HOKKAIDO UNIVERSITY

Title	Isolation and amino acid sequence of a dehydratase acting on d-erythro-3-hydroxyaspartate from Pseudomonas sp N99, and its application in the production of optically active 3-hydroxyaspartate
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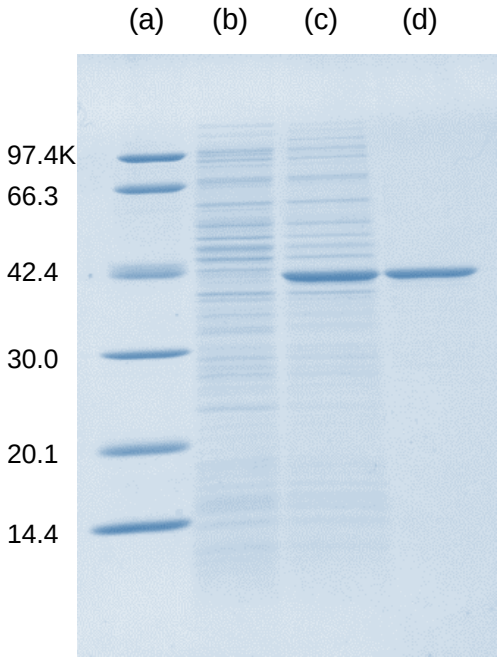


Fig. S1. SDS-PAGE analysis of the recombinant enzyme expressed in *E. coli* cells. Lane (a), molecular weight markers; Lane (b), cell extract of *E. coli* BL21 (DE3)/pET15b (empty vector); Lane (c), cell extract of *E. coli* BL21 (DE3)/pET15_*dehadh*; Lane (d), purified recombinant D-EHA DH.

Fig. S1. Nagano et al.

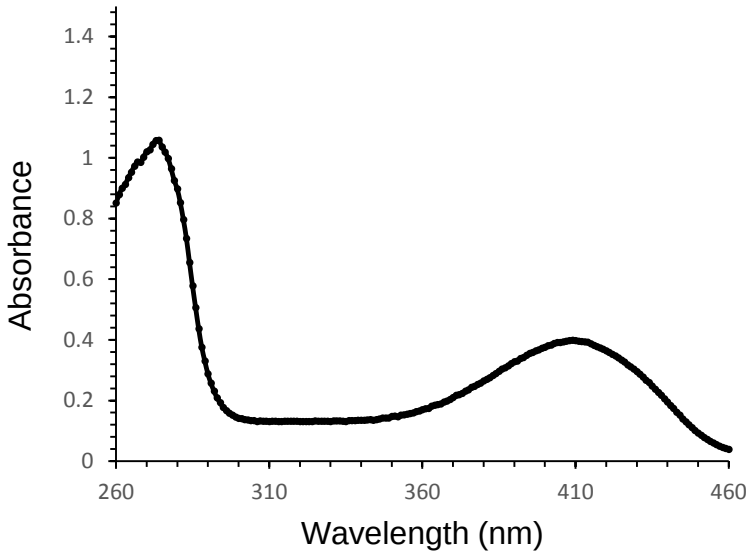


Fig. S2 Absorption spectrum of the recombinant enzyme. Absorption spectrum was obtained with a Beckman DU-800 spectrophotometer (Beckman Coulter, Inc., Brea, CA, USA). The recombinant enzyme (1.4 mg/ml) was dissolved in 10 mM Tris-HCl buffer (pH 8.5) containing 0.01mM PLP, 0.1mM $MnCl_2$ and 0.1mM dithiothreitol.

Fig. S2. Nagano et al.

