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Synthesis of optically active dihydrocarveol via a stepwise or one-pot enzymatic reduction of (*R*)- and (*S*)-carvone

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ABSTRACT

A recombinant enoate reductase **LacER** from *Lactobacillus casei* catalyzed the reduction of (*R*)-carvone and (*S*)-carvone to give (2*R*,5*R*)-dihydrocarvone and (2*R*,5*S*)-dihydrocarvone with 99% and 86% de, respectively, which were further reduced to dihydrocarveols by a carbonyl reductase from *Sporobolomyces salmonicolor* **SSCR** or *Candida magnolia* **CMCR**. For (*R*)-carvone, (1*S*,2*R*,5*R*)-dihydrocarveol was produced as the sole product with >99% conversion, while (1*S*,2*R*,5*S*)-dihydrocarveol was obtained as the major product, but with a lower de when (*S*)-carvone was used as the substrate. The one-pot reduction was performed at a 0.1 M substrate concentration, indicating that it might provide an effective synthetic route to this type of chiral compound.

1. Introduction

Carvone and related compounds are important components in the flavor and fragrance industry.[1] Dihydrocarvones are potential inhibitors of bacterial and fungal growth, as well as prospective insect repellents.[2] For example, they have shown insecticidal activity against the rice weevil *Sitophilus oryzae* (L.), one of the most widespread insect pests of stored cereals.[2,3] Recently, dihydrocarvones have also been used as important renewable building blocks in the synthesis of functional materials such as shape memory polyesters.[4] Dihydrocarveol is a fragrance ingredient used in decorative cosmetics, fine fragrances, shampoos, soaps and other toiletries as well as in household products such as cleaners and detergents.[5] Carvone is currently extracted from caraway, dill, and spearmint seeds.[1] While transformations of carvone into dihydrocarvones and dihydrocarveol have been widely studied, the chemo-, regio- and stereospecificity of these transformations still present a challenging task. (*R*)- or (*S*)-Carvone (Fig. 1), which possesses three unsaturated bonds and a C-5 stereogenic center, possesses many selectivity problems: (1) chemoselectivity between the C=C and C=O functional groups; (2) regioselectivity between the conjugated and isolated C=C bond; and (3) *cis* versus *trans* diastereoselectivity with respect to the C-5 substituent in the C=O and C=C bond reductions. With no control over selectivity, 17 different hydrogenation products are possible.[6] Bogel-Łukasik reported that at least 10 products were generated in the hydrogenation of carvone catalyzed by Pd, Rh, or Ru supported on alumina as the catalyst in supercritical carbon dioxide.[7]

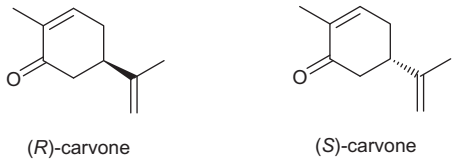

Figure 1.

As a result, biocatalytic transformation of carvone has recently been the focus of several papers.[8–17] The bioreduction of (*R*)-carvone and (*S*)-carvone using whole cells of bacteria,[13] fungi,[12] yeast,[17] plant cell cultures,[15] and marine microalgae[14] has already been reported. The microbial reduction of these compounds usually gives a mixture of saturated ketone, saturated alcohol, and occasionally the allylic alcohol, indicating that both enoate reductase and carbonyl reductase may catalyze the reduction of C=C and C=O double bonds competitively. The use of isolated enzymes offers several advantages including the elimination of undesirable side product formation mediated by contaminating enzymes in the whole cell system, the possibility of achieving a high substrate load, easy downstream product separation, and easy handling for organic chemists without microbiological knowledge. In recent years, isolated reductase enzymes have been demonstrated to be highly effective catalysts for the enantioselective reduction of a wide range of substrates.[18]

In this context, isolated enoate and carbonyl reductases were used in an attempt to address the chemo-, regio- and stereoselectivity in the transformations of carvone into dihydrocarvone, and then into dihydrocarveol.[9,10] Herein we report the

