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Short Research Article

Synthesis of isotopically labelled amino acids†

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Abstract: An efficient approach to the enantioselective synthesis of a series of amino acids from either bromoacetyl bromide or glycine is described using a [2,3]-sigmatropic rearrangement to establish the stereogenic centre at C-2 under mild conditions. Protected allylglycine **5** is a valuable building block to several amino acids e.g. hydrolytic cleavage of the auxiliary in **5** followed by deprotection gave L-allylglycine in 92% yield whilst oxidative cleavage of the terminal alkene followed by deprotection gave L-aspartic acid in 67% yield over the 2 steps. Furthermore alkene **5** may be converted to hydroxy ester **8** which is an intermediate for the synthesis of various amino acids including L-lysine and L-proline. Since the enantiomer of sultam **1** is commercially available, the analogous D-amino acids may be synthesised. This chemistry is readily adapted for the incorporation of isotopic labels for example for the synthesis of [1,2-$^{13}C_2$,^{15}N]-L-homoserine **14**. Copyright © 2007 John Wiley & Sons, Ltd.

Keywords: amino acids; isotope labelling; [2,3]-sigmatropic rearrangement

Introduction

Isotopically labelled amino acids are valuable for a range of studies in bioorganic chemistry including biosynthetic and metabolic studies. Recently the incorporation of stable labelled amino acids into proteins, either uniformly labelled or selectively labelled, has proved essential in the elucidation of the three-dimensional structure of proteins by NMR spectroscopy.[1] Many methods have been developed for the selective labelling of α-amino acids but there is a continuing need to refine approaches and improve efficiency to enable multigram quantities of specifically labelled amino acids to be prepared.[2] Herein the enantioselective synthesis of a series of amino acids is described using an approach which is readily adapted for the incorporation of isotopic labels.

[2,3]-Sigmatropic rearrangements of *N*-allyl ammonium ylids have been widely used in organic synthesis and the reactions are commonly carried out in two steps: preparation of the quaternary ammonium salt followed by treatment of the isolated salt with a strong base to generate an ylid which undergoes the rearrangement with creation of a new carbon–carbon bond.[3] Indeed using such a two step procedure, Oppolzer's camphor sultam has been employed as an auxiliary to induce intramolecular chirality transfer with excellent stereocontrol in the synthesis of D-allylglycine.[4] We have investigated an approach which would enable the stereogenic centre at C-2 of the α-amino acid to be created under mild conditions in a single step from a tertiary amine based upon the *in situ* generation of an *N*-allyl ammonium ylid as pioneered by Coldham and co-workers in their synthesis of racemic allylglycine methyl ester.[5]

Results and discussion

The substrate **3** required for the key [2,3]-Stevens rearrangement was prepared in two steps and 91% overall yield from (1*S*,2*R*)-bornane-10,2-sultam **1** (Scheme 1).[4] Initially the conditions reported by Coldham for the [2,3]-sigmatropic rearrangement of diallylglycine methyl ester were examined.[5] Treatment of **3** with allyl bromide, K_2CO_3, DBU and tetrabutylammonium iodide at 40°C in DMF gave the *C*-allylated product in 85% yield as a 12:1 mixture of diastereomers. The major isomer readily crystallized from