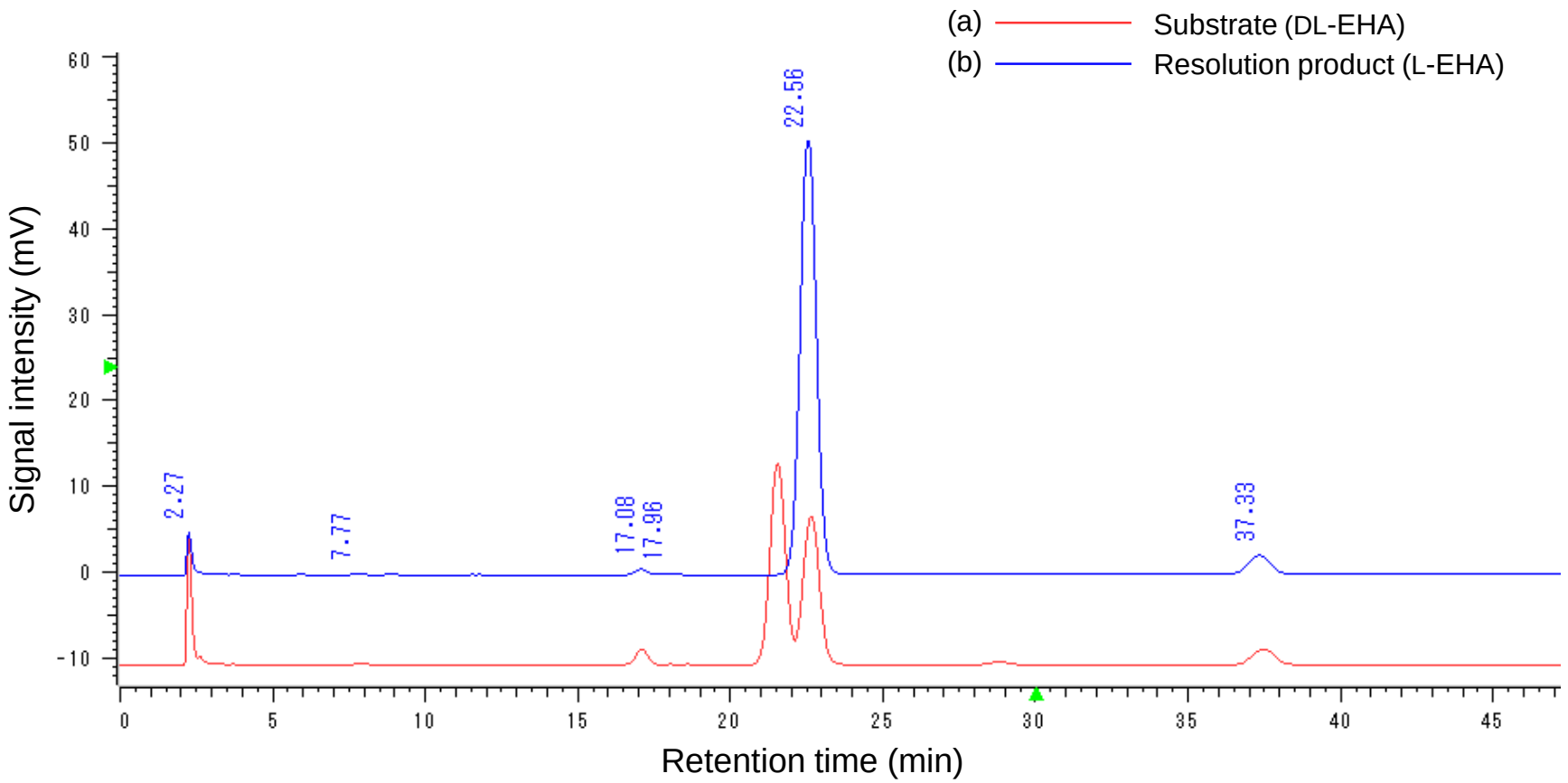


Fig. S3. Overlay chiral HPLC chromatograms of (a) DL-EHA synthesized by the protocol described in ref. 12, and (b) L-EHA obtained by enzymatic optical resolution.

Fig. S3. Nagano et al.

