A New Catch-and-Release Purification Method Using a 4-Azido-3-chlorobenzyl Group

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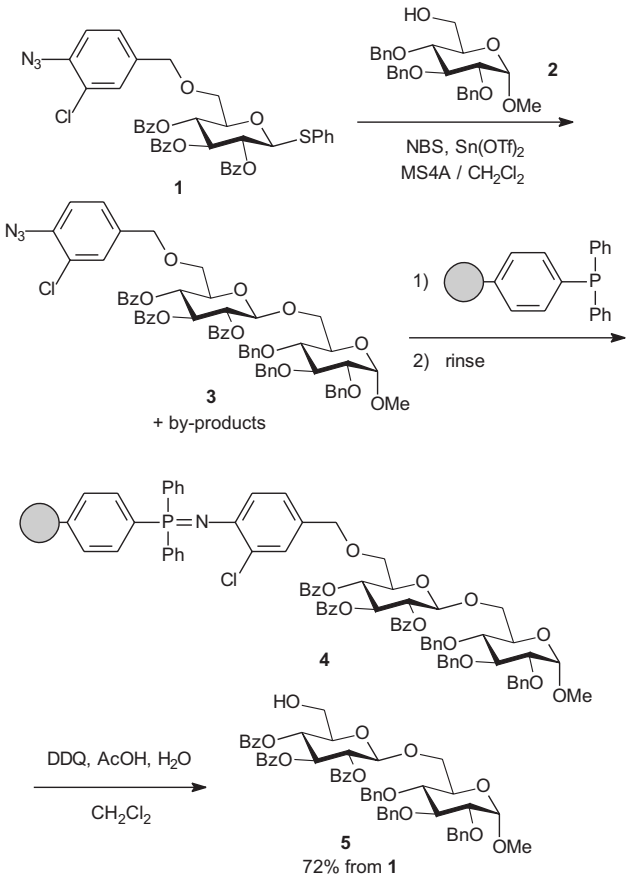
Abstract: Products that possess 4-azido-3-chlorobenzyl group were selectively isolated from reaction mixtures by using a solid-supported phosphine. Successive treatment with 2,3-dichloro-5,6-dicyanobenzoquinone readily liberated purified products. Application of this method to several oligosaccharide syntheses is described.

Key words: azides, combinatorial chemistry, glycosylations, phosphines, polymer-supported synthesis

In organic synthesis, the isolation of products is sometimes a tedious and time-consuming procedure. Especially in the area of combinatorial chemistry, a simple and effective isolation method is necessary because many compounds are handled at the same time. For instance, chromatography is a quite popular method for the isolation of products of organic synthesis. However, it is somewhat tedious and consumes a considerable amount of solvents and optimization of the conditions of the separation sometimes requires much time. Therefore, techniques for polymer-supported synthesis such as polymer-bound reagents, scavenger resins, catch-and-release purification have been developed recently.[1]

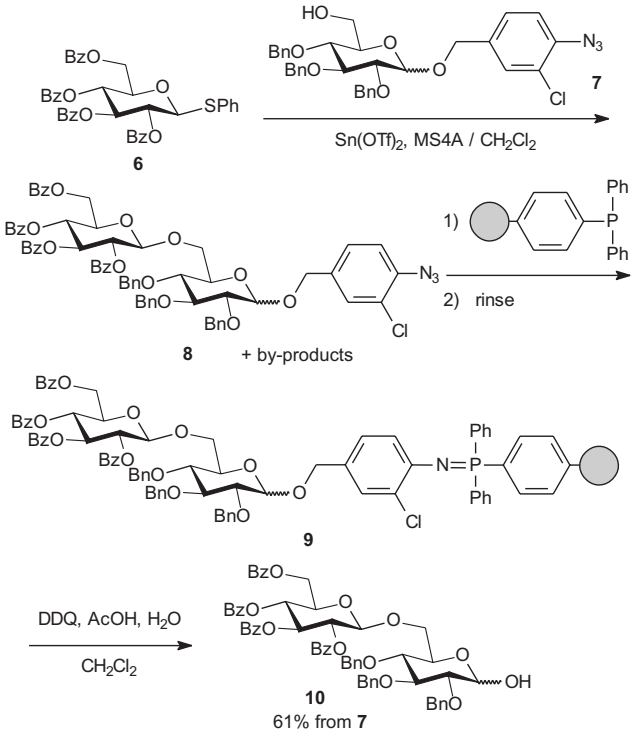
We have already reported the use of the 4-azido-3-chlorobenzyl (ClAzb) group for practical protection of hydroxy functions.[2,3] The ClAzb group is stable under various conditions but is readily removed by a safety catch procedure, i.e., conversion of the azido function to the corresponding iminophosphorane followed by oxidation using 2,3-dichloro-5,6-dicyanobenzoquinone (DDQ). We thought that the ClAzb group could be used not only as a protecting group but also as a tag of the product for catch-and-release purification. A compound possessing the ClAzb group can be selectively caught by a solid-supported phosphine based on the specific reaction between the azido function and the phosphine, separated from the other compounds by simple rinsing, and then released from the solid-support by treatment with DDQ. This method was examined in several oligosaccharide syntheses.