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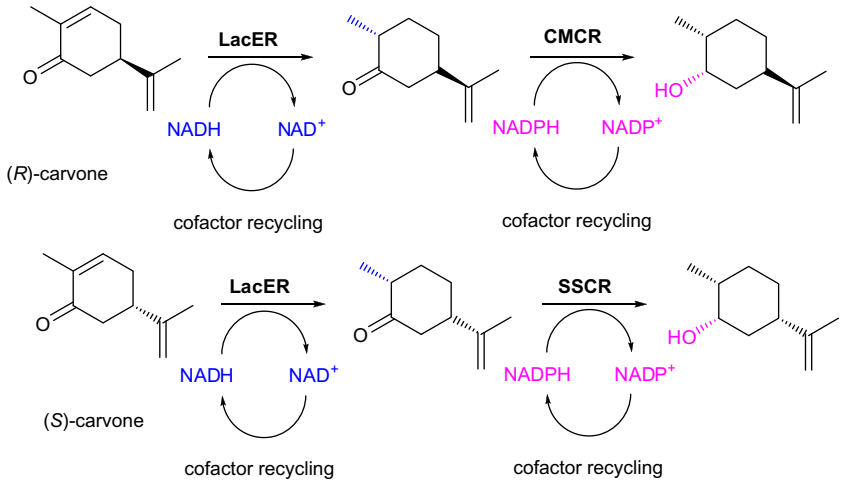

Scheme 1. The tandem reduction of carvone by enoate reductase and carbonyl reductase.

two-step enzymatic reduction of (*R*)- and (*S*)-carvone to dihydrocarveol (Scheme 1). For the first hydrogenation step, carbon–carbon double bond reduction, enoate reductase **LacER** from *Lactobacillus casei* was used; for the second step, the carbonyl reduction, a reductase from *Sporobolomyces salmonicolor* **SSCR** or *Candida magnolia* **CMCR** was employed. The reaction was evaluated by adding the two enzymes sequentially or simultaneously, with the aim of developing a more effective process for the conversion of carvones into enantiomerically pure dihydrocarveols.

2. Results and discussion

2.1. Enzymatic reduction of (*R*)- and (*S*)-carvone to dihydrocarvone

We have recently reported that **LacER** catalyzed the reduction of (*R*)-carvone to give (2*R*,5*R*)-dihydrocarvone with 99% diastereoselective excess (de), which is one of the best results obtained so far.[19] When (*S*)-carvone was used as the substrate for this enzyme, the configuration of newly formed C-2 stereomeric center of dihydrocarvone was also (*R*), with the de value being 86%. The results showed that the C-5 configuration did not have much impact on the stereochemistry of the hydride transfer in the **LacER**-catalyzed reduction of carvone. This is consistent with the observations for other enoate reductases such as pentaerythritol tetranitrate reductase **PETNR** from the anaerobic microorganism *Enterobacter cloacae st.* PB2,[9] a thermophilic 'ene' reductase (*TOYE*) isolated from *Thermoanaerobacter pseudethanolicus* E39[11] and *Saccharomyces pastorianus* old yellow enzyme.[10] Stewart et al. reported that (2*S*,5*S*)-dihydrocarvone was obtained in 88% de from the reduction of (*S*)-carvone catalyzed by a mutant enzyme (W116I) of *Saccharomyces pastorianus* **OYE**.[10]

2.2. Stepwise enzymatic reduction of (*R*)- and (*S*)-carvone to dihydrocarveol

(*R*)-Carvone was first reduced to (2*R*,5*R*)-dihydrocarvone by using **LacER** as the catalyst. To the resulting reaction mixture, D-glucose dehydrogenase, $NADP^+$, and the carbonyl reductase were added to reduce the carbonyl group in dihydrocarvone, and the results are presented in Table 1. As shown in Table 1, **CMCR**, **SSCR**, and its mutant enzymes catalyzed the reduction of (2*R*,5*R*)-dihydrocarvone to give (1*S*,2*R*,5*R*)-dihydrocarveol with high stereoselectivity. In the cases of **CMCR** and **SSCR mutant M242CQ245L**, (1*S*,2*R*,5*R*)-dihydrocarveol was obtained with 99% conversion and 99% de. The