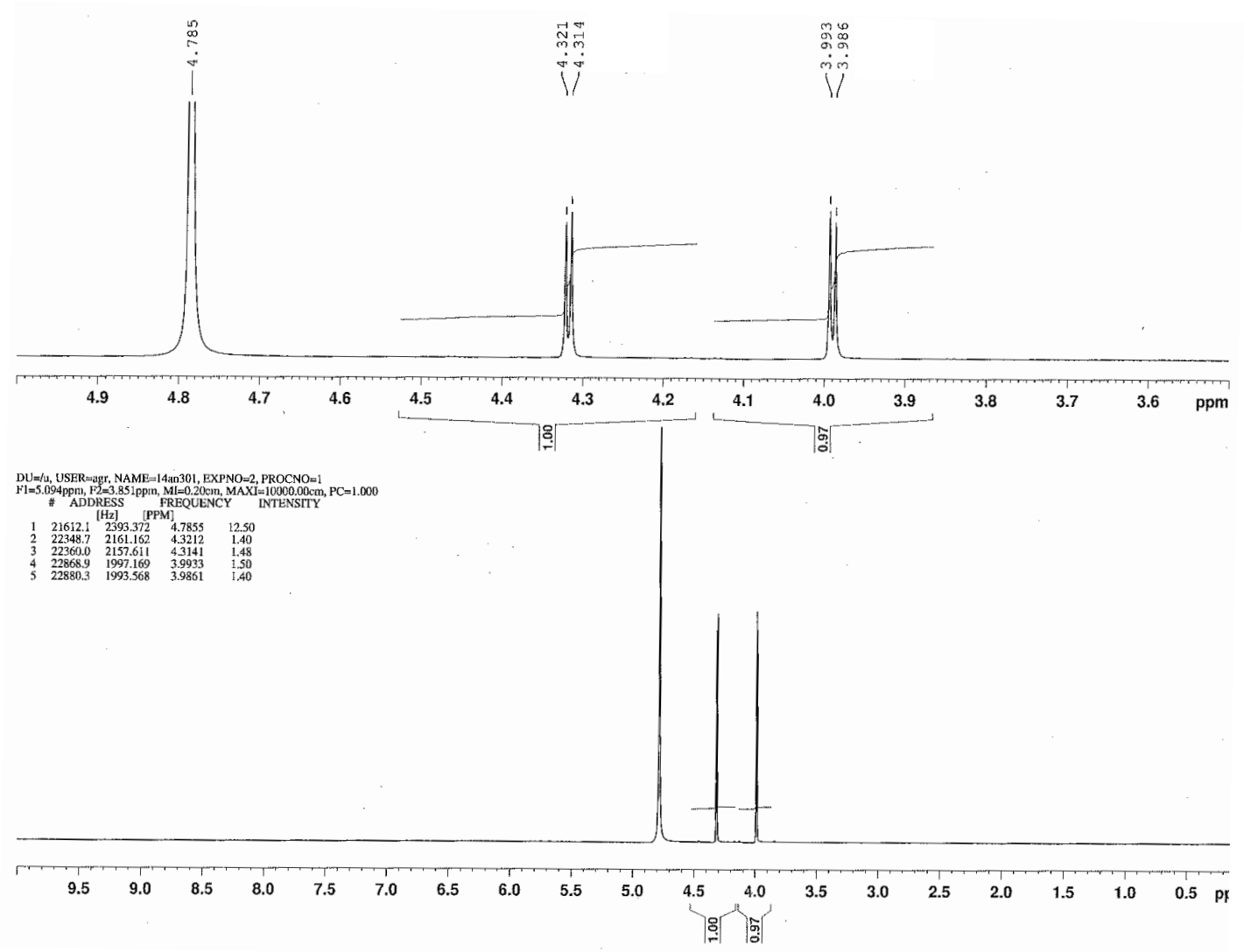


Fig. S4. ^{1}H-NMR spectra of L-EHA obtained by enzymatic optical resolution from DL-EHA recorded at 500 MHz. The ^{1}H signal at 4.79 ppm is attributed to deuterium oxide (D_2O).

