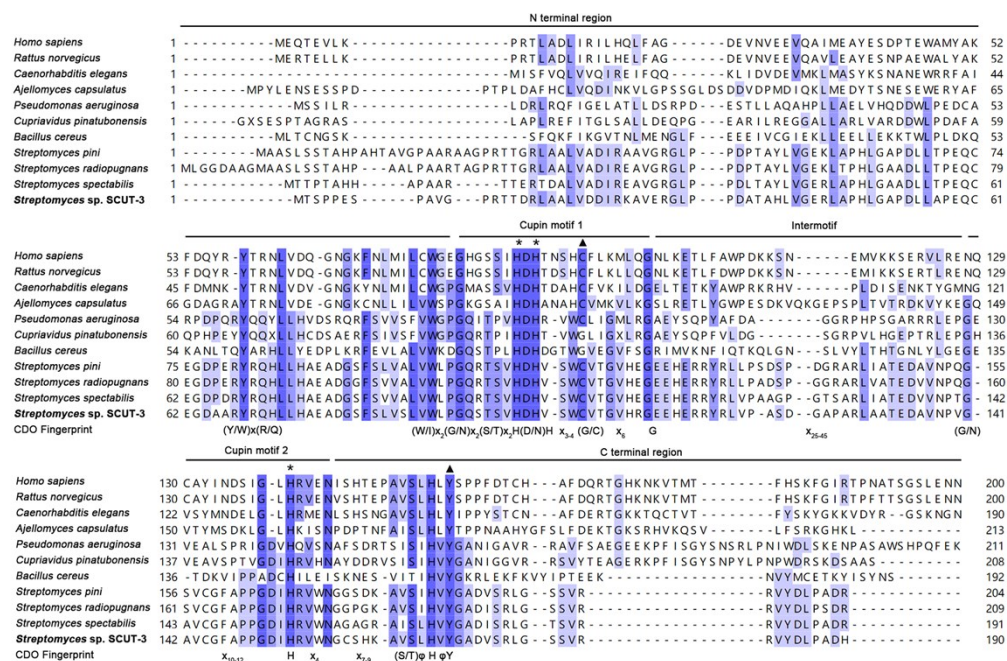
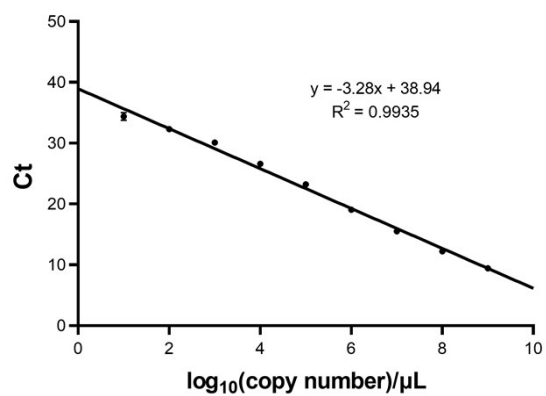
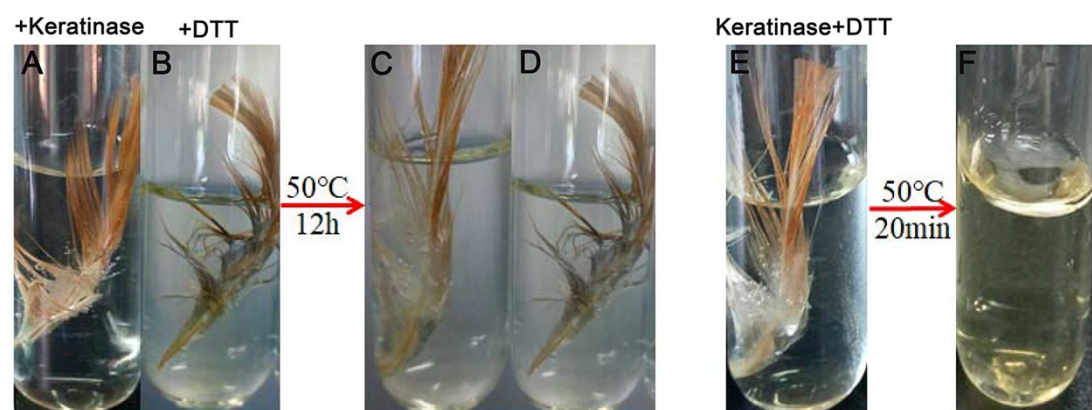




Supplementary Figure 1. Multiple sequence alignment of CDOs. The sequences are colored with percentage identity. Active site residues in CDOs known to chelate iron are marked as “*”. The residues that are able to form a cross-linked amino acid cofactor are marked as “▲”. Hydrophobic residues are indicated by “⊙”. Conserved CDO family fingerprint was listed under the corresponding base. From top to bottom, the sequences are from *Homo sapiens* (UniProt entry: Q16878), *Rattus norvegicus* (UniProt entry: P21816), *Caenorhabditis elegans* (UniProt entry: Q20893), *Ajellomyces capsulatus* (UniProt entry: Q5RLY7), *Pseudomonas aeruginosa* (UniProt entry: A0A140UH61), *Cupriavidus pinatubonensis* (UniProt entry: Q46R41), *Bacillus cereus* (UniProt entry: Q81CX4), *Streptomyces pini* (UniProt entry: A0A113XHB0), *Streptomyces radiopugnans* (UniProt entry: A0A1H9DPU4), *Streptomyces spectabilis* (UniProt entry: A0A516RDT4) and CDO1 from *Streptomyces* sp. SCUT-3 (UniProt entry: A0A2N5XEW9) (bold).



