Structural Determinants of Selective Thyromimetics

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The thyromimetic GC-1 shows a preference for binding the β form of the thyroid hormone receptor (TR). GC-1 was designed as an analogue of the thyromimetic DIMIT, which has a lower affinity for TR and is not selective. GC-1 has a methylene group linking its two aromatic rings and an oxyacetic acid polar side chain, while DIMIT has an ether oxygen linking its aromatic rings and an L-alanine polar side chain. The structural features of GC-1 that confer its greater affinity and selectivity compared to DIMIT were analyzed with the preparation of analogues that bear only one of their two different structural features. The analogue of GC-1 with a biaryl ether has selectivity comparable to that of GC-1, while the analogue of DIMIT with a methylene group linking its aromatic rings is only slightly selective. These results demonstrate that the oxyacetic acid side chain of GC-1 is critical in conferring TR-β selectivity.

Introduction

Thyroid hormone is a classical endocrine hormone that plays important roles in the development and regulation of homeostasis.[1] Thyroxine (T_4, **2**, Figure 1) is the major secreted form that is enzymatically deiodinated in peripheral tissues to the more active form 3,5,3′-triiodo-L-thyronine (T_3, **1**, Figure 1). Most of thyroid hormone's actions are believed to be mediated through nuclear thyroid hormone receptors (TRs) that regulate transcription of target genes either positively or negatively in response to hormone binding.[2] The two genes for TR (TRα and TRβ) are each expressed in most tissues in two alternatively spliced variants: TRα_1 and TRα_2; TRβ_1 and TRβ_2.[3] All these forms of the receptor except TRα_2 are activated by T_3, which binds the ligand binding domain (LBD) of the receptor with high affinity. While TRα and TRβ are widely expressed, they have distinct patterns of expression.[3] Mice deficient in either TRα or TRβ show different phenotypes,[4−8] suggesting that each receptor isoform has significantly different regulatory roles.

T_3 binds both TRα_1 and TRβ_1 with near-equal affinity.[9] Synthetic ligands that bind preferentially to only one isoform could be powerful tools for studying each isoform's physiological roles. T_3 is a potent serum cholesterol lowering agent;[10−12] however, its therapeutic use in treating hypercholesterolemia is prevented by various side effects. Tachycardia, increased cardiac output, and the increased risk of cardiac arrhythmia[13,14] are the most serious side effects that arise from an excess of T_3. Mice deficient in TRα_1 have a 20% lower heart rate than wild-type mice, and this does not increase to wild-type levels with T_3 treatment.[15] TRβ is the predominant isoform expressed in the liver,[16] and upon T_3 treatment, mice deficient in TRβ do not up-regulate CYP7A, the rate-limiting enzyme for the conversion of cholesterol to bile acids.[17] These results suggest that TRβ-selective thyromimetics with the expected tissue-selective action could function as useful therapeutic agents.

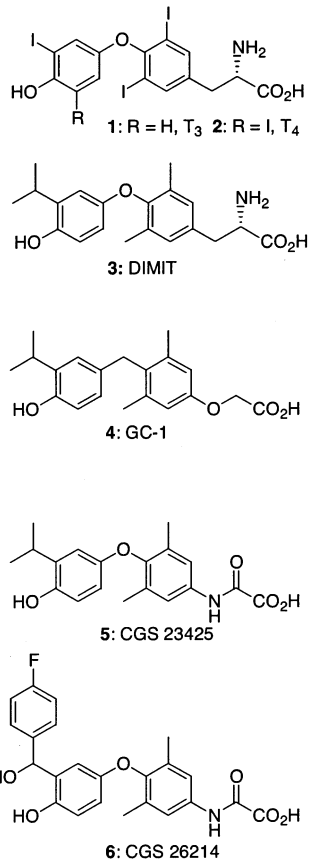

Figure 1. Structures of thyroid hormones and thyromimetics.

A number of thyromimetics have been reported in recent years to have tissue-selective action. Among these compounds are CGS 23425[18] (**5**, Figure 1), CGS 26214[19] (**6**, Figure 1), and GC-1[9,20] (**4**, Figure 1). CGS 23425 has

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a structure resembling the nonselective thyromimetic 3,5-dimethyl-3′-isopropyl-L-thyronine[21] (DIMIT, **3**, Figure 1) but with an oxamic acid polar side chain replacing DIMIT's alanine side chain. In hypercholesterolemic rats, CGS 23425 lowers low-density lipoprotein cholesterol by 44% without inducing cardiotoxic effects,[18] demonstrating that the compound is a liver-selective, cardiac-sparing thyromimetic. The selective action of CGS 23425 has been attributed to the preferential activation of TRβ over TRα. In assays of TR-mediated transcription activation from a rat apoAI promoter in HuH-7 cells, CGS 23425 has EC_{50} values of 0.002 nM for hTRβ_1 and 0.1 nM for hTRα_1, a 50-fold preference for activation of TRβ over TRα in this system.[18] It has not been reported whether this difference in potency for the TR isoforms reflects differences in affinity.

CGS 26214 resembles CGS 23425 structurally but has a 4-fluorophenylhydroxymethyl substituent at the 5′ position. When tested in rats and dogs, CGS 26214 is a potent serum cholesterol lowering agent free of cardiovascular effects. In nuclear binding assays using intact neonatal rat cardiac myocytes, CGS 26214 has an apparent 100-fold lower affinity than T_3, while the affinities of the ligands are comparable when the assay is performed using cultured liver (HepG2) cells.[19] These results suggest that the tissue-selective thyromimetic effects of CGS 26214 may be due to preferential TR binding in the liver.

The thyromimetic GC-1 was developed in our laboratory and binds hTRβ_1 with an affinity comparable to T_3 but binds hTRα_1 with a 10-fold lower affinity.[9] This 10-fold selectivity and ability to preferentially activate TRβ is reflected in several in vivo studies where it exhibits a distinct subset of physiological responses to T_3 in such TR-mediated processes as tadpole metamorphosis,[22] lipid metabolism,[23] cerebellar development,[24] and adaptive thermogenesis.[25] The structural features of GC-1 that confer its TRβ selectivity are not well defined. GC-1 was designed as a synthetically accessible thyromimetic that would serve as a platform for the development of TR antagonists. Its design was based on DIMIT, which binds TR with approximately 100-fold lower affinity than T_3.[9,26] For ease of synthesis, GC-1 was designed with a methylene group joining the two aromatic rings, instead of the ether oxygen of DIMIT and T_3. The α-amino group of T_3 is not required for high affinity binding to TR,[26] and the oxyacetic acid polar side chain was intended as a readily synthesized isostere of propionic acid. Comparing the structures and activities of GC-1 and DIMIT, one concludes that the structural changes introduced by the methylene bridge or oxyacetic acid side chain are responsible for the GC-1's higher affinity and selectivity.

Crystallographic studies have shown that only one amino acid residue (Ser277 in hTRα_1, Asn331 in hTRβ_1) is different in the ligand binding cavity of the two receptors.[4,27] This residue does not directly contact the ligand, but a comparison of the cocrystal structures of TRβ-LBD with GC-1 bound and with T_3 bound shows that Asn331 participates in a hydrogen bonding network with neighboring arginine residues that is arranged differently depending on the ligand present.[28] The role of Asn331/Ser277 in GC-1's TRβ selectivity has been confirmed with mutagenesis experiments showing that TRβ Asn331Ser has reduced affinity for GC-1 while TRα Ser277Asn has increased affinity.[28]

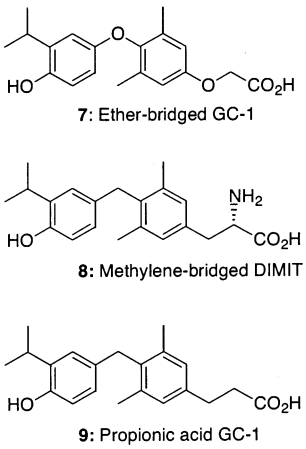


Figure 2. Structures of target compounds to study effects of methylene bridge and oxyacetic acid side chain on TR selectivity.

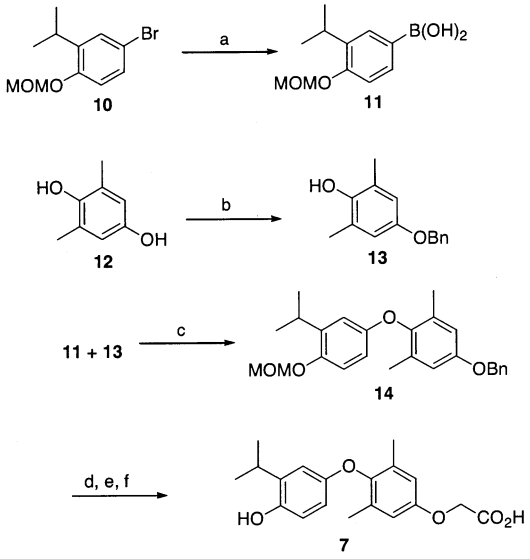
When the structures of T_3, DIMIT, GC-1, CGS 23425, and CGS 26214 are compared, a correlation emerges between selective action and the identity of the polar side chain. DIMIT and T_3 have an alanine polar side chain and are not selective, while the TR ligands with oxyacetic or oxamic acid side chains have selective action in vivo. For GC-1, this selective action correlates with its preferential binding to TRβ.

