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A concise synthesis of maleic anhydride and maleimide natural products found in *Antrodia camphorata*

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Abstract—Recently, the natural products maleic anhydride **1** and maleimides **2** and **3** have been isolated from the mycelium of *Antrodia Camphorata*, each displaying activity in LLC cell lines. We describe a concise synthesis of each of these natural compounds utilizing C–C cross-coupling methodology, namely, the Negishi and Suzuki reactions. This efficient and high yielding synthesis is convergent lending itself to production of medicinal analogues.

Antrodia camphorata is a parasitic fungus which grows on the inner heartwood of the native and endangered Taiwanese tree *Cinnamomun kanehirai*. This highly sought after fungus is used in traditional Chinese tribal medicine for the treatment of hypertension, liver cancer, drug intoxication and inflammation and can retail at ca. AUD 19,000 per kg.[1–6] Previous studies indicate that the *A. camphorata* mycelium extract consists of many compounds such as steroids, triterpenoids, sesquiterpene lactones, diterpenes and polysaccharides[5] some of which are in the early stages of investigation for activity against the growth of tumour cell lines.[1–5,7] Recently, a new class of compounds (Fig. 1), bearing either maleic anhydride **1** or maleimide **2** and **3** carbocyclic cores, has also been isolated and these compounds were found to be active against Lewis lung carcinoma (LLC) cell lines.[1] However, as yet, there is no additional biological evaluation and little is known about their mode of action.

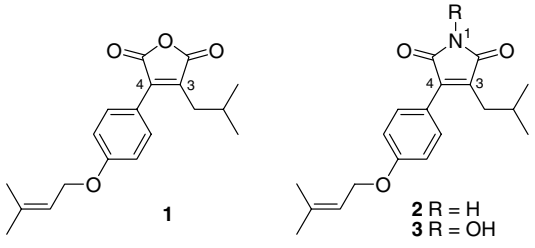

Figure 1. Maleic anhydride and maleimide natural products of *Antrodia camphorata*.

Natural product maleic anhydrides and maleimides have attracted the interest of several groups exploring their biological activity.[8,9] In conjunction with these studies, there have been various synthetic approaches targeting compounds bearing various R groups at positions C3 and C4.[10–12] More recently, the above natural products **1**, **2** and **3** have been prepared by the group of Argade[13] through a seven-step linear based approach. Alternatively, we envisaged a more convergent connection of the alkyl and aryl fragments through Negishi[14] and Suzuki[15] cross-coupling protocols. In this manner, it was intended that analogues could be easily prepared by simply altering the respective coupling partners.

The synthesis of natural product **1** began with 3,4-dichloromaleic anhydride (**4**, Aldrich) (Scheme 1).[16] As cross-coupling reactions involving 3,4-halogenated maleic anhydrides have proven to be difficult in this laboratory a protected maleimide was utilised. Thus, condensation of maleic anhydride **4** using benzylamine in acetic acid afforded the benzyl protected maleimide **5** in 81% yield. Negishi cross-coupling of compound **5** with isobutylzinc bromide, $Pd_2(dba)_3$ (10 mol %) and PPh_3 (20 mol %) in THF at room temperature (optimum conditions), furnished 60% of only mono alkylated maleimide **6**.[16] Bearing in mind that this transformation

