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Synthesis of all four stereoisomers of 5-hydroxy-4-methyl-3-heptanone using plants and oyster mushrooms

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ABSTRACT

All four possible stereoisomers of 5-hydroxy-4-methyl-3-heptanone were synthesized from common achiral reagents using fast, straightforward organic synthesis, including the use of whole tissue of *Daucus carota*, *Solanum melongena*, and *Pleurotus ostreatus*.

1. Introduction

(4*R*,5*S*)-5-Hydroxy-4-methyl-3-heptanone ((4*R*,5*S*)-**1a**), known as Sitophilure (Fig. 1), has been found to be an aggregation pheromone for *Sitophilus* weevils, such as the rice weevil *Sitophilus oryzae* and the maize weevil *Sitophilus zeamais*.[1] One of the *anti* isomers of this compound, (4*S*,5*S*)-**2a**, was found to be a compound specific to males, when volatile emissions from individuals of the lucerne weevil *Sitona discoideus* were analyzed, in order to attempt the identification of its sex pheromone.[2] To be able to determine whether (4*S*,5*S*)-**2a** was in fact a pheromone for this species, the compound had to be synthesized. We also wanted to obtain the remaining three stereoisomers for tests of synergetic or inhibitory effects.

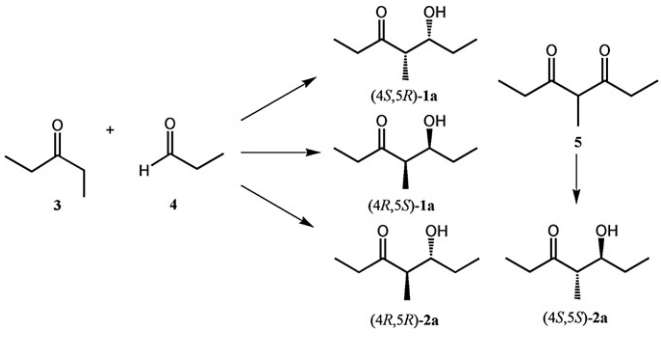

Figure 1. Starting materials and target compounds.

Numerous routes to the various stereoisomers of 5-hydroxy-4-methyl-3-heptanone (**1a** and **2a**) have been reported over the last decades.[3] For example Mori and Ebata have prepared all possible isomers from methyl (*R*)-3-hydroxypentanoate[3g] and more recently new biocatalytic routes have been presented, among some are: Pilli's and Riatto's preparation of (+)-Sitophilure from methyl-3-oxopentanoate using *S. cerevisiae*[3b] and the preparation of the same compound by Kalaitzakis et al. utilizing isolated NADPH-dependent ketoreductases.[3f] Our approach is based on the well known fact that diastereoselectivity of aldol additions of aldehydes to ketones can be directed by the choice of base in combination with added Lewis acids. We wanted to obtain the stereochemically pure isomers of **1a** and **2a** by utilizing diastereoselective aldol additions in combination with purifications by a common, commercially available lipase, whole tissue of vegetables and mushrooms, and column chromatography. By combining these methods, inexpensive and commercially available achiral reagents could be used to prepare substances highly enantioselectively with relative ease. Our aim of this project was to obtain the two *syn* isomers: (4*R*,5*S*)-**1a** and (4*S*,5*R*)-**1a** as well as the two *anti* isomers: (4*R*,5*R*)-**2a** and (4*S*,5*S*)-**2a** as four separate samples in an isomeric purity greater than 95%.

2. Results and discussion

In order to evaluate the relationship between stereochemistry and bioactivity, a synthesis of all four possible stereoisomers of

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5-hydroxy-4-methyl-3-heptanone ((4*R*,5*S*)-**1a**, (4*S*,5*R*)-**1a**, (4*R*,5*R*)-**2a**, and (4*S*,5*S*)-**2a**) was carried out. These syntheses consisted of 11 steps (Figs. 1 and 2) and total yields of the individual isomers ranged from 6 to 23%. The crude *syn* products **1a** and *anti* products **2a** of 5-hydroxy-4-methyl-3-heptanone were synthesized by the aldol addition between 3-pentanone (**3**) and *n*-propanal (**4**). Transesterification using lipase Amano PS-D from *Pseudomonas cepacia* gave further enrichment of one pair of enantiomers and column chromatography on silica gel gave partial separation of the two pairs of diastereomers. Finally, the choice of plant tissue in the stereoselective hydrolysis of the acetate esters **1b** and **2b** formed by the lipase further enhanced the isomeric purity of the final products.

