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# Synthesis of Glycocinnasperimicin D

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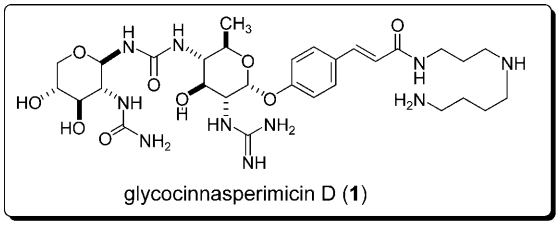
**Abstract:** The first total synthesis of amino sugar antibiotic glycocinnasperimicin D (**1**) has been achieved by a convergent, three-component coupling strategy. The key steps involve the Heck–Mizoroki reaction by using the iodophenyl glycoside **50** and acryl amide **32** to furnish the right core structure of **1**, and the construction of the urea glycoside employing the reaction of glycosyl isocyanate **8** with amino sugar **9**. Glycosyl isocyanate **8** was prepared by the oxidation of isonitrile **10**, which displayed excellent reactivity in the coupling event. Synthetic roadblocks, encountered during this synthetic effort, have led to the development of the α-selective, Lewis acid catalyzed phenyl glycosylation process with 2-amino-hexopyranose and a procedure for acetonide deprotection without affecting the silyl ethers.

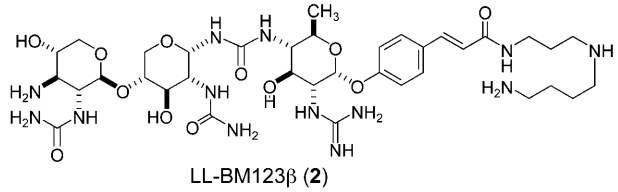
**Keywords:** amino sugars · antibiotics · glycosylation · Heck reaction · isonitriles · sigmatropic rearrangement

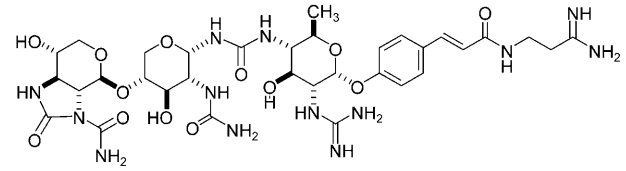
## Introduction

In 1985, Umezawa and co-workers at the Institute of Microbial Chemistry (Tokyo) in Japan reported the isolation and characterization of glycocinnasperimicin D (**1**), an antibiotic produced by the *Nocardia* sp. strain MG615-7F6 isolated from a soil sample collected in Fuchu City.[1] Analysis of its spectroscopic properties demonstrated that this substance belongs to the glycocinnaspermidine family of amino sugar antibiotics that includes LL-BM123β (**2**)[2] and coumamidine γ1 (**3**),[3] all of which were isolated and identified by researchers at the Lederle (New York) and Abbott (Illinois) Laboratories in the United States of America.

The structurally elaborate, nitrogen-rich and highly functionalized glycocinnasperimicin D possesses two unusual carbohydrates, including a 2-ureido-pentose in its left portion and a 2-guanidino-4-ureido-6-deoxy-α-D-glucopyranose with *p*-cinnamoylspermidine aglycon on the right side. In addition, these two amino sugars are joined by a unique glycosyl urea linkage. The stereochemistry of the urea glycoside

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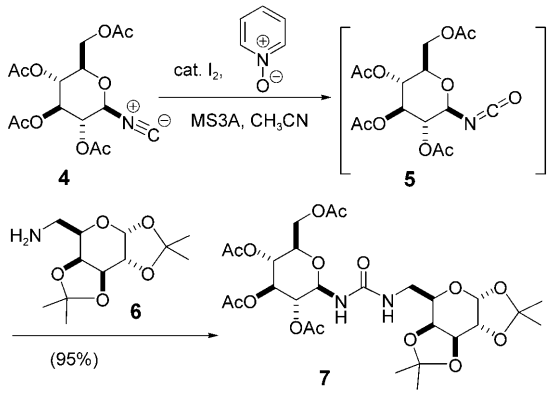
on the left-hand amino sugar was determined to be a β-linkage, assignment of which is based on the observation that the anomeric proton appearing at $\delta = 4.84$ ppm in the $^1$H NMR spectrum of **1** (40 °C, $D_2O$) has a $J$ value of 9.0 Hz. Although glycocinnasperimicin D is of special interest, owing to its broad-spectrum activity against gram-negative organisms and its undetermined mechanism of action, this antibiotic is no longer available from natural sources as a result of a mutation of the *Nocardia* strain.[4] The structural complexity, potent biological activity, and lack of availability of glycocinnasperimicin D have combined to stimulate the interest of synthetic chemists.[5] We now report a full account of our studies and observations leading to the first synthesis of glycocinnasperimicin D.[6,7]

## Results and Discussion

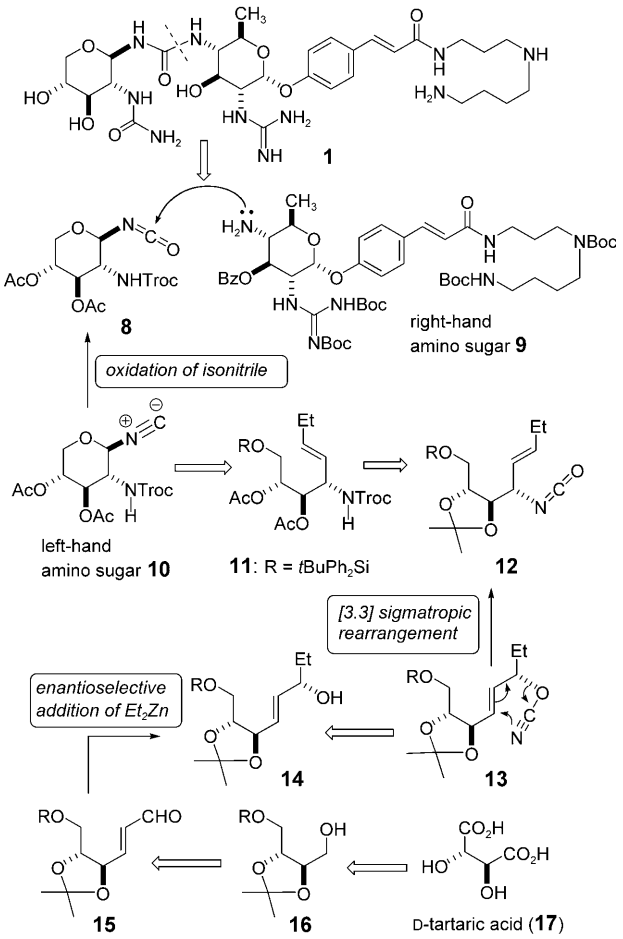
**Synthetic plan**: The major synthetic challenge posed by glycocinnasperimicin D is related to the difficulty associated with selective installation of the glycosyl urea linkage when a variety of nitrogen-containing functional groups are present. To surmount this problem, we have developed a new approach for the preparation of urea glycosides (Scheme 1).[8] The key features of this approach include




Scheme 1. A new approach to the synthesis of urea glycosides.

1) generation of a highly reactive glycosyl isocyanate **5** by the oxidation of glycosyl isonitrile **4** under mild conditions (catalytic iodine, pyridine *N*-oxide, acetonitrile, room temperature) and 2) without isolation, reaction of **5** with amino sugar **6** to furnish the urea glycoside **7** in excellent yield. Armed with this method, the convergent tactic for the synthesis of glycocinnasperimicin D and the left-hand amino sugar, outlined a retrosynthetic style in Scheme 2, was evolved.

