Pergamon

Tetrahedron Letters, Vol. 35, No. 30, pp. 5421-5424, 1994
Elsevier Science Ltd
Printed in Great Britain
0040-4039/94 $7.00+0.00

0040-4039(94)01096-X

MILD AND RAPID AZIDE- MEDIATED, PALLADIUM CATALYZED CLEAVAGE OF ALLYLESTER BASED PROTECTING GROUPS

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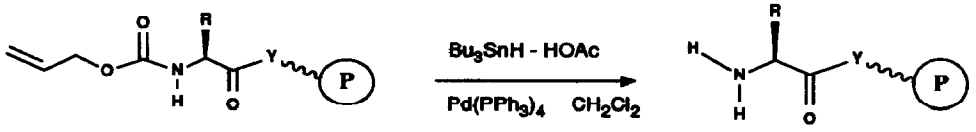
Summary: Under palladium catalysis various allylester protecting groups are rapidly cleaved in high yield with the reagent 8:3 trimethylsilylazide/tetrabutylammonium fluoride. The efficiency of this method makes it particularly useful for solid phase deprotections.

The development of the chemistry of π-allyl palladium complexes has led to a powerful and increasingly applied protection strategy in organic synthesis.[1] Thus, allyl esters, carbamates[2], carbonates[3] or other functions with an acidic leaving group are readily cleaved using zero valent palladium (Pd^0) catalyzed transfer of the allyl moiety to an acceptor nucleophile via the corresponding π-allyl palladium complex. Kunz has introduced the allyloxycarbonyl (Alloc) based protection strategy for the α-amino group in solution phase peptide synthesis,[4] and a few Alloc solid phase peptide syntheses (SPPS) have been described.[2d,5] For the solution phase peptide synthesis the standard nucleophilic acceptor agents have been active C-H acids (dimedone, N.N-dimethylbarbituric acid) or amines (eg. morpholine, N-methylaniline). These agents were found unsatisfactory in the case of SPPS for which an efficient rapid and side reaction free cleavage of the α-amino allyloxycarbonyl group is essential.[2d,6] The use of a tributyltinhydride-acetic acid-Pd^o mediated



reductive cleavage of the allyloxycarbonyl-α-amino protecting group was found to satisfy the above criteria in the Alloc SPPS synthesis of substance P.[2d] In the course of our studies on the SPPS synthesis of serine phosphopeptides we have experienced difficulties with this method. Therefore, we have developed a simple, rapid and mild method for the cleavage of the Alloc-α-amino protecting group of a solid phase bound peptide using azide as acceptor. The method also works well for the solid phase deprotection of allyl phosphonic acid esters and appears to be quite general.

Serine phosphopeptides are base sensitive and are not amenable to preparation via standard Fmoc-SPPS using a protected serine phosphate building block.[7] Although the Boc-SPPS of a serine phosphopeptide has been reported, difficulties were encountered during the strong acid cleavage of the peptide from the resin which required optimization of the aryl phosphate ester protecting groups.[8] A general SPPS building block method for synthesizing serine phosphopeptides is under investigation in our laboratories.[9] The

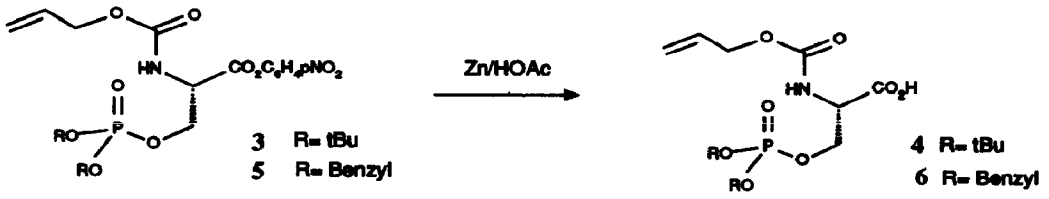
Alloc strategy is very attractive in this regard due to the mild conditions employed. Indeed, an SPPS of tetrapeptides 1 and 2 with Alloc-amino acids and the tributyltinhydride-Pd° deprotection method has appeared.[6] The synthesis was performed on Wang resin[10] using the building block Alloc-Ser[-

