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Described is a continuous, divergent synthesis system which is coupled to continuous purification and is capable of producing four anti-malarial APIs. The system is comprised of three linked reaction modules for photooxidation/cyclization, reduction, and derivatization. A fourth module couples the crude reaction stream with continuous purification to yield pure API.

As the global demand for medicines has increased, efforts to implement continuous Active-Pharmaceutical Ingredient (API) production have intensified.¹ Continuous processes hold many potential benefits including increased safety and product quality, as well as lower environmental impact.² While proof-of-concept continuous API syntheses^{3,4} focused on linear transformations, we describe the combination of a divergent, multi-step continuous synthesis with continuous in-line purification to prepare several artemisinin-derived APIs that are key components in Artemisinin Combination Therapies (ACT), the World Health Organization's (WHO) recommended first-line treatment for malaria.

Today, the key active pharmaceutical ingredients of all ACT anti-malarials are produced in one or two chemical steps from artemisinin (3) (Fig. 1). The majority of artemisinin (~200 tons per year) is extracted from the plant *Artemisia annua* and prices fluctuate with harvest yields.⁵ The plant waste material dihydroartemisinic acid (DHAA) 2 has also been used to synthesise artemisinin *via* continuous flow photochemistry with subsequent

Continuous synthesis of artemisinin-derived medicines†

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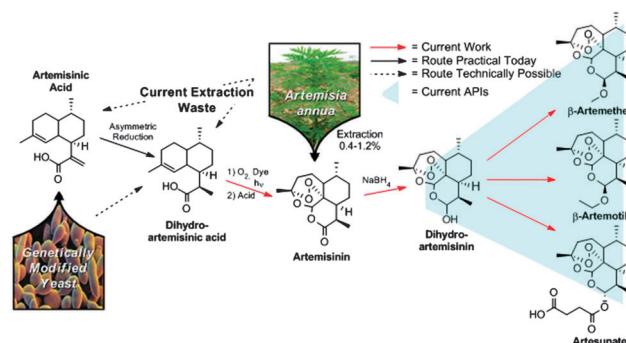


Fig. 1 Overall production scheme of anti-malaria APIs from artemisinin obtained by traditional extraction from *Artemisia annua* and genetically modified yeast combined with chemical modification. Dihydroartemisinin (**4**, combined with piperaquine in Eurartesim, Artekin and Duo-Cotecxin), β-artemether (**5**, combined with lumefantrine in Coartem), β-arteether (**6**, Artemotil), and α-artesunate (**7**, combined with amodiaquine in Coarsucam and ASAQ-Winthrop).

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† Electronic supplementary information (ESI) available: Experimental details of reaction parameters, purification, and NMR spectra are available free of charge, see DOI: 10.1039/c4cc05098c

acid-induced rearrangement, resulting in a fast (residence time less than 12 minutes) and efficient (69% yield) transformation.^{6,7}

ACT medications rely not only on the procurement of artemisinin, but also its conversion to the corresponding APIs (**4-7**). To achieve a completely continuous synthesis and purification of the three main anti-malarial APIs, we built upon the previously described continuous synthesis of artemisinin **3** from DHAA **2** (Fig. 3, module 1).⁶ While lithium triethylborohydride (Super-hydride[®]) effects the reduction of pure artemisinin **3** to **4** in a continuous flow chemistry setting,⁸ this reagent is an order of magnitude more expensive than the NaBH₄ used in traditional batch syntheses. Super-hydride[®] is even more expensive than artemisinin, making it unsuitable for the production of a price-sensitive API. Consequently, a continuous process using inexpensive NaBH₄ had to be developed.⁹

Several challenges had to be overcome, including the insolubility of NaBH₄ in the toluene-TFA solvent mixtures exiting module 1. While NaBH₄ suspensions have been used in batch to prepare **DHA**, the use of slurries in flow remains a major challenge,¹⁰ particularly



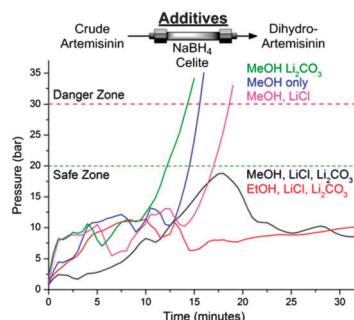


Fig. 2 System pressure observed during reduction of crude artemisinin 3 as a function of cosolvent and solid additives in the Celite[®]/NaBH₄ column.