In the retrosynthetic route, disconnection of the urea glycosyl linkage in the target **1** generates glycosyl isocyanate **8** and the highly functionalized right-hand amino sugar **9**. Isocyanate **8** would arise by oxidation of the isonitrile group in left-hand amino sugar **10**, and oxidative cleavage of the

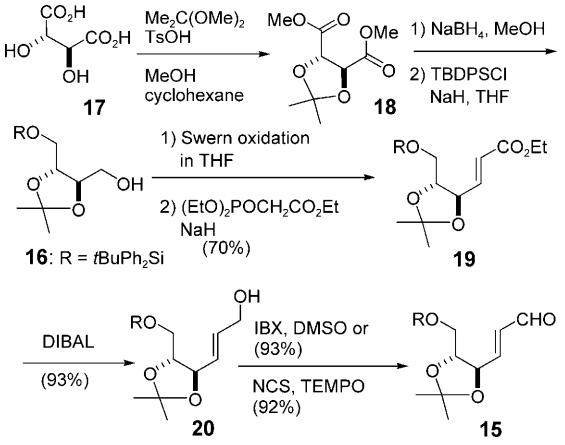



Scheme 2. Retrosynthetic plan of glycocinnasperimicin D (**1**). Boc: *tert*-butyloxycarbonyl; Bz: benzoyl; Troc: 2,2,2-trichloroethoxycarbonyl.

alkene moiety in allyl amine **11** would deliver the pyranose ring of **10**. The allyl amine **11** was to be constructed from the chiral, selectively protected butantetraol derivative **16**, prepared from D-tartaric acid (**17**). Based on the results of our previous studies exploring stereoselective methods for allyl amine synthesis,[9] we anticipated that enantioselective addition of diethylzinc ($Et_2Zn$) to aldehyde **15** would provide the allyl alcohol **14**, which in turn would be converted to allyl isocyanate **12** by [3.3] sigmatropic rearrangement of the allyl cyanate **13**. Based on this idea, we launched a synthetic venture toward glycocinnasperimicin D.

**Studies directed at the preparation of the left-hand amino sugar 10**: The synthesis of the left-hand amino sugar started with the chiral C-4 synthon **16**, which was prepared from D-tartaric acid (**17**) in three steps (Scheme 3) by using known procedures.[10] The robust TBDPS group was selected for protection of the primary hydroxyl, since we believed it would survive the conditions used for all of the reactions in the sequence. Swern oxidation of **16** in THF followed by Horner–Wadsworth–Emmons olefination in a one-pot operation afforded ester **19** in 70% yield.[11] DIBAL reduction of **19** gave the allyl alcohol **20**, which was subjected to IBX[12]

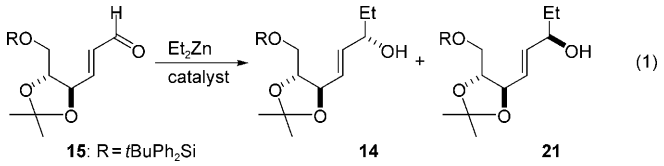
  




Scheme 3. Preparation of the unsaturated aldehyde **15**. DIBAL: diisobutylaluminun hydride; IBX: 1-hydroxy-1,2-benziodoxol-3(1*H*)one 1-oxide; NCS: *N*-chlorosuccinimide; TBDPS: *tert*-butyldiphenylsilyl; TEMPO: 2,2,6,6-tetramethylpiperidine *N*-oxyl.

or TEMPO oxidation[13] to produce the α,β-unsaturated aldehyde **15** in good yields, respectively.

**Enantioselective addition of $Et_2Zn$ to aldehyde 15**: Enantioselective addition of $Et_2Zn$ to the α,β-unsaturated aldehyde **15** was examined in the presence of the catalyst (*S*)-diphenyl(1-methylpyrrolidin-2-yl)methanol (DPMPM), reported by Soai [Eq. (1)].[14]



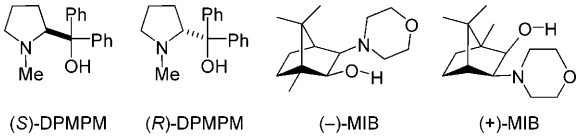

The reaction with a 5 mol% loading of (*S*)-DPMPM and 100 mg of **15** led to the generation of a mixture of allyl alcohols in 75–80% yields and with a 93:7 dr (favoring **14**; determined by $^1$H NMR spectroscopic analysis of the corresponding acetates, Table 1, entry A; dr = diastereromeric ratio). Disappointingly, the yield of this process decreased considerably (62%, entry B) when 600 mg of the substrate was employed. An increase of the catalyst loading to 10 mol% in a reaction carried out on a 200 mg scale enabled attainment of the small-scale yield (82%, entry C). More importantly, the use of 10 mol% of the catalyst realized an efficient reaction on a 5 g scale (79% yield with a 94:6 dr, entry D).

The catalyst, 3-*exo*-morpholinoisoborneol (MIB) developed by Nugent, was also probed.[15] Although the preparation of MIB is somewhat more laborious than that of DPMPA, better results were obtained when 8 mol% loading of (−)-MIB was employed for a 500 mg scale reaction (90% yield, 96:4 dr, entry E).[16] To explore the scope and limitation of the enantioselective addition of $Et_2Zn$ for stereoselective introduction of an allyl carbinol stereocenter, we carried out reactions with (*R*)-DPMPM and (+)-MIB, and

Table 1. Stereoselective construction of a new allyl carbinol stereocenter by using the enantioselective addition of diethylzinc.

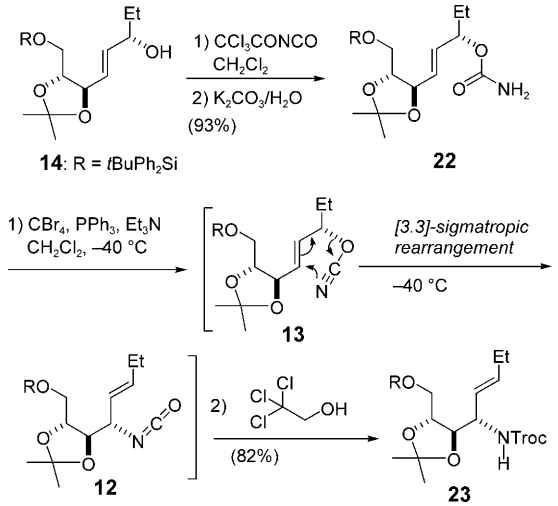
| Entry | Scale[a] [mg] | Catalyst ([mol%]) | Yield [%] | Selectivity[b] (dr, **14**/**21**) |
|---|---|---|---|---|
| A | 100 | (*S*)-DPMPM (5) | 75–80 | 93:7 |
| B | 600 | (*S*)-DPMPM (5) | 62 | 94:6 |
| C | 200 | (*S*)-DPMPM (10) | 82 | 94:6 |
| D | 5000 | (*S*)-DPMPM (10) | 79 | 94:6 |
| E | 500 | (−)-MIB (8) | 90 | 96:4 |
| F | 200 | (*R*)-DPMPM (10) | 77 | 7:93 |
| G | 300 | (+)-MIB (8) | 90 | 5:95 |

[a] Scale given by mg substrate. [b] Determined by $^1$H NMR spectroscopic analysis of the products after acetylation.




found that a complete reversal of stereospecificity took place without affecting the yield (77% yield with 7:93 dr in entry F, and 90% with 5:95 selectivity in entry G). Since the yields and selectivities in each pair of processes, (entries A/F) by using DPMPM and (entries E/G) employing MIB, are comparable, reagent control appears to be dominant and no matched/mismatched issues exist. In summary, enantioselective addition of $Et_2Zn$ to the α,β-unsaturated aldehydes can be performed in a highly selective manner at remote positions in which the effect of chiral centers in the substrates are negligible.

The next task in this synthetic sequence involves the preparation of allyl amine **23** through exploitation of an allyl cyanate-to-isocyanate rearrangement (Scheme 4).[17] For this purpose, the allyl alcohol **14** was transformed to the corresponding carbamate **22** by treatment with trichloroacetyl isocyanate followed by hydrolysis with potassium carbonate in aqueous methanol. Dehydration of carbamate **22** with triphenylphosphine, carbon tetrabromide, and triethylamine at



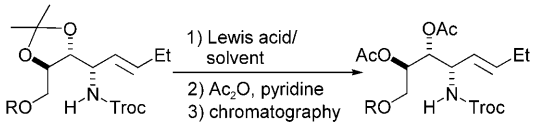

Scheme 4. Synthesis of allyl amine **23** by using an allyl cyanate-to-isocyanate rearrangement.

−40 °C generated the allyl cyanate **13**, which spontaneously underwent [3.3] sigmatropic rearrangement to afford allyl isocyanate **12**.[18] Due to the pronounced lability of isocyanates toward hydrolysis, transformation of **12** was carried out without using an aqueous workup; the reaction mixture was treated with a large excess of 2,2,2-trichloroethanol (12 equiv). Workup and purification then furnihsed the Troc-carbamate **23** in 76% overall yield from allyl alcohol **14**.

