

Unusual stereoselectivity of methionine- γ -lyase from *Citrobacter freundii* toward diastereomeric (*S*)-methionine *S*-oxide

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¹H NMR spectra of the obtained compounds were recorded on a Bruker Avance 400 instrument (operating frequency 400 MHz), Agilent 400-MR (operating frequency 400 MHz) in CDCl₃, and Bruker Avance™ 600 for ¹³C NMR (150.93 MHz). The progress of the reactions and the purity of the products were monitored by thin layer chromatography (TLC) on Silufol UV254 plates with a fixed layer of silica gel. Eluent CHCl₃ - MeOH (10 : 1), visualization by iodine vapor and the Kovac's reagent. Solvents were purified by standard methods. Liquid chromatograph Stayer (Akvilov, Russia). Chiral column - Chirobitic TAG column 250 x 4.6mm; eluent: MeOH / H₂O (60 / 40) + 0.2% DEA; 0.70 ml min⁻¹, UV-detector 210 nm.

(*S*)-Methionine *S*-oxide was purchased from Sigma. The initial ratio of diastereomers according to ¹H NMR was 37:63, ¹H NMR spectrum (D₂O, 400 MHz, δ , ppm); 2.13-2.19 (dt, $J = 8.0$ Hz, 2H, CH₂CH); 2.61 (s, 3H, CH₃); 2.76-3.02 (m, $J = 8.0$ Hz, $J = 4.0$ Hz, 2H, CH₂CH₂S); 3.69 - 3.74 (dd, $J = 4.0$ Hz, $J = 8.0$ Hz, 1H, CH).