Seeking to understand better the specific roles of GC-1's methylene bridge and oxyacetic acid side chain in its TRβ selectivity, we synthesized analogues of GC-1 and DIMIT bearing only one of the two different structural features. By comparison of the binding affinities and selectivities of methylene-bridged DIMIT (**8**, Figure 2) and GC-1 ether (**7**, Figure 2) to those of DIMIT and GC-1, the effects of the ether to methylene and the alanine to oxyacetic acid substitutions can be evaluated individually. The methylene-bridged analogue of T_3 has a 2.5-fold higher affinity for TR than T_3; however, this result was from a binding assay based on a nuclear extract, making it difficult to evaluate isoform-selective binding.[26] Additionally, the propionic acid side chain analogue of GC-1 (**9**, Figure 2) was prepared to assess the effect of the ether oxygen in the polar side chain of GC-1. To ensure the most valid comparisons between thyromimetic ligands, i.e., those with small perturbations, the study was limited to GC-1, DIMIT, and their related analogues.

Results

Synthesis of GC-1 Ether. The synthesis of the biaryl ether analogue of GC-1 was achieved in seven linear steps starting from 2-isopropylphenol and 2,6-dimethylhydroquinone. The key step for joining the two aromatic rings in a biaryl ether linkage from arylboronic acid and phenolic intermediates was achieved using the copper(II) catalyzed coupling reaction developed by Evans[29] (Scheme 1). 4-Benzyloxy-2,6-dimethylphenol (**13**) was prepared in low yield from the alkylation of 2,6-dimethylhydroquinone by a Williamson ether synthesis. The site of alkylation was assigned on the basis of ^{1}H NMR chemical shift perturbations of the 3,5-protons and the 2,6-methyl protons compared to the

Scheme 1. Preparation of the Ether-Bridged Analogue of GC-1[a]



[a] Reagents and conditions: (a) (i) *n*-BuLi, THF, −78 °C, (ii) $B(OMe)_3$, THF, −78 °C to room temp, (iii) H_3O^+; (b) BnBr, K_2CO_3, DMF, 9%; (c) $Cu(OAc)_2$ Et_3N, CH_2Cl_2; (d) H_2, Pd/C, EtOAc, 27% from **13**; (e) methyl bromoacetate, Cs_2CO_3, DMF, 81%; (f) LiOH, H_2O, MeOH, 30%.

corresponding chemical shifts in the starting material and the other alkylated products.

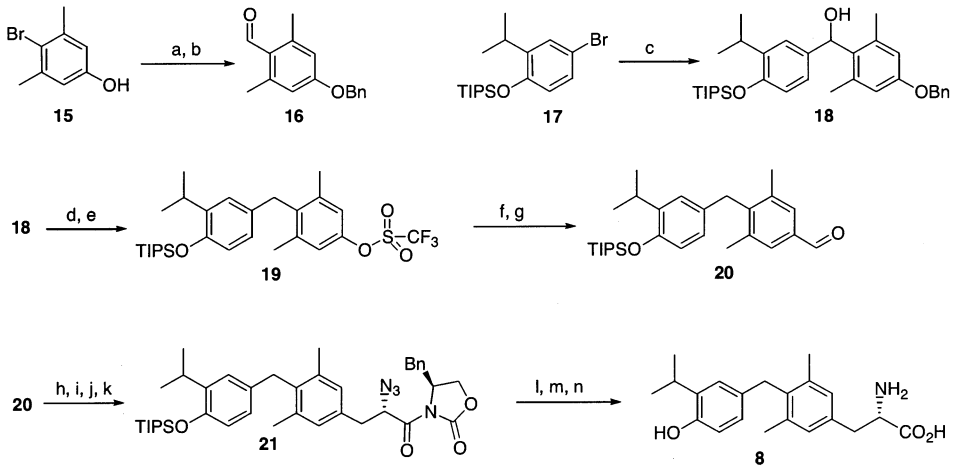
The arylboronic acid intermediate **11** for the coupling reaction was prepared from the para bromination of 2-isopropylphenol, followed by the protection of the phenol as a methoxymethyl (MOM) ether. The aryl bromide, **10**,[20] was converted to the arylboronic acid with the sequential addition of *n*-butyllithium and trimethylborate, followed by hydrolysis with aqueous HCl. The coupling of **11** and **13** was effected with copper(II) acetate and triethylamine as the base. The oxyacetic acid polar side chain was installed by deprotection of the 1-hydroxyl of **14** with catalytic hydrogenation, alkylation with methyl bromoacetate, and hydrolysis of the ester with lithium hydroxide. The MOM group protecting the 4′-hydroxyl was unexpectedly cleaved under saponification conditions, providing **7**, the biaryl ether analogue of GC-1.

Synthesis of Methylene-Bridged L-DIMIT. The synthesis of the methylene-bridged analogue of L-DIMIT was achieved in 14 linear steps starting from 4-bromo-2-isopropylphenol and 4-bromo-3,5-dimethylphenol (Scheme 2). The synthesis of the biphenylmethane scaffold employed the same strategy used to make GC-1,[9,20] with the coupling of the aromatic rings effected by the 1,2-addition of an aryllithium intermediate to a benzaldehyde intermediate. The hydroxyl of 4-bromo-3,5-dimethylphenol was protected as a benzyl ether, which was removed during the catalytic hydrogenolysis of the hydroxyl on the bridging carbon of **18**. The L-alanine side chain was synthesized from a propionic acid side chain containing intermediate, using Evans's chiral auxiliary.

4-Benzyloxy-2,6-dimethylbromobenzene was prepared by reaction of 4-bromo-3,5-dimethylphenol (**15**) with benzyl bromide and potassium carbonate in DMF. The benzaldehyde **16** was prepared in the usual fashion by treatment first with *n*-butyllithium and then DMF as a formylating reagent, followed by an aqueous acid workup. The coupling of the rings by the addition of the aryllithium reagent, derived from bromobenzene **17** and *n*-butyllithium, to benzaldehyde **16** proceeded in good yield. Catalytic hydrogenation of **18** yielded a phenolic biarylmethane intermediate, which was reacted with trifluoromethanesulfonic anhydride to produce the aryltrifluoromethanesulfonate (aryltriflate) **19** in high yield. When a Stille coupling with vinyltributylstannane was used, aryltriflate **19** was converted to a styrene intermediate, which was cleaved to the benzaldehyde **20** via ozonolysis and reduction using triphenylphosphine.

The propionic acid side chain was built from the benzaldehyde by a Wittig olefination with benzyl (triphenylphosphoranylidene)acetate, followed by catalytic hydrogenation to reduce the acrylate ester to propionate and hydrogenolyze the benzyl ester. To install the chiral auxiliary (*S*)-4-benzyl-2-oxazolidinone, a mixed anhy-

Scheme 2. Preparation of the Methylene-Bridged Analogue of DIMIT[a]



[a] Reagents and conditions: (a) BnBr, K_2CO_3, DMF, 78%; (b) (i) 1.1 equiv of *n*-BuLi, THF, −78 °C, (ii) DMF, THF, −78 °C, (iii) H_3O^+, 80%; (c) (i) 1.1 equiv of *n*-BuLi, THF, −78 °C, (ii) **16**, THF, −78 °C, 77%; (d) H_2, Pd/C, EtOH/AcOH, 100%; (e) Tf_2O, pyr, DCM, 99%; (f) vinyl-$SnBu_3$, $Cl_2Pd(PPh_3)_2$, LiCl, DMF, 85%; (g) (i) O_3, CH_2Cl_2, −78 °C, (ii) PPh_3, −78 °C to room temp, 86%; (h) benzyl (triphenylphosphoranylidene)acetate, THF, 100%; (i) H_2, Pd/C, EtOAc, 86%; (j) (i) pivaloyl-Cl, Et_3N, THF, (ii) lithium (*S*)-(−)-4-benzyl-2-oxazolidinone, THF, −78 °C, 89%; (k) (i) KHMDS, THF, −78 °C, (ii) trisylazide, THF, −78 °C, (iii) AcOH, 73%; (l) LiOOH, 3:1 THF/H_2O, 83%; (m) 100 equiv $Et_3N{\cdot}3HF$, THF, 44%; (n) H_2, Pd/C, 2:1 AcOH/H_2O, 95%.

Table 1. Binding Affinities of Thyromimetics for TR

compd	K_d(hTRα_1) (nM)	K_d(hTRβ_1) (nM)	selectivity	ΔG_{bind}(hTRα_1) (kcal/mol)	ΔG_{bind}(hTRβ_1) (kcal/mol)
T_3 (**1**)	0.060 ± 0.004	0.087 ± 0.006	1.0 ± 0.2	−12.96 ± 0.04	−12.76 ± 0.04
GC-1 (**4**)	0.66 ± 0.05	0.10 ± 0.01	10 ± 2	−11.64 ± 0.05	−12.71 ± 0.06
ether-bridged GC-1 (**7**)[a]	2.4 ± 0.4	0.36 ± 0.01	10 ± 2	−10.94 ± 0.08	−11.98 ± 0.02
propionic acid GC-1 (**9**)[a]	0.69 ± 0.05	0.22 ± 0.01	4.5 ± 0.6	−11.61 ± 0.04	−12.24 ± 0.04
methylene-bridged DIMIT (**8**)[a]	2.02 ± 0.09	1.26 ± 0.03	2.3 ± 0.3	−11.03 ± 0.03	−11.29 ± 0.01
DIMIT (**3**)	8.5 ± 0.8	6.2 ± 0.2	2.0 ± 0.3	−10.24 ± 0.05	−10.41 ± 0.02

[a] Compound purity >97% by HPLC.

