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Synthesis of (*S*)-5-(1-aminoethyl)-2-(cyclohexylmethoxy)benzamide

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Abstract

Three short syntheses of the title compound, a peptidomimetic for the Glu-Glu-Ile-NH_2 portion of Ac-pTyr-Glu-Glu-Ile-NH_2, a high affinity peptide for the Src SH2 domain, are described. The most efficient route produces the title compound in a final enantiopurity of 94% ee.

1. Introduction

In recent years, Src homology 2 (SH2) domain proteins have been targeted by a number of drug discovery programs for the development of therapeutic agents against a number of diseases.[1] As part of our efforts to develop novel Src SH2 inhibitors,[2] we were interested in identifying novel replacements for the phosphotyrosine (pTyr) portion of tetrapeptide **1** (Fig. 1), a high affinity ligand for the Src family SH2 domains. The ultimate goal is to develop a completely non-peptidic Src SH2 inhibitor; therefore, we were also interested in initially examining these pTyr replacements attached to a novel non-peptidic (1-aminoethyl)benzamide replacement for the Glu-Glu-Ile-NH_2 'tail' portion of **1** recently reported by workers at Parke-Davis.[1,3] Compound **2**, which is simply pTyr attached to this novel non-peptidic 'tail', displays improved binding affinity over **1**,[1] while compound **3**, which possesses the same 'tail' but attached to a 4-carboxyphenyl phosphate 'head' group, displays comparable binding affinity as **1**.[3] The syntheses of **2** and **3** employed racemic amine **4**, and the resulting stereoisomers were not separated.[3,4] It was assumed in the case of **3**, based on modeling, that the eutomer possesses the (*S*) stereochemistry at the stereogenic benzylic center.

We first wanted to confirm the findings of Parke-Davis; therefore, a small amount of **2** was prepared as a diastereomeric mixture from L-Tyr and *rac*-**4** in a manner analogous to that described previously.[3,4] The two diastereomers were carefully separated by reversed-phase HPLC and tested in an Src SH2 binding assay.[5] It turns out that one diastereomer displays more than 100-fold higher affinity over the other.[6] We

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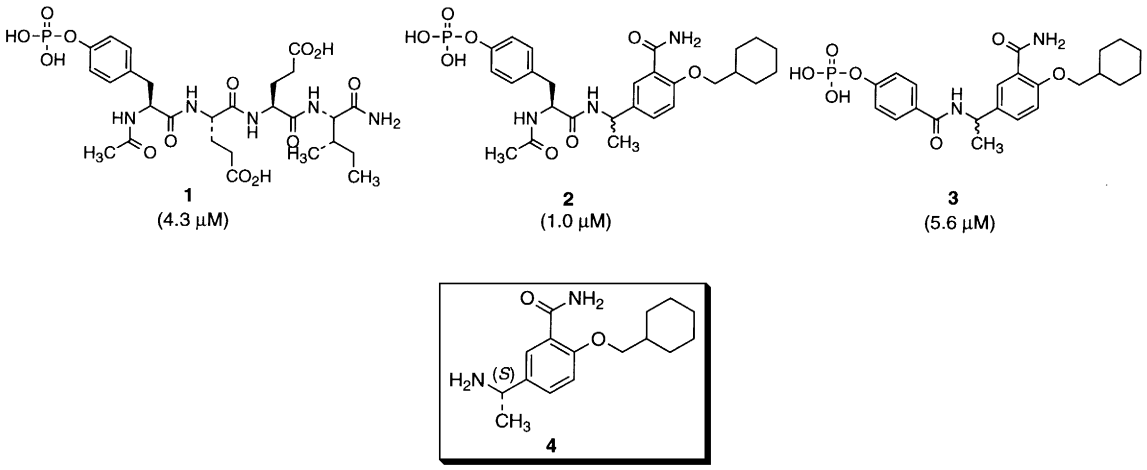


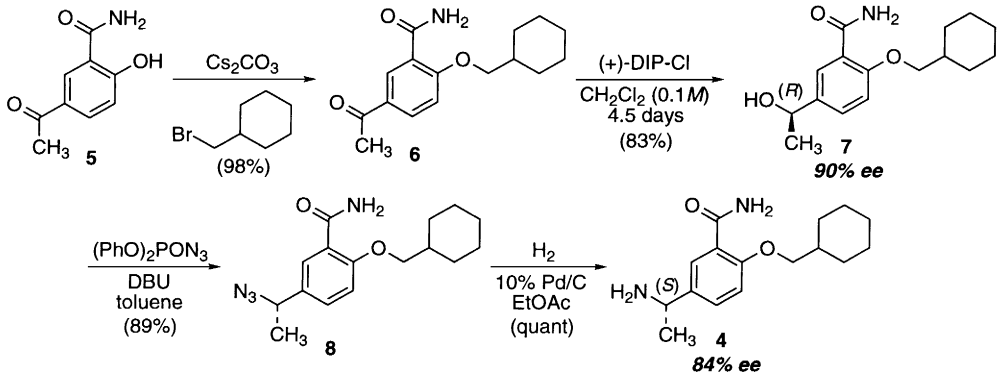
Fig. 1.

still, though, needed to determine conclusively that the diastereomer that exhibits the strongest affinity for the Src SH2 possesses the (*S*) stereochemistry at the stereogenic benzylic center.[7] Furthermore, in order to expedite our study of pTyr replacements in **2**, we required a supply of enantiomerically enriched (*S*)-amine **4**. This paper describes a few of the routes we examined for the preparation of (*S*)-**4**.

