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Total Synthesis of Depsilairdin

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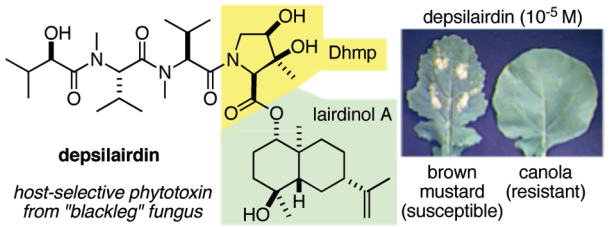

The total synthesis of depsilairdin, a host-selective phytotoxin isolated from *Leptosphaeria maculans* (the causal agent of blackleg disease of oilseed Brassicas), has been achieved by *N*-terminal extension of a suitably protected derivative of the hitherto unknown amino acid (2*S*,3*S*,4*R*)-3,4-dihydroxy-3-methyl-proline (Dhmp) followed by esterification with lairdinol A. The latter esterification, complicated by the sterically hindered nature of the carboxyl group, was accomplished by a novel method involving reaction of the 1-hydroxybenzotriazole (HOBt) derived active ester with the bromomagnesium alkoxide of lairdinol A. Three depsilairdin analogues were also prepared by replacing the Dhmp residue with L-proline and *cis*- and *trans*-4-hydroxy-L-proline. Phytotoxicity assays showed that the analogues were nontoxic to both blackleg-susceptible (brown mustard) and -resistant (canola) plants, suggesting that the presence of the Dhmp residue in depsilairdin is important for its host-selective toxicity toward brown mustard.

Introduction

Many microbial pathogens produce host-selective toxins (HSTs) to facilitate infection of plants.[1] By definition, these compounds show selective toxicity to host plants and are relatively harmless to nonhosts. HSTs are valuable probes to study the complex metabolic pathways that govern the interaction of plants with their microbial pathogens.[1] Recently, Pedras et al. reported[2] that depsilairdin (**1**) (Figure 1) is a highly selective toxin produced by the "blackleg" fungus (*Leptosphaeria maculans*; asexual stage *Phoma Lingam*), one of the most devastating pathogens of the oilseed crops rapeseed/canola (*Brassica napus*, *B. rapa*).[3] Recently, strong evidence has been accumulating that this pathogen is expanding its host range to include mustard (*B. juncea*), a crop traditionally resistant to blackleg disease.[4] In particular, fungal isolates that produce despilairdin (**1**) were found to infect mustard. Plant leaves of mustard treated with depsilairdin (**1**) showed strong necrotic and chlorotic lesions similar to those caused by the pathogen, but such symptoms were not observed on canola or rapeseed leaves.[2] Thus, due to its very selective phytotoxicity, depsilairdin (**1**) is a most

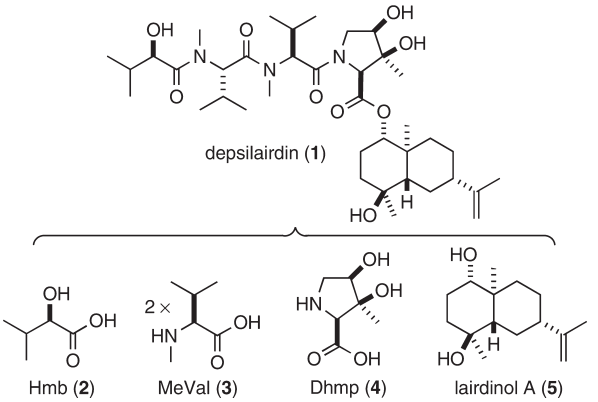

FIGURE 1. Structures of depsilairdin (**1**) and its component residues.

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(2) Pedras, M. S. C.; Chumala, P. B.; Quail, J. W. *Org. Lett.* **2004**, *6*, 4615–4617.
(3) (a) Howlett, B. J.; Idnurm, A.; Pedras, M. S. C. *Fungal Genet. Biol.* **2001**, *33*, 1–14. (b) Fitt, B. D. L.; Brun, H.; Barbetti, M. J.; Rimmer, S. R. *Eur. J. Plant Pathol.* **2006**, *114*, 3–15.
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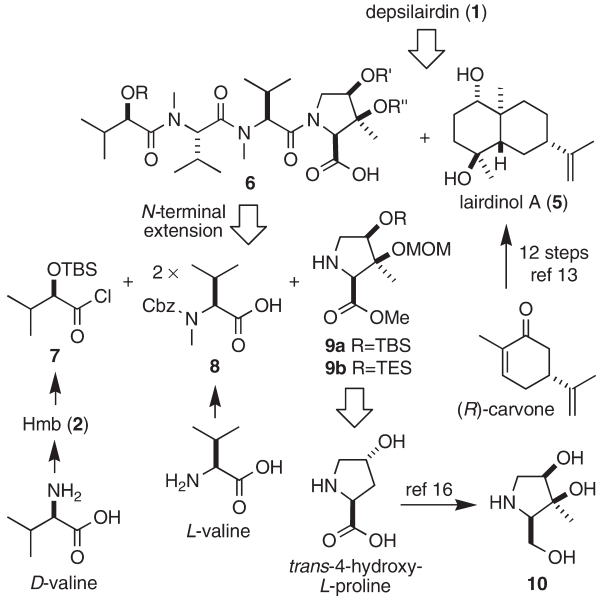
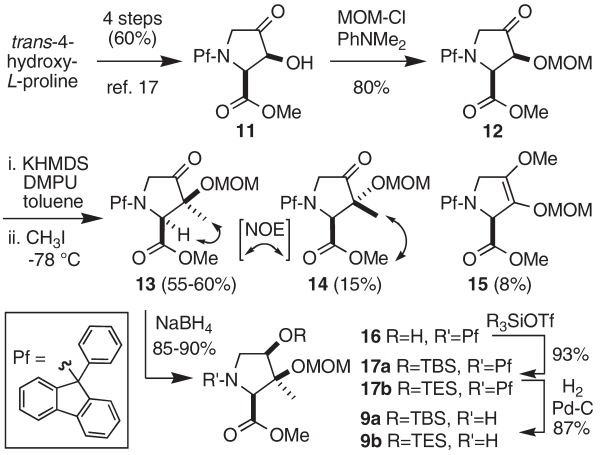
SCHEME 1. Retrosynthetic Analysis of Depsilairdin (1)



SCHEME 2. Synthesis of the Protected Dhmp Residue 9



desirable compound to probe the susceptibility and resistance of mustard and canola, respectively, to the blackleg pathogen. To date these studies have been impeded because insufficient quantities of **1** are available from fungal cultures. Herein, we report the total syntheses and relative phytotoxicities of depsilairdin (**1**) and three analogues where the Dhmp residue is replaced by proline and (4*R*)- and (4*S*)-4-hydroxyproline.

Depsilairdin (**1**) is a structurally interesting depsipeptide composed of five residues: (2*R*)-2-hydroxy-3-methylbutanoic acid (Hmb, **2**), two *N*-methyl-L-valines (MeVal, **3**), (2*S*,3*S*,4*R*)-3,4-dihydroxy-3-methyl-proline (Dhmp, **4**), and the sesquiterpene lairdinol A (**5**) (Figure 1). Dhmp (**4**) is a novel amino acid that has not been observed previously. Similarly, lairdinol A (**5**) is a novel sesquiterpene that was isolated[5] from the same fungal cultures as **1**; *ent*-**5** (cyperusol C) has been isolated from the plants *Cyperous longus*[6] and *Erigeron annus*.[7] Although there are several peptide natural products that contain a MeVal-MeVal sequence, the MeVal-MeVal-Pro sequence is rare[8] and the Hmb-MeVal-MeVal sequence is unknown. Finally, (depsi)peptide-sesquiterpene conjugates are highly unusual among natural products.

Syntheses of linear (depsi)peptides typically proceed by linear or convergent coupling of suitably protected intact residues followed by deprotection.[9] Our retrosynthetic analysis of depsilairdin (**1**) is outlined in Scheme 1. We chose to disconnect the ester linkage because C-terminal proline residues in oligopeptide chains are resistant to isomerization during chain extension and chemoselective acylation of the secondary alcohol in lairdinol A (**5**) is expected. The tetradepsipeptide **6** would be assembled by standard N-terminal extension of a suitably protected Dhmp residue (e.g., **9**).[9] Although incorporation of *N*-methylamino acids is difficult using standard peptide coupling methods,[10a] several specialized reagents are available for this purpose.[10] Of the required four residues, **7** and **8** are readily available from D- and L-valine, respectively,[11,12] and we have recently reported[13] the synthesis of lairdinol A (**5**) in 12 steps from (*R*)-carvone without the use of protecting groups. As noted above, Dhmp (**4**) is a hitherto unknown amino acid. In contemplating possible strategies to prepare a suitably protected derivative of **4**, we were cognizant of the numerous syntheses of 3,4-dihydroxyproline[14] and 3-hydroxy-3-methylproline[15] derivatives that have appeared in the literature. Moreover, Blanco and Sardina disclosed a seemingly straightforward synthesis of the closely related triol **10** from *trans*-4-hydroxy-L-proline,[16] and easy access to **9** seemed feasible by adaptation of this route.