Table 1
Stepwise enzymatic reduction of (*R*)-carvone to dihydrocarveol

Entry	Enzyme[a]	Conversion[b] (%)	de[c] (%)
1	CMCR	99	>99
2	SSCR	73	98
3	Q245P	42	93
4	Q245L	81	99
5	Q245H	73	66
6	M242Y	73	99
7	M242C	68	99
8	M242D	80	99
9	M242Q	88	99
10	M242CQ245L	99	99
11	M242LQ245T	93	97
12	M242FQ245T	99	98
13	M242LQ245P	94	96
14	Ymr226c	—	—
15	Gre2	—	—

[a] See Section 4 for the source of these enzymes.
[b] The conversion was measured by GC analysis.
[c] The de values were measured by chiral GC analysis.

carbonyl reductases **Ymr226c** and **Gre2** from Baker's yeast showed no detectable activity toward (2*R*,5*R*)-dihydrocarvone.

In the case of (*S*)-carvone as the substrate, it was reduced to give both (2*S*,5*S*)-dihydrocarvone (7%) and (2*R*,5*S*)-dihydrocarvone (93%). After the reaction was completed, the resulting mixture was treated with the carbonyl reductase and a **NADPH** regeneration system. The conversion for the carbonyl reduction and the de value for the reduction of (2*R*,5*S*)-dihydrocarvone (major product) and (2*S*,5*S*)-dihydrocarvone (minor product) are summarized in Table 2. From Table 2, it can be seen that **SSCR** and its mutant enzymes showed higher activity than **CMCR**, **Ymr226c**, and **Gre2**. In all cases, the stereoselectivity for the reduction of (2*R*,5*S*)-dihydrocarvone was higher than that for (2*S*,5*S*)-dihydrocarvone. For example, when **CMCR** was used as the catalyst, the product (1*S*,2*R*,5*S*)-dihydrocarveol was obtained in 99% de for the reduction of (2*R*,5*S*)-dihydrocarvone, but was obtained in only 48% de for the reduction of (2*S*,5*S*)-dihydrocarvone. **SSCR** and some mutant enzymes also catalyzed the reduction of (2*R*,5*S*)-dihydrocarvone to give (1*S*,2*R*,5*S*)-dihydrocarveol in 99% de.

2.3. One-pot enzymatic reduction of (*R*)- and (*S*)-carvone to dihydrocarveol

The C=C bond reduction of (*R*)-carvone and (*S*)-carvone by **LacER** and the following C=O reduction by carbonyl reductases were

Table 2
Stepwise enzymatic reduction of (*S*)-carvone to dihydrocarveol

Entry	Enzyme	Conversion[a] (%)	de[b] (%)	de[c] (%)
1	CMCR	68	99	48
2	SSCR	99	99	75
3	Q245P	80	87	68
4	Q245L	96	79	4
5	Q245H	83	82	65
6	M242Y	97	99	50
7	M242C	94	99	46
8	M242D	99	97	20
9	M242Q	97	98	28
10	M242CQ245L	99	94	24
11	M242LQ245T	99	78	10
12	M242FQ245T	85	91	41
13	M242LQ245P	99	89	21
14	Ymr226c	7	76	—[d]
15	Gre2	26	18	—[d]

[a] The conversion for the carbonyl reduction was measured by GC analysis.
[b] The de values for the reduction of (2*R*,5*S*)-dihydrocarvone catalyzed by carbonyl reductase.
[c] The de values for the reduction of (2*S*,5*S*)-dihydrocarvone catalyzed by carbonyl reductase.
[d] The conversion was so low that the de value was not determined.