2. Results and discussion

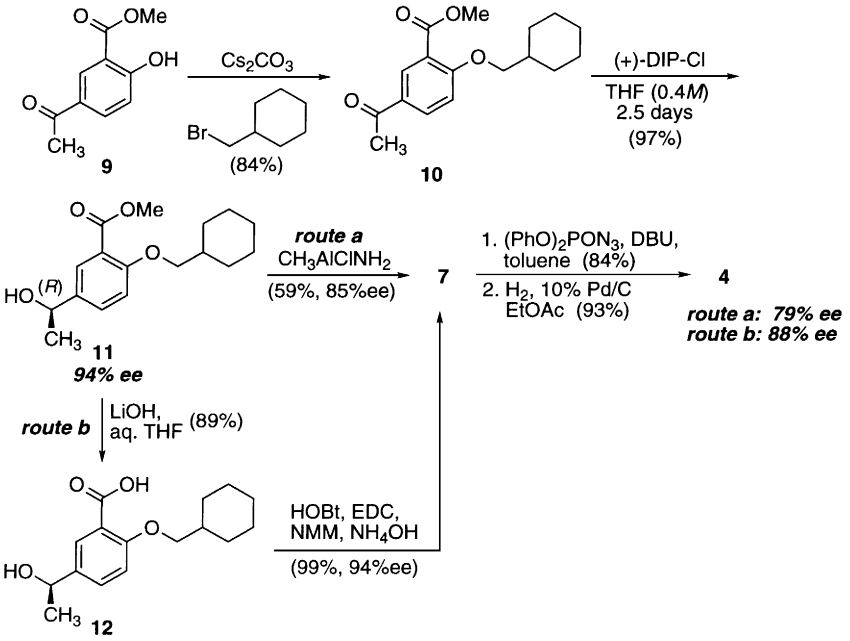
From the outset, it appeared that the most expedient route to (*S*)-**4** should employ an asymmetric reduction of acetophenone **6**, which is readily prepared by alkylation of commercially available 5-acetylsalicylamide **5** (Scheme 1).[3,4] The highly stereoselective reduction of phenyl ketones using (+)- or (−)-chlorodiisopinocampheylborane (DIP-Cl) is well precedented,[9] and for this reason, we chose to employ this methodology. The prescribed procedure for such reductions employs THF as the solvent, and the reactions are typically carried out at high concentration. Unfortunately, acetophenone **6** was only very slightly soluble in THF, so we had to resort to CH_2Cl_2 as the solvent.[10] Still, **6** was only modestly soluble in CH_2Cl_2, and the reaction therefore had to be carried out at a final concentration of 0.1 M. Thus, **6** was reduced using (+)-DIP-Cl using these modified conditions, affording alcohol **7** in 90% ee. Alcohol **7** was then converted to azide **8** following the Merck protocol,[11] and the azide was finally reduced, producing amine **4** in 84% ee. The erosion in enantiomeric purity is consistent with the observations by the Merck group, which saw some erosion for electron rich substrates. The absolute configurations and enantiomeric purities of alcohol **7** and amine **4** were confirmed by analysis of the ^1H and ^{19}F NMR spectra of the respective Mosher esters and amides.[12] Additionally, the enantiomeric purity of amine **4** was confirmed using a capillary electrophoresis method.[13]

We wanted to improve the enantiomeric purity of amine **4**, and rather than investigate alternative asymmetric reduction procedures, which typically employ THF as the solvent,[9c] we decided instead to modify the starting acetophenone so as to improve its solubility in THF. Suspecting that the primary amide functionality was the source of the insolubility, we chose to replace it with an ester functionality. Thus, commercially available methyl 5-acetylsalicylate **9** was converted to ether **10**, which turned out to be readily soluble in THF (Scheme 2). Reduction of acetophenone **10** using (+)-DIP-Cl produced alcohol **11** in a somewhat improved 94% ee. Furthermore, the reaction time for the reduction was shortened from 4.5 days to 2.5 days. We next needed to convert the ester functionality to the primary amide. We



Scheme 1.

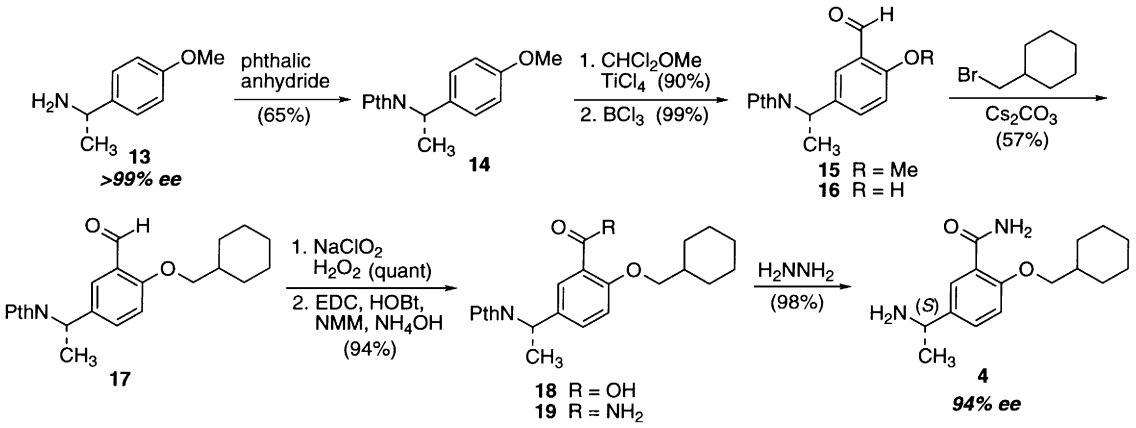
first examined a one step protocol using methylchloroaluminum amide.[14] Indeed, amide **7** was readily produced utilizing this methodology but with some erosion in enantiomeric purity, and amine **4** was ultimately obtained in 79% ee. We therefore resorted to a two step hydrolysis–amination conversion of ester **11** to amide **7**. Following this sequence, amine **4** was ultimately obtained in 88% ee.



