Electronic Supplementary Material (ESI) for Green Chemistry.

Supplementary Information

Parallel Anti-Sense Two Step Cascade for Alcohol Amination leading to ω-Amino Fatty Acids and α,ω-Diamines

Sihyong Sung[#a], Hyunwoo Jeon[#a], Sharad Sarak[a], Md Murshidul Ahsan[b], Mahesh D. Patil[a], Wolfgang Kroutil[c], Byung-Gee Kim[d], Hyungdon Yun*[a]

[a]*Department of Systems Biotechnology, Konkuk University, 120 Neungdong-ro, Gwanjin-gu, Seoul-050-29, South Korea*

[b]*School of Biotechnology, Yeungnam University, Gyeongsan 38541, South Korea*

[c]*Institute of Chemistry, University of Graz, Heinrichstrasse 28,8010 Graz, Austria*

[d]*School of Chemical and Biological Engineering, Seoul National University, Seoul 08826, South Korea*

[#] Sihyong Sung and Hyunwoo Jeon contributed equally

Table of Contents

Representative Biotransformation methods

Representative procedure exemplified for the ω-amino dodecanoic acid (ω-AmDDA) **5b:** Substrate **5a** (200 mM, final concentration) was dissolved in 100mM Tris-HCl buffer (pH 8.0) containing 20% DMSO, 400 mM benzyl amine, the pH was adjusted to pH 8.0 by addition of a 5 M NaOH solution and 0.1 mM PLP), the 36 mg_{CDW}/mL AHR/PMTA cell, 37°C, 200 rpm for 24 h. the product was separated from the reaction mixture and analysed by GC and GC-MS. For details see the Supporting Information.

Enzyme preparation

Aldehyde reductase (AHR) from Synechocystis (slr1192) [1]*:*

For expression, transformed *E. coli* BW25113, Δ fadD cells were grown in a 500 mL flask supplemented with LB/KAN medium (10 mL) at 37 °C, 200 rpm for 16 h. A sample (1 mL) of this culture was used to inoculate the experimental LB/KAN medium (100 mL). The culture was shaken at 37 °C 200 rpm for 2 h. IPTG solution (20 µL, 500 mM) was added per 100 mL culture. The cultures were shaken overnight at 20 °C, 120 rpm. Cells were harvested by centrifugation (5000 rpm, 10 min, 4°C), washed with Tris-HCl buffer (100 mM, pH 8.0), centrifuged (5000 rpm, 10 min, 4 °C) and resuspended in Tris-HCl buffer (100 mM, pH 8.0).

The gene sequence is given below.

ATGATTAAAGCCTACGCTGCCCTGGAAGCCAACGGAAAACTCCAACCCTTTGAATACGACCCCGGTGCCCTGGGTGCTAATGAGGTGGAGAT
TGAGGTGCAGTATTGTGGGGTGTGCCACAGTGATTTGTCCATGATTAATAACGAATGGGGCATTTCCAATTACCCCCTAGTGCCGGGTCATGA
GGTGGTGGGTACTGTGGCCGCCATGGGCGAAGGGGTGAACCATGTTGAGGTGGGGGATTTAGTGGGGCTGGGTTGGCATTCGGGCTACTG
CATGACCTGCCATAGTTGTTTATCTGGCTACCACAACCTTTGTGCCACGGCGGAATCGACCATTGTGGGCCACTACGGTGGCTTTGGCGATCG
GGTTCGGGCCAAGGGAGTCAGCGTGGTGAAATTACCTAAAGGCATTGACCTAGCCAGTGCCGGGCCCCTTTTCTGTGGAGGAATTACCGTTT
TCAGTCCTATGGTGGAACTGAGTTTAAAGCCCACTGCAAAAGTGGCAGTGATCGGCATTGGGGGCTTGGGCCATTTAGCGGTGCAATTTCTC
CGGGCCTGGGGCTGTGAAGTGACTGCCTTTACCTCCAGTGCCAGGAAGCAAACGGAAGTGTTGGAATTGGGCGCTCACCACATACTAGATTC
CACCAATCCAGAGGCGATCGCCAGTGCGGAAGGCAAATTTGACTATATTATCTCCACTGTGAACCTGAAGCTTGACTGGAACTTATACATCAG
CACCCTGGCGCCCCAGGGACATTTCCACTTTGTTGGGGTGGTGTTGGAGCCTTTGGATCTAAATCTTTTTCCCCTTTTGATGGGACAACGCTCC
GTTTCTGCCTCCCCAGTGGGTAGTCCCGCCACCATTGCCACCATGTTGGACTTTGCTGTGCGCCATGACATTAAACCCGTGGTGGAACAATTTA
GCTTTGATCAGATCAACGAGGCGATCGCCCATCTAGAAAGCGGCAAAGCCCATTATCGGGTAGTGCTCAGCCATAGTAAAAAT

The amino acid sequence is as follows:

MIKAYAALEANGKLQPFEYDPGALGANEVEIEVQYCGVCHSDLSMINNEWGISNYPLVPGHEVVGTVAAMGEGVNHVEVGDLVGLGWHSGYC
MTCHSCLSGYHNLCATAESTIVGHYGGFGDRVRAKGVSVVKLPKGIDLASAGPLFCGGITVFSPMVELSLKPTAKVAVIGIGGLGHLAVQFLRAWGC
EVTAFTSSARKQTEVLELGAHHILDSTNPEAIASAEGKFDYIISTVNLKLDWNLYISTLAPQGHFHFVGVVLEPLDLNLFPLLMGQRSVSASPVGSPATI
ATMLDFAVRHDIKPVVEQFSFDQINEAIAHLESGKAHYRVVLSHSKN

Transaminase (ω -TA) from Silicibacter pomeroyi [2]*:*

For expression, transformed *E. coli* BW25113, Δ fadD cells were grown in a 500 mL flask supplemented with LB/amp medium (10 mL) at 37 °C, 200 rpm for 16 h. A sample (1 mL) of this culture was used to inoculate the experimental LB/amp medium (100 mL) The culture was shaken at 37 °C 200 rpm for 2 h. IPTG solution (20 µL, 500 mM) was added per 100 mL culture. The cultures were shaken overnight at 20 °C, 120 rpm. Cells were harvested by centrifugation (5000 rpm, 10 min, 4°C), washed with Tris-HCl buffer (100 mM, pH 8.0), centrifuged (5000 rpm, 10 min, 4°C) and resuspended in Tris-HCl buffer (100 mM, pH 8.0).

The gene sequence is given below:

ATGGCTACTATCACCAACCACATGCCTACCGCGGAACTGCAGGCTCTGGATGCCGCCCATCATCTGCACCCATTCTCTGCAAACAACGCTCTG
GGTGAAGAGGGTACTCGTGTAATCACGCGTGCACGTGGTGTATGGCTGAACGATTCTGAGGGTGAGGAGATCCTGGACGCAATGGCAGGTC
TGTGGTGTGTAAACATCGGTTACGGTCGTGACGAGCTGGCAGAAGTAGCAGCACGTCAAATGCGTGAGCTGCCTTACTACAACACGTTCTTC
AAGACCACGCACGTCCCAGCAATCGCACTGGCTCAGAAGCTGGCAGAGCTGGCTCCAGGTGATCTGAACCATGTATTCTTCGCTGGCGGTGG
TTCTGAGGCAAACGACACTAACATCCGCATGGTCCGTACTTACTGGCAGAACAAGGGCCAGCCTGAAAAAACCGTCATCATCTCCCGTAAAAA
CGCCTACCACGGCTCCACCGTCGCTTCCTCCGCTCTGGGTGGTATGGCTGGTATGCATGCTCAGTCTGGTCTGATCCCGGATGTTCATCACATC
AACCAGCCGAACTGGTGGGCTGAAGGTGGTGATATGGACCCGGAAGAATTTGGCCTGGCTCGTGCTCGTGAACTGGAAGAAGCTATCCTGG
AACTGGGTGAAAACCGCGTTGCTGCTTTCATCGCGGAACCGGTTCAGGGTGCGGGTGGTGTTATTGTTGCCCCGGATTCTTACTGGCCGGAA
ATCCAACGTATCTGCGACAAATACGACATCCTGCTGATCGCGGACGAAGTGATCTGCGGTTTCGGCCGTACTGGTAACTGGTTCGGCACTCAG
ACCATGGGCATCCGTCCGCACATTATGACCATCGCGAAAGGTCTGTCTTCTGGCTACGCGCCGATTGGCGGCAGCATTGTTTGTGATGAAGTG
GCCCACGTTATCGGCAAAGACGAATTTAACCACGGCTATACCTATAGCGGCCACCCGGTGGCCGCAGCAGTTGCGCTGGAAAATCTGCGTAT
TCTGGAAGAGGAGAACATCCTGGACCACGTTCGTAACGTGGCGGCGCCGTATCTGAAAGAAAAATGGGAAGCGCTGACCGACCACCCGCTG
GTTGGCGAAGCGAAAATTGTTGGCATGATGGCGAGCATCGCGCTGACCCCGAATAAAGCCAGCCGTGCGAAATTTGCGTCCGAACCGGGCA
CCATTGGCTATATTTGCCGCGAACGCTGCTTTGCGAATAATCTGATCATGCGCCACGTGGGCGATCGCATGATTATCTCCCCGCCGCTGGTGA
TTACCCCGGCGGAAATCGATGAAATGTTCGTGCGCATTCGCAAAAGCCTGGACGAAGCCCAGGCGGAAATTGAAAAACAGGGCCTGATGAA
AAGCGCCT