Compounds (4*R*,5*S*)-**1a** and (4*S*,5*R*)-**1a**, the two *syn* isomers, were derived from **3** and **4**, where the selectivity for the *syn* products was achieved by formation of a titanium enolate from $TiCl_4$ and diisopropylethylamine.[4] Further enhancement of the isomeric purity was achieved by a lipase-mediated transacetylation with Amano PS-D lipase and vinyl acetate.[5] The main product (4*S*,5*R*)-**1b** was enriched further by column chromatography,[6] taking advantage of the partial separation found for the two diastereomers on silica gel. The remaining alcohol was predominantly (4*R*,5*S*)-**1a**, which was treated with lipase a second time, then acetylated and subjected to chromatography. The two acetate esters (4*R*,5*S*)-**1b** and (4*S*,5*R*)-**1b** were hydrolyzed selectively by the means of whole plant tissue of eggplant *Solanum melongena*,[7] yielding (4*R*,5*S*)-**1a** and (4*S*,5*R*)-**1a** in >95% isomeric purity.

Compound (4*R*,5*R*)-**2a** was synthesized via a Mg(II)-mediated aldol addition between **3** and **4**, selectively forming the *anti* isomers.[8] The initial aldol product, formed by an LDA reaction in THF, was treated with $MgBr_2$-etherate at low temperature followed by warming to rt, thus converting the *syn* product to a predominantly *anti* product. Analogously as for the *syn* products, a further increase in isomeric purity was achieved by a lipase-mediated