Table 4
One-pot enzymatic reduction of (*S*)-carvone to dihydrocarveol

Entry	Enzyme	Conversion[a] (%)	de[b] (%)	de[c] (%)
1	CMCR	36	99	32
2	SSCR	87	99	76
3	Q245P	43	86	84
4	Q245L	56	57	32
5	Q245H	53	73	83
6	M242Y	29	92	74
7	M242C	58	55	75
8	M242D	34	85	75
9	M242Q	66	91	77
10	M242CQ245L	76	87	87
11	M242LQ245T	86	72	61
12	M242FQ245T	27	66	79
13	M242LQ245P	72	86	61
14	Ymr226c	6	75	—
15	Gre2	17	22	—

[a] The conversion was for the carbonyl reduction of dihydrocarvone to dihydrocarveol and measured by GC analysis. No (*S*)-carvone was detected after reaction.
[b] The de values were for the reduction of (2*R*,5*S*)-dihydrocarvone by carbonyl reductase, and measured by chiral GC analysis.
[c] The de values were for the reduction of (2*S*,5*S*)-dihydrocarvone by carbonyl reductase, and measured by chiral GC analysis.

also performed in a mode in which **LacER**, carbonyl reductase, and the cofactor regeneration system were added into the reaction mixture at the same time. **LacER** required **NADH** for the (*R*)- or (*S*)-carvone reduction, while carbonyl reductase needed **NADPH** for the dihydrocarvone reduction; hence **GDH** and glucose were used for the regeneration of both **NADH** and **NADPH**. Since the optimal pH for LacER was 8.0 and the pH was not adjusted after the C=C bond reduction in the stepwise procedure described above, while the following C=O reduction proceeded smoothly, the one-pot reaction was thus performed at pH 8.0. The results are presented in Tables 3 and 4. For both (*R*)-carvone and (*S*)-carvone, the C=C bond reduction proceeded faster than the carbonyl reduction, as evidenced by the fact that both (*R*)-carvone and (*S*)-carvone were converted into the dihydrocarvones in about 4 h. The stereoselectivity for the C=C bond reduction was the same as that in the stepwise reaction, and was not affected by the reaction mode. The conversions in Tables 3 and 4 are for the carbonyl reduction of dihydrocarvone to dihydrocarveol.

As shown in Table 3, (*R*)-carvone was reduced to give (1*S*,2*R*,5*R*)-dihydrocarveol and (1*R*,2*R*,5*R*)-dihydrocarveol with the former being the major stereoisomer with de values of 52–99%.

Table 3
One-pot enzymatic reduction of (*R*)-carvone to dihydrocarveol

Entry	Enzyme	Conversion[a] (%)	de[b] (%)
1	CMCR	>99	>99
2	SSCR	83	94
3	Q245P	67	88
4	Q245L	83	93
5	Q245H	88	67
6	M242Y	90	75
7	M242C	80	74
8	M242D	87	69
9	M242Q	99	52
10	M242CQ245L	89	88
11	M242LQ245T	96	75
12	M242FQ245T	98	66
13	M242LQ245P	99	82
14	Ymr226c	—	—
15	Gre2	—	—

[a] The conversion was for the carbonyl reduction of dihydrocarvone to dihydrocarveol and measured by GC analysis. No (*R*)-carvone was detected after reaction.
[b] The de values were measured by chiral GC analysis.

When **LacER** was combined with **CMCR** as the catalysts, (*R*)-carvone was exclusively converted into (1*S*,2*R*,5*R*)-dihydrocarveol. For **SSCR** and its mutants, the product dihydrocarveol was obtained with a lower de than those in stepwise reactions, implying that the reaction mode had some effects on the stereoselectivity of the carbonyl reduction of dihydrocarvone to dihydrocarveol catalyzed by **SSCR** and its mutants. The carbonyl reductases **Ymr226c**, and **Gre2** showed no activity toward (*2R*,*5R*)-dihydrocarvone in the one-pot reaction mode (Table 3, entries 14 and 15).

