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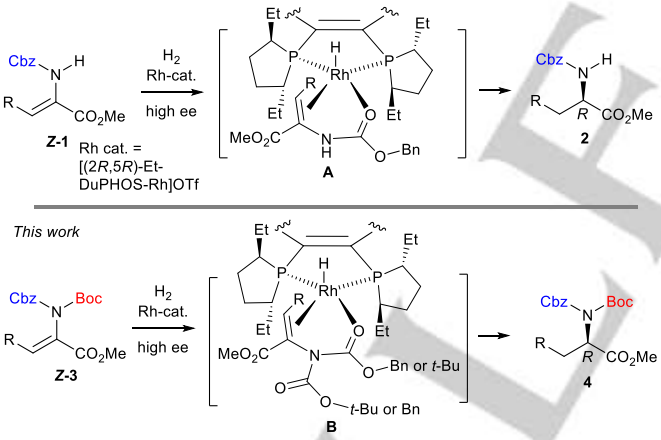
COMMUNICATION

Catalytic Asymmetric Hydrogenation of Dehydroamino Acid Ester with Biscarbamate Protection and Its Application to Synthesis of xCT inhibitor

Yoko Yasuno,[a] Iho Mizutani,[a] Yuki Sueuchi,[a] Yuuka Wakabayashi,[a] Nozomi Yasuo,[a] Keiko Shimamoto,[b] and Tetsuro Shinada* [a]

Abstract: Catalytic asymmetric hydrogenation of dehydroamino acid esters with biscarbamate protection was examined for the first time to prepare optically active amino acids. The new method was successfully applied to the synthesis of novel cystine-glutamate exchanger inhibitors.

Catalytic asymmetric hydrogenation of dehydroamino acid ester (DhaaE) **1** has been recognized as a well-established synthetic method to prepare optically active amino acids **2** (Scheme 1).[1] Enormous application has been demonstrated in the synthesis of biologically active nitrogen-containing compounds. In this study, we envisioned a new synthetic variant using biscarbamate **3** in the catalytic asymmetric hydrogenation with a chiral 1,2-Bis[(2,5)-diethylphospholano]benzene(1,5-cyclooctadiene)rhodium(I) trifluoromethanesulfonate [Et-DuPHOS Rh] catalyst. Although



Scheme 1. Catalytic asymmetric reduction of DhaaE **1** and **3** using chiral Et-DuPHOS Rh catalyst. **A**: transition state model with *Z*-**1**.[2] **B**: a hypothetical transition state model with *Z*-**3**.

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Z-DhaaE **3** has not been employed for the catalytic asymmetric reduction, we expected that **3** would be acceptable as a substrate for the Et-DuPHOS Rh catalyzed hydrogenation according to the following consideration. Burk et al. proposed the reaction mechanism of the asymmetric hydrogenation reaction of *Z*-**1** using (2*R*,5*R*)-Et-DuPHOS Rh catalyst to give (*R*)-**2** (model **A**).[2] The hydride on the Rh center in **A** is selectively transferred to the *si*-face of the coordinating enamide bond to give (*R*)-**2**.[3]It is conceivable that *Z*-DhaaE **3** could bind to the Rh center to give transition state model **B** in analogy with model **A** to undergo selective hydrogenation reaction to provide (*R*)-**4** in a stereoselective manner.

The above synthetic challenge is motivated by our original design and synthesis of cystine-glutamate exchanger (xCT) inhibitors[4] using *N*-benzoyl diaminosuberic acid (DAS) as a new template (Figure 1). xCTs are an amino acid transporter which imports extracellular cystine coupled with the efflux of intracellular glutamate (Figure 1, A).[4,5] The cystine is converted to glutathione via cysteine which play a crucial role in drug metabolism and removal of active oxygen species in the cell. Recent studies focusing on cancer metabolism suggest that xCTs are overexpressed in many cancers to promote tumor survival and enhancement of drug metabolism when cancer cells are exposed