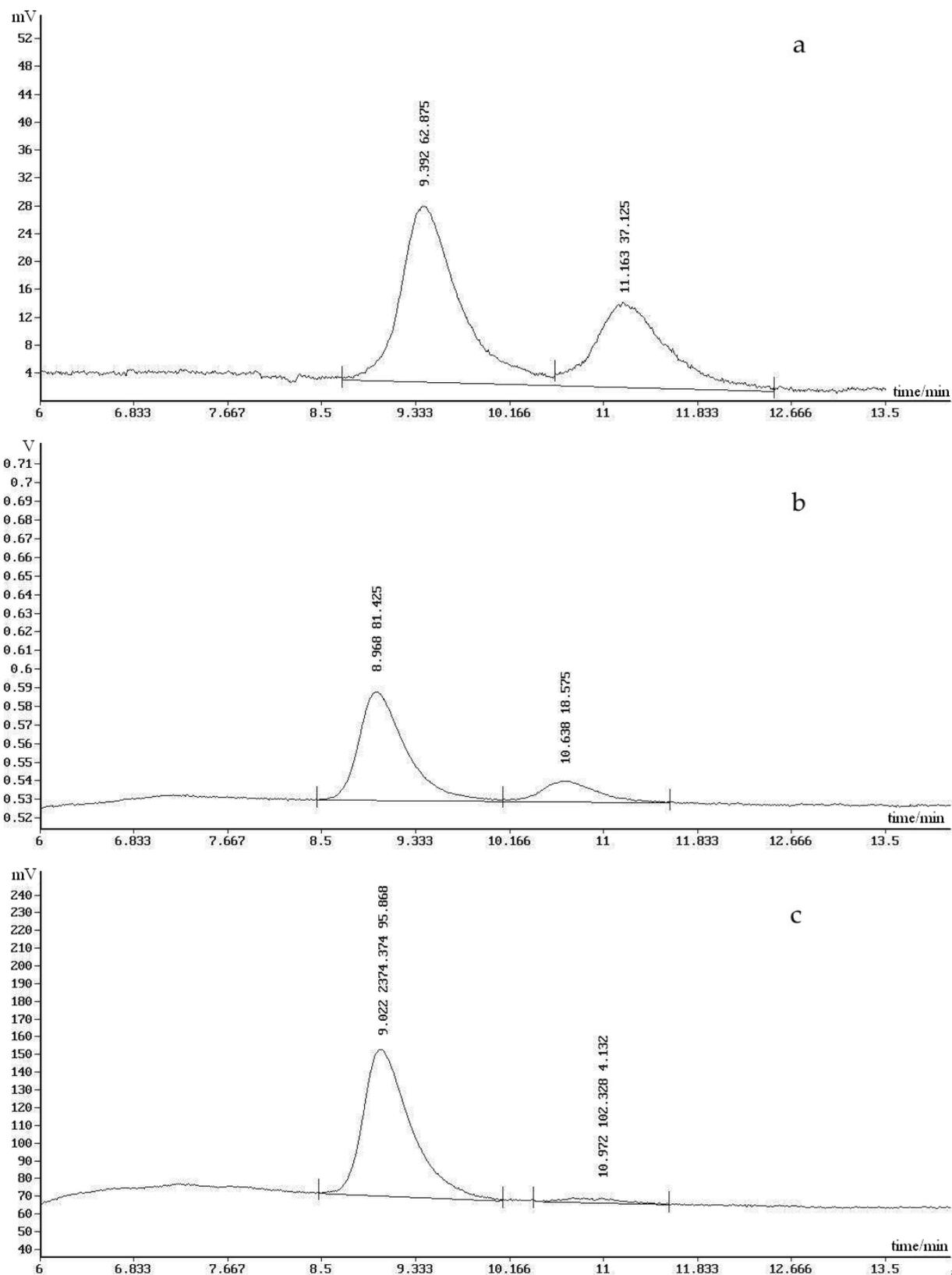


Figure S1 HPLC data. The action of methionine- γ -lyase on (S)-Methionine S-oxide. a) the initial mixture; b) after 5 h; c) after 24 h.

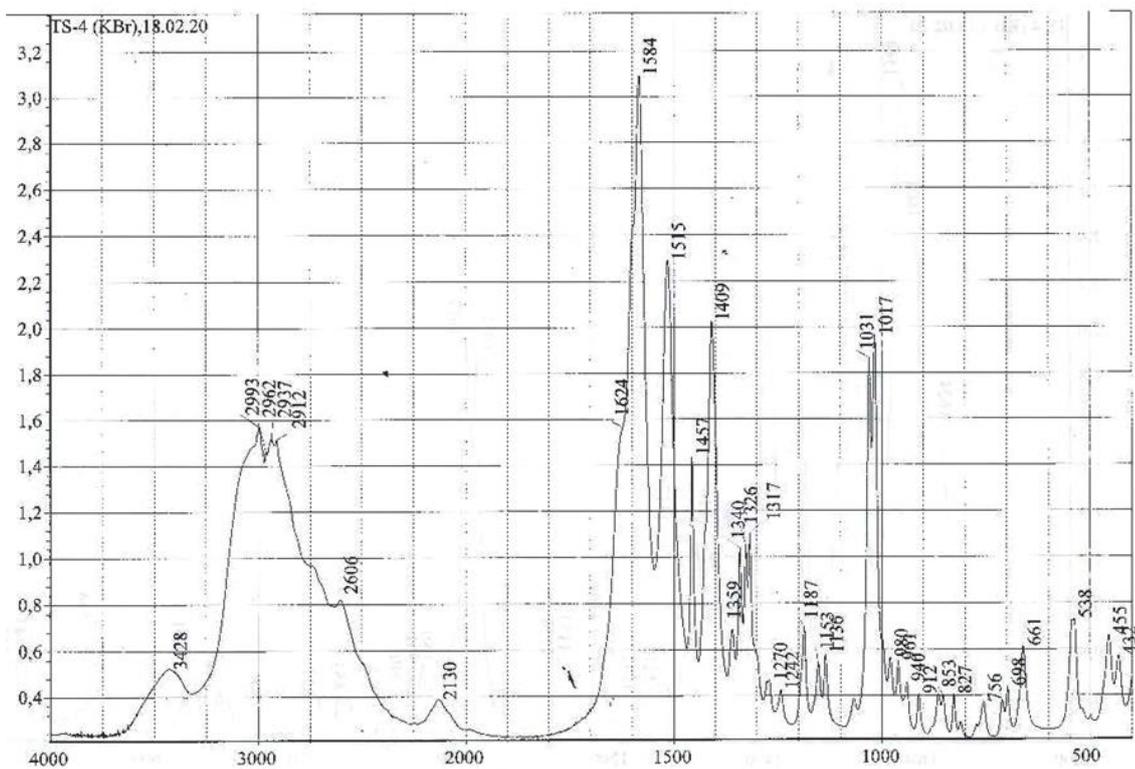


Figure S2 IR spectrum of the initial diastereomeric mixture of (*S*)-methionine *S*-oxide.

