The Ketosynthase Domain Constrains the Design of Polyketide Synthases

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	110	120	130	140	150	160	170	180
DEBSKS3	DPHSLRGTAT	G V F L <mark>G</mark> V A K F G	YG E DT A - #	AEDV <mark>EGY</mark> S	VTGVAPAVAS	GRISYTMGL	EGPSISVDTACS	SL VAL HL AVE SL
DEBSKS1	PPTSLOASPT	GVEVGLIPQE	G PRLAEG	GEGVEGYL PEDGVDGYL	I TONTASVAS	GRIAYTLGI.	EGPAISVDTACSS	
DEBS KS4	DPDALEGTOT	GVEVGMNGQS	MQLLAGE	AERVDGYQ	GLGNSASVLS	GRIAYTEG	EGPALTVDTACS	LVGIHLAMQAL
DEBS KS5	DPVSLRGSAT	G V I T <mark>G</mark> V G T V D	YG PRPDE	A P D E V L <mark>G Y</mark> V	G TG T A S S V A S	GRVAYCLGI	EGP A MT V DT A C S S C	ITAL HLAMESL
MYCI AKS	DPHSLRGSDT	GVELGAAYQG	GQDAVVF	PED-SEGYL	LTGNSSAVVS	GRVAYVLGL	EGPAVTVDTACS	SLVAL HSACGSL
MYCL_AKS	5 PAHTLAGTST	GVEVGAGAQS	G ATN	SDDAEGYA	MTGGATSVMS	GRIAYTLGL	EGPAITVDTACS	SLVAIHLACQSL
MYCL_AKS	7 PAHT <mark>LA</mark> GTST	G V I A G A WAQ S	<mark>۲</mark> G A T M	NS D D A <mark>E G Y</mark> A	M <mark>TG</mark> GSTSVMS	GRIAYTLGI	EGPAITVDTACS	SL VA I HL A CQ SL
MYCL_AK9	PAHTLAGTST	GVEVGAWAQS	G ATN	IS DG A EGY A	MTGGSTSVMS	GRIAYTLGL	EGPAITVDTACS	SL VA I HL ACQSL
MYCL_BKS	5 PAHTLAGTST	GVEAGAWAOS	G ATN	SDDAEGYA	MTGGATSVMS	GRIATTLGL	EGPAITVDTACS	SLVAIHLACOSL
MYCL_BKS	7 PAHTLAGTST	GVFAGAWAQS	YG AT M	NS D D A <mark>E G Y</mark> A	MTGGATSVMS	GRIAYTLG	EGPAITVDTACS	SLVAIHLACQSL
PIKSKS1	DPTSLRGRQV	GVI TGAMTHE	GPSLRDGGE	GLD <mark>GY</mark> L	LTGNTASVMS	GRVSYTLGL	EGPALTVDTACS	LVALHLAVQAL
PIKSKS5	DPTALKGSGL	GVEVGGWHTG	TSGOTTAVO	SPELEGHL	VSGAALGFLS	GRIAYVLG	DGPALTVDTACS	SLVALHLAVOAL
PIKS KS6	DPASVRGTDV	G V Y V G C G Y Q D	APDIRVAPE	G TG <mark>GY</mark> V	VTGNSSAVAS	GRIAYSLGI	EGPAVTVDTACS	LVAL HLALKGL
RIFSKS1	DP IAL KGT DT	GVISGLMGQG	GSGA	APELEGEV	TTGVASSVAS	GRVSYVLGL	EGPAVTVDTACS	LVAMHLAAQAL
RIFSKS10	DPGSLKGADV	GVFAGVSNOG	GMGA	DPAELAGYA	STAGASSVVS	GRVSYVFGF	EGPAVTIDTACSS	LVAMHLAGOAL
0.000	190	200 \star	210	220	230	240	250 2	60
DEBSKS3	RKGESSMAVV	GGAAVMATPG			AFGAGADGFG	FSEGVILVL	LERLSEARRNGHEN	LAVVRGSAL
DEBSKS2	RDGDCGLAVA	GGVSVMAGPE	FTEFSROG		PESDEADGEG	LGEGSAFVV	LORLSDARREGRR	LGVVAGSAV
DEBSKS4	RRGECSLALA	GG V T <mark>V</mark> M S D P Y	FVDFSTQRO	ILASDGRCK	AF SAR ADGFA	LSEGVAALV	LEPLSRARANGHQ	/LAVLRGSAV
DEBSKS5	RRDECGLALA	GGVTVMSSPG	AFTEFRSQGO		PESKAADGEG	LAEGAGVLV	LQRLSAARREGRP	LAVLRGSAV
MYCL_AKS	1 RNNESOLALA	GGVTVMSTPA	FTDFSRORO	SLAP DGRCK	AFAATADGTG	WGEGAAVLV	LERLSEARRNNHP	LAIVAGSAI
MYCL_AKS	5 RNNESQLALA	GGVT <mark>V</mark> MSTPA	FTEFSRORO	G L A P DG R C K	AFAATADGTG	FGEGAAVLV	LERLSEARRNNHP	I A I VAGSA I
MYCL_AKS	7 RNNESQLALA	GGVT <mark>VM</mark> STPA	FTEFSRORO	SLAPDGRCK	AFAATADGTG	FGEGAAVLV	LERLSEARRNNHP	LAIVAGSAI
MYCL_BKS	1 RNNESOLALA	GGVTVMSTPA	FTEFSRORG		AFAATADGTG	WGEGAAVLV	LERLSEARRNNHP	LAIVAGSAI
MYCL_BKS	5 RNNESQLALA	GGVT <mark>V</mark> MSTPA	FTEFSRORO	I AP DGRCK	AFAATADGTG	FGEGAAVLV	LERLSEARRNNHP	LA IVAGSA I
MYCL_BKS	7 RNNESQLALA	GGVT <mark>V</mark> MSTPA	FTEFSRORO		AFAATADGTG	FGEGAAVLV	LERLSEARRNNHP	LAIVAGSAI
PIKSKS4	RKGECGLALA	GGVTVMSTPT	FVEFSRORG		AFAASADGFG	PAEGVGMLL	VERLSDARRNGHOV	LAVVRGSAL
PIKS KS5	RKGECDMALA	GG V T <mark>V M</mark> P N A D I	FVQFSRQRO	ILA A DG R S K	AFATSADGFG	PAEGAGVLL	VERLSDARRNGHRI	LAVVRGSAV
PIKSKS6 RIFSKS1	RNGDCSTALV	GGVA <mark>VLATP</mark> GA	AFIEFSSQQ#		GFASAADGLA	WGEGVAVLL	LERLSDARRKGHR	LAVVRGSAI
RIFSKS6	ROGECSMALA	GGATVMANPG	AFVEFSRORG	LAV DGRCK	AFAAAADGTG	WAEGVGVVI	LERLSVARERGHRI	LAVLRGSAV
RIFSKS10	RQGECSMALA	GGVT <mark>V</mark> MG TP G	FVEFAKORO	ELAGDGRCK	AYAEGADGTG	WAEGVGVVV	LERLSVARERGHRV	I A V L RG S A V
	270	280 2	90	300	310	320	330 3	40
DEBSKS3	NODGASNOLS	APSGPAORBV		FPGDVDAV	FAHGTGTALG	DP TEANAL L		
DEBSKS1	NSD <mark>GASNG</mark> LS	A P NG R A Q V R V	IQQALAESGL	GPADIDAV	EAHGTGTRLG	DP IEARAL F	EAYGRDR EQPLH	ILGSVK <mark>SNL</mark> G
DEBSKS2	NQ D <mark>G A S NG</mark> L S	A P S G V A Q Q R V	I R R <mark>A WA</mark> R <mark>A</mark> G I	TGADVAVV	EAHGTGTRLG	DPVEASALL	<mark>A T Y G</mark> K S R G S S G <mark>P</mark> V L	LGSVK <mark>SNI</mark> G
DEBSKSS	NODGASNGLA	A P NG P SQ E R V		RAGDVDVV	FAHGTGTELG	DP IEAGAL I	STYGAFROPDOPLV	
DEBSKS6	NQ D G A S NG L A	A P S G V A Q Q R V	I R K <mark>A WA</mark> R A G I	TGADVAVV	EAHGTGTRLG	DPVEASALL	A T Y G K S R G S S G P V L	LGSVKSNIG
MYCL_AKS	1 NQDGASNGLT.	A P HG P SQ Q R V	INQALANAGL	THDQVDAV	EAHGTGTTLG	DP IEAGALH	ATYGHHHTPDQPLV	I G S I K S N I G
MYCL_AKS	7 NODGASNGLT	APHGPSOORV	INOALANAGL	THOOVDAY	EAHGTGTTLG	DP IEAGALH	ATYGHHHTPDOPLV	VLGSIKSNIG
MYCL_A KS	9 NQDGASNGLT	A P HG P S Q Q R V	INQALANAGL	T H DQ V D A V	EAHGTGTTLG	DPIEASALH	A TYGHHHTPDQPLV	VLGSIK <mark>SNI</mark> G
MYCL_BKS	NQDGASNGLT.	A P HG P SQ Q R V	INQALANAGL	THDQVDAV	EAHGTGTTLG	DP IEASALH	ATYGHHHTPDQPLV	LGSIKSNIG
MYCL_BKS	NQDGASNGLT.	APHGPSQQRV	INQALANAGL	THDQVDAV	EAHGTGTTLG	DP IEASALH	ATYGHHHTPDQPLV	VLGSIKSNIG
PIKSKS1	NQ D G A S NG L T	A P NG P S Q Q R V	IRRALADARL	TTSDVDVV	EAHGTGTRLG	DP IEAQAL I	A T Y G Q G R D D E Q P L P	L G S L K <mark>S N I</mark> G
PIKS KS4 PIKS KS5	NODGASNGLT	A P NG P SQ Q R V		ARGDVDVV	EAHGTGTRLG	DP IEAQAL I	ATYGQGRDTEQPL P	LGALKSNIG
PIKS KS6	NQDGASNGLT	APHGPSQQHL	IRQALADARL	TSSDVDVV	EGHGTGTRLG	DP IEAQALL	ATYGQGRAPGQPLR	LGTLKSNIG
RIFSKS1	NQ D <mark>G A S NG</mark> L T	A P NG L S Q Q R V	IRR <mark>ALA</mark> AAGL	. A P S D V D V V	EAHGTGTTLG	DP IEAQALL	ATYGQER KQPLV	VLGSLK <mark>SNI</mark> G
RIFSKS0	NODGASNGLT.	A P NG P SQ Q R V	IRRALVSAGL	EPSDVDVV	EAHGTGTTLG	DP I EAQALL	ATYGKDR ESPLV	
DEDGLASS	350	360	370	380	390	400 ,	410 * 4	20
DEBSKS3 DEBSKS1	HTQAAAGVTG		GELPATLHVE	EPTPHVDW	SSGGVALLAG	NQPWR RG	ERTRRARVSAFG IS	SGTNAHVIVE
DEBSKS2	HAQAAAGVAG	VIKVLLGLER	GVVPPMLCRO	GERSGL IDV	ISSGEIELADG	VREWSP - AA	DGVRRAGVSAFGVS	GTNAHVIIA
DEBSKS4	HTQAAAGAAG	V I K V V L AMRH	GML P R S L H A I	DELSPHIDW	/ES <mark>G</mark> A <mark>V</mark> EVLRE	EVP <mark>WP</mark> AG	E R <mark>P R R A</mark> G V S S F G V S	5GTNAH <mark>VI</mark> VE
DEBSKS5	HTQAAAGVAG	VMKAVLALRH	GEMPRTLHF	DEPSPQIEW	DLGAVSVVSQ	ARSWP AG	ERPRRAGVSSFG IS	SGTNAHVIVE
MYCL_A KS	1 HTQAAAGAAG	VVKMIQAITH	ATLPATLHVI	QPSPHID	SGTVRLLTE	PIQWP NT	DHPRTAAVSSEGIS	GTNAHL ILQ
MYCL_AKS	HTQAAAGAAG	VVKM IQA ITH	ATLPATLHVI	DQ P S P H I DW	<mark>IS SG T V R L L</mark> T E	PIQWP NT	DHPRTAAVSSFG IS	SGTNAHL ILQ
MYCL_AKS	HTQAAAGAAG	VVKM IQA ITH	ATLPATLHVI	DOPSPHID	SSGTVRLLTE	PIQWP NT	DHPRTAAVSSEG IS	GTNAHL ILQ
MYCL_BKS	1 HTQAAAGAAG	VVKMIQAITH	ATLPATLHVI	DQPSPHID	SGTVRLLTE	PIQWP NT	DHPRTAAVSSEGIS	GTNAHL ILQ
MYCL_BKS	HTQAAAGAAG	VVKM IQA ITH	ATLPATLHVI	DQPSPHIDW	ISSGTVRLLTE	PIQWP NT	DHPRTAAVSSFG I	GTNAHL ILQ
PIKSKSI	HTQAAAGAAG HTQAAAGVSC	VIKMVOAMPH	ATLPATLHVI GLIPKTLHVI		SAGAVELLTE	PIQWP NT	DHPRTAAVSSEG IS	SGTNAHL ILQ
PIKS KS4	HTQAAAGVSG	IIKMVQAMRH	GVLPKTLHV	DRPSDQID	SAGTVELLTE	AMDWPRKQE	GGLRRAAVSSFGIS	GTNAHIVLE
PIKS KS5	HTQAAAGVAG	VIKMVQAMRH	GLLPKTLHVI	DEPSDQIDW	SAGTVELLTE	A V D <mark>WP</mark> E KQ D	GGL <mark>RRAA</mark> VSSFG I	GTNAHVVLE
PIK5K30	The second se		THE REAL PROPERTY AND ADDRESS OF ADDRES	T DOM DU	THE REPORT OF THE PARTY OF THE		CHILD D D D C DEE A E C V/	
RESEST	HADAAAGVAG	VIKMVOALRH	ETLPPTLHVI	DKPTLEVD	SAGAIELITE	ARAWP RN	GRPRRAGVSSEGV	SGTNAHL ILF
RIFSKS6	HAQAAAGVAG	V I KMVQAL RH V I KMVQAL RH	ETLPPTLHVI EVLPPTLHVI	DKPTLEV DV DRPTPEV DV	/SAGAIELLTE /SAGAIELLTE /SAGAVELLTE	ARAWP RN AREWP RN	IGRPRRAGVSAFGVS IGRPRRAGVSAFGVS	GTNAHL ILE