Scheme 2.

Subsequent to our investigations of the asymmetric reductions, we became aware of the commercial availability of both enantiomers of nonracemic 1-(4-methoxyphenyl)ethylamine **13** (Scheme 3).[15] We felt that the (*S*)-enantiomer could be readily elaborated to our desired amine **4**. The phthaloyl protecting group was chosen in anticipation of the use of rather harsh Lewis acids later in the sequence. Thus, amine **13** was first protected as phthalimide **14**, which was then formylated *ortho* to the ether functionality. Ether **15** was then de-methylated, and the resulting salicylaldehyde **16** was alkylated to produce ether **17**. The aldehyde was then oxidized[16] to produce acid **18**, which was then aminated affording amide **19**. The phthaloyl protecting group was finally removed using hydrazine, producing amine **4** in 94% ee.

In order to confirm that (*S,S*)-**2** is the most active of the two diastereomers that were originally



Scheme 3.

separated, we next prepared a sample of (*S*,*S*)-**2** from L-Tyr and (*S*)-**4**. Indeed, (*S*,*S*)-**2** prepared from (*S*)-**4** is identical (NMR and RP-HPLC) to the most active diastereomer isolated from the diastereomeric mixture prepared from *rac*-**4**.

3. Conclusion

In summary, we have described three stereoselective syntheses of (*S*)-5-(1-aminoethyl)-2-(cyclohexylmethoxy)benzamide **4**. The most efficient utilizes commercially available nonracemic (*S*)-1-(4-methoxyphenyl)ethylamine **13** and produces **4** in 94% ee. It has been confirmed that the most active diastereomer in the case of **2** is the one possessing the (*S*) stereochemistry at the stereogenic benzylic center. Results of an examination of novel pTyr replacements in the context of (*S*,*S*)-**2** will be discussed in subsequent papers.

4. Experimental

*4.1. 5-Acetyl-2-(cyclohexylmethoxy)benzamide **6***

To a mixture of 5-acetylsalicylamide (4.48 g, 25.0 mmol) in MeOH (200 mL) containing H_2O (5 mL) was added Cs_2CO_3 (8.23 g, 25.25 mmol). After 30 min, the yellow solution was concentrated in vacuo. Next, toluene (150 mL) was added to the residue and then removed in vacuo in order to azeotropically remove the water; this was repeated once more. The resulting solid was then suspended in DMF (150 mL) and treated with (bromomethyl)cyclohexane (3.66 mL, 26.25 mmol). The mixture was then stirred at 90°C for 24 h. Additional (bromomethyl)cyclohexane (1.7 mL) was added, and the mixture was stirred at 90°C for an additional 18 h. After cooling to rt, the reaction mixture was poured into 5% aq NaOH (100 mL) and extracted with EtOAc (not all of the product dissolved in the EtOAc, but simply stayed suspended in the organic layer). The organic layer was washed with H_2O (3×100 mL) and brine (1×100 mL). The aqueous washes were re-extracted once with EtOAc, and the combined extracts were concentrated in vacuo. The solid was dissolved in $CHCl_3$, and the resulting solution was dried over $MgSO_4$ and concentrated in vacuo. The solid was then thoroughly washed with Et_2O to remove the excess bromide and trace DMF. The solid was dried over P_2O_5 under high vacuum to yield 6.75 g (98%) of pure ether **6**. ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.79 (d, J=2.3 Hz, 1H), 8.12 (dd, J=8.8, 2.3 Hz, 1H), 7.72

(br, 1H), 7.05 (d, J=8.8 Hz, 1H), 6.22 (br, 1H), 4.03 (d, J=6.0 Hz, 2H), 2.62 (s, 3H), 1.91–1.73 (m, 6H), 1.40–1.06 (m, 5H).

4.2. (R)-2-(Cyclohexylmethoxy)-5-(1-hydroxyethyl)benzamide **7** from **6**

To a solution of (+)-DIP-Cl (5.98 g, 18.6 mmol) in CH_2Cl_2 (25 mL) at −35°C under N_2 was added, via cannula, a solution of ketone **6** (2.56 g, 9.30 mmol) in CH_2Cl_2 (66 mL). The resulting solution was kept at −15°C for 4.5 days (reaction checked after 2.5 days, but some unreacted **6** remained). The reaction mixture was then concentrated, and the residue was dissolved in Et_2O (100 mL) and treated with diethanolamine (3.6 mL, 37.2 mmol). The mixture was vigorously stirred for 3.5 h and then filtered through a pad of Celite. The filtrate was allowed to stand overnight at rt. An additional needle-like precipitate formed. The mixture was filtered and the filtrate was concentrated. The residue was dissolved in a small amount of $CHCl_3$ and purified by flash chromatography on silica gel. Elution with 2:1 EtOAc:hexanes followed by 3:1 EtOAc:hexanes and finally 100% EtOAc afforded 2.15 g (83%) of alcohol **7** as a white solid. Enantiomeric excess=90% (as determined by Mosher ester analysis). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.17 (d, J=2.4 Hz, 1H), 7.82 (br, 1H), 7.52 (dd, J=8.6, 2.4 Hz, 1H), 6.97 (d, J=8.6 Hz, 1H), 5.89 (br, 1H), 4.91 (m, 1H), 3.94 (d, J=5.9 Hz, 1H), 1.91–1.72 (m, 6H), 1.49 (d, J=6.4 Hz, 3H), 1.38–1.04 (m, 5H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 167.4, 156.7, 138.6, 130.3, 129.6, 120.2, 112.5, 74.7, 69.4, 37.5, 30.0, 26.2, 25.6, 25.0.