The amino acid sequence is:

MATITNHMPTAELQALDAAHHLHPFSANNALGEEGTRVITRARGVWLNDSEGEEILDAMAGLWCVNIGYGRDELAEVAARQMRELPYYNTFFKT
THVPAIALAQKLAELAPGDLNHVFFAGGGSEANDTNIRMVRTYWQNKGQPEKTVIISRKNAYHGSTVASSALGGMAGMHAQSGLIPDVHHINQP
NWWAEGGDMDPEEFGLARARELEEAILELGENRVAAFIAEPVQGAGGVIVAPDSYWPEIQRICDKYDILLIADEVICGFGRTGNWFGTQTMGIRP
HIMTIAKGLSSGYAPIGGSIVCDEVAHVIGKDEFNHGYTYSGHPVAAAVALENLRILEEENILDHVRNVAAPYLKEKWEALTDHPLVGEAKIVGMMA
SIALTPNKASRAKFASEPGTIGYICRERCFANNLIMRHVGDRMIISPPLVITPAEIDEMFVRIRKSLDEAQAEIEKQGLMKSA

ω -Amino fatty acids (ω-AmFAs)

AHR activity towards benzaldehyde

To confirm the AHR activity, A reaction with benzaldehyde was carried out in presence of 10% EtOH at varying pH values from 7.0 to 9.0. The results confirmed the highest reactivity of AHR at pH 8.0 with efficiently converting benzaldehyde into benzyl alcohol

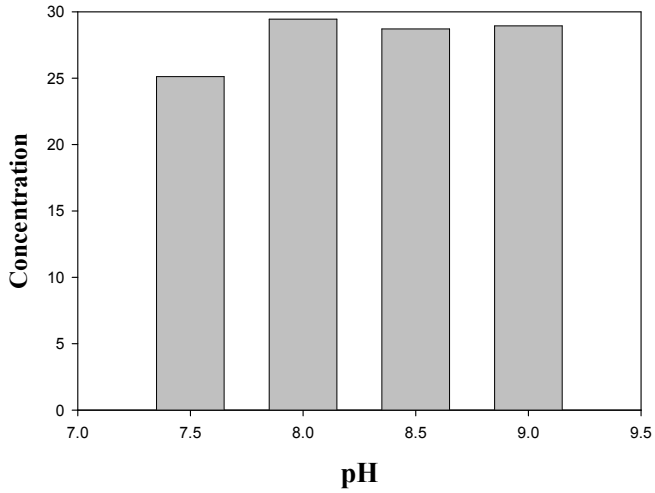


Figure S1. Reaction condition: 30 mM benzaldehyde,1 mM NAD^+, 10% (v/v) EtOH, 0.05 mg/mL AHR purified protein, 100 mM Tris-HCl buffer, 37 °C.

Screening and optimization of ω-TA:

Table S1. List of .in-house ω-TAs

Entry	Organism	Abbreviation	Protein ID No.
1	*Synechocystis sp. PCC 6714*	*AHR*	*WP_028949165.1*
2	*Silicibacter pomeroyi*	*PMTA*	*WP_011049154.1*
3	*Chromobacterium violaceum*	*CVTA*	*WP_011135573.1*
4	*Vibrio fluvialis JS17*	*VFTA*	*WP_040602310.1*
5	*agrobacterium tumefaciens*	*ATTA*	*WP_010972924.1*
6	*Pseudomonas putida*	*SpuTA*	*WP_016502144.1*
7	*Polaromonas sp. JS666*	*POTA*	*WP_011482414.1*
8	*Sphaerobacter thermophilus*	*STTA*	*WP_012871332.1*
9	*Burkholderia plantarii*	*BGTA*	*WP_006050635.1*
10	*Aspergillus fischeri NRRL 181*	*NFTA*	*XP_001261640.1*
11	*Mesorhizobium loti*	*MLTA*	*WP_010909990.1*

1) ω -Amino fatty acids (ω-AmFAs)

Optimization of ω-TA was performed by using previously developed strategy for the screening of reported ω-TA enzymes and the best results were found for four active ω-transaminases (*Silicibacter pomeroyi*, *Chromobacterium violaceum*, *Vibrio Fluvialis and Agrobacterium Tumefaciens*)

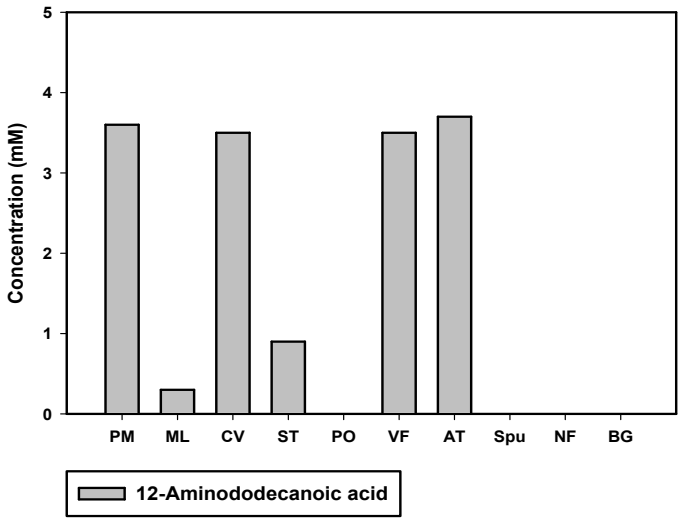


Figure S2. Reaction Condition: 5 mM Substrate (12-hydroxyldodecanoic acid) 20 mM benzylamine, 100 mM Tris-HCl buffer, 8.0 pH, AHR 0.5mg/mL, ω-TA 0.1 mg/mL purified protein, 0.5 mM NAD^+, 0.1 mM PLP, 5% (v/v) DMSO, 37 °C, 24 h.

Optimization of pH-value

A reaction was carrying with pH-values varying from 7.5 to 9.0 using 5 mM 12-hydroxy dodecanoic acid as a substrate. The highest conversion to the 12-hydroxydodecanoic acid (ω-HDDA) was obtained at pH 8.0. Therefore, further experiments were conducted at the optimum pH value of 8.0.

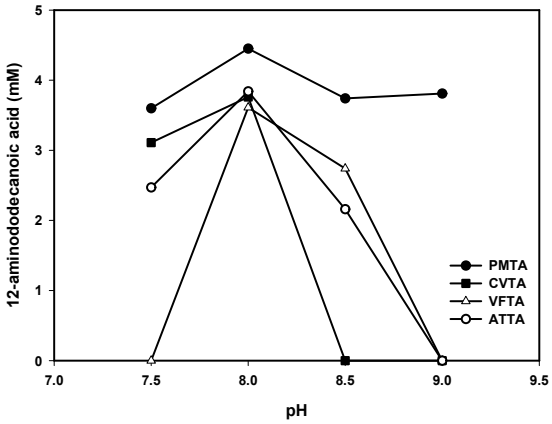


Figure S3. Reaction condition: 5 mM Substrate (12-hydroxydodecanoic acid), 20 mM Benzylamine, 100 mM Tris-HCl buffer, 7.5-9.0 pH, AHR 0.5 mg/mL, ω-TA 0.5 mg/mL Purified protein, 0.5 mM NAD^+, 0.1 mM PLP, 5% v/v DMSO, 37 °C, 24 h.