Figure S1. Sequence alignment of KS domains to identify residues for site-directed mutagenesis of DEBS KS3. Sequences were obtained from DEBS, mycolactone synthase (MYCL), PIKS, and RIFS. Residues within 12 Å of the active cysteine of DEBS KS3 (C175; Uniprot EryA2 Q03132 position 202) are highlighted in orange. Green circles- catalytic triad, red stars- residues selected for multipoint mutagenesis (A124, F126, A203, F236, F238, S279, and A416). Nomenclature of selected residues according to PDB 20Q3: A154, F156, A230, F263, F265, S306, A441.

	110 1	20 + +	130 140	150	160	170 , 180	
DEBS KS3	DPHSLRGTATGVE	LGVAKFGYG	EDTA - AAEDVEG	YSVTGVAPAVASGE	ISYTMGLEGPS	ISVDTACSSSLVA	HLAVESL
DEBSKS1	P P T S L Q A S P T G V F	VGLIPQE <mark>Y</mark> G	P R L A EGGEGVEG	YLM <mark>TG</mark> TTT <mark>SVA</mark> SGF	IAYTLGLEGPA	ISVDTACSSSLVA	HLACQSL
DEBSKS2	P P E T L R <mark>G</mark> S D T G V F	VGMSHQGYA	T G R P R P E D G V D G	YLL TGNTASVASGR	IAYVLGLEGPA	LTVDTACSSSLVA	HTACGSL
DEBS KS4	DPDALRGTDTGV	VGMNGQSYM	QLLAGEAERVDG	YQGLGNSASVLSGR	TAYTEGWEGPA	LTVDTACSSSLVG	IHLAMQAL
DEBSKS6	DPHSLRGSDTGVE	LGAAYOGYG	ODAVVPED - SEG	YLLTGNSSAVVSGR	VAYVLGLEGPA	VTVDTACSSELVA	HSACGSL
MYCL_A KS1	PAHTLAGTSTGVE	VGAWAQSYG	ATNSDDAEG	YAMTGGATSVMSGR	IAYTLGLEGPA	ITVDTACSSSLVA	IHLACQSL
MYCL_A KS5	∮ P A H T <mark>L</mark> A <mark>G</mark> T S T G V F	VGAGAQS <mark>YG</mark>	<mark>A</mark> TNSDDA <mark>EG</mark>	Y AMTGGATSVMSGR	IAYTLGLEGPA	ITVDTACSSSLVA	IHLACQSL
MYCL_AKS7	PAHTLAGTSTGVE	AGAWAQSYG	ATNSDDAEG	YAMTGGSTSVMSGR	IAYTLGLEGPA	ITVDTACSSSLVA	IHLACQSL
MYCL BKS1	PAHILAGISIGV	VGAWAQSYG		YAMIGGSISVMSGR	TAYTLGLEGPA	ITVDIACSSSLVA	THLACQSE
MYCL_BKS5	PAHTLAGTSTGVE	AGAWAQSYG	ATNSDDAEG	YAMTGGATSVMSGR	IAYTLGLEGPA	ITVDTACSSSLVA	IHLACOSL
MYCL_BKS7	' PAHT <mark>L</mark> A <mark>G</mark> TS <mark>TGV</mark>	AGAWAQSYG	<mark>A</mark> TNSDDA <mark>E</mark> G	Y AMTGGATSVMSGR	IAYTLGLEGPA	ITVDTACSSSLVA	IHLACQSL
PIKSKS1	DPTSLRGRQVGVF	TGAMTHEYG	PSLRDGGEGLD	YLLTGNTASVMSGR	VSYTLGLEGPA	LTVDTACSSSLVA	HLAVQAL
PIKSKS5		VGGWHTGYT	SCOTTAVOSPELEC	HIVSGAALGELSGE		V V DI ACSSSL VA	HLAVOAL
PIKSKS6	DPASVRGTDVGV	VGCGYQDYA	PDIRVAPEG TG	YVVTGNSSAVASGR	IAYSLGLEGPA	VTVDTACSSSLVA	HLALKGL
RIFSKS1	DP IA <mark>L</mark> K <mark>G</mark> TD <mark>TGV</mark> F	SGLMGQG <mark>Y</mark> -	G SG A V A P E L EG	F V T <mark>T G</mark> V A S S V A S G F	VSYVLGLEGPA	. VTVDTACSSSLVA	MHLAAQAL
RIFSK96	DPLSLKGNDVGV	TGMFGQGYV	- APGDSVVTPELEC	FAGTGGSSSVASGR	VSYVFGFEGPA	VTIDSACSSSLVA	HLAAQSL
RESISIU	DPGSLKGADVGV	AGVSNUGY -	GMGADPAELA	YASIAGASSVVSGR	VSYVEGEGPA		
	190 200	+ 210	220	230 🔹 🔹	240 2	50 260	
DEBS KS3	RKGESSMAVVGG		VDFSRORALAADGR	SKAFGAGADGFGFS	EGVTLVLERL	SEARRNGHEVLAV	RGSAL
DEBSKS1	RRGESSLAMAGG	/TVMPTPGML	V D F S R M N S L A P DG R	CKAFSAGANGFGMA	EGAGMLLLERL	SDARRNGHPVLAV	LRGTAV
DEBSKS2	R DG DCG L A V AGG V	/SVMAGPEVF	TEFSRQGALSPDGR	CKPFSDEADGFGLG	EGSAFVVLQRL	SDARREGRRVLGV	VAGSAV
DEBSKS5	RRGECSLALAGG	TVMSDPYTE	V D F STORGLASDGR	CKAFSARADGFALS	EGVAALVLEPL	SRARANGHQVLAVI	
DEBS KS6	RDGDCGLAVAGG	SVMAGPEVE	TEFSROGGLAVDGR	CKAFSAEADGFGFA	EGVAVVLLQRL	SDARRAGROVLGV	AGSAI
MYCL_AKS1	RNNESQLALAGG	/T <mark>V</mark> MSTPAVF	T D F S R Q R G L A P DG R	CKAFAATADGTGWG	EGAAVLVLERL	SEARRNNHPVLAI	VAGSAI
MYCL_A KS5	RNNESQLALAGG	TVMSTPAVF	T E F S R Q R G L A P D G R	CKAFAATADGTGFG	EGAAVLVLERL	SEARRNNHPVLAI	VAGSAI
MYCL A K99		TYMSTPAVE	TEESPORGLAPDGE	CKAFAATADGTGFG	EGAAVLVLERL	SEARRNNHPVLAI	AGSAI
MYCL_BKS1	RNNESQLALAGG	TVMSTPAVE	TEFSRORGLAPDGR	CKAFAATADGTGWG	EGAAVLVLERL	SEARRNNHPVLAI	AGSAI
MYCL_BKS5	RNNESQLALAGG	/T <mark>V</mark> MSTPAVF	T E <mark>F S R Q</mark> R G L A P DG R	C <mark>KAF</mark> AATA <mark>D</mark> GTGFG	EGAAVLVLERL	SEARRNNHPVLAI	VAGSAI
MYCL_BKS7	RNNESQLALAGG	TVMSTPAVF	TEFSRQRGLAPDGR	CKAFAATADGTGFG	EGAAVLVLERL	SEARRNNHPVLAI	VAGSAI
PIKSKS4	RKGECGLALAGGI	TVMSTPTTF		SKAFAASALGISWS	EGVGVLLVERL	SDARRNGHQVLAV	RGSAL
PIKS KS5	RKGECDMALAGG	TVMPNADLE	VQFSRORGLAADGR	SKAFATSADGFGPA	EGAGVLLVERL	SDARRNGHRILAV	RGSAV
PIKSKS6	RNGDCSTALVGG	AVLATPGAF	IEFSSQQAMAADGR	T KG F A S A A D G L A WG	EGVAVLLLERL	SDARRKGHRVLAV	V R <mark>G S</mark> A I
RIFSKS1 DIFSKS6	RQGECSMALAGG	TVMATPGSF	VEFSRQRALAPDGR	CKAFAAAADGTGWS	EGVGVVVLERL	SVARERGHR ILAVI	
RIFSKS10	ROGECSMALAGG		VEFAKORGLAGDGR	CKAPAAAACGIGWA	EGVGVVVLERL	SVARERGHRILAVI	
	270 🗙 280	290	300	310	320 3	30 340	
DEBSKS3	270 ± 280	290 GPAQRRVIR	300 QALESCGLEPGDVD		320 33	30 340 RDRDADRPLWLGS	
DEBSKS3 DEBSKS1 DEBSKS2	270 ± 280 N Q D G A S NG L S A P S N S D G A S NG L S A P S N Q D G A S NG L S A P S	290 G P AQ R R V I R G R AQ V R V I Q G V AO O R V I R	300 Q A L E S C G L E P G D V D Q A L A E S G L G P A D I D R A WA R A G I T G A D V A	310 AVEAHGTGTALGDP AVEAHGTGTRLGDP VVEAHGTGTRLGDP	320 33 IEANALLDTYG IEARALFEAYG VEASALLATYG	30 340 R D R D A D R P L WLGS R D R E Q P L HLGS K S R G S S G P V L LGS	/K <mark>SN</mark> IG /K <mark>SN</mark> LG /KSNIG
DEBSKS3 DEBSKS1 DEBSKS2 DEBSKS4	270 ± 280 NSDGASNGLSAPS NSDGASNGLSAPS NQDGASNGLSAPS NQDGASNGLAAPS	290 G P A Q R R V I R G R A Q V R V I Q G V A Q Q R V I R G P S Q E R V I R	300 QALESCGLEPGDVD QALAESGLGPADID RAWARAGITGADVA QALAASGVPAADVD	310 AVEAHGTGTALGD AVEAHGTGTRLGD VVEAHGTGTRLGD VVEAHGTGTELGD	320 IEANALLDTYG IEARALFEAYG VEASALLATYG IEAGALIATYG	0 340 R D R D A D R P L W L G S R D R E Q P L H L G S K S R G S S G P V L L G S Q D R D R P L R L G S	/K <mark>SN</mark> IG /K <mark>SNL</mark> G /K <mark>SN</mark> IG /KTNIG
DEBSKS3 DEBSKS1 DEBSKS2 DEBSKS4 DEBSKS5	270 280 N 2 D G A S N G L S A P S N 5 D G A S N G L S A P S N 2 D G A S N G L S A P S N 2 D G A S N G L S A P S N 2 D G A S N G L A A P S	290 G P AQ R R V I R G R AQ V R V I Q G V AQ Q R V I Q G V AQ Q R V I R G P AQ Q R V I R G P AQ Q R V I R	300 QALESCGLEPGDVD QALAESGLGPADID RAWARAGITGADVA QALAASGVPAADVD RALENAGVRAGDVD	310 A V E A HG TG T A L G D P A V E A HG TG T R L G D P V V E A HG TG T R L G D P V V E A HG TG T R L G D P Y V E A HG TG T R L G D P	320 IEANALLDTYG IEARALFEAYG VEASALLATYG IEAGALIATYG IEVHALLSTYG	30 340 R D R D A D R P L W L G S R D R E Q P L H L G S K S R G S S G P V L L G S Q D R D R P L R L G S A E R D P D D P L W I G S	/K <mark>SN</mark> IG /K <mark>SNL</mark> G /K <mark>SN</mark> IG /KTNIG /K <mark>SN</mark> IG
