Organic Syntheses

Working with Hazardous Chemicals

The procedures in *Organic Syntheses* are intended for use only by persons with proper training in experimental organic chemistry. All hazardous materials should be handled using the standard procedures for work with chemicals described in references such as "Prudent Practices in the Laboratory" (The National Academies Press, Washington, D.C., 2011; the full text can be accessed free of charge at http://www.nap.edu/catalog.php?record_id=12654). All chemical waste should be disposed of in accordance with local regulations. For general guidelines for the management of chemical waste, see Chapter 8 of Prudent Practices.

In some articles in *Organic Syntheses*, chemical-specific hazards are highlighted in red "Caution Notes" within a procedure. It is important to recognize that the absence of a caution note does not imply that no significant hazards are associated with the chemicals involved in that procedure. Prior to performing a reaction, a thorough risk assessment should be carried out that includes a review of the potential hazards associated with each chemical and experimental operation on the scale that is planned for the procedure. Guidelines for carrying out a risk assessment and for analyzing the hazards associated with chemicals can be found in Chapter 4 of Prudent Practices.

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September 2014: The paragraphs above replace the section "Handling and Disposal of Hazardous Chemicals" in the originally published version of this article. The statements above do not supersede any specific hazard caution notes and safety instructions included in the procedure.

ENANTIOSELECTIVE HYDROLYSIS OF cis-3,5-DIACETOXYCYCLOPENTENE: (1R,4S)-(+)-4-HYDROXY-2-CYCLOPENTENYL ACETATE

[4-Cyclopentene-1,3-diol, monoacetate, (1R-cis)-]
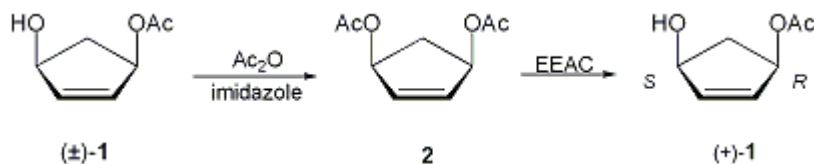
Submitted by Donald R. Deardorff, Colin Q. Windham, and Chris L. Craney[1].
Checked by Renaud Beaudegnies and Leon Ghosez.

1. Procedure

A. *cis-3,5-Diacetoxycyclopentene.* A flame-dried, 100-mL, single-necked, round-bottomed flask is equipped with a serum cap, Teflon-coated magnetic stirring bar, and an 18-gauge needle attached to a dry source of argon vented through a mineral oil bubbler. The apparatus is flushed with argon and charged with 16.70 g (118 mmol) of (±)-cis-4-acetoxy-1-hydroxycyclopent-2-ene [(±)-**1**] (Note 1) and 20 mL of methylene chloride (Note 2). To this stirred solution is added 8.81 g (129 mmol) of imidazole in one portion (Note 3). When dissolution is complete, the reaction mixture is cooled to 0°C with the aid of an ice-water bath. Acetic anhydride (13.20 g, 12.20 mL, 129 mmol) (Note 4) is added dropwise over 5 min by means of an oven-dried syringe and needle. The cooling bath is removed and the reaction is allowed to stir at ambient temperature until judged complete by TLC analysis (Note 5). The contents of the flask are transferred to a 250-mL separatory funnel and diluted with 60 mL of reagent grade ethyl ether. The contents of the separatory funnel are washed with 60 mL of ice-cold 1 N hydrochloric acid which results in noticeable removal of all color from the organic phase. The acidic aqueous phase is back-extracted with two 50-mL portions of ether (Note 6). The combined organic phases are washed with a single 50-mL portion of saturated sodium bicarbonate solution followed by back-extraction of the aqueous phase with three 50-mL portions of ether (Note 6). The combined organic extracts are dried over anhydrous magnesium sulfate, filtered, and concentrated under rotary evaporation at 30 mm. The resulting viscous tan oil is distilled through a 10-cm Vigreux column to afford 20.9–21.1 g (96–98%) of cis-3,5-diacetoxycyclopentene **(2)** (Note 7) as a colorless liquid, bp 55–56°C at 0.1 mm.