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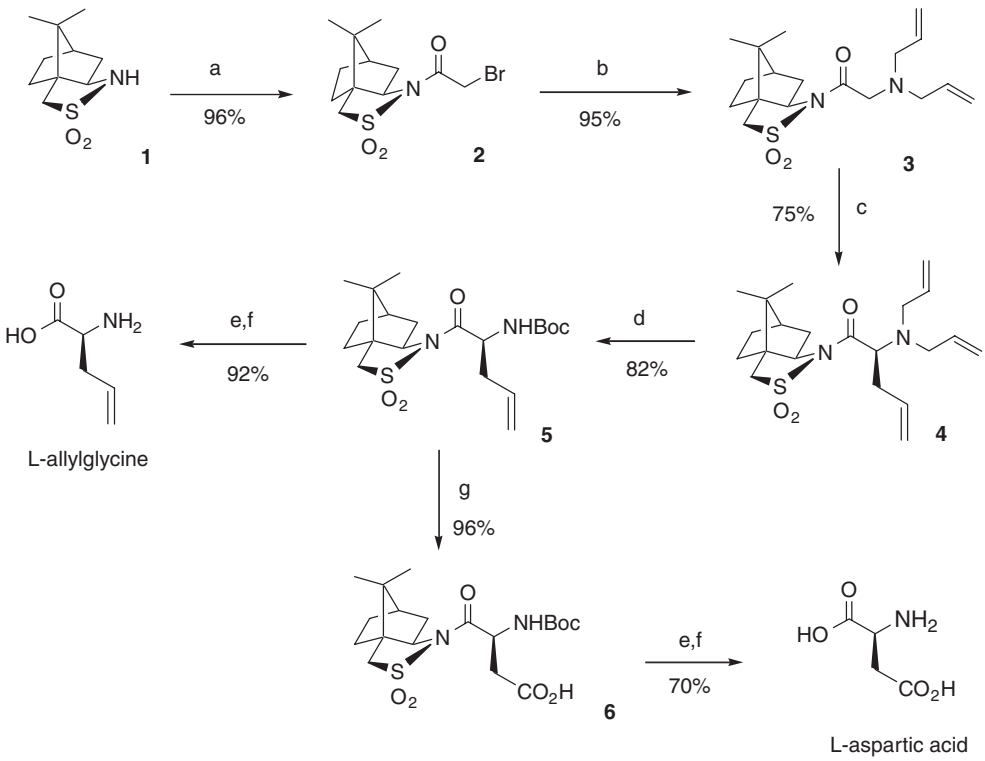

Scheme 1

ethanol giving exclusively the (2′*S*) stereoisomer **4** in 75% isolated yield. Following further investigations the optimum conditions were found to be reaction of **3** with allyl iodide, K_2CO_3 in DMF at 40°C giving the 12:1 mixture of diastereomers in 90% yield.[6]

The palladium(0) mediated removal of the *N*-allyl groups[7] gave the primary amine which was protected *in situ* as the *t*-butyloxycarbonyl (Boc) derivative **5** in 82% yield. Protected allylglycine **5** is a valuable intermediate for the synthesis of a number of α-amino acids. For example, hydrolytic cleavage of the auxiliary using lithium hydroxide and hydrogen peroxide followed by removal of the Boc group with acid gave L-allylglycine in 92% yield from **5**. L-aspartic acid was prepared in 67% overall yield from **5** using ruthenium dioxide and sodium periodate to oxidatively cleave the double bond to acid **6** followed by removal of the auxiliary and Boc protecting group.

A further demonstration of the versatility of this chemistry is in the conversion of alkene **5** to primary alcohol **8** which is a valuable intermediate in the synthesis of further amino acids e.g. L-proline and L-lysine (Scheme 2).[8] Hydrolytic cleavage of the auxiliary and treatment of the resultant acid with *t*BuOH, DCC (*N*,*N*′-dicyclohexylcarbodiimide) and DMAP (4-dimethylaminopyridine) gave ester **7**. Hydroboration of the olefin under standard conditions then gave the required alcohol **8**. Two approaches were examined for the conversion of alcohol **8** to protected proline. The first involved treatment with Ph_3P/Br_2 which led to spontaneous cyclization to the target compound **10** but in our hands in a disappointing 40% yield. A better approach was to use a 2 step procedure in which the alcohol was converted to tosylate **9** and then on treatment with sodium hydride, protected L-proline **10** was isolated in 84% yield over the 2 steps. Finally, removal of the protecting groups using TFA gave L-proline.

Since the enantiomer, (1*R*, 2*S*)-bornane-10,2-sultam, is commercially available this chemistry may be readily adapted for the enantioselective synthesis of D-amino acids.