As shown in Scheme 1, ClAzb-thioglycoside **1** was reacted with 3 equiv of glycosyl acceptor **2** using *N*-bromosuccineimide (NBS) and $Sn(OTf)_2$ as activating reagents in the presence of molecular sieves 4Å. In this reaction, MS4A and excess amount of the acceptor were used to avoid formation of the hydrolyzate of the thioglycoside **1**. The reaction mixture was neutralized with Amberlite® IRA-68, and then filtered. To the filtrate was added triphenylphosphine-polystyrene resin to catch the ClAzb-disaccharide **3** onto the resin. The reagents and by-products without the ClAzb moiety were removed by simple rinsing with dichloromethane and the solid-supported disaccharide **4** was obtained. The disaccharide part of the resin **4** was finally released by treatment with DDQ. Excess DDQ was removed by ion-exchange resin Amberlite® IRA-68 after conversion of DDQ to the corresponding hydroquinone using L-ascorbic acid. After passing the solution of the product through a silica gel short column (1 × 2 cm) to remove 4-amino-3-chlorobenzaldehyde derived from the ClAzb group, pure disaccharide **5** was obtained in 72% yield.[4] Disaccharide **5**, which has a free hydroxy group, can be used as a glycosyl acceptor for the subsequent glycosylation.



Scheme 1 Glycosylation using ClAzb-thioglycoside

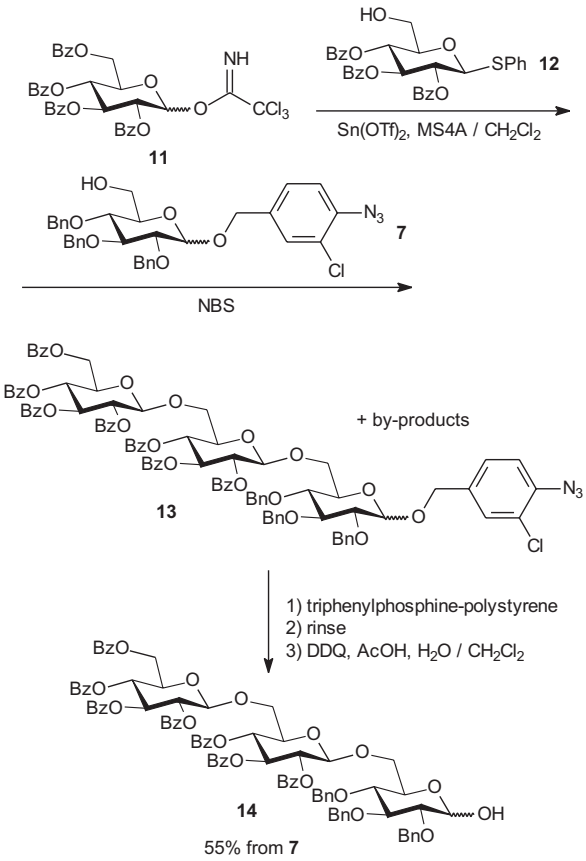
We then carried out disaccharide synthesis using a glycosyl acceptor having the ClAzb group. As shown in Scheme 2, 1-*O*-ClAzb-glycoside **7** was glycosylated with 3 equiv of thioglycoside **6** as above. The reaction mixture containing disaccharide **8** was neutralized and treated with triphenylphosphine-polystyrene similarly to give solid-supported disaccharide **9**. After the treatment with DDQ and the operation described above, disaccharide **10** was obtained in 61% yield in a pure state. Disaccharide **10** can be used as a glycosyl donor for the subsequent glycosylation after conversion of the 1-hydroxy group to a proper functional group such as trichloroacetimidate.



Scheme 2 Glycosylation using 1-*O*-ClAzb-glycoside

Next, we examined one-pot trisaccharide synthesis.[5] One problem in this approach is the separation of the desired products from the reaction mixtures containing by-products derived from excess glycosyl donors. As shown in Scheme 3, glycosyl trichloroacetoimidate **11** was reacted with thioglycoside **12** using $Sn(OTf)_2$ in the presence of molecular sieves 4Å at r.t. for 30 min and then 1-ClAzb-glycoside **7** and NBS were added. The reaction was continued at r.t. for 30 min. to give a complex reaction mixture containing trisaccharide **13**. The mixture was first treated with triphenylphosphine-polystyrene resin and then with DDQ to give the pure trisaccharide **14** in 55% yield from **7**.