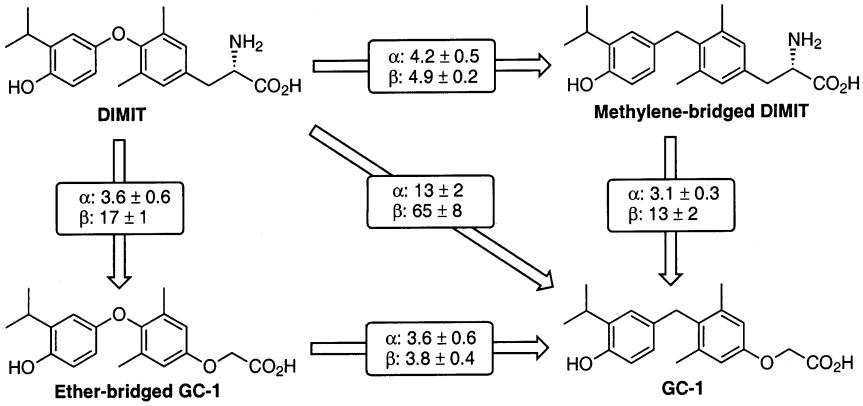
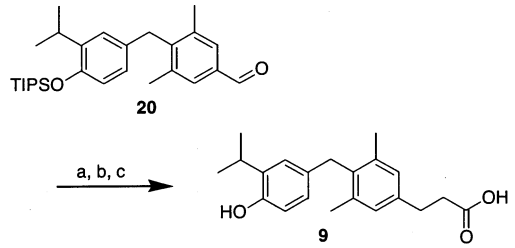


Figure 3. Ether oxygen to methylene and alanine to oxyacetic acid side chain substitutions have distinct independent effects on TR affinity and selectivity. Values indicate fold gain in affinity for TRα and TRβ.

Scheme 3. Preparation of the Propionic Acid Side Chain Analogue of GC-1[a]



[a] Reagents and conditions: (a) benzyl (triphenylphosphoranylidene)acetate, THF, 100%; (b) 100 equiv of $Et_3N{\cdot}3HF$, THF, 88%; (c) H_2, Pd/C, 2:1 AcOH/H_2O, 90%.

dride was generated from the propionic acid intermediate and pivaloyl chloride, which was reacted with the lithium salt of (*S*)-4-benzyl-2-oxazolidinone. The chiral *N*-acyloxazolidinone was deprotonated with potassium bis(trimethylsilyl)amide and quickly reacted with 2,4,6-triisopropylbenzenesulfonylazide, followed rapidly by an acetic acid quench to give the single diastereomer (by ^{1}H and ^{13}C NMR) of the *N*-(α-azidoacyl)oxazolidinone **21**. The chiral auxiliary was removed with lithium peroxide followed by deprotection of the TIPS ether with triethylamine trihydrofluoride. Finally, the α-azide was reduced to the α-amine with catalytic hydrogenation to yield the methylene-bridged analogue of L-DIMIT (**8**).

Synthesis of Propionic Acid Side Chain Analogue of GC-1. The GC-1 analogue with a propionic acid polar side chain (**9**, Scheme 3) was prepared from the benzaldehyde intermediate (**20**, Scheme 2) from the synthesis of methylene-bridged DIMIT. The TIPS protecting group was removed using the standard procedure, and the product was hydrogenated to generate the propionic acid side chain.

TR Binding. The affinities and selectivities of T_3, DIMIT, GC-1, methylene-bridged DIMIT, and GC-1 ether for TRα_1 and TRβ_1 are summarized in Table 1. To ensure a valid comparison of the affinities of different ligands, all the compounds were assayed simultaneously using the same preparation of purified TR. The selectivity was determined by the ratio of K_d(hTRα_1) to K_d(hTRβ_1) and normalized to the ratio of K_d(hTRα_1) to K_d(hTRβ_1) for T_3.

While none of the synthetic thyromimetics, except for GC-1, bind to hTRβ_1 as tightly as T_3, the thyromimetics containing the methylene bridge (GC-1, methylene-bridged DIMIT, and propionic acid GC-1) bind with greater affinities than those containing the ether bridge. The ether to methylene substitution thus results in increased affinity for TR with thyromimetics of the 3,5-dimethyl-3′-isopropyl substitution pattern, as it does with the 3,5,3′-triiodo substitution pattern of T_3. Comparing the affinities and selectivities of DIMIT and T_3 shows that 3,5-dimethyl-3′-isopropyl substitution results in an approximate loss of affinity of 140-fold for hTRα and 70-fold for hTRβ, resulting in a 2-fold gain in TRβ selectivity.

To analyze the role of the bridging moiety and polar side chain in influencing TRα/TRβ selectivity, pairwise comparisons were made among GC-1, DIMIT, methylene-bridged DIMIT, and ether-bridged GC-1. As shown in Figure 3, the ether to methylene substitution results in the same change in affinity for TRα and TRβ. Methylene-bridged DIMIT shows (4.2 ± 0.5)-fold and (4.9 ± 0.2)-fold increases in affinity for TRα and TRβ, respectively, compared to DIMIT, while GC-1 correspondingly has (3.6 ± 0.6)-fold and (3.8 ± 0.4)-fold greater affinities than ether-bridge GC-1. Thus, the ether to methylene substitution has little effect on selectivity.

The gains in affinity for ether-bridged GC-1 compared to DIMIT ((3.6 ± 0.6)-fold for TRα and (17 ± 1)-fold for TRβ) and correspondingly for GC-1 compared to meth-

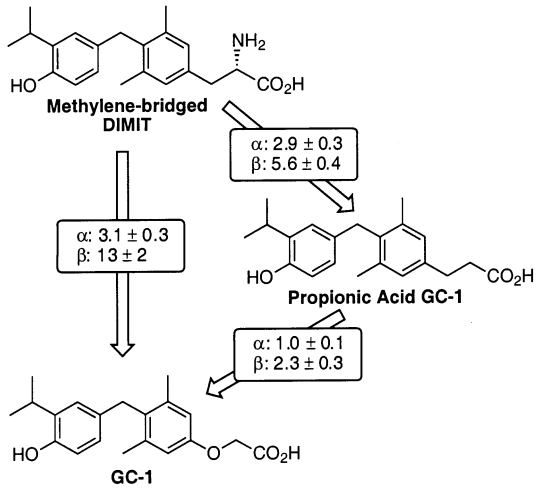


Figure 4. Loss of the α-amino group and the side chain ether for methylene substitution both contribute toward TRβ selectivity. Values indicate fold gain in affinity for TRα and TRβ.

ylene-bridged DIMIT ((3.1± 0.3)-fold for TRα and (13 ± 2)-fold for TRβ) show a strong differential gain in affinity for TRβ associated with the alanine to oxyacetic acid polar side chain substitution. In comparison with the results of the ether to methylene substitution, these data clearly demonstrate the key role of the oxyacetic acid side chain in GC-1's preferential binding to TRβ.

The change of alanine side chain to oxyacetic acid can be thought of as occurring in two steps: first, the replacement of the α-amine with hydrogen and, second, the ether replacement of the side chain methylene. To determine the role of these specific structural changes in influencing affinity and selectivity, the preceding analysis was repeated with methylene-bridged DIMIT, GC-1, and propionic acid GC-1. The comparison, shown in Figure 4, demonstrates that both changes differentially increase affinity for TRβ. The loss of the amino group increases affinity (2.9 ± 0.3)-fold and (5.6 ± 0.4)-fold for TRα and TRβ, respectively, while a corresponding (2.3 ± 0.3)-fold gain in affinity for TRβ is observed in the side chain methylene to ether substitution.

The free energies of binding associated with the binding affinities of the compounds (calculated with units of mol/L) are listed in Table 1. The changes in selectivity associated with the various structural alterations of the thyromimetics are expressed in Figure 5 as the differential gain in free energy of binding, or $\Delta\Delta\Delta G_{bind}$. Inspection of the ratios of affinities in Figure 3 shows the independent effects of the structural changes on affinity. The ether bridge to methylene substitution has practically the same effect on binding affinity regardless of the identity of the polar side chain. Similarly, the side chain substitutions have almost the same effect on affinity and selectivity within the context of either bridging moiety.

Discussion

The preceding results clearly demonstrate that the TRβ selectivity of GC-1 is determined by its oxyacetic acid side chain. In a comparison of the GC-1 analogues with alanine and propionic acid side chains, it is also apparent that both the loss of the α-amine and the methylene to ether substitution make significant contributions to differentially increase the affinity for TRβ.

That the polar side chain substitution has the same effect on binding and selectivity in the context of either bridging moiety strongly implies that the side chain and the adjacent aromatic ring (A-ring) interact with the receptors in the same fashion regardless of bridging moiety. Accordingly, the small shift in the relative positions of the A-ring and B-ring caused by the longer carbon−carbon bonds of the methylene bridge (1.54 versus 1.39 Å for C−O) would be borne out by the B-ring with the A-ring remaining in the same position relative to the receptor.

