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An efficient synthesis of oosporein

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ABSTRACT

Fungal metabolite oosporein was prepared in four steps and 24% overall yield starting from 2,5-dimethoxytoluene. It was demonstrated that treatment of phoenicin with pyrrolidine and copper(II) acetate led to oosporein, whereas similar treatment of the isomeric 'isophoenicin' produced a benzofuran diquinone. No chromatography was required during any step of the synthesis.

The fungal metabolite oosporein **8** has been isolated from numerous sources.[1] It has shown antifungal,[1d] antibiotic,[2] and antiviral[3] activities, in addition to being the major secondary metabolite of some fungal biological control agents.[1c] Although its physicochemical properties have been studied extensively,[4] and there has been considerable interest in analytical methods for its detection,[5] an efficient synthesis of this compound has been lacking. One of the earliest syntheses was reported by Kögl and van Wessem,[6] in which the natural product phoenicin **6**, prepared by the method of Posternak,[7] was converted in two steps and 15% overall yield into oosporein. Many years later, Dallacker and Löhnert[8] prepared oosporein in seven steps and 10% overall yield starting from catechol, and a few years after that, Posternak and coworkers reported a synthesis starting from benzoquinone.[9] Posternak's approach required seven steps and produced oosporein in something less than 3% yield (the yield for the final step was not given). A key step in both of these syntheses was the copper-catalyzed dimerization of tetraoxygenated toluene derivatives to give the corresponding biaryls (via the aryllithium in Dallacker and Löhnert's synthesis, and via an aryl iodide in Posternak's synthesis). Although 35 years have passed, we are unaware of any new syntheses of oosporein having been reported since Posternak's paper.[10]

Recently we reported an improved protocol for the preparation of diquinones via oxidation of dimethoxyarenes with ceric ammonium nitrate (CAN).[11] As this methodology allows the preparation of ditoluquinone **1** in a high state of purity and >90% yield starting from commercially available[12] 2,5-dimethoxytoluene, we investigated the synthesis of oosporein **8** starting from this material, following much the same approach used by Kögl and van Wessem (Scheme 1).

Subjecting **1** to a Thiele-Winter acetoxylation[14] provided a mixture of 4,4′-bitoluene hexaacetates **2** and **3** in an approximately 1.2:1 ratio (**2**:**3**, by NMR) and 95% overall yield. This isomer ratio was consistent with that reported earlier in the literature.[7,14] Since ultimately our goal was to oxygenate four of the six carbons on each ring, this product mixture initially did not concern us, apart from the inconvenience of dealing with a mixture instead of a pure substance.

Acid-catalyzed methanolysis of the mixture of **2** and **3** provided products which were most easily isolated by simply allowing the solvent and by-products to evaporate over a period of several days.[15] The product mixture thus obtained consisted of a mixture of biaryl **4** and its isomer **5**, along with traces of methanol and HCl. As there were no other side products formed in the reaction (as evidenced by NMR), and there were no losses due to extraction or filtration, the yield was essentially quantitative.

Oxidation of biaryls **4** and **5** to phoenicin **6** and 'isophoenicin' **7**, respectively, could be accomplished in 41% yield through the use of iron chloride, according to Posternak's procedure.[7] No attempt was made to separate the two isomers at this point, either.

Kögl and van Wessem's[6] route to oosporein **8** involved treatment of phoenicin **6** with ethanolic methylamine (yielding tetraamino derivative **10**), followed by aqueous acid hydrolysis, and we attempted to replicate these results. Despite several attempts, we were never able to obtain significant amounts of oosporein following this procedure. Finding no examples of this reaction having been successfully repeated by others, we investigated other similar processes for the conversion of **6** and **7** into **8**. Since copper(II) acetate is known to facilitate addition of amines to benzoquinones,[16] a mixture of **6** and **7** was allowed to stir with copper(II) acetate and an excess of pyrrolidine. This reaction produced a very fine green-