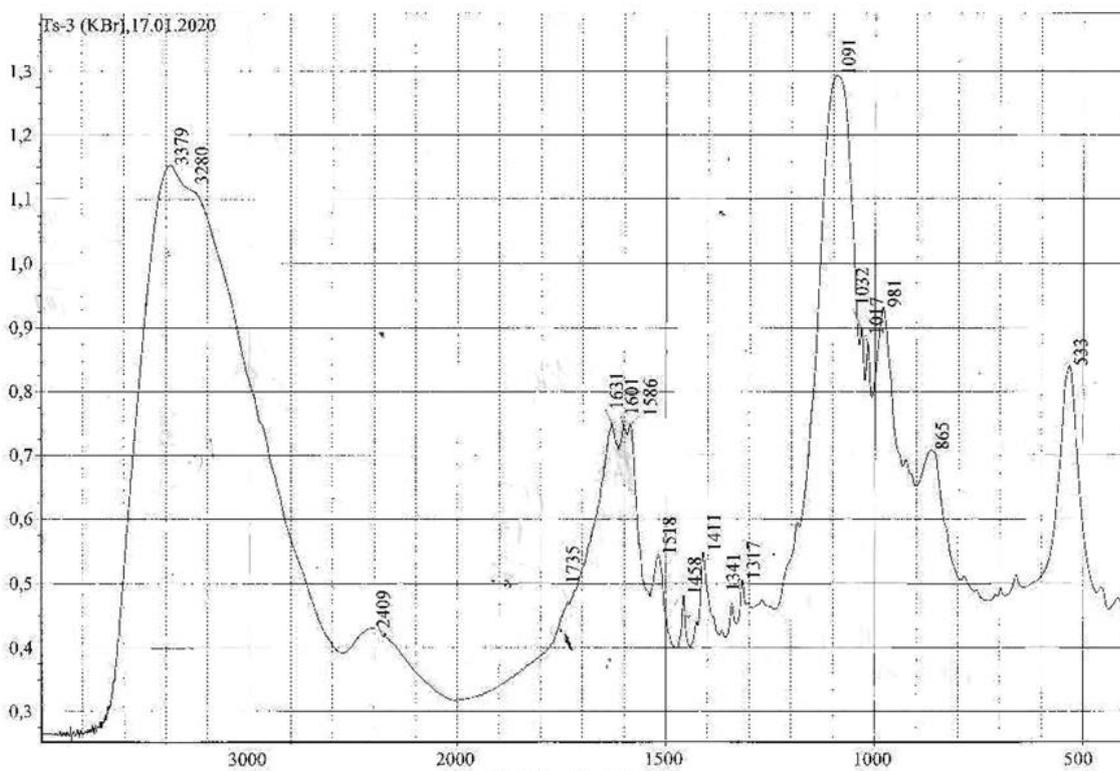


Figure S3 IR spectrum of the reaction mixture after 48 hours of enzymatic reaction.