Deprotection of the isopropylidene ketal moiety in **23** proved to be unexpectedly difficult (Table 2). Application of a number of protocols resulted in hydrolysis of both the acetonide and TBDPS groups. Further experimentation found that Kim's method was appropriate for this task (entry A).[19] Treatment of **23** with ferric chloride hexahydrate ($FeCl_3{\cdot}6H_2O$) in chloroform followed by immediate acetylation of the resultant diol, to prevent silyl-group migration, afforded the requisite acetate **11** in 70% yield (based on the consumed starting material) along with recovered **23**. Further efforts to improve this deprotection step led to the discovery of unusual reaction conditions involving Lewis acid catalyzed thiolysis with thiols as the solvent.[20] For example, combinations of trichloroborane–dimethyl sulfide complex with either ethanethiol (entry B) or 1,2-ethanedithiol (entry C), and zinc triflate with 1,2-ethanedithiol (entry D) were found to promote smooth deprotection reactions. As a result, facile reactions leading to complete consumption of **23** on a 500 mg scale were observed to take place in these cases, affording **11** in good yields (84–89% after acetylation and purification). Since excess thiol can be removed during aqueous workup, this procedure appears to be an excellent method for acetonide deprotection, with the exception of the disagreeable odors.

Table 2. New procedures for unmasking the acetonide group in **23** without affecting the TBDPS silyl ether moiety.

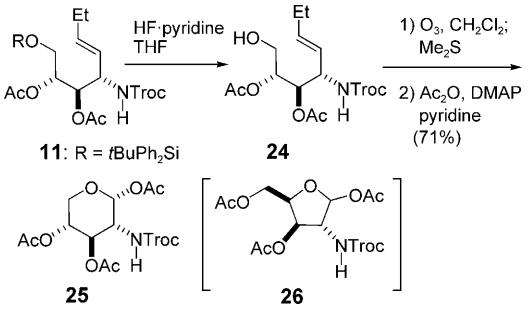



**23**: R = *t*BuPh$_2$Si **11**

| Entry | Lewis acid | Solvent | Yield [%] |
|---|---|---|---|
| A | $FeCl_3{\cdot}6H_2O$ | $CHCl_3$ | 70 (17)[a] |
| B | $BCl_3{\cdot}Me_2S$ | EtSH | 84 |
| C | $BCl_3{\cdot}Me_2S$ | $HSCH_2CH_2SH$ | 86 |
| D | $Zn(OTf)_2$ | $HSCH_2CH_2SH$ | 89 |

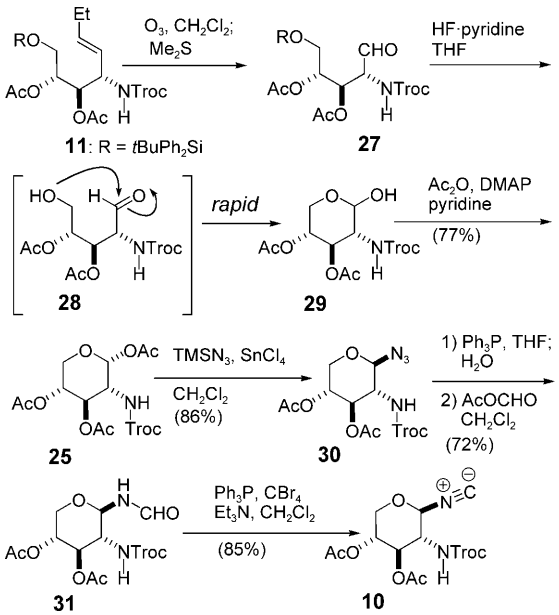
[a] Value in parentheses indicates the recovery of starting material.

Procedures to transform the allyl amine **11** into the left-hand amino sugar were investigated next (Scheme 5). Although removal of the TBDPS group in **11** with tetrabutylammonium fluoride ($n$Bu$_4$NF) buffered with AcOH[21] was complicated by acetyl-group migration, treatment of **11** with HF/pyridine in THF[22] cleanly provided a fragile intermediate alcohol, which was immediately subjected to ozonolysis ($CH_2Cl_2$, −78 °C). Acetylation gave the acetyl α-glycoside **25** predominantly (71%, for three steps). In practice, this reaction sequence could be performed conveniently and efficiently on a 30 mg scale, but on larger scales the yields dropped to less than 50% and the process was accompanied by a significant amount of formation of a byproduct, the structure of which was tentatively assigned as the pentose **26**, presumably stemming from 1,2-migration of the acetyl group.




Scheme 5. Transformation of allyl amine **11** into the pyranose **25**. DMAP: 4-dimethylaminopyridine.

As a result of the problem encountered in this sequence, an alternative approach was investigated (Scheme 6). We envisaged that byproduct formation could be avoided by carrying out ozonolysis prior to silyl deprotection. It was speculated that rapid hemiacetal formation (**28**→**29**) would block the primary hydroxyl in **28** and prevent both acetyl 1,2-migration and epimerization of the α-amino aldehyde. Indeed, this change led to a reliable transformation of **11** into **25** on a 1 gram scale in 77% overall yield.




Scheme 6. Synthesis of the left-hand amino sugar **10**. TMS: trimethylsilyl.

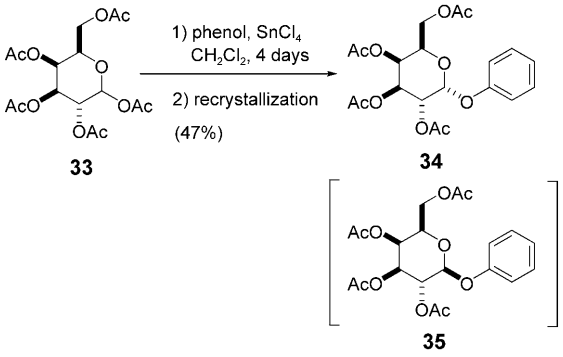
Treatment of the acetyl glycoside **25** with trimethylsilyl azide in the presence of tin(VI) chloride furnished the β-gly-

cosyl azide **30** exclusively (86%).[23] The isonitrile group was introduced at the anomeric position by using a four-step sequence involving the Staudinger reaction of glycosyl azide **30** with triphenylphosphine followed by hydrolysis of the resultant iminophosphorane to give the glycosylamine, which was immediately treated with acetic formic anhydride to give the formamide **31** (72% yield). Dehydration of **31** with triphosgene generated the desired isonitrile, albeit in modest and variable yields (40–60%). However, implementation of the more reliable, modified Appel's procedure ($PPh_3$, $CBr_4$, $Et_3N$, $CH_2Cl_2$), furnished the left-hand amino sugar **10** in 85% yield.[24]

**Synthetic plan for the construction of the right-hand amino sugar 9**: The strategy designed for preparation of the right-hand amino sugar **9** relies on the Heck–Mizoroki reaction, which would couple the iodophenyl-6-deoxy-2-amino-α-D-glycopyranoside **A** and acrylamide **32** subunits (Scheme 7). This convergent tactic was selected to alleviate concerns about potential problems that might be encountered in the glycosylation reaction of the poorly nucleophilic phenol with a highly functionalized pyranose.[25] In studies, described in detail elsewhere,[26] we demonstrated that the Heck–Mizoroki reaction of iodophenyl glycopyranoside with acrylamide **32** proceeded smoothly to introduce the cinnamoyl structure and sperimidine moiety on the iodophenyl glycopyranoside in one step. The remaining task in the execution of this plan is the multigram synthesis of phenyl 2-amino-α-D-glycopyranoside **B** and its transformation into subunit **A** by deoxygenation of the C-6 hydroxy group and iodination of the phenyl ring. Owing to the fact that our preliminary experiments showed that catalytic hydrogenolysis of 6-iodo-glycopyranose is the most simple and high-yielding method for multigram synthesis of 6-deoxy-glycopyranoside, we did not chose a route involving glycosylation of iodophenol followed by deoxygenation of the C-6 hydroxyl. An iodophenyl glycopyranoside would not be compatible with the conditions employed in the deoxygenation step.[27]

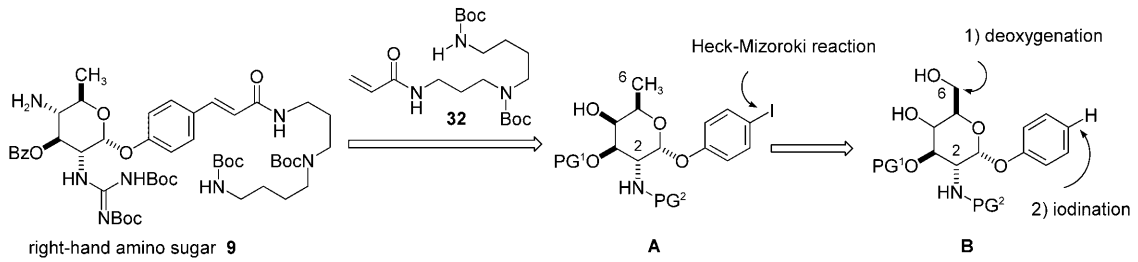
**Synthesis of the phenyl 2-amino-α-D-glycopyranoside B**: In studies performed to find deoxygenation and iodination reaction conditions, we initially planned to prepare phenyl acetyl galactopyranoside as model compounds. Although synthesis of aryl α-glycopyranosides and isomerization of the phenyl β-glycopyranosides into the α-isomers (anomerization) have been described,[28] the yields of the reported procedures are relatively low (23–32%). In contrast, β-selective syntheses of phenyl glucosides[29] and galactosides[30] are well documented. Prompted by these earlier reports, preliminary investigations began with the Lewis acid catalyzed glycosylation of phenol with penta-*O*-acetyl-D-galactopyranoside **33** to obtain the β-anomer **35** (Scheme 8). During



