

A novel, robust peptidyl-lys metalloendopeptidase from *Trametes coccinea* recombinantly expressed in *Komagataella phaffii*

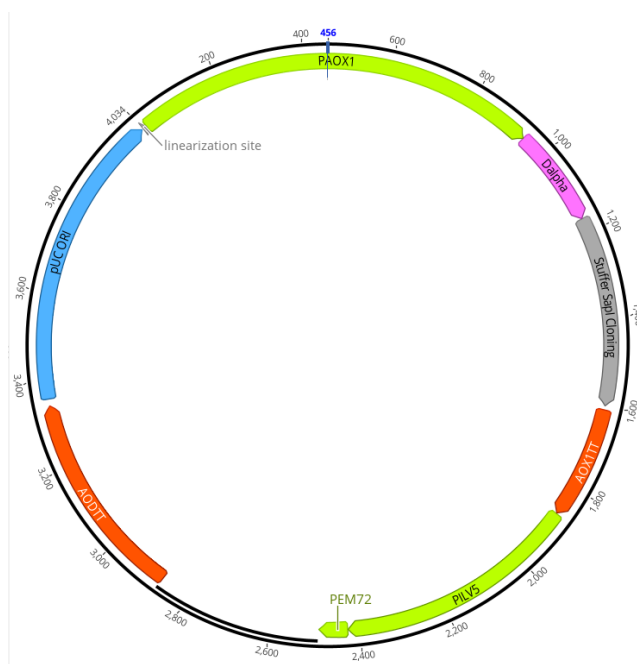
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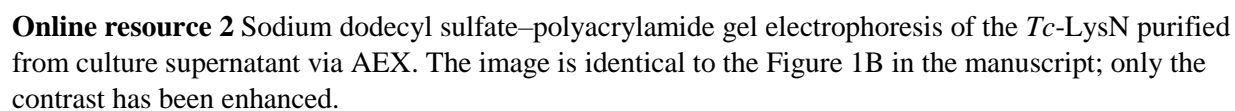
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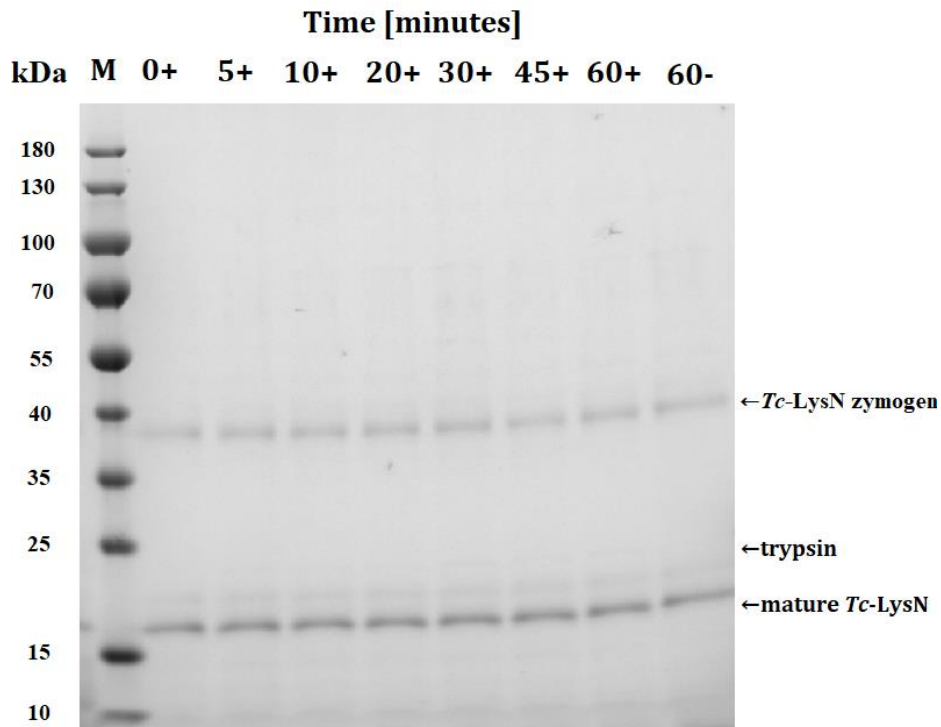


Online resource 1 Plasmid map of pBSY2S1Z.