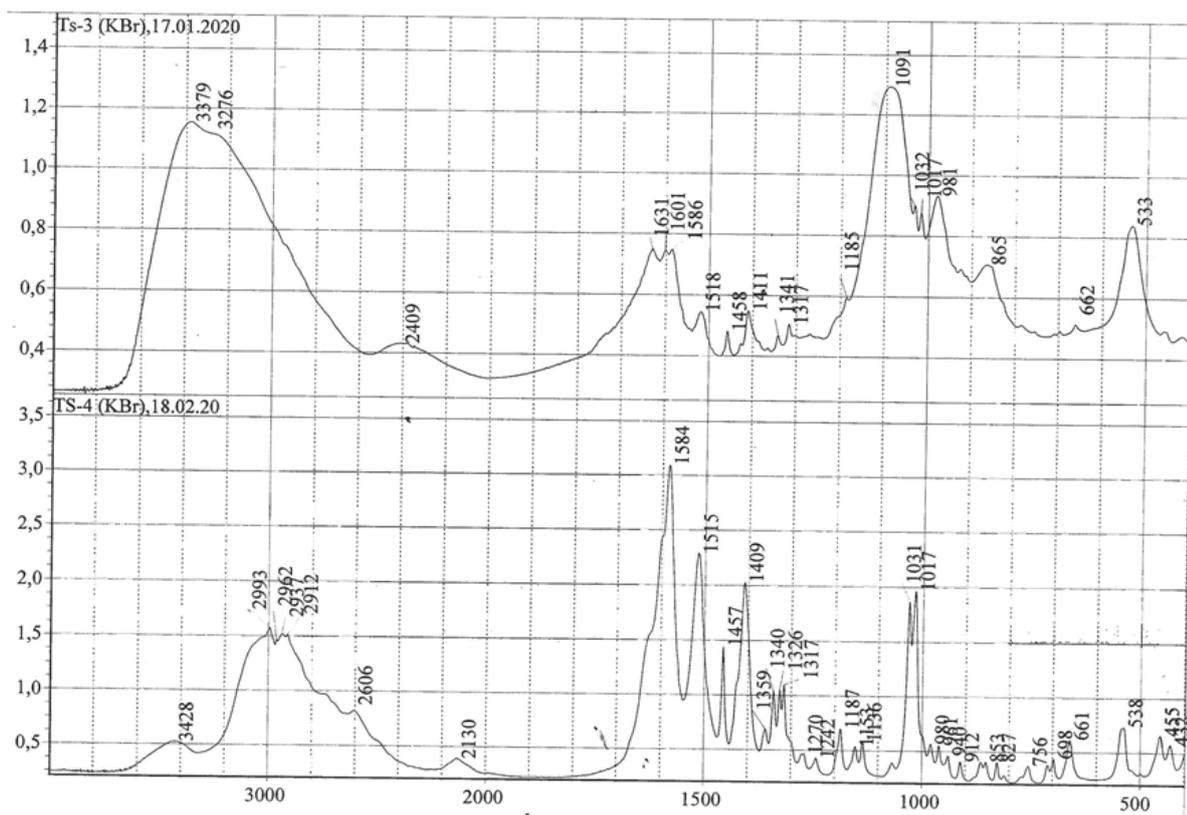


Figure S4 IR spectra. Top is the initial mixture of diastereomers, bottom is after reaction.

Display Report

Analysis Info

Analysis Name D:\Data\Kolotyorkina\2020\Kochetkov\0114016.d
Method tune_50-1600.m
Sample Name /NGKO Koch001
Comment C3H7NO3 mH 106.0498, calibrant added CH3OH

Acquisition Date 14.01.2020 15:30:57

Operator BDAL@DE
Instrument / Ser# micrOTOF 10248

Acquisition Parameter

Source Type	ESI	Ion Polarity	Positive	Set Nebulizer	1.0 Bar
Focus	Not active			Set Dry Heater	200 °C
Scan Begin	50 m/z	Set Capillary	4500 V	Set Dry Gas	4.0 l/min
Scan End	1600 m/z	Set End Plate Offset	-500 V	Set Divert Valve	Waste