Comparison of amine donor

For optimization and selection of a suitable amine donor, relative screening analysis was performed between (*S*)-α-methyl benzylamine ((*S*)-α-MBA) and benzylamine as amine donor. The results showed that benzyl amine had a considerable reactivity in comparison with that of (*S*)-α-MBA.

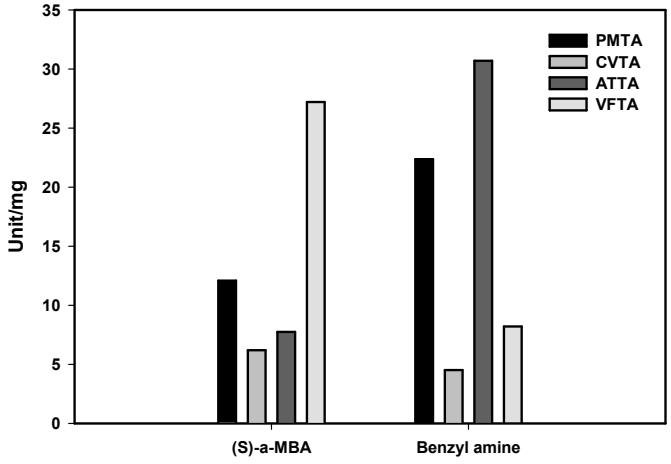


Figure S4. Reaction condition: 10 mM substrate (pyruvate), 20 mM amine, pH 8.0, AHR/ω-TA 0.001 mg/mL, 100 mM Tris-HCl buffer, 0.1 mM PLP, 37°C, 24 h.

SDS PAGE for expression of AHR and ω-TA

AHR and ω-TA were separately expressed in (*E. coli*, BW25113 (Δ fadD) cells using pET-24ma as vector.

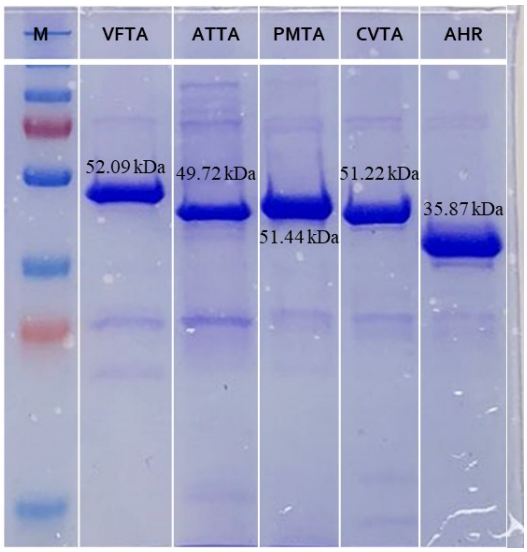


Figure S5. SDS PAGE for expression of AHR and ω-TA

20 mM reaction using whole-cell system, seperatly expressing AHR and ω-TA

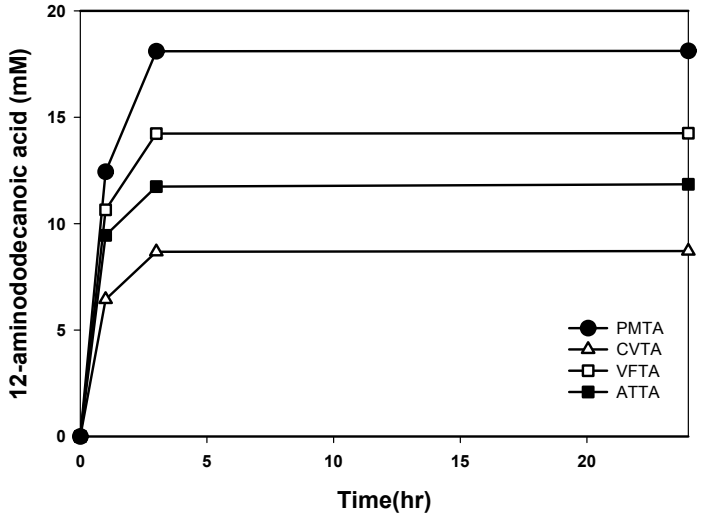


Figure S6. Reaction condition: 20 mM substrate (12-hydroxydodecanoic acid) 60 mM benzylamine, 100 mM Tris-HCl buffer pH 8.0, 9 mg_{CDW}/mL AHR/ ω-TA cell conc, 5% v/v DMSO, 37°C, 24 h.

Optimization at 100 mM concentration using whole-cell system, seperatly expressing AHR and ω-TA

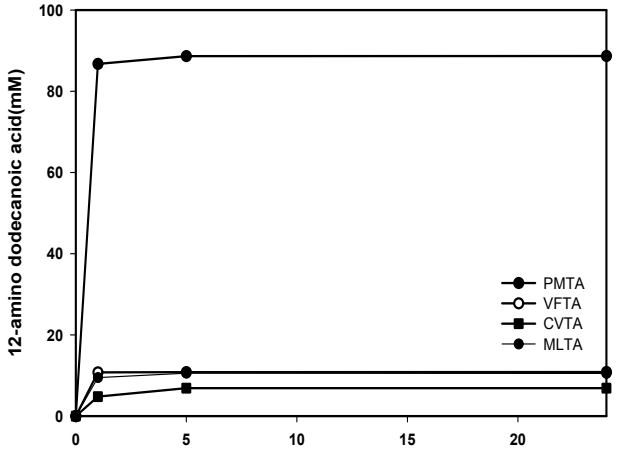
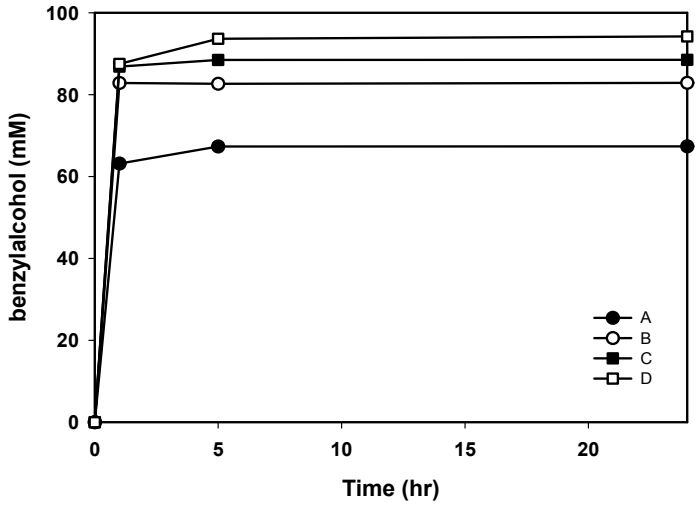


Figure S7. Reaction condition: 100 mM Substrate (12-hydroxydodecanoic acid), 200 mM benzylamine, 100 mM Tris-HCl buffer pH 8.0, 18 mg_{CDW}/mL AHR/ω-TA cell conc, 5% v/v DMSO, 37 °C, 24 h.

3) Confirmation of transamination with benzylamine as an amine donor

To confirm the transamination reaction using benzyl amine as an amine donor, consumption of benzyl amine at varying cell concentrations was calculated over the reaction time.



A: AHR: 9 mg_{CDW}/mL
PMTA: 9 mg_{CDW}/mL
B: AHR: 18 mg_{CDW}/mL
PMTA: 9 mg_{CDW}/mL
C: AHR: 18 mg_{CDW}/mL
PMTA: 18 mg_{CDW}/mL
D: AHR: 27 mg_{CDW}/mL
PMTA: 27 mg_{CDW}/mL

Figure S8. Reaction condition: 100 mM Substrate (12-hydroxydodecanoic acid), 200 mM benzylamine, 100 mM Tris-HCl buffer pH 8.0, AHR 9-27 mg_{CDW}/mL, ω-TA 9-27 mg_{CDW}/mL cell conc, 10% v/v DMSO, 37 °C, 24 h.

SDS-PAGE for expression of AHR and ω-TA in a single cell system

The AHR and PMTA were cloned using three different combinations of vectors (**A** pQE-80L and pET-24ma **B** pET-24ma and pET-Duet1 **C** pET-Duet1 and pET-24ma for AHR and PMTA, respectively). Amongst these systems, system **C** showed good expression for both the enzymes.