B. *(1R,4S)-(+)-4-Hydroxy-2-cyclopentenyl acetate.* A rigorously clean 1-L Erlenmeyer flask, equipped with a 2-in. long Teflon-coated stirring bar (Note 8), is charged with 320 mL of a 1.45 M sodium dihydrogen phosphate buffer concentrate (Note 9). The solution is diluted to a final volume of 800 mL with the addition of 480 mL of glass-distilled water. To the gently stirred solution (Note 10) is added 78 mg of sodium azide (Note 11) followed by 18.6 mg (200 units/mg) of lyophilized electric eel acetyl cholinesterase (EEAC) (Note 12). The enzyme completely dissolves within 5 min after which time 16.01 g (87.0 mmol) of cis-3,5-diacetoxycyclopentene is added in one portion. A few extra mL of distilled water are used to facilitate a quantitative transfer of the diacetate. Parafilm is placed over the mouth of the flask and the two-phase system (Note 13) is allowed to stir at room temperature (Note 14). The progress of the reaction is conveniently monitored by periodic TLC analysis. The reaction is terminated when only a trace of diacetate remains and the corresponding diol begins to appear (Note 15). The aqueous phase is first extracted three times with 150-mL portions of ether followed by fifteen 150-mL extractions with a 1:1 mixture of ether/ethyl acetate (Note 16). The organic extracts are combined, dried over anhydrous magnesium sulfate, and concentrated by rotory evaporation at ~30 mm (Note 17). The residue is passed through a 60-g plug of silica gel (Note 18) with approximately 750 mL of ether. Removal of the solvent by rotary evaporation followed by exposure to oil pump vacuum affords 11.32 g of nearly colorless material [mp 43–49°C (corrected)]. Vacuum distillation (Note 19) of the product through a short-path apparatus at 64°C/0.060 mm furnishes 10.64–10.76 g (86–87%, 96%

e.e.) of colorless solid (+) hydroxy acetate **1** (mp 44.5–49.5°C (corrected), $[\alpha]_D$ +62.9° (chloroform, c 1.465)) (Note 20). A sample recrystallized from 1:1 pentane/ether (approximately 5 mL/g) provides high purity (≥99% e.e.), colorless needles of **1** (mp 50.7–51.3°C (corrected), $[\alpha]_D^{23}$ +73.8° (chloroform, c 1.25) [lit.[2] mp 49° − 51°C, $[\alpha]_D^{23}$ +68.0° (chloroform, c 1.64)] (Note 21).

2. Notes

1. (±)-cis-4-Hydroxy-2-cyclopentenyl acetate [(±)-**1**] was prepared according to the method described in *Org. Synth*.[3] This material must be recrystallized (1:1 ether-pentane) before use to remove impurities (e.g., trans-monoacetate) that otherwise lead to reduced optical rotations in the final product [(+)-**1**]. The submitters have found that material having a melting point of 37–39°C is of satisfactory purity. The racemic monoacetate is hygroscopic so the proper precautions should be taken to prevent exposure to moisture. The NMR spectrum is as follows: ^{1}H NMR of (±) **1** (200 MHz, CDCl$_3$) δ: 1.64 (dt, 1 H, J = 14.7, 3.7), 2.02 (s, 3 H), 2.78 (overlapping dt, 1 H, J = 14.7, 7.5), 4.70 (overlapping s, 1 H and broad s), 5.45 (m, 1 H), 5.95 (m, 1H), 6.10 (m, 1H); ^{13}C NMR of (+) **1** (50 MHz, CDCl$_3$) δ: 21.2, 40.5, 74.9, 77.0, 132.6, 138.5, 170.8.

2. HPLC grade methylene chloride was purchased from Fisher Scientific Company and distilled from calcium hydride through a Vigreux column prior to use.

3. Imidazole (99% indicated purity) was purchased from Aldrich Chemical Company, Inc., and used without any further purification.

