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The Synthesis of (±)-Oxyisocyclointegrin

Robert J. Smith,[a] Rebekah L. Bower,[b] Scott A. Ferguson,[c] Rhonda J. Rosengren,[b] Greg M. Cook[c] and Bill C. Hawkins*[a]

Abstract: The total synthesis of oxyisocyclointegrin is described. The key steps in the synthesis included a Tsuji-Trost allyl migration to exclusively afford the corresponding monoallylated 1,3-diketone and a photochemical initiated oxidative cyclization to forge the oxepine core.

Introduction

The wood of the *moraceae* family, which includes the mulberry and fig tree, is a rich source of phenol and flavone-derived natural products. Most notable is a series of prenylated flavones and several oxidised derivatives with a common tricyclic or tetracyclic core.[1] One of the most studied subsets of these natural products are the morusins (morusin (**1**), cyclomorusin (**2**), neocyclomorusin (**3**), Figure 1). Investigations into the biological activity of these compounds have discovered a wealth of potential medical applications, with activity including anti-cancer (neocyclomorusin, morusin, cyclomorusin) and antibacterial properties (neocyclomorusin, morusin), inhibition of platelet aggregation (neocyclomorusin) and radical scavenging ability (neocyclomorusin).[2] Among the first reported compounds from this family of structurally related molecules are integrin (**4**) and oxyisocyclointegrin (**5**).[3] It is not clear if oxyisocyclointergrin (**5**) exists as a racemate or as a single stereoisomer as no optical rotation data has been reported. To the best of our knowledge no biological activity data for either **4** or **5** has been reported in the literature. Given the promising biological activity associated with other structurally similar members of this natural product family (*vide supra*), a robust and facile entry into these natural products would be of great interest. Herein we report the first total synthesis of the natural products integrin (**4**) and oxyisocyclointegrin (**5**) as well as preliminary biological data indicating both compounds possess moderate to weak antibacterial and anticancer properties.

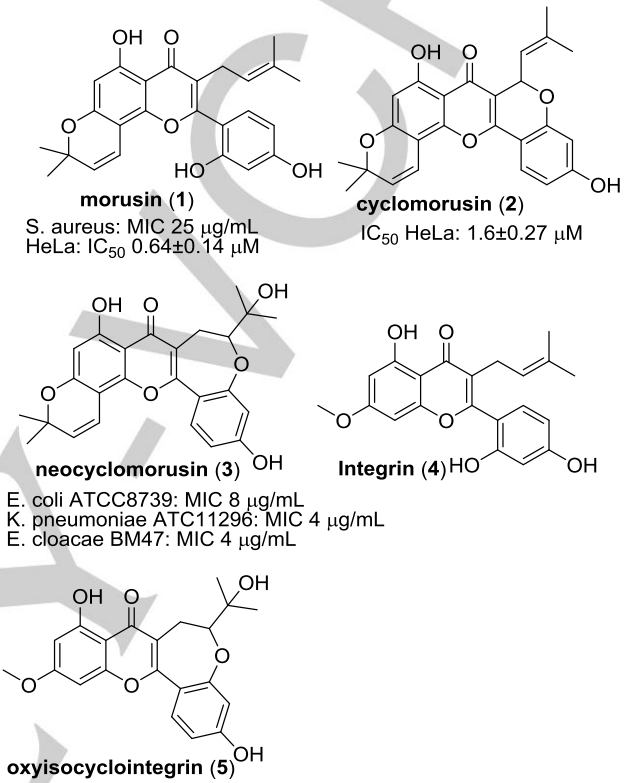

Figure 1. Representative natural products from the morusin family.

Results and Discussion

Unsurprisingly, retrosynthetic analysis of oxyisocyclointegrin (**5**) revealed integrin (**4**) to be a direct precursor, which could be converted to the natural product by an oxidative cyclization strategy (Scheme 1). Preparation of the natural product integrin could be performed by cross metathesis of the 3-allylflavone **6**. Synthesis of this key intermediate necessitates the preparation of the 2-allyl-1,3-diketone **7**. We propose that a Tsuji-Trost type allyl migration of the 1-(phenyl-(2-O-allyl)-1,3-diketone (**8**) would provide a concise method to access this substrate, whilst avoiding the potential for over alkylation of the 1,3-diketone.[4]