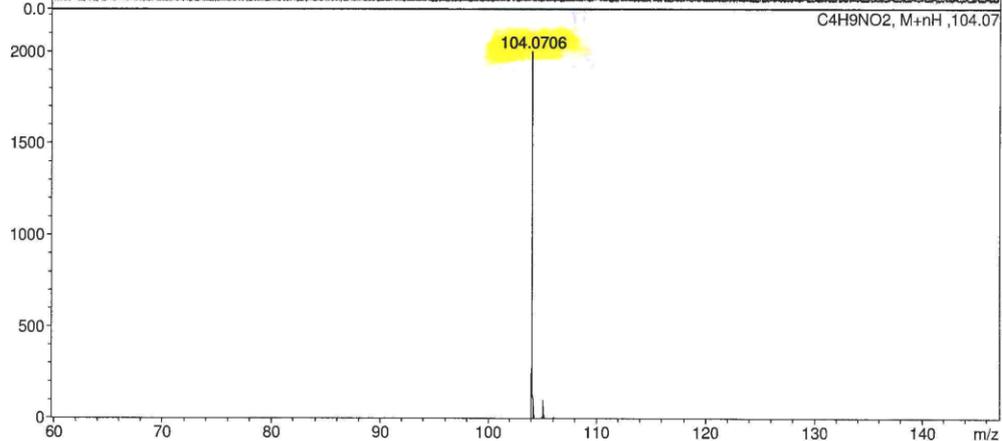
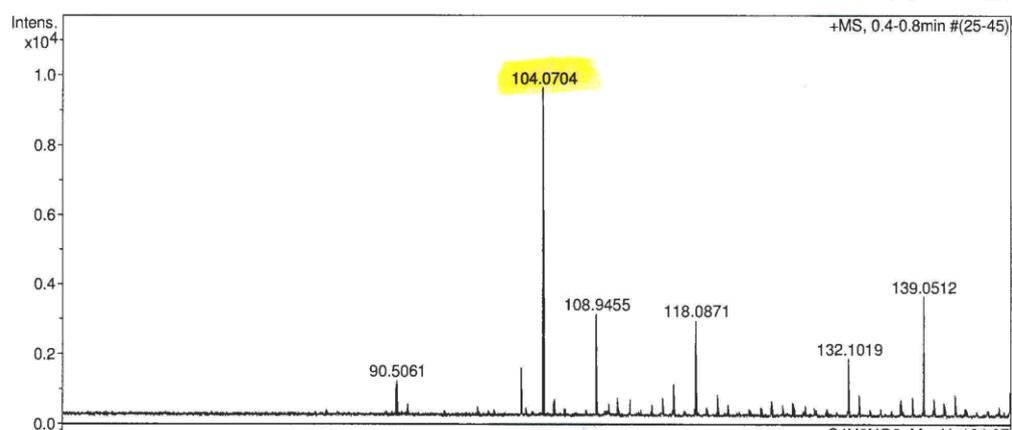
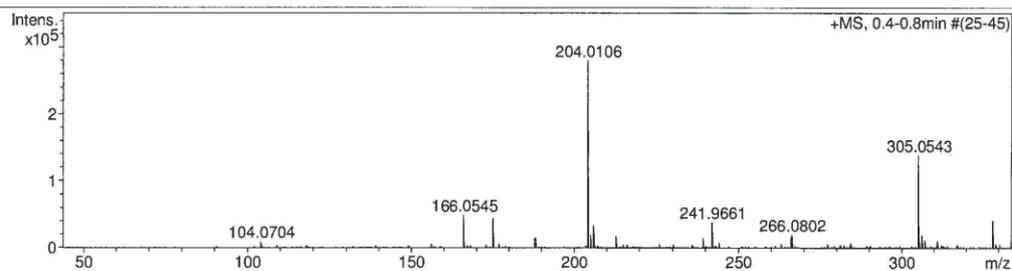


Figure S5 MALDI data of reaction mixture after 48 h.