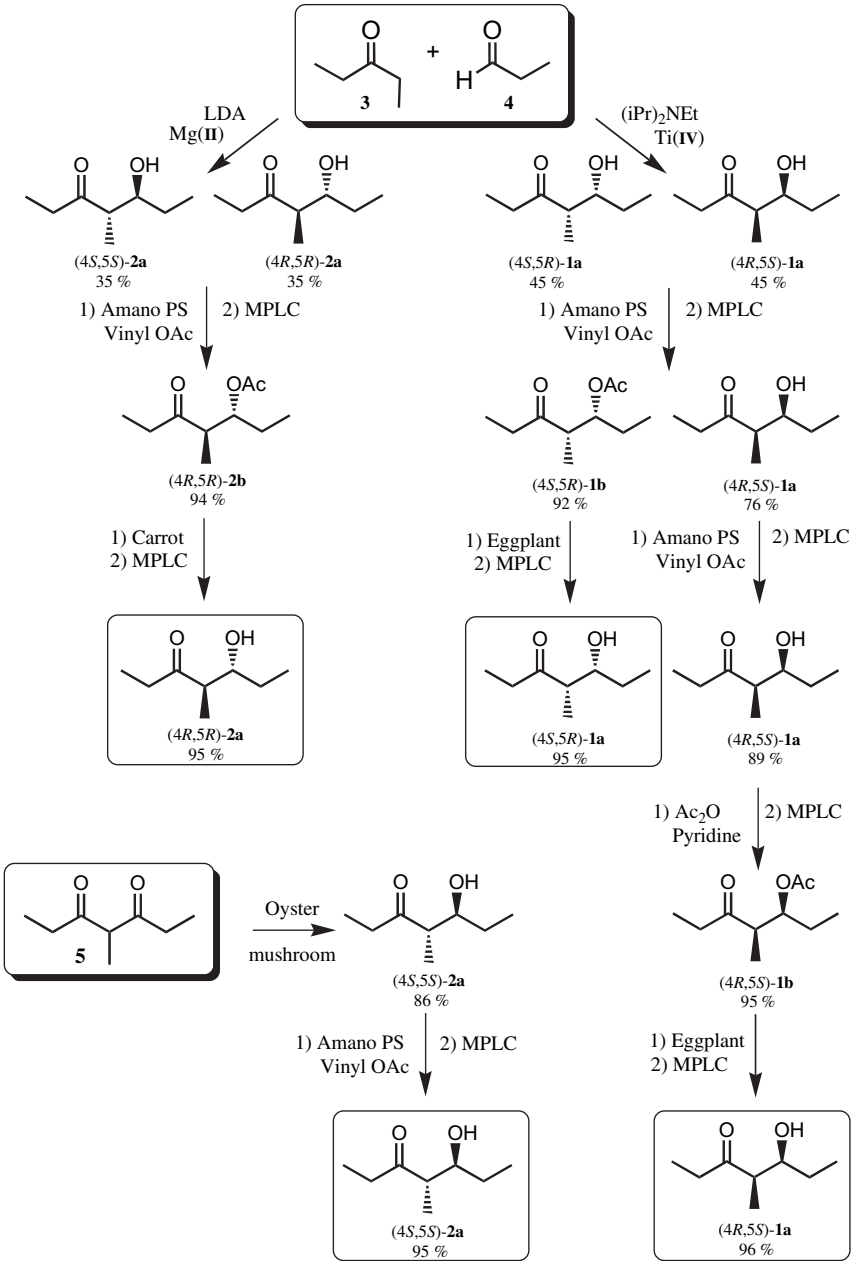

Figure 2. Synthesis of all four stereoisomers of 5-hydroxy-4-methyl-3-heptanone.

transacetylation followed by chromatography of the acetate (4*R*,5*R*)-**2b**. The selective hydrolysis of this ester was performed using whole tissue of carrot *Daucus carota* yielding the product (4*R*,5*R*)-**2a** in 95% isomeric purity.

Compound (4*S*,5*S*)-**2a** was synthesized by a regio- and enantioselective reduction of the diketone **5** by the means of whole tissue of oyster mushrooms *Pleurotus ostreatus*. The crude product was purified by chromatography and subjected to a lipase transacetylation. The enriched hydroxyketone was isolated from the formed acetate esters by chromatography yielding (4*S*,5*S*)-**2a** in 95% isomeric purity. In theory this stereoisomer could have been prepared by a similar method as for (4*R*,5*S*)-**1a**, using *rac*-**2a** instead of *rac*-**1a** as starting material, but our method for preparation of (4*S*,5*S*)-**2a** is shorter.

It is important to note that the hydrolysis of the acetates **1b** and **2b** proceeded smoothly to the β-hydroxyketones **1a** and **2a** when the whole tissue of carrot and eggplant was used. This is in stark contrast to our attempts to execute chemical hydrolysis under alkaline standard conditions, which instantly yielded the elimination product 4-methyl-4-heptene-3-one. We also tried chemical hydrolysis under acidic conditions, as well as various commercial lipases (lipase B from *Candida antarctica* and Amano PS-D from *Pseudomonas cepacia*), esterase (Pig liver esterase), and protease (Subtilisin A) in attempts to hydrolyze and alcoholyze **1b** and **2b** without success.

The regio- and enantioselective reduction of the diketone **5** to (4*S*,5*S*)-**2a** by the use of oyster mushrooms (or other mushrooms) is an attractive alternative to the use of baker's yeast. Although excellent enantioselectivity and reasonable yields have been reported for reductions of β-diketones by baker's yeast,[9] other authors report much lower yields (10–15%) of the *anti* product (>96% diastereomeric purity, no enantiomeric purity given) for the reduction using **5** as substrate.[10] In our laboratory the yields and stereoselectivity of this reaction were substantially lower than those reported above, while in experiments using oyster mushrooms we have obtained a crude product containing 82% of the desired enantiomer in 95% chemical purity and 40% yield, without any use of commercial lipases (unpublished data).

3. Conclusions

We have described methods of stereoselective synthesis and separation, such as aldol addition, lipase transesterification, and column chromatography as well as ester hydrolysis mediated by plants and mushrooms. When these methods were employed individually they gave moderate isomeric purity of certain isomers of 5-hydroxy-4-methyl-3-heptanone **1a** and **2a** but when combined all stereoisomers of **1a** and **2a** could be prepared from common achiral starting materials in good enantiomeric and diastereomeric purity.