When (*S*)-carvone was used as the substrate, it was first reduced to (2*R*,5*S*)-dihydrocarvone and (2*S*,5*S*)-dihydrocarvone with a ratio of about 93:7. The results in Table 4 show that the conversions were generally lower than those in the stepwise reaction (Table 2), while the stereoselectivity for the carbonyl reduction was also influenced by the reaction mode for some carbonyl reductases. For example, with **CMCR** as the reductase, the product dihydrocarveol was obtained in only 36% conversion while the de for the reduction of (2*S*,5*S*)-dihydrocarvone decreased from 48% to 32%. In the case of **SSCR**, while the product dihydrocarveol was obtained with 87% conversion compared to the 99% conversion in the stepwise reaction, the stereoselectivity was not affected. The (*S*)-carvone could not be completely transformed into dihydrocarveol via a one-pot reaction even with larger amount of the glucose, **GDH**, **SSCR,** or **CMCR**.

2.4. Synthesis of (1*S*,2*R*,5*R*)-dihydrocarveol

The concomitant reduction of the C=O group was observed in the reduction of (*R*)- and (*S*)-carvone by whole cells of *Nicotiana tabacum*, Baker's yeast, and other strains, but the conversions and stereoselectivity were low.[8,16,17,20] As described above, the one-pot reduction of (*R*)-carvone by combining the enoate reductase **LacER** and the carbonyl reductase **CMCR** generated exclusively (1*S*,2*R*,5*R*)-dihydrocarveol with >99% conversion. This reaction should be very useful for the synthesis of (1*S*,2*R*,5*R*)-dihydrocarveol from the readily available (*R*)-carvone. The reaction was thus carried out on a 30 mL scale with the substrate concentration being 0.02 M. (*R*)-Carvone was thus completely transformed into the product. The substrate concentration was then increased to 0.1 M, the substrate was converted completely, while (1*S*,2*R*,5*R*)-dihydrocarveol was isolated in 93% yield and with >99% de.

The enzymatic asymmetric reduction of (*R*)- or (*S*)-carvone to dihydrocarvone has been previously reported, and many reports have described the enzymatic asymmetric reduction of ketones to the corresponding enantiomerically pure alcohols. To the best of our knowledge, only one report is available with regard to the combination of the asymmetric hydrogenation of a carbon–carbon double bond and a carbon–oxygen double bond using isolated enzymes.[21] Wada et al. reported that (4*R*,6*R*)-4-hydroxy-2,2,6-trimethylcyclohexanone was produced at a concentration of 9.5 mg/ml by a two-step enzymatic reduction of ketoisophorone with 94% enantiomeric excess in the presence of glucose, NAD^+, and glucose dehydrogenase. In that case, the enzymes must be added stepwise because ketoisophorone could be reduced by levodione reductase; the resulting 4-hydroxy-2,6,6-trimethyl-2-cyclohexenone would not be converted into actinol if the levodione reductase was added with the old yellow enzyme at the same time.[21] Herein, the enoate and carbonyl reductases could be added at the same time to allow the conversion of (*R*)- or (*S*)-carvone into dihydrocarveols. (*R*)-Carvone was completely converted into (1*S*,2*R*,5*R*)-dihydrocarveol at a substrate concentration of 0.1 M in 13 h. Under the same conditions, if the enoate and carbonyl reductases were added in two steps, the reaction was finished after 24 h. This suggests that the 'one-pot' reduction is more effective than the stepwise procedure.