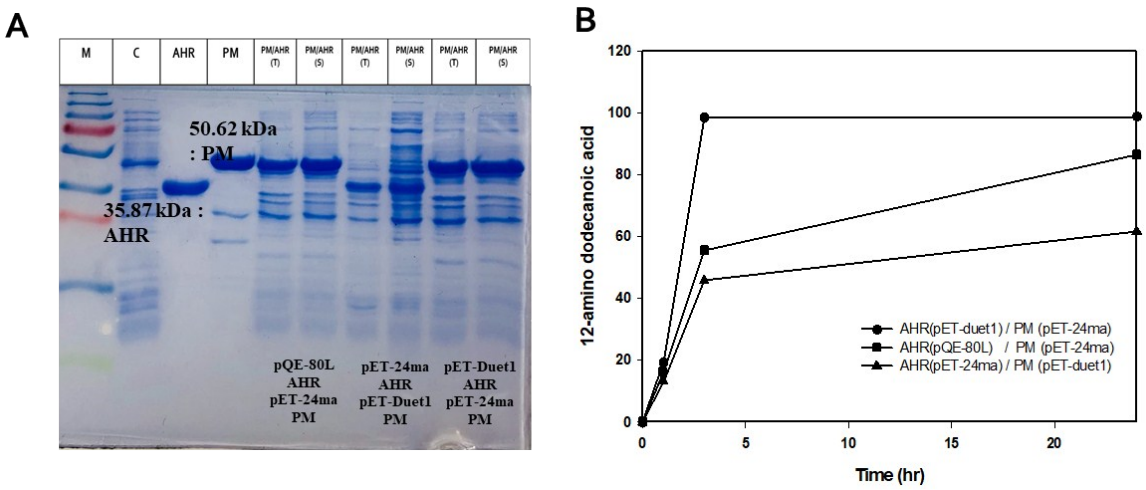


Figure S9. Reaction condition: 100 mM substrate (12-hydroxydodecanoic acid), 200 mM benzylamine, 100 mM Tris-HCl buffer pH 8.0, 9-27 mg_{CDW}/mL AHR /ω-TA, 10% v/v DMSO, 37 °C, 24 h.

Comparison with separately expressed AHR/ ω-TA system:

In comparison with in two different cell system the co-expression in single cell showed good expression results see the **Fig S10**

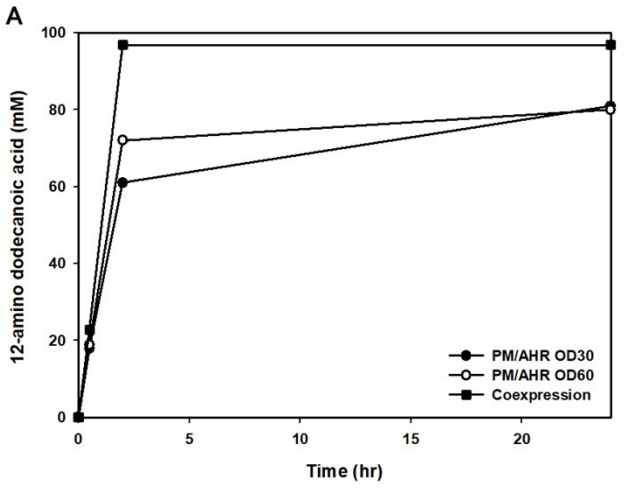


Figure S10. Reaction condition: 100 mM substrate (12-hydroxydodecanoic acid), 200 mM benzylamine, AHR-PMTA 36 mg_{CDW}/mL Cell conc, 100 mM Tris-HCl buffer pH 8.0, 37°C, 10% v/v DMSO.

Comparison of productivity with respect to cell concentration

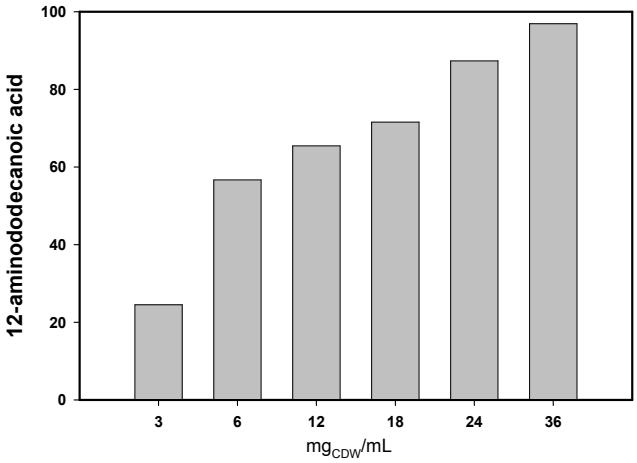


Figure S11. Reaction condition: 5 mM substrate (12-hydroxydodecanoic acid), 20 mM benzylamine, 100 mM Tris-HCl buffer pH 8.0, 37 °C, 5% v/v DMSO.

Table S2. 20 mM reaction using different ω-HFAs[a].

No	Substrate	*n*	R	Amine	[mM]
1	**1a**	4	H	**1b**	19.9
2	**2a**	6	H	**2b**	19.9
3	**3a**	8	H	**3b**	18.5
4	**4a**	10	H	**4b**	18.3
5	**5a**	12	H	**5b**	18.0
6	**6a**	14	H	**6b**	16.5

[a] Reaction conditions: 20 mM substrate (6-16-hydroxy fatty acids), 60 mM benzylamine, 18 mg$_{CDW}$/mL AHR-PMTA cell conc, 100 mM Tris-HCl buffer, pH 8.0, 37 °C, 10% v/v DMSO

200 mM reaction using ω-OHDDA as substrate

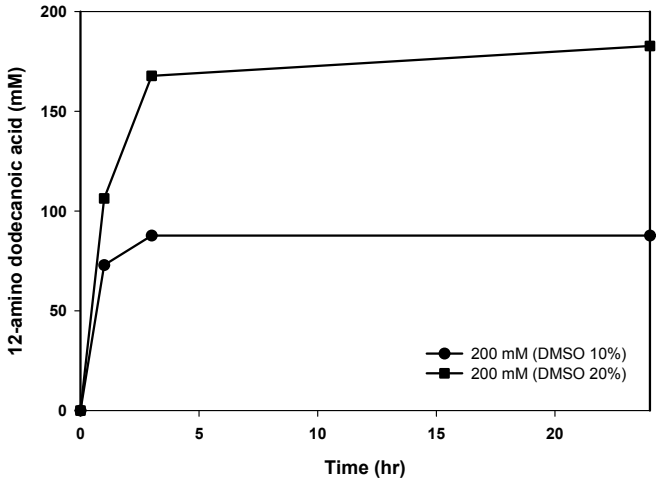


Figure S12. Reaction condition: 200 mM substrate (12-hydroxydodecanoic acid), 400 mM benzylamine, 200 mM Tris-HCl buffer pH 8.0, AHR-ω-TA 36 mg_{CDW}/mL cell conc, 20 % v/v DMSO, 37°C, 24 h.

Reaction using ω-hydroxydodecanoic acid methyl ester (ω-ADAME) as a substrate

Optimization of ω-TA:

Screening of ω-TA for 12-amino dodecanoic acid methyl ester (12-ADAME) was performed using above optimized strategy for screening of active ω-TA enzymes and best results were achieved with *Silicibacter pomeroyi* ω-TA.

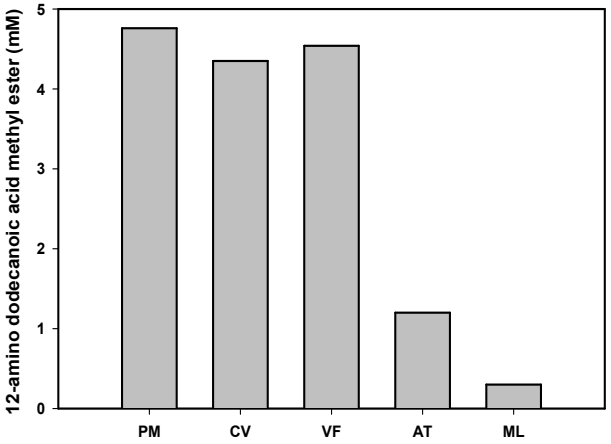


Figure S13. Reaction condition: 5 mM substrate (12-hydroxydodecanoate), 20 mM benzylamine, AHR-PMTA 0.1mg/mL purified proteins, 100 mM Tris-HCl buffer pH 8.0, 0.1 mM PLP, 37 °C, 5% v/v DMSO.