4. Experimental

4.1. General

For all synthesized compounds ^{1}H NMR and ^{13}C NMR spectra of $CDCl_3$ solutions were recorded at 500 MHz and 125 MHz, using a Varian Unity spectrometer. Chemical shifts were expressed in ppm in relation to tetramethylsilane, multiplicity (s, singlet; d, doublet; t, triplet; q, quintet and m, multiplet), coupling constants (Hz), and number of protons. The starting materials employed were obtained from commercial suppliers and used without further purification. All vegetables and mushrooms were obtained from the local supermarket. Mass spectra were obtained with an HP 6890GC interfaced to an HP 5973 mass selective detector, in electron impact mode (70 eV), with helium as the carrier gas. The GC was equipped with a BPX-70 column (30 m×0.25 mm, i.d.×0.25 μm, SGE Australia). Enantioselective GC was performed with an HP 5890 GC fitted with a CYCLOSILB column (30 m×0.25 mm, i.d.×0.25 μm, J & W Scientific, USA). Column chromatography on silica gel (Merck 60, 0.040–0.063 mm) was carried out using a Separo medium pressure liquid chromatography (MPLC) system.[6] The column inner diameter was 15, 20, or 25 mm and gradient elution was applied, using cyclohexane and increasing amounts of ethyl acetate. All chemicals used as starting materials in the syntheses, were used as delivered from Sigma–Aldrich (Sweden), and Alfa Aesar (Germany). Anhydrous solvents were used and reactions were carried out under nitrogen when appropriate.

4.2. Synthesis of *syn* 5-hydroxy-4-methyl-3-heptanone (1a)

In dichloromethane (20 mL), while stirring, 3-pentanone (**3**) (0.43 g, 5.0 mmol) was dissolved and the solution was cooled to −80 °C. Titanium tetrachloride (0.60 mL, 5.5 mmol) was added dropwise. After 5 min, diisopropylethylamine (1.02 mL, 6.0 mmol) was added dropwise and the mixture was stirred for 1.5 h at the same temperature. Propanal (**4**) (0.44 mL, 6.0 mmol) was added dropwise and stirring was continued for 1.5 h under the same conditions. NH_4Cl (satd, aq, ca. 20 mL) was added, the cooling bath was removed, and water (ca. 10 mL) and diethyl ether (ca. 20 mL) were added. The aqueous phase was separated and extracted with diethyl ether (ca. 2×20 mL), the combined organic phases were washed with brine (ca. 3×20 mL), dried over $MgSO_4$, and the solvents were removed in vacuo, yielding the product **1a** as a slightly yellow oil of 98% purity (0.62 g, 90% *syn*, 84% yield). GC–MS: 126(15), 97(14), 86(37), 70(18), 69(11), 59(16), 57(100), 55(15). For NMR see individual isomers.

4.3. Synthesis of *anti* 5-hydroxy-4-methyl-3-heptanone (2a)

Diisopropylamine (12.6 mL, 90 mmol) was dissolved in THF (200 mL) and cooled to 0 °C. Butyllithium (2.0 M in pentane, 30.0 mL, 60 mmol) was added slowly while stirring and after 20 min at rt, the mixture was cooled to −70 °C. 3-Pentanone (**3**) (5.16 g, 60 mmol) was added slowly and stirring was continued for 30 min, whereafter propanal (**4**) (3.83 g, 66 mmol) was added. At the same temperature, after 20 min, $MgBr_2 \cdot OEt_2$ (19.4 g, 76 mmol) was added in one portion. After continued stirring for 5 min, the mixture was allowed to reach rt and after another 3 h NH_4Cl (satd, aq, ca. 200 mL) was added followed by diethyl ether (ca. 100 mL) and water (ca. 50 mL). The aqueous phase was extracted with diethyl ether (ca. 3×50 mL) and then the combined organic phases were washed with brine (ca. 2×50 mL) and dried over $MgSO_4$. The solvents were removed in vacuo yielding the product **2a** as a yellow oil of 54% purity (8.78 g, 71% *anti*). Yield: 55%. After purification by MPLC 4.35 g product of 89% purity was collected, to be used for the next step. For GC–MS see **1a** and for NMR see individual isomers.

