Pivotal Role of (*E*)-3-Carbamoyl-1-propenylboronic Acid in the Combination of Suzuki-Miyaura Coupling and Enzyme Reactions: Synthesis of (*3E,5E*)- and (*3E,5Z*)-6-Phenyl-3,5-hexadienoic Acid

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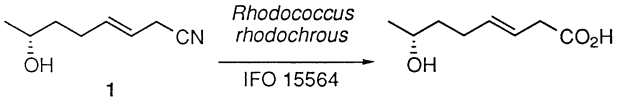
(Received April 24, 1997; CL-970307)

The first example of the enzyme-catalyzed hydrolysis of a nitrile bearing alkenylboronic acid functionality and its application to the synthesis of dienecarboxylic acids are described. (*E*)-3-Carbamoyl-1-propenylboronic acid was obtained by an incubation of the corresponding nitrile with *Rhodococcus rhodochrous* IFO 15564. Palladium(0)-catalyzed cross-coupling reaction of the product with (*E*)- and (*Z*)-bromoalkenes afforded the (3*E*,5*E*)- and (3*E*,5*Z*)-unsaturated amides, respectively. The second incubation of the above amides with the same microorganism provided the carboxylic acids with the defined configuration of conjugated double bonds.

Enzyme-mediated hydrolysis of nitriles allows their transformation into amides and carboxylic acids under mild conditions.[1] Indeed, the hydrolysis of β,γ-unsaturated nitrile **1** proceeded without any migration or isomerization of double bond, which was exploited as the key-step of a natural product synthesis.[2] The successful result prompted us a preparation of novel building blocks **2** and **3**. These boronic acids would be coupled with alkenyl halides with a catalytic action of a palladium(0) complex under Suzuki-Miyaura cross-coupling conditions[3] to provide dienes with a defined (*E*,*E*)- or (*E*,*Z*)-configuration.



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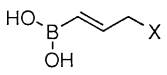
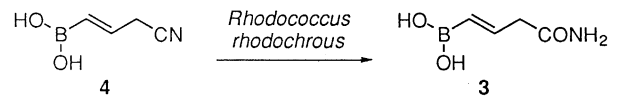


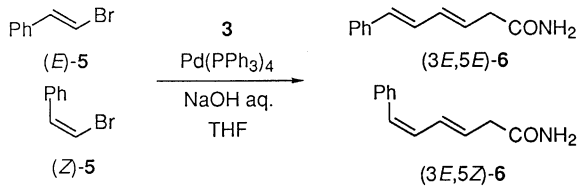
2: X = CO_2H
3: X = $CONH_2$
4: X = CN

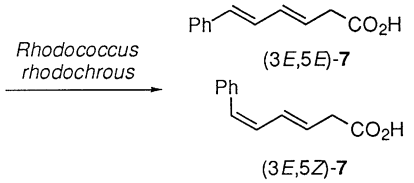
Toward this end, nitrile **4**[4] was prepared from propargyl bromide via hydroboration with catecholborane[5] and the subsequent treatment with tetraethylammonium cyanide.[6] Hydrolysis of **4** by an incubation with the resting cells of *Rhodococcus rhodochrous* IFO 15564 smoothly proceeded to afford the corresponding amide **3** (75%).[7] This result indicated that the major product of the microbial hydrolysis was ascribed to the action of nitrile hydratase. In contrast, the further action of amidase on **3** was slow; even after a prolonged incubation (7 days), the yield of the carboxylic acid **2** reached only as low as 30%. Previous examples showed that amides possessing another carboxylic acid functionality in the same molecules were poor substrates of amidase,[8] thus the total result would be explained by an analogy of boronic acid to carboxylic acid.



4 3

The subsequent palladium(0)-catalyzed cross-coupling reaction of **3** with bromoalkenes (*E*)-**5**[9] and (*Z*)-**5**[10] worked to give (3*E*,5*E*)-**6** (49%) and (3*E*,5*Z*)-**6** (70%)[11] under an elaborated condition; a careful addition of base (0.7N aqueous NaOH solution)[12] in several portions was neccesary with monitoring the progress of the reaction. The further incubation of the coupling products **6** yielded the corresponding carboxylic acids (3*E*,5*E*)-**7** (87%) and (3*E*,5*Z*)-**7** (79%) without any disarrangement of the configuration of the double bonds.[13]





In conclusion, (3*E*,5*E*)- and (3*E*,5*Z*)-6-phenyl-3,5-hexadienoic acids were synthesized by the combination of *R. rhodochrous*-mediated hydrolysis and palladium(0)-catalyzed coupling reaction. As the key intermediate, (*E*)-3-carbamoyl-1-propenylboronic acid can be isolated as very stable crystallines, the present synthesis will lead to a convenient preparation of various amides and carboxylic acids with diene systems of defined configuration.