H-Arg-Ser(PO_3H_2)-Leu-Asn-OH

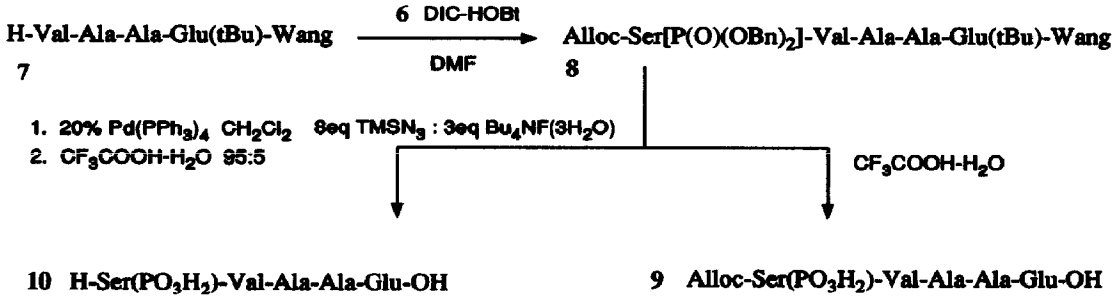
1

H-Arg-Leu-Ser(PO_3H_2)-Ile-OH

2



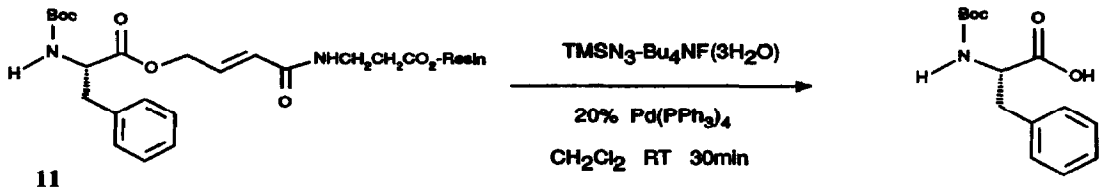
P(O)(OtBu)$_2$]-OH, 4, with t-butyl phosphate ester protection enabling resin cleavage and concomitant side chain deprotection with trifluoroacetic acid (TFA). We desired to add an Alloc serine phosphate ester building block to the tetrapeptide sequence H-Val-Ala-Ala-Glu(tBu)-Wang, 7,[11] cleave the Alloc group and then extend the chain by coupling an amino acid or a peptide fragment. Unfortunately, we were unable to repeat the last step in the described synthesis of 4 in which the nitrobenzyl ester 3, is reduced to the free acid 4. In our hands the phosphate t-butyl ester functionality proved sensitive to the reduction conditions and was cleaved to a large extent to the phosphonic acid. At this point we turned to the building block Alloc-Ser[P(O)(OBn)$_2$]-OH 6 with the TFA removable benzyl phosphate ester protection.[12] This compound was readily prepared using the method



described for 4 and could be stored indefinitely. Diisopropylcarbodiimide-hydroxybenzotriazole (DIC-HOBt) mediated coupling of 6 onto 7 was performed and checked by TFA cleavage of the peptide from the resin which gave Alloc-Ser(PO_3H_2)-Val-Ala-Ala-Glu-OH 9 as the major product. Alloc group cleavage from 8 was performed using the described Bu_3SnH-HOAc-Pd^0 protocol, followed by peptide liberation with TFA:H_2O 95:5. HPLC analysis of the crude reaction mixture indicated incomplete reaction to give 10, 9 and an unsatisfactory array of products.[13] This result could not be significantly improved by manipulation of the reaction conditions and led us to develop our own Alloc-cleavage method.

We reasoned that for rapid solid phase allyl cleavage, a nucleophile which is highly reactive, but also small would be desirable. These criteria are fulfilled by the azide anion. In addition electrophilic activation[14] could be achieved by using a conjugate Lewis acid of azide ideally from which the azide anion could be readily generated namely trimethylsilyl azide. Indeed, we have found that a 8:3 mixture of $TMSN_3/Bu_4NF$ is a superior reagent for the rapid and clean Pd^0 catalyzed removal of allyl ester protecting groups. Treatment of 1mol equivalent of **8** with premixed 8eq.:3eq. $TMSN_3/Bu_4NF(3H_2O)$ and 20mol% $Pd(PPh_3)_4$ in dichloromethane at room temperature under argon for 30min resulted in quantitative cleavage of the Alloc group. After standard resin washing and TFA cleavage, **10** was the major product which was readily isolated by HPLC. At this point it was clear that an excellent method for removing allyl ester protecting groups had been achieved.