4.4. Synthesis of 4-methyl-3,5-heptanedione (5)

To acetone (250 mL), 3,5-heptanedione (12.8 g, 0.098 mol) and potassium carbonate (oven dried, 12.8 g, 0.094 mol) were added at rt while stirring. After 15 min, iodomethane (7.8 mL, 0.12 mol) was added dropwise. The solution was heated to reflux overnight. After 16 h, the heating was discontinued and at rt diethyl ether (ca. 200 mL) was added and stirring was continued for 30 min. Solids were filtered off and the remaining solution was concentrated in vacuo to yield **5** as a yellow oil of 90% purity (14.8 g), containing some residue of solvent. The product was used as such in the following steps. GC–MS[11] and NMR[3f] corresponded to literature data. GC–MS: 142(5), 114(5), 113(8), 86(45), 57(100). ^{1}H NMR δ: 3.68

(q, 1H, J=7.1 Hz), 2.49 (m, 4H), 1.31 (d, 3H, J=7.1 Hz), 1.04 (t, 6H, J=7.2 Hz); ^{13}C NMR δ: 207.9, 60.5, 35.0, 13.1, 7.8 ppm.

4.5. Synthesis of (4*R*,5*S*)-5-hydroxy-4-methyl-3-heptanone ((4*R*,5*S*)-1a)

Compound *syn*-**1a** [0.60 g, 4.2 mmol (sum of isomers), 90% *syn*] was mixed with vinyl acetate (2.0 g, 23.2 mmol) and Amano PS-D lipase (0.60 g) was added. The mixture was incubated at 25 °C/120 rpm in a shaking water bath. After 4 days the lipase was filtered off and rinsed with dichloromethane. The solvents were removed in vacuo and the enriched alcohol was isolated from the formed acetate by MPLC yielding a fraction of 331 mg of 76% enantiomeric purity. This product (331 mg, 2.3 mmol) was treated again with Amano PS-D lipase (0.30 g) in vinyl acetate (1.0 g, 11.6 mmol) under the same conditions for 8 days. Analogous work-up and MPLC yielded 225 mg (75%) of (4*R*,5*S*)-**1a** of 89% isomeric purity.

The crude product (4*R*,5*S*)-**1a** (225 mg, 1.6 mmol) was treated with acetic anhydride (4.05 g, 40 mmol) in pyridine (11.0 g, 139 mmol) at rt overnight. Then the mixture was poured on ice and extracted with pentane (ca. 3×15 mL), washed with $CuSO_4$ (10%, aq) until no change in color was observed (ca. 50 mL), water (ca. 15 mL) and brine (ca. 15 mL), and dried over $MgSO_4$. The solvents were removed in vacuo to yield a colorless product (234 mg). This product was purified by repetitive MPLC (two times) to yield the ester (4*R*,5*S*)-**1b** in 95% isomeric purity (178 mg, some solvent residue remaining).

The acetate (4*R*,5*S*)-**1b** (178 mg, including solvent residue) was mixed with water (150 mL). Eggplant *S. melongena* (70 g) was cut into thin slices (c. 5 mm) and added to the mixture, which was incubated in an open container at 25 °C/120 rpm in a shaking water bath for 14 h. The eggplant tissue was filtered off and rinsed with water (ca. 4×60 mL). The remaining aqueous solution was extracted with diethyl ether (ca. 5×50 mL), washed with water (ca. 2×50 mL) and brine (ca. 50 mL), dried over $MgSO_4$, and concentrated in vacuo to yield 53 mg crude product. This product was subjected to MPLC and 42 mg of (4*R*,5*S*)-**1a** was isolated in 96% isomeric purity and 98% chemical purity. Overall yield from 3-pentanone: 12%. ^{1}H NMR δ: 3.82 (m, 1H), 2.45–2.60 (m, 3H), 1.52 (m, 1H), 1.38 (m, 1H), 1.13 (d, 3H, J=7.2 Hz), 1.06 (t, 3H, J=7.2 Hz), 0.95 (t, 3H, J=7.4 Hz); ^{13}C NMR δ: 217.2, 72.8, 49.5, 35.3, 27.1, 10.6, 10.1, 7.8 ppm. For MS see *syn*-**1a**.