Comparison of two cell system with a co expression system

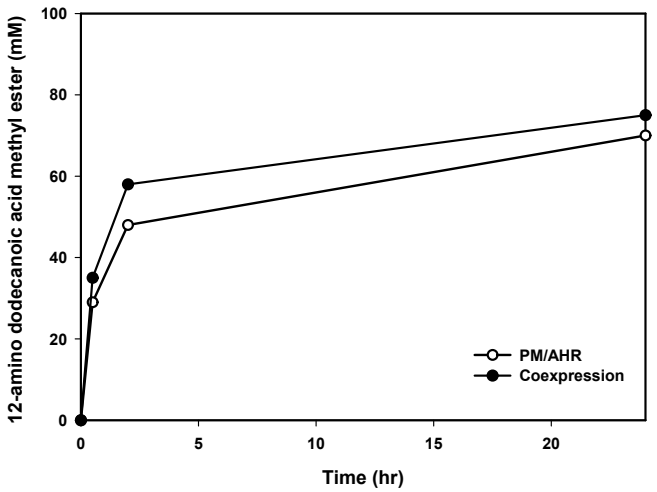


Figure S14. Reaction condition: 100 mM substrate (12-hydroxydodecanoate), 200 mM benzylamine, AHR-PMTA 36 mg/mL, 100 mM Tris-HCl buffer pH 8.0, 37 °C, 5% v/v DMSO.

Biosynthesis of α, ω-diamines

Optimization of ω-TA

Considering our previously reported strategy for screening of active ω-TA enzymes, the ω-TA obtained from *Silicibacter pomeroyi* (Sp ω -TA, PMTA) was identified as the most suitable TA for the biosynthesis of α, ω-diamines.

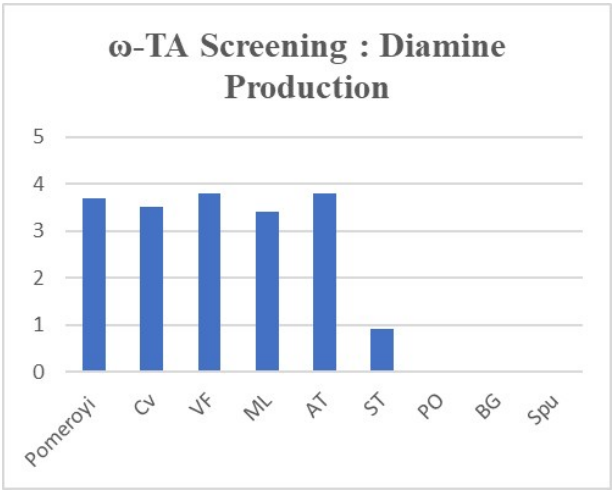


Figure S15. Reaction condition: 5 mM substrate (1,10 decandiol), 20 mM benzylamine, AHR-PMTA 0.1mg/mL purified proteins, 100 mM Tris-HCl buffer pH 8.0, 1 mM NAD^+, 37 °C, 5% v/v DMSO.

Optimization of Co-solvent

A) DMSO

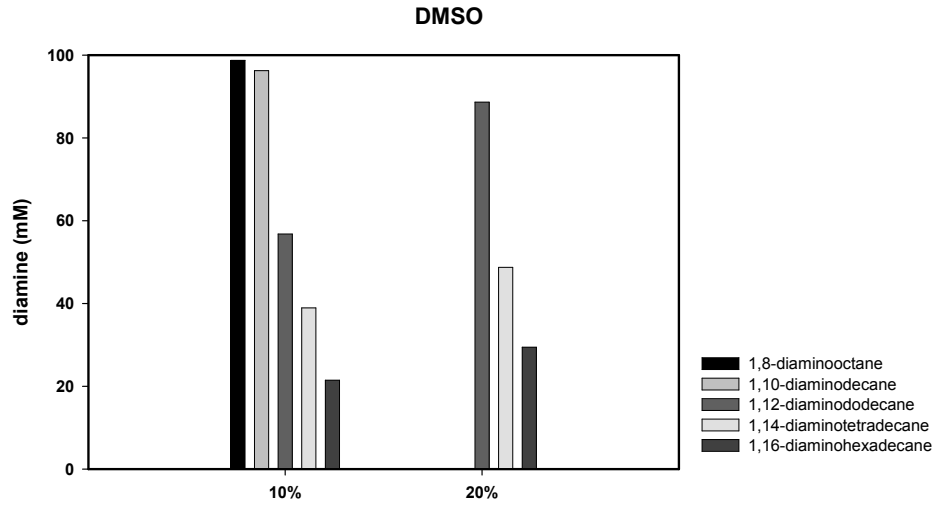


Figure S16. Reaction condition: 100 mM substrate (C_8 to C_{16} diols), 400 mM benzylamine, 200 mM Tris-HCl buffer pH 8.0, 36 mg_{CDW}/mL AHR/ ω-TA cell conc, 37°C, 24 h.

B) DME

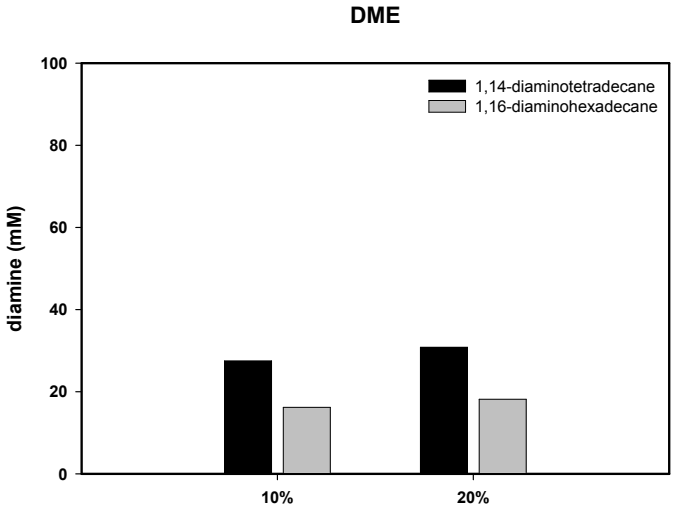


Figure S17. Reaction condition: 100 mM substrate (C_{14} to C_{16} diols), 400 mM benzylamine, 200 mM Tris-HCl buffer pH 8.0, 36 mg_{CDW}/mL AHR/ ω-TA cell conc, 37°C, 24 h.

Relative activity of amino donor

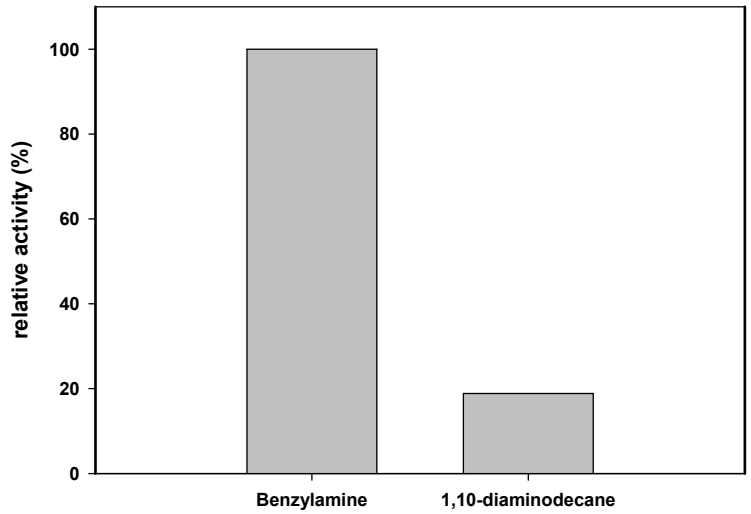


Figure S18. Reaction condition: 10 mM pyruvate substrate, 20 mM (benzylamine and 1,10-diaminodecane), 0.005 mg/mL for benzylamine and 0.02 mg/mL for 1,10-diaminodecane purified enzymes, 100 mM Tris-HCl buffer, pH 8.0, 37°C, 24 h.