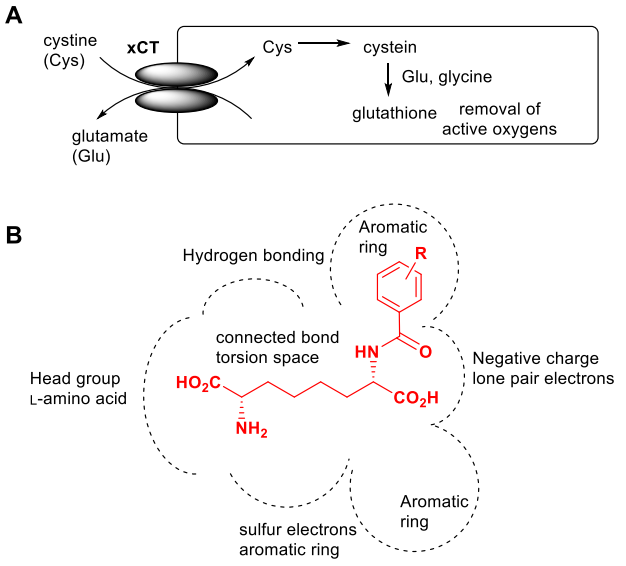

Figure 1. A: a model for cystine uptake via xCT. **B**: Pharmacophore model based on the 3D superposition of low energy conformation of substrates[4c] and a proposed biding model of a newly designed *N*-Benzoyl DAS derivative (red).

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to anticancer drugs.[4a,6] Therefore, xCTs are expected to be a promising molecular target for drug discovery of new anticancer drugs. Along this line, development of xCT inhibitors have been extensively studied.[4,6] Bridges et al. proposed a pharmacophore model of xCT inhibitors based on the three-dimensional superposition of low energy conformation of xCT inhibitors (Figure 1B).[4c] Inspired by their analysis, we envisioned structurally simple *N*-benzoyl DAS derivatives **5**–**8** as a new inhibitor in which their substituents could interact with the proposed aromatic biding site, negative charge ion pair site, and head group L-amino acid site.

We emerged a synthetic route to access all of the diastereoisomers of **5**-**8** via common synthetic precursor **9** (Figure 2). The synthesis of **9** offers challenging opportunity to test the feasibility of the proposed asymmetric hydrogenation. The distal amino acid chiral centers with orthogonal protection would be installed by the reduction of dehydroamino acid ester with mono- or bis-carbamate protection.

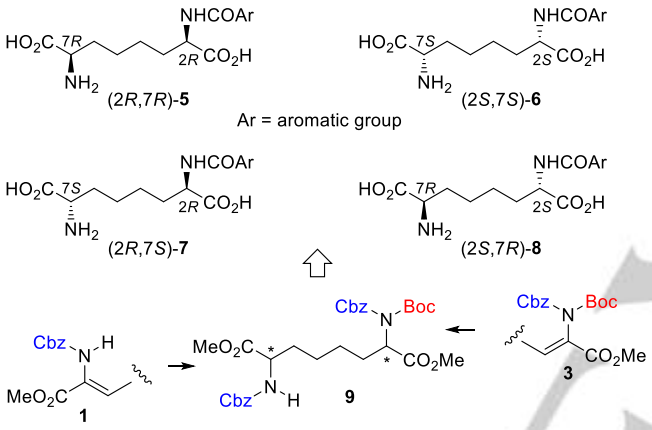
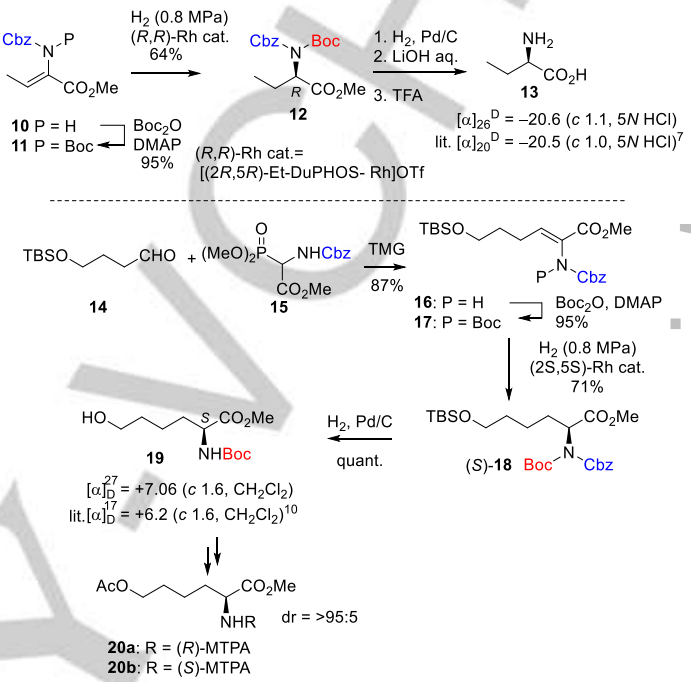