It was of further interest to us to establish the generality and scope for this method with regard to solid phase deprotections. We have established the utility of the method for the liberation of Boc-Phe-Hycram **11**[15] and Abu[P(O)(OCH$_2$CH=CH$_2$)$_2$][16] containing peptides. The use of peptide allyl resin linkers is of use for preparing peptide fragments, but a straightforward, efficient cleavage method is desirable.[17] When **11** was cleaved under our standard conditions (*vide supra*), Boc-Phe-OH was obtained with excellent recovery.[18] For



11

12 **13**

the deprotection of allyl phosphate or phosphonate esters Noyori has developed particular conditions: butylamine(4eq):formic acid(16eq) in tetrahydrofuran for several hours at 50°C.[19] We have successfully employed the Noyori method in the preparation of several phosphonopeptide isosteres of serine phosphopeptides.[16b] However; complete deprotection typically required reaction for several hours at 50°C as well as 20mol% $Pd(PPh_3)_4$. Using our standard $TMSN_3/Bu_4NF$ and 20mol% $Pd(PPh_3)_4$ method with 30min reaction time followed by standard resin washing, the previously described phosphono-hexapeptide **13**[16b] was isolated in high yield from precursor **12** after cleavage from the Wang resin.[20]

In summary an efficient and apparently general method for the cleavage of allyl based ester groups has been developed which is particularly useful for solid phase applications. The method may be useful as a global side chain deprotection strategy in SPPS. Recently the potential danger of using azide in dichloromethane ascribed to the formation of diazidomethane which can explode upon concentration has been reported and should be kept in mind.[21] In the solid phase application where the product remains bound to the

solid phase, there should be little danger of using the $TMSN_3/Bu_4NF$ reagent in dichloromethane since it is discarded in dilute solution with the standard resin washing procedures.