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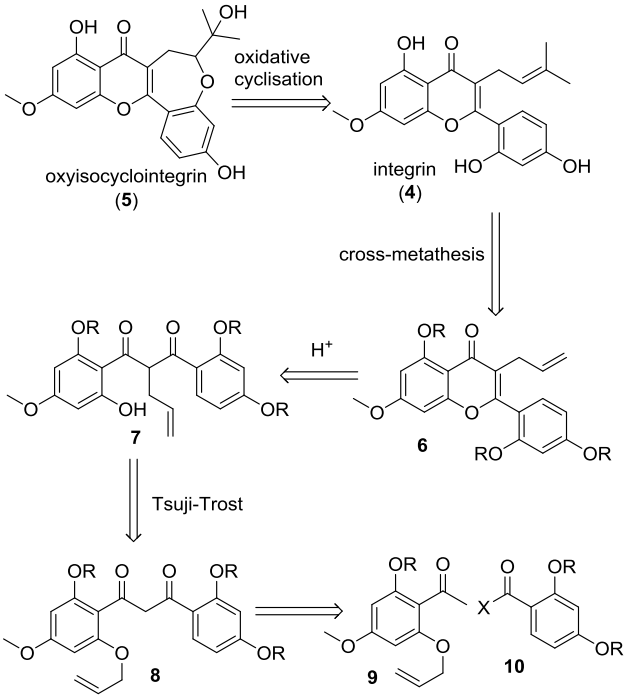
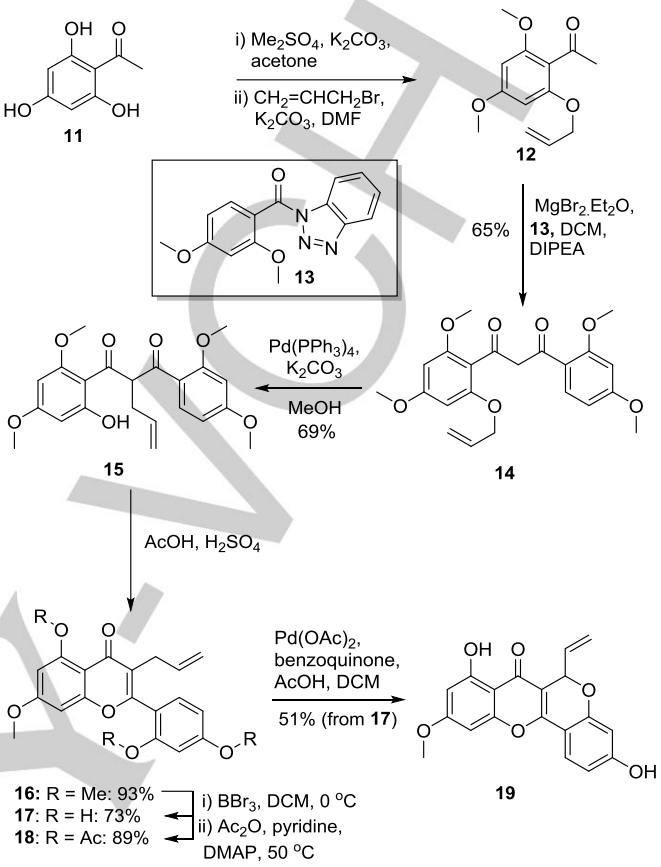


Scheme 1. Retrosynthetic analysis of oxyisocyclointegrin

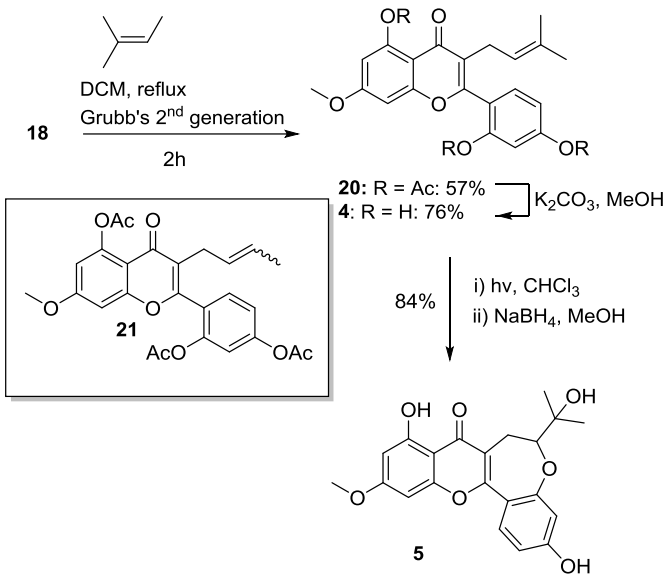


Scheme 2. Construction of the flavone core

In a forward synthetic sense, the synthesis began with the known ketone **12**, which was prepared from the commercially available 2,4,6-trihydroxyacetophenone following literature procedures (Scheme 2).[5] Standard Claisen condensation protocols failed to deliver significant quantities of the corresponding 1,3-diketone. Recently, soft enolization of ketones followed by acylation using acylbenzotriazoles has been shown to be an effective way of generating 1,3-diketones.[6] Pleasingly, treatment of **12** and the acylbenzotriazole **13** with magnesium bromide diethyletherate and diisopropyl ethyl amine smoothly provided the electron rich diketone **14** in a 65% yield.[6] With this substrate in hand we turned our attention to the Tsuji-Trost type allyl migration, exposure of the ketone **14** to $Pd(PPh_3)_4$ and potassium carbonate in methanol resulted in exclusive formation of the mono α-allyl 1,3-diketone **15** in a 69% yield.[7] Subsequent treatment of **15** with 10% H_2SO_4 in AcOH provided the 3-allylflavone **16** in an excellent yield. Selective demethylation was achieved upon careful treatment of the tetramethylether **16** with three equivalents of BBr_3 providing the desired mono-methylether flavone **17** in a 73% yield (Scheme 2). Flavone **17** was found to readily undergo palladium-mediated intramolecular oxidative cyclization,[8] to afford the vinylpyran **19**, a synthetic analog of cyclomorusin (**2**), in a 51% yield.