4. Acetic anhydride (98% indicated purity) was purchased from Aldrich Chemical Company, Inc., and distilled from phosphorus pentoxide through a Vigreux column prior to use.

5. Thin layer chromatographic (TLC) analysis was performed on Baker Si250F precoated plates with ethyl ether as the solvent. The approximate R_f values for the monoacetate and diacetate under these conditions are 0.44 and 0.72, respectively. The compounds were visualized using p-anisaldehyde stain.

6. This step is necessary to retrieve the slightly water-soluble product diester that migrates into the aqueous layer during the wash process. The meso-diester is easily detected in the water phase by TLC analysis (see (Note 5)).

7. cis-3,5-Diacetoxycyclopentene has the following spectral data: ^{1}H NMR of **2** (200 MHz, CDCl$_3$) δ: 1.74 (dt, 1 H, J = 15, 3.85), 2.05 (s, 6 H), 2.87 (overlapping dt, 1 H, J = 15, 7.6), 5.55 (m, 2 H), 6.1 (s, 2 H); ^{13}C NMR of **2** (50 MHz, CDCl$_3$) δ: 21.1, 37.1, 76.6, 134.6, 170.7.

8. The stirring bar should be free of any slivers of magnetic material or heavy metals that could inactivate the enzyme.

9. The buffer was prepared by dissolving 100 g of sodium dihydrogen phosphate monohydrate (Alfa Johnson Matthey) into 200 mL of glass-distilled water. The pH of the solution was adjusted to 6.9 with the addition of concentrated sodium hydroxide solution (MCB Reagent) and verified with a pH meter recently calibrated between 4–7 pH units using Fisher standards. The buffer solution was diluted to a final volume of 500 mL (1.45 M) and stored in a clean plastic bottle.

10. The stirring rate was adjusted so as to minimize the size of the vortex, which would introduce air into the solution and jeopardize the enzyme.

11. Sodium azide (Sigma Chemical Company) was used to protect the solution's components from microbial attack during the prolonged reaction period.

12. Lyophilized electric eel acetyl cholinesterase (C-3389, EC 3.1.1.7) was purchased from Sigma Chemical Company and stored below 0°C.

13. As the reaction nears completion, all the diacetate dissolves.

14. The success of this reaction may be temperature dependent. Shortly after starting this reaction on a particularly hot day, the building air conditioning failed. The temperature of the reaction flask quickly reached 28.7°C. The optical rotation of the distilled product was 4° off and the melting point was lower than expected. In all reactions prior to or since that incident the reaction temperatures never climbed above 23°C and the results have been uniformly superior.

15. Under these conditions, the reaction lasts 9–12 hr with a final pH of approximately 6.5. Nonspecific hydrolysis of **2** over this time accounts for less than 0.5% of the product as measured by control experiments. If cost is of little concern, the reaction time can be shortened dramatically by the addition of extra enzyme. cis-3,5-Dihydroxycyclopentene has an R_f value in ether of approximately 0.1.

16. HPLC grade ethyl acetate was purchased from J. T. Baker Inc. and distilled from phosphorus pentoxide prior to use. Although the product, (1R,4S)-(+)-4-hydroxy-2-cyclopentenyl acetate [(+)-**1**], is more soluble in ethyl acetate, emulsion concerns dictated the use of pure ether for the first three

extractions and then a 1:1 ether/ethyl acetate mixture for all subsequent extractions. When emulsions did occur during the extraction process, the unemulsified portion of the aqueous phase was drained and a few mL of brine solution was added to the emulsified phase. Two or three quick shakes alleviated the problem.

17. The residual oil may crystallize during this process. If so, it will be necessary to dissolve the solid in about 10 mL of ether (with the aid of a hot air gun) prior to passing the material through the silica gel plug.

18. Baker Analyzed Reagent silica gel 40-140 mesh was used in a 4.5-cm diameter column. This step removes the small amounts of diol present. TLC analysis was helpful in determining when all the monoacetate had been stripped from the plug.