4.6. Synthesis of (4*S*,5*R*)-5-hydroxy-4-methyl-3-heptanone ((4*S*,5*R*)-1a)

Compound *syn*-**1a** [0.60 g, 4.2 mmol (sum of isomers), 90% *syn*] was mixed with vinyl acetate (2.0 g, 23.2 mmol) and Amano PS-D lipase (0.30 g) was added. The mixture was incubated at 25 °C/120 rpm in a shaking water bath. After 4 days the lipase was filtered off and rinsed with dichloromethane. The solvents were removed in vacuo, the product was isolated from the remaining starting material and enriched by repetitive MPLC (two times) yielding a fraction of 185 mg (47%) (4*S*,5*R*)-**1b** in 92% isomeric purity.

The acetate (4*S*,5*R*)-**1b** (185 mg, 0.99 mmol) was mixed with water (150 mL). Eggplant (*S. melongena*, 70 g) was cut into thin slices (ca. 5 mm) and added to the mixture, which was incubated in an open container at 25 °C/120 rpm in a shaking water bath for 20 h. The biological tissue was filtered off and rinsed with water (ca. 4×50 mL). The remaining aqueous solution was extracted with diethyl ether (ca. 3×30 mL), washed with water (ca. 2×30 mL) and brine (ca. 30 mL), dried over $MgSO_4$, and concentrated in vacuo to yield 61 mg crude product. This product was subjected to MPLC and 42 mg (4*S*,5*R*)-**1a** was isolated in 95% isomeric purity and 98% chemical purity. Overall yield from 3-pentanone: 12%. For spectral data see (4*R*,5*S*)-**1a**.

4.7. Synthesis of (4*R*,5*R*)-5-hydroxy-4-methyl-3-heptanone ((4*R*,5*R*)-2a)

Compound *anti*-**2a** [1.80 g, 12.5 mmol (sum of enantiomers), 70% *anti*] was mixed with vinyl acetate (7.2 g, 84 mmol) and Amano PS-D lipase (1.80 g) was added. The mixture was incubated at 25 °C/120 rpm in a shaking water bath. After two days the lipase was filtered off and rinsed with dichloromethane. The solvents were removed in vacuo and the product was isolated from the remaining starting material and enriched by repetitive MPLC (three times) yielding a fraction of 426 mg (4*R*,5*R*)-**2b** of 94% isomeric purity (36%[†]).

The acetate (4*R*,5*R*)-**2b** (426 mg, 2.3 mmol) was mixed with water (300 mL). Carrot *D. carota* (90 g) was cut into thin slices (ca. 3 mm) and added to the mixture, which was incubated in an open container at 25 °C/120 rpm in a shaking water bath for 64 h. The biological tissue was filtered off and rinsed with water (ca. 4×100 mL). The remaining aqueous solution was extracted with diethyl ether (ca. 3×50 mL), washed with brine (ca. 50 mL), dried over $MgSO_4$, and concentrated in vacuo to yield 240 mg crude product. This product was subjected to MPLC and a fraction of 85 mg (4*R*,5*R*)-**2a** was isolated in 95% isomeric purity and 98% chemical purity. Overall yield from 3-pentanone: 6%. ^{1}H NMR δ: 3.62 (m, 1H), 2.40–2.70 (m, 3H), 1.58 (m, 1H), 1.41 (m, 1H), 1.13 (d, 3H, J=7.2 Hz), 1.05 (t, 3H, J=7.2 Hz), 0.98 (t, 3H, J=7.4 Hz); ^{13}C NMR δ: 217.1, 75.2, 50.8, 36.3, 27.8, 14.5, 10.1, 7.8 ppm. For MS see *syn*-**1a**.

4.8. Synthesis of (4*S*,5*S*)-5-hydroxy-4-methyl-3-heptanone ((4*S*,5*S*)-2a)

4-Methyl-3,5-heptanedione **5** [1.20 g, 7.6 mmol (calcd from 90% purity)] was mixed with water (1.8 L). Oyster mushrooms *P. ostreatus* (600 g) were cut into thin slices (c. 5 mm) and added to the mixture, which was incubated in six open 250 mL Pyrex bottles at 25 °C/120 rpm in a shaking water bath for 72 h. The biological material was filtered off and rinsed with water (ca. 4×250 mL). The remaining aqueous solution was extracted with diethyl ether (ca. 3×200 mL), washed with brine (ca. 200 mL), dried over $MgSO_4$, and concentrated in vacuo to yield 1.27 g crude product. This product was subjected to MPLC and a fraction of 287 mg of (4*S*,5*S*)-**2a** was isolated in 86% isomeric purity and 96% chemical purity.