Cross metathesis of flavone **17** with 2-methyl-2-butene using Grubb's second-generation catalyst was met with failure, presumably due to the free phenol functional groups interacting with the catalyst. Acetylation of **17** under standard reaction conditions provided the triacetate derivative **18** in good yield. The acetate derivative **18** proved to be more amenable to cross-metathesis, affording the desired 3-prenylflavone **20** in a modest 57% yield (Scheme 3). The low yield was the result of the formation of by-products, including the alternate cross metathesis product **21**. Methanolysis of **20** provided the natural product integrin (**4**) in a 76% yield.

We proposed that cyclization of integrin (**4**) to form oxyisocyclointegrin (**5**) would be possible through an oxidative cyclization reaction. After extensive screening,[9] it was found that irradiation of integrin (**4**) in the presence of oxygen, with a high pressure mercury lamp for 7 hours, provided a mixture of oxyisocyclointegrin and its hydroperoxide (1:4 ratio, respectively determined by ^{1}H NMR).[10] The crude residue was then treated with sodium borohydride in methanol to provide the natural product oxyisocyclointegrin (**5**) in 84% yield over 2 steps. The synthetic material was spectroscopically equivalent to that reported for the isolated natural product.[3]



Scheme 3. The synthesis of isoxycyclointegrin (**5**)

Vinylpyran **19**, integrin (**4**) and oxyisocyclointegrin (**5**) were found to possess moderate to weak activity against *Staphylococcus aureus* (Table 1). Oxyisocyclointegrin (**5**) possessed weak activity against prostate cancer cells (LnCap).

Table 1. Antibacterial activity

Compound[a]	***S. aureus* ATCC 6538 MIC (μg/mL)**	**LnCap IC_{50} (μM)**
Vinylpyran **19**	16-32	60
Integrin (**4**)	32-64	60
Oxyisocyclointegrin (**5**)	128	16
Penicillin G (control)	0.03125	

Conclusions

In conclusion, we have reported the first total synthesis of integrin and oxyisocyclointegrin, these compounds were found to possess moderate antibacterial activity against *S. aureus*. Future work focusing on the synthesis of related natural products is underway and will be reported in due course.

Experimental Section

Method: Antimicrobial susceptibility testing was performed by measuring the minimum inhibitory concentration (MIC) by broth microdilution. *S. aureus* ATCC 6538 was grown overnight (16 h) at 37°C with shaking at 200 rpm in Cation Adjusted Mueller Hinton II Broth (CAMHB) (BD BBL™). The following day a cell suspension was prepared by diluting the overnight culture to an optical density (OD_{600}) of 0.003 in CAMHB. A 96 well polystyrene plate was then set up to contain a 2-fold dilution series of each compound in 50 µL CAMHB. The cell suspension (50 µL) was then added to the diluted compounds yielding an OD_{600} (final) of 0.0015 in 100 µL (final) to ensure that the cell concentration was adjusted to approximately 5×10^5 CFU/ml. Media, compound-free (untreated), and DMSO vehicle controls were also included. The plates were then incubated for 24 h at 37°C with shaking at 200 rpm. The MIC was reported as the lowest concentration of the test compound for which no growth occurred. Experiments were performed with biological replicates.

Keywords: flavone • natural product • total synthesis • oxepine• antibacterial

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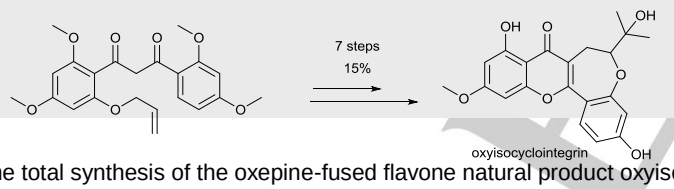

The total synthesis of the oxepine-fused flavone natural product oxyisocyclointegrin is described. The synthesis was achieved in 7 steps and an overall yield of 15%, oxyisocyclointegrin was found to possess moderate antibacterial and modest cytotoxicity.

Total Synthesis*

*Robert J. Smith, Rebekah L. Bower, Scott A. Ferguson, Rhonda J. Rosengren, Greg M. Cook and Bill C. Hawkins**

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