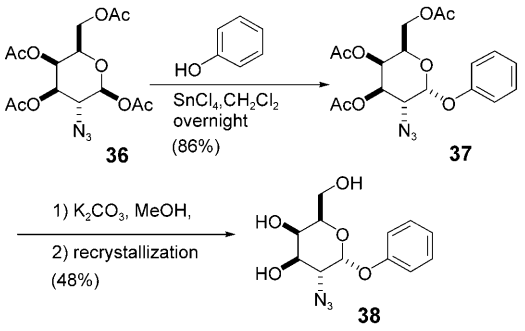

Scheme 8. The unexpected α-selective galactosylation of phenol.

Lewis acid screening studies, a reaction mixture containing pentaacetyl galactopyranoside **33**, phenol, and tin(IV) chloride in $CH_2Cl_2$ was accidentally left at room temperature for several days and then subjected to workup. Surprisingly, the predominant product was found to be the α-galactopyranoside **34**. The significance of this fortuitous experimental result led us to carry out this process under controlled conditions, and we found that the glycosylation reaction initially led to a mixture of anomers, which, with a prolonged reaction time, underwent a gradual decrease in the amount of the β-isomer and a concomitant increase in the α-anomer. Contrary to our initial plan, phenyl α-D-galactopyranoside **34** was obtained in a modest yield (47%, unoptimized) after recrystallization.[31]

These unexpected results led us to explore the reaction of the 2-azido-D-galactose derivative **36**[32] with phenol in the presence of tin(VI) chloride (Scheme 9).[33] This reaction proceeded smoothly in $CH_2Cl_2$ at room temperature overnight to produce a 4:1 mixture ($^1H$ NMR spectroscopic analysis of the crude product mixture) of phenyl galactopyranosides favoring the α-anomer **37** in 86% yield. Removal of



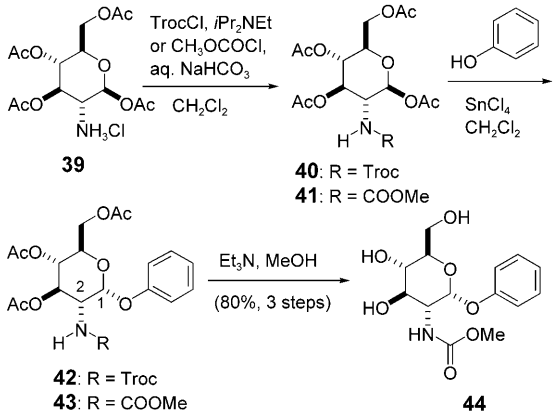

Scheme 7. Plan for synthesis of the right-hand amino sugar **9**.




Scheme 9. α-Selective glycosylation of phenol with 2-azido-D-galactose derivative **36**.

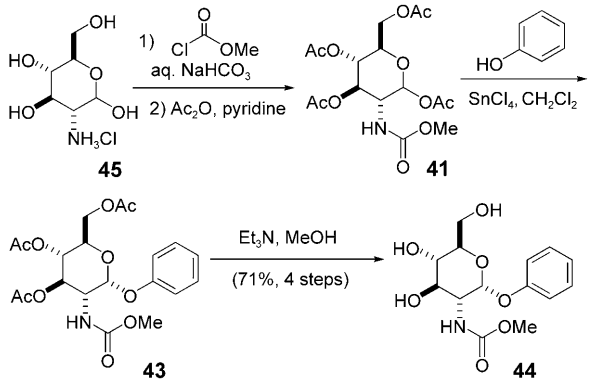
acetyl groups in **37** afforded the highly crystalline triol **38**, and the β-isomer was conveniently removed by recrystallization. As a result, multigram quantities of the α-anomer **38**, a synthetic equivalent to phenyl 2-amino-α-D-glycopyranoside **B**, were obtained in 48% yield.

At first, we had excluded a plan to synthesize phenyl α-glycopyranosides by starting from *N*-alkoxycarbonyl derivatives of glucosamine, because literature precedent clearly indicated that the neighboring effect of the *N*-alkoxycarbonyl group results in formation of 1,2-*trans*-glycosylation products.[34] However, the results uncovered in studying Lewis acid catalyzed α-selective glycosylation reactions with phenol guided us to reconsider this plan (Scheme 10). Accordingly, the *N*-trichloroethoxy carbonyl derivative **40** was prepared from tetraacetyl glucosamine **39**[35] by a conventional method (TrocCl, $iPr_2NEt$, $CH_2Cl_2$) and then subjected to the glycosylation reaction conditions. We were pleased to find that this process was also attended by anomerization of the initially formed β-isomer into the α-anomer; as a result, it furnished the phenyl α-D-glycopyranoside **42** predominantly ($^1H$ NMR spectroscopic analysis). Unfortunately, removal of the acetyl groups in **42** was complicated by loss of the Troc protecting group. To circumvent this problem, the more robust methoxycarbonyl-protected derivative **41** was prepared and then employed in the Lewis acid catalyzed glycosylation to afford the phenyl α-glycopyranoside **43** with α/β = 10:1 selectivity. Treatment of **43** with triethylamine in methanol produced the crystalline triol **44**, which was easily purified by recrystallization (80% yield, three steps from **41**).




Scheme 10. 1,2-*cis*-Glycosylation of phenol with a *N*-alkoxycarbonyl derivative of glucosamine.

Further efforts established a four-step synthesis of **44** from commercially available and inexpensive D-glucosamine hydrochloride (**45**) without the need for chromatographic purifications (Scheme 11). By using the procedure reported by Schmidt,[36] **45** was transformed into a mixture of *N*-methoxycarbonyl-protected *O*-acetyl glucosamine **41**. Glycosylation of phenol with **41**, followed by removal of the acetyl groups in the product **43** and recrystallization led to production of the triol **44** in 71% yield (four steps).

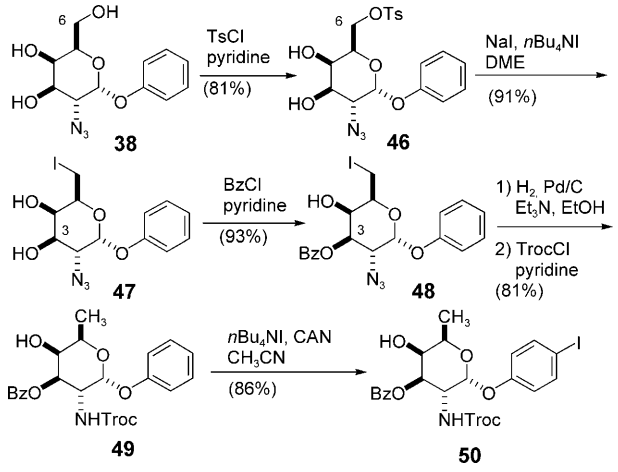



Scheme 11. Synthesis of phenyl 2-amino-α-D-glycopyranoside **44** from D-glucosamine hydrochloride.