Figure 2. Synthetic plan for **5**–**8**.

To realize the new synthetic route, we initially investigated the stereochemical outcomes of (2*R*,5*R*)-Et-DuPHOS Rh-catalyzed hydrogenation reactions using DhaaEs **11** and **17** as substrates (Scheme 2). *Z*-Bis-carbamate **11** was prepared from *Z*-**10** by treatment with Boc_2O in the presence of 4-[*N*,*N*-(dimethylamino)]pyridine (DMAP). Pleasingly, the key asymmetric hydrogenation proceeded smoothly under the conditions [H_2 (0.8 MPa), (2*R*,5*R*)-Et-DuPHOS Rh catalyst (5 mol%), THF, rt] to give (*R*)-**12** in a stereoselective manner. The newly created stereocenter was determined to be *R* by conversion of **12** to amino acid **13**[7] and comparison of the optical rotation with that of the authentic **13**. In a similar manner, *Z*-**17**, prepared from **14** by olefination using the Schmidt reagent **15**,[8,9] followed by *tert*-butoxycarbonylation of the resulting *Z*-DhaaE **16**, underwent stereoselective reduction in the presence of (2*S*,5*S*)-Et-DuPHOS Rh catalyst to give (*S*)-**18** in high yield. The chirality of **18** was unambiguously determined to be *S* by comparison of the optical rotation value of **19**[10] derived from **18** as well as inspection of the MTPA amide **20a** and **20 b** derived from **19**. Each amide revealed *dr* = >95:5 (Supporting Information) implying the minor diastereomer was less than the analytical detection limit. It is noteworthy that the observed *R* stereoselectivity from **11** and **17** using (2*R*,5*R*)-Et-DuPHOS Rh catalyst is the same as those of the previously reported asymmetric reduction of DhaaEs **1**. These results suggested that the hydrogenation stereoselectively proceeded via the proposed transition state model **B** described in Figure 1.



Scheme 2. Catalytic asymmetric hydrogenation of bis-carbamate **11** and **17** using chiral Et-DuPHOS-Rh catalyst.

The above new methodology was successfully applied to the synthesis of common synthetic precursors (2*S*,7*S*)-**23** and (2*R*,7*S*)-**26** with orthogonal protection (Figure 2). The synthesis of (2*S*,7*S*)- **23** was achieved by the simultaneous hydrogenation of DhaaE **22** which was prepared from **17** by a series of sequential transformations: (i) removal of the TBS group, (ii) TEMPO oxidation, and (iii) olefination with **15** in good yield. DhaaE **22** was subjected to the catalytic asymmetric hydrogenation in the presence of (2*S*,5*S*)-DuPHOS Rh catalyst to give (2*S*,7*S*)-**23** as a single diastereoisomer.