Results and Discussion

The synthesis commenced with the preparation of keto-alcohol **11** that proceeded without incident using the reported protocols (Scheme 2).[17] Surprisingly, attempted protection of

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(6) Xu, F.; Morikawa, T.; Matsuda, H.; Ninomiya, K.; Yoshikawa, M. *J. Nat. Prod.* **2004**, *67*, 569–576.
(7) Li, X.; Yang, M.; Han, Y.-F.; Gao, K. *Planta Med.* **2005**, *71*, 268–272.
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(11) (a) Deechongkit, S.; You, S.-L.; Kelly, J. W. *Org. Lett.* **2004**, *6*, 497–500. (b) Gonzalez, I.; Jou, G.; Caba, J. M.; Albericio, F.; Lloyd-Williams, P.; Giralt, E. *J. Chem. Soc., Perkin Trans. 1* **1996**, 1427–1433. (c) Wissner, A.; Grudzinskas, C. V. *J. Org. Chem.* **1978**, *43*, 3972–3974. (d) Ward, D. E.; Vazquez, A.; Pedras, M. S. C. *J. Org. Chem.* **1999**, *64*, 1657–1666.
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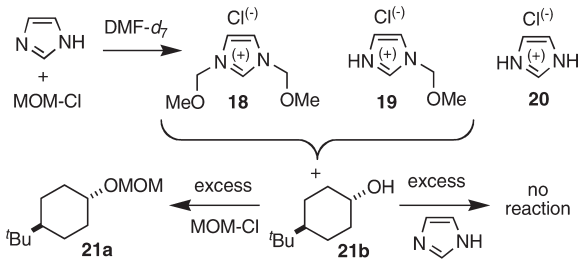
(14) Selected recent examples: (a) Taylor, C. M.; Jones, C. E.; Bopp, K. *Tetrahedron* **2005**, *61*, 9611–9617. (b) Soengas, R. G.; Estevez, J. C.; Estevez, R. J. *Tetrahedron: Asymmetry* **2003**, *14*, 3955–3963. (c) Garcia, A. L. L.; Correia, C. R. D. *Tetrahedron Lett.* **2003**, *44*, 1553–1557. (d) Taylor, C. M.; Barker, W. D.; Weir, C. A.; Park, J. H. *J. Org. Chem.* **2002**, *67*, 4466–4474. (e) Fujii, M.; Miura, T.; Kajimoto, T.; Ida, Y. *Synlett* **2000**, 1046–1048.
(15) Selected recent examples: (a) Yoshitomi, Y.; Makino, K.; Hamada, Y. *Org. Lett.* **2007**, *9*, 2457–2460. (b) Chen, Z.; Ye, T. *Synlett* **2005**, 2781–2785. (c) Makino, K.; Nagata, E.; Hamada, Y. *Tetrahedron Lett.* **2005**, *46*, 8159–8162. (d) Davis, F. A.; Ramachandar, T.; Liu, H. *Org. Lett.* **2004**, *6*, 3393–3395. (e) Shen, J.-W.; Qin, D.-G.; Zhang, H.-W.; Yao, Z.-J. *J. Org. Chem.* **2003**, *68*, 7479–7484. (f) Qin, D.-G.; Zha, H.-Y.; Yao, Z.-J. *J. Org. Chem.* **2002**, *67*, 1038–1040.
(16) Blanco, M.-J.; Sardina, F. J. *J. Org. Chem.* **1998**, *63*, 3411–3416.
(17) Blanco, M.-J.; Sardina, F. J. *J. Org. Chem.* **1996**, *61*, 4748–4755.

SCHEME 3. Reaction of MOM-Cl with Imidazole in DMF-d_7



the OH group in **11** according to the published procedure (5 equiv each of MOM-Cl and imidazole in DMF)[16] failed to give detectable amounts of the expected MOM ether **12**. Varying the amounts of imidazole (1−7 equiv; MOM-Cl, 5 equiv) clearly showed that formation of **12** occurred only with excess MOM-Cl, and apparent high conversions required ratios of MOM-Cl to imidazole of 3:1 or higher. The reaction of **11** (0.25 M in DMF) with MOM-Cl (5 equiv) and imidazole (1 equiv) was complete within 48 h. Although the ^{1}H NMR spectrum of the crude product after workup was fairly "clean" and indicated that full conversion to **12** had been achieved, the isolated yield after chromatography was only 55%. This suggested that **12** and/or **11** were converted into unknown water-soluble compounds under the reaction conditions and lost upon workup. To investigate this process further, the reaction of MOM-Cl with imidazole in DMF-d_7 was monitored by ^{1}H NMR. Within 10 min, a near equimolar mixture of MOM-Cl and imidazole (0.25 M) was converted into a ca. 2:1:2 mixture of **18**, **19**, and **20**, respectively (Scheme 3). Using 5 equiv of MOM-Cl compared to imidazole produced a similar mixture that, over time (72 h), was slowly converted to mainly **18** (> 80%). Addition of *trans*-4-*tert*-butylcyclohexanol (**21b**; 1 equiv) to this mixture produced the corresponding MOM ether **21a** (ca. 80% after 2.5 h)[18] that slowly decomposed over 24 h (ca. 40% of **21a**) by which time MOM-Cl was no longer present.[19] Alternatively, a 3.5:1 of **19** and **18**, respectively, was formed using 2 equiv of imidazole with respect to MOM-Cl. This mixture was stable over 48 h and, importantly, did not react with **21b** even after 24 h. These experiments explain our failure to obtain **12** under the reported conditions. Imidazole rapidly consumes 1 equiv of MOM-Cl and, over time, can consume 2 equiv of MOM-Cl. The resulting adducts **18** and **19** are not sufficiently reactive to alkylate an alcohol. Thus, with equimolar amounts imidazole and MOM-Cl (i.e., the reported protocol)[16] or with an excess of imidazole, alkylation of alcohols does not occur (or is exceedingly slow). Using excess MOM-Cl, alkylation is possible, but greater than 3 equiv of MOM-Cl (relative to imidazole) are required for significant conversion. However, under these conditions the reaction mixture becomes highly acidic, and this aspect can explain the moderate yields of **12** obtained (e.g., by acid-catalyzed deprotection of the amine, elimination, etc.). Attempted formation of **12** from **11** using the standard conditions (MOM-Cl, DIPEA)[20] was very sluggish (< 50% yield after 72 h) and not clean, as previously noted[16] by Blanco and Sardina. Speculating that the apparent higher reactivity of **11** toward MOM-Cl (5 equiv) under acidic conditions (1 equiv of imidazole) compared to basic conditions (7 equiv of DIPEA) might be due to protonation of the tertiary amine in **11**, we decided to employ the weaker tertiary amine base *N*,*N*-dimethylaniline (DMA)[21] ($pK_a \approx 5.1$) to allow reaction via the conjugate acid of **11**. Treatment of **11** (0.25 M in DMF) with MOM-Cl (5 equiv) and DMA (7 equiv) gave **12** (65%) along with recovered **11** (30%) after 24 h. Improved conversions were obtained using CH_2Cl_2 as solvent, and under optimized conditions, **12** was obtained in 80% isolated yield after 24 h.

Blanco and Sardina reported the methylation of ketone **12** using *n*-BuLi in presence of HMPA at −78 °C to form the enolate followed by reaction with CH_3I at −78 to 0 °C (Scheme 2).[16] The authors indicated that the resulting **13** was unstable, and consequently they treated the crude with $NaBH_4$ to obtain **16** (62%), apparently as the sole product.[16] This reaction proved very capricious in our hands, and the reported result could not be reproduced. We verified that enolate formation was essentially quantitative under the reported conditions by quenching with D_2O (i.e., > 95% deuterium incorporation at C-3). However, similar experiments established that this enolate was unstable above −50 °C and rapidly decomposed on warming to 0 °C. Despite extensive experimentation including varying the reaction time, temperature, solvent, and additives, our best result was obtained by reaction of the enolate with CH_3I at −50 °C for 4.5 h to give **13** (35%) along with the C-3 diastereomer **14**, the *O*-alkylation product **15**, and several unidentified byproducts. The formation of **14** could result from methylation of the enolate of **12** from its *si* face; alternatively, *ent*-**14** could result from epimerization of **13** at C-2. We did not establish the absolute configuration or enantiopurity of **14** or **15**. Lubell and co-workers have reported the alkylation of 4-oxoproline derivatives via the enolate generated by reaction with KHMDS in THF and 1,3-dimethyltetrahydropyrimidin-2(1*H*)-one (DMPU).[22] We optimized that procedure for the methylation of **12** and identified toluene as a superior solvent (cf. THF) and DMPU as the most effective additive (cf. HMPA and TMEDA). Thus, reaction of **12** with KHMDS (1.05 equiv) in a 1:1 mixture of toluene and DMPU at −78 °C for 1 h followed by addition CH_3I and reaction for 2 h gave the desired **13** (55−60%) along with **14** (15%) and **15** (8%). In contrast to the previous report,[16] we found that **13** was routinely isolated and not especially prone to decomposition. Reduction of **13** with $NaBH_4$ followed by conversion of the resulting alcohol **16** (dr > 10:1) to the corresponding TBS ether **17a** and hydrogenolysis of the 9-phenylfluorenyl (Pf) group delivered the desired proline fragment **9a**.