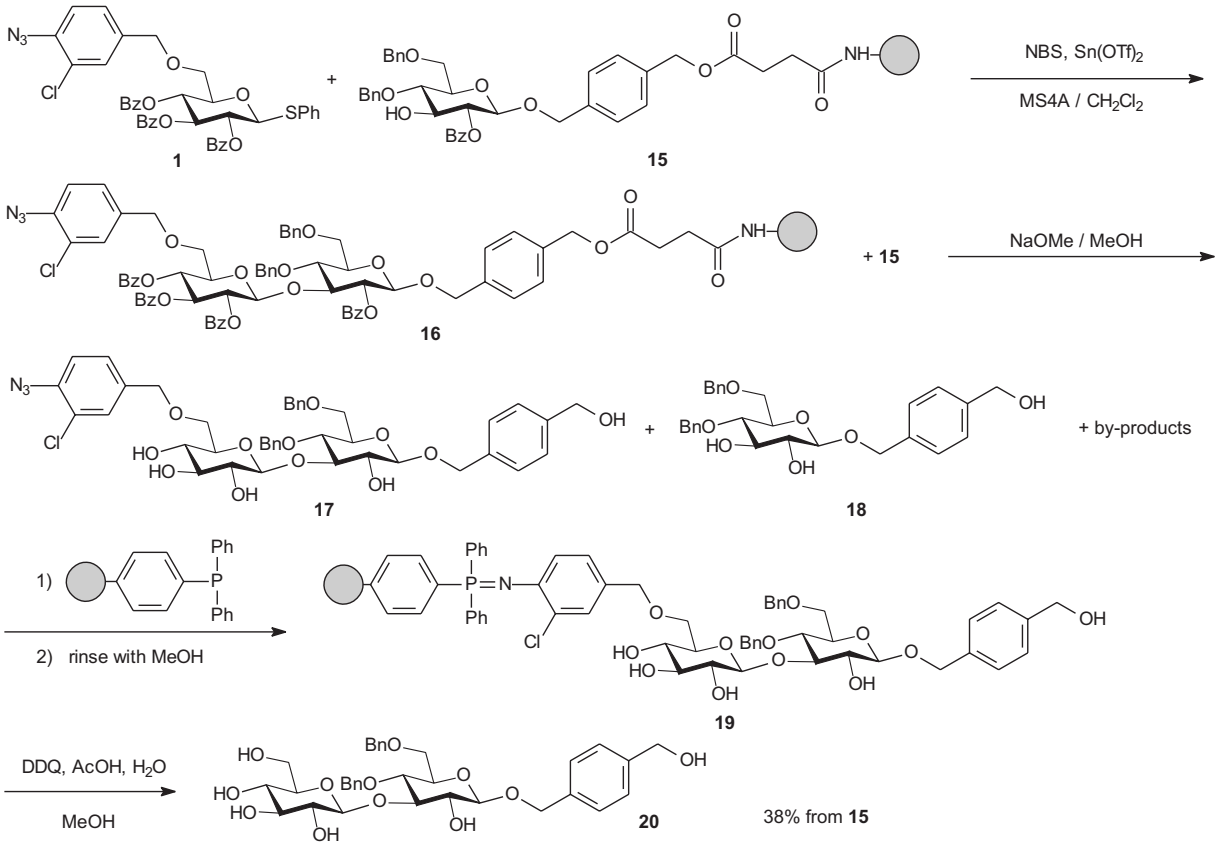
Finally, we examined a combined application of this method with solid-phase glycosylation.[6] In the final cleavage step of solid-phase synthesis, solid-bound impurities formed in each reaction step are released with the desired



Scheme 3 One-pot trisaccharide synthesis

compound, and such impurities must be removed. As shown in Scheme 4, a glycosyl acceptor bound to a macroporous polystyrene (ArgoPore™-NH_2[7]) **15** was glycosylated with thioglycoside **1** using NBS and $Sn(OTf)_2$ in the presence of MS4A to give a mixture of disaccharide **16** and unreacted **15**.[8] The resin was treated with sodium methoxide in methanol to cleave the linker, and a mixture of disaccharide **17**, monosaccharide **18** derived from **15** and other by-products was obtained. Then, the mixture was treated with triphenylphosphine-(polyethyleneglycol-polystyrene-copolymer) resin[9] in methanol instead of the triphenylphosphine-polystyrene resin employed above. This was because disaccharide **17** was insoluble in dichloromethane and the polystyrene resin does not swell in methanol. The solid-supported disaccharide **19** obtained was treated with DDQ. Excess DDQ was then quenched with L-ascorbic acid. The mixture was treated with ion exchange resins Amberlyst® A-26 (OH^- form, to remove hydroquinone) and Amberlyst® 15 (H^+ form, to remove 4-amino-3-chlorobenzaldehyde derived from the ClAzb group) to afford highly pure disaccharide **20** in 38% yield from **15**.

