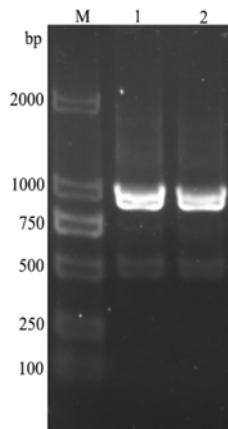


M G S S	H H H H H H	S S G L V P ↓ R G S H M
ATGGGCAGCAGCCATCATCATCATCACAGCAGCGGCTGGTGCAGCGCAGCCATATG		
Protein	K T Y E A C E L A R E L V D K G A A M D Q V G T W V C I A K	
Original 1	AAGACTTACGAGGCCCTGCGAGCTCGCCGGAGCTGGTGGACAAGGGGGCGCGATGGACCAGGTGGCACCTGGGCTGCGATCGCAAG	
Optimized	AAGAC <b>C</b> TATGAAG <b>C</b> TGTGA <b>A</b> CTGG <b>C</b> AC <b>G</b> TAAAG <b>G</b> CG <b>C</b> ATGG <b>A</b> T <b>C</b> AG <b>G</b> TTGG <b>T</b> AC <b>T</b> GG <b>T</b> TT <b>G</b> TATT <b>G</b> CAAA	
Protein	Y E S D F N T S A V G R L N G D G S A D H G I F Q I S D R Y	
Original 91	TACGAGTCCGACTTCACACCTCCGCGTGGGAGGTTGAACGGGACGGCAGCGCCGACCCGGCATCTCCAGATCAGCGACCCCTAC	
Optimized	TAT <b>G</b> AAAG <b>C</b> GATTCAAC <b>A</b> CC <b>A</b> GG <b>C</b> CG <b>T</b> TGG <b>C</b> CG <b>C</b> CTGA <b>A</b> AT <b>G</b> GC <b>A</b> T <b>G</b> GT <b>A</b> GG <b>T</b> AT <b>T</b> TCAG <b>A</b> TT <b>G</b> CG <b>A</b> T <b>C</b> C <b>T</b> AT	
Protein	W C S P Q G W A C N I P C D L L E D D D I S D D W K C A K R	
Original 181	TGGTGCTCCCCCAGGGATGGCCTGCAACATACCCCTGCACCTCTGGAGGACGACGACATCTCGACGACTGGAAGTGCGCCAAGAGG	
Optimized	TGG <b>T</b> GTAG <b>T</b> CCG <b>C</b> AGGG <b>T</b> GG <b>G</b> CAT <b>G</b> TAA <b>T</b> AT <b>T</b> CC <b>G</b> TGC <b>G</b> AT <b>T</b> CT <b>G</b> GA <b>A</b> GT <b>T</b> AT <b>T</b> AG <b>G</b> AT <b>G</b> TGG <b>A</b> AT <b>T</b> GT <b>G</b> AAA <b>C</b> G	
Protein	I F R Q H K V L S G N G F R A W A V Y S S R C S G D N S G Y	
Original 271	ATATTCAAGCAGCACAAAGTCTCAGCGGAATGGTTCAAGGGCTGGCGCTACTCTCCAGGTGTCGGGTGATAACAGTGGTAT	
Optimized	ATTTTC <b>G</b> T <b>C</b> AG <b>C</b> ATA <b>A</b> AG <b>T</b> CT <b>G</b> AG <b>C</b> GG <b>A</b> AT <b>G</b> CT <b>T</b> TC <b>G</b> CG <b>C</b> AT <b>G</b> GG <b>C</b> GT <b>T</b> T <b>A</b> T <b>G</b> C <b>A</b> GT <b>C</b> G <b>T</b> GTAG <b>T</b> GG <b>G</b> ATA <b>A</b> T <b>G</b> CG <b>T</b> TAT	
Protein	V E G C F D S K A S V S Q S N T T V I T L P A F T T S T T T	
Original 361	GTGGAGGGTGTGGAGCTAAGGCTCTGTTACAGTCAACACCACAGTCATTACACTGCCCTGCCTCACAACTCGACCCACA	
Optimized	GTGG <b>A</b> AG <b>G</b> IT <b>G</b> CT <b>T</b> GA <b>T</b> AG <b>C</b> AA <b>G</b> CA <b>A</b> GT <b>G</b> TA <b>G</b> CC <b>A</b> GA <b>C</b> AA <b>I</b> AC <b>C</b> AC <b>C</b> GT <b>A</b> TT <b>A</b> CC <b>C</b> IG <b>C</b> CG <b>C</b> CT <b>T</b> AC <b>C</b> AC <b>C</b> AG <b>T</b> AC <b>C</b> AC <b>C</b> AC	
Protein	S T S T T T E P E R V E G S L S A R E R N N A E K V Q V I	
Original 451	TCCACTTCTACCACAACTGAAGAACCTGAGCGTGTGGAGGATCCCTAGTGCCCAGAACGAAACATGCAGAGAAAGTACAAGTTATA	
Optimized	<b>A</b> GC <b>A</b> CC <b>A</b> GC <b>A</b> CC <b>A</b> CC <b>A</b> CC <b>A</b> GA <b>A</b> AC <b>C</b> GA <b>A</b> CC <b>G</b> TA <b>G</b> CT <b>G</b> AG <b>T</b> GC <b>A</b> CG <b>C</b> GA <b>A</b> TA <b>A</b> T <b>G</b> CAG <b>A</b> AA <b>A</b> GT <b>T</b> GC <b>A</b> GG <b>T</b> TAT	
Protein	D L E M M E K K G K S F V N V A S S S K K K W K P W T W E I	
Original 541	GATCTAGAAATGATGGAAAAGAAGGCAAGTCATTGTAATGTGGCATCCTCATAAAAAGAAATGGAAACCATGGACTGGGAGATA	
Optimized	GAT <b>C</b> TGG <b>A</b> AT <b>G</b> TGG <b>A</b> AA <b>G</b> GA <b>A</b> AG <b>G</b> TAA <b>A</b> AG <b>C</b> TT <b>G</b> TT <b>A</b> AG <b>T</b> GG <b>C</b> AG <b>C</b> AG <b>C</b> AAA <b>A</b> GA <b>A</b> AT <b>G</b> GA <b>A</b> CC <b>G</b> TGG <b>A</b> CT <b>G</b> GG <b>A</b> ATT	
*		
TAAGAATT <sub>C</sub>		