Because the improvement in affinity observed with the introduction of the methylene bridge is nearly equal for both receptors in the context of either side chain, it is possible that a factor external to the interaction of ligand with receptor is mostly responsible for the increased affinity. One such factor could be the increased hydrophobicity of the ligands containing the methylene bridge. Increased hydrophobicity of a ligand increases the free energy in the unbound state by increasing the energetic cost of solvation and results in a greater free energy of binding and higher affinity. This effect may not be functioning in this case because diphenylmethane has a reported log *P* value of 4.14 while the corresponding value for diphenyl ether is 4.21,[30] suggesting that a significant increase in hydrophobicity does not occur with the methylene for oxygen substitution. Alternatively, the methylene bridge may cause the B-ring to interact with the receptor in a more energetically favorable fashion to cause the observed 3-

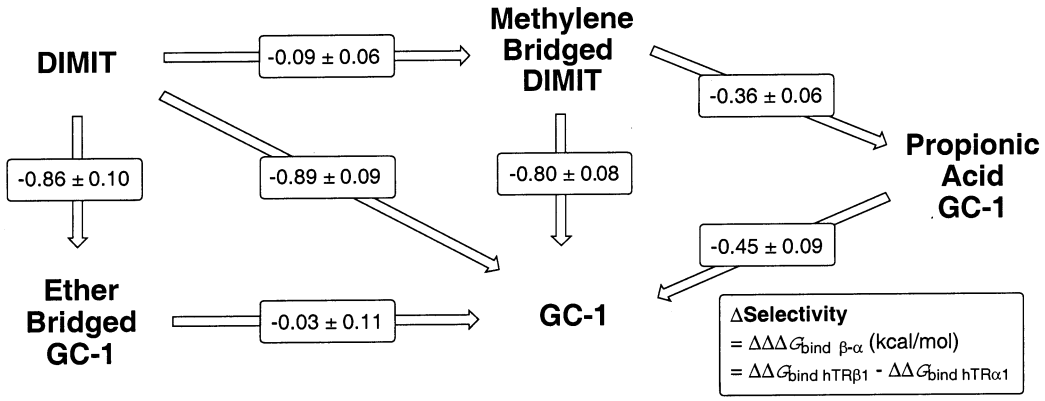


Figure 5. Oxyacetic acid polar side chain is the key determinant of TRβ selectivity. Values indicate gain in TRβ selectivity (kcal/mol).

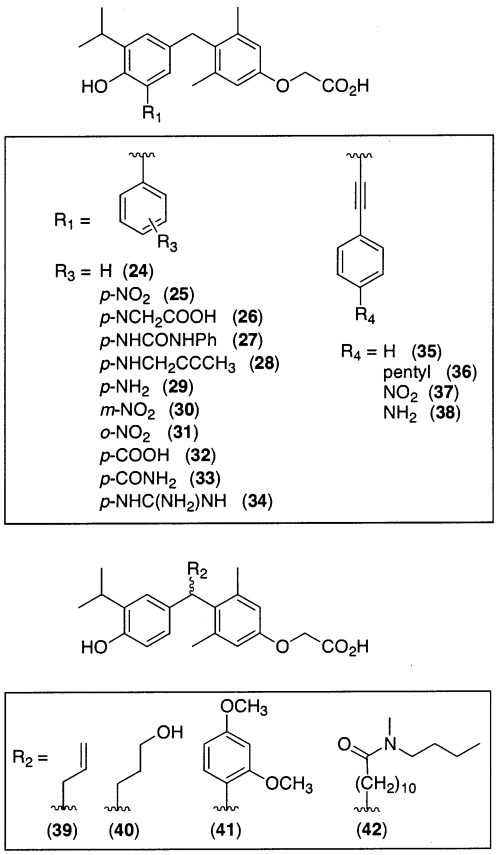


Figure 6. Structures of previously reported GC-1 analogues with substituents at the 5′-position and at the bridging carbon.

to 5-fold increase in affinity. Interestingly, the B-ring of GC-1 shows a slightly shifted position, while the B-rings of triac, DIMIT, and T_3 all closely overlap in structural alignments of the crystal structures of liganded TR-LBDs.[27,28,31]

While we have shown the importance of the character of the polar side chain in determining selectivity toward binding TRβ, we do know that other features of the ligand can influence selectivity. Our laboratory has prepared over 20 analogues of GC-1 in studies of TR antagonism.[32−34] Their structures are summarized in Figure 6. Most of these analogues bear substituted phenyl or phenylethynyl substituents at the 5′-position (**24**−**38**), while a racemic series with alkyl, allyl, and aryl substiuents at the bridging carbon (**39**−**42**) has also been prepared. All show reduced affinity for TR of about 200- to 7000-fold lower than GC-1. TRβ selectivity is observed with most of these GC-1 analogues; however, the degree of TRβ selectivity varies considerably between 0.33- and 20-fold in a manner that is not easily explained by the character of the extra substituent. The analogues with 5′ substituents all bind better to TRβ than to TRα, though the degree of selectivity varies between 3- and 20-fold.[33,34] Analogues with hydroxyalkyl, aryl, and alkylamide substitutents at the bridging carbon (**40**−**42**) show 3.6- to 1.5-fold TRα selectivity, while the allyl-substituted analogue (**39**) is 7-fold TRβ-selective.[32] Substitution at the bridging carbon can, but does not necessarily, confer slight TRα selectivity. The binding characteristics of these analogues indicate that their perturbations of the ligand binding cavity are not equally tolerated by TRα and TRβ, presumably because of structural differences in the ligand binding domains.

This study demonstrates that subtle changes to the structures of the polar side chains of high-affinity thyromimetics can have significant effects on selectivity. Our understanding of selectivity from these results, along with the earlier structural and mutagenesis studies,[28] emphasizes the importance of polar interactions between ligand and receptor in the vicinity of the one varying amino acid residue in the ligand binding cavity. While the precise ligand−receptor interactions that govern selectivity remain elusive, structural features of both the receptor and ligand that influence selectivity have been identified.

Experimental Section

Thyroid Hormone Receptor Binding Assays. hTRα_1 and hTRβ_1 were expressed in *Escherichia coli* and purified as described previously.[35] Competition ligand binding affinities were determined using 0.1 nM [^{125}I]-T_3 in gel filtration binding assays performed in duplicate as described.[35] The K_d and standard error (SE) values were calculated by fitting the competition data using the GraphPad Prism computer program (GraphPad Software, Inc.)

General Methods. Unless otherwise indicated, solvents and reagents were purchased from the Aldrich Chemical Co. and used without further purification. Tetrahydrofuran was distilled from sodium benzophenone ketyl prior to use. Other anhydrous solvents were from Aldrich SureSeal bottles. Water-sensitive reactions were performed using oven- or flame-dried glassware. Unless indicated, reactions were performed under argon inert atmosphere. NMR spectra were obtained using a Varian Inova 400 MHz spectrometer. Target compounds **7**−**9** were purified by reverse-phase preparative HPLC.

4-Benzyloxy-2,6-dimethylphenol (13). To a stirred suspension of 2,6-dimethylhydroquinone (Rütgers Organics) (3.00 g, 21.7 mmol) and potassium carbonate (6.30 g, 45.6 mmol) in DMF (anhydrous, 10 mL) was added benzyl bromide (2.6 mL, 22 mmol). The reaction mixture was cooled in a cold water bath and was stirred overnight at room temperature. After 21 h, the reaction was quenched by the cautious addition of 1 M HCl (50 mL), followed by extraction with ethyl acetate (4 × 50 mL). The combined organic fractions were washed with saturated aqueous NH_4Cl (2 × 20 mL) and brine (2 × 20 mL), then dried ($MgSO_4$), filtered through a Celite pad, and concentrated under reduced pressure. The residue was purified by flash chromatography (silica, 5% → 10% ethyl acetate in hexanes) to give the product (440 mg, 9% yield) as a white solid. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.30−7.43 (m, 5 H), 6.63 (s, 2 H), 4.97 (s, 2 H), 4.25 (s, 1 H), 2.21 (s, 6 H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 152.23, 146.32, 137.48, 128.49, 127.77, 127.43, 124.09, 114.90, 70.56, 16.24. HRMS exact mass calcd for $C_{12}H_{16}O_2$: 228.1150. Found: 228.1148.

3-Isopropyl-4-methoxymethoxyphenylboronic Acid (11). A stirred solution of 3-isopropyl-4-methoxymethoxybromobenzene (2.5 g, 9.6 mmol) in THF (25 mL) was chilled to −78 °C, and *n*-butyllithium (2.5 M in hexanes, 4.0 mL, 10 mmol) was added via syringe over several minutes. After 15 min of stirring, trimethyl borate (1.2 mL, 11 mmol) was added via syringe. The mixture was stirred overnight, slowly warming to room temperature. Then 1 M HCl (25 mL) was added and stirring was continued for another hour. The aqueous and organic layers were separated, and the aqueous layer was extracted with methylene chloride (2 × 40 mL). Combined organic fractions were dried ($MgSO_4$), filtered through a Celite pad, and concentrated under reduced pressure to yield the product (2.0 g) as a thick clear syrup that solidified over a few days. The product was used directly in the next reaction without further purification.

α-[4-(3-Isopropyl-4-methoxymethoxyphenoxy)-3,5-dimethylphenoxy]toluene (14). To a stirred solution of 3-isopropyl-4-methoxymethoxyphenylboronic acid (**11**) (approximately 290 mg, 1.3 mmol) and 4-benzyloxy-2,6-dimethylphenol (**13**) in methylene chloride (anhydrous, 10 mL) was added

copper(II) acetate (80 mg, 0.44 mmol), triethylamine (305 μL, 2.19 mmol), and powdered 4 Å molecular sieves (spatula tip). A drying tube was attached, and the mixture was stirred under ambient atmosphere. After 25 h, the mixture was filtered through a pad of Celite, which was rinsed with ethyl acetate. Rotary evaporation yielded a brown-green semisolid that was purified by flash chromatography to yield the enriched product (149 mg), which was used directly in the next reaction.