(18) Sawyer, J. S.; Kucerovy, A.; Macdonald, T. L.; McGarvey, G. J. *J. Am. Chem. Soc.* **1988**, *110*, 842–853.

(19) Major byproducts included dimethoxymethane and bis(4-*tert*-butylcyclohexyloxy)methane in addition to **18** and **21**. For acid-catalyzed exchange of alcohol groups in methylene acetals, see: Ledneczki, I.; Molnar, A. *Synth. Commun.* **2004**, *34*, 3683–3690.

(20) Wuts, P. G. M.; Greene, T. W. *Protective Groups in Organic Synthesis*, 4th ed.; Wiley: New York, 2007.

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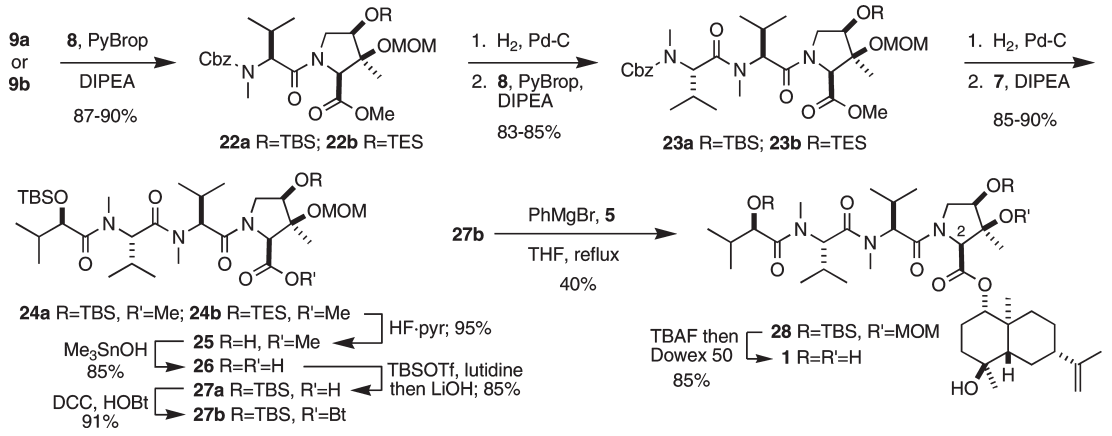
(22) Sharma, R.; Lubell, W. D. *J. Org. Chem.* **1996**, *61*, 202–209.

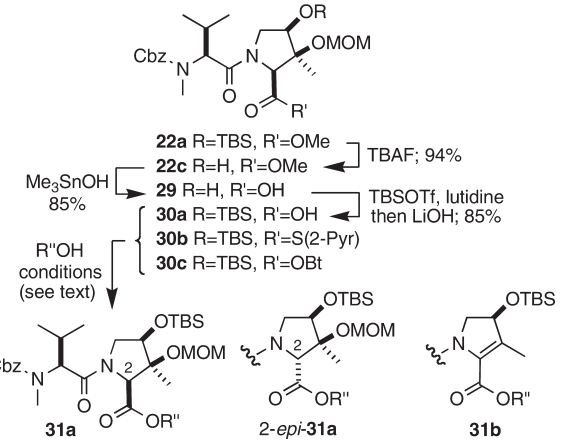
SCHEME 4. Synthesis of Depsilairdin (1) from Its Component Residues



Acylation of **9a** with **8** to give the dipeptide fragment **22a** proceeded smoothly using bromotri(pyrrolidino)phosphonium hexafluorophosphate (PyBroP), a reagent known to be effective for coupling sterically hindered amino acids (Scheme 4).[23] Removal of the Cbz protecting group in **22a** and iteration of the above coupling procedure produced the tripeptide fragment **23a**. Subjecting **23a** to hydrogenolysis followed by reaction with **7** gave the tetradepsipeptide **24a**. Unfortunately, all efforts to obtain the carboxylic acid **27** by reactions of **24a** with LiOH/H_2O/MeOH or $KOSiMe_3$/ether[24] or $Me_3SnOH/(CH_2Cl)_2$[25] were unsuccessful, presumably because of steric hindrance of the ester carbonyl group. Similar reactions of dipeptide **22a** also failed to hydrolyze the methyl ester (Scheme 5). Hypothesizing that the ester carbonyl might be activated by intramolecular hydrogen bonding, we attempted hydrolysis of the ester in **22c**. Gratifyingly, the reaction of **22c** with Me_3SnOH in $(CH_2Cl)_2$ at 80 °C gave the desired acid **22d** in good yield.[25] To facilitate application of this hydrolysis strategy to the tetradepsipeptide **24**, we prepared the differentially protected derivative **24b** by a route analogous to that used to obtain **24a** (Scheme 4).[26] The triethylsilyl group in **24b** was selectively removed by reaction with HF·pyridine to give **25** that was subjected to ester hydrolysis using Me_3SnOH to provide the acid **26** in 85% yield. Reaction of **26** with TBSOTf followed by hydrolysis of the intermediate TBS-ester gave acid **27a** in high yield.

Given the difficulties experienced in the hydrolysis of the methyl ester in **24a**, it was anticipated that esterification of the hindered acid in **27a** with lairdinol A (**5**) might prove challenging. To examine that process, we attempted esterification of the model dipeptide **30a** under a variety of conditions (Scheme 5). Using simple unhindered alcohols (e.g., (4-methoxyphenyl)-methanol), attempted esterification of **30a** using Mukaiyama's reagent (*O*,*O*-di(2-pyridyl) thionocarbonate)[27] failed, and the use of more standard reagents (e.g., DCC/DMAP, EDCI, or

SCHEME 5. Model Study for Methyl Ester Hydrolysis and Esterification



PyBrop/Et_3N) resulted in significant isomerization (i.e., **31a**:2-*epi*-**31a**, 1−3:1).[28] Similarly, esterification of the 2-pyridylthiol ester **30b** by heating with the alcohol in toluene[29] or by reaction with the Li alkoxide (ROH + BuLi) at ambient temperature also gave extensive isomerization. Alternatively, reaction of **30b** with the bromomagnesium alkoxide (ROH + CH_3MgBr) in THF produced **31a** (>80%) with minimal isomerization; however, similar reactions with hindered alcohols (e.g., borneol) were unproductive at ambient temperature and heating under reflux resulted in elimination products (**31b**).[30] Reasoning that the use of activated esters with lower acidity at the α-CH might suppress isomerization and elimination, we investigated the HOBt ester **30c**.[31] Gratifyingly, reactions of **30c** with the bromomagnesium

(23) Coste, J.; Frerot, E.; Jouin, P.; Castro, B. *Tetrahedron Lett*. **1991**, *32*, 1967–1970.

(24) (a) Laganis, E. D.; Chenard, B. L. *Tetrahedron Lett*. **1984**, *25*, 5831–5834. (b) Lovric, M.; Cepanec, I.; Litvic, M.; Bartolincic, A.; Vinkovic, V. *Croat. Chem. Acta* **2007**, *80*, 109–115.

(25) Nicolaou, K. C.; Estrada Anthony, A.; Zak, M.; Lee Sang, H.; Safina Brian, S. *Angew. Chem., Int. Ed.* **2005**, *44*, 1378–1382.

(26) Protection of the Hmb hydroxyl group was thought prudent considering the facile degradation of depsilairdin to form 3,6-diisopropyl-2,5-morpholindione (ref 2).

(27) This reagent is particularly useful for the synthesis of hindered esters. Saitoh, K.; Shiina, I.; Mukaiyama, T. *Chem. Lett*. **1998**, 679–680.