4.3. (R)-MTPA ester of **7**

^{1}H NMR (300 MHz, $CDCl_3$) δ 8.24 (d, J=2.4 Hz, 1H), 7.86 (br, 1H), 7.49–7.31 (m, 6H), 6.96 (d, J=8.6 Hz, 1H), 6.10 (m, 2H), 3.95 (d, J=5.8 Hz, 2H), 3.47 (s, 3H), 1.88–1.73 (m, 6H), 1.57 (d, J=6.6 Hz, 3H), 1.40–1.05 (m, 5H); ^{19}F NMR (282 MHz, $CDCl_3$) δ −72.3.

4.4. (S)-MTPA ester of **7**

^{1}H NMR (300 MHz, $CDCl_3$) δ 8.15 (d, J=2.3 Hz, 1H), 7.84 (br, 1H), 7.44–7.30 (m, 6H), 6.91 (d, J=8.6 Hz, 1H), 6.09 (m, 2H), 3.93 (d, J=5.8 Hz, 2H), 3.54 (s, 3H), 1.88–1.72 (m, 6H), 1.64 (d, J=6.6 Hz, 3H), 1.40–1.05 (m, 5H); ^{19}F NMR (282 MHz, $CDCl_3$) δ −72.5.

4.5. (S)-5-(1-Azidoethyl)-2-(cyclohexylmethoxy)benzamide **8**

To a mixture of alcohol **7** (1.76 g, 6.35 mmol) and diphenylphosphoryl azide (1.64 mL, 7.61 mmol) in toluene (26 mL) at 0°C under N_2 was added DBU (1.14 mL, 7.61 mmol). The reaction mixture was stirred overnight while slowly warming to rt, and then diluted with EtOAc and washed with 5% aq HCl (2×50 mL), H_2O (1×50 mL), and brine (1×50 mL). The aqueous washes were re-extracted once with EtOAc, and the combined extracts were dried over $MgSO_4$ and concentrated. The crude material was purified by flash chromatography on silica gel. Elution with 2:1 hexanes:EtOAc followed by 3:2 hexanes:EtOAc afforded material that was still not quite pure. The material was purified again by flash chromatography on silica gel using the same eluents affording 1.71 g (89%) of pure azide **8**. ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.17 (d, J=2.3 Hz, 1H), 7.83 (br, 1H), 7.45 (dd, J=8.5, 2.3 Hz, 1H), 6.99 (d, J=8.6 Hz, 1H), 5.89 (br, 1H), 4.64 (q, J=6.8 Hz, 1H), 3.95 (d, J=5.8 Hz, 2H), 1.89–1.73 (m, 6H), 1.52 (d, J=6.8 Hz, 3H), 1.39–1.05 (m, 5H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 166.9, 157.2, 133.4, 131.1, 130.7, 120.8, 112.7, 74.8, 60.3, 37.5, 30.0, 26.2, 25.6, 21.4.

4.6. (S)-5-(1-Aminoethyl)-2-(cyclohexylmethoxy)benzamide **4** from **8**

A solution of azide **8** (4.04 g, 13.4 mmol) in EtOAc (130 mL) containing 10% Pd/C (700 mg, 0.67 mmol) was vigorously stirred under an atmosphere of H_2 (double stuffed balloon) for 4 h. The mixture was then filtered through a pad of Celite. The filtrate was concentrated and the residue was dissolved in 0.5 M aq HCl (100 mL). The solution was then washed with EtOAc (2×50 mL). The aqueous layer was basified by the addition of 6 M aq NaOH and then extracted twice with CH_2Cl_2. The combined extracts were dried over K_2CO_3 and concentrated to 3.44 g (93%) of amine **4** as a white solid. ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.17 (d, J=2.1 Hz, 1H), 7.85 (br, 1H), 7.47 (dd, J=8.5, 2.2 Hz, 1H), 6.94 (d, J=8.5 Hz, 1H), 6.01 (br, 1H), 4.15 (q, J=6.6 Hz, 1H), 3.93 (d, J=5.9 Hz, 2H), 1.92–1.72 (m, 8H), 1.39 (d, J=6.6 Hz, 3H), 1.34–1.04 (m, 5H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 167.4, 156.3, 140.3, 130.4, 129.6, 120.5, 112.5, 74.7, 50.4, 37.5, 29.9, 26.2, 25.6, 25.5. HRMS-Electrospray (*m/z*): $[M+H]^+$ calcd for $C_{16}H_{25}N_2O_2$, 277.1916; found, 277.1924.