REFERENCES AND NOTES

1. Trost, B. M. *Acc. Chem. Res.* **1980**, *13*, 385. b. Tsuji, J. *Organic Synthesis with Palladium Compounds*, Springer Verlag Berlin
2. a.Minami, I.; Ohashi, Y.; Shimizu, I. and Tsuji, J. *Tetrahedron Lett.* **1985**, *26*, 2449. b. Kinoshita, H.; Inomata, K.; Kameda, T., and Kotake, H. *Chem. Lett.* **1985**, 515. c. Guibe, F.; Dangles, O.; Balavoine, G. *Tetrahedron Lett.* **1986**, *27*, 2368. d. Dangles, O.; Guibé, F.; Balavoine, G.; Lavielle, S. and Marquet, A. *J. Org. Chem.* **1987**, *52*, 4984. e. Merzouk, A; Guibe, F. and Loffet, A. *Tetrahedron Lett.* **1992**, *33*, 477.
3. a. Minami, I.; Ohashi, Y.; Shimizu, I. and Tsuji, J. *Tetrahedron Lett.* **1982**, *23*, 4809. b. Guibe, F. and Saint M Leux, Y. *Tetrahedron Lett.* **1981**, *22*, 3591.
4. Kunz, H. and Unverzagt, C. *Angew. Chem. Int. Ed. Engl.* **1984**, *23*, 436. Kunz, H. and Waldmann, H. *Angew. Chem. Int. Ed. Engl.* **1984**, *23*, 71.
5. Allyl transfer to the liberated alpha amino function must be suppressed see ref. 2.
6. Lacombe, J. M.; Andriamanampisoa, F. and Pavia, A. A. *Int. J. Peptide Protein Res.* **1990**, *36*, 275.
7. Interestingly there is one report of the synthesis of the threonine phosphopeptide, Pro-Arg-Gly-Asp-Thr(P)-Tyr, with the building block Fmoc-Thr[$PO(OBn)_2$]-OH using Fmoc-SPPS and morpholine as a base to perform the Fmoc cleavage: Larsson, E.; Lünig, B.; Mickos, H.; and Heinegård, D. "Synthesis of Phosphopeptides by the Solid Phase Technique" in *Innovation and Perspectives in Solid Phase Synthesis* (1991) R. Epton, SPPC (U.K.)
8. Tsukamoto, M.; Kato, R.; Ishiguro, K.; Uchida, T. and Sato, K. *Tetrahedron Lett.* **1991**, *32*, 7083.
9. The synthesis of phosphoserine peptides by Fmoc-SPPS synthesis followed by on resin phosphorylation of the free hydroxyl residue of serine has been reported by several groups see: Shapiro, G.; Stauss, U. and Swoboda, R. *Tetrahedron Lett.* **1994**, *35*, 869. and references therein.
10. Wang, S. *J. Am. Chem. Soc.* **1973**, *95*, 1328. Wang-Asp was purchased from Novabiochem AG, Weidenmattweg 4, Läufelfingen, CH-4448, Switzerland.
11. Prepared by standard Fmoc-SPPS on Wang resin.
12. Andrews, D. M.; Kitchin, J.; and Seale, P. W. *Int. J. Peptide Protein Res.* **1991**, *38*, 469.
13. Determined by HPLC purification of **9**, **10** and FAB-MS analysis.
14. After performing this work we found that the concept of using electrophilic activation had been described in the deprotection of Alloc protected amines using silylamines as the nucleophilic acceptor or amines in the presence of trimethylsilylacetate.[2e]
15. Hycram= hydroxycrotonylamidomethyl, Kunz, H. and Dombo, B. *Ang. Chem. Int. Ed.* **1988**, *27*, 710. Boc-Phe-Hycram was purchased from Orpegen Inc., Czernyring 20, Heidelberg, Germany D-69115.
16. For the synthesis of the building block L-Fmoc-Abu[$PO(OCH_2CH{=}CH_2)_2$]-OH and its incorporation into peptides see a. Shapiro, G.; Buechler, D.; Ojea, V.; Pombo-Villar, E.; Ruiz, M. and Weber, H.P. *Tetrahedron Lett.* **1993**, *34*, 6255. b. Shapiro, G.; Buechler, D.; Enz, A. and Pombo-Villar, E. *Tetrahedron Lett.* **1994**, *35*, 1173.
17. a. For other examples of allyl linkers see Guibé, F.; Dangles, O.; Balavoine, G. and Loffet, A. *Tetrahedron Lett.* **1989**, *30*, 2641. and Blankemeyer-Menge, B. and Frank, R. *Tetrahedron Lett.* **1988**, *29* 5871. b. For cleavage of allyl resins see Lloyd Williams, P.; Jou, G.; Albericio, F and Giralt, E. *Tetrahedron Lett.* **1991**, *32*, 4207.
18. In the HPLC analysis (Merck LiChroCartRP-18, 125mm X 4mm, LiChrosphere 5µm, solution A 1000 H_2O, 20 tetramethylammonium hydroxide, 2 H_3PO_4(85%), solution B 700 CH_3CN, 300 H_2O, 20 tetramethylammonium hydroxide, 2 H_3PO_4(85%); gradient 5%B to 95%B in 20min, 1.5mLmin^{-1}, 40°C, 205nM) of the crude product peaks originating from the reagents and a major peak corresponding to >95% (using pure Boc-Phe standard, t_R=15.3min) recovery of Boc-Phe were found.
19. a. Hayakawa, Y; Kato, H.: Uchiyama, M.; Rajino, H and Noyori, R. *J. Org. Chem.* **1986**, *51*, 2402. b. Hayakawa, Y; Wakabashi, S.; Kato, H., and Noyori, R. *J. Am. Chem. Soc.* **1990**, *112*, 1691.
20. General HPLC conditions gradient[18] 0%B to 50%B in 20min, 13, t_R=10.2min.
21. Hruby, V. J., Boteju L. and Guigen, L. in Letter to *Chem. Eng. News* Oct. 11, 1993.

(*Received in Germany* 4 *May* 1994; *accepted* 3 *June* 1994)