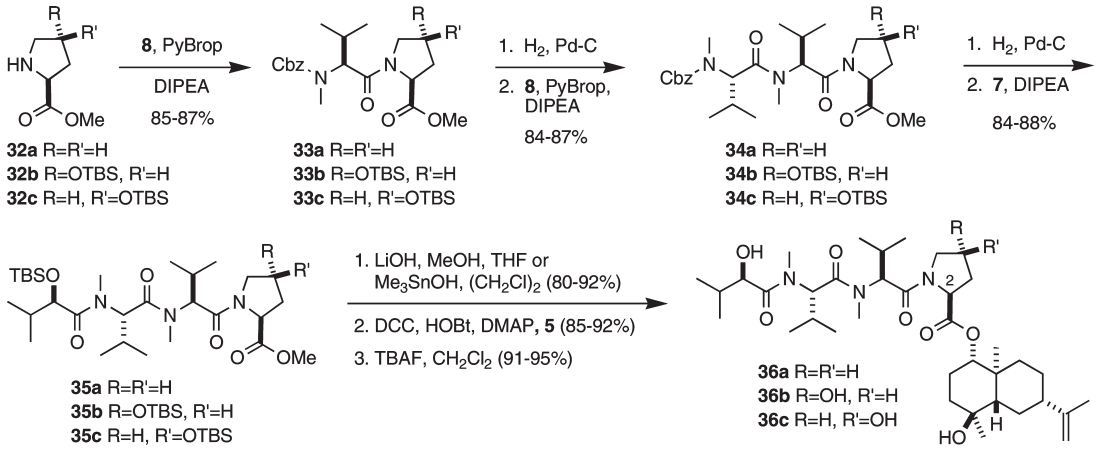
(28) The esters **31** were not fully characterized; the relative configurations at C-2 were established by NOE (e.g., irradiations of HC-2, HC-4, and H_3CC-3).

(29) Corey, E. J.; Nicolaou, K. C. *J. Am. Chem. Soc*. **1974**, *96*, 5614–5616.

(30) For reactions of 2-pyridylthiol esters with Grignard reagents to form ketones, see: (a) Mukaiyama, T.; Araki, M.; Takei, H. *J. Am. Chem. Soc*. **1973**, *95*, 4763–4765. For reactions of 2-pyridylthiol esters with lithium dialkylcuprates in the presence of oxygen to form esters, see: (b) Kim, S.; Lee, J. I.; Chung, B. Y. *J. Chem. Soc., Chem. Commun*. **1981**, 1231–1232.

(31) We did not firmly establish whether this product was the *O*- or *N*-acyl 1-hydroxybenzotriazole derivative; however, the IR spectrum showed an absorption at ca. 1825 cm^{-1}, indicative of the *O*-acyl isomer. For example, see: (a) Li, P.; Xu, J. C. *J. Chem. Soc., Perkin Trans. 2* **2001**, 113–120. (b) Katritzky, A. R.; Malhotra, N.; Fan, W.-Q.; Anders, E. *J. Chem. Soc., Perkin Trans. 2* **1991**, 1545–1547.

SCHEME 6. Synthesis of Depsilairdin Analogues 36a−c



alkoxides of secondary alcohols (e.g., borneol) in refluxing THF gave the corresponding esters **31a** in good yield.

The HOBt ester **27b** was readily prepared from **27a** using the standard method (Scheme 4).[31] Reaction of **27b** with the bromomagnesium alkoxide prepared from **5** (2 equiv) and freshly prepared PhMgBr (1.2 equiv) in refluxing THF gave the desired ester **28** in 40% yield along with recovered **5** (75%). Deprotection of **28** was achieved by sequential reaction with TBAF in CH_2Cl_2 (to remove the TBS groups) and then treatment with Dowex 50 in refluxing aqueous MeOH (to hydrolyze the MOM ether)[32] to give **1** (85%; $[\alpha]_D$ −45 (*c* 0.15, CH_2Cl_2), lit.[2] −65 (*c* 0.9, CH_2Cl_2)). ^1H and ^{13}C NMR spectra for synthetic **1** in $CDCl_3$ were essentially superimposable with those for natural **1** kindly provided by Prof. Pedras.[33]

Using an analogous approach, we prepared some simple analogues of depsilairdin (**1**) by replacing the Dhmp (**4**) residue with L-proline and *cis*- and *trans*-4-hydroxy-L-proline (Scheme 6). Assembly of the tetradepsipeptides **35a−c** proceeded smoothly and with yields comparable to those obtained in the synthesis of **24**. Compared to our difficulties with **24a**, hydrolyses of the methyl esters in **35a−c** were straightforward and could be accomplished with LiOH in methanolic THF. With **32b** and **32c**, ester hydrolysis was accompanied by partial hydrolysis of the proline TBS ether; the latter was avoided by using Me_3SnOH. In sharp contrast to **27a**, esterifications of the resulting acids with lairdinol A (**5**) were readily accomplished under standard conditions to give the corresponding analogues **36a−c** in good yields after removal of the TBS ether(s).[34]

The relative phytotoxicities of depsilairdin (**1**) and analogues **36a−c** (10^{-4}−10^{-5} M in aqueous methanol) to plants resistant [canola (*B. napus*) cv. Westar] and susceptible [brown mustard (*B. juncea*) cv. Cutlass] to blackleg isolates Mayfair 2 and Laird 2 were evaluated by a punctured leaf assay as previously described.[2] Analogous to the results reported using a natural sample,[2] synthetic depsilairdin (**1**) caused strong necrotic and chlorotic lesions on leaves of brown mustard (blackleg-susceptible), whereas canola leaves (blackleg-resistant) were not affected.[33] In contrast, the three analogues **36a−c** did not produce significant lesions on either brown mustard or canola leaves under the same conditions. We conclude that the presence of the Dhmp (**4**) residue in **1** is important for its selective toxicity.

Summary and Conclusions

In summary, the total synthesis of depsilairdin (**1**), a host-selective phytotoxin isolated from *L. maculans* (the causal agent of blackleg disease of oilseed Brassicas), has been achieved by sequential coupling of its five component residues. The reagents **7** and **8**, protected derivatives of the Hmb (**2**) and MeVal (**3**) residues, are readily available from D- and L-valine, respectively. The (2*S*,3*S*,4*R*)-3,4-dihydroxy-3-methyl-proline residue (Dhmp; **4**) is a previously unknown amino acid, and the suitably protected Dhmp derivative **9a** was prepared from *trans*-4-hydroxy-L-proline with significant modifications of Sardina's synthetic route to a related compound. The synthesis of lairdinol A (**5**) from (*R*)-carvone was described previously. Sequential PyBrop-mediated N-terminal extension of **9a** with **8** followed by reaction with **7** gave the tetradepsipeptide TBS-Hmb-MeVal-MeVal-(4-*O*-TES)-(3-*O*-MOM)Dhmp-OMe (**24b**) in excellent overall yield. Hydrolysis of the methyl ester in **24b** proved difficult and could be achieved only by reaction of Me_3SnOH with TBS-Hmb-MeVal-MeVal-(3-*O*-MOM)Dhmp-OMe (**25**), a derivative presumably activated by intramolecular hydrogen bonding. Similarly, esterification of the tetradepsipeptide acid with lairdinol A (**5**) was complicated by the sterically hindered nature of the carboxyl group and required a novel method involving reaction of the 1-hydroxybenzotriazole (HOBt) derived active ester with the bromomagnesium alkoxide of **5**. Three depsilairdin analogues **36a−c** were similarly prepared by replacing the Dhmp residue with L-proline and *cis*- and *trans*-4-hydroxy-L-proline; however, these analogues proved to be biologically inactive. The next step is to prepare probes (e.g., radiolabeled **1**)[35] to study the underlying mechanism(s) responsible for the host-selective

(32) Seto, H.; Mander, L. N. *Synth. Commun.* **1992**, *22*, 2823–2828.
(33) See Supporting Information for additional information.
(34) These conditions (i.e., 1 equiv each of DCC and DMAP in CH_2Cl_2 at rt for 15−20 h) produce the HOBt ester in situ. Attempted esterification of **27a** with **5** with this protocol (i.e., via **27b**), failed; however, heating the reaction at 50 °C in a sealed tube for 72 h produced an ester (70%). Although not fully characterized, NOE experiments (ref 28) suggest that this ester is 2-*epi*-**28**, and subjecting it to the deprotection sequence used for **28** gave a product that was clearly different from **1** by ^1H NMR.
(35) Pedras, M. S. C.; Zaharia, I. L.; Gai, Y.; Zhou, Y.; Ward, D. E. *Proc. Natl. Acad. Sci. U.S.A.* **2001**, *98*, 747–752.

phytotoxiciity of depsilairdin (**1**) and our results will be reported in due course.