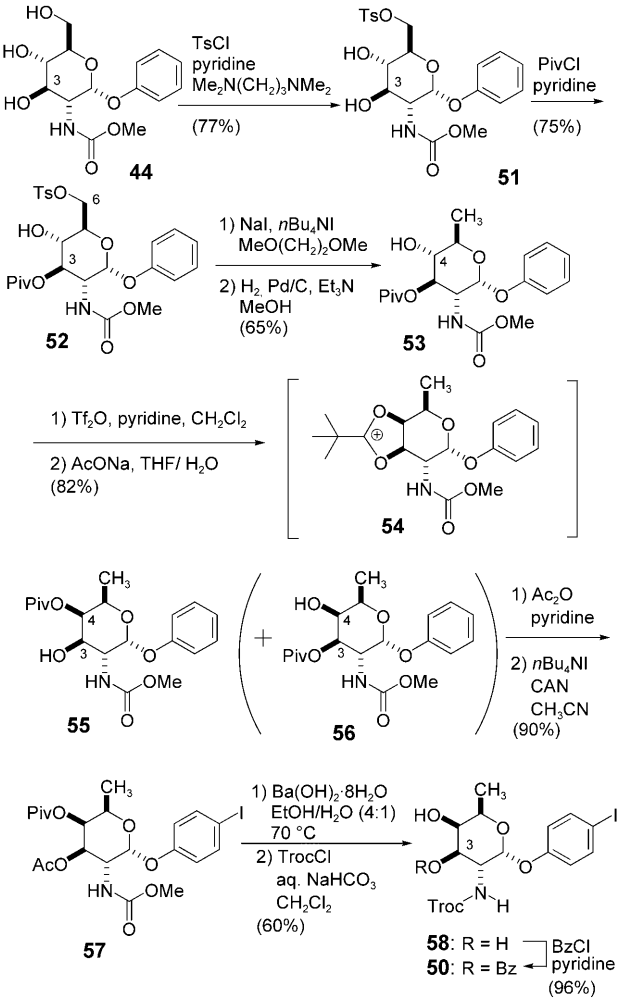
**Synthesis of the right-hand amino sugar 9**: A synthetic route for the preparation of subunit **A**, by starting from galactose derivative **38**, was explored (Scheme 12). Tosylation of the primary C-6 hydroxyl in **38** was accomplished by using the conventional method employing TsCl in pyridine. A subsequent displacement reaction of the resulting tosylate **46** with sodium iodide and tetrabutylammonium iodide ($nBu_4NI$) in refluxing DME provided the iodide **47** in 73% yield (two steps). After selective protection of the equatorial C-3 hydroxy group in **47** as a benzoate (benzoyl chloride, pyridine), reductive hydrogenolysis of the iodide **48** was carried out with concomitant reduction of the azide group ($H_2$, 5% Pd/C, $Et_3N$). It should be noted that while the use of AcOEt or MeOH as solvents gave poor results, the reduction proceeded smoothly in EtOH. Subsequent protection of the resulting amine as a Troc-carbamate (TrocCl, pyridine) gave rise to the 6-deoxy-pyranose **49** in 81% yield (two steps). Finally, iodination of **49** with CAN and $nBu_4NI$ in acetonitrile at 70°C furnished the iodophenyl galactoside **50** (86%),[37] a synthetic equivalent of subunit **A**.

A second route for the synthesis of subunit **A** was developed (Scheme 13). The initial step in this sequence, which




Scheme 12. Synthesis of subunit **A** from D-galactose derivative **38**. CAN: ceric ammonium nitrate; DME: dimethoxyethane; TsCl: *p*-toluenesulfonyl chloride.



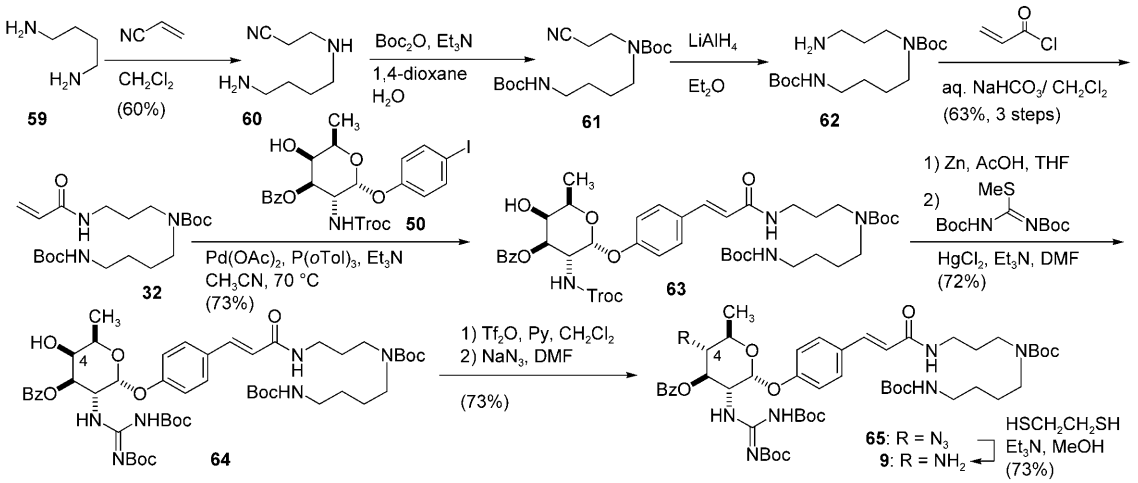

Scheme 13. A second approach to the synthesis of subunit **A** (**50**) from D-glucosamine derivative **44**. Tf: trifluoromethanesulfonyl.

involves selective tosylation of the primary hydroxyl in **44**, was unexpectedly difficult. Since this reaction, when using standard conditions (TsCl, pyridine), was sluggish and resulted in low conversion and poor yield, the diamine-catalyzed tosylation method developed by Tanabe was investigated.[38] Although original protocols by using toluene or acetonitrile as the solvent were problematic due to the poor solubility of the triol **44**, employment of pyridine as a solvent led to the smooth reaction of **44** with TsCl in the presence of one equivalent of tetramethyl-1,3-diaminopropane to afford the tosylate **51** in 77% yield. As a consequence of the lower reactivity of C-4 hydroxy groups in pynanosides that have the *gluco* configuration,[39] selective protection of the C-3 hydroxyl in the diol **51** could be carried out by treatment with pivaloyl chloride in pyridine to afford **52** in 75% yield.

Deoxygenation of the C-6 hydroxy group was then accomplished by using reaction conditions similar to those shown in Scheme 12 to afford the 6-deoxypyranose **53** in 65% yield (two steps). Inversion of the C-4 hydroxyl stereochemistry in **53** was accomplished by using an intramolecular displacement reaction of the triflate with the carbonyl oxygen atom in the pivalate group at C-3, which involves the dioxolenium-ion intermediate **54**.[40] Following this strategy, the triflate was prepared by using the standard method ($Tf_2O$, pyridine, $CH_2Cl_2$). Unfortunately, the previously described solvolytic conditions, which involve addition of water to the reaction mixture followed by heating, was complicated by hydrolysis of the phenyl glycoside. An exploration of this process revealed that treatment of triflate with sodium acetate in a mixture of THF and $H_2O$ (4:1) at room temperature effectively promoted formation of the pivaloyl-group migration product **55** (82%) along with a minor amount of the C-3 pivalate **56** (ca. 3%). The small $J_{3,4}$ value of 2.0 Hz observed in the $^1$H NMR spectrum of **55** is consistent with the assignment of axial stereochemistry of the C-4 hydroxyl. After protection of the hydroxy group in **55** as an acetate, iodination of the phenyl group as described before (Scheme 12) provided the iodophenyl glycopyranoside **57** in 90% yield. It should be noted that the yield of this iodination reaction was considerably lower (46%) when the C-4 hydroxyl unprotected substrate was used. Removal of the acetyl, pivaloyl, and *N*-methoxycarbonyl groups in **57** with barium hydroxide in a mixture of ethanol and $H_2O$ (4:1) at 70°C gave the amino triol, which was then subjected to Schotten–Baumann reaction conditions (TrocCl, aq. $NaHCO_3/CH_2Cl_2$) to furnish the Troc-carbamate **58** in 60% yield. Finally, selective protection of the equatorial C-3 hydroxyl in **58** as a benzoate (benzoyl chloride, pyridine) then delivered subunit **A** (**50**, 96%), which is identical to the product synthesized earlier in Scheme 12.