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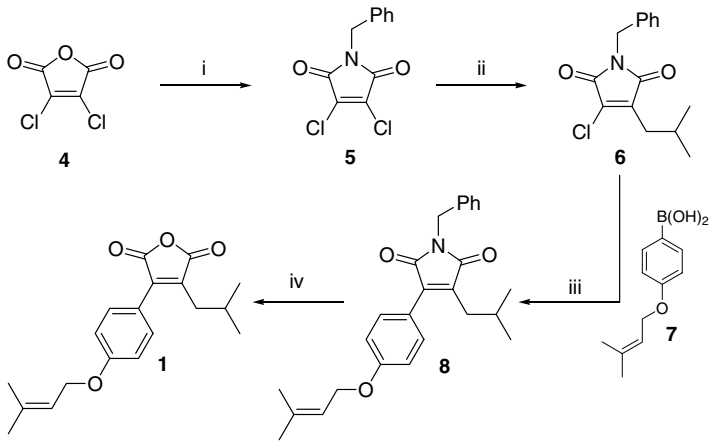
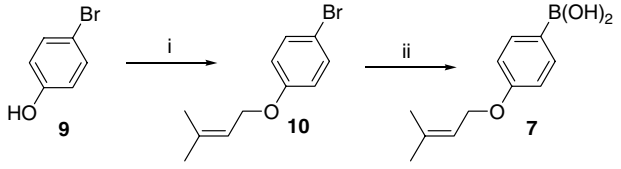


Scheme 1. Reagents and conditions: (i) $BnNH_2$, AcOH, 50 °C, 16 h, 81%; (ii) $Pd_2(dba)_3$ (10 mol %), PPh_3 (20 mol %), ^tBuZnBr, THF, 20 °C, 16 h, 60%; (iii) **7**, $Pd_2(dba)_3$ (10 mol %), $HP(t\text{-}Bu)_3BF_4$ (20 mol %), Cy_2NMe, dioxane, 20 °C, 3 h, 76%; (iv) KOH, THF/MeOH (1:2), 78 °C, 12 h then HCl (2 M), 20 °C, 63%.

also falls into the category of palladium mediated conjugate substitution reactions,[17] we decided to try a wide range of catalysts in order to improve this yield. Unfortunately, using other palladium catalysts, that is, $PdCl_2(dppf)$ (11% yield) or even nickel based catalysts, that is, $Ni(PPh_3)_2Cl_2$ (24% yield), were either lower yielding or failed to produce any of the desired product.

With chloromaleimide **6** in hand, we examined the Suzuki cross-coupling reaction in order to attach the aryl ring fragment. Boronic acid **7**, required for this transformation, was prepared (Scheme 2) by initially treating the commercially available 4-bromophenol **9** with K_2CO_3 and 3,3-dimethylallyl bromide to provide ether **10** in 77% yield. In a modified procedure to that reported by Sellès,[11] halogen metal exchange of **10** using *n*-BuLi followed by treatment with trimethyl borate afforded the respective boronic ester. This latter compound was hydrolysed (HCl 2 M) to provide boronic acid **7** (45% from **9**). Suzuki cross-coupling of chloromaleimide **6** with boronic acid **7** using $Pd_2(dba)_3$ and in situ generated $P(t\text{-}Bu)_3$ afforded the highly fluorescent aryl maleimide **8** in 76% yield.[18–20] Treatment of compound **8** with KOH in THF/MeOH at reflux followed by acidic work-up afforded the desired natural product maleic anhydride **1** in 63% yield. The spectral data for compound **1** matched those reported previously.[1,13]

In order to compare the relative reactivity of the chloro-substituted maleimides with their brominated counterparts, a second pathway (Scheme 3) using bromomaleic anhydride **11** was devised. Under the same conditions



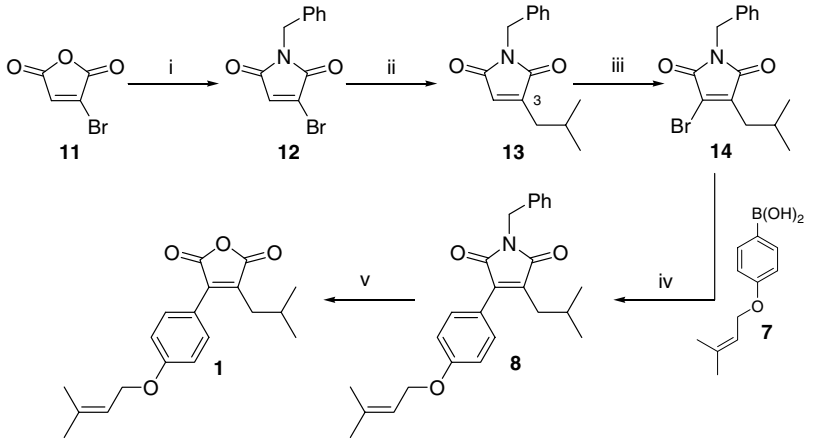
Scheme 2. Reagents and conditions: (i) K_2CO_3, 3,3-dimethylallyl bromide, acetone, 56 °C, 16 h, 77%; (ii) (a) *n*-BuLi, THF, −78 °C, 20 min then $B(OMe)_3$, 1.5 h, −78→20 °C; (b) NH_4Cl (aq), 20 °C, 0.5 h, 59%.