MFTISTVRATL IALAASAVAV SAERALSLKLV SGPEAVDGVQ NFKVTTTTLTN TGDESLKLLN DPRGALNTLP ANTFDIT TDS
GEKPSFIVGK VKY VPRHAAQ STNEKSFVTL AGPESTSVTH DLSAAAYNFTL PGAAGYTVEP ANLFHYVDPE TNEPVEIRAD
SEPHVAAVSG KLVAVRTPPT LTKRETYNGK SSSQKTGIOA AAPAAQTYVA NALSYLQTHT ASTARYTTFW GSYTSSRHST
VLSHYTNNNG NDYSSEFYDC TCTESDTHAY VYPDDFGYIY LCGAFWQAPT TGTDS KGGTL VHESSHFTRN AGTQDYVYGQ
SAAKSLAKSN PSEAI MNADN HEYFAENNPA OS

Online resource 3 *Tc*-LysN sequence coverage of TMT-labeling experiment. E185 is the N-terminal TMT-labeled amino acid.



Online resource 4 Sodium dodecyl sulfate–polyacrylamide gel electrophoresis of the concentrated *Tc-LysN* culture supernatant (24 hr induction) that was incubated with trypsin for 60 minutes. Samples were withdrawn at indicated time points and mixed with reducing sample loading at heated for 5 minutes at 90°C before being loaded onto the gel. The “+” indicates samples that were incubated with trypsin and the “-” sign indicates samples that weren’t incubated with trypsin. Lane M contains the marker proteins (PageRuler Prestained 10–180 kDa, Thermofisher).

Albumin OS=Bos taurus OX=9913 GN=ALB PE=1 SV=4
78 exclusive unique peptides, 99 exclusive unique spectra, 205 total spectra, 507/607 amino acids (84% coverage)

MKWVTFISLL	LLFSSAYSRG	VFRRDTHKSE	IAHRFKDLGE	EHFKGLVLIA	FSQYLQCCPF	DEHVKLVLNEL	TEFAKT	C VAD
ESHAGCEKSL	HTLFGDELCK	VASLRETYGD	MADCCCEKQEP	ERNECFLLSHK	DDSPDLPLKLL	PDPTNLCDEF	KADEKK	FWGK
YLYEIAARRHP	YFYAPELLLY	ANKYNGVFQEH	CCQAECKGAC	LLPKIETMBE	KVLASSARQR	LRCAISIQKFG	ERALKAWSVA	
RLSQKFPKAE	FVEVTKLVTYD	LTKVHKECCH	GDLLLECAADR	ADLAKYICDN	QDTISSKLKE	CCDKPLLEKS	HCIAEVEKDA	
IPENLPPLTA	DFAEDKDVCK	NYQEAQDAFL	GSFLYEYSRR	HPEYAVSVLL	RLAKEYEATL	EECCAKDDPH	ACYSTVFQDL	
KHLVDEPQNL	IKQNCDDQFEK	LGEYGFQNAL	IVRYTRKVPO	VSTPTLVEVS	RSLGKVGTRC	CTKPESERMP	CTEDYLSLIL	
NRLCVLHEKT	PVSEKVTKCC	TESLVNRRPC	FSALTPDETY	VPKAFDEKLF	TFHADICTLP	DTEKQIKKQT	ALVELLKHKP	
KATEEQLKTV	MENFVAFVDK	CCAADDKEAC	FAVEGPKLVV	STQTALA				

Online resource 5 Peptide sequence coverage of the in-solution digest of bovine serum albumin with *Tc-LysN*.