when envisioning prolonged production runs. A NaBH₄-containing packed-bed column would be an attractive means of reducing 3 to 4. While packed-beds containing NaBH₄/Celite[®] mixtures (1:1 (w/w))¹¹ successfully reduced artemisinin from the outflow of module 1 to DHA, columns packed in this way proved unstable (see ESI[†]). Different column additives to both accelerate the reduction and stabilise the column were screened (Fig. 2). Lithium chloride (LiCl) accelerates NaBH₄ reductions *via* the *in situ* formation of LiBH₄,¹² but clogging remained a problem. Buffering the trifluoroacetic acid (TFA) contained in the reaction stream by the addition of lithium carbonate (Li₂CO₃) also did not remedy the clogging problems. However, upon combining the two additives and mixing NaBH₄, Celite[®], Li₂CO₃, and LiCl (1:1:1:0.76 (w/w)) as packed-bed materials and using an alcohol co-solvent such as ethanol,¹² complete and clean reduction of crude artemisinin was achieved (Fig. 2, red line). By combining modules 1 and 2, the first API (4) is produced from DHAA 2, with module 2 representing a general solution to achieve efficient reductions of carbonyl compounds in continuous flow, with a reduction capacity which is stoichiometric with respect to NaBH₄.¹³

The mixed methyl acetal 5, API of the drug Coartem, is accessed by combining artemisinin reduction and methyl ether formation (modules 2 and 3, Fig. 3). Previous attempts at

Table 1 The effect of hydride source and intermediate wash on the epimeric ratio of artemether following treatment with Feed B_{AM} in batch

Entry	Hydride source	Intermediate wash	β:α
1	Superhydride [®] (LiEt ₃)	None	50:50
2	Superhydride [®] (LiEt ₃)	H ₂ O	50:50
3	Superhydride [®] (LiEt ₃)	H ₂ O/ethanolamine (3/1, v/v)	80:20
4	NaBH ₄ Column	None	75:25
5	NaBH ₄ Column	H ₂ O	81:19
6	NaBH ₄ Column	H ₂ O/ethanolamine (3/1, v/v)	82:18

developing a combined continuous process were unsuccessful due to sensitivity following NaBH₄ reduction.¹⁴ As such, the effects of both hydride source (1 M LiEt₃BH or reduction column) and workup conditions on the resulting epimeric ratio of β:α artemether were examined in batch following treatment with Feed B_{AM} (1.8 M HCl in 2:1 v/v methanol/trimethyl orthoformate). The ratio was determined by ¹H NMR of the crude reaction mixture following basic workup of the second step, comparing the doublets at 4.65 ppm (β-artemether 5, *J* = 4 Hz) and 4.32 ppm (α-artemether, *J* = 8 Hz).¹⁵

As expected, the telescoping of the reduction and mixed acetal formation without any intermediate workup resulted in a poor mixture of artemether epimers for both hydride sources (Table 1, entries 1 and 4).¹⁴ It appears the intermediate alkoxyborane species that is formed upon reduction using LiEt₃BH is stronger than that emanating from Na/LiBH₄, thus requiring an ethanolamine wash to break it down and give acceptable epimeric ratios (entries 2 and 3). However, upon exiting the reduction column, a simple aqueous wash (entry 5) was sufficient to obtain epimeric ratios equal to those resulting from pure dihydroartemisinin.¹⁴

In flow, the reaction stream exiting module 2 (pH ~ 10) was continuously degassed¹⁶ and extracted¹⁷ with water before the organic stream was combined with Feed B_{AM} *via* a T-mixer. Complete etherification occurred within a 25 minute residence time at 25 °C, providing artemether 5 as a 81:19 ratio of β:α epimers, similar to that obtained *via* traditional batch synthesis.¹⁴ β-Artemether 5 was obtained in 25% overall yield from DHAA 2 following basic workup and isolation. In a similar manner, artemotil 6 was obtained as a 5:1 mixture of epimers (β:α) in 22% yield from 2 when Feed B_{AE} consisted of a 1.8 M HCl solution in ethanol/triethyl orthoformate (2:1 v/v).

Artesunate 7, the active ingredient in Coarsucam, was continuously prepared from DHAA using a succinic anhydride solution (1.6 M containing one equiv. triethylamine in dichloromethane) in Feed B_{AS} at a 25 minute residence time at 25 °C. Exclusively the desired α-epimer was formed, to yield 7 in 28% from DHAA 2. The crude solution of this derivative was used to demonstrate continuous purification (*vide infra*).

The entire three module continuous flow process to convert 2 to the APIs 5–7 requires a reaction time of 46 minutes (3.4 mmol per hour through the laboratory-scale system). Importantly, not only can 5, 6, or 7 be synthesised simply by exchanging the solution in Feed B and the acid/base quench (Fig. 3), any further artemisinin C10 derivatives that are under investigation as future anti-malarial APIs¹⁸ can be prepared by using this modular synthesis system.