Production of ω- AmHA using Lactonase, AHR and ω- TA

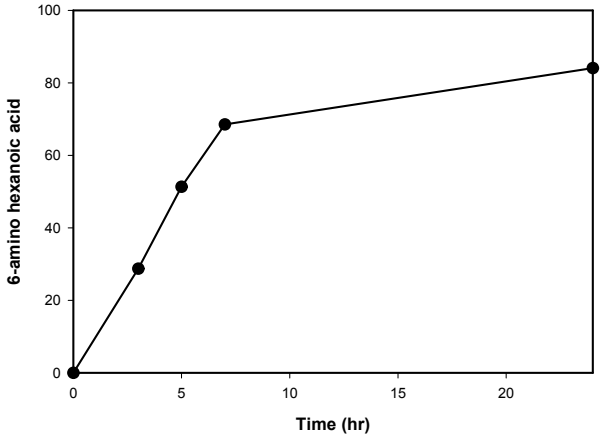


Figure S19. Reaction condition: 100 mM substrate (ε-caprolactone), 200 mM benzylamine, 100 mM Tris-HCl buffer pH 8.0, 9 mg_{CDW}/mL lactonase, 9 mg_{CDW}/mL AHR/ ω-TA cell conc, 10% DMSO, 37°C, 24 h

Table S3. List of reagents and products with retention times observed

	Sr. No	Substrate	Abbre-viation	Company	Purity (%)	Retention time [min]	Method
ω-Hydroxy Fatty acids	1	6-hydroxyhexanoic acid	**1a**	Alfa-aesar	95	6.34	1
	2	8-hydroxyoctanoic acid	**2a**	Sigma Aldrich	98	7.91	1
	3	10-hydroxydecanoic acid	**3a**	Sigma Aldrich	Technical grade	9.67	1
	4	11-hydroxyundecanoic acid	**4a**	Sigma Aldrich	96	10.63	1
	5	12-hydroxydodecanoic acid	**5a**	Sigma Aldrich	97	11.79	1
	6	16-hydroxyhexadecanoic acid	**6a**	Sigma Aldrich	98	16.9	1
	7	12-hydroxydodecanoic acid methyl ester	**7a**	-	-	17.68	2
ω-Amino Fatty acids	8	6-aminohexanoic acid	**1b**	Alfa-aesar	98	8.91	1
	9	8-aminooctanoic acid	**2b**	Sigma Aldrich	99	10.94	1
	10	10-aminodecanoic acid	**3b**	-	-	13.28	1
	11	11-aminoundecanoic acid	**4b**	-	-	14.57	1
	12	12-aminododecanoic acid	**5b**	Sigma Aldrich	95	16.01	1
	13	16-aminohexadecanoic acid	**6b**	-	-	18.13	1
	14	12-aminododecanoic acid methyl ester	**7b**	-	-	17.33	2
α, ω-diols	15	1,8-octanediol	**1c**	Sigma Aldrich	98	11.76	2
	16	1,10-decanediol	**2c**	Sigma Aldrich	98	14.73	2
	17	1,12-dodecanediol	**3c**	Tokyo Chemical Industry	>97	17.08	2
	18	1,14-tetradecanediol	**4c**	Sigma Aldrich	97	19.19	2
	19	1,16-hexadecanediol	**5c**	Tokyo Chemical Industry	>95	22.12	2
α, ω-diamines	20	1,8-diaminooctane	**1d**	-	98	11.35	2
	21	1,10-diaminodecane	**2d**	Sigma Aldrich	97	13.24	2
	22	1,12-diaminododecane	**3d**	Sigma Aldrich	98	15.6	2
	23	1,14-diaminotetradecane	**4d**	-	-	18.77	2
	24	1,16-diaminohexadecane	**5d**	-	-	21.17	2
ω-amino-α-alcohol	25	8-amino-1-octanol	**1e**	Tokyo Chemical Industry	>98	13.12	2
	26	10-amino-1-decanol	**2e**	Tokyo Chemical Industry	>98	14.53	2
	27	12-amino-1-dodecanol	**3e**	Tokyo Chemical Industry	>98	16.84	2
	28	14-amino-1-tetradecanol	**4e**	-	-	18.97	2
	29	16-amino-1-hexadecanol	**5e**	-	-	21.69	2

Analytical Conditions:

Derivatization

For derivatization, reaction mixture was evaporated, 10 volumes of pyridine as a solvent was added. This mixture was subjected to sonication. 10 volumes of the derivatization solution (N-Methyl-N-(trimethylsilyl) trifluoro-acetamide) was added, and incubated at 50 °C for 30 min.

Determination of conversion

Conversion of ω- amino fatty acids and α, ω-Diamines were measured by using GC and GC-MS.

Column: Agilent J&W HP-5 column (30.0 m, 320mm, 0.25 mm);

GC program parameters:

Method 1: (For ω- amino fatty acids):

Injector 230°C; flow 1.5 mL/min; Temperature program 90°C/hold 0 min; 15°C per min to 200 °C/ hold 0 min and 5°C per min to 280°C / hold 5 min.

Method 2: (α, ω-diamines, ω- amino fatty acid methyl ester)

Injector 260°C; flow 1.5 mL/min; Temperature program 50°C/hold 1 min. and 10°C per min to 250 °C / hold 0 min and 30°C per min to 280 °C / hold 5 min.

Characterization of ω- amino fatty acids and α, ω-diamines

After derivatization of the compounds, the biotransformations were characterized by GC-MS. The corresponding results are described in below figures