DEBSKS3 DEBSKS1 DEBSKS2 DEBSKS4 DEBSKS5 DEBSKS6 MYCL AKS1	270 ±280 NG D G A S NG L S A P S NG D G A S NG L S A P S NG D G A S NG L S A P S NG D G A S NG L S A P S NG D G A S NG L T A P S	290 G P A O R R V I R O G R A O V R V I Q G V A O O R V I R G P S O E R V I R G P A O Q R V I R G V A O Q R V I R	300 QALESCGLEPGDVD QALAESGLGPADID RAWARAGITGADVA QALAASGVPAADVD RALENAGVRAGDVD KAWARAGITGADVA	310 A V E A HG TG T A L G D P V V E A HG TG T R L G D P V V E A HG TG T R L G D P V V E A HG TG T R L G D P V V E A HG TG T R L G D P	320 IEANALLDIYG VEASALLATYG IEAGALIATYG IEAGALIATYG IEVHALLSTYG VEASALLATYG	30 340 RDR DA DR PL WLG S KSRGSSG PVLLG S QDR - DR PLRLG S A ER DP DD PL WLG S KSRGSSG PVLLG S	KSN IG KSN IG KSN IG KSN IG KSN IG
DEBS/KS3 DEBS/KS1 DEBS/KS2 DEBS/KS4 DEBS/KS5 DEBS/KS6 MYCL_A/KS1 MYCL_A/KS1	270 ±280 NG D G A S NG L S A P S NG D G A S NG L S A P S NG D G A S NG L S A P NG D G A S NG L A A P NG D G A S NG L A A P S NG D G A S NG L T A P	290 G P AQ R R V I R G R AQ V R V I Q G V AQ Q R V I R G P SQ E R V I R G P SQ E R V I R G P SQ Q R V I R G P SQ Q R V I R G P SQ Q R V I N G P SQ Q R V I N	300 Q A L E S C G L E P G D V D Q A L A E S G L G P A D I D R A WA R AG I T G A D V A Q L L A AS G V P A A D V D R A L E N AG V R AG D V D K A WA R AG I T G A D V A Q A L A N AG L T H D Q V D O A L A N AG L T H D Q V D	310 AVEAHGTGTALGD VVEAHGTGTRLGD VVEAHGTGTLGD VVEAHGTGTLGD VVEAHGTGTRLGD AVEAHGTGTLGD AVEAHGTGTLGD	320 IEANAL L DTYG IEARAL FEAYG IEAGAL LATYG IEAGAL IATYG VEASAL LATYG IEAGAL HATYG IEAGAL HATYG	20 340 R D R D A D R P L W L G S R D R - E Q P L H L G S K S R G S S G P V L L G S Q D R - D R P L R L G S K S R G S S G P V L L G S K S R G S S G P V L G S H H H T P D Q P L W L G S H H H T P D Q P L W L G S	/KSNIG /KSNIG /KSNIG /KSNIG /KSNIG IKSNIG IKSNIG
DEBSIKSI DEBSIKSI DEBSIKS2 DEBSIKS5 DEBSIKS6 MYCL_AIKS1 MYCL_AIKS7	270 280 NG CGASNGL SAP NG CGASNGL SAP NG CGASNGL SAP NG CGASNGL SAP NG CGASNGL TAP NG CGASNGL TAP NG CGASNGL TAP	290 G P AQ R R V I R G R AQ V R V I Q G V AQ R V I R G P SQ R V I R G P SQ R V I R G P SQ R V I N G P SQ R V I N G P SQ R V I N	300 QALESCGLEPGUD QALAESGLGPADI QALAESGLGPADI QALAASGVPAADVD KALWARAGITGADVA QALAASGVPAADVD QALANAGLTHDQVD QALANAGLTHDQVD QALANAGLTHDQVD	310 AV EAHGTGTALGCP AV EAHGTGTRLGCP VV EAHGTGTRLGCP VV EAHGTGTRLGCP VV EAHGTGTRLGCP AV EAHGTGTTLGCP AV EAHGTGTTLGCP AV EAHGTGTTLGCP	320 I E ANAL L D TYG J E ARAL F E AYG VE ASAL L ATYG I E AGAL I ATYG I E VHAL L STYG VE ASAL L ATYG I E AGAL HATYG I E ASAL HATYG	20 340 R D R D A D R P L WLG S R D R - E Q P L HLG S V L LG S V L G S	/KSNIG /KSNIG /KSNIG /KSNIG /KSNIG /KSNIG IKSNIG IKSNIG
DEBSKS3 DEBSKS1 DEBSKS2 DEBSKS5 DEBSKS6 MYCL_AKS3 MYCL_AKS3 MYCL_AKS3 MYCL_AKS3	270 280 NSDGASNGLSAP NQDGASNGLSAP NQDGASNGLSAP NQDGASNGLAP NQDGASNGLAP NQDGASNGLAP NQDGASNGLAP NQDGASNGLAP NQDGASNGLAP NQDGASNGLAP	GP AQ R RV I R G RAQ VRV I Q G VAQ RV I R GP SQ RV I R GP SQ RV I R GP SQ RV I R GP SQ RV I N GP SQ RV I N GP SQ RV I N GP SQ RV I N GP SQ RV I N	QALESCGLEPGU QALAESGLGPADIQ RAWARAGITGADVA QALAASGVPAADVC KAWARAGITGADVA QALANGVRAGDVA QALANAGLTHQVD QALANAGLTHQVD QALANAGLTHQVD QALANAGLTHQVD	310 AV EAHGTGTAL GDP VV EAHGTGTAL GDP VV EAHGTGTAL GDP VV EAHGTGTAL GDP VV EAHGTGTAL GDP AV EAHGTGTAL GDP AV EAHGTGTTL GDP AV EAHGTGTTL GDP AV EAHGTGTTL GDP AV EAHGTGTTL GDP	1 EANALL DTYG I EARALFEAYG VEASALL ATYG I EAGAL IATYG I EVAALLSTYG VEASALLATYG I EAGALHATYG I EAGALHATYG I EASALHATYG I EASALHATYG	340 R D R D A D R P L WLG S R D R - E Q P L H G S K S G S S G V L LG S Q D R - D R P L R LG S A E R D P D P L WLG S H H T P D Q P L WLG S H H H T P D Q P L WLG S H H H T P D Q P L WLG S H H H T P D Q P L WLG S H H H T P D Q P L WLG S	KSN IG KSN IG KSN IG KSN IG KSN IG KSN IG IKSN IG IKSN IG IKSN IG
DEBS KS3 DEBS KS1 DEBS KS4 DEBS KS5 DEBS KS5 DEBS KS5 MYQL_A KS5 MYQL_A KS5 MYQL_A KS9 MYQL_B KS5	270 280 NO CASNELSAM NO CASNELSAM NO CASNELSAM NO CASNELSAM NO CASNELAM NO CASNELAM NO CASNELTAM NO CASNELTAM NO CASNELTAM NO CASNELTAM NO CASNELTAM	290 G P AQ R R V I R G R AQ V R V I Q G V AQ Q R V I R G P SQ E R V I R G P SQ R V I R G P SQ R V I N G P SQ R V I N	200 QALESCLEFGDV QALAESSLFPGDV RAWARAGITGADVA QALAASGVPAADVQ XALAASGVPAADVQ QALAAGTTADVA QALAAGLTHDQVD QALANAGLTHDQVD QALANAGLTHDQVD QALANAGLTHDQVD QALANAGLTHDQVD QALANAGLTHDQVD	310 AVEANGTGALGD VVEANGTGTRLGD VVEANGTGTRLGD VVEANGTGRLGD VVEANGTGRLGD VVEANGTGRLGD AVEANGTGTLGD AVEANGTTLGD AVEANGTTLGD AVEANGTTLGD AVEANGTTLGD	20 I EANALL DTYG I EARALFEAYG VEASALLATYG I EAGAL IATYG I EAGAL IATYG VEASALLATYG I EAGAL HATYG I EASAL HATYG I EASAL HATYG I EASAL HATYG	0 340 R DR DADR PLWLGS R DR - E DPLHLGS K S RGS SG PVLLGS V Q DR - DR PLRLGS K S RGS SG PVLLGS HHT P DQ PLWLGS HHT P DQ PLWLGS	KSNIG KSNLG KSNIG KSNIG KSNIG KSNIG KSNIG KSNIG KSNIG
DEBS KS3 DEBS KS1 DEBS KS4 DEBS KS5 DEBS KS5 DEBS KS5 MYCL_A KS1 MYCL_A KS1 MYCL_B KS1 MYCL_B KS1 MYCL_B KS1	270 280 NO CASNELSAM NO CASNELSAM NO CASNELSAM NO CASNELSAM NO CASNELSAM NO CASNELSAM NO CASNELTAM NO CASNELTAM NO CASNELTAM NO CASNELTAM	GP AQ RV I R GR AQ V V I Q GR AQ V V I Q GP AQ RV I R GP AQ RV I R GP AQ RV I N GP SQ RV I N GP SQ Q RV I N	200 QALESCGLEPGDU RAWARGITCADVA QALASSCHADU QALASSCHADVA RAURAGUTCADVA QALANGUTHQVD QALANAGUTHQVD QALANAGUTHQVD QALANAGUTHQVD QALANAGUTHQVD QALANAGUTHQVD QALANAGUTHQVD QALANAGUTHQVD QALANAGUTHQVD QALANAGUTHQVD	310 AVEAHGTGTALGD VVEAHGTGTALGD VVEAHGTGTALGD VVEAHGTGTALGD VVEAHGTGTLGD AVEAHGTGTLGD AVEAHGTGTLGD AVEAHGTGTLGD AVEAHGTGTLGD AVEAHGTGTLGD AVEAHGTGTLGD AVEAHGTGTLGD AVEAHGTGTLGD	120 1 EA NALL DIYG 1 EA NALL DIYG VEASALL ATYG 1 EA GAL 1 ATYG 1 EA GALL ATYG 1 EA GAL HATYG 1 EA SALL ATYG 1 EA SALL HATYG 1 EA SALL HATYG	D 340 R DR DA DR PLWLGS KS RGSSG PVL GS KS RGSSG PVL GS AE RDP DP LWLGS HHHT PO PLWLGS HHHT PO PLWLGS HHHT PO PLWLGS HHHT PO PLWLGS HHHT PO PLWLGS HHHT PO PLWLGS	KSNIG KSNIG KTNIG KSNIG KSNIG KSNIG IKSNIG IKSNIG IKSNIG IKSNIG IKSNIG