**Synthesis of the right-hand amino sugar 9**: With sufficient quantities of iodophenyl galactoside **50** in hand, we embarked on a study of the Heck–Mizoroki reaction of **50** with acryl amide **32** (Scheme 14). The preparation of **32** began with conjugate addition of 1,4-diaminobutane to acrylonitrile (60%).[41] Protection of the amine moieties in **60** as Boc-carbamates followed by reduction of the nitrile **61** with

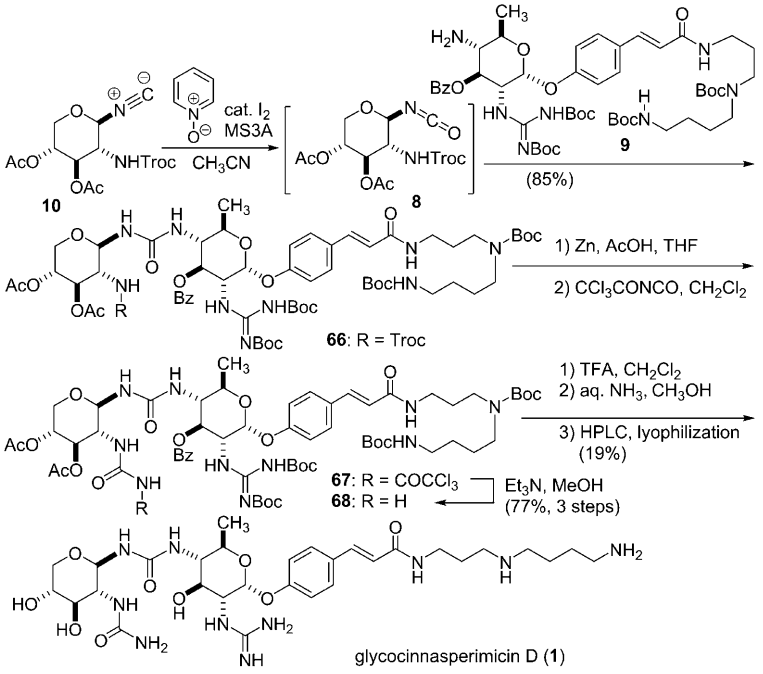
  



Scheme 14. Synthesis of the right-hand amino sugar **9**.

lithium aluminum hydride ($LiAlH_4$) gave rise to the protected spermidine **62**, which was then treated with acryl chloride under Schotten–Baumann conditions to afford the acryl amide **32** in 63% yield (three steps). As foreshadowed by the successful model experiments,[26] the Heck–Mizoroki reaction enabled the uneventful construction of the right core structure of glycocinnasperimicin D. Thus, joining iodophenyl glycoside **50** and acryl amide **32** in the presence of palladium catalyst ($Pd(OAc)_2$ (0.19 equiv), $P(oTol)_3$ (0.33 equiv), $Et_3N$, $CH_3CN$, 70°C) led to the production of the cinnamoyl galactoside **63** in 73% yield. The amine, formed by deprotection of the Troc-carbamate in **63** (Zn, AcOH, THF), was treated with *N*,*N*-di-(*tert*-butoxycarbonyl)-(*S*)-methyl isothiourea and mercuric chloride in DMF to provide bis(Boc-protected) guanidine **64** in 72% yield (two steps).[42] Finally, the C-4 amino group was introduced by using a $S_N2$ displacement reaction of the triflate, prepared from **64** by the standard method ($Tf_2O$, pyridine, $CH_2Cl_2$) with sodium azide in DMF to furnish **65** in 73% yield (two steps). A number of methods, involving the use of $PPh_3$, $SnCl_2$, $nBu_3SnH$, or $H_2$/Pd/C, were applied for the reduction of the azide **65**, but none led to formation of the desired product. However, treatment of **65** with propanedithiol and triethylamine in MeOH[43] successfully yielded the desired left-hand amino sugar **9** in 73% yield.

**Completion of the total synthesis of glycocinnasperimicin D**: With the fully elaborated amino sugars **9** and **10** in hand, we were well positioned to attempt the critical coupling reaction of the two intermediates (Scheme 15). In the event, oxidation of the isonitrile **10** with pyridine *N*-oxide in the presence of a catalytic amount of iodine and MS3A in acetonitrile generated the isocyanate **8**, which was subsequently treated with amino sugar **9**. The high reactivity associated with the glycosyl isocyanate **8** was manifested in this crucial coupling reaction; as a result, the stereochemically congested 4-amino-pyranose **9** reacted smoothly with isocyanate **8** to furnish the urea glycoside **66** in good yield (85%).



Scheme 15. Completion of the total synthesis of glycocinnasperimicin D.

Installation of the urea group on the left-hand amino sugar unit in **66** was then carried out by using three operations involving 1) deprotection of the Troc group in **66** (Zn, AcOH, THF), 2) treatment of the resulting amine with trichloroacetyl isocyanate ($CCl_3CONCO$, $CH_2Cl_2$), and 3) methanolysis ($Et_3N$, MeOH) of the trichloroacetyl urea **67** giving rise to the fully protected glycocinnasperimicin D **68** in a 77% overall yield.

All that remained to complete the total synthesis was to remove the seven protecting groups present in **68**. Observations made in studies of a model compound indicated that removal of Boc protection followed by hydrolysis of acetyl and benzoyl groups would be an optimal deprotection protocol.[44] Accordingly, the Boc group was removed by exposure of **68** to TFA in $CH_2Cl_2$. Subsequent cleavage of the acetyl and benzoyl groups was accomplished by treatment with aqueous ammonia in methanol at 85°C in a sealed tube overnight. An extended reaction time and an elevated temperature were needed for this process to remove the obstinate benzoyl group. Lyophilization of the reaction mixture furnished a residue that was analyzed by $^1$H NMR spectroscopy. While we were excited to find resonances corresponding to glycocinnasperimicin D in the crude mixture, several riddle signals were noted in the region of $\delta = 5$–8 ppm (indicated by arrows in Figure 1). To our surprise, these weak resonaces are also observed in the reported $^1$H NMR spectrum of the natural product.[2]

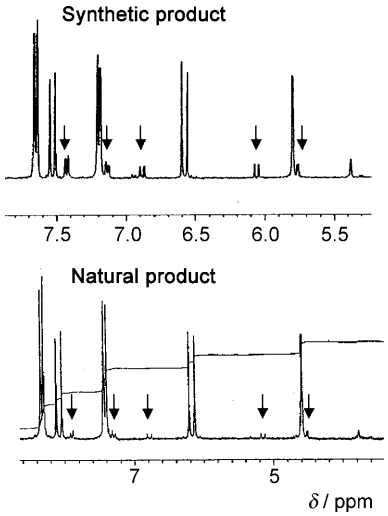
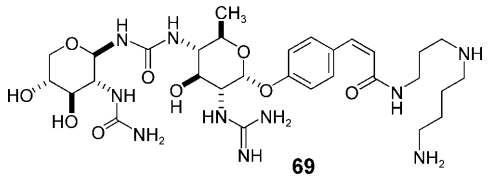
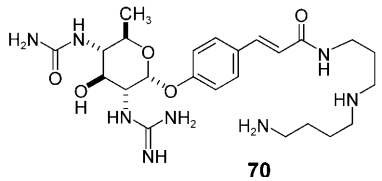



Figure 1. $^1$H NMR spectra of synthetic and natural glycocinnasperimicin D.

Extensive analysis of the crude product mixture unraveled the following key clues; the puzzling small peaks in the $^1$H NMR spectra were finally worked out by analysis of the coupling constants of the weak alkene proton resonances, which have a *J* value of 12.5 Hz. This observation suggested that the photochemically induced olefin isomerization of the *trans*-cinnamoyl moiety took place during and/or after removal of the protecting groups to from the *cis*-isomer **69**. In addition, ESI Mass spectrometric analysis of the crude product mixture revealed the presence of a peak at 521, which is not observed in the spectrum of the natural product. This result indicates that partial hydrolysis of urea glycoside occurred to form product **70**, arising from the right-hand amino sugar.[45] Finally, reverse-phase HPLC analysis of the crude product mixture showed the presence of four bands, comprised by a major peak associated with glycocinnasperimicin D and three minor peaks attributed to the *cis*-isomer **69**, an unidentified substance, and the hydrolyzed products corresponding to **70** and its *cis*-alkene isomer.