Supplementary Figure 2. *Streptomyces* sp. SCUT-3 quantitative real-time PCR standard curve using plasmids of *bla* gene. Error bars are one standard deviation from the mean.



Supplementary Figure 3. Identification the role of keratinase and reducing power in feather degradation. (A, C) Feather incubated with keratinase KerK (200U/mL) before and after 12 h, (B, D) Feather incubated with DTT (10 mM) before and after 12 h, (E, F) Feather incubated with KerK and DTT before and after 20 min.

Supplementary Table 1. Comparison of liquid fermentation and solid-state fermentation of SCUT-*Ocdo1-sep39* and wild type SCUT-3 processing 1g of feather waste

Type of fermentation	Liquid-state fermentation		Solid-state fermentation	
	5% CFM-day 2		40% CFM-day 6	
Strains	SCUT-3	SCUT- <i>Ocdo1-sep39</i>	SCUT-3	SCUT- <i>Ocdo1-sep39</i>
Recovered peptide (g)	0.09	0.13	0.12	0.20
Recovered amino acid (g)	0.13	0.19	0.26	0.29
Recovery rate	25.9%	37.6%	44.7%	57.6%

Supplementary Table 2. Inventory of procedures in different recycling methods to process 6,500 tonnes feather waste yearly in Guangzhou.

Process	Agent	Feather Pretreatment			Feather treatment		Product preparation	
Puffing method (PM)	Procedure	Clean		Dry		Puffing		-
	Chemicals/enzymes	-		-		-		-
	Reaction conditions	195,000 m ³ Water; 25°C/1 atm		60 °C/1 atm/48 h		200-230°C/9.87-14.80 atm		-
	Energy exhausted	-		48,000 kWh		2,437,500 kWh		-
	Waste water	195,000 tonnes		-		-		-
	CO ₂ emissions	-		7,180 kg		364,650 kg		-
	Valuable products	-		-		6,500 tonnes puffed feather meal (3,008,850 USD)		-
High-density steam flash-explosion-assisted protease (HDSFP) ¹	Procedure	Clean		Dry		HDSF	Enzymatic hydrolysis	Dry
	Chemicals/enzymes	-		-		-	1,300 tonnes protease (1,300,000 USD)	-
	Reaction conditions	195,000 m ³ Water; 25°C/1 atm		60 °C/1 atm/48 h		15.79 atm/1 min	650,000 m ³ Water; 50°C/1 atm/140 min	100°C/1 atm/10 min
	Energy exhausted	-		48,000 kWh		1,334,667 kWh	18,958,333 kWh	48,000 kWh
	Waste water	195,000 tonnes		-		-	-	-
	CO ₂ emissions	-		7,180 kg		199,666 kg	2,836,167 kg	7180 kg
	Valuable products	-		-		-	-	6,500 tonnes feather meal (10,725,000 USD)
<i>B. licheniformis</i> with <i>S. maltophilia</i> co-fermentation (BSC) ²	Procedure	Clean	Dry	Sterilization		Fermentation		Dry
	Chemicals/enzymes	-	-	-		195 tonnes yeast extract (962,814 USD) 390 tonnes glucose (108,316 USD) 91 tonnes KH ₂ PO ₄ (84,246 USD) 52 tonnes K ₂ HPO ₄ (40,117 USD) 65 tonnes NaCl (5,015 USD) 13 tonnes MgSO ₄ (1,103 USD)		-
	Reaction conditions	195,000 m ³ Water; 25°C/1 atm	65°C/1 atm/24 h	130,000 m ³ Water; 121°C/1 atm/ 15 min		37°C /1atm/12 h, 30°C 1atm/36 h		100°C/1 atm

	Energy exhausted	-	24,000 kWh	14,560,000 kwh	2,881,667 kWh	11,375,000 kWh
	Waste water	195,000 tonnes	-	-	-	-
	CO ₂ emissions	-	3,590 kg	2,178,176 kg	431,097 kg	1,701,700 kg
	Valuable products	-	-	-	-	6,500 tonnes feather meal (10,725,000 USD)
Reconstructed SCUT-3 solid-state fermentation (rSCUT-3 SSF)	Procedure		Sterilization		Fermentation	Dry
	Chemicals/enzymes		-	6.5 tonnes KH ₂ PO ₄ (6,017 USD)	4.9 tonnes K ₂ HPO ₄ (3,760 USD) 8.1 tonnes NaCl (627 USD)	-
	Reaction conditions	16,250 m ³ Water;	121°C/1 atm/ 30min		40°C/1 atm/6 d	100°C/1 atm
	Energy exhausted		1,820,000 kWh		284,375 kWh	1,421,875 kWh
	Waste water		-		-	-
	CO ₂ emissions		272,272 kg		42,543 kg	212,713 kg
	Valuable products		-		-	6,500 tonnes feather meal (10,725,000 USD)

Supplementary Table 3. Strains and plasmids used in this study.