4-(3-Isopropyl-4-methoxymethoxyphenoxy)-3,5-dimethylphenol (14a). A solution of α-[4-(3-isopropyl-4-methoxymethoxyphenoxy)-3,5-dimethylphenoxy]toluene (**14**) (approximately 0.25 mmol) in ethyl acetate was sparged with Ar, and a spatula tip of palladium on carbon (10%) was added. The flask was purged with H_2, and the mixture was stirred overnight under H_2 atmosphere maintained with balloon pressure and then was filtered through a Celite pad that was rinsed with ethyl acetate. After rotary evaporation, the crude product was purified by flash chromatography (silica, 5% → 20% ethyl acetate in hexanes) to yield the product (36 mg, 27% yield from **13**). ^{1}H NMR (400 MHz, $CDCl_3$): δ 6.89 (d, J = 9.3 Hz, 1 H), 6.74 (d, J = 2.9 Hz, 1 H), 6.54 (s, 2 H), 6.36 (dd, J = 8.8, 2.9 Hz, 1 H), 5.12 (s, 2 H), 4.93 (s, 1 H), 3.49 (s, 3 H), 3.29 (heptet, J = 6.8, Hz, 1 H), 2.06 (s, 6 H), 1.18 (d, J = 6.8 Hz, 6 H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 153.19, 152.06, 148.81, 145.10, 139.47, 132.70, 115.41, 115.17, 112.91, 111.14, 95.38, 55.97, 27.05, 22.76, 22.63, 16.47. HRMS exact mass calcd for $C_{19}H_{24}O_3$: 316.1675. Found: 316.1665.

[4-(3-Isopropyl-4-methoxymethoxyphenoxy)-3,5-dimethylphenoxy]acetic Acid Methyl Ester (14b). To a stirred suspension of **14a** (30 mg, 0.095 mmol) and cesium carbonate (46 mg, 0.14 mmol) in DMF (anhydrous, 200 μL) was added methyl bromoacetate (13 μL, 0.14 mmol) via syringe. The mixture was stirred overnight at room temperature. Then the reaction was quenched with half-saturated NH_4Cl (8 mL) and the mixture was extracted with diethyl ether (2 × 10 mL). The combined organic fractions were washed with brine (2 × 2 mL), dried ($MgSO_4$), filtered through a Celite pad, and concentrated under reduced pressure to yield the product (30 mg, 81% yield). ^{1}H NMR (400 MHz, $CDCl_3$): δ 6.88 (d, J = 9.3 Hz, 1 H), 6.73 (d, J = 2.9 Hz, 1 H), 6.63 (s, 2 H), 6.34 (dd, J = 9.0, 3.2 Hz, 1 H), 5.11 (s, 2 H), 4.62 (s, 2 H), 3.82 (s, 3 H), 3.48 (s, 3 H), 3.29 (heptet, J = 6.8, Hz, 1 H), 2.09 (s, 6 H), 1.18 (d, J = 7.3 Hz, 6 H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 169.55, 154.30, 153.02, 148.90, 146.00, 139.44, 132.68, 115.34, 114.63, 112.94, 111.12, 95.35, 65.64, 55.94, 52.21, 27.05, 22.73, 16.66. HRMS exact mass calcd for $C_{22}H_{28}O_6$: 388.1886. Found: 388.1895.

[4-(4-Hydroxy-3-isopropylphenoxy)-3,5-dimethylphenoxy]acetic Acid (7). To a stirred suspension of methyl ester **14b** (30 mg, 0.077 mmol) and lithium hydroxide monohydrate (7.0 mg, 0.17 mmol) in methanol (2 mL) was added water (7.6 μL, 0.42 mmol). The mixture was stirred at room temperature for 4 h, and then solvent was removed under reduced pressure. The residue was resuspended in a 1:1 mixture of saturated NH_4Cl/1 M HCl (6 mL) and extracted with ethyl acetate (3 × 6 mL). The combined organic fractions were dried ($MgSO_4$), filtered through a Celite pad, and concentrated under reduced pressure. The crude product was purified by flash chromatography (silica, 7.5% ethyl acetate, 3% acetic acid in chloroform) and preparative TLC (silica, 20 cm × 20 cm, 15% ethyl acetate, 3% acetic acid in chloroform, eluted with 10% methanol in chloroform) to give a white solid (7 mg, 30% yield). ^{1}H NMR (400 MHz, CD_3OD): δ 6.70−6.65 (m, 2 H), 6.67 (s, 2 H), 6.25 (dd, J = 8.8, 2.9 Hz, 1 H), 4.61 (s, 2 H), 3.21 (heptet, J = 6.8 Hz, 1 H), 2.05 (s, 6 H), 1.13 (d, J = 7.3 Hz, 6 H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 172.94, 155.94, 152.78, 149.66, 147.25, 137.52, 133.64, 116.52, 115.65, 113.28, 112.33, 66.22, 28.11, 22.95, 16.75. HRMS exact mass calcd for $C_{19}H_{22}O_5$: 330.1467. Found: 330.1471.

4-Benzyloxy-2,6-dimethylbromobenzene (15a). A suspension 4-bromo-3,5-dimethylphenol (20.0 g, 99.5 mmol), benzyl bromide (17 g, 99 mmol), and potassium carbonate (20.6 g, 149 mmol) in DMF (anhydrous, 20 mL) was stirred overnight at room temperature. The reaction mixture was diluted with diethyl ether (150 mL), and the reaction was quenched with the cautious addition of 1 M HCl (250 mL). The aqueous and organic phases were separated, and the aqueous phase was extracted with diethyl ether (2 × 180 mL). The combined organic phases were washed with saturated NH_4Cl (2 × 50 mL) and brine (2 × 50 mL), then dried ($MgSO_4$), filtered through a Celite pad, and concentrated under reduced pressure. The crude product was purified with two rounds of KugelRohr distillation, giving an off-white solid (22.3 g, 78% yield). ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.43−7.32 (m, 5 H), 6.73 (s, 2 H), 5.01 (s, 2 H), 2.38 (s, 6 H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 157.25, 139.12, 136.84, 128.57, 127.97, 127.40, 118.49, 114.72, 70.04, 24.06. HRMS exact mass calcd for $C_{15}H_{15}BrO$: 290.0306. Found: 290.0320.

4-Benzyloxy-2,6-dimethylbenzaldehyde (16). A stirred solution of bromobenzene **15a** (21.0 g, 72.1 mmol) in THF (200 mL) was cooled to −78 °C. Then *n*-butyllithium (2.5 M in hexanes, 32 mL, 79 mmol) was added via syringe over several minutes. A thick precipitate formed, and stirring was maintained manually. After 10 min, DMF (anhydrous, 8.4 mL, 110 mmol) was added via syringe over a period of 10 min with frequent shaking of the flask as the precipitate dissolved. Stirring at −78 °C was maintained for 20 min, and then the reaction was quenched by the addition of 1 M HCl (150 mL). The aqueous and organic layers were separated, and the aqueous phase was extracted with diethyl ether (3 × 100 mL). The combined organic fractions were washed with saturated $NaHCO_3$ (100 mL) and brine (100 mL) and then dried ($MgSO_4$), filtered through a Celite pad, and concentrated under reduced pressure. Purification by short-path vacuum distillation, followed by KugelRohr distillation (1 Torr, 160 °C) gave the product (14.0 g, 80% yield) as an off-white solid. ^{1}H NMR (400 MHz, $CDCl_3$): δ 10.47 (s, 1 H), 7.42−7.32 (m, 5 H), 6.67 (s, 2 H), 5.09 (s, 2 H), 2.60 (s, 6 H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 191.56, 161.85, 144.44, 136.17, 128.64, 128.18, 127.42, 126.13, 115.61, 69.84, 21.06. HRMS exact mass calcd for $C_{16}H_{16}O_2$: 240.1150. Found: 240.1151.

3-Isopropyl-4-triisopropylsilyloxybromobenzene (17). To a stirred solution of triisopropylsilyl chloride (12 mL, 56 mmol) in anhydrous 1,2-dichloroethane (70 mL) was added 4-bromo-2-isopropylphenol (9.6 g, 46 mmol) and imidazole (7.8 g, 114 mmol). The reaction mixture was refluxed for 30 min and then was stirred overnight at room temperature. The reaction mixture was refluxed 1 h further, and then 150 mL of 0.6 M HCl was added, layers were separated, and the aqueous phase was extracted with diethyl ether. The combined organic fractions were washed with saturated $NaHCO_3$, dried ($MgSO_4$), and filtered through a Celite plug and solvent was removed by rotary evaporation to yield an oil (18.3 g). Purification by fractional distillation (bp 137 °C, 0.3 mm) yielded bromobenzene **17** as a white solid (12.3 g, 72%). ^{1}H NMR (600 MHz, $CDCl_3$): δ 7.27 (d, J = 2.6 Hz, 1 H), 7.11 (dd, J = 8.4, 2.6 Hz, 1 H), 6.64 (d, J = 8.8 Hz, 1 H), 3.33 (heptet, J = 6.8 Hz, 1 H), 1.30 (heptet, J = 7.5 Hz, 3 H), 1.19 (d, J = 6.6 Hz, 6 H), 1.10 (d, J = 7.7 Hz, 18 H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 152.29, 140.98, 129.27, 128.93, 119.36, 113.14, 26.85, 22.61, 18.04, 13.05. HRMS exact mass calcd for $C_{18}H_{31}BrOSi$: 370.1328. Found: 370.1336.