as described previously commercially available bromomaleic anhydride **11** was converted to the *N*-benzyl derivative **12** in 83% yield. The corresponding metal mediated conjugate substitution reaction using the Negishi protocol provided alkyl maleimide **13** in 49% yield. Surprisingly, under various catalytic conditions this conversion was consistently lower than when using the chlorinated derivative **5**. Bromination of **13** using bromine and a catalytic amount of aluminium tribromide afforded vinyl bromide **14** in reasonable yield.[8] Attachment of the aryl side chain was slightly lower yielding (56%) when treating the brominated product **14** under the same Suzuki cross-coupling conditions as described previously to give compound **8**. Overall the described alterations illustrated in Scheme 3 compared to the earlier pathway (Scheme 1) were disadvantaged because of the additional bromination step and lower yields associated with the initial attachment of the alkyl side chain at position C3.

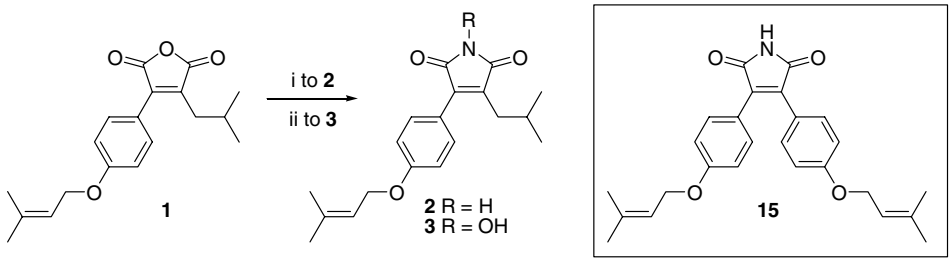
The synthesis of the natural product maleimide **2** (Scheme 4) was complete on treatment of anhydride **1** with urea to afford **2** in 60% yield. Similarly, the treatment of **1** with *N*-hydroxylamine hydrochloride gave **3** in 79% yield.[12,21] The spectral data corresponding to both maleimides were consistent with those reported in the literature.[1,13]

The aforementioned synthetic sequence is currently being used as a template for the synthesis of a series of derivatives for biological evaluation as liver cancer agents. These derivatives which contain various attachments to both the maleimide and maleic anhydride core are being provided by simply changing the transmetalation partners in both the Negishi and Suzuki cross-coupling reactions. Interestingly, if the Suzuki reaction is carried out on compound **5** a series of bis-arylated compounds such as **15** are obtained and none of the mono-substituted product.[8] Compound **15** is one of a library of compounds currently being screened within our laboratories.

In summary, we have developed a novel and concise synthesis of three natural products found in *A. campho-*



Scheme 3. Reagents and conditions: (i) $BnNH_2$, AcOH, 50 °C, 16 h, 83%; (ii) $Ni(PPh_3)_2Cl_2$ (5 mol %), ^iBuZnBr, THF, 20 °C, 4 h, 49%; (iii) Br_2, $AlBr_3$, CH_2Cl_2, 1 h, 0→20 °C, 55%; (iv) **7**, $Pd_2(dba)_3$ (10 mol %), $HP(t\text{-}Bu)_3BF_4$ (20 mol %), Cy_2NMe, dioxane, 20 °C, 3 h, 56%; (v) KOH, THF/MeOH (1:2), 78 °C, 12 h then HCl (2 M), 20 °C, 63%.