Strain or plasmid	Description	Source or reference
<i>Streptomyces</i> sp.		
SCUT-3	Parent strain; isolated feather piles up soil in Shaoguan (Guangdong, China)	This laboratory ³
SCUT-3-pSET152	SCUT-3 integrated with pSET152	This study
SCUT-3- <i>Ocd01</i>	SCUT-3 integrated with pSET152- <i>cdo1</i>	This study
SCUT-3- <i>Osep39</i>	SCUT-3 integrated with pSET152- <i>sep39</i>	This study
SCUT-3- <i>Ocd01-sep39</i>	SCUT-3 integrated with pSET152- <i>sep39</i>	This study
SCUT-3- pSET-dCas9	SCUT-3 integrated with pSET-dCas9	This study
SCUT-3- <i>Dcd01</i>	SCUT-3 integrated with pSET-dCas9- <i>cdo1</i>	This study
<i>Escherichia coli</i>		
DH5 α	Cloning host	Takara, China
ET12567(pUZ8002)	Methylation defective; cloning host for conjugal transfer of DNA from <i>E. coli</i> to <i>Streptomyces</i>	MacNeil, et al. ⁴
Plasmids		
pSET152	<i>Streptomyces</i> integration vector, containing ϕ C31 int, <i>attP</i> , <i>oriT</i> of RK2, <i>aac (3) IV</i> and constitutive promoter <i>PerME*</i>	Bierman ⁵
pSET152- <i>cdo1</i>	<i>cdo1</i> inserted into <i>EcoRI/NdeI</i> sites of pSET152, under the control of the constitutive promoter <i>PerME*</i> , Apr ^r	This study
pSET152- <i>sep39</i>	<i>sep39</i> inserted into <i>EcoRI/NdeI</i> sites of pSET152, under the control of the constitutive promoter <i>PerME*</i> , Apr ^r	This laboratory ³
pSET152- <i>Ocd01-sep39</i>	The DNA fragment <i>PerME*-sep39</i> inserted into the <i>NdeI</i> sites of pSET152- <i>cdo1</i>	This study
pSET-dCas9- <i>actII4</i>	pSET152 containing dCas9, under the control of the constitutive promoter; <i>actII4</i> sgRNA, under the control of the promoter <i>j223119 PerME*</i> , Apr ^r	Zhao, et al. ⁶
pSET-dCas9- <i>cdo1</i>	pSET-dCas9 containing <i>cdo1</i> sgRNA, under the control of the promoter <i>j223119</i> , Apr ^r	This study

Gene	Primers used for gene amplification (5'-3')
<i>cdo1</i> -Gibson-F	TAGGTATAATACTAGTGGACGACATACGCAAGGCCGTTTTAGAGCTAGAAATA (Specific N ₂₀ target sequence of <i>cdo1</i> is underlined)
<i>gRNA</i> -R	GCTATGACATGATTACG
<i>cdo1</i> -F	TTTACACATATGACTTCCCCGCCGAATCACC (<i>Nde</i> I restriction site is underlined)
<i>cdo1</i> -R	GAGCGAGAATTCTCAGTGGTCGGCGGGCAG (<i>Eco</i> RI restriction site is underlined)
<i>sep39</i> -F-2	CCGCCGACCACTGAGCTAGTATGCATGCGAG
<i>sep39</i> -R-2	ACAGCTATGACATGATTACGTCAGAGGCCGGACTTGAAC
16S rRNA-F	ACGGGCAGGCTAGAGTTCGGT
16S rRNA-R	GCTCCTCAGCGTCAGTATCGG
<i>bla</i> -qpcr-F	ACGCACGGATGAAGGAGT
<i>bla</i> -qpcr-R	TCAGCACGTTGGTGATG
<i>cdo1</i> -qpcr-F	GCACCTGGTCGGCGAGAGA
<i>cdo1</i> -qpcr-R	CGAGGGAGACGAGGGAGAA
<i>sep39</i> -qpcr-F	GGCAACGACAACCAGG
<i>sep39</i> -qpcr-R	TTGCTGAACGACGCCT

Supplementary Table 4. Primers used in this study.

Supplementary Table 5. The unit price of materials (Guangzhou local market) used in four feather treatment processes.

Materials	Unit price (USD/t)
Protease	1,000
Yeast extract	4,937.5
Glucose	277.7
KH ₂ PO ₄	925.8
K ₂ HPO ₄	771.5
NaCl	77.1
MgSO ₄	84.9
PM feather meal	462.9
HDSFP feather meal	1,650
BSC feather meal	1,650
rSCUT-3 SSF feather meal	1,650

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