19. The receiving flask should be submerged in an ice-water bath since the product has a considerable vapor pressure under these conditions. It is wise to have a heat gun nearby in the event that the condensate begins to crystallize in the condenser.

20. The enantiomeric excess was unambiguously determined via gas chromatographic separation of diastereomers obtained from derivatization of the chiral alcohol with Mosher's reagent,[4] (R)-(+)-α-methoxy-α-(trifluoromethyl)phenylacetic acid, (MTPA) 99% (Aldrich Chemical Company, Inc.). The checkers used the procedure of Noyori: Kitamura, M.; Tokunaga, M.; Ohkuma, T., Noyori, R. *Org. Synth., Coll. Vol. IX* **1998**, 592 for derivatization with Mosher's reagent. The MTPA esters derived from the (−)- and (+)-monoacetate enantiomers have retention times of 108 and 111.5 min, respectively, on a 25-m OV-101, crosslinked, methylsilicone capillary column operating at 150°C and flow rate of 22 cm/sec. Although less reliable than GC, the ratio of MTPA esters can be estimated via the use of high field (≥200 MHz) NMR in CDCl$_3$ solvent.[5]

21. One paper reports[5] an optical rotation of $[\alpha]_D^{22}$ +75.0° (chloroform, *c* 1.16) for crystals of (+)-**1** melting at only 47.5–48°C. We find these data to be inconsistent with our own experience and with the absolute values of the reported rotations for optically pure (−)-**1**. The polarimeter used in the submitter's analysis, a JACSO DIP-360, had been previously calibrated with NBS Standard Reference Sucrose 17d in accordance with NBS guidelines.

Waste Disposal Information

All toxic materials were disposed of in accordance with "Prudent Practices in the Laboratory"; National Academy Press; Washington, DC, 1995.

3. Discussion

In the 1980's there was a great increase in the development and use of enzymatic procedures by synthetic chemists.[6,7,8,9,10,11] Previously regarded more as scientific curiosities of limited scope than of practical utility, biological-chemical transformations are now used regularly by synthetic chemists. The ability to induce optical activity in molecules where none existed before is the most useful property of these chiral catalysts. Hydrolase enzymes are generally preferred over other kinds of enzymes for transformations of this nature because they are more easily handled and do not require cofactors for activity. In cases where enantiotopic differentiation between ester functions is desired, prochiral meso diesters are more efficient substrates than racemic esters. In the former case it is possible for all starting material to be converted into a single enantiomer, whereas in the latter example only enzymatic resolution is possible.

(1R,4S)-(+)-4-Hydroxy-2-cyclopentenyl acetate [(+)-**1**] is an important synthetic precursor. It provides optically active starting material, via the versatile intermediate 4-oxo-2-cyclopentenyl acetate,[12] for important cyclopentanoids such as the prostaglandins[13] and carbocyclic nucleosides.[14] Because of the medicinal significance of these compounds, more efficient routes with better enantioselectivities have been devised to nonracemic **1**. Enzymatic catalysis has become the dominant methodology for induction of this optical activity.

In addition to the title procedure,[15] other enzymatic preparations of nearly optically pure (1R,4S)-(+)-4-hydroxy-2-cyclopentenyl acetate [(+)-**1**][5,21] and its optical antipode (−)-**1**[16,17,18,19,20] are known. These enzyme-catalyzed procedures are derivatives of two basic strategies: (1) the enantioselective hydrolysis of the meso-diacetate **2**,[5,15,21,16,17] or (2) the enantioselective transacetylation of the parent

meso-diol.[18,19,20] Although (−)-**1** has been successfully prepared by either route, the (+)-enantiomer is available only via the hydrolytic approach.