Experimental Section[33]

Methyl (2*S*,3*S*)-3-(Methoxymethoxy)-4-oxo-1-(9-phenyl-9*H*-fluoren-9-yl)pyrrolidine-2-carboxylate (12). *N*,*N*-Dimethylaniline (1.9 mL, 1.8 g, 15 mmol) and MOM-Cl (0.81 mL, 0.86 g, 11 mmol) were sequentially added to a stirred solution of **11** (0.85 g, 2.1 mmol) in CH_2Cl_2 (8.5 mL) at room temperature under argon. After 1 day, the reaction mixture was diluted with diethyl ether and washed with 10% aq HCl and satd $NaHCO_3$. The organic layer was dried over Na_2SO_4, concentrated, and fractionated by FCC (30% ethyl acetate in hexane) to afford the title compound as a pale yellow foam (0.75 g, 80%): $[\alpha]_D$ −170 (*c* 1.1, $CHCl_3$) [lit. −158.2 (*c* 1.1, $CHCl_3$)];[16] ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.75 (1H, ddd, J = 1, 1, 7.5 Hz), 7.69 (1H, dd, J = 1, 8 Hz), 7.44 (1H, dd, J = 1, 7.5 Hz), 7.43−7.35 (5H, m), 7.33 (1H, ddd, J = 1, 7.5, 8 Hz), 7.30−7.23 (4H, m), 4.65 (1H, d, J = 6.5 Hz), 4.57 (1H, d, J = 6.5 Hz), 4.49 (1H, dd, J = 1, 7.5 Hz), 3.98 (1H, d, J = 7.5 Hz), 3.90 (1H, d, J = 17.5 Hz), 3.60 (1H, dd, J = 1, 17.5 Hz), 3.30 (3H, s), 3.11 (3H, s); ^{13}C NMR (125 MHz, $CDCl_3$) δ 209.3, 170.7, 146.6, 144.9, 141.6, 141.2, 139.9, 129.23, 129.19, 128.9, 128.3, 128.1, 127.7, 126.9, 125.5, 120.4, 120.4, 96.5, 77.7, 75.1, 61.4, 56.2, 52.2, 51.4; HRMS m/z calcd for $C_{27}H_{25}NO_5$ 443.1733, found 443.1717 (EI). ^{1}H and ^{13}C NMR data (in CD_2Cl_2) were consistent with those previously reported.[16]

Methyl (2*S*,3*S*)-3-(Methoxymethoxy)-3-methyl-4-oxo-1-(9-phenyl-9*H*-fluoren-9-yl)pyrrolidine-2-carboxylate (13). DMPU (5 mL) was added to a stirred solution of $KN(SiMe_3)_2$ (0.45 M in toluene; 3.0 mL, 1.4 mmol) in 5 mL of toluene at 0 °C under Ar. After 10 min, the mixture was cooled to −78 °C, and a solution of **12** (0.60 g, 1.4 mmol) in toluene and DMPU (1:1 (v/v); 8 mL) was added. After 1 h, CH_3I (0.84 mL, 1.9 g, 1.4 mmol) was added via syringe. After 2 h, the reaction was quenched by addition of KH_2PO_4 (1 M; 20 mL). The mixture was allowed to warm to ambient temperature and then was extracted with ethyl acetate. The combined organic layers were washed sequentially with H_2O and brine, dried over Na_2SO_4, concentrated, and fractionated by FCC (gradient elution; 10−20% ethyl acetate in hexane) to afford the diastereomer **14** (0.093 g, 15%) and a 8:1 mixture of the titled compound **13** and **15** (0.42 g, 68%), respectively. The mixture was used in the next step without further purification. In a smaller scale experiment (50 mg of **12**), fractionation of the crude by PTLC (35% ethyl acetate in hexane; 2 elutions) gave pure **13** (28 mg, 54%): $[\alpha]_D$ −8 (c 0.3, CH_2Cl_2); ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.75 (1H, ap d, J = 7.5 Hz), 7.70 (1H, ap d, J = 7.5 Hz), 7.21−7.47 (11H, m), 4.91 (1H, d, J = 7.5 Hz), 4.63 (1H, d, J = 7.5 Hz), 4.08 (1H, d, J = 17.5 Hz), 3.72 (1H, d, J = 17.5 Hz), 3.58 (1H, s), 3.24 (3H, s), 3.00 (3H, s), 1.68 (3H, s); ^{13}C NMR (125 MHz, $CDCl_3$) δ 211.1, 170.8, 146.4, 144.8, 141.5, 141.4, 140.1, 129.2, 129.1, 129.0, 128.3, 128.0, 127.5, 127.4, 126.9, 125.5, 120.50, 120.46, 92.8, 81.7, 74.8, 68.7, 56.0, 52.1, 51.1, 20.7; HRMS m/z calcd for $C_{28}H_{27}NO_5$ 457.1889, found 457.1889.

Methyl (2*S*,3*S*,4*R*)-4-Hydroxy-3-(methoxymethoxy)-3-methyl-1-(9-phenyl-9*H*-fluoren-9-yl)pyrrolidine-2-carboxylate [Pf-(3-*O*-MOM)Dhmp-OMe] (16). $NaBH_4$ (0.35 g, 0.92 mmol) was added to a stirred solution of an 8:1 mixture of **13** and **15**, respectively, (0.42 g, 0.092 mmol) in a mixture of CH_2Cl_2 and MeOH (1:1 (v/v); 9 mL) at −78 °C under argon. After 16 h, the reaction was quenched by dropwise addition of acetone (5 mL). The mixture was diluted with CH_2Cl_2, washed with satd $NaHCO_3$, dried over Na_2SO_4, concentrated, and fractionated by FCC (30% ethyl acetate in hexane) to afford **15** (0.046 g, 11%) and the titled compound **16** as a foam (0.32 g, 76%): $[\alpha]_D$ +200 (*c* 0.6, $CHCl_3$) [lit. 161.7 (*c* 0.6, $CHCl_3$)];[16] ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.82 (1H, d, J = 8 Hz), 7.67 (1H, d, J = 8 Hz), 7.60−7.64 (2H, m), 7.47−7.53 (2H, m), 7.38 (1H, dd, J = 8, 8 Hz), 7.31−7.36 (2H, m), 7.26−7.31 (4H, m), 7.14 (1H, dd, J = 8, 8 Hz), 4.74 (1H, d, J = 7.5 Hz), 4.64 (1H, d, J = 12 Hz), 4.60 (1H, d, J = 7.5 Hz), 3.81 (1H, dd, J = 3.5, 12 Hz), 3.53 (1H, d, J = 10.5 Hz), 3.38 (3H, s), 3.29 (3H, s), 3.21 (1H, dd, J = 3.5, 10.5 Hz), 2.83 (1H, s), 0.89 (3H, s); ^{13}C NMR (125 MHz, $CDCl_3$) δ 176.1, 148.2, 144.7, 142.1, 141.0, 139.4, 129.1, 128.7, 128.6, 127.9, 127.8, 127.7, 127.3, 127.2, 126.5, 120.6, 120.2, 92.8, 83.4, 75.8, 75.5, 69.7, 55.8, 55.0, 51.9, 23.4; HRMS m/z calcd for $C_{28}H_{29}NO_5$ 459.2046, found 459.2044. ^{1}H and ^{13}C NMR data (in CD_2Cl_2) were consistent with those previously reported.[16]

Methyl (2*S*,3*S*,4*R*)-3-(Methoxymethoxy)-3-methyl-1-(9-phenyl-9*H*-fluoren-9-yl)-4-(triethylsilyloxy)pyrrolidine-2-carboxylate [Pf-(4-*O*-TES)(3-*O*-MOM)Dhmp-OMe] (17b). 2,6-Lutidine (0.110 mL, 0.105 g, 0.980 mmol) and $Et_3SiOSO_2CF_3$ (0.17 mL, 0.194 g, 0.735 mmol) were sequentially added to a stirred solution of alcohol **16** (0.225 g, 0.489 mmol) in dry CH_2Cl_2 (5 mL) at 0 °C. After 15 min, the mixture was diluted with ethyl acetate and washed sequentially with satd $NaHCO_3$, 5% aq HCl, satd $NaHCO_3$, and brine. The organic layer was dried over Na_2SO_4, concentrated, and fractionated by FCC (20% ethyl acetate in hexane) to afford the titled compound **17b** (0.260 g, 93%): $[\alpha]_D$ +200 (*c* 1.2, CH_2Cl_2); ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.76 (1H, d, J = 7.5 Hz), 7.75−7.65 (3H, m), 7.45−7.51 (2H, m), 7.18−7.37 (6H, m), 7.07 (1H, dd, J = 7.5, 7.5 Hz), 5.13 (1H, d, J = 7.5 Hz), 4.56 (1H, d, J = 7.5 Hz), 3.47−3.57 (2H, m), 3.44 (3H, s), 3.28 (1H, dd, J = 6, 10 Hz), 3.21 (3H, s), 2.78 (1H, s), 1.56 (3H, s), 0.90 (9H, t, J = 8 Hz), 0.52 (6H, ap q, J = 8 Hz); ^{13}C NMR (125 MHz, $CDCl_3$) δ 172.2, 147.6, 146.5, 143.6, 142.6, 139.2, 129.2, 128.58, 128.55, 128.4, 128.1, 127.7, 127.5, 127.1, 125.5, 120.3, 119.8, 92.5, 83.1, 77.9, 77.4, 70.7, 55.0, 54.5, 51.3, 19.9, 6.9, 5.0; HRMS m/z calcd for $C_{34}H_{43}NO_5Si$ 573.2911, found 573.2906 (EI).