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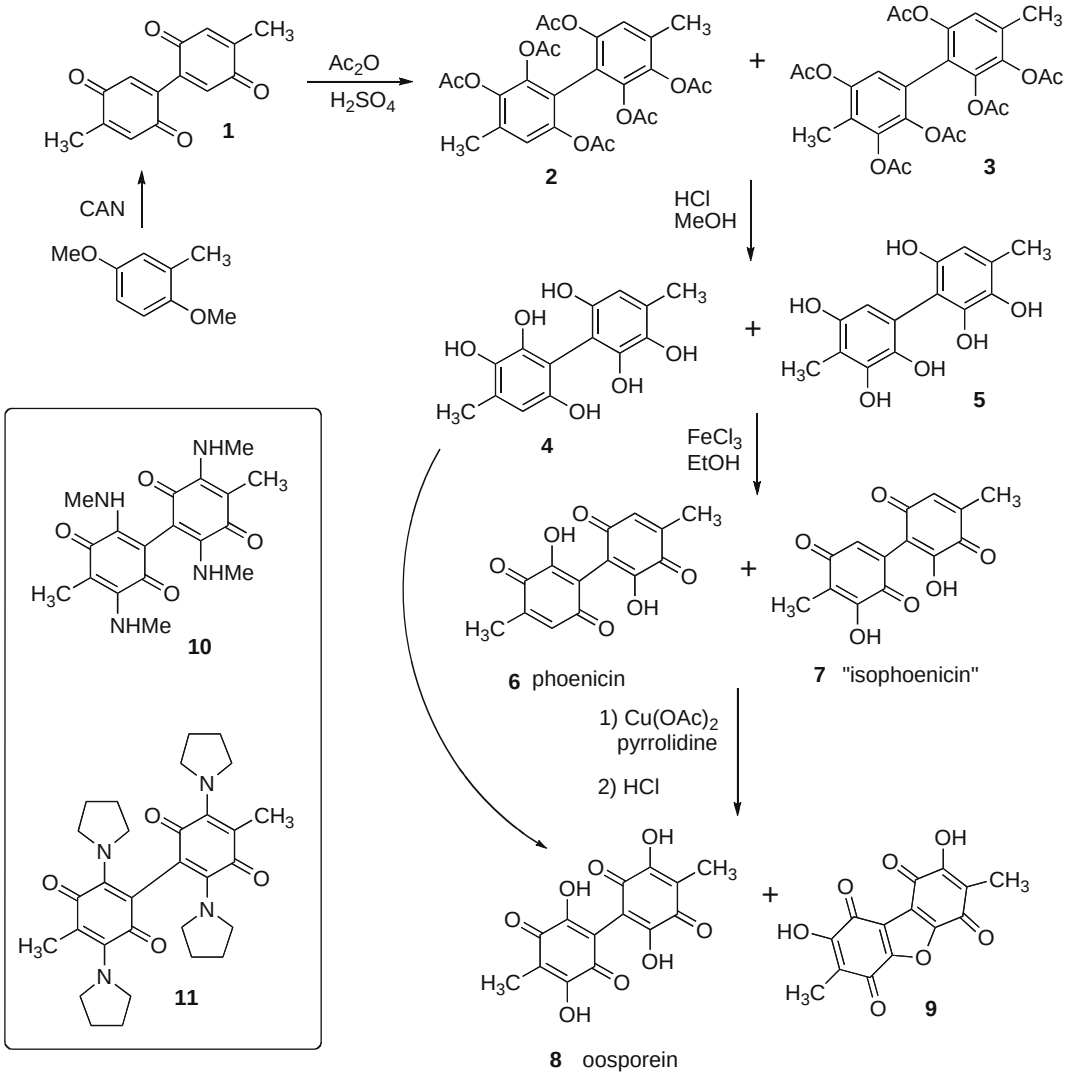

Scheme 1. Synthetic routes to oosporein.

black precipitate that was difficult to isolate by filtration, as it tended to rapidly clog the pores of the filter paper. Further, this solid was found to be insoluble in all typical organic solvents and was difficult to analyze by NMR. In addition to poor solubility in typical NMR solvents, spectra obtained from dilute solutions of this product displayed very broad peaks, suggesting perhaps the presence of a paramagnetic substance.[17] Replacement of the pyrrolidine with 1-methylpiperazine produced a product with increased solubility, but NMR spectra of this product still suffered from significant line broadening. In the end, it was found advantageous to simply proceed with the hydrolysis step without isolation of the intermediate product(s).

Hydrolysis of the crude product mixture with 6 M HCl produced a mixture of oosporein **8** and dibenzofuran diquinone **9** in approximately 60% yield, suggesting that reaction of **6** and **7** with pyrrolidine and copper acetate had indeed led to addition of pyrrolidine to the unsubstituted carbons of these compounds, and perhaps by analogy to the literature precedent,[6] yielded the tetrapyrrolidino compound **11**. Attempts to separate **8** from **9** by recrystallization were not successful, and chromatography was not attempted. Treatment with activated charcoal was only partially successful at removing **9** from a mixture of **8** and **9**.

As the conversion of **6** and **7** into **8** and **9** occurred under oxidative conditions, as did the conversion of **4** and **5** into **6** and **7**, we investigated the direct conversion of **4** and **5** into **8** and **9**. We were pleased to find that treatment of **4** and **5** with copper(II) acetate and an excess of pyrrolidine yielded a fine precipitate as observed previously, which on hydrolysis yielded a mixture of **8** and **9**. This modification thus removed the need to separately prepare and isolate phoenicin en route to oosporein (though we assume phoenicin was an intermediate in this process).