The other enzymatic routes[5,21] to (+)-**1** follow roughly similar procedures. Both employ pH 7 buffer solutions of the hydrolytic enzyme porcine pancreas lipase (PPL) as opposed to electric eel acetyl cholinesterase (EEAC) used here. Although EEAC does afford better enantioselection than PPL (96% vs. 92% e.e.[21]), this apparent advantage may be viewed as somewhat tenuous since (+)-**1** can be optically enriched via repeated recrystallizations. However, EEAC is clearly superior to PPL in terms of chemical yield. PPL mediated hydrolyses lead to yields of (+)-**1** in the 50% range (plus recovered starting material), while the EEAC conversion rate is 90–95%. More importantly, the EEAC route eliminates the need to recycle starting material.
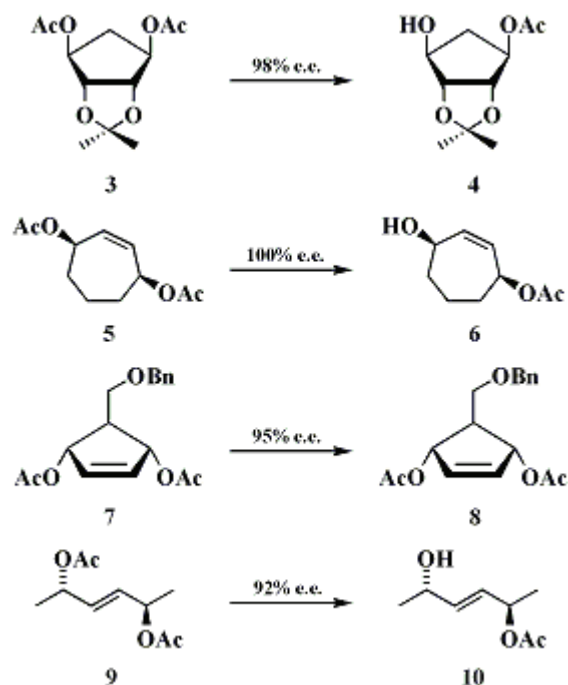
EEAC has been used successfully in other enantiotopic differentiations. Johnson, et al.[22] have reported that diester **3** can be readily transformed into hydroxy acetate **4** via this enzymatic process in 98% e.e. and an 80% chemical yield. Similarly, hydroxy acetate **6** was prepared from its parent diester **5** by Pearson, et al.[23] in 100% e.e., although in 39% yield (50–55% recovered starting material). The enzyme also appears effective on 4-substituted cis-3,5-diacetoxycyclopentenes as Danishefsky[24] demonstrated with the conversion of **7** into **8** in 95% yield and 95% e.e. Finally, the successful enantioselective hydrolysis of **9** into **10** (77%, 92% e.e.) extends the range of useful EEAC substrates to acyclic cases.[25]

Immobilized EEAC (Sigma Chemical Company) has been found equally effective and allows economical, large-scale hydrolyses of **2**. The immobilized enzyme, which is covalently bonded to agarose beads, is easily recovered through centrifugation. Reuse does not compromise the enantioselectivity (unpublished results).

This preparation is referenced from:

- Org. Syn. Coll. Vol. 9, 132
- Org. Syn. Coll. Vol. 9, 136

References and Notes

1. Department of Chemistry, Occidental College, Los Angeles, CA 90041. We gratefully acknowledge the National Science Foundation (CHE-8908212, CHE-8804037 & DMB-9005512) for financial support of this work. C. Q. W. wishes to thank the Ford Foundation for his summer undergraduate research stipend.
2. Busato, S.; Tinembart, O.; Zhang, Z. D.; Scheffold, R. *Tetrahedron* **1990**, *46*, 3155.
3. Deardorff, D. R.; Myles, D. C. *Org. Synth., Coll. Vol. VIII* **1993**, 13.
4. Dale, J. A.; Dull, D. L.; Mosher, H. S. *J. Org. Chem.* **1969**, *34*, 2543.
5. Sugai, T.; Mori, K. *Synthesis* **1988**, 19.
6. For recent reviews see: Chen, C.-S.; Sih, C. J. *Angew. Chem., Int. Ed. Engl.* **1989**, *28*, 695;
7. Ohno, M.; Otsuka, M. *Org. React.* **1989**, *37*, 1;
8. Jones, J. B.; *Tetrahedron 1986, 42,* 3351;
9. Whitesides, G. M.; Wong, C.-H. *Angew. Chem., Int. Ed. Engl.* **1985**, *24*, 617;
10. Suckling, C. J. In "Enzyme Chemistry, Impact and Applications;" Suckling, C. J., Ed., Chapman and Hall, New York, **1984**, pp. 78–118;
11. Whitesides, G. M.; Wong, C.-H. *Aldrichim. Acta* **1983**, *16*, 27.
12. Harre, M.; Raddatz, P.; Walenta, R.; Winterfeldt, E. *Angew. Chem., Int. Ed. Engl.* **1982**, *21*, 480.
13. Noyori, R.; Suzuki, M. *Angew. Chem., Int. Ed. Engl.* **1984**, *23*, 847.
14. Deardorff, D. R. ; Shambayati, S.; Myles, D. C.; Heerding, D. *J. Org. Chem.* **1988**, *53*, 3614.
15. Deardorff, D. R.; Matthews, A. J.; McMeekin, D. S.; Craney, C. L. *Tetrahedron Lett.* **1986**, *27*, 1255.
16. Wang, Y.-F.; Chen, C.-S.; Girdaukas, G.; Sih, C. J. *J. Am. Chem. Soc.* **1984**, *106*, 3695.
17. Laumen, K.; Schneider, M. *Tetrahedron Lett.* **1984**, *25*, 5875.
18. Theil, F.; Ballschuh, S.; Schick, H.; Haupt, M.; Häfner, B.; Schwarz, S. *Synthesis* **1988**, 540.
19. Jommi, G.; Orsini, F.; Sisti, M.; Verotta, L. *Gazz. Chim. Ital.* **1988**, *118*, 863.
20. Babiak, K. A.; Ng, J. S.; Dygos, J. H.; Weyker, C. L.; Wang, Y.-F.; Wong, C.-H. *J. Org. Chem.* **1990**, *55*, 3377.
21. Laumen, K.; Schneider, M. P. *J. Chem. Soc., Chem. Commun.* **1986**, 1298.
22. Johnson, C. R.; Penning, T.D. *J. Am. Chem. Soc.* **1986**, *108*, 5655.
23. Pearson, A. J.; Bansal, H. S.; Lai, Y.-S. *J. Chem. Soc., Chem. Commun.* **1987**, 519.
24. Griffith, D. A.; Danishefsky, S. J. *J. Am. Chem. Soc.* **1991**, *113*, 5863.
25. Schink, H. E.; Bäckvall, J-E. *J. Org. Chem.* **1992**, *57*, 1588.

Appendix
Chemical Abstracts Nomenclature (Collective Index Number); (Registry Number)

brine

acetyl cholinesterase

EEAC

hydrochloric acid (7647-01-0)

ethyl acetate (141-78-6)

ether,
ethyl ether (60-29-7)

acetic anhydride (108-24-7)

sodium hydroxide (1310-73-2)

chloroform (67-66-3)

sodium bicarbonate (144-55-8)

sucrose

Pentane (109-66-0)

sodium azide (26628-22-8)

methylene chloride (75-09-2)

magnesium sulfate (7487-88-9)

Imidazole (288-32-4)

argon (7440-37-1)

sodium dihydrogen phosphate (7558-80-7)

calcium hydride (7789-78-8)

p-anisaldehyde (123-11-5)

4-oxo-2-cyclopentenyl acetate (768-48-9)

phosphorus pentoxide (1314-56-3)

(1R,4S)-(+)-4-Hydroxy-2-cyclopentenyl acetate,
4-Cyclopentene-1,3-diol, monoacetate, (1R-cis)-,
(±)-cis-4-Hydroxy-2-cyclopentenyl acetate (60410-16-4)

cis-3,5-Diacetoxycyclopentene (54664-61-8)

(±)-cis-4-acetoxy-1-hydroxycyclopent-2-ene (60410-18-6)

sodium dihydrogen phosphate monohydrate (10049-21-5)

cis-3,5-Dihydroxycyclopentene

(R)-(+)-α-methoxy-α-(trifluoromethyl)phenylacetic acid (20445-31-2)