4-Benzyloxy-2,6-dimethylphenyl[3-isopropyl-4-(triisopropylsilyloxy)phenyl]methanol (18). A stirred solution of **17** (13.9 g, 37.5 mmol) in THF (250 mL) was cooled to −78 °C, and *n*-butyllithium (2.5 M in hexanes, 16 mL, 40 mmol) was added via syringe over 10 min. After 10 min of stirring at −78 °C, **16** (9.00 g, 37.5 mmol), dissolved in THF (10 mL), was added dropwise via cannula over 20 min, followed by rinses with THF (2 × 5 mL). After the mixture was stirred for 30 min at −78 °C, the reaction was quenched with 1 M HCl (45 mL). The aqueous and organic layers were separated, and the aqueous phase was extracted with diethyl ether (4 × 40 mL). The combined organic fractions were washed with saturated $NaHCO_3$ (30 mL) and brine (30 mL), then dried ($MgSO_4$), filtered through a Celite pad, and concentrated under reduced pressure. Flash chromatography of the residue (2% → 7% ethyl acetate in hexanes) gave the purified product (15.5 g, 77% yield). ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.45−7.32 (m, 4 H), 7.23

(d, J = 1.5 Hz, 1 H), 6.75 (dd, J = 8.2, 1.5 Hz, 1 H), 6.67−6.65 (m, 3 H), 6.24 (d, J = 4.3 Hz, 1 H), 5.04 (s, 2 H), 3.36 (heptet, J = 7.0, Hz, 1 H), 2.24 (s, 6 H), 1.29 (heptet, J = 7.6 Hz, 3 H), 1.17 (dd, J = 8.2, 7.0 Hz, 6 H), 1.10 (d, J = 7.3 Hz, 18 H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 157.62, 151.72, 138.72, 138.13, 137.16, 135.07, 132.49, 128.53, 127.87, 127.48, 123.50, 123.39, 117.46, 115.20, 70.95, 69.74, 26.74, 22.83, 20.99, 18.09, 13.07. HRMS exact mass calcd for $C_{34}H_{48}O_3Si$: 532.3373. Found: 532.3379.

4-[3-Isopropyl-4-(triisopropylsilyloxy)benzyl]-3,5-dimethylphenol (18a). A solution of **18** (9.35 g, 17.8 mmol) in 1:1 ethyl acetate/ethanol (30 mL) in a glass Parr hydrogenator flask was sparged with Ar. Palladium on carbon (10%, 1.3 g) was added, and the flask was attached to a Parr hydrogenator and shaken for 24 h under H_2 at 35 psi. The mixture was then filtered through a pad of Celite, which was then rinsed with ethyl acetate, and the filtrate was concentrated to yield the product (7.77 g, 100% yield) as a white solid. ^{1}H NMR (400 MHz, $CDCl_3$): δ 6.92 (s, 1 H), 6.61 (d, J = 8.3 Hz, 1 H), 6.54−6.51 (m, 3 H), 4.96 (s, 1 H), 3.87 (s, 2 H), 3.33 (heptet, J = 6.9 Hz, 1 H), 2.18 (s, 6 H), 1.27 (heptet, J = 7.3 Hz, 3 H), 1.16 (d, J = 6.9 Hz, 6 H), 1.09 (d, J = 7.6 Hz, 18 H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 153.20, 150.96, 138.65, 138.10, 131.96, 130.02, 125.82, 124.83, 117.60, 114.72, 33.74, 26.62, 22.83, 20.32, 18.09, 13.05. HRMS exact mass calcd for $C_{27}H_{42}O_2Si$: 426.2954. Found: 426.2950.

4-[3-Isopropyl-4-(triisopropylsilyloxy)benzyl]-3,5-dimethylphenyltrifluoromethanesulfonate (19). A stirred solution of **18a** (10.0 g, 23.4 mmol) and pyridine (anhydrous, 20 mL, 250 mmol) in methylene chloride (anhydrous, 150 mL) was cooled in an ice bath. Triflic anhydride (4.3 mL, 26 mmol) was added via syringe over 5 min, with vigorous stirring maintained. The mixure was stirred for 4 h, slowly warming to room temperature. Then a 1:1 mixture of saturated NH_4Cl and 1 M HCl (100 mL) was added. The aqueous and organic layers were separated, and the aqueous phase was extracted with diethyl ether (3 × 100 mL). The combined organic fractions were dried ($MgSO_4$), filtered through a Celite pad, and concentrated under reduced pressure. Flash chromatography (silica, 9% diethyl ether in hexanes) gave the product (9.2 g, 99% yield with recovered starting material) and unreacted phenol (2.9 g). ^{1}H NMR (400 MHz, $CDCl_3$): δ 6.96 (s, 2 H), 6.88 (d, J = 2.0 Hz, 1 H), 6.62 (d, J = 8.3 Hz, 1 H), 6.46 (dd, J = 8.3, 2.4 Hz, 1 H), 3.94 (s, 2 H), 3.32 (heptet, J = 6.8, Hz, 1 H), 2.27 (s, 6 H), 1.27 (heptet, J = 7.8, Hz, 3 H), 1.15 (d, J = 6.8 Hz, 6 H), 1.09 (d, J = 7.3 Hz, 18 H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 151.37, 147.41, 139.74, 138.50, 138.25, 130.23, 125.82, 124.76, 120.20, 118.77 (qt, J = 320.7 Hz), 117.78, 34.10, 26.64, 22.78, 20.42, 18.09, 13.07. HRMS exact mass calcd for $C_{28}H_{41}F_3O_4SSi$: 558.2457. Found: 558.2447.

[4-(2,6-Dimethyl-4-vinylbenzyl)-2-isopropylphenoxy]-triisopropylsilane (19a). To a stirred suspension of triflate **19** (5.00 g, 8.95 mmol), lithium chloride (1.14 g, 26.8 mmol), and dichlorobis(triphenylphosphine)palladium(II) (314 mg, 0.447 mmol) in DMF (anhydrous, degassed, 25 mL) was added vinyltributyltin (2.8 mL, 9.8 mmol). The reaction flask was fitted with a heating mantle, and a thermocouple needle was attached to a temperature controller. The reaction mixture was stirred for 2 h at 60 °C, and then most of the solvent was removed under reduced pressure. The residue was resuspended in 1:1 diethyl ether/hexanes (50 mL) and aqueous saturated potassium fluoride (10 mL). The mixture was stirred for 2 h and then filtered through a pad of Celite, which was rinsed with diethyl ether (200 mL). Hexanes (200 mL) were added to the filtrate, and the aqueous and organic phases were separated. The organic layer was washed with saturated $NaHCO_3$ (25 mL) and half-saturated brine (4 × 25 mL), then dried ($MgSO_4$), filtered through a Celite pad, and concentrated under reduced pressure. Flash chromatography (silica, hexanes), followed by KugelRohr vacuum distillation to remove the organotin residue, gave a cloudy syrup that contained ~13 mol % organotin residue (^{1}H NMR) (3.80 g, 85% yield). A small sample was repurified for characterization. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.10 (s, 2 H), 6.94 (d, J = 2.0 Hz, 1 H), 6.66 (dd, J = 17.6, 10.7 Hz, 1 H), 6.59 (d, J = 8.3 Hz, 1 H), 6.50 (dd, J = 8.3, 2.0 Hz, 1 H), 5.71 (d, J = 17.6 Hz, 1 H), 5.18 (d, J = 11.7 Hz, 1 H), 3.94 (s, 2 H), 3.32 (heptet, J = 6.8 Hz, 1 H), 2.24 (s, 6 H), 1.27 (heptet, J = 7.3, Hz, 3 H), 1.16 (d, J = 6.8 Hz, 6 H), 1.08 (d, J = 7.3 Hz, 18 H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 151.06, 138.17, 137.54, 137.24, 136.88, 135.20, 131.44, 125.99, 125.94, 124.90, 117.65, 112.65, 34.43, 26.66, 22.84, 20.29, 18.11, 13.07. HRMS exact mass calcd for $C_{29}H_{44}OSi$: 436.3161. Found: 436.3163.

4-[3-Isopropyl-4-(triisopropylsilyloxy)benzyl]-3,5-dimethylbenzaldehyde (20). A stirred solution of styrene **19a** (2.15 g, 4.9 mmol) in methylene chloride (anhydrous, 200 mL) was cooled to −78 °C and sparged with an oxygen/ozone mixture until the solution took on a light-violet color. The solution was then sparged with Ar for 15 min, and then triphenylphosphine (7.5 g, 29 mmol) dissolved in methylene chloride was added via cannula. After the mixture was stirred for 50 min, the solvent was removed under reduced pressure. The residue was purified by flash chromatography (silica, 0.5% → 1% diethyl ether in hexanes) to give **20** (1.79 g, 86% yield) as a white solid. ^{1}H NMR (400 MHz, $CDCl_3$): δ 9.94 (s, 1 H), 7.56 (s, 2 H), 6.90 (d, J = 2.0 Hz, 1 H), 6.61 (d, J = 8.3 Hz, 1 H), 6.48 (dd, J = 8.3, 2.44 Hz, 1 H), 4.02 (s, 2 H), 3.32 (heptet, J = 6.8 Hz, 1 H), 2.33 (s, 6 H), 1.26 (heptet, J = 7.3 Hz, 3 H), 1.15 (d, J = 7.3 Hz, 6 H), 1.09 (d, J = 7.3 Hz, 18 H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 192.51, 151.35, 145.30, 138.50, 138.18, 134.44, 130.07, 129.38, 125.93, 124.84, 117.78, 34.92, 26.63, 22.79, 20.23, 18.08, 13.05. HRMS exact mass calcd for $C_{28}H_{42}O_2Si$: 438.2954. Found: 438.2959.