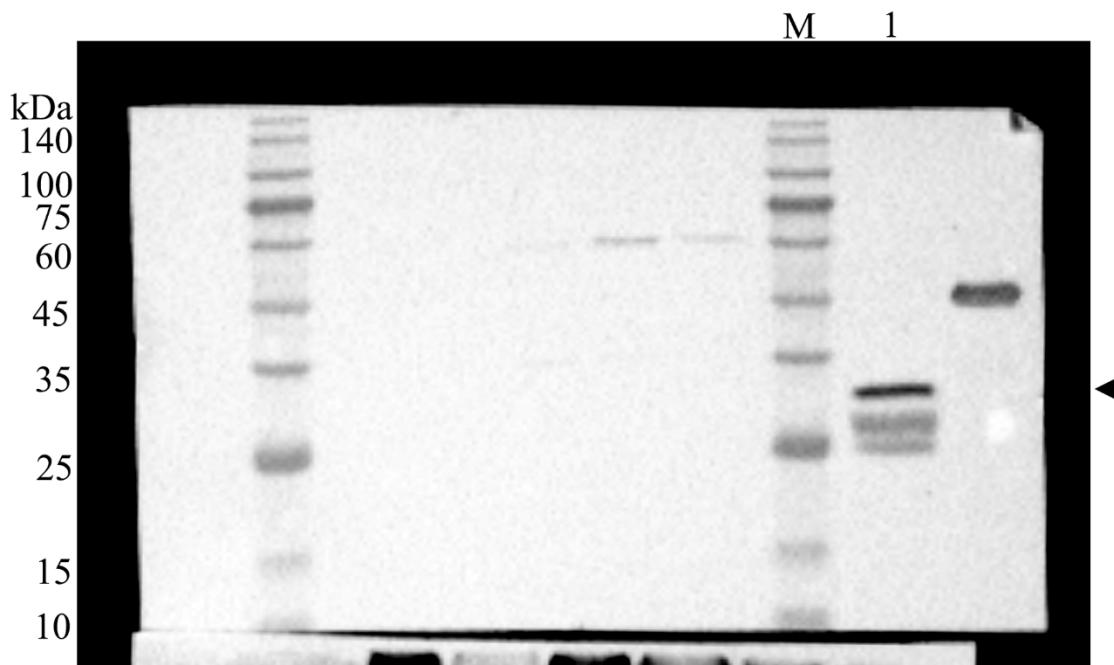
**Figure S1.** Optimized nucleotide and amino acid sequences of the mature CcLys2. The total length of the optimized gene is 633 bp, and red marks the mutated bases. The recognition sites for *Nde* I and *Eco* R I are underlined, and the red arrow marks the thrombin cleavage site. The N-terminal of the amino acid sequence contains a 6×His-tag. The asterisk marks the stop codon.



**Figure S2.** Identification of the recombinant expression vector by PCR. M: DL2000 DNA marker; Lanes 1 and 2: the *CcLys2* gene with a part of the pET-28b(+) vector sequence.

**Table S1.** Species name, protein name and GenBank accession number for multiple sequence alignment and phylogenetic analysis.

Protein name	Species name	GenBank accession number
CcLys2	<i>Coridius chinensis</i>	QPB70596
CcLLP3	<i>Coridius chinensis</i>	MW250790
HEWL	<i>Gallus gallus domesticus</i>	1LSZ_A
HLYZ1	<i>Homo sapiens</i>	NP_000230
PsLys	<i>Plautia stali</i>	BBE08150
HhLys	<i>Halyomorpha halys</i>	KAE8573139
TcLys1	<i>Tribolium castaneum</i>	ACV32411
TcLys2	<i>Tribolium castaneum</i>	ACV32412
HaLLP1	<i>Harmonia axyridis</i>	AIZ72680
HaLLP2	<i>Harmonia axyridis</i>	AIZ72679
HaLys3	<i>Harmonia axyridis</i>	AIZ72681
HaLys4	<i>Harmonia axyridis</i>	AIZ72682
AgLYSC1	<i>Anopheles gambiae</i>	Q17005
AgLys c-6	<i>Anopheles gambiae</i>	AAV21238
AgLys c-8	<i>Anopheles gambiae</i>	AAV21241
CqLys	<i>Culex quinquefasciatus</i>	XP_001843429
FoLys	<i>Frankliniella occidentalis</i>	KAE8737425
NIlys	<i>Nilaparvata lugens</i>	AGK40894
BmLZ	<i>Bombyx mori</i>	NP_001037448
BLLP1	<i>Bombyx mori</i>	ABP52099
ALLP1	<i>Antheraea mylitta</i>	ABP52098
CfLys3	<i>Coptotermes formosanus</i>	AFZ78838
SeLys3	<i>Spodoptera exigua</i>	AKJ54522
OfLys5	<i>Ostrinia furnacalis</i>	AGV28584



**Figure S3.** M: Protein molecular weight marker (10-140 kDa). Lane 1: blotting band of fusion CcLys2. The arrow indicates the band of fusion CcLys2.