Fig. S4. Nagano et al.

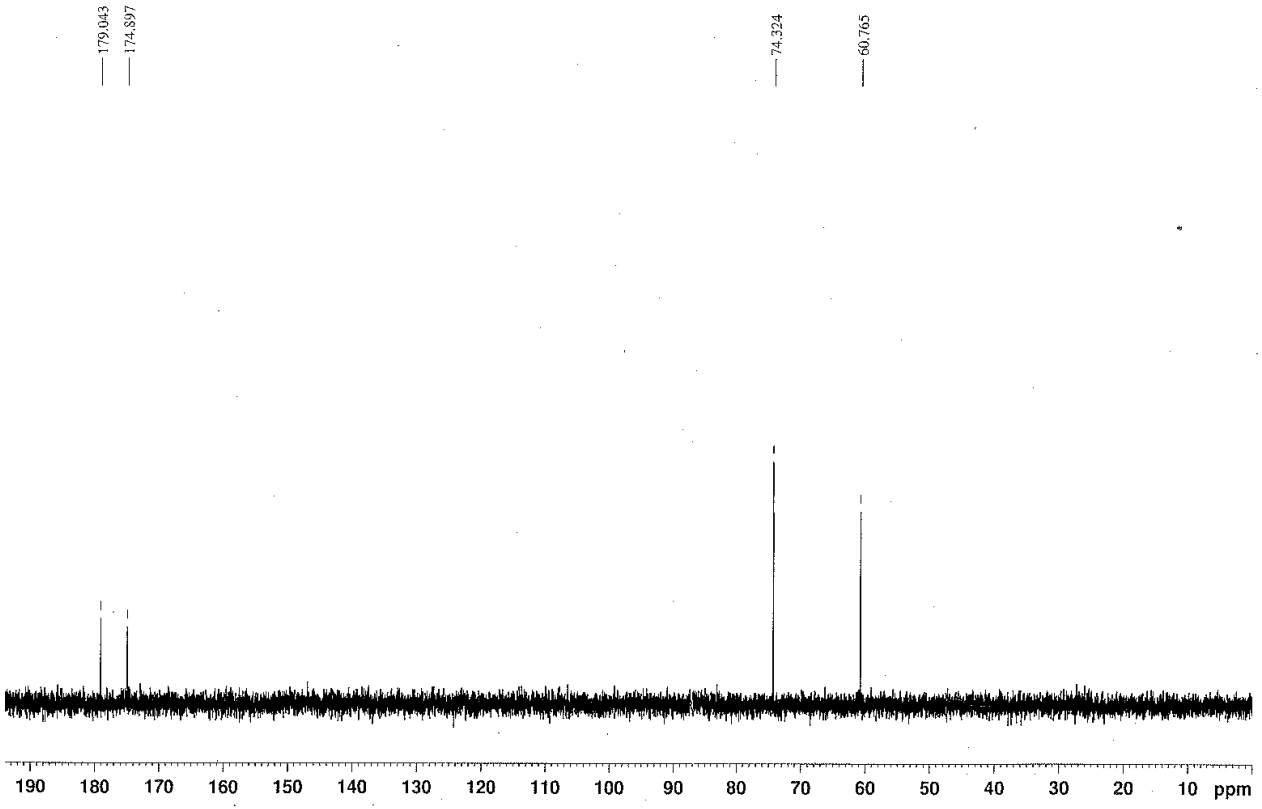


Fig. S5. ^{13}C-NMR spectra of L-EHA obtained by enzymatic optical resolution from DL-EHA recorded at 125 MHz.

Fig. S5. Nagano et al.