The synthesis of (2*R*,7*S*)-**26** was accomplished by a stepwise introduction of the (*R*)- and the (*S*)-chirality starting from **17**. Biscarbamte **17** was converted to (*R*)-**18** under the hydrogenation conditions in the presence of (2*R*,5*R*)-Et-DuPHOS Rh catalyst. The optical rotation value of (*R*)-**18** was opposite in sign to that of (*S*)-**18**. (*R*)-Carbamate **18** was converted dehydroamino acid ester **25** in a similar manner to the synthesis of **22** from **17**. The asymmetric hydrogenation of **25** using (2*S*,5*S*)-Et-DuPHOS Rh catalyst provided (2*R*,7*S*)-**26** in 70% yield. We carefully examined the diastereomeric ratio of (2*S*,7*S*)-**23** and (2*R*,7*S*)-**26** by ^{1}H-NMR analysis (Supporting information). The NMR signal patterns of **23** and **26** were distinguishable in C_6D_6, indicating that the distal amino acid chiral centers of (2*S*,7*S*)-**23** and (2*R*,7*S*)-**26** were selectively induced under the hydrogenation conditions.

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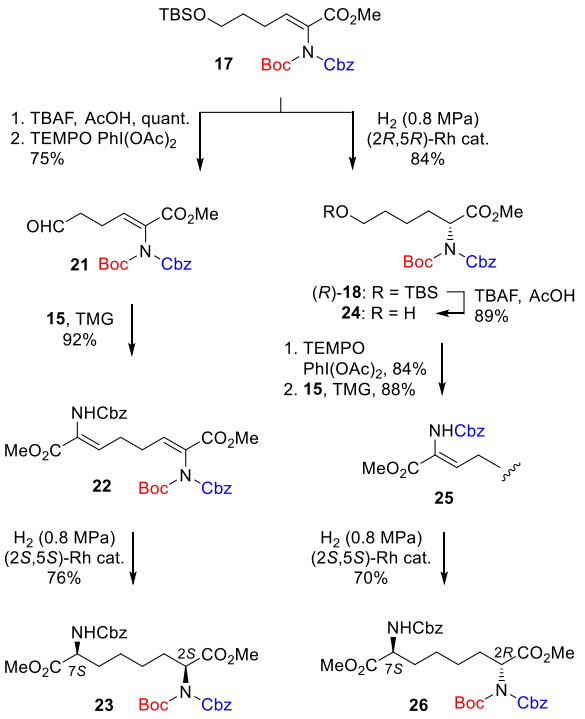
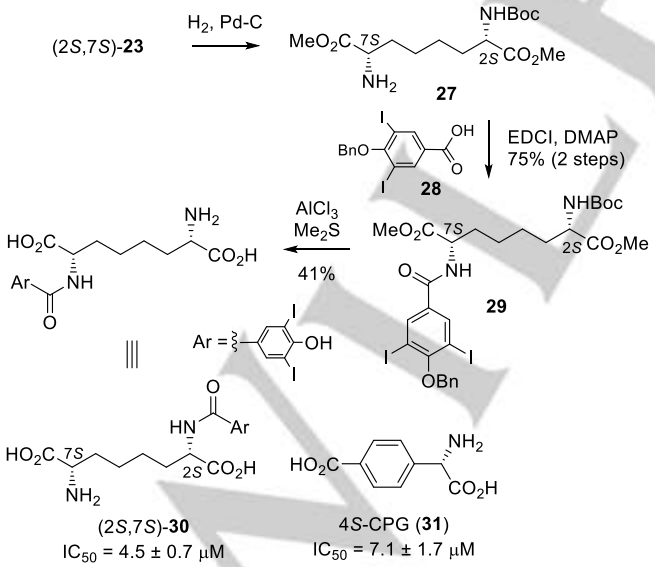


Scheme 3. Stereoselective preparation of **23** and **26** with orthogonal protections.

N-benzoyl DAS derivatives were prepared from **9** via **17**. A typical example is displayed in Scheme 4 and Figure 3.[12,13] Deprotection of the Cbz groups of **23** under the hydrogenation conditions followed by acylation with **28** to give **29** in 75% yield.