**69**



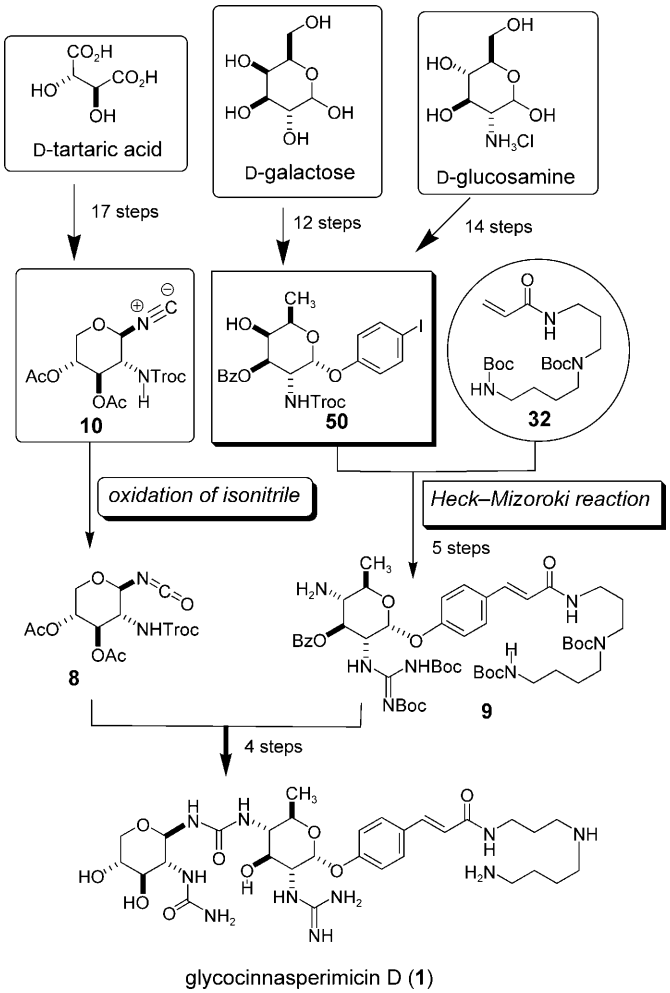
**70**

Although a detailed understanding of the facility of the *cis*/*trans* isomerization of the cinnamoyl group that took place during or/after deprotection is not currently in hand,[46] this obstacle could be circumvented by implementation of a simple experimental technique. The deprotection and purification steps were carried out in glass apparatus wrapped with aluminum foil. After an exhaustive and labor-intensive task of purification, which led to loss of a considerable amount of our precious compound, we were able to isolate pure compound **1** as its TFA salt. Careful treatment of this TFA salt in $H_2O$ with Amberlite IRA 410 ion-exchange resin ($Cl^-$ form) followed by filtration and lyophilization afforded the glycocinnasperimicin D (**1**) as its hydrochloride salt (19% yield).[47] The spectroscopic data ($^1$H NMR in $D_2O$ at 40°C and $^{13}$C NMR spectra), TLC behavior, and antimicrobial activity of the synthetic material were in good agreement with those of naturally occurring glycocinnasperimicin D.

## Conclusions

The total synthesis of glycocinnasperimicin D has been accomplished with a highly convergent, three-component coupling strategy (Scheme 16). The key features in the sequence involve a Heck–Mizoroki reaction by using the iodophenyl glycoside **50** and acryl amide **32**, and the construction of the urea glycoside employing glycosyl isocyanate **8** and amino sugar **9**. Oxidation of isonitrile **10** generated the reactive glycosyl isocyanate **8**, which displayed excellent reactivity in its coupling event. Several synthetic roadblocks, encoun-




Scheme 16. Summary of the three-component coupling route to glycocinnasperimicin D.

tered in this synthetic route, have led to the development of new methodologies including the α-selective, Lewis acid catalyzed phenyl glycosylation process and an interesting procedure for acetonide deprotection without affecting the silyl ethers.

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[1] a) K. Dobashi, K. Nagaoka, Y. Watanabe, M. Nishida, M. Hamada, H. Naganawa, T. Takita, T. Takeuchi, H. Umezawa, *J. Antibiot.* **1985**, 1166–1170; b) K. Dobashi, PhD Thesis, Kyushu University (Japan), **1990**.

[2] a) G. A. Ellestad, D. B. Cosulich, R. W. Broschard, J. H. Martin, M. P. Kunstmann, G. O. Morton, J. E. Lancaster, W. Fulmor, F. M. Lovell, *J. Am. Chem. Soc.* **1978**, *100*, 2515–2524; b) H. D. Tresner, J. H. Korshalla, A. A. Fantini, J. D. Korshalla, J. P. Kirby, J. J. Goodman, R. A. Kele, A. J. Shay, D. B. Borders, *J. Antibiot.* **1978**, *31*, 394–397; c) J. H. Martin, M. P. Kunstmann, F. Barbatschi, M. Hertz, G. A. Ellestad, M. Dann, G. S. Reiden, A. C. Dornbushi, N. A. Kuck, *J. Antibiot.* **1978**, *31*, 398–404; d) S. H. L. Chiu, R. Fiala, R. Kennett, L. Wozniak, M. W. Bullock, *J. Antibiot.* **1984**, 1000–1006; e) S. H. L. Chiu, R. Fiala, M. W. Bullock, *J. Antibiot.* **1984**, *37*, 1079–1081; f) H.-R. Tsou, R. R. Fiala, P. C. Mowery, M. W. Bullock, *J. Antibiot.* **1984**, 1382–1387. Biological activity; g) M. Greenstein, J. L. Speth, W. M. Maise, *Antimicrob. Agents Chemother.* **1981**, *20*, 425-432; h) M. S. Osburne, W. M. Maiese, M. Greenstein, *Antimicrob. Agents Chemother.* **1990**, *34*, 1450- 1452.

[3] R. H. Chen, D. N. Whittern, A. M. Buko, J. B. McAlpine, *J. Antibiot.* **1989**, 533–537.

[4] Private communication from Dr. H. Naganawa and Dr. Y. Takahashi.

[5] For synthetic studies of glycocinnamoylspermidine antibiotics, see: a) K. Araki, K. Miyazawa, J. Yoshimura, *Tetrahedron Lett.* **1982**, *23*, 1705–1708; b) H. Hashimoto, K. Araki, H. Hashimoto, J. Yoshimura, *Carbohydr. Res.* **1982**, *99*, 59–69; c) K. Araki, H. Hashimoto, J. Yoshimura, *Carbohydr. Res.* **1982**, *109*, 143–160; d) K. Araki, M. Kawai, K. Miyazawa, H. Hashimoto, J. Yoshimura, *Bull. Chem. Soc. Jpn.* **1986**, *59*, 3137–3143.

[6] This work has been communicated in a preliminary form: T. Nishiyama, M. Isobe, Y. Ichikawa, *Angew. Chem.* **2005**, *117*, 4446–4449; *Angew. Chem. Int. Ed.* **2005**, *44*, 4372–4375.

[7] Taken in part from: T. Nishiyama, PhD Thesis, Nagoya University (Japan), **2005**.

[8] a) Y. Ichikawa, T. Nishiyama, M. Isobe, *Synlett* **2000**, 1253–1256; b) Y. Ichikawa, T. Nishiyama, M. Isobe, *J. Org. Chem.* **2001**, *66*, 4200–4205.

[9] a) Y. Ichikawa, T. Ito, T. Nishiyama, M. Isobe, *Synlett* **2003**, 1034–1036; b) Y. Ichikawa, T. Ito, M. Isobe, *Chem. Eur. J.* **2005**, *11*, 1949–1957.

[10] For the preparation of **18**: E. A. Mash, K. A. Nelson, S. V. Deusen, S. B. Hemperly, *Org. Synth. Coll. Vol. 8* **1993**, 155–161; for the synthesis of **16**: K. Ucihda, K. Kato, H. Akita, *Synthesis* **1999**, 1678–1686; see also: H. Iida, N. Yamazaki, C. Kibayashi, *J. Org. Chem.* **1987**, *52*, 3337–3342.

[11] R. E. Ireland, W. Norbeck, *J. Org. Chem.* **1985**, *50*, 2198–2200.

[12] M. Frigerio, M. Santagostino, S. Sputore, G. Palmisano, *J. Org. Chem.* **1995**, *60*, 7272–7276.

[13] J. Einhorn, C. Einhorn, F. Ratajczak, J.-L. Pierre, *J. Org. Chem.* **1996**, *61*, 7452–7454.

[14] K. Soai, A. Ookawa, T. Kaba, K. Ogawa, *J. Am. Chem. Soc.* **1987**, *109*, 7111–7115.

[15] W. A. Nugent, *Chem. Commun.* **1999**, 1369–1370.

[16] Recently, a simple three-step synthesis of MIB has appeared: Y. K. Chen, S.-J. Jeon, P. J. Walsh, W. A. Nugent, *Org. Synth.* **2005**, *82*, 87–92.

[17] For representative examples of the allyl cyanate-to-isocyanate rearrangement, see: a) C. Christophersen, A. Holm, *Acta Chem. Scand.* **1970**, *24*, 1512–1526; b) L. E. Overman, M. Kakimoto, *J. Org. Chem.* **1978**, *43*, 4564–4567; c) K. Banert, *Angew. Chem.* **1992**, *104*, 865–867; *Angew. Chem. Int. Ed. Engl.* **1992**, *31*, 866–868.

[18] a) Y. Ichikawa, *Synlett* **1991**, 238–240; b) Y. Ichikawa, *Synlett* **2007**, 2927–2936.