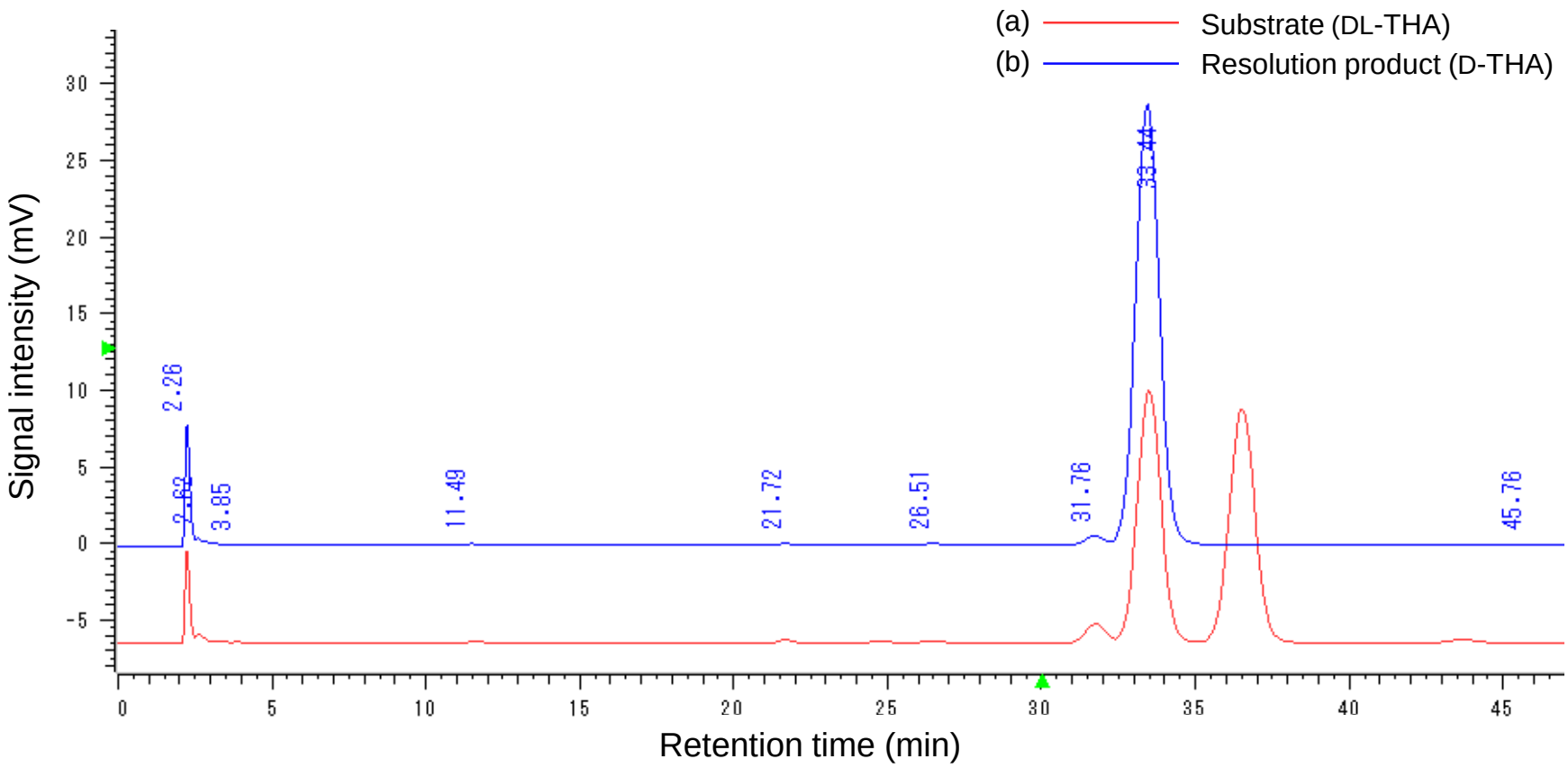


Fig. S6. Overlay chiral HPLC chromatograms of (a) DL-THA purchased from Tokyo Chemical Industry, and (b) D-THA obtained by enzymatic optical resolution.

Fig. S6. Nagano et al.