Scheme 4. Reagents and conditions: (i) Urea, 140 °C, 4 h, 60%; (ii) $NH_2OH{\cdot}HCl$, pyridine, 100 °C, 12 h, 79%.

rata. Compound **1** was synthesised in four steps in 23% overall yield while the maleimide natural products **2** and **3** were prepared in five steps and 14% and 18% overall yields, respectively. We expect this synthesis utilising Negishi and Suzuki reaction protocol to be highly amenable to the production of a range of similar analogues bearing this basic structure. Currently work is underway to produce such compounds and develop a structure–activity relationship (SAR) study for further biological evaluation.

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References and notes

1. Nakamura, N.; Hirakawa, A.; Gao, J.-J.; Kakuda, H.; Shiro, M.; Komatsu, Y.; Sheu, C.; Hattori, M. *J. Nat. Prod.* **2004**, *67*, 46.
2. Hsu, Y.-L.; Kuo, Y.-C.; Kuo, P.-L.; Ng, L.-T.; Kuo, Y.-H.; Lin, C.-C. *Cancer Lett.* **2005**, *221*, 77.
3. Hattori, M.; Sheu, C.-C. US Patent & Trademark Office, 2005, US, 7109232, CAN 143:284829.
4. (a) Liu, J. J.; Huang, T. S.; Hsu, M.-L.; Chen, C. C.; Lin, W. S.; Lu, F. J.; Chang, W.-H. *Toxicol. Appl. Pharm.* **2004**, *201*, 186; (b) Shu, C.-H.; Lung, M.-Y.; Xu, C.-J. *J. Chem. Technol. Biotechnol.* **2005**, *80*, 216.
5. Chen, C.-C.; Shiao, Y.-J.; Lin, R.-D.; Shao, Y.-Y.; Lai, M.-N.; Lin, C.-C.; Ng, L.-T.; Kuo, Y.-H. *J. Nat. Prod.* **2006**, *69*, 689, and references cited therein.
6. Hseu, Y.-C.; Wu, F.-Y.; Wu, J.-J.; Chen, J.-Y.; Chang, W.-H.; Lu, F.-J.; Lai, Y.-C.; Yang, H.-L. *Int. Immunopharmacol.* **2005**, *5*, 1914.
7. Yang, H.-L.; Chen, C.-S.; Chang, W.-H.; Lu, F.-J.; Lai, Y.-C.; Chen, C.-C.; Hseu, T.-H.; Kuo, C.-T.; Hseu, Y.-C. *Cancer Lett.* **2006**, *231*, 215.
8. Dubernet, M.; Caubert, V.; Guillard, J.; Viaud-Massuard, M.-C. *Tetrahedron* **2005**, *61*, 4585.
9. Wang, J.; Soundarajan, N.; Liu, N.; Zimmermann, K.; Naidu, B. N. *Tetrahedron Lett.* **2005**, *46*, 907.
10. Easwar, S.; Argade, N. P. *Synthesis* **2006**, 831.
11. Sellès, P. *Org. Lett.* **2005**, *7*, 605.
12. Trost, B. M.; Tang, W. *Org. Lett.* **2001**, *3*, 3409.
13. Baag, M. M.; Argade, N. P. *Synthesis* **2006**, 1005.
14. Negishi, E.-I.; Hu, Q.; Huang, Z.; Qian, M.; Wang, G. *Aldrichim. Acta* **2005**, *38*, 71.
15. Miyaura, N.; Suzuki, A. *Chem. Rev.* **1995**, *95*, 2457.
16. *Preparation of 1-benzyl-3-chloro-4-isobutylpyrrole-2,5-dione* (**6**): Triphenylphosphine (100 mg, 0.38 mmol) was added in one portion to a magnetically stirred solution of $Pd_2(dba)_3$ (178 mg, 0.19 mmol) and the solution degassed and stirred at rt for 0.5 h. The ensuing dark brown mixture was treated with isobutylzinc bromide (3.9 mL, 0.5 M in THF) followed by 3,4-dichloromaleimide **5** (500 mg,

