to *trans*-cinnamaldehyde (10) and the acylation reaction of the resulting allylic alcohol 12 could be combined in a telescoped process leading to the formation of ester 16 in 96% yield, for two steps. Running this sequence by continuously pumping the starting materials over a period of 3 h led to the preparation of 20.9 g of material,

giving a productivity of 7 g·h<sup>-1</sup>. An asymmetric Brown allylation reaction was also demonstrated in a continuous flow, leading to excellent yield (98%) and ee (91.5%) for (*S*)-12. The ring-closing metathesis reaction was evaluated in batch and flow conditions using three different ruthenium catalysts. In the batch mode, the reaction could be performed with good to excellent selectivities to give goniotalamin (1) in excellent yield (up to 96%). The use of the Grubbs second-generation catalyst proved to be very efficient, and the reaction time decreased from 7 to 1.5 h in comparison to the use of the Grubbs first-generation catalyst, even at lower catalyst loadings.

Even though full conversion could be obtained in a short residence time under continuous flow conditions, selectivity and yield were negatively affected due to the entrapment of ethylene or propylene. In the closed flow system, the evolving gases underwent an olefin cross-metathesis reaction with the styryl moiety of goniotalamin (1) affording lactones 19 or 20, in up to a 1:1 molar ratio. Different flow strategies, including a tube-in-tube reactor for ethylene/propylene removal and sacrifice of olefin, were unsuccessfully employed. On the other hand, good yield and selectivity could be achieved in flow at reduced residence times at the cost of incomplete conversion (16 min, 80% conversion, and 75% yield) using a more dilute solution. In view of the formation of by-products and high dilution issues, which in turn presents a limitation for throughput and productivity, a preparative-scale synthesis of goniotalamin (1) was performed under the optimized batch conditions. In this way, 7.75 g of goniotalamin (1) was obtained in 78% yield. Our work demonstrates that the combination of batch and continuous flow processes is a useful strategy when one or more steps of a target molecule can be maximized in terms of selectivity and yield.

## 4. EXPERIMENTAL SECTION

**4.1. Materials and Methods.** Reagents were obtained from commercial sources and used as received unless otherwise stated. All reactions were carried out with anhydrous solvents, oven-dried glassware (200 °C), and manipulated under an argon atmosphere unless otherwise stated. Tetrahydrofuran (THF) was freshly distilled under argon from sodium/benzophenone ketyl prior to use. Allylmagnesium chloride, at a concentration of 2.0 M in THF, was obtained from Sigma-Aldrich and titrated before its use. When necessary, dilution was performed by the addition of anhydrous THF. Compounds 1, 12, and 16 were purified by flash column chromatography using a Biotage Isolera system using prepacked silica gel 60 Å cartridges. TLC analysis was performed utilizing 0.25 mm plates precoated with silica gel 60 UV254, with the compounds being visualized by the use of UV light and/or aqueous potassium permanganate dip when appropriate. <sup>1</sup>H NMR and <sup>13</sup>C NMR data were recorded on a Bruker Avance (400 MHz for <sup>1</sup>H and 100 MHz for <sup>13</sup>C NMR) spectrometer using the residual solvent peak as the internal reference (CDCl<sub>3</sub> = 7.26 ppm for <sup>1</sup>H and 77.0 ppm for <sup>13</sup>C).



The data are reported as follows: chemical shift in ppm ( $\delta$ ), multiplicity (s = singlet, d = doublet, t = triplet, q = quartet, quint = quintet, m = multiplet, br = broad signal, or combinations of thereof), coupling constant (Hz), integration. Enantiomeric excess (% ee) for Roush and Brown allylation reactions was determined by chiral HPLC: 99:01 *n*-hexanes/*i*-PrOH, CHIRALPAK AD-H, 0.9 mL·min<sup>-1</sup>, 46 and 49 min.

Reactors and interconnecting lines were constructed using a PFA polymer tubing of 1.0 mm i.d., the exception being the fluoropolymer peristaltic tubes, which were supplied by Vapourtec. Most of the experiments were carried out using a Vapourtec E-series flow system and the cooled reactor assembly, except for the preparation of compound **16**, where an external pump (K-120 Knauer pump) was used to deliver the Grignard Reagent. A Uniqsis FlowSyn System was used in the study of the RCM reaction. Please see the SI for pictures of the flow equipment and setups discussed and used.