GC-MS spectra

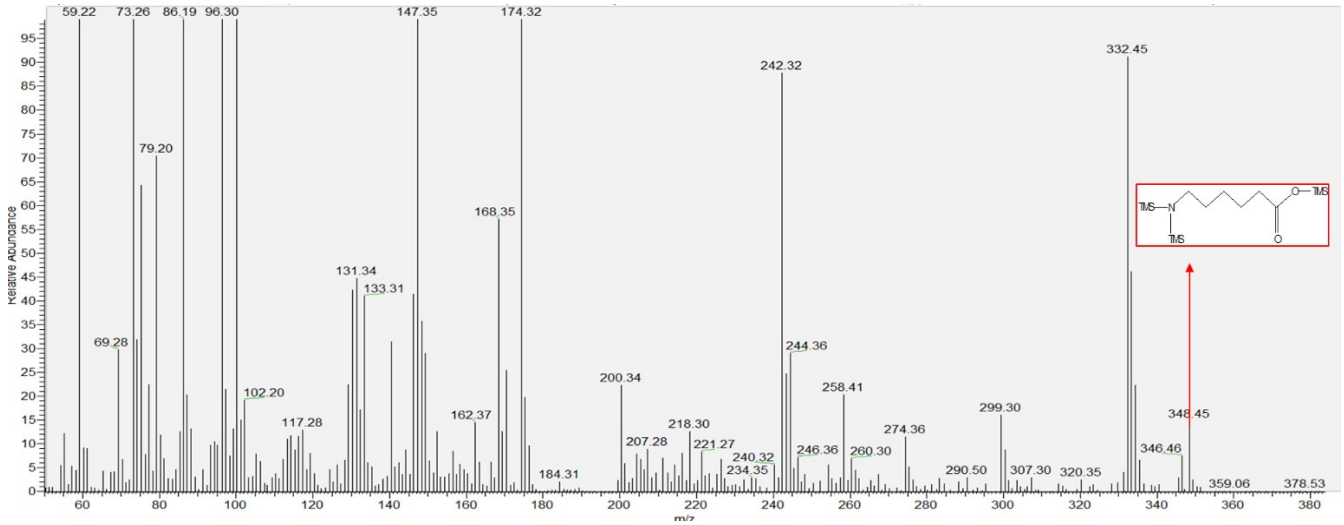


Figure S19. Mass spectrum of 6-aminohexanoic acid

Figure S20. Mass spectrum of 8-aminooctanoic acid

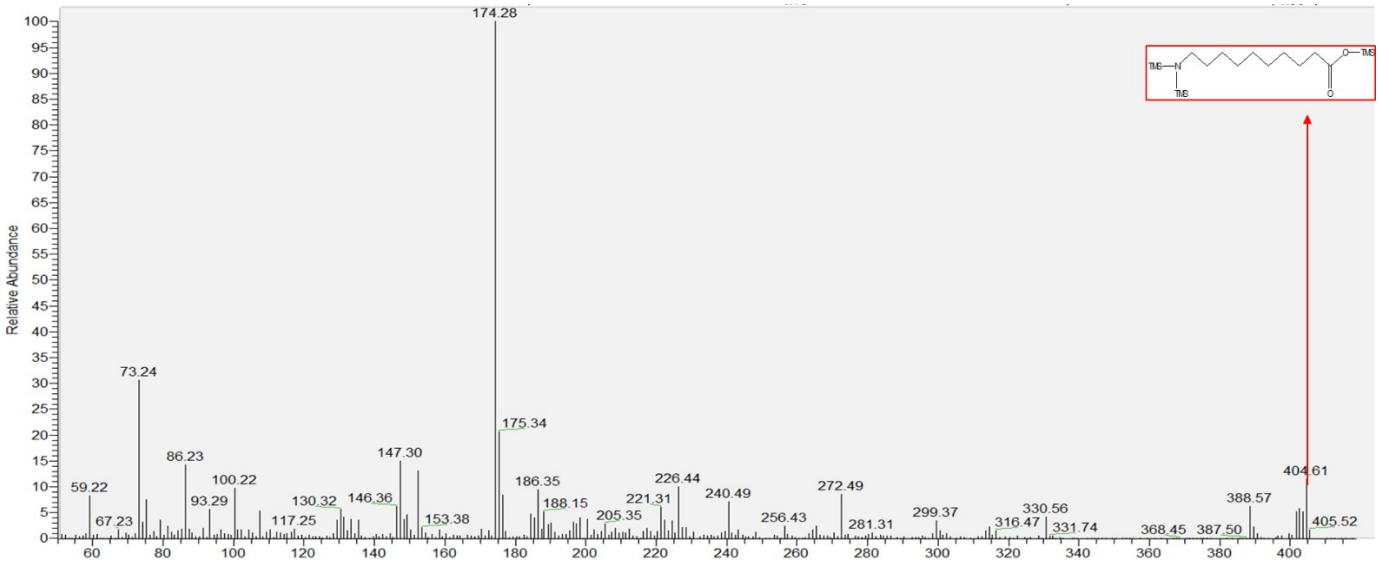


Figure S21. Mass spectrum of 10-aminodecanoic acid

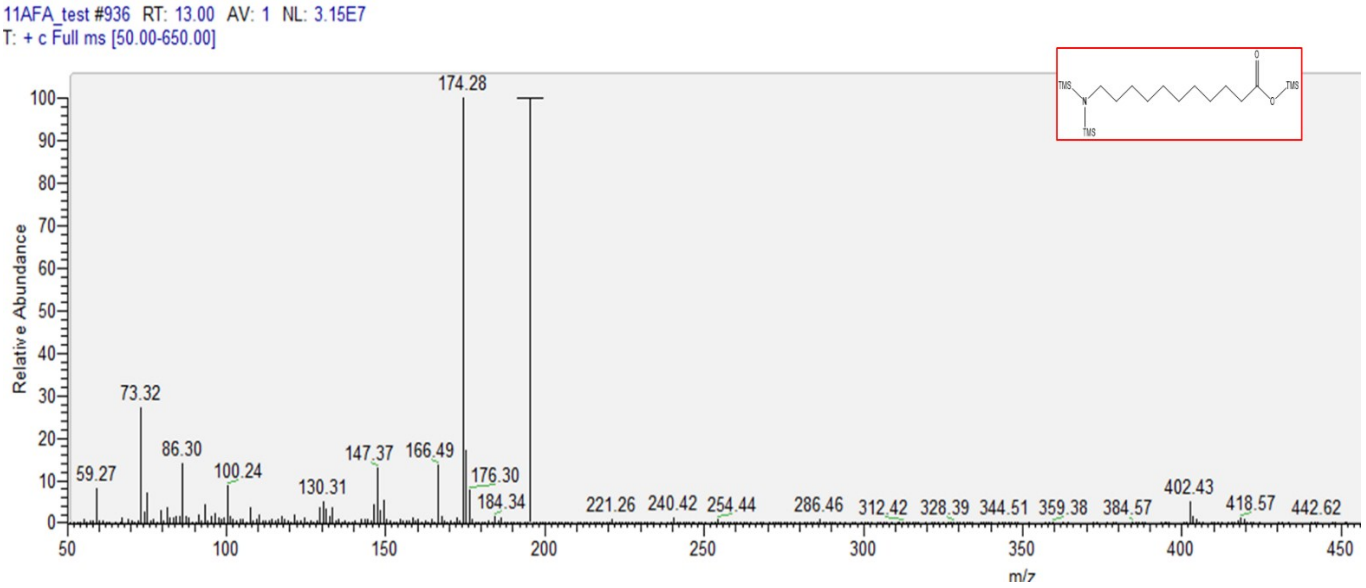


Figure S22. Mass spectrum of 11-aminoundecanoic acid

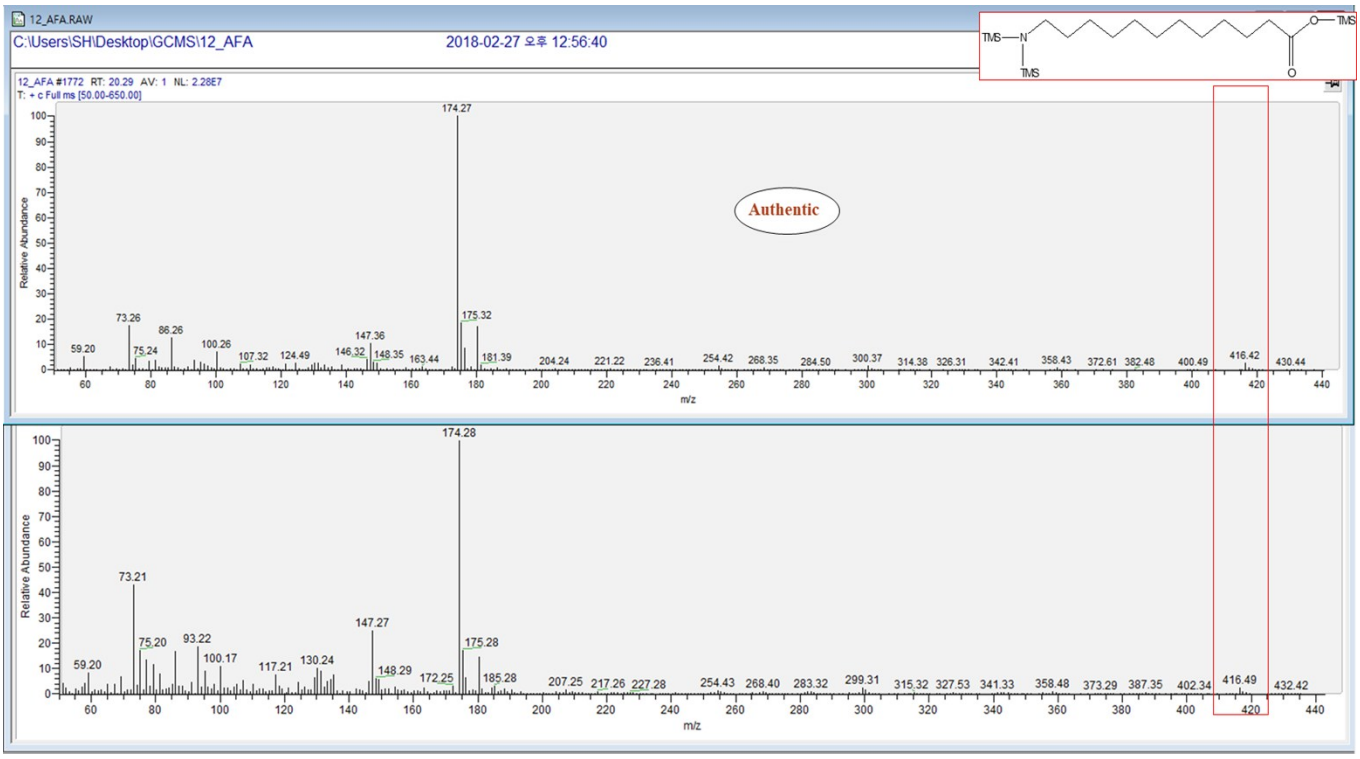


Figure S23. Mass spectrum of 12-aminododecanoic acid

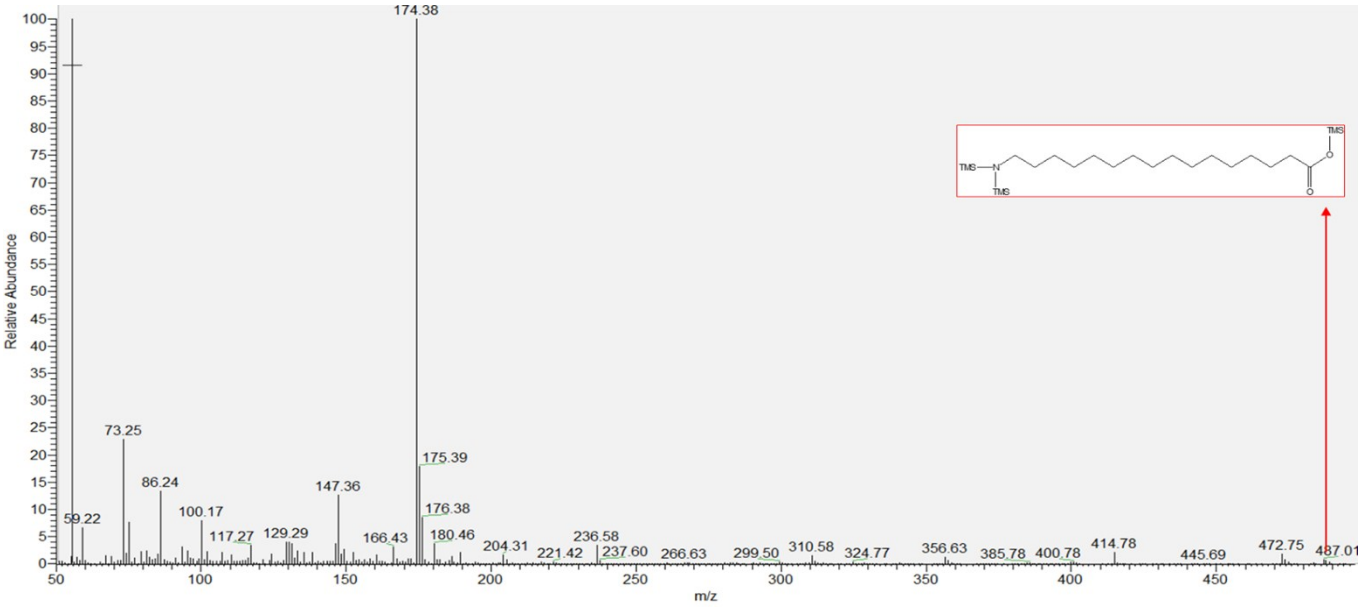


Figure S24. Mass spectrum of 16-amino hexadecanoic acid

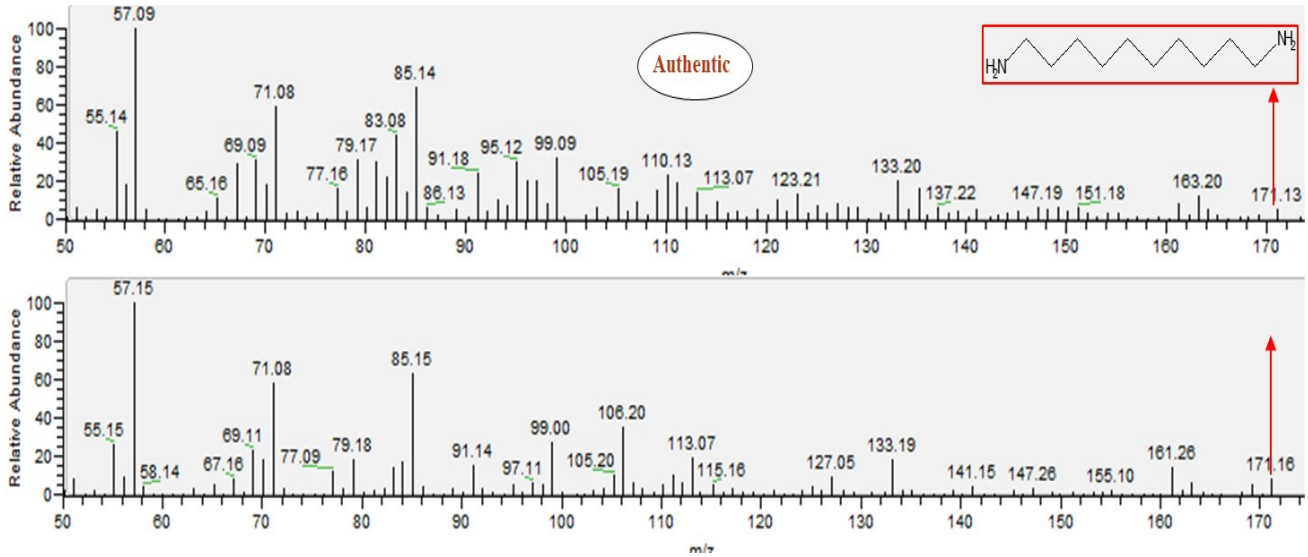