(*E*)-3-{4-[3-Isopropyl-4-(triisopropylsilyloxy)benzyl]-3,5-dimethylphenyl}acrylic Acid Benzyl Ester (20a). A solution of benzaldehyde **20** (500 mg, 1.13 mmol) and benzyl (triphenylphosphoranylidene)acetate (834 mg, 2.03 mmol) in THF (25 mL) was stirred at reflux overnight. Solvent was removed under reduced pressure, and the residue was purified by flash chromatography (silica, 2% → 4% ethyl acetate in hexanes) to give **20a** (646 mg, 100% yield) as a syrup. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.68 (d, J = 16.1 Hz, 1 H), 7.42−7.31 (m, 5 H), 7.21 (s, 2 H), 6.92 (d, J = 1.5 Hz, 1 H), 6.60 (d, J = 8.3 Hz, 1 H), 6.50−6.44 (m, 2 H), 5.25 (s, 2 H), 3.96 (s, 2 H), 3.32 (heptet, J = 6.8 Hz, 1 H), 2.25 (s, 6 H), 1.27 (heptet, J = 7.3 Hz, 3 H), 1.15 (d, J = 6.8 Hz, 6 H), 1.08 (d, J = 7.3 Hz, 18 H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 167.03, 151.18, 145.46, 140.71, 138.32, 137.76, 136.19, 132.01, 130.76, 128.53, 128.17, 128.13, 127.86, 125.94, 124.82, 117.69, 116.63, 66.17, 34.59, 26.63, 22.80, 20.25, 18.08, 13.03. HRMS exact mass calcd for $C_{37}H_{50}O_3Si$: 570.3529. Found: 570.3524.

3-[4-(4-Hydroxy-3-isopropylbenzyl)-3,5-dimethylphenyl]-acrylic Acid Benzyl Ester (20b). A solution of aryl silyl ether **20a** (60 mg, 0.11 mmol) and triethylamine trihydrofluoride (1.7 mL, 11 mmol) in THF (3 mL) was stirred overnight and then cooled in an ice bath and quenched with the cautious addition of potassium carbonate (2.0 g, 15 mmol) dissolved in water (5 mL). After addition of saturated $NaHCO_3$ (5 mL), the mix was extracted with diethyl ether (3 × 15 mL), then dried ($MgSO_4$), filtered through a Celite pad, and concentrated under reduced pressure. Purification of the residue with flash chromatography (silica, 10% → 20% ethyl acetate in hexanes) gave **20b** (40 mg, 88% yield). ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.68 (d, J = 16.1 Hz, 1 H), 7.42−7.32 (m, 5 H), 7.20 (s, 2 H), 6.59 (d, J = 8.3 Hz, 1 H), 6.53 (d, J = 8.3 Hz, 1 H), 6.46 (d, J = 16.1 Hz, 1 H), 5.25 (s, 2 H), 3.96 (s, 2 H), 3.17 (heptet, J = 6.8 Hz, 1 H), 2.24 (s, 6 H), 1.20 (d, J = 6.8 Hz, 6 H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 167.26, 151.07, 145.62, 140.50, 137.72, 136.05, 134.45, 132.00, 130.89, 128.53, 128.46, 128.17, 127.90, 126.16, 125.27, 116.57, 115.22, 66.30, 34.46, 27.07, 22.52, 20.18. HRMS exact mass calcd for (M − benzyl) $C_{21}H_{23}O_3$: 323.1647. Found: 323.1632.

3-{4-[3-Isopropyl-4-(triisopropylsilyloxy)benzyl]-3,5-dimethylphenyl}propionic Acid (20c). A solution of benzyl ester **20b** (490 mg, 0.86 mmol) in ethyl acetate (30 mL) and acetic acid (2 mL) was put in a Parr hydrogenator flask and sparged with Ar. Palladium on carbon (10%, 90 mg) was added, and the mixture was shaken for 20 h under H_2 atmosphere at

33 psi. The mixture was then filtered through a Celite pad, which was rinsed with ethyl acetate and ethanol. The filtrate was concentrated under reduced pressure, with toluene azeotroping to remove residual acetic acid. Flash chromatography (silica, 10% ethyl acetate, 1% acetic acid in hexanes) gave **20c** (355 mg, 86% yield) as a crystalline solid. ^{1}H NMR (400 MHz, $CDCl_3$): δ 6.94 (s, 1 H), 6.89 (s, 2 H), 6.59 (d, J = 7.8 Hz, 1 H), 6.49 (d, J = 8.3 Hz, 1 H), 3.92 (s, 2 H), 3.32 (heptet, J = 6.8 Hz, 1 H), 2.90 (t, J = 7.8 Hz, 2 H), 2.69 (t, J = 8.3 Hz, 2 H), 2.22 (s, 6 H), 1.27 (heptet, J = 7.8 Hz, 3 H), 1.16 (d, J = 6.8 Hz, 6 H), 1.08 (d, J = 7.3 Hz, 18 H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 178.73, 151.01, 138.12, 137.65, 137.31, 135.69, 131.57, 127.87, 126.00, 124.82, 117.61, 35.62, 34.23, 30.18, 26.63, 22.83, 20.25, 18.11, 13.06. HRMS exact mass calcd for $C_{30}H_{46}O_3Si$: 482.3216. Found: 482.3217.

3-[4-(4-Hydroxy-3-isopropylbenzyl)-3,5-dimethylphenyl]-propionic Acid (9). A solution of benzyl ester **20b** (21 mg, 0.051 mmol) in ethyl acetate (2 mL) was sparged with Ar, and then palladium on carbon (10%, small spatula tip) was added. After being stirred overnight under H_2 atmosphere maintained with balloon pressure, the mixture was filtered through a Celite pad, which was rinsed with ethyl acetate and ethanol. The filtrate was concentrated under reduced pressure, and the residue was purified by preparative TLC (silica, 20 cm × 20 cm × 1000 μm, 4% methanol, 1% acetic acid in chloroform) to give **9** (15 mg, 90% yield). ^{1}H NMR (400 MHz, CD_3OD): δ 6.88 (s, 2 H), 6.81 (s, 1 H), 6.56 (d, J = 8.3 Hz, 1 H), 6.49 (dd, J = 8.3, 2.0 Hz, 1 H), 3.89 (s, 2 H), 3.19 (heptet, J = 7.3 Hz, 1 H), 2.82 (t, J = 7.6 Hz, 2 H), 2.57 (t, J = 7.8 Hz, 2 H), 2.18 (s, 6 H), 1.12 (d, J = 6.8 Hz, 6 H). ^{13}C NMR (100 MHz, CD_3OD): δ 176.98, 153.39, 139.61, 138.08, 136.65, 135.82, 131.78, 128.94, 126.70, 126.33, 115.82, 36.92, 34.85, 31.67, 28.01, 23.05, 20.30. HRMS exact mass calcd for $C_{21}H_{26}O_3$: 326.1882. Found: 326.1888.

(*S*)-(4)-Benzyl-3-(3-{4-[3-isopropyl-4-(triisopropylsilyloxy)benzyl]-3,5-dimethylphenyl}propionyl)oxazolidin-2-one (20d). A stirred solution of carboxylic acid **20c** (304 mg, 0.63 mmol) in THF (15 mL) was chilled to −78 °C, and triethylamine (114 μL, 0.82 mmol) and pivaloyl chloride (85 μL, 0.69 mmol) were added via syringe. The mixture was stirred for 15 min at −78 °C, then for 30 min on an ice bath, and then at −78 °C again. To a separate stirred solution of (*S*)-4-benzyl-2-oxazolidinone (200 mg, 1.13 mmol) in THF (9 mL) cooled to −78 °C was added *n*-butyllithium (2.5 M in hexane, 450 μL, 1.13 mmol). After 10 min of stirring, the oxazolidinone lithium salt was added via cannula to the mixed anhydride solution. After the mixture was stirred for 30 min at −78 °C and another 30 min while slowly warming to room temperature, the reaction was quenched with the addition of 1 M $NaHSO_4$ (15 mL). After removal of most of the THF under reduced pressure, the residue was extracted with ethyl acetate (3 × 20 mL). The combined organic fractions were washed with saturated $NaHCO_3$ (15 mL) and brine (10 mL), then dried ($MgSO_4$), filtered through a Celite pad, and concentrated under reduced pressure. Purification of the residue by flash chromatography (silica, 5% ethyl acetate in hexanes) gave **20d** (359 mg, 89% yield) as a colorless syrup. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.31−7.23 (m, 3 H), 7.18 (d, J = 6.8 Hz, 2 H), 6.96 (d, J = 2.4 Hz, 1 H), 6.94 (s, 2 H), 6.58 (d, J = 8.3 Hz, 1 H), 6.48 (dd, J = 8.3, 2.4 Hz, 1 H), 4.67 (td, J = 6.4, 3.4 Hz, 1 H), 4.19−4.13 (m, 2 H), 3.92 (s, 2 H), 3.36−3.21 (m, 4 H), 3.02−2.90 (m, 2 H), 2.76 (dd, J = 13.4, 9.5 Hz, 1 H), 2.22 (s, 6 H), 1.26 (m, J = 7.8 Hz, 3 H), 1.17 (d, J = 6.4 Hz, 6 H), 1.08 (d, J = 7.3 Hz, 18 H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 172.59, 153.38, 150.96, 138.04, 137.86, 137.14, 135.54, 135.19, 131.61, 129.37, 128.89, 128.15, 127.29, 126.02, 124.79, 117.56, 66.11, 55.07, 37.81, 37.22, 34.21, 29.85, 26.62, 22.81, 20.22, 18.08, 13.01. HRMS exact mass calcd for $C_{40}H_{55}NO_4Si$: 641.3900. Found: 641.3905.