4.7. (R)-MTPA amide of **4**

^{1}H NMR (300 MHz, $CDCl_3$) δ 8.16 (d, J=2.4 Hz, 1H), 7.91 (br, 1H), 7.42–7.31 (m, 6H), 7.08 (d, J=7.9 Hz, 1H), 6.90 (d, J=8.6 Hz, 1H), 6.32 (br, 1H), 5.12 (pent, J=7.1 Hz, 1H), 3.92 (d, J=5.9 Hz, 2H), 3.38 (d, J=1.2 Hz, 3H), 1.87–1.72 (m, 6H), 1.55 (d, J=6.9 Hz, 3H), 1.38–1.04 (m, 5H); ^{19}F NMR (282 MHz, $CDCl_3$) δ −69.7.

4.8. (S)-MTPA amide of **4**

^{1}H NMR (300 MHz, $CDCl_3$) δ 8.20 (d, J=2.4 Hz, 1H), 7.92 (br, 1H), 7.55–7.40 (m, 6H), 7.00 (d, J=8.3 Hz, 1H), 6.95 (d, J=8.6 Hz, 1H), 6.31 (br, 1H), 5.15 (pent, J=7.5 Hz, 1H), 3.94 (d, J=5.9 Hz, 2H), 3.37 (d, J=1.2 Hz, 3H), 1.89–1.72 (m, 6H), 1.51 (d, J=6.9 Hz, 3H), 1.39–1.04 (m, 5H); ^{19}F NMR (282 MHz, $CDCl_3$) δ −69.5.

4.9. Methyl 5-acetyl-2-(cyclohexylmethoxy)benzoate **10**

To a solution of methyl 5-acetylsalicylate (5.05 g, 26.0 mmol) in MeOH (200 mL) containing some H_2O (5 mL) at rt was added Cs_2CO_3 (8.56 g, 26.3 mmol). The mixture was stirred at rt for 45 min and then concentrated in vacuo. Toluene was added to the residue and then removed in vacuo in order to azeotropically remove H_2O. This was repeated once more. The resulting light yellow solid was suspended in DMF (150 mL) and treated with (bromomethyl)cyclohexane (3.8 mL, 27.3 mmol). The suspension was stirred at 90 °C for 16 h whereupon additional bromide (0.38 mL) was added. The reaction mixture was stirred at 90°C for an additional 2 h. After cooling to rt, the reaction mixture was poured into 5% aq NaOH (100 mL) and extracted with Et_2O. The Et_2O extract was washed with additional H_2O (2×100 mL) and brine (1×100 mL). The aqueous washes were re-extracted once with Et_2O, and the combined extracts were dried over $MgSO_4$ and concentrated. The crude oil was purified by flash chromatography on silica gel. Elution with 7:1 hexanes:EtOAc, 6:1 hexanes:EtOAc, and finally 5:1 hexanes:EtOAc afforded 6.33 g (84%) of ether **10**. ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.39 (d, J=2.3 Hz, 1H), 8.07 (dd, J=8.8, 2.3 Hz, 1H), 6.99 (d, J=8.8 Hz, 1H), 3.91 (s, 3H), 3.90 (d, J=6.0 Hz, 2H), 2.57 (s, 3H), 1.91–1.70 (m, 6H), 1.38–1.06 (m, 5H).

4.10. (R)-Methyl 2-(cyclohexylmethoxy)-5-(1-hydroxyethyl)benzoate **11**

To a solution of (+)-DIP-Cl (5.64 g, 17.6 mmol) in THF (17 mL) at ~−55°C was slowly added, via cannula, a solution of acetophenone **10** (3.32 g, 11.4 mmol) in 10 mL of THF. The reaction mixture, as well as the cold bath in which the reaction flask was sitting, was placed in a freezer at −13°C. After 2.5 days, the reaction mixture was warmed to 0°C and slowly treated with acetaldehyde (2 mL). The reaction mixture was then stirred at rt for an additional 4 h and then treated with 1 M aq NaOH (60 mL). The resulting mixture was stirred vigorously for 10 min and then extracted with Et_2O. The extract was washed with water and brine. The aqueous washes were re-extracted once with Et_2O, and the combined extracts were dried over $MgSO_4$ and concentrated. The crude material was purified by flash chromatography on silica gel. Elution with 4:1 hexanes:EtOAc, 3:1 hexanes:EtOAc, and finally 2:1 hexanes:EtOAc afforded 3.23 g (97%) of alcohol **11**. Enantiomeric excess=94% (as determined by Mosher ester analysis). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.77 (d, J=2.3 Hz, 1H), 7.46 (dd, J=8.6, 2.3 Hz, 1H), 6.93 (d, J=8.6 Hz, 1H), 4.87 (q, J=6.4 Hz, 1H), 3.89 (s, 3H), 3.82 (d, J=6.0 Hz, 2H), 1.91–1.70 (m, 6H), 1.48 (d, J=6.4 Hz, 3H), 1.37–1.03 (m, 5H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 166.9, 158.1, 137.4, 130.4, 128.7, 120.1, 113.2, 74.4, 69.4, 51.8, 37.6, 29.7, 26.5, 25.8, 25.0.

4.11. (R)-MTPA ester of **11**

^{1}H NMR (300 MHz, $CDCl_3$) δ 7.80 (br, 1H), 7.42–7.34 (m, 6H), 6.92 (d, J=8.6 Hz, 1H), 6.08 (q, J=6.4 Hz, 1H), 3.88 (s, 3H), 3.82 (d, J=5.5 Hz, 2H), 3.45 (s, 3H), 1.91–1.63 (m, 6H), 1.56 (d, J=6.4 Hz, 3H), 1.37–1.04 (m, 5H); ^{19}F NMR (282 MHz, $CDCl_3$) δ −72.2.