For setting up the peristaltic flow system for each experiment and cleaning down afterward, we followed the general protocol reported in our previous study on the pumping of organometallic reagents.<sup>13a</sup>

**4.2. Continuous Flow Synthesis of (*E*)-1-Phenylhexa-1,5-dien-3-ol (**12**).** The flow system was set up according to Scheme 3. A 1.0 M solution of *trans*-cinnamaldehyde (**10**) in anhydrous THF (100 mL), under argon, was combined at a T-piece with a 1.97 M solution of allylmagnesium chloride (**11**) in anhydrous THF (100 mL), under the flow rates indicated in Scheme 3. The combined stream was passed through a coil reactor (10 mL) kept at 25 °C with continuous cooling and pressurized by a 75 psi back-pressure regulator. After the steady-state operation, the output stream was collected over 11.5 min into a conical flask containing a saturated aqueous ammonium chloride solution (50 mL) at room temperature under stirring. The collected material was transferred to a separatory funnel, and the aqueous phase was extracted with diethyl ether (3 × 50 mL). The combined organic phase was dried over anhydrous magnesium sulfate, filtered, and concentrated to dryness. The crude product was subjected to flash column chromatography on silica using the Biotage autocolumn system, eluting in 5–10% ethyl acetate/petroleum ether 40–60 to give alcohol **12** in 91% yield (1.83 g), obtained as a colorless viscous oil.

**4.3. Telescoped Synthesis of (*E*)-((*E*)-1-Phenylhexa-1,5-dien-3-yl) but-2-enoate (**16**).** **4.3.1. Procedure A: Using the Vapourtec E-Series (Pumps A–C).** The flow system was set up according to Scheme 5. A 0.5 M solution of *trans*-cinnamaldehyde (**10**) in anhydrous THF (250 mL), under argon, was combined at a T-piece with a 0.89 M solution of allylmagnesium chloride (**11**) in anhydrous THF (250 mL). The combined stream was passed through a PFA reactor coil (10 mL) kept at 25 °C with continuous cooling. The output stream was combined in-line at a T-piece with a 0.5 M solution of crotonic anhydride in anhydrous THF (250 mL). The new combined stream was passed through a second PAF reactor coil (10 mL) held at 50 °C, and the whole system was pressurized by a 75 psi back-pressure regulator. The three solutions were pumped at the flow rates indicated in Scheme 5. After the steady-state operation, the output stream was collected over 10 min into a conical flask containing a saturated aqueous ammonium chloride solution (100 mL) at room temperature under stirring. The collected material was transferred to a separatory funnel, and the aqueous phase was extracted with diethyl ether (3 × 30 mL). The combined

organic phase was dried over anhydrous magnesium sulfate, filtered, and concentrated to dryness. The crude product was subjected to flash column chromatography on silica using the Biotage autocolumn system, eluting in 4–30% ethyl acetate/petroleum ether 40–60, to afford ester **16** in 73% yield (0.89 g), obtained as a light-yellow oil.

**4.3.2. Procedure B: Using the Vapourtec E-Series (Pumps A and C) and the K-120 Knauer Pump.** The flow system was set up according to Scheme 5. A 0.5 M solution of *trans*-cinnamaldehyde (**10**) in anhydrous THF (250 mL), under argon, was combined at a T-piece with a 0.88 M solution of allylmagnesium chloride (**11**) in anhydrous THF (100 mL). The combined stream was passed through a PFA reactor coil (10 mL) kept at 25 °C with continuous cooling. The output stream was combined in-line at a T-piece with a 0.5 M solution of crotonic anhydride in anhydrous THF (250 mL). The new combined stream was passed through a second PAF reactor coil (10 mL) held at 50 °C, and the whole system was pressurized by a 75 psi back-pressure regulator. The three solutions were pumped at the flow rates indicated in Scheme 5. After the steady-state operation, the output stream was collected over 3 h into a conical flask containing a saturated aqueous ammonium chloride solution (400 mL) at room temperature under stirring. The collected material was transferred to a separatory funnel, and the aqueous phase was extracted with diethyl ether (2 × 200 mL). The combined organic phase was dried over anhydrous magnesium sulfate, filtered, and concentrated to dryness. The crude product was subjected to flash column chromatography on silica using the Biotage autocolumn system, eluting in 1–30% ethyl acetate/petroleum ether 40–60, to afford ester **16** in 96% yield (20.9 g), obtained as a light-yellow oil.