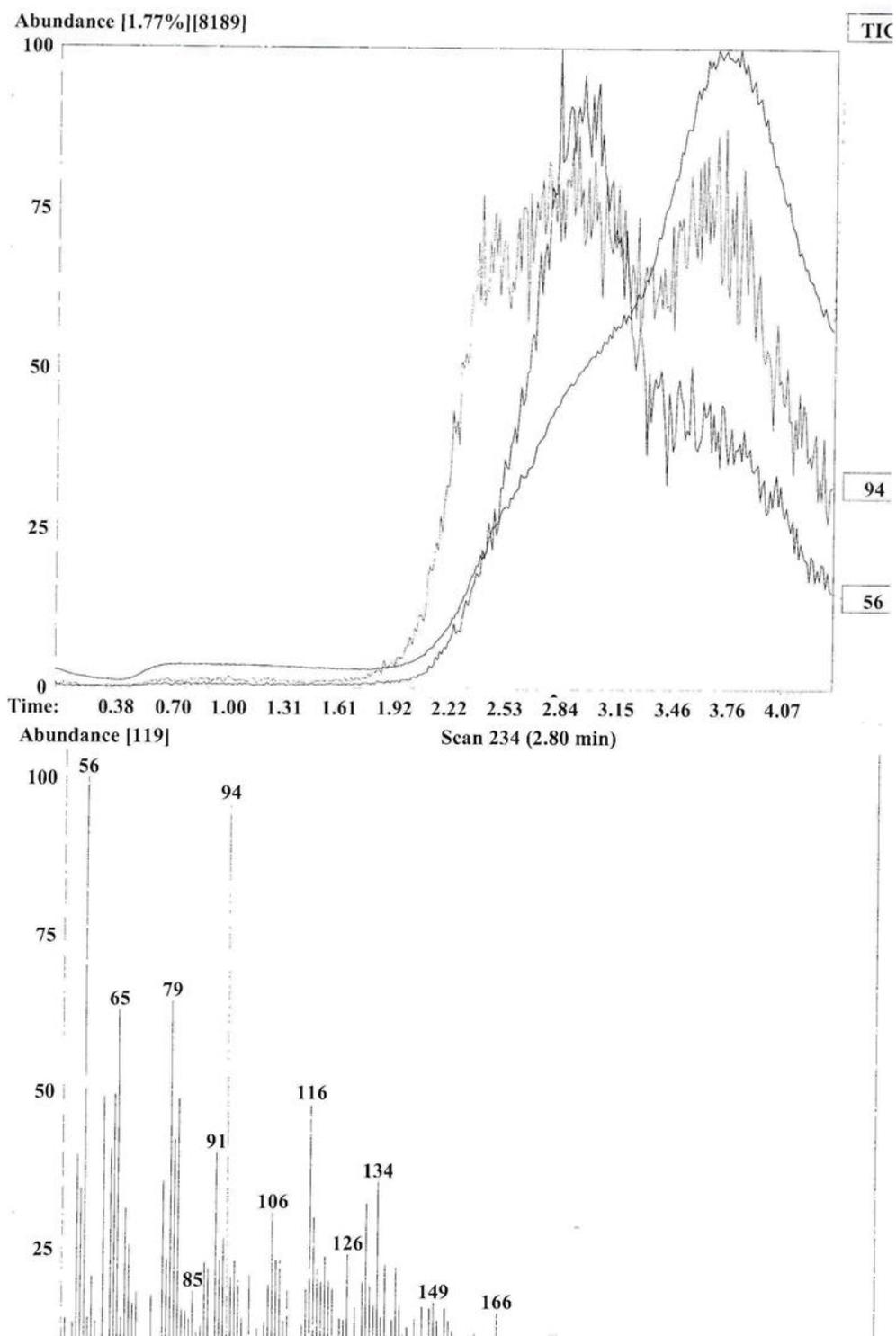


Figure S6 Mass spectral decomposition of the reaction mixture after 48 h (direct input).