The crude product (4*S*,5*S*)-**2a** (287 mg, 2.0 mmol) was mixed with vinyl acetate (1.0 g, 12 mmol) and Amano PS-D lipase (0.20 g) was added. The mixture was incubated at 25 °C/120 rpm in a shaking water bath. After 4 days the lipase was filtered off and rinsed with dichloromethane. The solvents were removed in vacuo and the enriched alcohol was isolated from the formed ester by MPLC yielding a fraction of 253 mg (4*S*,5*S*)-**2a** of 95% isomeric purity. Total yield from 3,5-heptanedione: 23%.

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References and notes

1. Schmuff, N. R.; Phillips, J. K.; Burkholder, W. E.; Fales, H. M.; Chen, C.-W.; Roller, P. P.; Ma, M. *Tetrahedron Lett.* **1984**, *25*, 1533.
2. Unelius, C. R.; Wee, S. L.; McNeill, M.; Daly, J.; Bunn, B. J.; Gibb, A. R.; Bohman, B.; Suckling, D. M. In preparation.

[†] Yields calculated from the amount of (4*R*,5*R*)-**2a** present in the racemic starting material, i.e., the yield obtained if (4*S*,5*S*)-**2a** would have been retrieved from the reaction mixture.

3. Some examples are (a) Mori, K.; Yoshimura, T.; Sugai, T. *Liebigs Ann. Chem.* **1988**, 899; (b) Pilli, R. A.; Riatto, V. B. *J. Braz. Chem. Soc.* **1999**, *10*, 363; (c) Delas, C.; Szymoniak, J.; Thery, N.; Moïse, C. *Synth. Commun.* **1998**, *28*, 2613; (d) Fujisawa, T.; Mobele, B. I.; Shimizu, M. *Tetrahedron Lett.* **1992**, *33*, 5567; (e) Fauve, A.; Veschambre, H. *Tetrahedron Lett.* **1987**, *28*, 5037; (f) Kalaitzakis, D.; Rozzell, J. D.; Kambourakis, S.; Smonou, I. *Eur. J. Org. Chem.* **2006**, 2309; (g) Mori, K.; Ebata, T. *Tetrahedron* **1986**, *42*, 4421.
4. Evans, D. A.; Rieger, D. L.; Bilodeau, M. T.; Urpi, F. *J. Am. Chem. Soc.* **1991**, *113*, 1047.
5. Pontes, G. B.; Bohman, B.; Unelius, C. R.; Lorenzo, M. G. *J. Chem. Ecol.* **2008**, *34*, 450.
6. Baeckstrom, P.; Stridh, K.; Li, L.; Norin, T. *Acta Chem. Scand.* **1987**, *B41*, 447.
7. Bohman, B.; Cavonius, L. R.; Unelius, C. R. *Green Chemistry* **2009**. doi:10.1039/b913936b
8. Swiss, K. A.; Choi, W.-B.; Liotta, D. C.; Abdel-Magid, A. F.; Maryanoff, C. A. *J. Org. Chem.* **1991**, *56*, 5978.
9. (a) Bolte, J.; Gourcy, J.-G.; Veschambre, H. *Tetrahedron Lett.* **1986**, *27*, 565; (b) Fauve, A.; Veschambre, H. *J. Org. Chem.* **1988**, *53*, 5215.
10. Koutek, B.; Korblova, E.; Kreckova, J.; Vrkoc, J. In *Endocrinological Frontiers in Physiological Insect Ecology*; Sehnal, F., Zabza, A., Denlinger, D. L., Eds.; Wroclaw Technical University: Wroclaw, 1988; pp 767–770.
11. Blight, M. M.; Pickett, J. A.; Smith, M. C.; Wadhams, L. J. *Naturwissenschaften* **1984**, *71*, 480.