4.12. (S)-MTPA ester of **11**

^{1}H NMR (300 MHz, $CDCl_3$) δ 7.67 (br, 1H), 7.37–7.32 (m, 6H), 6.87 (d, J=8.6 Hz, 1H), 6.04 (q, J=6.5 Hz, 1H), 3.86 (s, 3H), 3.81 (d, J=5.5 Hz, 2H), 3.55 (s, 3H), 1.91–1.69 (m, 6H), 1.62 (d, J=6.5 Hz, 3H), 1.37–1.07 (m, 5H); ^{19}F NMR (282 MHz, $CDCl_3$) δ −72.5.

4.13. (R)-2-(Cyclohexylmethoxy)-5-(1-hydroxyethyl)benzoic acid **12**

To a solution of ester **11** (3.14 g, 10.7 mmol) in THF (22 mL) at 0°C was added a solution of $LiOH \cdot H_2O$ (496 mg, 11.8 mmol) in H_2O (12 mL). The biphasic mixture was vigorously stirred for 20 h at rt. The yellow solution was then washed with Et_2O (2×50 mL). The aqueous layer was then acidified with 1.0 M aq HCl and extracted with EtOAc. The extract was washed with H_2O (25 mL) and brine (25 mL). The aqueous washes were re-extracted once with EtOAc, and the combined extracts were dried over $MgSO_4$ and concentrated to 2.64 g (89%) of acid **12** as an off-white solid. ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.15 (d, J=2.1 Hz, 1H), 7.62 (dd, J=8.6, 2.1 Hz, 1H), 7.03 (d, J=8.6 Hz, 1H), 4.92 (q, J=6.4 Hz, 1H), 4.05 (d, J=6.0 Hz, 2H), 1.89–1.73 (m, 6H), 1.50 (d, J=6.4 Hz, 3H), 1.35–1.09 (m, 5H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 165.6, 156.8, 139.7, 132.1, 130.6, 117.1, 112.7, 75.6, 69.1, 37.3, 29.7, 26.1, 25.5, 25.1.

4.14. (R)-2-(Cyclohexylmethoxy)-5-(1-hydroxyethyl)benzamide **7** from **12**

To a solution of acid **12** (4.87 g, 17.5 mmol) in THF (50 mL) at 0°C under N_2 was added NMM (2.9 mL, 26.2 mmol), HOBt (3.55 g, 26.2 mmol), and EDC (5.03 g, 26.2 mmol). After stirring at 0°C for 1

h, the reaction mixture was treated with ~4–4.5 mL of concentrated NH_4OH. The reaction mixture was allowed to slowly warm to rt. After 5 h, the reaction mixture was diluted with EtOAc and washed with 1.0 M aq HCl (2×50 mL), half saturated aq $NaHCO_3$ (2×50 mL), H_2O (1×50 mL), and brine (1×50 mL). The EtOAc extract was dried over $MgSO_4$ and concentrated to 4.80 g (99%) of amide **7**, which needed no further purification.

4.15. (S)-2-[1-(4-Methoxyphenyl)ethyl]-1H-isoindole-1,3(2H)-dione **14**

A solution of amine **13** (2.44 g, 16.1 mmol) and phthalic anhydride (2.34 g, 15.8 mmol) in $CHCl_3$ (40 mL) was stirred at reflux. The reaction was monitored by ^1H NMR. Formation of the amide-acid was relatively fast (a few hours); however, formation of the phthalimide was very slow. After stirring at reflux for 4 days (reaction still not complete), the reaction mixture was concentrated. The residue was diluted with EtOAc and washed with 1.0 M aq HCl (1×50 mL), half saturated aq $NaHCO_3$ (2×50 mL), H_2O (1×50 mL), and brine (1×50 mL). The EtOAc extract was dried over $MgSO_4$ and concentrated to 2.96 g (65%) of phthalimide **14**, which needed no further purification. ^1H NMR (300 MHz, $CDCl_3$) δ 7.80–7.76 (m, 2H), 7.70–7.66 (m, 2H), 7.45 (d, J=8.7 Hz, 2H), 6.85 (d, J=8.7 Hz, 2H), 5.53 (q, J=7.3 Hz, 1H), 3.77 (s, 3H), 1.90 (d, J=7.3 Hz, 3H).

4.16. (S)-5-[1-(1,3-Dioxoisoindolin-2-yl)ethyl]-2-methoxybenzaldehyde **15**

To a solution of aryl ether **14** (1.59 g, 5.65 mmol) in CH_2Cl_2 (15 mL) at 0°C under N_2 was added $TiCl_4$ (1.92 mL, 17.5 mmol). The resulting thick brown slurry was diluted with additional CH_2Cl_2 (15 mL) (in order to facilitate stirring) and then cooled to −78°C. Next, $CHCl_2OMe$ (0.77 mL, 8.48 mmol) was added. After 5 min, the reaction mixture was warmed to 0°C. After stirring at 0°C for 75 min, the dark green solution was diluted with 1.0 M aq HCl (50 mL). The mixture was then extracted with EtOAc. The extract was washed with H_2O (1×50 mL) and brine (1×50 mL), dried over $MgSO_4$, and concentrated to 1.58 g (90%) of aldehyde **15** as a colorless solid. ^1H NMR (300 MHz, $CDCl_3$) δ 10.43 (s, 1H), 7.93 (d, J=2.4 Hz, 1H), 7.82–7.77 (m, 2H), 7.74–7.66 (m, 3H), 6.96 (d, J=8.7 Hz, 1H), 5.55 (q, J=7.3 Hz, 1H), 3.91 (s, 3H), 1.90 (d, J=7.3 Hz, 3H).