Methyl (2*S*,3*S*,4*R*)-3-(Methoxymethoxy)-3-methyl-4-(triethylsilyloxy)pyrrolidine-2-carboxylate [(4-*O*-TES)(3-*O*-MOM)Dhmp-OMe] (9b). A stirred suspension of **17b** (0.26 g, 0.45 mmol) and 10% Pd/C (0.10 g) in ^{i}PrOH (4 mL) was evacuated, and H_2 gas was introduced using a balloon. After 6 h, the reaction mixture was passed through pad of Celite, and the combined filtrate and CH_2Cl_2 washings were concentrated and fractionated by FCC (5% MeOH in CH_2Cl_2) to afford the titled compound **9b** (0.13 g, 88%) that was somewhat unstable and used immediately in the next step: $[\alpha]_D$ −54 (*c* 1.1, CH_3OH); ^{1}H NMR (500 MHz, $CDCl_3$) δ: 5.17 (1H, d, J = 7.5 Hz), 4.59 (1H, d, J = 7.5 Hz), 3.99 (1H, dd, J = 7.5, 9.5 Hz), 3.78 (3H, s), 3.60 (1H, s), 3.26 (3H, s), 3.10−2.99 (2H, m), 2.51 (1H, br s), 1.55 (3H, s), 0.94 (9H, t, J = 8 Hz), 0.58 (6H, ap q, J = 8 Hz); ^{13}C NMR (125 MHz, $CDCl_3$) δ: 171.9, 92.6, 82.3, 79.5, 68.6, 55.2, 52.2, 50.3, 18.8, 6.9, 4.9; HRMS m/z calcd for $C_{15}H_{31}NO_5Si$ 333.1971, found 333.1974 (EI).

Cbz-MeVal-(4-*O*-TES)(3-*O*-MOM)Dhmp-OMe (22b). PyBroP (0.27 g, 0.58 mmol) and DIPEA (0.14 mL, 0.10 g, 0.78 mmol) were sequentially added to a stirred solution compound **9b** (0.13 g, 0.39 mmol) and Cbz-MeVal-OH (**8**) (0.13 g, 0.51 mmol) in CH_2Cl_2 (5.5 mL) at 0 °C. The mixture was stirred for 10 min at 0 °C and then for 18 h at room temperature. The mixture was diluted with CH_2Cl_2, washed sequentially with aq citric acid (0.5 M) and satd aq $NaHCO_3$, dried over Na_2SO_4, concentrated, and fractionated by FCC (30% ethyl acetate in hexane) to give the titled compound **22b** (0.020 g, 90%): $[\alpha]_D$ −92 (*c* 1.5, CH_2Cl_2); ^{1}H NMR (500 MHz, $CDCl_3$) (a ca. 5.5:1 mixture of rotamers; signal for the major rotamer only) δ 7.28−7.39 (5H, m, Ph), 5.14 (d, J = 12.8 Hz), 5.10 (1H, d, J = 12.8 Hz), 5.17 (1H, d, J = 7.5 Hz), 4.60 (1H, d, J = 7.5 Hz), 4.54 (1H, d, J = 11 Hz), 4.38 (1H, dd, J = 7. 9.5 Hz), 4.26 (1H, s), 3.78 (1H, dd, J = 7, 9.5 Hz), 3.75 (3H, s), 3.52 (1H, dd, J = 9.5, 9.5 Hz), 3.28 (3H, s), 2.94 (3H, s), 2.20−2.34 (1H, m), 1.60 (3H, s), 1.03 (3H, d, J = 6.5 Hz), 0.96 (9H, t, J = 8 Hz), 0.88 (3H, d, J = 6.5 Hz), 0.62 (6H, ap q, J = 8 Hz); ^{13}C NMR (125 MHz, $CDCl_3$) (a ca. 5.5:1 mixture of rotamers; signals for the major rotamer only) δ 170.2, 167.6, 157.2, 136.9, 128.7, 128.2, 128.0, 92.7, 80.7, 77.0, 67.9,

67.5, 61.5, 55.3, 52.0, 50.1, 29.6, 28.0, 19.6, 19.1, 18.9, 6.9, 4.8; HRMS *m*/*z* calcd for $C_{29}H_{49}N_2O_8Si$ (M + H) 581.3253, found 581.3259 (ESI).

Cbz-MeVal-MeVal-(4-*O*-TES)(3-*O*-MOM)Dhmp-OMe (23b). A stirred suspension of **22b** (0.070 g, 0.12 mmol) and 10% Pd/C (20 mg) in ^{i}PrOH (1.2 mL) was evacuated, and H_2 gas was introduced using a balloon. After 4 h, the reaction mixture was passed through pad of Celite. The combined filtrate and CH_2Cl_2 washings were concentrated to give the crude deprotected amine (0.028 g). PyBroP (0.84 g, 0.18 mmol) and DIPEA 0.042 mL, 0.031 g, 0.24 mmol) were sequentially added to a stirred solution of Cbz-MeVal-OH (**8**) (0.041 g, 1.6 mmol) and the above crude amine in CH_2Cl_2 (2 mL) at 0 °C under argon. After 10 min, the mixture was allowed to warm to ambient temperature. After 18 h, the mixture was diluted with CH_2Cl_2, washed sequentially with aq citric acid (0.5 M) and satd aq $NaHCO_3$, dried over Na_2SO_4, concentrated, and fractionated by FCC (30% ethyl acetate in hexane) to give **23b** (0.071 g, 85%): $[\alpha]_D$ −110 (*c* 1.4, CH_2Cl_2); ^{1}H NMR (500 MHz, $CDCl_3$) (a ca. 2.2:1 mixture of rotamers; signals for the major rotamer only) δ 7.28−7.38 (5H, m), 5.17−5.20 (3H, m), 5.00 (1H, d, *J* = 11 Hz), 4.73 (d, *J* = 11 Hz), 4.61 (1H, d, *J* = 7.5 Hz), 4.42 (1H, dd, *J* = 7, 9.5 Hz), 4.27 (s), 3.76 (3H, s), 3.67−3.74 (2H, m), 3.54 (1H, dd, *J* = 9.5, 9.5 Hz), 3.28 (3H, s), 3.09 (3H, s), 2.87 (3H, s), 2.13−2.40 (2H, m), 1.60 (3H, s), 1.03 (d, *J* = 7 Hz), 0.93−0.99 (9H, m, $H_3CCSi \times 3$), 0.88 (3H, d, *J* = 7 Hz), 0.86 (3H, d, *J* = 7 Hz), 0.77 (3H, d, *J* = 7 Hz), 0.58−0.66 (6H, m); ^{13}C NMR (125 MHz, $CDCl_3$) (a ca. 2.2:1 mixture of rotamers; signals for the major rotamer only) δ 171.4, 169.8, 167.6, 157.1, 136.9, 128.7, 128.2, 127.8, 92.7, 80.8, 77.1, 67.9, 67.6, 60.7, 59.2, 55.3, 52.0, 50.3, 30.7, 29.5, 28.1, 27.6, 19.9, 19.7, 19.1, 18.6, 18.3, 6.9, 4.8; HRMS *m*/*z* calcd for $C_{35}H_{59}N_3O_9Si$ 693.4021, found 693.4010 (EI).

TBS-Hmb-MeVal-MeVal-(4-*O*-TES)(3-*O*-MOM)Dhmp-OMe (24b). Using the modified[11d] procedure of Wissner,[11c] oxalyl chloride (0.013 mL, 0.018 g, 0.15 mmol) was added dropwise to a solution of *tert*-butyldimethylsilyl (*R*)-2-((*tert*-butyldimethylsilyl)-oxy)-3-methylbutanoate (0.052 g, 0.15 mmol)[11b] and DMF (ca. 1 μL, 0.01 mmol) in CH_2Cl_2 (1 mL) at 0 °C under argon. The mixture was allowed to warm to ambient temperature and, after 4 h, was diluted with dry hexane (5 mL), and the precipitated solids were removed by filtration. The combined filtrate and hexane washings were concentrated to give the crude acid chloride **7**. A stirred suspension of **23b** (0.051 g, 0.073 mmol) and 10% Pd/C (9 mg) in ^{i}PrOH (0.5 mL) was evacuated, and H_2 gas was introduced using a balloon. After 4 h, the reaction mixture was passed through pad of Celite. The combined filtrate and CH_2Cl_2 washings were concentrated to give the crude deprotected amine (0.018 g) that was dissolved in CH_2Cl_2 (1 mL), and the resulting solution was added to the above crude **7**. DIPEA (0.019 mL, 0.014 g, 0.11 mmol) was added to the stirred mixture at 0 °C. The mixture was allowed to warm to ambient temperature and after 4 h, diluted with ethyl acetate, washed sequentially with water, 10% aq citric acid, satd aq $NaHCO_3$ and brine, dried over Na_2SO_4, and concentrated. The resulting yellow oil was fractionated by FCC (30% ethyl acetate in hexane) to give **24b** as a colorless oil (0.048 g, 85%): $[\alpha]_D$ −95 (*c* 2.06, CH_2Cl_2); ^{1}H NMR (500 MHz, $CDCl_3$) δ 5.13 (1H, d, *J* = 7.5 Hz), 5.10 (1H, d, *J* = 11 Hz), 5.02 (1H, d, *J* = 11 Hz), 4.60 (1H, d, *J* = 7.5 Hz), 4.43 (1H, dd, *J* = 6.5, 9.5 Hz), 4.26 (1H, s), 4.10 (1H, d, *J* = 6.5 Hz), 3.75 (3H, s), 3.71 (1H, dd, *J* = 6.5, 9.5 Hz), 3.53 (1H, dd, *J* = 9.5, 9.5 Hz), 3.28 (3H, s), 3.19 (3H, s), 3.15 (3H, s), 2.22−2.37 (2H, m), 1.91−2.00 (1H, m), 1.60 (3H, s), 1.02 (3H, d, *J* = 6.5 Hz), 0.97 (9H, dd, *J* = 7.5, 7.5 Hz), 0.87−0.95 (18H, m), 0.83 (3H, d, *J* = 6.5 Hz), 0.79 (3H, d, *J* = 6.5 Hz), 0.63 (6H, ddd, *J* = 7.5, 7.5, 7.5 Hz), 0.03 (3H, s), 0.02 (3H, s); ^{13}C NMR (125 MHz, $CDCl_3$) δ 173.5, 172.2, 169.7, 167.6, 92.7, 80.7, 79.4, 77.2, 67.8, 58.9, 58.5, 55.3, 52.0, 50.3, 31.9, 30.9, 30.2, 27.9, 27.8, 26.0, 19.8, 19.59, 19.57, 19.0, 18.7, 18.4, 18.2, 6.9, 4.8, −4.4, −5.0; HRMS *m*/*z* calcd for $C_{38}H_{75}N_3O_9Si_2$ 773.5042, found 773.5045 (EI).