DEBSKS3 DEBSKS2 DEBSKS4 DEBSKS5 DEBSKS6 MYCL_AKS5 MYCL_AKS5 MYCL_BKS1 MYCL_BKS1	270 280 NQ C & S NG L S A IP NQ C & S NG L S A IP NQ C & S NG L S A IP NQ C & S NG L A A IP NQ C & S NG L A A IP NQ C & S NG L A A IP NQ C & S NG L T A IP NQ C & S NG L T A IP NQ C & S NG L T A IP NQ C & S NG L T A IP NQ C & S NG L T A IP NQ C & S NG L T A IP NQ C & S NG L T A IP NQ C & S NG L T A IP NQ C & S NG L T A IP NQ C & S NG L T A IP NQ C & S NG L T A IP NQ C & S NG L T A IP NQ C & S NG L T A IP NQ C & S NG L T A IP NQ C & S NG L T A IP NQ C & S NG L T A IP NQ C & S NG L T A IP NQ C & S NG L T A IP NQ C & S NG L T A IP NQ C & S NG L T A IP NQ C & S NG L T A IP NQ C & S NG L T A IP	200 G P A Q R V I R G C A Q V V Q Q G V A Q R V I R G P S Q R V I R G P S Q R V I R G P S Q R V I N G P S Q R V I N	QALESCGLEPGDU QALESSCGLPGDU RAWARGITGADU RAWARGITGADU RALASSUPADU QALASSUPADU QALASSUPADU QALANAGUTHOU QALANAGUTHOU QALANAGUTHOU QALANAGUTHOU QALANAGUTHOU QALANAGUTHOU QALANAGUTHOU QALANAGUTHOU QALANAGUTHOU QALANAGUTHOU QALANAGUTHOU	210 A V A ANGIGI A L GO P V A ANGIGI A L GO P A A ANGIGI T L GO P	120 TY 124 NALL DTYG 124 RALF ATYG VEASALL ATYG 124 GALT ATYG 124 GALT ATYG 124 GALTATYG 124 SALL ATYG 124 SALHATYG 124 SALHATYG 124 SALHATYG 124 SALHATYG 124 SALHATYG 124 SALHATYG 124 SALHATYG	0 340 R DD AD R PLWLGS' KS RGSSG PVLGS' KS RGSSG PVLGS' KS RGSSG PVLGS' AE RDPDD PLWIGS' KS RGSSG PVLGS' HHHT POQPLWLGS' HHHT POQPLWLGS'	KSN IG KSN IG KSN IG KSN IG KSN IG IKSN IG IKSN IG IKSN IG IKSN IG IKSN IG KSN IG
DEBSKS3 DEBSKS2 DEBSKS4 DEBSKS6 DEBSKS6 MYCL_AKS7 MYCL_AKS7 MYCL_AKS7 MYCL_BKS2 MYCL_BKS7 PIKSKS4 PIKSKS4 DIBK5KS5	270 280 NQ C GAS NG L S A PI NQ C GAS NG L T A PI	200 G P A Q R V I Q G V A Q R V I Q G V A Q R V I R G P S Q R V I N G P S Q R V I N R G P S Q R V I N G R S Q R V I N R V I V I N G R S Q R V I N R V I V I N R V I V I V I N G R S Q R V I N R V I V I V I V I V I V I V I V I V I V	200 Q A L A S G L C P G D D R AWA R G I T G A D M R AWA R G I T G A D M Q A L A S G V A A D Y R A L M A G V A A D Y Q A L A A S G V A A D Y Q A L A N G L T H D Y Q A L A D A R L T T A D Y Q A L A D Y Q A L A N G L T H D Y H D Y Q A L A N G L T H D Y H D Y Q A L A N G L T H D Y H		300 3 I E ANAL DIYG I E ARAL FE AYG VE AS ALL ATYG I E AGAL I ATYG I E AGAL I ATYG I E AGAL HATYG I E AGAL HATYG I E AS AL HATYG	0 340 R D R D A D R P L WL G S R D R D A D R P L WL G S WL S R S S C P VL L G S WL S R S S C P VL L G S M S R S S C P VL L G S M S R S S C P VL L G S H H T P D P L WL G	KSN IG KSN IG KTN IG KSN IG
DEBSKS3 DEBSKS1 DEBSKS2 DEBSKS5 DEBSKS5 DEBSKS5 MYQL_AKS3 MYQL_AKS7 MYQL_BKS1 MYQL_BKS1 PIKSKS1 PIKSKS5 PIKSKS65	270 +200 NG G A S NG S A P NG G A S NG T A P	200 G P A Q R V I Q G V A Q R V I Q G V A Q R V I Q G V A Q R V I R G P A Q R V I R G P A Q R V I R G P A Q R V I N G P S Q R V I N I G P S Q R V I N I I I I I I I I I I I I I I I I I I I	20 20 20 20 20 20 20 20 20 20	210 A V A ANG C A G A G A V A ANG C A G A G A G A V A ANG C A G A G A G A V A ANG C A G A G A G A V A ANG C A G A G A G A V A ANG C A G A G A G A V A ANG C A G A G A G A V A ANG C A G A G A G A V A ANG C A G A G A G A G A V A ANG C A G A G A G A G A G A V A ANG C A G A G A G A G A G A A V A ANG C A G A G A G A A G A A G A A A G A A A G A	1 E ANAL DTYG 1 E ANAL DTYG 1 E ARAL FEANG 1 E ARAL FEANG 1 E ARAL STYG 1 E AGAL IATYG 1 E AGAL HATYG 1 E AGAL HATYG 1 E ASAL HATYG 1	20 340 R D R D A D R P W K G S R D R D A D R P W K G S K S R G S G V L G S K S R G S G V L G S R D R - 2 R P V L G S K S R G S G V L G S H H T P D C V K G H H T P D C V K G H H T P D C V K G G R D C P R G S G G D C P R G S Q G R S C P R G S C P R G S C P R G S Q G R S C P R G S C P R C P R G S C P R C P R C P R C P R C P R C P R C P R C P R C P R C	KSN IG KSN IG KSN IG KSN IG KSN IG IKSN IG IKSN IG IKSN IG IKSN IG IKSN IG KSN IG KSN IG KSN IG KSN IG KSN IG KSN IG
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DEESIGS DEESIGS DEESIGS DEESIGS DEESIGS DEESIGS DEESIGS MYCL_AISS MYCL_AISS MYCL_AISS MYCL_AISS MYCL_BISS DEESIGS PPISISS DEESIGS DEES	270 +200 ND C GAS NG L S A B A ND C GAS NG L S A B A ND C GAS NG L S A B A ND C GAS NG L S A B A ND C GAS NG L S A B A ND C GAS NG L S A B A ND C GAS NG L S A B A ND C GAS NG L S A B A ND C GAS NG L S A B A ND C GAS NG L S A B A ND C GAS NG L S A B A ND C GAS NG L T A B A ND C GAS NG L T A B A ND C GAS NG L T A B A ND C GAS NG L T A B A ND C GAS NG L T A B A ND C GAS NG L T A B A ND C GAS NG L T A B A ND C GAS NG L T A B A ND C GAS NG L T A B A ND C GAS NG L T A B A ND C GAS NG L T A B A ND C GAS NG L T A B A ND C GAS NG L T A B A ND C GAS NG L T A B A ND C GAS NG L T A B A ND C GAS NG	1200 1200	700 710 710 710 710 710 710 710	310 A V A ANG I GI A GO V A ANG A A GO V A ANG I GI A GO V A ANG I GO V A	320 32 32 1 C A NAL L D'TG C A RAL L D'TG C A RAL L C TG C	20 340 R D R D A D R P W W G S S W L G S S R D R D A D R P L W G S S V L G S S K S R S S G V L G S S W L G S S R D R D A D R P L W G S S W L G S S K S R S S G V L G S S W L G S S H H T P D O P L W G S S W L G S S H H T P D O P L W G S S W L G S S H H T P D O P L W G S S H W T S O P L W G S S G G D D C P L W G S S G G D C P L W G S S G G D D C P L W G S S G G D C P L W G S S G G A D C P L W G S S G C A S G C P L W G S S G G A S G C P L W G S S G C S S G C S G S G S G S G S G S G S G	K SN IG K SN IG A KU I I E