References and Notes

1 J. Crosby, J. Moilliet, J. S. Parratt, and N. J. Turner, *J. Chem. Soc., Perkin 1*, **1994**, 1679; T. Sugai, T. Yamazaki, M. Yokoyama, and H. Ohta, *Biosci. Biotech. Biochem.*, submitted.

2 T. Sugai, O. Katoh, and H. Ohta, *Tetrahedron*, **51**, 11987 (1995).

3 N. Miyaura and A. Suzuki, *Chem. Rev.*, **95**, 2457 (1995) and references cited therein.

4 Experimental procedure and properties of nitrile **4**: The adduct of propargyl bromide and catecholborane was prepared as a solid according to the reported procedure.[5] To a solution of the adduct (5.00 g, 20.9 mmol) in dry dichloromethane (4.2 ml) was added dropwise a solution of tetraethylammonium cyanide (6.60 g, 42.2 mmol) in dichloromethane (21 ml). The mixture was stirred at room temperature for 1 h, and the reaction was quenched by adding 1N hydrochloric acid until the pH of the mixture reached 2. The mixture was saturated with NaCl and the product was extracted with ethyl acetate. The combined organic layer was dried over Na_2SO_4, concentrated *in vacuo* and the residue was purified by silica gel chromatography (100 g). Elution with hexane-ethyl acetate-ethanol (60 : 40 : 3) followed by recrystallization from hexane-ethyl acetate afforded **4** as colorless fine

needles (1.03 g, 44%); mp 109.0-109.3 °C; IR νmax (KBr) 3370, 2930, 2520, 2290, 1660, 1390, 1315, 1240, 1120, 1015, 960, 910, 870, 810, 740, 630 cm^{-1}; ^{1}H NMR (270 MHz, D_2O) δ = 3.39 (d, J = 5.0 Hz, 2H), 5.81 (d, J = 18.2 Hz, 1H), 6.37 (dt, J = 18.2, 5.0 Hz, 1H); ^{13}C NMR (100 MHz, CD_3OD) δ = 23.3, 118.5, 137.8, 139.5.

5 H. C. Brown and S. K. Gupta, *J. Am. Chem. Soc.*, **97**, 5249 (1975).

6 S. Andreades and E. W. Zahnow, *J. Am. Chem. Soc.*, **91**, 4181 (1969).

7 The harvested wet cells of *Rhodococcus rhodochrous*[14] (15 g) were re-suspended in a phosphate buffer solution (pH 6.0, 1 mM, 160 ml) and **4** (817 mg, 7.37 mmol) was added. The mixture was stirred at 30 °C for 20 h. During the incubation, its pH was kept at 6.0 by a pH controller. After the removal of cell mass by suction filtration through a pad of Celite, the filtrate was concentrated *in vacuo*. The residue was purified by silica gel chromatography (25 g). Elution with ethyl acetate-ethanol (4 : 1) followed by recrystallization from water afforded **3** as colorless prisms (708 mg, 75%); mp 131.5–131.8 °C; IR νmax (KBr) 3420, 3350, 3220, 2930, 1670, 1620, 1355, 1275, 1245, 1190, 1150, 1120, 1000, 940, 800, 770, 620 cm^{-1}; ^{1}H NMR (270 MHz, D_2O) δ = 3.19 (d, J = 6.6 Hz, 2H), 5.66 (d, J = 18.2 Hz, 1H), 6.56 (dt, J = 18.2, 6.6 Hz, 1H); ^{13}C NMR (100 MHz, CD_3OD) δ = 44.3, 144.5, 146.2, 177.1. Found: *m/z* 129.0726 (M^++1). Calcd for $C_4H_8BNO_3$: 129.0711.

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10 M. Karpaty, M. Davidson, M. Hellin, and F. Coussemant, *Bull. Soc. Chim. Fr.*, **1971**, 1731.