It was not known if both **4** and **5** were precursors to **9**, or just **5**, though it was noted that the ratio of **8**:**9** (as determined by NMR) closely matched the ratio of **4**:**5**, which, not surprisingly, also correlated with the ratio of **2**:**3**. Acting on the hypothesis that cyclization of **7** (or other intermediates derived from **5**) would be faster than cyclization of **6** (owing to the lesser steric hindrance at the unsubstituted C-2 of **7**), we sought to obtain a sample of **2** uncontaminated by **3** in order to determine if this would lead to pure oosporein.

Purification of **2** by recrystallization from methanol gave either a 30% yield of pure **2** or a 50% yield of a 9:1 mixture of **2**:**3**, depending on the volume of methanol used. A more convenient purification method consisted of suspending the mixture of **2** and **3** in boiling methanol, filtering the mixture while still hot, and briefly rinsing the precipitate with either ice cold or room temperature methanol. Pure product could be obtained by this method, in approximately 50% yield.[18] Methanolic hydrolysis of pure **2** (as

previously described) provided a quantitative yield of **4** uncontaminated by **5**. This product could then be treated with iron chloride to give phoenicin **6** in 76% yield uncontaminated by **7**, which then provided pure oosporein **8** in 59% yield upon treatment with pyrrolidine and copper(II) acetate, followed by hydrolysis. Similarly, treatment of pure **4** directly with pyrrolidine and copper(II) acetate (once again followed by hydrolysis), provided **8** in 52% yield, uncontaminated by **9**. These results suggested that the source of dibenzofuran diquinone **9** was isophoenicin **7**, and that pure oosporein **8** could indeed be prepared simply by starting from pure biaryl **4**, which in turn could be prepared from pure hexaacetate **2**. Thus, a synthesis of oosporein was achieved in four steps and 24% overall yield starting from 2,5-dimethoxytoluene. Identity of the product was confirmed by preparation of the tetraacetate via treatment with acetic anhydride and sulfuric acid. Interestingly, the ^{13}C NMR spectrum of **8** obtained at room temperature in DMSO-d_6 showed only two sharp peaks above 100 ppm (attributable to C-1 and C-4) along with a broad peak centered around 169 ppm, presumably caused by slow tautomerization between the carbonyl- and hydroxyl-bearing carbons. Addition of excess triethylamine resolved this broad peak into two sharp peaks at 173.3 and 171.9 ppm, presumably through formation of the corresponding anionic species.

Having found success in removing one step from the synthesis of oosporein, we considered further shortening the synthetic sequence. As aryl acetates can be hydrolyzed under base-promoted conditions, treatment of hexaacetate **2** with pyrrolidine and copper acetate (followed by acidic hydrolysis) was investigated. While this gave a 51% yield of product (yield essentially identical to that obtained in the conversion of **4** into **8**) the product was a mixture consisting of approximately 85% **8** and 15% cyclized product **9**. This indicated that even pure hexaacetate **2** (or intermediates derived from it) could lead to **9** under some conditions. Variation in reaction conditions failed to improve the purity of **8** while still maintaining comparable yields.[19]

While the synthesis described above was proceeding, the reaction of pyrrolidine and copper acetate with ditoluquinone **1** was also investigated in an attempt to achieve an even shorter synthesis of oosporein **8**. Treatment of **1** with copper(II) acetate and an excess of pyrrolidine once again produced a fine green-black precipitate which was difficult to isolate due to its tendency to clog filter paper or fritted glass during filtration, though hydrolysis of this material gave a 69% yield of benzofuran diquinone **9**.[20] All attempts to hydrolyze **9** to convert it into **8** were unsuccessful.

Since a large loss of usable product occurred in the Thiele-Winter transformation of **1** into **2**, variations in the reaction protocol were investigated in an attempt to improve the ratio of **2**:**3** in the product mixture. Use of both triflic acid[21] and boron trifluoride etherate[22] as substitutes for concentrated sulfuric acid was investigated, but the product ratio remained constant at approximately 1.2:1. (The lack of acid catalyst influence on product distributions has been noted previously).[14]

In summary, a short, efficient synthesis of oosporein **8** has been developed, which provides this compound in 24% overall yield in four steps from 2,5-dimethoxytoluene. No chromatography was required during any step of the synthesis.

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Supplementary data

Experimental procedures, characterization data and NMR spectra for compounds **2**, **4**, **6** and **8**. Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.tetlet.2009.06.103.