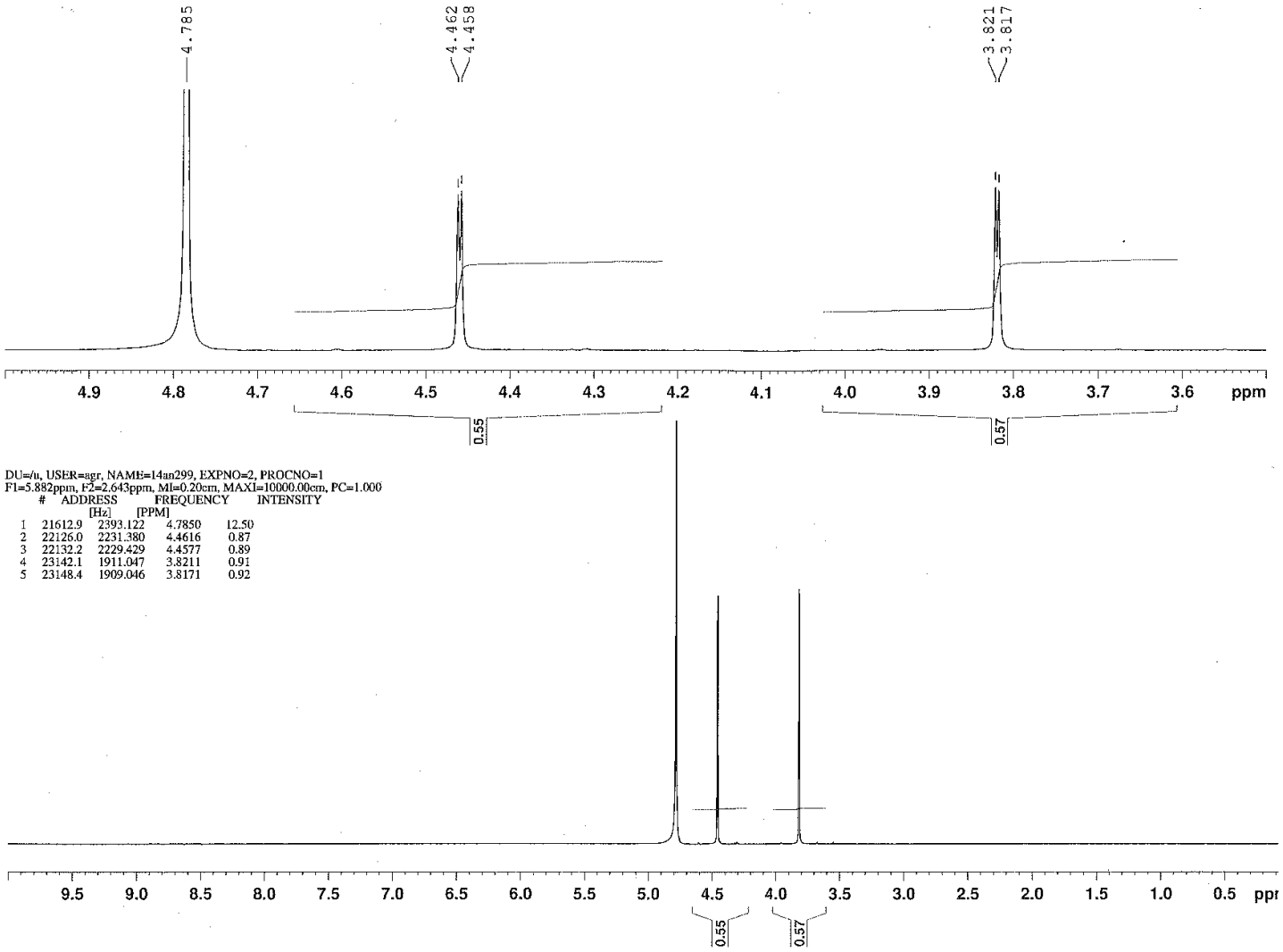


Fig. S7. ^{1}H-NMR spectra of D-THA obtained by enzymatic optical resolution from DL-THA recorded at 500 MHz. The ^{1}H signal at 4.79 ppm is attributed to deuterium oxide (D_2O).