4.17. (S)-5-[1-(1,3-Dioxoisoindolin-2-yl)ethyl]salicylaldehyde **16**

To a solution of aryl ether **15** (1.53 g, 4.95 mmol) in CH_2Cl_2 (20 mL) at −10°C under N_2 was slowly added BCl_3 (25mL of 1.0 M solution in CH_2Cl_2). The dark solution was stirred at rt for 24 h. It was then cooled to 0°C and quenched by the careful addition of 1.0 M aq HCl (50 mL). The mixture was stirred vigorously for 5 min and then extracted with EtOAc. The EtOAc extract was washed with H_2O (50 mL) and brine (50 mL). The aqueous washes were re-extracted once with EtOAc, and the combined extracts were dried over $MgSO_4$ and concentrated. The crude material was purified by flash chromatography on silica gel. Elution with 5:1 hexanes:EtOAc followed by 4:1 hexanes:EtOAc, and finally 3:1 hexanes:EtOAc afforded 1.46 g (99%) of salicylaldehyde **16** as a colorless solid. ^1H NMR (300 MHz, $CDCl_3$) δ 10.98 (s, 1H), 9.90 (s, 1H), 7.83–7.78 (m, 2H), 7.74–7.67 (m, 4H), 6.95 (d, J=8.6 Hz, 1H), 5.55 (q, J=7.3 Hz, 1H), 1.93 (d, J=7.3 Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 196.5, 168.0, 161.0, 136.5, 134.0, 132.6, 131.9, 131.8, 123.2, 120.1, 117.7, 48.6, 17.4. Electrospray mass spectrum (50:50 acetonitrile:water + 0.1% NH_4OH): *m/z* 294 [M−H].

4.18. (S)-5-[1-(1,3-Dioxoisoindolin-2-yl)ethyl]-2-(cyclohexylmethoxy)benzaldehyde **17**

To a mixture of salicylaldehyde **16** (1.395 g, 4.72 mmol) in MeOH (40 mL)–H_2O (1 mL) at rt was added Cs_2CO_3 (1.62 g, 4.96 mmol). After stirring for 30 min, the resulting yellow solution was concentrated in vacuo. Toluene was added to the residue and then removed in vacuo in order to azeotropically remove the H_2O. This was repeated once more. The residue was then suspended in DMF (20 mL) and treated with (bromomethyl)cyclohexane (0.79 mL, 5.67 mmol). The mixture was stirred at 90°C for 2 h. After cooling to rt, the reaction mixture was diluted with H_2O (50 mL) and extracted with EtOAc. The extract was washed with additional H_2O (2×50 mL) and brine (1×50 mL). The aqueous washes were re-extracted once with EtOAc, and the combined extracts were dried over $MgSO_4$ and concentrated. The crude material was purified by flash chromatography on silica gel. Elution with 7:1 hexanes:EtOAc followed by 6:1 hexanes:EtOAc afforded 1.05 g (57%) of ether **17**. Some impure fractions were not collected. ^{1}H NMR (300 MHz, $CDCl_3$) δ 10.49 (s, 1H), 7.92 (d, J=2.4 Hz, 1H), 7.81–7.77 (m, 2H), 7.71–7.66 (m, 3H), 6.93 (d, J=8.7 Hz, 1H), 5.54 (q, J=7.3 Hz, 1H), 3.85 (d, J=5.8 Hz, 2H), 1.94–1.69 (m, 6H), 1.89 (d, J=7.3 Hz, 3H), 1.32–1.03 (m, 5H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 189.5, 167.9, 161.0, 134.9, 133.9, 132.3, 131.9, 127.1, 124.5, 123.2, 112.6, 73.9, 48.5, 37.6, 29.7, 26.3, 25.7, 17.4. Electrospray mass spectrum (50:50 acetonitrile:water + 0.1% NH_4OH): *m/z* 392 [M+H].

4.19. (S)-5-[1-(1,3-Dioxoisoindolin-2-yl)ethyl]-2-(cyclohexylmethoxy)benzoic acid **18**

A solution of aldehyde **17** (222 mg, 0.567 mmol) in CH_3CN (1.2 mL) was diluted with a solution of $NaH_2PO_4 \cdot 2H_2O$ (24 mg, 0.153 mmol) in H_2O (0.5 mL). The mixture was cooled to 0°C and treated with 50% H_2O_2 (34 μL, 0.595 mmol) followed by a solution of $NaClO_2$ (90 mg, 0.794 mmol) in H_2O (1.6 mL). The mixture was then stirred at rt for 20 h. Next, some solid $NaHSO_3$ was added to the reaction mixture to destroy the unreacted HOCl and H_2O_2. The mixture was acidified with 1.0 M aq HCl and extracted with EtOAc. The extract was washed with H_2O and brine, dried over $MgSO_4$, and concentrated to 230 mg (100%) of acid **18** as a foam. ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.27 (d, J=2.4 Hz, 1H), 7.82–7.78 (m, 2H), 7.72–7.68 (m, 3H), 7.00 (d, J=8.7 Hz, 1H), 5.57 (q, J=7.3 Hz, 1H), 4.02 (d, J=6.0 Hz, 2H), 1.96–1.71 (m, 6H), 1.90 (d, J=7.3 Hz, 3H), 1.39–1.03 (m, 5H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 167.8, 165.1, 157.0, 134.0, 133.9, 132.6, 131.8, 123.2, 117.4, 112.6, 75.6, 48.3, 37.2, 29.7, 26.1, 25.5, 17.4. Electrospray mass spectrum (50:50 acetonitrile:water + 0.1% NH_4OH): *m/z* 406 [M−H].