Figure S25. Mass spectrum of 1, 10-diaminodecane

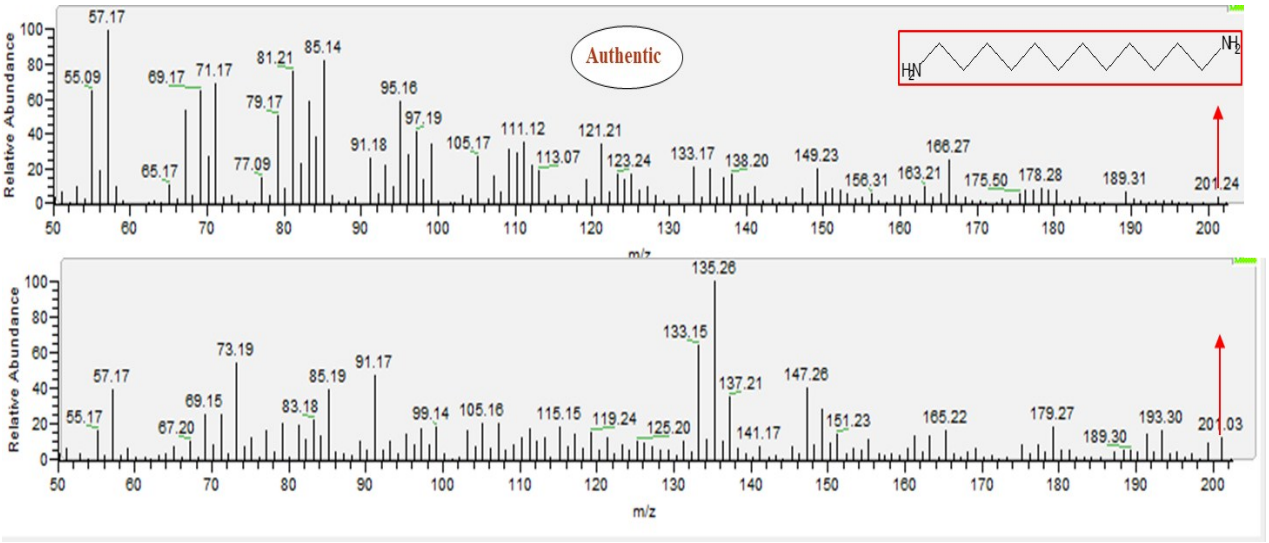


Figure S26. Mass spectrum of 1, 12-diaminododecane

References

[1] a) M.K. Akhtar, N.J. Turner, P.R. Jones. *Proc. Natl. Acad. Sci*. USA 2013, *110*, 87-92; b) R. Vidal, L. López-Maury, M.G. Guerrero, F.J. Florencio. *J. Bacteriol*. 2009, *191*, 4383-91

[2] a) M. M. Ahsan, H. Jeon, P. S. Nadarajan., C Taeowan, H. W. Yoo, B.G. Kim, M. D. Patil, H. Yun, *Biotechnol. J*. 2017. DOI: 10.1002/biot.201700562; b) M. M. Ahsan, S Sung, H. Jeon, M. D. Patil, T. Chung, H. Yun, *Catalysts.* 2017, *8*, 4.