References and notes

1. (a) Vining, L. C.; Kelleher, W. J.; Schwarting, A. E. *Can. J. Microbiol.* **1962**, *8*, 931; (b) Takeshita, H.; Anchel, M. *Science* **1965**, *147*, 152; (c) Strasser, H.; Vey, A.; Butt, T. M. *Biocontrol Sci. Technol.* **2000**, *10*, 717; (d) Nagaoka, T.; Nakata, K.; Kouno, K.; Ando, T. *Z. Naturforsch.* **2004**, *59c*, 302.
2. Brewer, D.; Jen, W. C.; Jones, G. A.; Taylor, A. *Can. J. Microbiol.* **1984**, *30*, 1068.
3. Terry, B. J.; Liu, W.-C.; Cianci, C. W.; Proszynski, E.; Fernandes, P.; Meyers, E. *J. Antibiot.* **1992**, *2*, 286.
4. Seger, C.; Erlebach, D.; Stuppner, H.; Griesser, U. J.; Strasser, H. *Helv. Chim. Acta* **2005**, *88*, 802.
5. (a) Seger, C.; Laengle, T.; Pernfuss, B.; Stuppner, H.; Strasser, H. *J. Chromatogr., A* **2005**, *1092*, 254; (b) Seger, C.; Sturm, S.; Laengle, T.; Wimmer, W.; Stuppner, H.; Strasser, H. *J. Agric. Food Chem.* **2005**, *53*, 1364; (c) Michelitsch, A.; Rueckert, U.; Rittmannsberger, A.; Seger, C.; Strasser, H.; Likussar, W. *J. Agric. Food Chem.* **2004**, *52*, 1423; (d) Strasser, H.; Abendstein, D.; Stuppner, H.; Butt, T. M. *Mycol. Res.* **2000**, *104*, 1227.
6. Kögl, F.; van Wessem, G. C. *Recl. Trav. Chim. Pays-Bas* **1944**, *63*, 5.
7. Posternak, T. *Helv. Chim. Acta* **1938**, *21*, 1326.
8. Dallacker, F.; Löhnert, G. *Chem. Ber.* **1972**, *105*, 614.
9. Kalamar, J.; Steiner, E.; Charollais, E.; Posternak, T. *Helv. Chim. Acta* **1974**, *57*, 2368.
10. A SciFinder Scholar® search failed to locate any newer syntheses of oosporein than the one described by Posternak in Ref. 9.
11. Love, B. E.; Bonner-Stewart, J.; Forrest, L. A. *Synlett* **2009**, 813.
12. Though commercially available, 2,5-dimethoxytoluene is moderately expensive. If cost is a concern, however, it can readily be prepared in 97% yield from inexpensive methylhydroquinone.[13]
13. Sudhir, U.; James, B.; Joly, S.; Nair, M. S. *Res. Chem. Intermed.* **2003**, *29*, 523.
14. McOmie, J. F. W.; Blatchly, J. M. *Org. React.* **1972**, *19*, 199.
15. More rapid isolation of the products (in approximately 80% yield) could be accomplished by removal of most of the methanol under reduced pressure, followed by dilution with water and isolation of the products by suction filtration. Failure to remove most of the methanol prior to dilution with water greatly diminished the yield of the product obtained.
16. (a) Baltzly, R.; Lorz, E. *J. Am. Chem. Soc.* **1948**, *70*, 861; (b) Luly, J. R.; Rapoport, H. *J. Org. Chem.* **1981**, *46*, 2745.
17. Mixtures of quinones and tertiary amines have been observed to form stable radical anion/radical cation pairs. See, for example: Dworniczak, M. *React. Kinet. Catal. Lett.* **2003**, *78*, 65.
18. The ratio of solvent volume to mixture weight (10 mL of methanol per gram of mixture) appeared to be more critical than the number of washings. For identical one gram samples, one trituration with 10 mL of methanol produced **2** in 45% yield, two triturations with 5 mL each of methanol produced **2** in 49% yield, and three triturations with 3 mL each of methanol produced **2** in 51% yield, all uncontaminated by **3**.
19. As yields for the conversion of **2** into **4** are quantitative, this modification, had it been successful, would have only resulted in a saving of time, not an improvement of yield.
20. While 6 equiv of pyrrolidine (relative to diquinone) were sufficient for the conversion of **4** into **8**, the conversion of **1** into **9** required 12 equiv of pyrrolidine. Use of lesser amounts of pyrrolidine led to contamination of **9** with an unidentified impurity.
21. Villemin, D.; Bar, N.; Hammadi, M. *Tetrahedron Lett.* **1997**, *38*, 4777.
22. (a) Fieser, L. *J. Am. Chem. Soc.* **1948**, *70*, 3165; (b) Blatchly, J. M.; Green, R. J. S.; McOmie, J. F. W.; Searle, J. B. *J. Chem. Soc. (C)* **1969**, 1353.