Scheme 4. Synthesis of (2*S*,7*S*)-**30**. IC_{50} values was estimated by the incorporation of extracellular ^{14}C-glutamate by the C6 cells[12] in which xCT is expressed by the pretreatment with diethyl maleate. 4-Carboxyphenyl)glycine (4*S*-CPG) was used for a positive control (IC_{50} = 7.1 μM).[14]

Global deprotection of the two methyl esters, Boc, and Bn groups using $AlCl_3$ and Me_2S gave **30** in 41% yield. In our delight, (2*S*,7*S*)-**30** displayed potent xCT inhibitory effect at the IC_{50} values of 4.5 μM. It is noteworthy that (2*S*,7*S*)-**30** (4.5 μM) is 1.6-fold more potent than that of 4-(carboxyphenyl)glycine (4-CPG, **31**) known to be one of the most potent xCT inhibitors.[14] We prepared other distereoisomers, (2*R*,7*R*)-**30**, (2*S*,7*R*)-**32** and (2*R*,7*S*)-**32** from **17** to compare their biological activities (Figure 3). These diastereoisomers displayed much lower inhibitory effects at the IC_{50} values of 50–80 μM, indicating that the stereochemistry of the designed molecules plays an important role in the inhibitory effects.

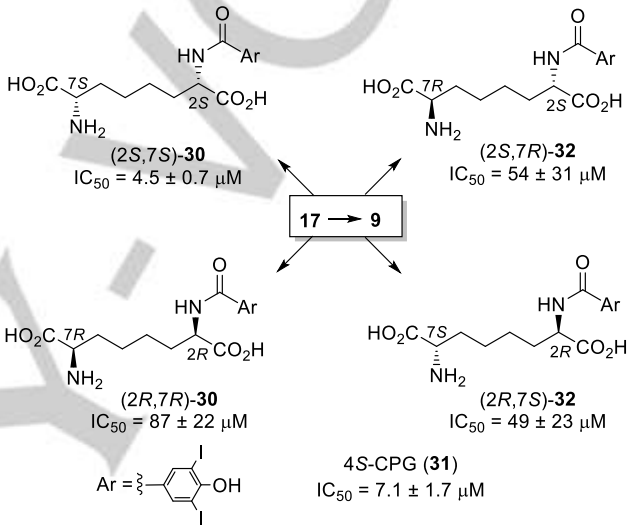


Figure 3. Comparison of biological activities of **30** and **32**.

In summary, we have developed a new entry for the catalytic asymmetric hydrogenation reaction using dehydroamino acids with biscarbamate protection. The experimental results demonstrated in the above revealed that the degree of stereoselectivity and product yield were found to be the same as those of the previously established catalytic asymmetric reduction using *Z*-**1**.[1] Successful application was demonstrated by the synthesis of all of the optically active *N*-Benzoyl DAS derivatives **30** and **32**. Among them, we found a potent xCT inhibitor (2*S*,7*S*)-**30** which was more potent to the powerful inhibitor 4-CPG (**31**). Compound **30** would be a new lead compound to develop anticancer agent based on the cysteine uptake inhibition. Recently, DAS and its analogues have received significant attention as a chemically stable and robust cystine surrogate.[15,16] Previous synthetic methods to prepare DAS derivatives have been mainly performed by chiral pool approach using amino acids as a starting material and diastereoselective alkylation using glycine derivatives with a chiral auxiliary.[17] However, the previous synthesis was performed in multi-steps to give a limited number of the diastereoisomers. In contrast, the present catalytic asymmetric hydrogenation reaction offers short and flexible accesses to all of the stereoisomers. The new catalytic hydrogenation reaction would open a new way to prepared not

only optically active amino acid derivatives but also nitrogen containing compounds. Further synthetic application is in progress in our laboratory.

Acknowledgements

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Keywords: catalytic asymmetric hydrogenation• dehydroamino acid • diaminosuberic acid • orthogonal protection • cystine-glutamate exchanger

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