Figure S2. Sequence alignment of KS domains to identify residues for site-directed mutagenesis of DEBS KS6. Sequences were obtained from DEBS, mycolactone synthase (MYCL), PIKS, and RIFS. Residues within 12 Å of the active cysteine of DEBS KS6 (C175; Uniprot EryA3 Q03133 position 1661) are highlighted in orange. Green circles- catalytic triad, red stars- residues selected for multipoint mutagenesis (A124, Q126, S177, S203, V210, F236, F238, A279, A416). Nomenclature of selected residues according to homology model: A124, Q126, S174, S200, V207, F233, F235, A276, A412. Note that S177 (S174 in homology model) was accidently included in the diversified sequence space, although it is a conserved residue. According to the phylogenetic analysis implemented in FuncLib both Ala, and Gly can also be tolerated at this position (see Table S1).



Figure S3. Structural alignment of wild-type and mutant KS3 binding sites. Alignment of wild-type KS3 (PDB 2Q03) and predicted structures of mutant KS3 proteins. Catalytic triad in gray, residues selected for mutagenesis in orange, and residues mutant in the respective design in red.



Figure S4. Structural alignment of wild-type and mutant KS6 binding sites. Alignment of wild-type KS6 (homology model) and predicted structures of mutant KS6 proteins. Catalytic triad in gray, residues selected for mutagenesis in orange, and residues mutant in the respective design in red.



Figure S5. Purity of proteins used in this study. (A) SZ4-M3-TE and its mutants (MW 189 kDa), (B) SZ4-M6-TE and its mutants (MW 184 kDa), and (C) substrate-donating modules MK178 (Donor 4, 161 kDa), MK179 (Donor 6, 163 kDa), MK183 (Donor 2, 161 kDa), MK184 (Donor 3, 164 kDa), MK185 (Donor 5, 164 kDa).



Figure S6. Analysis of proteins by SEC – M3 substrate-accepting modules. Wild-type M3-TE and its mutants analyzed by SEC. All proteins eluted in a predominately single peak from SEC.



Figure S7. Analysis of proteins by SEC – M6 substrate-accepting modules. Wild-type M6-TE and its mutants analyzed by SEC. All proteins eluted in a predominately single peak from SEC.



Figure S8. LC-MS analysis of unreduced TKL 2 produced by chimeric bimodular PKSs using (5)M1-SZ3 and SZ4-M3-TE. Unreduced TKL **2** ($C_9H_{14}O_3$, calculated MW = 170.22 g/mol) was detected in all reaction mixtures after overnight incubation. The extracted ion chromatograms were obtained by extraction of the [M+H]⁺ species and one chromatogram per compound is shown as an example. Labeled peaks from left to right correspond to [M+H-H2O]⁺, [M+H]⁺, and [M+Na]⁺ ions. TKL **2** eluted at 4.7 min.



Figure S9. LC-MS analysis of reduced TKL 3 and unreduced TKL 5 produced by chimeric bimodular PKSs using (5)M1-SZ3 and SZ4-M6-TE. Reduced TKL 3 ($C_9H_{16}O_3$, calculated MW = 172.22 g/mol) and unreduced TKL 5 ($C_9H_{14}O_3$, calculated MW = 170.22 g/mol) were simultaneously detected in reaction mixtures shown in Figure 3B. The extracted ion chromatograms were obtained by extraction of the [M+H]⁺ species (TKL 5) or [M+Na]⁺ species (TKL 3) and one chromatogram per compound is shown as an example. Labeled peaks from left to right correspond to [M+H-H2O]⁺, [M+H]⁺, and [M+Na]⁺ ions. TKL 5 eluted at 4.7 min and TKL 3 at 4.2 min.



Figure S10. Analysis of substrate-donating modules by SEC. All proteins eluted in multiple oligomeric species from SEC.



Figure S11. LC-MS analysis of unreduced TKL 2 (in case of Donor 3) and unreduced TKL 4 (in case of Donor 2 & 4) produced by chimeric bimodular PKSs using SZ4-M3-TE and different substrate-donating modules. Either (5)KS1-AT1-KR1-ACP2-SZ3 (Donor 3), (5)KS1-AT1-KR2-ACP1-SZ3 (Donor 2), or <u>(5)KS1-AT1-KR2-ACP2-SZ3 (Donor 4)</u> was used as the substrate-donating module. Unreduced TKL **2** or the diastereomeric

form unreduced TKL **4** ($C_9H_{14}O_3$, calculated MW = 170.22 g/mol) was detected in all reaction mixtures after overnight incubation. The extracted ion chromatograms were obtained by extraction of the [M+H]⁺ species and one chromatogram per substrate-donating module is shown as an example. Labeled peaks from left to right correspond to [M+H-H2O]⁺, [M+H]⁺ and [M+Na]⁺ ions.



Figure S12. Structural organization of ACP docking interfaces and hydrophobic patch in DEBS KS3 and KS3_Mut13. The catalytic triad is depicted as black sticks. Position S306 which enlarged the conserved hydrophobic patch when mutated to S306P is depicted as orange stick. Docking interfaces of the upstream and intramodular ACP¹ are shown as yellow and green surfaces, respectively. The hydrophobic patch² is indicated as purple surface. Chain A and chain B of the KS3-AT3 didomain are colored in blue and grey, respectively.

Α	selected positions for mutagenesis in FuncLib mutated residues in Mut26: S174A, V207L, F235M
KS2AT2 1	EPIAIVGMACRLPGEVDSPERLWELITSGRDSAAEVPDDRGWVPDELMASDAAGTR
KS6AT6 1	DPIAIVGMACRFPGGVHNPGELWEFIVGRGDAVTEMPTDRGWDLDALFDPDPQRHGTSYS
¥20100 57	
KSCATC 57 KSCATC 61	RHGAFLDGAADFDAAFFGISPREALAMDPOOROVLETTWELFENAGIDPHSLRGSDTGVF
	** * ** ******************************
KS2AT2 117	VGM <mark>SHQ</mark> GYATGRPRPEDGVDGYLLTGNTASVASGRIAYVLGLEGPALTVDTAC <mark>S</mark> SSLVAL
KS6AT6 121	LGAAYQGYGQDAVVPEDS-EGYLLTGNSSAVVSGRVAYVLGLEGPAVTVDTACSSSLVAL
KS2AT2 177	HTACGSLRDGDCGLAVAGGVSVMAGPEVFTEFSROGALSPDGRCKPFSDEADGFGLGEGS
KS6AT6 180	HSACGSLRDGDCGLAVAGGV <mark>S</mark> VMAGPEVFTEFSRQGGLAVDGRCKAFSAEADGFGFAEGV
KS2AT2 237	AFVVLQRLSDARREGRRVLGVVAGSAVNQDGASNGL <mark>S</mark> APSGVAQQRVIRRAWARAGITGA
KS6AT6 240	AVVLLQRLSDARRAGRQVLGVVAGSAINQDGASNGL <mark>A</mark> APSGVAQQRVIRKAWARAGITGA * * ******** ** ******** ** ********* ****
KS2AT2 297	${\tt DVAVVEAHGTGTRLGDPVEASALLATYGKSRGSSGPVLLGSVKSNIGHAQAAAGVAGVIK}$
KS6AT6 300	DVAVVEAHGTGTRLGDPVEASALLATYGKSRGSSGPVLLGSVKSNIGHAQAAAGVAGVIK
KS2AT2 357	VLLGLERGVVPPMLCRGERSGLIDWSSGEIELADGVREWSPAADGVRRAGVSAFGVSGTN
KS6AT6 360	VVLGLNRGLVPPMLCRGERSPLIEWSSGGVELAEAVSPWPPAADGVRRAGVS <mark>A</mark> FGVSGTN
D	* *** ** *****************************
KS2 (yeinov) a	Highed to KSO (blue) KSZ (yellow) aligned to Mul2o (blue) KSZ (yellow) aligned to Mul2o (blue) KSZ (yellow) aligned to Mul2o (blue) F233 V207L F235 V207L F233 F233 F233 F235 F25 F25 F25 F255 F255 F25 F25 F255 F255 F255 F25 F2

Figure S13. Sequence and structural alignment of DEBS KS2 and KS6. (A) Sequence alignment of KS2 and KS6. Residues selected for mutagenesis are highlighted in orange and those mutated in Mut26 in red. Structural alignment of KS2 to KS6 (B) and of KS2 to Mut26 (C). Catalytic triad in gray, residues selected for mutagenesis in orange, and positions mutated in Mut26 in red.