4.20. (S)-5-[1-(1,3-Dioxoisoindolin-2-yl)ethyl]-2-(cyclohexylmethoxy)benzamide **19**

To a solution of acid **18** (223 mg, 0.547 mmol) and NMM (0.09 mL, 0.821 mmol) in THF (5 mL) at 0°C was added HOBt·H_2O (126 mg, 0.821 mmol) followed by EDC (157 mg, 0.821 mmol). The reaction mixture was stirred at 0°C for 44 min and then treated with 14 drops of concentrated NH_4OH. The reaction mixture was stirred overnight while slowly warming to rt. The mixture was then diluted with EtOAc and washed with 1.0 M aq HCl (2×10 mL), half saturated aq $NaHCO_3$ (2×10 mL), H_2O (1×10 mL), and brine (1×10 mL). The EtOAc extract was dried over $MgSO_4$ and concentrated. The crude material was purified by flash chromatography on silica gel. Elution with 1:1 EtOAc:hexanes followed by 2:1 EtOAc:hexanes afforded 208 mg (94%) of amide **19** as a colorless solid. ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.28 (d, J=2.4 Hz, 1H), 7.81–7.74 (m, 3H), 7.71–7.65 (m, 2H), 7.62 (dd, J=8.6, 2.5 Hz, 1H), 6.93 (d, J=8.7 Hz, 1H), 5.96 (br, 1H), 5.57 (q, J=7.3 Hz, 1H), 3.91 (d, J=5.9 Hz, 2H), 1.90 (d, J=7.3 Hz, 3H), 1.86–1.71 (m, 6H), 1.37–1.02 (m, 5H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 167.9, 167.0, 156.8, 133.8,

132.7, 132.1, 131.9, 131.5, 123.1, 120.6, 112.3, 74.6, 48.5, 37.4, 29.9, 26.2, 25.6, 17.6. Electrospray mass spectrum (50:50 acetonitrile:water+0.1% NH_4OH): *m/z* 405 [M−H].

*4.21. (S)-5-(1-Aminoethyl)-2-(cyclohexylmethoxy)benzamide **4** from **19***

To a mixture of phthalimide **19** (177 mg, 0.435 mmol) in EtOH (5 mL) at rt was added 10 drops of hydrazine hydrate. The mixture was stirred at rt overnight and then concentrated. The residue was partitioned between EtOAc and 0.5 M aq HCl (25 mL). The layers were separated and the EtOAc wash was re-extracted once with 0.5 M aq HCl (8 mL). The aqueous extracts were washed once more with EtOAc. The combined aqueous extracts were basified with 6.0 M aq NaOH and then extracted twice with CH_2Cl_2. The combined extracts were dried over K_2CO_3 and concentrated to 118 mg (98%) of amine **4** as a colorless solid. Enantiomeric excess=94% (as determined by Mosher amide analysis and capillary electrophoresis analysis[9]). $[\alpha]_D^{25}$ −22.3 (*c* 1.06, $CHCl_3$).

*4.22. NMR data for (S,S)-**2***

^{1}H NMR (300 MHz, DMSO-d_6) δ 8.46 (d, J=7.8 Hz, 1H), 8.02 (d, J=8.5 Hz, 1H), 7.78 (d, J=2.2 Hz, 1H), 7.53 (br d, J=6.4 Hz, 2H), 7.20–7.13 (m, 3H), 7.06–7.00 (m, 3H), 4.86 (m, 1H), 4.51 (m, 1H), 3.93 (d, J=5.8 Hz, 2H), 2.86, 2.65 (ABX, J_{AB}=13.7 Hz, J_{AX}=4.8 Hz, J_{BX}=9.5 Hz, 2H), 1.82–1.64 (m, 6H), 1.77 (s, 3H), 1.33 (d, J=7.0 Hz, 3H), 1.28–1.01 (m, 5H); ^{31}P NMR (121 MHz, DMSO-d_6) δ −0.92.

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6. Src SH2 binding affinity was determined using a fluorescence polarization method (Lynch, B. A.; Loiacono, K. A.; Tiong, C. L.; Adams, S. E.; MacNeil, I. A. *Anal. Biochem.* **1997,** *247*, 77–82). IC_{50}s for **1** and the two diastereomers of **2** are 6 μM, 3 μM, and 527 μM using this method.
7. The diastereomer that exhibited the strongest binding affinity to the Src SH2 was also co-crystallized with the Lck SH2.[8] The X-ray crystal structure of this complex, which was determined at 2.5 Å resolution, is consistent with the prediction that it is the (*S,S*) diastereomer that shows the greatest affinity (Hatada, M.; Lu, X., unpublished results).

8. The SH2 binding sites for Lck and Src are nearly identical with only conservative modifications in a few residues. The strongest binding diastereomer was co-crystallized with Lck SH2 (S162C), which was used as an Src SH2 surrogate due to greater success with ligand co-crystallization. In addition, binding to Lck SH2 was equivalent to Src SH2. See also Ref. 2(a).
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