Thus, the products possessing the ClAzb group were selectively isolated from the reaction mixture using triphenylphosphine-resin, and the successive treatment with DDQ readily afforded purified products. Since a new free



Scheme 4 Solid-phase saccharide synthesis

hydroxyl function is generated through this purification procedure, this method can be regarded as a combination of purification and deprotection.

In conclusion, the catch-and-release purification using the ClAzb group was proved to be simple but quite effective. This method offers a new approach to the purification of products in organic synthesis, especially in the field of combinatorial synthesis.

Acknowledgement

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(3) 4-Azido-3-chlorobenzyl bromide is commercially available from Wako Pure Chemicals Industries, Ltd., http://www.wako-chem.co.jp/.

(4) Representative procedure: Preparation of 2,3,4-tri-*O*-benzoyl-β-D-glucopyranosyl-2,3,4-tri-*O*-benzyl-β-D-glucopyranoside **5**. After a mixture of ClAzb-thioglycoside **1** (50 mg, 67 μmol), glycosyl acceptor **2** (93 mg, 200 μmol), $Sn(OTf)_2$ (6 mg, 13 μmol) and molecular sieves 4 Å (ca. 500 mg) in 2 mL of dichloromethane was stirred at r.t. for 15 min., the reaction was started by addition of NBS (13 mg, 70 μmol). The mixture was stirred at r.t. for 30 min. and then ion exchange resin Amberlite® IRA-68 (ca. 200 mg) was added and filtered. To the filtrate was added triphenylphosphine-polystyrene resin (278 mg, 333 μmol), and the mixture was stirred at r.t. for 3 h, and filtered. The resin was washed with dichloromethane twice and suspended in 2 mL of dichloromethane. DDQ

(91 mg, 400 μmol), acetic acid (100 μL) and water (100 μL) were added and the mixture was shaken at r.t. for 3 h, then filtered. To the filtrate was added a solution of L-ascorbic acid (70 mg, 400 μmol) in methanol and the mixture was stirred at r.t. for 5 min. After passing through a column of ion exchange resin Amberlite® IRA-68 (2 × 5 cm), the mixture was concentrated in vacuo. The residue was dissolved in dichloromethane and charged on a short silica gel column (1 × 2 cm). After flushing the column with 5 mL of dichloromethane, the product was eluted with hexane/ethyl acetate 1:1 and the solvent was removed in vacuo to give disaccharide **5** (45 mg, 48 μmol, 72% yield) as a colorless oil. HPLC (column: Inertsil ODS-2 5 μm, 4.6 × 150 mm, 30 °C, mobile phase: acetonitrile/water 85:15, flow rate: 1.0 mL/min, detection: UV210 nm) Rt = 6.2 min., purity: 96% (calcd. from the peak area); ^{1}H NMR (270 MHz, $CDCl_3$): δ = 7.95-7.80 (m, 6H), 7.56-7.49 (m, 1H), 7.44-7.20 (m, 21H), 7.09-7.05 (m, 2H), 5.89 (t, 1H, *J* = 9.57 Hz), 5.84 (dd, 1H, *J* = 9.57, 7.92 Hz), 5.50 (t, 1H, *J* = 9.57 Hz), 4.90 (d, 1H, *J* = 10.89 Hz), 4.81 (d, 1H, *J* = 7.59 Hz), 4.73 (d, 1H, *J* = 12.21 Hz), 4.70 (d, 1H, *J* = 10.89 Hz), 4.60 (d, 1H, *J* = 12.21 Hz), 4.85 (d, 1H, *J* = 9.21), 4.52 (d, 1H, *J* = 3.63 Hz), 4.33 (d, 1H, *J* = 11.21), 4.13-4.08 (m, 1H), 3.90 (t, 1H, *J* = 9.24), 3.78-3.70 (m, 5H), 3.45 (dd, 1H, *J* = 9.57, 3.63 Hz), 3.42 (t, 1H, *J* = 9.90), 3.23 (s, 3H), 2.50 (broad, 1H); ESI-MS m/z: Calcd for $C_{55}H_{54}NaO_{14}$ [$(M+Na)^+$] 961.3, found 961.4.

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(7) Commercially available from Argonaut Technologies, http://www.argotech.com.

(8) The macroporous resin, ArgoPore™, is heterogeneous in the pore size. It was thought that the glycosylation in smaller pores was interfered by steric hindrance of the resin itself and some of the glycosyl acceptors remained unchanged.

(9) Commercially available from Wako Pure Chemicals Industries, Ltd., http://www.wako-chem.co.jp/.

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