Supplementary Tables

KS Domain	Position	Theoretical sequence space
	A154	ADEGIKLMNQSTVW
	F156	FADEGHKMNQSTY
	A230	ACST
KS3	F263	FALSY
	F265	FMWY
	S306	SAEGKMNPQRTV
	A441	AS
	A124	AGNQSTW
	Q126	QAGHNST
	S174	SAG
	S200	SAHMNT
KS6	V207	VACHILMNQST
	F233	FAS
	F235	FM
	A276	AEGKNQRST
	A412	ADEGS

Table S1. Sequence space of chosen binding site residues of KS3 and KS6 calculated by FuncLib

Construct	Protein	Yield/ mg/L of <i>E. coli</i> culture
MK149	SZ4-M3-TE	5.9
LB001	SZ4-M3-TE_Mut01	7.9
LB003	SZ4-M3-TE_Mut03	11.0
LB004	SZ4-M3-TE_Mut04	5.4
LB005	SZ4-M3-TE_Mut05	7.2
LB006	SZ4-M3-TE_Mut06	8.5
LB007	SZ4-M3-TE_Mut07	8.4
LB008	SZ4-M3-TE_Mut08	7.9
LB009	SZ4-M3-TE_Mut09	8.6
LB010	SZ4-M3-TE_Mut10	11.0
LB011	SZ4-M3-TE_Mut11	17.0
LB013	SZ4-M3-TE_Mut13	5.7
LB014	SZ4-M3-TE_Mut14	10.0
LB015	SZ4-M3-TE_Mut15	6.9
MK147	SZ4-M6-TE	6.3
LB002	SZ4-M6-TE_Mut02	5.5
LB016	SZ4-M6-TE_Mut16	9.1
LB017	SZ4-M6-TE_Mut17	11.0
LB018	SZ4-M6-TE_Mut18	11.0
LB019	SZ4-M6-TE_Mut19	8.2
LB021	SZ4-M6-TE_Mut21	9.6
LB022	SZ4-M6-TE_Mut22	5.1
LB023	SZ4-M6-TE_Mut23	6.2
LB024	SZ4-M6-TE_Mut24	6.5
LB026	SZ4-M6-TE_Mut26	7.2
LB027	SZ4-M6-TE_Mut27	7.9
BL12	LDD(4)	2.9
MK150	(5)M1-SZ3	4.7
MK148	SZ4-M2-TE	2.5
MK178	(5)KS1-AT1-KR2-ACP2-SZ3	4.1
MK179	(5)KS1-AT1-KR5-ACP5-SZ3	5.4
MK183	(5)KS1-AT1-KR2-ACP1-SZ3	2.7
MK184	(5)KS1-AT1-KR1-ACP2-SZ3	3.6
MK185	(5)KS1-AT1-KR1-ACP5-SZ3	6.5

Table S2. Yields of proteins used in this study. Typical yields are presented.

Construct	Im/ C
MK147	41.7±0.3
MK149	41.5±0.0
LB001	40.0±0.0
LB002	41.5±0.0
LB003	41.7±0.3
LB004	40.7±0.3
LB005	41.5±0.0
LB006	42.5±0.0
LB007	N/A
LB008	41.2±0.3
LB009	43.7±0.3
LB010	N/A
LB011	41.5±0.7
LB013	41.2±0.3
LB014	46.0±0.0
LB015	41.7±0.3
LB016	41.2±0.3
LB017	36.7±1.0
LB018	40.7±0.3
LB019	42.7±0.3
LB021	41.0±0.0
LB022	40.5±0.0
LB023	38.5±0.0
LB024	40.0±0.7
LB026	40.2±0.3
LB027	42.0±0.7

Table S3. Melting temperatures of wild-type and mutant modules. N/A.- Not applicable due to higheroligomerization of protein in the sample (see Figure S6).Construct $T_m / °C$

Table S4. Distribution of reduced and unreduced triketide lactone products in bimodular chimeric PKSs using LDD(4), (5)M1-SZ3 and the listed substrate-accepting modules. Peak areas were calculated from LC-MS measurements after the reaction time of 10 min. Peaks were obtained by searching for the respective [M+H]⁺-species. Reduced TKL **3** eluted at 4.2 min and unreduced TKL **5** at 4.7 min.

acceptor module	red. TKL, peak area	unred. TKL, peak area	red. TKL: unred. TKL
M2-TE	2.3x10 ⁷	7.1x10 ⁶	3.2 : 1
M6-TE_WT	5.9x10 ⁶	1.7×10^{8}	1:28.8
M6-TE_Mut02	4.2x10 ⁶	1.1×10^{8}	1:26.1
M6-TE_Mut16	6.3x10 ⁷	2.0×10^{8}	1:3.1
M6-TE_Mut17	2.2x10 ⁶	1.0×10^{8}	1:45.4
M6-TE_Mut18	4.2x10 ⁶	1.3x10 ⁸	1:30.9
M6-TE_Mut19	1.8x10 ⁶	5.4x10 ⁷	1:30.0
M6-TE_Mut21	4.5x10 ⁶	1.2×10^{8}	1:26.6
M6-TE_Mut22	2.1x10 ⁶	3.9x10 ⁷	1:18.5
M6-TE_Mut23	2.4x10 ⁶	6.4x10 ⁷	1:26.6
M6-TE_Mut24	6.9x10 ⁶	2.3x10 ⁸	1:33.3
M6-TE_Mut26	1.3x10 ⁷	5.1x10 ⁸	1:39.2
M6-TE_Mut27	5.1x10 ⁵	3.0x10 ⁷	1:58.8

Table S5. Peak area of reduced and unreduced triketide lactone products in bimodular chimeric PKSs using SZ4-M6-TE or Mut26 in the presence of different substrate-donating modules. The identity of the substrate on the upstream ACP is indicated. Peak areas were calculated from LC-MS measurements after a reaction time of 10 min. Peaks were obtained by searching for the respective [M+H]+-species. Reduced TKL 3 & 6 eluted at 4.2 min and unreduced TKL 5 & 7 at 4.7 min. Note that the ratio does not reflect the absolute amount of TKL product in the sample as no TKL standard was available to capture different ionization behaviors of the two compounds.

Acceptor module	Donor module (Abbreviation and full name)	Substrate bound to upstream ACP	red. TKL, peak area	unred. TKL, peak area
M6-TE	1, M1	NDK	5.9x10 ⁶	1.7×10^{8}
	5, KS1-AT1-KR1-ACP5	NDK	none	5.8x10 ⁷
	2, KS1-AT1-KR2-ACP1	EDK	1.1x10 ⁷	6.4x10 ⁶
	6, KS1-AT1-KR5-ACP5	EDK	3.6x10 ⁶	2.6x10 ⁶
M6-	1, M1	NDK	1.3x10 ⁷	5.1x10 ⁸
TE_Mut26	5, KS1-AT1-KR1-ACP5	NDK	3.1x10 ⁶	9.3x10 ⁷
	2, KS1-AT1-KR2-ACP1	EDK	1.1x10 ⁷	1.5x10 ⁶
	6, KS1-AT1-KR5-ACP5	EDK	1.8x10 ⁷	2.1x10 ⁶

Table S6. Plasmids used in this study and their origin

pBL12_LDD(4)_pET28_kan	Lowry <i>et al.</i> ³
	-
pMK147_SZ4-M6-TE-H6_pET22_carb	Klaus <i>et al.</i> 4
pMK148_SZ4-M2-TE-H6_pET22_carb	Klaus <i>et al</i> 4
pMK149_SZ4-M3-TE-H6_pET22_carb	Klaus <i>et al.</i> 4
pMK150_(5)M1-SZ3-H6_pET22b_carb	Klaus <i>et al.</i> 4
pMK178_(5)KS1-AT1-KR2-ACP2-SZ3-H6_pET22b_carb	this study
pMK179_(5)KS1-AT1-KR5-ACP5-SZ3-H6_pET22b_carb	this study
pMK183_(5)KS1-AT1-KR2-ACP1-SZ3-H6_pET22b_carb	this study
pMK184_(5)KS1-AT1-KR1-ACP2-SZ3-H6_pET22b_carb	this study
pMK185_(5)KS1-AT1-KR1-ACP5-SZ3-H6_pET22b_carb	this study
pLB001_SZ4-M3-TE-H6_A189W_pET22_carb	this study
pLB002_SZ4-M6-TE-H6_A189W_pET22_carb	this study
pLB003_SZ4-M3-TE-H6_A189Q_F191Y_A265S_F298L_pET22_carb	this study
pLB004_SZ4-M3-TE-H6_A189L_F191Y_A265S_F298L_F300Y_pET22_carb	this study
pLB005_SZ4-M3-TE-H6_A19E_F191Y_A165T_F298Y_F300W_pET22_carb	this study
pLB006_SZ4-M3-TE-H6_A189Q_F191Y_A265T_F298L_F300M_pET22_carb	this study
pLB007_SZ4-M3-TE-H6_A189Q_F191K_F298W_S341T_A476S_pET22_carb	this study
pLB008_SZ4-M3-TE-H6_A189T_F191Q_F298L_S306P_pET22_carb	this study
pLB009_SZ4-M3-TE-H6_A189Q_F191N_F298L_S341R_pET22_carb	this study
pLB010_SZ4-M3-TE-H6_F191Y_A265T_F298A_S341T_A476S_pET22_carb	this study
pLB011_SZ4-M3-TE-H6_A189Q_F191Y_A265C_F298L_S341P_pET22_carb	this study
pLB013_SZ4-M3-TE-H6_A189Q_F156Y_A265S_S341P_pET22_carb	this study
pLB014_SZ4-M3-TE-H6_A189W_F191M_F298L_F300Y_pET22_carb	this study
pLB015_SZ4-M3-TE-H6_A189E_F191Y_F298L_F300Y_pET22_carb	this study
pLB016_SZ4-M6-TE-H6_A189S_Q191H_S239A_V272I_A341S_pET22_carb	this study
pLB017_SZ4-M6-TE-H6_A189W_Q191S_S239A_S265A_A341R_pET22_carb	this study
pLB018_SZ4-M6-TE-H6_A189W_Q191H_S239A_S265T_A341T_pET22_carb	this study
pLB019_SZ4-M6-TE-H6_A189S_Q191H_S239A_S265H_A341S_pET22_carb	this study
pLB021_SZ4-M6-TE-H6_A189W_Q191S_S265A_A341T_pET22_carb	this study
pLB022_SZ4-M6-TE-H6_Q191S_S239A_V272L_F300M_A341E_pET22_carb	this study
pLB023_SZ4-M6-TE-H6_A189G_Q191H_S239A_S265H_A341Q_pET22_carb	this study
pLB024_SZ4-M6-TE-H6_A189W_S239A_S265T_A341S_A477S_pET22_carb	this study
pLB026_SZ4-M6-TE-H6_S239A_V272L_F300M_pET22_carb	this study
pLB027_SZ4-M6-TE-H6_A189T_S239A_S265H_V272M_A341S_pET22_carb	this study

Table S7. Cloning strategy of plasmids generated in this study. Individual fragments were generated by overlap extension PCR, conventional PCR or ordered as gBlocks and assembled via In-Fusion cloning. .Single point mutations were introduced via QuickChange site-directed mutagenesis. See Table S8 for primer used to generate individual fragments and Table S9 for sequences of gBlocks.