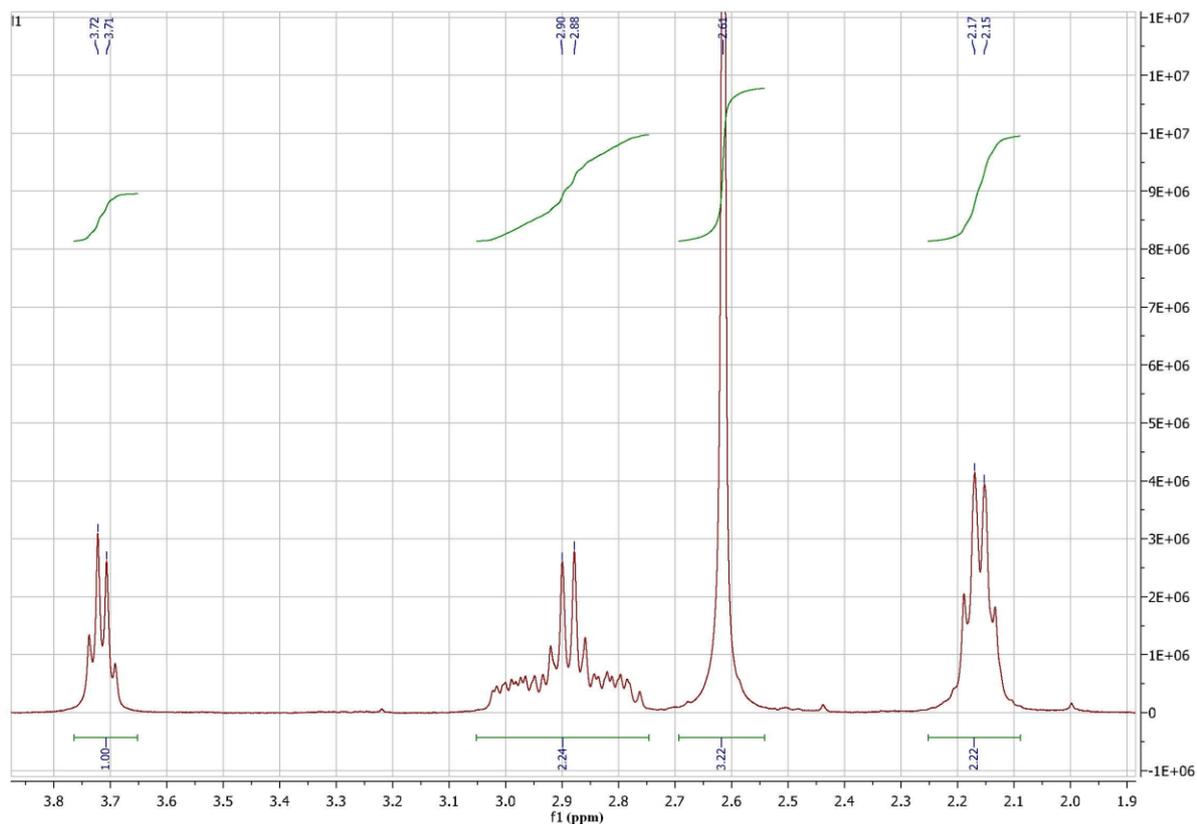


Figure S7 ¹H NMR spectrum of the initial diastereomeric mixture of (*S*)-methionine *S*-oxide.

