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Supporting information for article:

Crystallographic analysis of the *Staphylococcus epidermidis* lipase involved in esterification in aqueous solution

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S1. Materials and methods

S1.1. Enzyme kinetic characterization

Enzyme kinetic assays of purified rGehC were done using p-NP esters under optimum pH and temperature condition (Chang et~al., 2000). The p-NP esters of various chain-length fatty acids (p-NP butyrate, C4; p-NP caprylate, C8; p-NP caprate, C10; p-NP myristate, C14) were determined by measuring the amount of p-nitrophenol released. The catalytic rate constant k_{cat} (s- 1) was calculated from the initial velocity according to the equation $k_{cat} = V_{max}$ / [E].

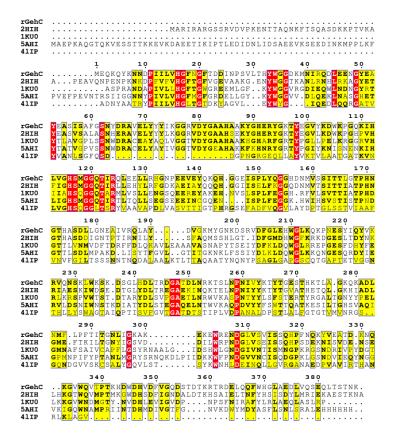


Figure S1 Sequences of lipases were aligned using MultAlin. Visualization of the multiple sequence alignment was performed with ESPript 3.0. The consensus regions are colored based on sequence conservation: red, highly conserved (more than 90% sequence identity); yellow, moderately conserved (more than 50% sequence similarity); and white, not conserved. Amino acid sequences of *S. epidermidis* lipase rGehC, *S. hyicus* lipase (2HIH), *Bacillus stearothermophilus* lipase (1KU0), *Clostridium botulinum* esterase (5AH1) and *Pseudomonas cepacia* lipase (4IIP) α/β hydroxylase were obtained from the PDB.

 Table S1
 Kinetic parameters of recombinant rGehC

Substrate	(chain)	$K_{\rm m}$ (mM)	k _{cat} (s ⁻¹)	$k_{\text{cat}}/K_{\text{m}}$ (s ⁻¹ mM ⁻¹)
<i>p</i> -NP butyrate	(C ₄)	1.03 ± 0.05	856.6 ± 34.6	836.6 ± 8.3
p-NP caprylate	(C_8)	1.59 ± 0.24	680.6 ± 77.2	431.8 ± 14.1
<i>p</i> -NP decanoate	(C_{10})	1.02 ± 0.04	524.9 ± 24.4	637.3 ± 12.1
<i>p</i> -NP myristate	(C_{14})	0.52 ± 0.03	41.0 ± 3.3	79.2 ± 1.2

The activity was assayed in 50 mM Good's buffer (pH 6.0) with different substrate concentrations of substrates and 5 μg of protein at 37°C. Each data was expressed as the mean \pm SE of three individual replication.