TBS-Hmb-MeVal-MeVal-(3-*O*-MOM)Dhmp-OMe (25). Pyridine (0.12 mL) and HF·pyridine (0.12 mL) were added to a stirred solution of **24b** (40 mg, 0.052 mmol) in THF (1 mL) at 0 °C. After 1 h, the mixture was quenched by addition of satd $NaHCO_3$ (aq) (**caution**: CO_2 evolution), and then was diluted with ethyl acetate, washed sequentially with 2% aq citric acid (× 3), satd $NaHCO_3$ (aq) and brine, dried over Na_2SO_4, concentrated, and fractionated by FCC (40% ethyl acetate in hexane) to give **25** (32 mg, 95%): $[\alpha]_D$ −90 (*c* 1.1, CH_2Cl_2); ^{1}H NMR (500 MHz, $CDCl_3$) δ 5.11 (1H, d, *J* = 11 Hz), 5.04 (1H, d, *J* = 11 Hz), 4.82 (1H, d, *J* = 7.3 Hz), 4.74 (1H, d, *J* = 7.3 Hz), 4.33 (1H, s), 4.24 (1H, dd, *J* = 5, 11 Hz), 4.12 (1H, d, *J* = 11 Hz), 4.10 (1H, d, *J* = 6.5 Hz), 3.90 (1H, ddd, *J* = 5, 5, 11 Hz), 3.81 (1H, dd, *J* = 5, 11 Hz), 3.79 (3H, s), 3.42 (3H, s), 3.21 (3H, s), 3.14 (3H, s), 2.22−2.28 (2H, m), 1.90−2.01 (1H, m), 1.46 (3H, s), 1.01 (3H, d, *J* = 6.5 Hz), 0.94 (3H, d, *J* = 6.5 Hz), 0.91 (9H, s), 0.89 (3H, d, *J* = 6.5 Hz), 0.88 (3H, d, *J* = 6.5 Hz), 0.82 (3H, d, *J* = 6.5 Hz), 0.81 (3H, d, *J* = 6.5 Hz), 0.04 (3H, s), 0.03 (3H, s); ^{13}C NMR (125 MHz, $CDCl_3$) δ 173.6, 172.5, 170.9, 170.7, 92.7, 82.3, 79.8, 75.7, 67.8, 58.6, 58.5, 56.2, 53.9, 52.7, 32.0, 31.0, 29.9, 27.8, 27.6, 26.0, 22.0, 19.5, 19.3, 19.04, 19.00, 18.8, 18.4, 18.3, −4.4, −5.0; HRMS *m*/*z* calcd for $C_{32}H_{61}N_3O_9Si$ 659.4177, found 659.4156 (EI).

TBS-Hmb-MeVal-MeVal-(3-*O*-MOM)Dhmp-OH (26). Me_3Sn-OH (55 mg, 0.30 mmol) was added to a stirred solution of **25** (20 mg, 0.030 mmol) in $(CH_2Cl)_2$ (0.5 mL), and the reaction mixture was heated in an oil bath at 80 °C. After 2 days, the mixture was diluted with ethyl acetate and washed sequentially with 5% aq HCl and brine. The organic layer was dried over Na_2SO_4, concentrated, and fractionated by FCC (10% MeOH in CH_2Cl_2) to give **26** that was taken up in CH_2Cl_2, washed with 5% aq HCl, dried over Na_2SO_4, and concentrated to give **26** (17 mg, 87%) (the latter process was required to obtain material whose NMR spectra showed sharp signals); $[\alpha]_D$ −100 (*c* 0.4, CH_3OH); ^{1}H NMR (500 MHz, $CDCl_3$) δ 5.10 (1H, d, *J* = 11 Hz), 5.02 (1H, d, *J* = 11 Hz), 4.87 (1H, d, *J* = 7.5 Hz), 4.73 (1H, d, *J* = 7.5 Hz), 4.36 (1H, s, Pro-C-2), 4.22 (1H, dd, *J* = 4.5, 11 Hz, Pro-HC-5), 4.10 (1H, d, *J* = 6.5 Hz), 3.92 (1H, dd, *J* = 4.5, 4.5 Hz), 3.80 (1H, dd, *J* = 4.5, 11 Hz), 3.43 (3H, s), 3.20 (3H, s), 3.13 (3H, s), 2.24−2.36 (2H, m), 1.90−1.99 (1H, m), 1.46 (3H, s), 0.98 (3H, d, *J* = 6.5 Hz), 0.93 (3H, s, *J* = 6.5 Hz), 0.90 (9H, s), 0.85−0.89 (6H, m), 0.82 (3H, d, *J* = 6.5 Hz), 0.79 (3H, d, *J* = 6.5 Hz), 0.03 (3H, s), 0.02 (3H, s); ^{13}C NMR (125 MHz, $CDCl_3$) δ 173.6, 172.5, 171.3, 170.6, 92.5, 82.7, 79.8, 75.7, 67.9, 58.70, 58.67, 58.3, 53.2, 32.0, 31.1, 30.2, 27.7, 27.6, 26.0, 21.5, 19.5, 19.3, 19.1, 19.0, 18.8, 18.4, 18.3, −4.4, −5.0; HRMS *m*/*z* calcd for $C_{31}H_{60}N_3O_9Si$ (M + H) 646.4093, found 646.4086 (ESI).

TBS-Hmb-MeVal-MeVal-(4-*O*-TBS)(3-*O*-MOM)Dhmp-OH (27a). 2,6-Lutidine (0.021 mL, 0.020 g, 0.18 mmol) and ^{t}BuMe$_2$-SiOSO$_2$CF$_3$ (0.030 mL, 0.035 g, 0.13 mmol) were sequentially added to a stirred solution of **26** (0.017 g, 0.026 mmol) in dry CH_2Cl_2 (0.5 mL) at 0 °C under Ar. After 15 min, the mixture was diluted with ethyl acetate and washed sequentially with satd $NaHCO_3$ and brine. The organic layer was dried over Na_2SO_4 and concentrated to get crude tris-TBS derivative that was taken up in THF (1 mL). A solution of aq LiOH (0.5 M; 0.70 mL, 0.35 mmol) was added to the above stirred solution at 0 °C. The reaction mixture was allowed to warm to room temperature and, after 3 h, was diluted with CH_2Cl_2 and washed with 10% aq HCl. The organic layer was dried over Na_2SO_4, concentrated, and fractionated by FCC (10% MeOH in CH_2Cl_2) to give **27a** that was taken up in CH_2Cl_2, washed with 5% aq HCl, dried over Na_2SO_4 and concentrated to give the titled compound (17 mg, 86%) (the latter process was required to obtain material whose NMR spectra showed sharp signals): $[\alpha]_D$ −80 (*c* 0.5, CH_3OH); ^{1}H NMR (500 MHz, $CDCl_3$) δ 5.07−5.13 (2H, m), 5.00 (1H, d, *J* = 11 Hz), 4.67 (1H, d, *J* = 7.5 Hz), 4.40 (1H, dd, *J* = 6.5, 9.5 Hz), 4.29 (1H, s), 4.09 (1H, d, *J* = 6.5 Hz), 3.72 (1H, dd, *J* = 6.5, 9.5 Hz), 3.56 (1H, dd, *J* = 9.5, 9.5 Hz), 3.32 (3H, s), 3.18 (3H, s), 3.13 (3H, s), 2.21−2.36 (2H, m), 1.90−1.99 (1H, m), 1.58 (3H, s),

0.97 (3H, d, J = 6.5 Hz), 0.92 (3H, d, J = 6.5 Hz), 0.84−0.91 (24H, m), 0.82 (3H, d, J = 6.5 Hz), 0.77 (3H, d, J = 6.5 Hz), 0.11 (3H, s), 0.09 (3H, s), 0.03 (3H, s), 0.01 (3H, s); ^{13}C NMR (125 MHz, $CDCl_3$) δ 173.6, 172.3, 170.5, 170.0, 92.9, 81.1, 79.8, 77.4, 67.6, 58.9, 58.6, 55.6, 50.8, 32.0, 31.0, 30.2, 27.85, 27.75, 26.0, 25.8, 20.1, 19.65, 19.58, 19.1, 19.0, 18.7, 18.4, 18.2, 18.1, −4.4, −4.7, −4.99, −5.02; HRMS m/z calcd for $C_{37}H_{74}N_3O_9Si_2$ 759.4885 (M + H) 760.4958, found 760.4967 (ESI).