Plasmid	Cloning Method	Fragments	Primer Name	Primer Sequence 5'-3'	Template
pLB003	In-Fusion	1 + 7	P-LB003	CTCGCCGTAGCCGAACTTCcaCACTCCGAGGAAGACGCC	pMK149
			P-LB050	TCCGCTCGCCGCGCC	
		5 + 3	P-LB051	GCGCGAGGGCGGTGAG	рМК149
			P-LB009	CGGGGTCGCCATCACCGaGGCACCGCCGACGACC	
		pLB003V	P-LB052	CTCACCGCCCTCGCGC	рМК149
			P-LB053	GGCGCGGCGAGCGGA	
pLB004	In-Fusion	4 + 6	P-LB051	GCGCGAGGGCGGTGAG	рМК149
			P-LB009	CGGGGTCGCCATCACCGaGGCACCGCCGACGACC	
		2 + 8	P-LB008	GGTCGTCGGCGGTGCCtCGGTGATGGCGACCCCG	рМК149
			P-LB050	TCCGCTCGCCGCGCC	
		pLB003V	P-LB052	CTCACCGCCCTCGCGC	рМК149
			P-LB053	GGCGCGGCGAGCGGA	
pLB005	In-Fusion	11 + 17 + 21 + 35	P-LB050	TCCGCTCGCCGCGCC	рМК149
			P-LB051	GCGCGAGGGCGGTGAG	
		pLB003V	P-LB052	CTCACCGCCCTCGCGC	pMK149
			P-LB053	GGCGCGGCGAGCGGA	
pLB006	In-Fusion	22 + 18	P-LB051	GCGCGAGGGCGGTGAG	pMK149
			P-LB019	CGGGGTCGCCATCACCGtGGCACCGCCGACGACC	
		12 + 36	P-LB018	GGTCGTCGGCGGTGCCaCGGTGATGGCGACCCCG	рМК149
			P-LB050	TCCGCTCGCCGCGCC	
		pLB003V	P-LB052	CTCACCGCCCTCGCGC	рМК149
			P-LB053	GGCGCGGCGAGCGGA	

Plasmid	Cloning Method	Fragments	Primer Name	Primer Sequence 5'-3'	Template
pLB007	In-Fusion	pLB007_gblock			
		pLB007V	P- LB062opt	TCCGAGGAATACGCCGGTCGCGGTACCGCGC	рМК149
			P- LB063opt	TTCGGTATTAGCGGGACGAATGCGCACGTGATCGTC	
pLB008	In-Fusion	25 + 27	P-LB051	GCGCGAGGGCGGTGAG	pMK149
			P-LB011	GACGCCTTCGGAGAAGCCtAACCCGTCGGCGCCGG	
		13 + 32	P-LB010	CCGGCGCCGACGGGTTaGGCTTCTCCGAAGGCGTC	рМК149
			P-LB050	TCCGCTCGCCGCGCC	
		pLB003V	P-LB052	CTCACCGCCCTCGCGC	рМК149
			P-LB053	GGCGCGGCGAGCGGA	
pLB009 In-Fusion	In-Fusion	pLB009gblock			
		pLB009V	P-MK550	CCCCAAGAATACGCCGGTCGCGGTACCGCGC	pMK149
			P-MK551	CGCGCACCCTCGGGGCCCGCGCAGCGCAGGG	
pLB010	In-Fusion	pLB010gblock			
		pLB010V	P-MK552	CGCCACACCAAGGAAGACGCCGGTCGCGGTACC	pMK149
			P-MK553	TTCGGGATCTCTGGGACGAATGCGCACGTGATCGTC	
pLB011	In-Fusion	pLB011gblock			
		pLB011V	P-MK554	TCCTAAGAAGACGCCGGTCGCGGTACCGCGC	pMK149
			P-MK555	CCTGCCCCTTCAGGGCCCGCGCAGCGCAGG	
pLB013	In-Fusion	16 + 32	P-LB008	GGTCGTCGGCGGTGCCtCGGTGATGGCGACCCCG	pMK149
			P-LB050	TCCGCTCGCCGCGCC	
		22 + 19	P-LB050	TCCGCTCGCCGCGCC	pMK149
			P-LB009	CGGGGTCGCCATCACCGaGGCACCGCCGACGACC	
		pLB003V	P-LB052	CTCACCGCCCTCGCGC	рМК149
			P-LB053	GGCGCGGCGAGCGGA	

Plasmid	Cloning Method	Fragments	Primer Name	Primer Sequence 5'-3'	Template			
			P-LB015	GGGTGACGCCTTCGGAGtAGCCtAACCCGTCGGCGCCGG				
pLB015	In-Fusion	21	21					
		30 + 37	P-LB016	GCGTCTTCCTCGGAGTGGaGAAGTaCGGCTACGGCGAGGAC	pMK149			
			P-LB050	TCCGCTCGCCGCGCC				
		pLB003V	P-LB052	CTCACCGCCCTCGCGC	pMK149			
			P-LB053	GGCGCGGCGAGCGGA				
pLB016 In-Fu	In-Fusion	pLB016gblock		-				
		pLB016V	P-LB073	GGCCCCCAAGAAGACGCCGGTGTCGC	pMK147			
			P-LB074	GCGCCTTCGGGCGTCGCCCAGCAGC				
pLB017 In-	In-Fusion	pLB017gblock		·	·			
		pLB016V	P-LB073	GGCCCCCAAGAAGACGCCGGTGTCGC	pMK147			
			P-LB074	GCGCCTTCGGGCGTCGCCCAGCAGC				
pLB018	In-Fusion	pLB018gblock		·	·			
		pLB016V	P-LB073	GGCCCCCAAGAAGACGCCGGTGTCGC	pMK147			
			P-LB074	GCGCCTTCGGGCGTCGCCCAGCAGC				
pLB019	In-Fusion	pLB019gblock		-				
		pLB016V	P-LB073	GGCCCCCAAGAAGACGCCGGTGTCGC	pMK147			
			P-LB074	GCGCCTTCGGGCGTCGCCCAGCAGC				
pLB021	In-Fusion	pLB021gblock		·	·			
		pLB016V	P-LB073	GGCCCCCAAGAAGACGCCGGTGTCGC	pMK147			
			P-LB074	GCGCCTTCGGGCGTCGCCCAGCAGC				
pLB022	In-Fusion	pLB022gblock		·				
		pLB016V	P-LB073	GGCCCCCAAGAAGACGCCGGTGTCGC	pMK147			
			P-LB074	GCGCCTTCGGGCGTCGCCCAGCAGC				

Plasmid	Cloning Method	Fragments	Primer Name	Primer Sequence 5'-3'	Template		
pLB023 In-Fusion		pLB023gblock					
		pLB016V	P-LB073	GGCCCCCAAGAAGACGCCGGTGTCGC	pMK147		
			P-LB074	GCGCCTTCGGGCGTCGCCCAGCAGC			
pLB024	In-Fusion	pLB024gblock					
		pLB024V	P-LB073	GGCCCCCAAGAAGACGCCGGTGTCGC	pMK147		
			P-LB074	GCGCCTTCGGGCGTCGCCCAGCAGC	_		
pLB026	In-Fusion	pLB026gblock	·		·		
		pLB016V	P-LB073	GGCCCCCAAGAAGACGCCGGTGTCGC	pMK147		
			P-LB074	GCGCCTTCGGGCGTCGCCCAGCAGC			
pLB027	In-Fusion	pLB027glbock					
		pLB016V	P-LB073	GGCCCCCAAGAAGACGCCGGTGTCGC	pMK147		
			P-LB074	GCGCCTTCGGGCGTCGCCCAGCAGC			
pMK178	In-Fusion	pMK178_I	P-MK517	GAGCGCGTCTGGCTCCTGCCGGACCGCACCAC	pMK175		
			P-MK518	GCCACCGGATCCGCCGACCTCACTAGTGAGG			
		pMK178_V	P-MK312	GGATCCGCCACCGGATCCGCCTTCAGCAACATCGTTCTCCAATCTG	pMK111		
			P-MK483	GGCGGATCCGGTGGCGGA			
рМК179	In-Fusion	pMK179_I	P-MK519	GAGCGCGTCTGGCTCCCCATCCCCACCGGCG	pMK168		
			P-MK520	GCCACCGGATCCGCCGACGAGCCGCTCCAGGTA	_		
		pMK179_V	P-MK312	GGATCCGCCACCGGATCCGCCTTCAGCAACATCGTTCTCCAATCTG	pMK111		
			P-MK483	GGCGGATCCGGTGGCGGA	1		
рМК183	In-Fusion	pMK183_I	P-MK260	GAAGGAGATATACATATGAGCGGTGACAACGGCATGACCGAGGAAAAG	pMK178		

Plasmid	Cloning Method	Fragments	Primer Name	Primer Sequence 5'-3'	Template
			P-MK560	CCGGTCGCGCAGGCTC	
		pMK183_V	P-MK559	ACGGAGAGCCTGCGCGACCGGCTGGCGTCGCTGCCCG	pMK150
			P-MK239	CATATGTATATCTCCTTCTTAAAGTTAAACAAAATTA	
рМК184	In-Fusion	pMK184_I	P-MK260	GAAGGAGATATACATATGAGCGGTGACAACGGCATGACCGAGGAAAAG	pMK150
			P-MK562	CGCGCCCACCCGCGG	
		pMK184_V	P-MK239	CATATGTATATCTCCTTCTTAAAGTTAAACAAAATTA	pMK178
			P-MK561	GCCGAACCGCGGGTGGGCGCGCGCGGGGTCTGCCGC	-
pMK185	In-Fusion	pMK185_I	P-MK260	GAAGGAGATATACATATGAGCGGTGACAACGGCATGACCGAGGAAAAG	pMK150
			P-MK562	CGCGCCCACCCGCGG	-
		pMK185_V	P-MK563	GCCGAACCGCGGGTGGGCGCGCGCGCGCGCGCGCGCGCGC	pMK179
			P-MK239	CATATGTATATCTCCTTCTTAAAGTTAAACAAAATTA	-
pLB001	QuickChange	-	P-LB002	GGCGTCTTCCTCGGAGTGtgGAAGTTCGGCTACGGCGAG	pMK149
			P-LB003	CTCGCCGTAGCCGAACTTCcaCACTCCGAGGAAGACGCC	-
pLB002	QuickChange	-	P-LB004	CGTCTTCCTCGGCGCCtgGTACCAGGGCTACGGCC	pMK147
			P-LB005	GGCCGTAGCCCTGGTACcaGGCGCCGAGGAAGACG	