11 To a solution of **3** (100 mg, 0.79 mmol) in water (2 ml) was added a solution of (*E*)-**5** (100 mg, 0.57 mmol) in THF (8 ml). After the atmosphere was replaced with argon, tetrakis(triphenylphosphine)-palladium(0) (45 mg, 0.040 mmol) and degassed 0.7 N aqueous sodium hydroxide solution (0.3 ml) were added and the mixture was stirred at 70 °C. While the progress of the reaction was monitored by TLC (ethyl acetate), four portions of sodium hydroxide solution as above were added at an interval of 30 min, until the disappearance of (*E*)-**5** was confirmed. After 3 h, the mixture was saturated with NaCl and extracted with ethyl acetate. The combined organic layer was dried over Na_2SO_4, concentrated *in vacuo* and the residue was purified by silica gel chromatography (10 g). Elution with ethyl acetate followed by recrystallization from ethyl acetate afforded (3*E*,5*E*)-**6** as colorless plates (52 mg, 49%); mp 161.8–162.5 °C; IR νmax (KBr) 3390, 3190, 1650, 1490, 1445, 1415, 1260, 1180, 1070, 990, 920, 795, 745, 690 cm^{-1}; ^{1}H NMR (400 MHz, CD_3OD) δ = 3.07 (d, J = 7.3 Hz, 2H), 5.91 (dt, J = 15.1, 7.3 Hz, 1H), 6.35 (dd, J = 10.5, 15.1 Hz, 1H), 6.53 (d, J = 15.6 Hz, 1H), 6.84 (dd, J = 10.5, 15.6 Hz, 1H), 7.17–7.41 (m, 5H); ^{13}C NMR (100 MHz, CD_3OD) δ = 40.5, 127.4, 128.0, 128.5, 129.6, 133.1, 135.3, 138.7, 176.9. Anal. Found: C, 76.69; H, 7.05; N, 7.78%. Calcd for $C_{12}H_{13}NO$: C, 76.98; H, 7.00; N, 7.48%. In a similar manner, (*Z*)-**5** was coupled with **3** to afford (3*E*,5*Z*)-**6** as colorless plates (70%); mp 133.1-133.4 °C; IR νmax (KBr) 3390, 3200, 1650, 1490, 1445, 1425, 1410, 1395, 1300, 1255, 1135, 990, 955, 930, 830, 805, 780, 710, 670, 640 cm^{-1}; ^{1}H NMR (400 MHz, CD_3OD) δ = 3.04 (d, J = 7.3 Hz, 2H), 5.95 (dt, J = 14.9, 7.3 Hz, 1H), 6.25 (dd, J = 11.2, 11.7 Hz, 1H), 6.41 (d, J = 11.7 Hz, 1H), 6.72 (dd, J = 11.2, 14.9 Hz, 1H), 7.20–7.35 (m, 5H); ^{13}C NMR (100 MHz, CD_3OD) δ = 40.5, 128.0, 129.3, 130.0, 130.4, 130.5, 130.6, 131.0, 138.8, 176.7. Anal. Found: C, 76.81; H, 6.90; N, 7.54%. Calcd for $C_{12}H_{13}NO$: C, 76.98; H, 7.00; N, 7.48%.

12 G. Cassani, P. Massardo, and P. Piccardi, *Tetrahedron Lett.*, **24**, 2513 (1983).

13 Amide (3*E*,5*E*)-**6** (50 mg, 0.27 mmol) was incubated with the harvested wet cells of *Rhodococcus rhodochrous* (1 g) for 6 h. Conventional workup and the treatment of the crude product with diazomethane afforded a methyl ester. This was purified by silica gel chromatography (2 g). Elution with hexane-ethyl acetate (4 : 1) followed by recrystallization from hexane afforded methyl ester of (3*E*,5*E*)-**7** as colorless plates (47 mg, 87%); mp 46.7–47.0 °C; IR νmax (KBr) 3030, 2950, 1730, 1590, 1485, 1460, 1435, 1400, 1370, 1340, 1310, 1290, 1200, 1170, 990, 930, 890, 800, 755, 700 cm^{-1}; ^{1}H NMR (270 MHz, $CDCl_3$) δ = 3.20 (d, J = 7.3 Hz, 2H), 3.71 (s, 3H), 5.89 (dt, J = 15.2, 7.3 Hz, 1H), 6.31 (dd, J = 10.2, 15.2 Hz, 1H), 6.51 (d, J = 15.8 Hz, 1H), 6.79 (dd, J = 10.2, 15.8 Hz, 1H), 7.20–7.41 (m, 5H); ^{13}C NMR (100 MHz, $CDCl_3$) δ = 38.0, 52.0, 125.5, 126.3, 127.5, 128.3, 128.6, 132.2, 134.0, 137.2, 172.0. Anal. Found: C, 77.06; H, 6.97%. Calcd for $C_{13}H_{14}O_2$: C, 77.20; H, 6.98%. In a similar manner, methyl ester of (3*E*,5*Z*)-**7** was obtained as colorless oil (79%); IR νmax (film) 3030, 2960, 1740, 1640, 1600, 1570, 1490, 1435, 1415, 1340, 1250, 1200, 1160, 990, 955, 920, 890, 835, 775, 700 cm^{-1}; ^{1}H NMR (270 MHz, $CDCl_3$) δ = 3.17 (d, J = 7.3 Hz, 2H), 3.70 (s, 3H), 5.94 (dt, J = 15.2, 7.3 Hz, 1H), 6.25 (dd, J = 10.9, 11.6 Hz, 1H), 6.43 (d, J = 11.6 Hz, 1H), 6.67 (dd, J = 10.9, 15.2 Hz, 1H), 7.21–7.38 (m, 5H); ^{13}C NMR (100 MHz, $CDCl_3$) δ = 38.1, 51.9, 127.0, 127.7, 128.3, 128.9, 129.4, 129.8, 130.0, 137.4, 171.9. Anal. Found: C, 77.02; H, 7.18%. Calcd for $C_{13}H_{14}O_2$: C, 77.20; H, 6.98%.

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