Fig. S7. Nagano et al.

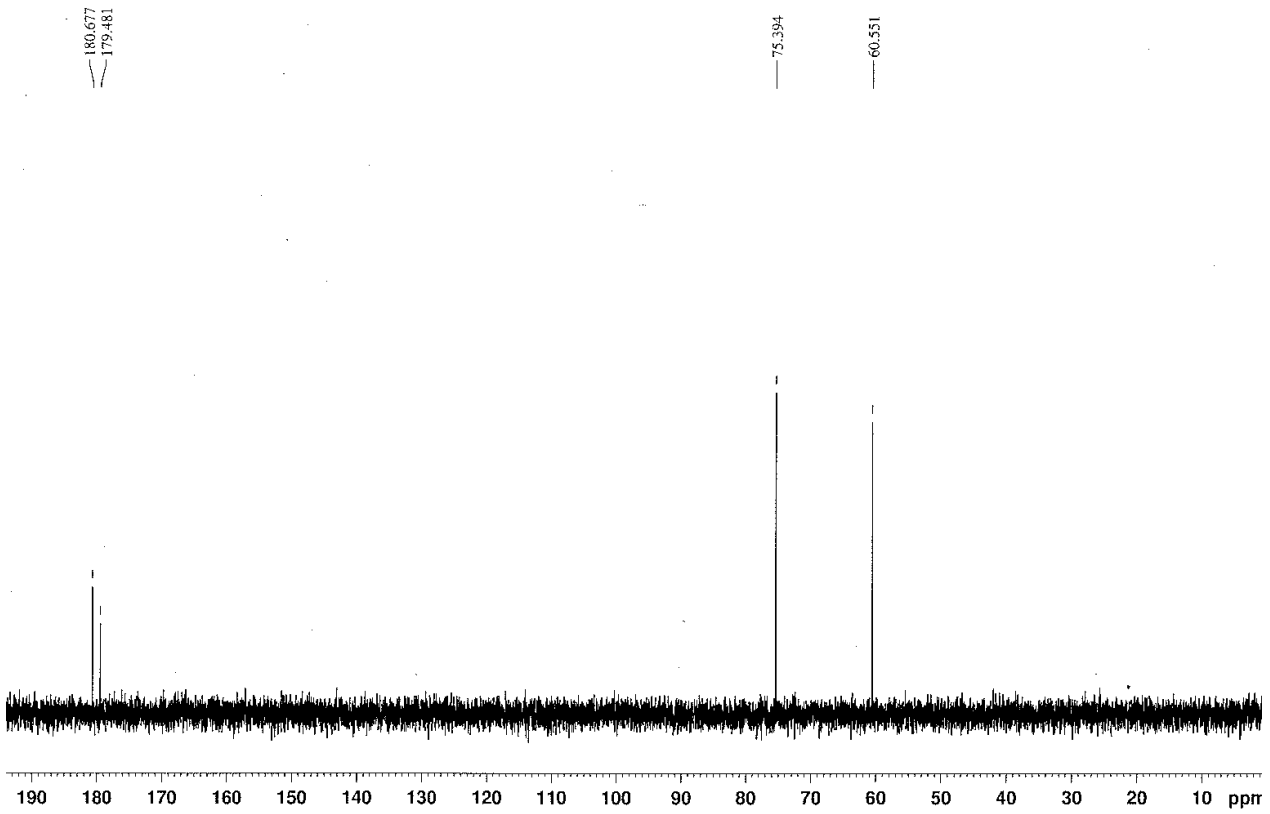


Fig. S8. ^{13}C-NMR spectra of D-THA obtained by enzymatic optical resolution from DL-THA recorded at 125 MHz.

Fig. S8. Nagano et al.