**4.4. Procedure for the Continuous Flow Asymmetric Allylborane Addition to *trans*-Cinnamaldehyde (**10**).** The flow system was set up according to Scheme 6, and the reactor assembly was kept at –78 °C, due to the exothermic reaction. A 0.5 M solution of *trans*-cinnamaldehyde (**10**) in anhydrous Et<sub>2</sub>O (100 mL), under argon, was combined at a T-piece with a 0.6 M solution of (–)-*B*-allyldiisopinocamphe-nylborane in anhydrous Et<sub>2</sub>O (100 mL). The combined stream was passed through a PFA reactor coil (10 mL) kept at –78 °C with continuous cooling. The output stream of the reactor was combined in-line with neat MeOH (250 mL), and the whole system was pressurized by a 100 psi back-pressure regulator. The three solutions were pumped at the flow rates indicated in Scheme 6. After the steady-state operation, the output stream was collected over 15 min into a round-bottomed flask (50 mL) containing 10 mL of 30% aq H<sub>2</sub>O<sub>2</sub>/3 M NaOH aq at 0 °C under stirring. Next, the resulting solution was warmed up to rt over 1 h. The collected material was transferred to a separatory funnel, and the aqueous phase was extracted with diethyl ether (3 × 50 mL). The combined organic phase was dried over anhydrous magnesium sulfate, filtered, and concentrated to dryness. The crude product was subjected to flash column chromatography on silica using the Biotage autocolumn system, eluting in 5–10% ethyl acetate/petroleum ether 40–60, to afford the alcohol (*S*)-**12** in 98% yield (391.5 mg), obtained as a colorless viscous oil.

**4.5. Segmented Continuous Flow Ring-Closing Metathesis Reaction.** Procedure for the flow preparation of goniothalamine (**1**) (exemplified for entry 12, Table 2).

The Uniqsis FlowSyn reactor was fitted with two 5 mL injection loopings and attached to a PFA reactor coil (16 mL,

1.0 mm i.d.), which was placed on a heating block. The system was primed with CH<sub>2</sub>Cl<sub>2</sub> at a flow rate of 0.5 mL/min per channel and the temperature was set to 100 °C. A solution of ester **16** in dry CH<sub>2</sub>Cl<sub>2</sub> (0.1 M, 0.5 mmol, 5.0 mL) and a solution of Grubbs second-generation catalyst in dry CH<sub>2</sub>Cl<sub>2</sub> (5 mol %, 0.025 mmol, 5 mL) were loaded into two 5 mL injection loopings (injection loopings A and B, Figure S3). To initiate, injection loopings A and B were simultaneously inserted into the main flow stream via a T-piece connector at a flow rate of 0.5 mL/min per channel. The combined stream was then directed through a PFA reactor coil (16 mL, 1.0 mm i.d.), which was maintained at 100 °C, giving a residence time of 16 min. After exiting the system via a BPR (6.0 bar), the output stream was collected during 30 min in a conical flask. The solution was passed through a plug of silica flash, and the volatiles were removed under vacuum. The crude product was subjected to flash column chromatography on silica using the Biotage autocolumn system, eluting in 20–60% ethyl acetate/petroleum ether 40–60, to afford an inseparable mixture of goniothalamin (**1**) and lactone **20** in 94% combined yield.

**4.6. Procedure for the Batch Preparation of Goniothalamin (1).** To a solution of ester **16** (50 mmol) in anhydrous CH<sub>2</sub>Cl<sub>2</sub> (5.0 L, 0.01 M) was added the Grubbs second-generation catalyst (5 mol %, 2.5 mmol, 2.1 g). After 1.5 h under reflux, the reaction was allowed to cool down to room temperature, DMSO (50 equiv relative to the catalyst) was added, and the mixture was kept under these conditions overnight. After that, the volatiles were removed under vacuum and the crude product was subjected to flash column chromatography (petroleum ether 40–60/ethyl acetate 3:1, 2:1, and 1:1 v/v) to afford goniothalamin (**1**) in 78% yield (7.75 g) and lactone **20** in 19% yield (0.81 g).

## ■ ASSOCIATED CONTENT

### Supporting Information

The Supporting Information is available free of charge at <https://pubs.acs.org/doi/10.1021/acsomega.0c02390>.

<sup>1</sup>H and <sup>13</sup>C NMR spectra, flow setup pictures, and optimization procedures can be found (PDF)

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## Notes

The authors declare no competing financial interest.

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