1.95 mmol) in one portion. The resulting mixture was stirred at ambient temperature for 16 h before being treated with silica (ca. 300 mg) and the suspension concentrated under reduced pressure. Subjection of this material to chromatography (EtOAc/hexane 1:19) afforded dione **6** (326 mg, 60%) as a colourless oil. $R_f = 0.3$ (EtOAc/hexane, 1:9); ν_{max} (NaCl)/cm^{-1} 2959, 1716, 1434, 1399, 1351, 1100, 1069, 741; ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.96 (d, $J = 7.3$ Hz, 6H), 2.10 (sep, $J = 7.0$ Hz, 1H), 2.36 (d, $J = 7.7$ Hz, 2H), 4.69 (s, 2H), 7.26–7.36 (m, 5H); ^{13}C NMR (75 MHz; $CDCl_3$): δ 22.6, 27.9, 32.7, 42.0, 127.9, 128.4, 128.6, 134.1, 135.9, 140.6, 164.8, 168.7.

17. *Handbook of Organopalladium Chemistry for Organic Synthesis*; Negishi, E., Ed.; Wiley: Interscience, New York, 2002; Vol. 1, Part III.2.15.
18. Netherton, M. R.; Fu, G. C. *Org. Lett.* **2001**, *3*, 4295.
19. Tietze, L. F.; Stewart, S. G.; Polomska, M. E. *Eur. J. Org. Chem.* **2005**, 1752.
20. *Preparation of 1-benzyl-3-isobutyl-4-[4-(3-methylbutenyloxy)phenyl]pyrrole-2,5-dione* (**8**): *N*,*N*-Dicyclohexylmethylamine (174 μL, 0.80 mmol) was added to a degassed and magnetically stirred solution of $Pd_2(dba)_3$ (36 mg, 40.0 μmol) and tri-*tert*-butylphosphonium tetrafluoroborate (24 mg, 82.0 μmol) in dioxane (400 μL) maintained at 20 °C under an atmosphere of argon. The resulting mixture was stirred for 1 h, before being treated dropwise with chloromaleimide **6** (100 mg, 0.36 mmol) in dioxane (200 μL) followed by boronic acid **7** (84 mg, 0.40 mmol). Stirring was continued for 14 h at 22 °C, before silica gel (~150 mg) was added and the suspension concentrated under reduced pressure. Subjection of this material to flash chromatography (EtOAc/hexane 1:19) afforded the desired product **8** (110 mg, 76%) as a fluorescent yellow oil. $R_f = 0.6$ (hexane/EtOAc, 9:1); ν_{max} (NaCl)/cm^{-1} 2959, 1703, 1605, 1511, 1434, 1402, 1246, 1179, 996; ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.89 (d, $J = 6.7$ Hz, 6H), 1.76 (s, 3H), 1.81 (s, 3H), 2.06 (sep, $J = 7.0$ Hz, 1H), 2.51 (d, $J = 7.2$ Hz, 2H), 4.55 (d, $J = 6.6$ Hz, 2H), 4.72 (s, 2H), 5.54 (brt, $J = 6.6$ Hz, 1H), 6.98 (d, $J = 8.5$ Hz, 2H), 7.23–7.41 (m, 5H), 7.52 (d, $J = 8.5$ Hz, 2H) ^{13}C NMR (75 MHz; $CDCl_3$): δ 18.1, 22.7, 25.8, 28.0, 32.8, 41.6, 64.7, 114.6, 119.1, 121.3, 127.1, 127.6, 128.4, 128.5, 130.8, 136.6, 138.0, 138.6, 159.8, 171.0, 171.8; MS (EI, 70 eV): m/z (%) = 403.5 ($M^{+\cdot}$, 10), 335 (100), 293 (25), 215 (33), 131 (58).
21. Shah, K. R.; Blanton, C. D. *J. Org. Chem.* **1982**, *47*, 502.