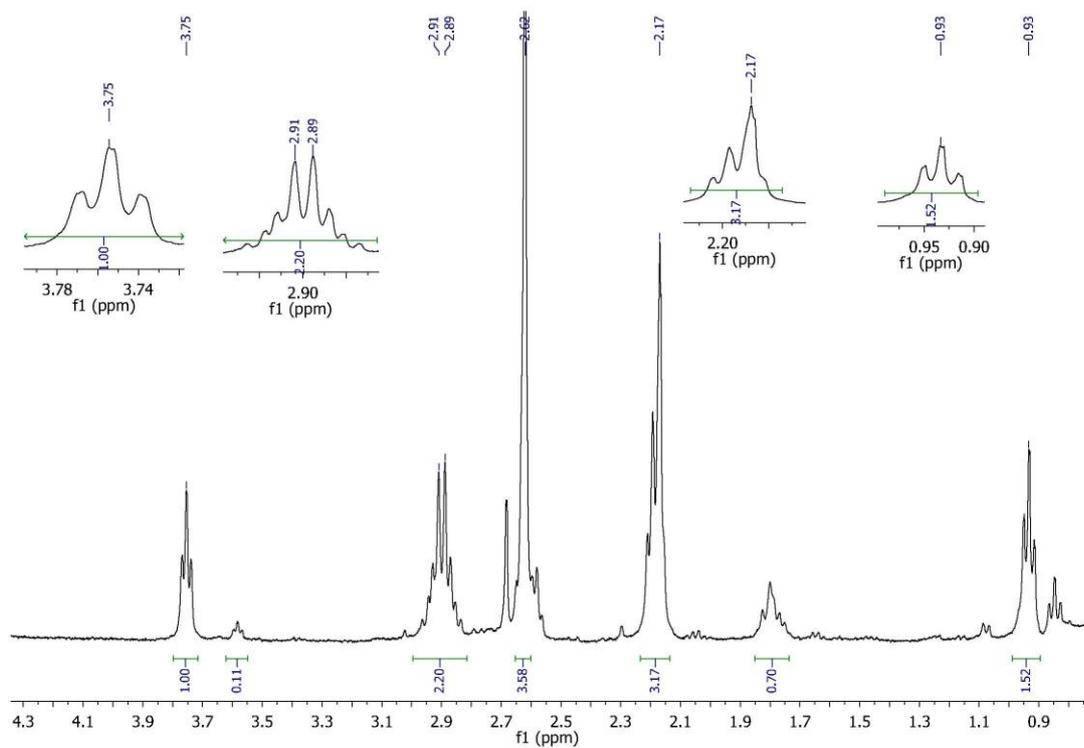


Figure S8 ¹H NMR spectrum of the reaction mixture after completion of the enzymatic process.

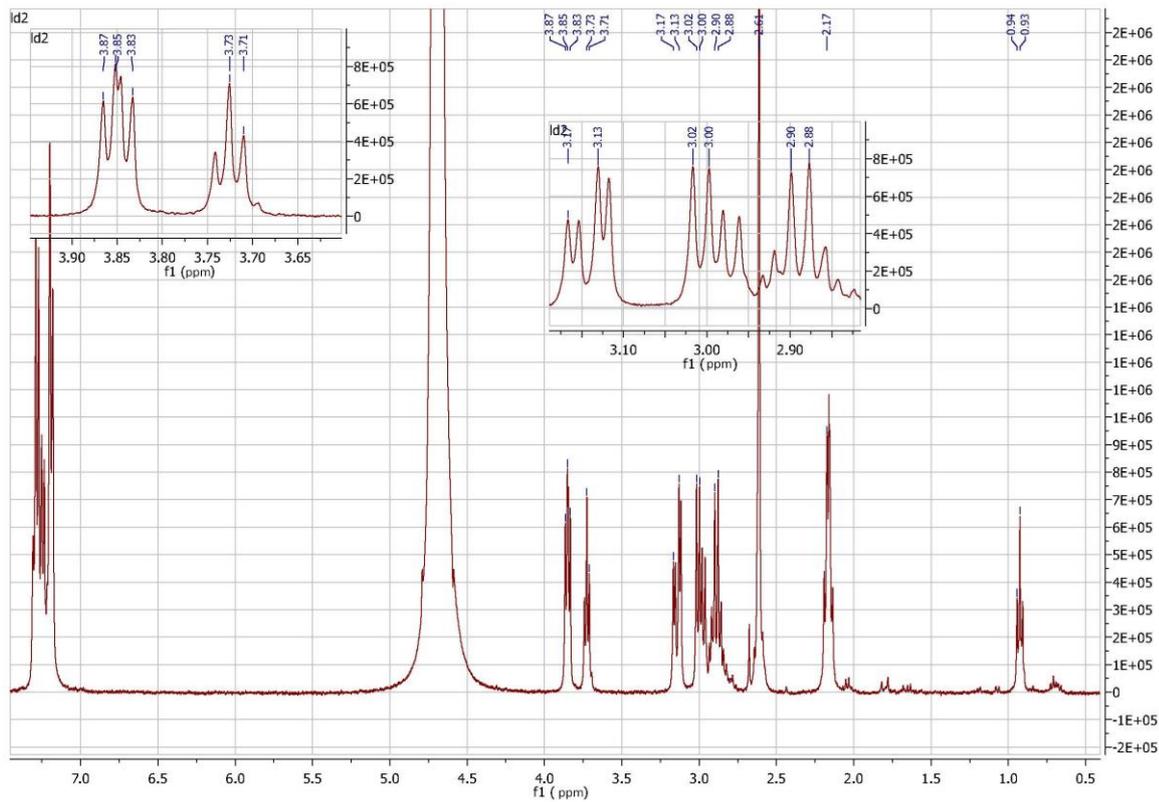


Figure S9 ¹H NMR spectrum of diastereomeric mixture of (*S*)-methionine *S*-oxide with the addition of an aliquot of (*R*)-Phe.