TBS-Hmb-MeVal-MeVal-(4-*O*-TBS)(3-*O*-MOM)Dhmp-OBt (27b).[31] DCC (16 mg, 0.080 mmol) was added to a stirred solution of acid **27a** (20 mg, 0.026 mmol) and HOBt (5 mg, 0.04 mmol) in CH_2Cl_2 (0.5 mL). After 6 h the reaction mixture was diluted with ethyl acetate (0.5 mL). The precipitated DCU was filtered off, and the combined filtrate and washings were concentrated and fractionated by PTLC (60% ethyl acetate in hexane) to give the HOBt ester (21 mg, 91%) as a thick oil: $[\alpha]_D$ −70 (c 0.4, CH_2Cl_2); ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.02 (1H, d, J = 8.5 Hz), 7.72 (1H, d, J = 8.5 Hz), 7.54 (1H, dd, J = 8.5, 8.5 Hz), 7.40 (1H, dd, J = 8.5, 8.5 Hz), 5.30 (1H, d, J = 7.5 Hz), 5.14 (1H, d, J = 11 Hz), 5.10 (1H, d, J = 11 Hz), 4.81 (1H, d, J = 7.5 Hz), 4.63 (1H, s), 4.57 (1H, dd, J = 6.5, 9.5 Hz), 4.12 (1H, d, J = 6.5 Hz), 3.86 (1H, dd, J = 6.5, 9.5 Hz), 3.72 (1H, dd, J = 9.5, 9.5 Hz), 3.45 (3H, s), 3.26 (3H, s), 3.15 (3H, s), 2.41−2.30 (2H, m), 1.96 (1H, dqq, J = 6.5, 6.5, 6.5 Hz), 1.73 (3H, s), 0.94 (3H, d, J = 6.5 Hz), 0.93 (9H, s), 0.92 (9H, s), 0.91 (3H, d, J = 6.5 Hz), 0.90 (3H, d, J = 6.5 Hz), 0.88 (3H, d, J = 6.5 Hz), 0.86 (3H, d, J = 6.5 Hz), 0.79 (3H, d, J = 6.5 Hz), 0.16 (3H, s), 0.14 (3H, s), 0.04 (3H, s), 0.03 (3H, s); ^{13}C NMR (125 MHz, $CDCl_3$) δ 173.7, 172.4, 170.4, 164.0, 143.6, 128.9 (× 2), 124.9, 120.3, 109.6, 93.3, 81.3, 79.7, 77.5, 67.1, 58.9, 58.7, 56.5, 50.4, 32.0, 31.0, 30.3, 27.9, 27.6, 26.0, 25.9, 19.82, 19.76, 19.6, 19.0 (× 2), 18.6, 18.4, 18.2 (× 2), −4.4, −4.6, −4.97, −4.99; HRMS m/z calcd for $C_{43}H_{77}N_6O_9Si_2$ (M + H) 877.5290, found 877.5259 (ESI).

TBS-Hmb-MeVal-MeVal-(4-*O*-TBS)(3-*O*-MOM)Dhmp-Lar (28). PhMgBr (0.60 M in THF; 0.068 mL, 0.040 mmol) was added to a stirred solution of lairdinol A (**5**) (16 mg, 0.068 mmol) in THF (0.25 mL) at 0 °C. After 30 min, a solution of the **27b** (30 mg, 0.034 mmol) in THF (0.25 mL) was added. The reaction mixture was allowed to reach ambient temperature and then was heated under reflux. After 5 h, the mixture was allowed to cool, quenched by addition of phosphate buffer (pH = 7), and then extracted with CH_2Cl_2. The combined organic layers were dried over Na_2SO_4, concentrated, and fractionated using PTLC (60% EtOAc in hexane) to afford lairdinol A (**5**) (12 mg, 75%) and **28** (13 mg, 40%): $[\alpha]_D$ −39 (c 0.20, CH_2Cl_2); ^{1}H NMR (500 MHz, $CDCl_3$) δ: 5.10 (1H, d, J = 11 Hz), 5.09 (1H, d, J = 7 Hz), 5.02 (1H, d, J = 11 Hz), 4.72−4.68 (2H, m), 4.65 (1H, d, J = 7 Hz), 4.59 (1H, dd, J = 4, 11.5 Hz), 4.41 (1H, dd, J = 7, 9.5 Hz, 4.19 (1H, s), 4.10 (1H, d, J = 6 Hz), 3.69 (1H, dd, J = 7, 9.5 Hz), 3.57 (1H, dd, J = 9.5, 9.5 Hz), 3.29 (3H, s), 3.20 (3H, s), 3.13 (3H, s), 2.40−2.18 (2H, m), 2.10−1.53 (10H, m), 1.74 (3H, s), 1.57 (3H, s), 1.40−1.16 (4H, m), 1.13 (3H, s), 1.03 (3H, d, J = 6.5 Hz), 0.99 (3H, s), 0.92 (3H, d, J = 6.5 Hz), 0.90 (9H, s), 0.89 (9H, s), 0.88 (3H, d, J = 6.5 Hz), 0.87 (3H, d, J = 6.5 Hz), 0.82 (3H, d, J = 6.5 Hz), 0.78 (3H, d, J = 6.5 Hz), 0.11 (3H, s), 0.08 (3H, s), 0.03 (3H, s), 0.01 (3H, s); ^{13}C NMR (125 MHz, $CDCl_3$) δ: 173.5, 172.1, 169.8, 166.3, 150.5, 108.6, 92.8, 82.5, 80.7, 79.6, 77.2, 71.6, 68.1, 59.0, 58.6, 56.2, 53.5, 50.5, 45.8, 40.88, 40.83, 38.3, 32.0, 31.1, 30.2, 27.8, 27.8, 26.5, 26.0, 25.93, 25.86, 25.3, 22.9, 21.3, 19.7, 19.6, 19.6, 19.3, 19.0, 18.7, 18.4, 18.1, 18.1, 14.5, −4.4, −4.6, −5.01, −5.06; HRMS m/z calcd for $C_{52}H_{98}N_3O_{10}Si_2$ (M + H) 980.6785, found 980.6761 (ESI).

Depsilairdin (1). TBAF (0.011 g, 0.041 mmol) was added to a stirred solution of **28** (4 mg, 0.004 mmol) in CH_2Cl_2 (0.1 mL) at room temperature. After 2 d, the mixture was diluted with CH_2Cl_2 and washed with water. The organic layer was dried over Na_2SO_4, concentrated, and fractionated using FCC (50% ethyl acetate in hexane) to afford corresponding diol (3 mg, > 90%). Dry Dowex 50 (50 mg)[32] was added to a solution of the above diol (3 mg) in a 5:1 (v/v) mixture of MeOH and H_2O, respectively (0.5 mL), and the stirred mixture was heated under reflux. After 36 h, the mixture was filtered. The combined filtrate and MeOH washings were concentrated and fractionated by PTLC (high performance cyano matrix TLC plates, 10 mm × 10 mm × 0.2 mm; 40% acetonitrile in H_2O) to afford **1** (2.5 mg, 85% from **28**) as a colorless oil, $[\alpha]_D$ −45 (c 0.15, CH_2Cl_2) [lit.[2] −65 (c 0.9, CH_2Cl_2)]. ^{1}H and ^{13}C NMR data (in C_6D_6 solution) were consistent with those reported previously.[2]

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Supporting Information Available: Experimental procedures, characterization data, and copies of ^{1}H and ^{13}C NMR spectra for all reported compounds; comparison of NMR data for natural and synthetic **1**; NMR spectra for the reaction of MOM-Cl with imidazole in DMF-d_7; procedures and results for phytotoxicity assays. This material is available free of charge via the Internet at http://pubs.acs.org.