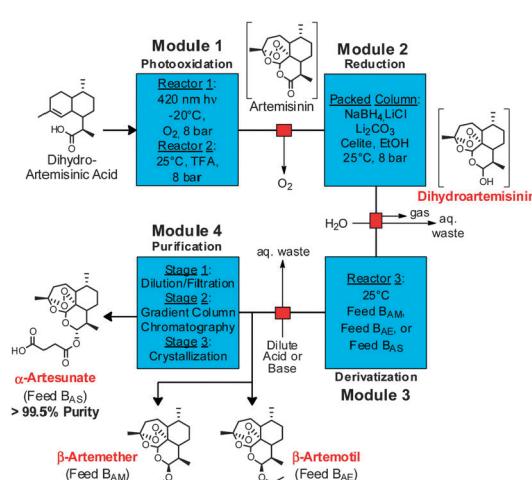


Fig. 3 Four module system for the continuous synthesis and purification of artemisinin APIs (in red text). TFA: trifluoroacetic acid, EtOH: ethanol.



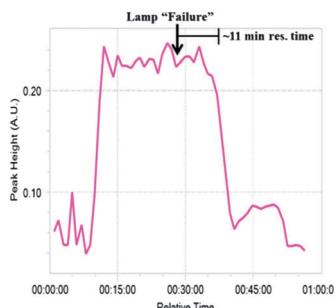


Fig. 4 The reaction exiting module 1 was monitored (FlowIR) at 1033 cm^{-1} . To simulate an equipment error, the lamp was turned off during the reaction, resulting in cessation of the monitored peak following the ~ 11 minute residence time of module 1.

With the chemical steps defined, continuous monitoring and purification were introduced. FlowIR has been shown to be efficient to observe continuous systems in real time¹⁹ and was utilized to monitor the system after each reaction module. In module 1, the plant waste material DHAA 2 is converted to artemisinin 3. When the lamp was turned off to simulate a “lamp failure”, the result is observed approximately 11 minutes later when product 3 (1033 cm^{-1}) was no longer observed in the IR trace (Fig. 4). Similarly, the outputs from modules 2 and 3 were also successfully monitored (see ESI†), allowing for facile quality control at each stage of production.

The continuous synthesis of drug substances is ideally complemented by continuous purification to yield pure APIs that meet the standards set by regulatory bodies such as the FDA²⁰ or WHO.²¹ To demonstrate the power of the fully continuous synthesis/purification regime, a continuous three-stage purification method (filtration/multi-column chromatography/crystallisation) was developed for α -artesunate 7. The first stage dilutes the stream exiting module 3 with *n*-hexane, which decreases the solvent strength and precipitates polar byproducts. The recovery yield

of 7 after dilution and filtration of the reaction mixture is 65%. Following filtration, the artesunate-containing stream can enter a separator consisting of five identical chromatography columns participating in the following steps: loading, elution, rinsing, and equilibration (see ESI†). This multi-column arrangement, together with the periodic switching between two filters, permits a permanent feed supply. The artesunate fraction can be subsequently fed directly into a continuous crystalliser to remove the remaining impurities (Fig. 5). With 73% of 7 recovered from the crystalliser, the overall recovery yield of α -artesunate from the crude exit stream of module 3 is 48% with an HPLC purity of greater than 99.5%, exceeding the requirements set by the WHO.²¹

This continuous, modular approach for the divergent synthesis of small molecules has attractive scientific and conceptual implications. Batch processes are significantly more labour-intensive, as they require solvent removal and purification of the intermediates following each step. The use of potentially dangerous reagents, such as oxygen or sodium borohydride, becomes possible in flow and allows for new reaction pathways,²² and the ability to combine/interchange different reaction modules allows for the production of a variety of APIs. The synthesis of the four anti-malarial APIs described here illustrates this principle and with this first example of a divergent, multi-step continuous synthesis and purification process, many routes to APIs of generic and patented drugs can be envisioned.

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Notes and references

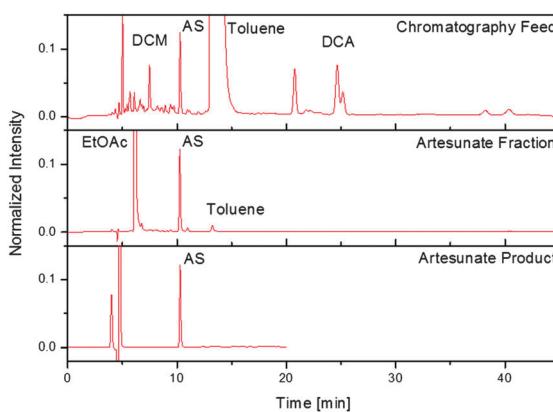


Fig. 5 Comparison of HPLC chromatograms exiting module 3 (top), following gradient elution chromatography (middle), and recrystallisation (bottom). Initial peaks in the bottom frame correspond to ethanol injection marks and the run was stopped after 20 minutes due to lack of peaks observed in the chromatogram following the previous purification step (middle). DCM: dichloromethane, EtOAc: ethyl acetate, AS: α -artesunate, DCA: 9,10-dicyanoanthracene.

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