3. Conclusion

(*R*)- and (*S*)-Carvone were reduced by **LacER** to (2*R*,5*R*)-dihydrocarvone and (2*R*,5*S*)-dihydrocarvone with de values of 99% and 86%, respectively. The resulting dihydrocarvones were reduced stereoselectively to give dihydrocarveols by carbonyl reductases, **CMCR**, **SSCR**, or its mutant enzymes. In particular, (1*S*,2*R*,5*R*)-dihydrocarveol was produced in high yield by combining the C=C and C=O reductions in one-pot reactions, in which **LacER** and **CMCR** were added at the same time or stepwise. The one-pot reduction was carried out at a 0.1 M substrate concentration, thus demonstrating its applicability in the synthesis of (*1S,2R,5R*)-dihydrocarveol from renewable (*R*)-carvone.

4. Experimental

4.1. General

LacER, **CMCR**, **SSCR**, and its mutants, **Gre2**, **Ymr226c**, and **GDH** were prepared as previously reported.[19,22–25] NAD^+, NADH, $NADP^+$, and NADPH were obtained from Codexis (USA). (*R*)-Carvone, (*S*)-carvone, (+)-(2*R*,5*R*)-dihydrocarvone, and (1*R*,2*R*,5*R*)-dihydrocarveol were obtained from Sigma–Aldrich Chemical Co. The GC analyses were performed on an Agilent 7890 gas chromatography with CD-Chiral-DEX CB (30 m × 0.25 mm × 0.25 μm) column, detector temperature 220 °C, split ratio 20:1. Programme: 70 °C, hold for 2 min, 5 °C/min to 120 °C, hold for 12 min.

4.2. Reduction of (*S*)-carvone to dihydrocarvone catalyzed by LacER

The general procedure was as follows: D-glucose (12 mg), D-glucose dehydrogenase (2 mg), NAD^+ (1 mg), the enoate reductase (LacER, 2 mg), and (*S*)-carvone solution in DMSO (100 μL, 0.20 M) were mixed in a potassium phosphate buffer (0.9 mL, 100 mM, pH 8.0) and the mixture was shaken at 37 °C for 4 h. The mixture was extracted with methyl *tert*-butyl ether (1 mL). The organic extract was dried over anhydrous sodium sulfate and then subjected to chiral GC analysis to determine the conversion and diastereomeric excess. Retention times: (2*R*,5*S*)-dihydrocarvone, 13.5 min; (2*S*,5*S*)-dihydrocarvone, 13.3 min. The absolute configurations of the product alcohols were identified by comparing the chiral GC data with the standard samples and the ^{1}H NMR data with those in the literature.[20] (2*R*,5*S*)-Dihydrocarvone ^{1}H NMR ($CDCl_3$, 400 MHz) δ (ppm) 4.82 (s, 1H), 4.68 (s, 1H), 2.59–2.52 (m, 2H), 2.41–2.37 (m, 2H), 1.86–1.82 (m, 3H), 1.72 (s, 3H), 1.62–1.58 (m, 1H), 1.08 (d, J = 4.8 Hz, 3H).