The approaches to the enantioselective synthesis of α-amino acids outlined in Schemes 1 and 2 are readily adapted for the incorporation of isotopic labels e.g. using [1,2-$^{13}C_2$]-bromoacetyl bromide to acylate (1S,2*R*)-bornane-10,2-sultam **1** in the initial step leads to [1,2-$^{13}C_2$]-amino acids. Furthermore, amino acids incorporating both carbon-13 and nitrogen-15 labels may be prepared starting with commercially available [1,2-$^{13}C_2$,^{15}N]-glycine **8** as illustrated in the synthesis of [1,2-$^{13}C_2$,^{15}N]-L-homoserine **14** (Scheme 3).[9]

This chemistry may be used for the synthesis of further amino acids e.g. Boc homoserine *t*-butyl ester

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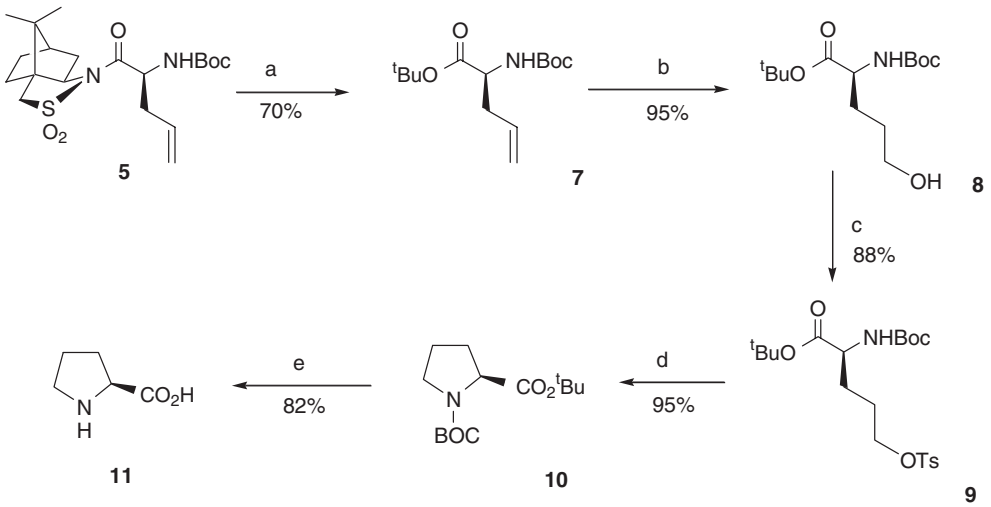
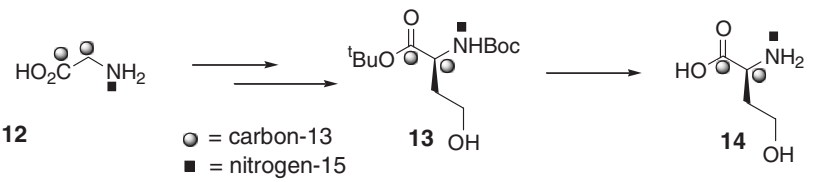


Scheme 2



Scheme 3

(unlabelled **13**) has been used as an intermediate in the synthesis of methionine and selenomethinoine.[10]

Conclusion

In conclusion we have described a simple and efficient method for the synthesis of protected allylglycine **4** using a base mediated [2,3]-sigmatropic rearrangement of an *N*-allyl ammonium ylide generated *in situ* from tertiary amine **3** using the camphor sultam to induce stereocontrol at C-2. Boc protected allyl glycine **5** is a valuable intermediate in the enantioselective synthesis of a number of amino acids including L-allylglycine, L-aspartic acid, L-proline, L-lysine and L-methionine. Since the enantiomer, (1*R*, 2*S*)-bornane-10,2-sultam, is commercially available this chemistry may be readily adapted for the enantioselective synthesis of a series of D-amino acids. The approach is flexible and has been used for the incorporation of isotopic labels for example in the synthesis of [1,2-$^{13}C_2$,^{15}N]-L-homoserine **14**.

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