3-((*S*)-2-Azido-3-{4-[3-isopropyl-4-(triisopropylsilyloxy)-benzyl]-3,5-dimethylphenyl}propionyl)-(*S*)-(4)-benzyloxazolidin-2-one (21). To a stirred solution of *N*-acyloxazolidinone **20d** (290 mg, 0.45 mmol) in THF (20 mL) cooled to −78 °C was added potassium bis(trimethylsilyl)amide (0.5 M in toluene, 0.90 mL, 0.45 mmol). After the mixture was stirred for 15 min, a prechilled solution of trisylazide (Omega, Inc., 175 mg, 0.57 mmol) in THF (15 mL) was added via Teflon cannula over 1 min. After approximately 45 s from the end of the addition, acetic acid (78 μL, 1.4 mmol) was added via syringe and the reaction mixture was slowly warmed to room temperature over 2.5 h. Solvent was removed under reduced pressure, and the residue was dissolved in methylene chloride (100 mL) and washed with half-saturated $NaHCO_3$ (20 mL) and brine (20 mL). The solution was dried ($MgSO_4$), filtered through a Celite pad, and concentrated under reduced pressure. The residue was purified by flash chromatography (silica, 5% → 10% ethyl acetate in hexanes) to give **21** (226 mg, 73%) as a thick syrup. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.37−7.27 (m, 3 H), 7.22−7.18 (m, 2 H), 6.98 (s, 2 H), 6.95 (d, J = 2.0 Hz, 1 H), 6.58 (d, J = 8.3 Hz, 1 H), 6.47 (dd, J = 8.1, 2.2 Hz, 1 H), 5.29 (dd, J = 8.8, 5.9 Hz, 1 H), 4.60−4.55 (m, 1 H), 4.13 (dd, J = 9.0, 2.7 Hz, 1 H), 3.99 (t, J = 8.3 Hz, 1 H), 3.92 (dd, J = 16.1, 5.9 Hz, 2 H), 3.35−3.28 (m, 2 H), 3.14 (dd, J = 13.7, 5.9 Hz, 1 H), 3.00 (dd, J = 13.4, 9.0 Hz, 1 H), 2.83 (dd, J = 13.4, 9.5 Hz, 1 H), 2.23 (s, 6 H), 1.31−1.24 (m, 3 H), 1.16 (dd, J = 6.8, 2.0 Hz, 6 H), 1.08 (d, J = 6.8 Hz, 18 H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 170.77, 152.69, 151.04, 138.14, 137.45, 136.79, 134.71, 132.92, 131.31, 129.38, 129.01, 128.85, 127.48, 126.04, 124.78, 117.54, 66.42, 61.17, 55.40, 37.58, 37.34, 34.26, 26.62, 22.79, 20.21, 18.07, 13.02. HRMS exact mass calcd for $C_{40}H_{54}N_4O_4Si$: 682.3914. Found: 682.3932.

(*S*)-2-Azido-3-{4-[3-isopropyl-4-(triisopropylsilyloxy)-benzyl]-3,5-dimethylphenyl}propionic Acid (21a). To a stirred solution of *N*-acyloxazolidinone **21** (58 mg, 0.085 mmol) in 3:1 THF/water (4 mL) cooled to 0 °C was added hydrogen peroxide (50 wt % solution, 20 μL, 0.34 mmol) and lithium hydroxide monohydrate (7.1 mg, 0.17 mmol). After the mixture was stirred for 2.5 h, the reaction was quenched with $Na_2S_2O_3$ (0.4 M, 935 μL, 0.37 mmol) and half-saturated $NaHCO_3$ (5 mL). After acidification to pH 1 with HCl, the mixture was extracted with methylene chloride (4 × 10 mL). The combined organic fractions were washed with 1 M $NaHSO_4$ (5 mL), then dried ($MgSO_4$), filtered through a Celite pad, and concentrated under reduced pressure. Purification of the residue with flash chromatography (silica, 7.5% ethyl acetate, 1% acetic acid in hexanes) gave **21a** (37 mg, 83% yield). ^{1}H NMR (400 MHz, $CDCl_3$): δ 6.94 (s, 2 H), 6.92 (d, J = 1.95 Hz, 1 H), 6.60 (d, J = 8.30 Hz, 1 H), 6.50 (dd, J = 8.30, 1.95 Hz, 1 H), 4.14 (dd, J = 9.28, 4.88 Hz, 1 H), 3.94 (s, 2 H), 3.32 (heptet, J = 6.84 Hz, 1 H), 3.19 (dd, J = 14.16, 4.88 Hz, 1 H), 2.95 (dd, J = 13.92, 9.52 Hz, 1 H), 2.24 (s, 6 H), 1.26 (heptet, J = 7.81 Hz, 3 H), 1.15 (d, J = 6.84 Hz, 6 H), 1.09 (d, J = 7.32 Hz, 18 H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 175.48, 151.06, 138.19, 137.61, 136.85, 133.13, 131.29, 128.74, 125.94, 124.85, 117.63, 63.23, 37.22, 34.28, 26.62, 22.80, 20.26, 18.10, 13.05. HRMS exact mass calcd for $C_{30}H_{45}N_3O_3Si$: 523.3230. Found: 523.3245.

(*S*)-2-Azido-3-{4-[3-isopropyl-4-hydroxybenzyl]-3,5-dimethylphenyl}propionic Acid (21b). To a stirred solution of silyl ether **21a** (35 mg, 0.067 mmol) in THF (3 mL) was added triethylamine trihydrofluoride (1.1 mL, 6.7 mmol). After the mixture was stirred overnight, most of the THF was removed under reduced pressure. The reaction mixture was cooled in an ice bath, and the reaction was quenched with the cautious addition of potassium carbonate (1.3 g, 9.0 mmol) dissolved in water (5 mL). The solution was reacidified to pH 3 with 6 M HCl and extracted with methylene chloride (3 × 10 mL). The combined organic fractions were dried ($MgSO_4$), filtered through a Celite pad, and concentrated under reduced pressure. The residue was purified by preparative TLC (silica, 20 cm × 20 cm × 1000 μm, 8% methanol, 1% acetic acid in chloroform) to give **21b** (11 mg, 44% yield). ^{1}H NMR (400 MHz, CD_3OD): δ 6.95 (s, 2 H), 6.80 (d, J = 2.0 Hz, 1 H), 6.56 (d, J = 8.3 Hz, 2 H), 6.50 (dd, J = 8.3, 2.0 Hz, 1 H), 4.13 (br s, 1 H), 3.91 (s, 2 H), 3.19 (heptet, J = 6.8 Hz, 1 H), 3.11 (dd, J = 13.9, 4.2 Hz, 1 H), 2.90 (dd, J = 13.4, 8.6 Hz, 1 H), 2.19 (s, 6 H), 1.11 (d, J = 6.8 Hz, 6 H). ^{13}C NMR (100 MHz, CD_3OD): δ 153.41, 138.26, 137.60, 135.86, 135.42, 131.57, 129.97, 126.66,

126.36, 115.83, 38.38, 34.87, 28.00, 23.04, 20.29. HRMS exact mass calcd for $C_{21}H_{25}N_3O_3$: 367.1896. Found: 367.1936.

(*S*)-2-Amino-3-{4-[3-isopropyl-4-hydroxybenzyl]-3,5-dimethylphenyl}propionic Acid (8). A solution of azidocarboxylic acid **21b** (11 mg, 0.11 mmol) in 2:1 acetic acid/water (3 mL) was sparged with Ar, then palladium on carbon (10%, 22 mg) was added, and the mixture was stirred overnight under H_2 atmosphere maintained with balloon pressure. The reaction mixture was filtered through a small plug of cotton, concentrated under reduced pressure, and dried in vacuo to give **8** (10 mg, 97% yield) as a white solid. ^{1}H NMR (400 MHz, CD_3OD): δ 6.99 (s, 2 H), 6.87 (s, 1 H), 6.56 (d, J = 8.3 Hz, 1 H), 6.51 (dd, J = 8.3, 2.0 Hz, 1 H), 3.90 (d, J = 7.8 Hz, 2 H), 3.76 (br s, 1 H), 3.27−3.16 (m, 2 H), 2.91 (dd, J = 13.7, 8.8 Hz, 1 H), 2.22 (s, 6 H), 1.12 (d, J = 7.3 Hz, 6 H). ^{13}C NMR (100 MHz, CD_3OD): δ 169.33, 153.54, 138.73, 138.12, 135.88, 134.73, 131.47, 130.03, 126.94, 126.36, 115.85, 34.98, 28.13, 23.04, 20.38. HRMS exact mass calcd for $C_{21}H_{27}NO_3$: 341.1991. Found: 341.1993.

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Supporting Information Available: Analytical HPLC chromatograms of compounds **7**−**9**. This material is available free of charge via the Internet at http://pubs.acs.org.

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