[19] K. S. Kim, Y. H. Song, B. H. Lee, C. S. Hahn, *J. Org. Chem.* **1986**, *51*, 404–407.

[20] Deprotection of either benzylidene acetal or acetonide in the presence of silyl protecting groups with ethanethiol and zinc triflate in $CH_2Cl_2$ has been reported: a) K. C. Nicolaou, M. E. Bunnage, D. G. McGarry, S. Shi, P. K. Somers, P. A. Wallace, X-J. Chu, K. A. Agrios, J. L. Gunzner, Z. Yang, *Chem. Eur. J.* **1999**, *5*, 599–617; b) K. Fuji-

wara, S. Yoshimoto, A. Takizawa, S, Souma, H. Mishima, A. Murai, H. Kawai, T. Suzuki, *Tetrahedron Lett.* **2005**, *46*, 6819–6822; in our case, use of $CH_2Cl_2$ as a solvent resulted in slow and incomplete deprotection accompanied by recovery of the starting material.

[21] A. B. Smith III, G. R. Ott, *J. Am. Chem. Soc.* **1996**, *118*, 13095–13096.

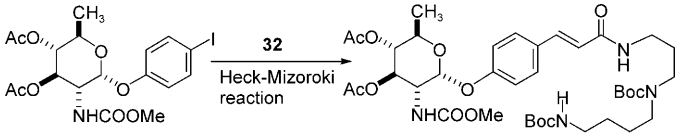
[22] D. A. Evans, S. W. Kaldor, T. K. Jones, J. Clardy, T. J. Stout, *J. Am. Chem. Soc.* **1990**, *112*, 7001–7031.

[23] H. Paulsen, Z. Gyorgydeak, M. Friedman, *Chem. Ber.* **1974**, *107*, 1568–1578.

[24] Y. Ichikawa, *J. Chem. Soc. Perkin Trans. 1* **1992**, 2135–2139.

[25] K. Oyama, T. Kondo, *Synlett* **1999**, 1627–1629, and references therein; see also reference [6c].

[26] Model studies by using iodophenyl glycoside **I** and acryamide **32** provided **II** in 75% yield, see: T. Nishiyama, Y. Ichikawa, M. Isobe, *Synlett* **2004**, 89–92.




I II

[27] For a recent example of C-6 deoxygenation by using catalytic hydogenolysis of 6-idodo-glycopyranose, see: A. Medgyes, E. Farkas, A. Liptak, V. Pozsgay, *Tetrahedron* **1997**, *53*, 4159–4178.

[28] a) W. E. Trevelyan, *Carbohydr. Res.* **1966**, *2*, 418–420; b) J. L. Bose, T. R. Ingle, *Chem. Ind.* **1967**, 1451; c) T. D. Audichya, T. R. Ingle, J. L. Bose, *Indian J. Chem.* **1971**, *9*, 315–317; d) T. D. Audichya, T. R. Ingle, J. L. Bose, *Indian J. Chem.* **1973**, *11*, 704–705; e) K. Honma, K. Nakazima, T. Uematsu, A. Hamada, *Chem. Pharm. Bull.* **1976**, *24*, 394–399.

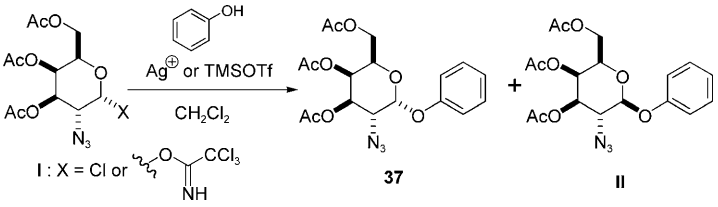
[29] a) R. U. Lemieux, W. P. Shyluk, *Can. J. Chem.* **1953**, *31*, 528–535; b) E. Smits, J. B. F. N. Engberts, R. M. Kellogg, H. A. van Doren, *J. Chem. Soc. Perkin Trans. 1* **1996**, 2873–2877.

[30] C. Murakata, T. Ogawa, *Carbohydr. Res.* **1992**, *235*, 95–114.

[31] During the preparation of this manuscript, an extensive literature search uncovered the following reference: T. R. Ingle, J. L. Bose, *Carbohydr. Res.* **1970**, *12*, 459–462. In this report, Bose described the stannic chloride catalyzed glycosylation reaction of phenol with penta-*O*-acetyl-β-D-galactopyranose. In this case, α- and β-anomers were isolated in 40 and 12% yields, respectively. In reference [28b], Bose reported the reaction of β-D-galactopyranose pentaacetate with phenol in the presence of stannic chloride to afford a mixture of phenyl α- and β-galactopyranoside in a 6:4 ratio.

[32] 2-Azido-D-galactose derivative **36** was prepared from D-galactal by using a two-step route involving azido nitration of D-galactal followed by acetolysis by using the method reported by Lemieux, see: R. U. Lemieux, R. M. Ratcliffe, *Can. J. Chem.* **1979**, *57*, 1244–1251.

[33] Initial efforts to prepare phenyl α-D-galactopyranoside **37** focused on glycosylation of phenol with 2-azido galactose derivatives **I**, because an azide group is known to show a small neighboring effect in the Konig–Knorr and the imidate glycosylation reactions. Unfortunately, this approach resulted in only moderate selectivity (α/β = 7:3), which is not amenable to a large-scale synthesis. For the pioneering work on glycosylation by using 2-azide glycopyranose, see: H. Paulsen, *Angew. Chem.* **1982**, *94*, 184–201; *Angew. Chem. Int. Ed. Engl.* **1982**, *21*, 155–224.




[34] a) S. Kusumoto, H. Yoshimura, M. Imoto, T. Shimamoto, T. Shiba, *Tetrahedron Lett.* **1985**, *26*, 909–912; b) P. Boullanger, J. Banoub, G. Descotes, *Can. J. Chem.* **1987**, *65*, 1343–1348.

[35] Tetra-*O*-acetyl-glucosamine **39** was prepared from glucosamine hydrochloride **45** in three steps, see reference [27].

[36] W. Dullenkopf, J. C. Castro-Palomino, L. Manzoni, R. R. Schmidt, *Carbohydr. Res.* **1996**, *296*, 135–147.

[37] T. Sugiyama, *Bull. Chem. Soc. Jpn.* **1981**, *54*, 2847–2848.

[38] Y. Yoshida, K. Shimonishi, Y. Sakakura, S. Okada, N. Aso, Y. Tanabe, *Synthesis* **1999**, 1633–1636.

[39] a) J. M. Williams, A. C. Richardson, *Tetrahedron* **1967**, *23*, 1369–1378; b) O. Varela, D. Cicero, R. M. de Lederkremer, *J. Org. Chem.* **1989**, *54*, 1884–1890; c) C. Gallo-Rodriguez, O. Varela, R. M. de Lederkremer, *J. Org. Chem.* **1996**, *61*, 1886–1889.

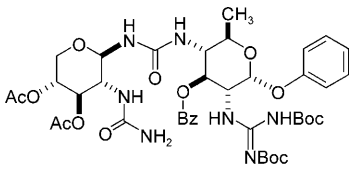
[40] a) R. W. Binkley, M. R. Sivik, *J. Org. Chem.* **1986**, *51*, 2619–2621; b) R. W. Binkley, *J. Org. Chem.* **1991**, *56*, 3892–3896; c) V. R. Bouvet, R. N. Ben, *J. Org. Chem.* **2006**, *71*, 3619–3622.

[41] a) M. Israel, J. S. Rosenfield, E. J. Modest, *J. Med. Chem.* **1964**, *7*, 710–716; b) M. Humora, J. Quick, *J. Org. Chem.* **1979**, *44*, 1166–1168.

[42] Z. X. Guo, A. N. Cammidge, D. Horwell, *Synth. Commun.* **2000**, *30*, 2933–2943.

[43] H. Bayley, D. N. Stadring, J. R. Knowles, *Tetrahedron Lett.* **1978**, *19*, 3633–3634.

[44] The model compound shown below was employed to examine the deprotection conditions.




model compound for deprotection

[45] In our exploration to find deprotection conditions with the model compound shown in reference [44], hydrolysis of urea glycoside was never observed under similar conditions to those used in Scheme 15.

[46] After completion of the synthetic efforts, we noticed the photolysis of a mixture of LL-BM123γ1 and γ2, see: J. J. Hlavka, P. Bitha, J. Boothe, T. Fields, *J. Antibiot.* **1978**, *31*, 477–479.

[47] This yield is calculated on the basis of the trihydrochloride of **1**. The previous value (23%) in reference [6] was estimated from the monohydrochloride.

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