4.3. Stepwise enzymatic reduction of (*R*)- and (*S*)-carvone to dihydrocarveol

The general experimental procedure was as follows: a (*R*)-carvone or (*S*)-carvone solution in DMSO (100 μL, 0.20 M) was mixed in a potassium phosphate buffer (0.9 mL, 100 mM, pH 8.0). Next, D-glucose (24 mg), D-glucose dehydrogenase (2 mg), NAD^+ (1 mg), and the enoate reductase (LacER, 2 mg) were added. After the mixture was shaken at 37 °C for 4 h, D-glucose dehydrogenase (2 mg), $NADP^+$ (1 mg), and the carbonyl reductase (2 mg) were added. The mixture was shaken at 37 °C for another 24 h. The mixture was extracted with methyl *tert*-butyl ether (1 mL). The organic extract was dried over anhydrous sodium sulfate and then subjected to chiral GC analysis to determine the diastereomeric excess. (2*R*,5*R*)-dihydrocarvone and (2*R*,5*S*)-dihydrocarvone were reduced by $NaBH_4$ to give two sets of diastereomer mixtures, which served as the standard samples for GC analysis. Their retention times were as follows: (1*S*,2*R*,5*R*)-dihydrocarveol, 16.6 min; (1*R*,2*R*,5*R*)-dihydrocarveol, 17.2 min; (1*S*,2*R*,5*S*)-dihydrocarveol, 20.0 min; (1*R*,2*R*,5*S*)-dihydrocarveol, 19.3 min. The absolute configurations of the product alcohols were identified by comparing the chiral GC data with the standard samples and the ^{1}H NMR spectra with those in the literature.[20] (1*S*,2*R*,5*R*)-Dihydrocarveol ^{1}H NMR ($CDCl_3$, 400 MHz) δ (ppm) 4.69 (s, 2H), 3.88 (s,1H), 2.29 (t, J = 8 Hz, 1H), 1.93–1.90 (dd, J = 1.6 Hz, 1H), 1.77–1.75 (m,1H), 1.71 (s, 3H), 1.50–1.40 (m, 5H), 1.84 (m,1H), 0.97 (d, J = 4.8 Hz, 3H). ^{13}C NMR ($CDCl_3$, 100 MHz) δ (ppm) 150.26, 108.38, 70.99, 38.69, 37.82, 36.08, 31.41, 28.14, 20.96, 18.29. (1*S*,2*R*,5*S*)-Dihydrocarveol ^{1}H NMR ($CDCl_3$, 400 MHz) δ (ppm) 4.69 (d, J = 2.8 Hz, 2H), 3.78 (m, 1H), 2.07 (m, 1H), 1.96(m, 1H), 1.71(s, 3H), 1.70–1.62 (m, 2H), 1.55–1.49 (m, 1H), 1.44–1.38 (m, 2H), 1.34–1.27 (m, 1H), 0.93–0.92 (d, J = 4.8 Hz, 3H). ^{13}C NMR ($CDCl_3$, 100 MHz) δ (ppm) 149.48, 108.55, 72.70, 44.02, 34.05, 33.68, 30.58, 24.59, 20.88, 10.69.

4.4. One-pot enzymatic reduction of (*R*)- and (*S*)-carvone to dihydrocarveol

The NADH and NADPH regeneration system containing NAD^+, $NADP^+$, GDH, and glucose was used for the reduction of carvone and dihydrocarvone. An (*R*)-carvone or (*S*)-carvone solution in DMSO (100 μL, 0.20 M) was mixed in a potassium phosphate buffer (0.9 mL, 100 mM, pH 8.0), after which D-glucose (24 mg), D-glucose dehydrogenase (4 mg), $NADP^+$ (1 mg), NAD^+ (1 mg), the enoate reductase (LacER, 2 mg), and the carbonyl reductase (CMCR, 2 mg) were added. The mixture was then shaken at 37 °C for 24 h. The mixture was extracted with methyl *tert*-butyl ether (1 mL). The organic extract was dried over anhydrous sodium sulfate and then subjected to chiral GC analysis to determine the diastereomeric excess. The absolute configurations of the product alcohols were identified as described above.

4.5. Synthesis of (1*S*,2*R*,5*R*)-dihydrocarveol

At first, D-glucose (110 g L^{-1}), D-glucose dehydrogenase (2 g L^{-1}), NAD^+ (1 g L^{-1}), $NADP^+$ (1 g L^{-1}), enoate reductase enzyme (2 g L^{-1}), and carbonyl reductase (2 g L^{-1}) were dissolved in potassium phosphate buffer (100 mM, pH 8.0, 27 mL). The resulting solution was mixed with 3 mL of (*R*)-carvone solution in DMSO (1.0 M).

The reaction mixture was shaken at room temperature for 24 h. The reduction was completed as shown by GC analysis. The mixture was extracted with methyl *tert*-butyl ether (3 × 30 mL). The organic extract was dried over anhydrous sodium sulfate and removal of the solvent gave a yellow oil (429.5 mg, 93% yield), which was identified as (1*S*,2*R*,5*R*)-dihydrocarveol with >99% de.

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