Fragment	Primer Name	Primer Sequence 5'-3'	Template	Used to generate
1	P-LB008	GGTCGTCGGCGGTGCCtCGGTGATGGCGACCCCG	pMK149	pLB003
	P-LB011	GACGCCTTCGGAGAAGCCtAACCCGTCGGCGCCGG		
2	P-LB008	GGTCGTCGGCGGTGCCtCGGTGATGGCGACCCCG	pMK149	pLB004
	P-LB015	GGGTGACGCCTTCGGAGtAGCCtAACCCGTCGGCGCCGG		
3	P-LB006	GGCGTCTTCCTCGGAGTGcaGAAGTaCGGCTACGGCGAGGAC	pMK149	pLB003
	P-LB009	CGGGGTCGCCATCACCGaGGCACCGCCGACGACC		
4	P-LB012	GGCGTCTTCCTCGGAGTGctGAAGTaCGGCTACGGCGAGGAC	pMK149	pLB004
	P-LB009	CGGGGTCGCCATCACCGaGGCACCGCCGACGACC		
5	P-LB051	GCGCGAGGGCGGTGAG	pMK149	pLB003
	P-LB007	GTCCTCGCCGTAGCCGtACTTCtgCACTCCGAGGAAGACGCC		
6	P-LB051	GCGCGAGGGCGGTGAG	pMK149	pLB004
	P-LB013	GTCCTCGCCGTAGCCGtACTTCagCACTCCGAGGAAGACGCC	_	
7	P-LB010	CCGGCGCCGACGGGTTaGGCTTCTCCGAAGGCGTC	pMK149	pLB003
	P-LB050	TCCGCTCGCCGCGCC	_	
8	P-LB014	CCGGCGCCGACGGGTTaGGCTaCTCCGAAGGCGTCACCC	pMK149	pLB004
	P-LB050	TCCGCTCGCCGCGCC		
11	P-LB018	GGTCGTCGGCGGTGCCaCGGTGATGGCGACCCCG	pMK149	pLB005
	P-LB021	CAGGGTGACGCCTTCGGAccAGCCGtACCCGTCGGCGCCGG		
12	P-LB018	GGTCGTCGGCGGTGCCaCGGTGATGGCGACCCCG	pMK149	pLB006
	P-LB023	CAGGGTGACGCCTTCGGAcAtGCCtAACCCGTCGGCGCCGG		
13	P-LB010	CCGGCGCCGACGGGTTaGGCTTCTCCGAAGGCGTC	pMK149	pLB008
	P-LB035	CCCGCTCGGCGCGGgAAGCCCGTTGCTGGCCC		
16	P-LB008	GGTCGTCGGCGGTGCCtCGGTGATGGCGACCCCG	pMK149	pLB013
	P-LB035	CCCGCTCGGCGCGGgAAGCCCGTTGCTGGCCC	_	
17	P-LB016	GCGTCTTCCTCGGAGTGGaGAAGTaCGGCTACGGCGAGGAC	pMK149	pLB005
	P-LB019	CGGGGTCGCCATCACCGtGGCACCGCCGACGACC		

 Table S8. Cloning strategy of fragments used in In-Fusion cloning and primer sequences

Fragment	Primer Name	Primer Sequence 5'-3'	Template	Used to generate
18	P-LB006	GGCGTCTTCCTCGGAGTGcaGAAGTaCGGCTACGGCGAGGAC	pMK149	pLB006
	P-LB019	CGGGGTCGCCATCACCGtGGCACCGCCGACGACC		
19	P-LB006	GGCGTCTTCCTCGGAGTGcaGAAGTaCGGCTACGGCGAGGAC	pMK149	pLB013
	P-LB009	CGGGGTCGCCATCACCGaGGCACCGCCGACGACC		
21	P-LB051	GCGCGAGGGCGGTGAG	pMK149	pLB015
	P-LB017	GTCCTCGCCGTAGCCGtACTTCtCCACTCCGAGGAAGACGC		
22	P-LB051	GCGCGAGGGCGGTGAG	pMK149	pLB006
	P-LB007	GTCCTCGCCGTAGCCGtACTTCtgCACTCCGAGGAAGACGCC		
25	P-LB051	GCGCGAGGGCGGTGAG	pMK149	pLB008
	P-LB033	GTGTCCTCGCCGTAGCCctgCTTCGtCACTCCGAGGAAGAC		
27	P-LB032	GTCTTCCTCGGAGTGaCGAAGcagGGCTACGGCGAGGACAC	pMK149	pLB008
	P-LB011	GACGCCTTCGGAGAAGCCtAACCCGTCGGCGCCGG		
30	P-LB016	GCGTCTTCCTCGGAGTGGaGAAGTaCGGCTACGGCGAGGAC	pMK149	pLB015
	P-LB015	GGGTGACGCCTTCGGAGtAGCCtAACCCGTCGGCGCCGG		
32	P-LB034	GGGCCAGCAACGGGCTTcCCGCGCCGAGCGGG	pMK149	pLB008
	P-LB050	TCCGCTCGCCGCGCC		
35	P-LB020	CCGGCGCCGACGGGTaCGGCTggTCCGAAGGCGTCACCCTG	pMK149	pLB005
	P-LB050	TCCGCTCGCCGCGCC		
36	P-LB022	CCGGCGCCGACGGGTTaGGCaTgTCCGAAGGCGTCACCCTG	pMK149	pLB006
	P-LB050	TCCGCTCGCCGCGCC		
37	P-LB014	CCGGCGCCGACGGGTTaGGCTaCTCCGAAGGCGTCACCC	pMK149	pLB015
	P-LB050	TCCGCTCGCCGCGCC		

Table S9. gBlocks used for In-Fusion cloning

gblock	Sequence 5'-3'
pLB007_	GGCGTATTCCTCGGAGTGCAGAAAAAGGGATATGGCGAAGACACGGCTGCAGCTGAGGACGTCGAGGGATACAGTGTTACCGGCGTGGCGCCTGCCGTGGCTAGTGGT
gblock	${\tt CGAATATCGTACACGATGGGTCTGGAGGGTCCAAGTATTTCCGTAGATACGGCCTGTAGTTCGTCCCTGGTAGCACTGCATTTGGCGGTAGAGTCGCTGCGAAAAGGT}$
	GAAAGTTCCATGGCCGTGGTAGGTGGTGCTGCGGTAATGGCAACACCTGGAGTATTCGTGGACTTTTCCCGTCAACGTGCGTTGGCAGCCGACGGTCGTAGTAAGGCG
	TTTGGCGCGGGAGCGGATGGTTGGGGGATTTAGTGAAGGCGTGACCCTGGTGCTCCTCGAGCGACGGAGCTCGACGCAATGGTCATGAGGTTTTGGCAGTGGTC
	CGCGGTTCGGCGCTCAATCAAGACGGAGCAAGCAATGGTTTGACTGCTCCTAGCGGCCCTGCACAAAGGCGCGTGATCCGGCAGGCGCTGGAGTCCTGCGGTCTGGAG
	CCGGGCGACGTTGATGCTGTCGAAGCTCACGGTACGGGTACTGCGTTGGGTGATCCGATTGAGGCGAATGCGCTCCTCGATACCTATGGACGAGATCGTGATGCCGAC
	CGACCGTTGTGGCTGGGATCGGTTAAGTCCAACATTGGACATACTCAAGCTGCGGCCGGTGTTACTGGTTTGCTGAAGGTAGTACTCGCAACCTCCGTAATGGCGAACTG
	CCAGCAACCCTCCATGTAGAGGAGGCCGACACCACATGTAGATTGGTCGTCCGGAGGTGTTGCGCTCCTGGCCGGAAATCAACCATGGAGGAGGGGAGAGGCGAACGCGA
LDOOD	
pLB009_	GCCGTATTCTTGGGGGTGCAGAAGAACGGCTATGGGCGAAGAATTGCGGCCGCCGCAGAAGATGCCGAGGGGTTACTCAGTTACCGGAGCGCCCCCGCCGTAGCGTAGCCG
gblock	
prenio_	
gblock	
	TGCCGGGGCCGACGGGGCTGGGGTTTTCTGAAGGGGTAACATTAGTGGTTGGT
	TAGTGCGCTGAACCAAGATGGACCAGCAACGGTCTGACGGCCTCGGCCCCCAGCGCCGTGATCGCCCAAGCGCTGGATCTTGCGCTTAGAGGCCCGG
	GGATGTCGACGCCGTAGAAGCCCATGGCACTGGAACAGCTTTGGGTGACCCAATCGAGGCTAACGCTCTTCTTGACACTTATGGTCGCGATCGTGACGCCGATCGTCGCCC
	GCTGTGGTTAGGTTCGGTCAAATCAAACATTGGCCACACGCAGGCTGCAGCAGGCGTAACGGGACTTTTGAAAGTAGTGCTGGCGCGCTTCGTAATGGGGAGTTACCAGC
	TACTTTGCACGTTGAGGAACCCACTCCTCATGTAGATTGGTCGTCGGGCGGCGTTGCCTTACTGGCGGGTAACCAACC
	GCGTGTTAGTTCGTTCGGGATCTCTGGG
pLB011	GGCGTCTTCTTAGGAGTCCAGAAGTATGGCTATGGGGAGGACACCGCTGCTGCGGAGGACGTAGAAGGTTATTCTGTCACAGGTGTAGCGCCTGCCGTGGCGTCAGGC
gblock	${\tt CGCATCTCTTACACAATGGGGTTAGAAGGTCCTTCAATTAGCGTAGACACCGCTTGCTCATCGTCGTTGGTAGCTCTTCACCTTGCAGTAGAGTCTTTACGCAAGGGG$
8510011	GAGTCTTCCATGGCCGTAGTGGGTGGTGGCTGCGTAATGGCAACCCCTGGCGTGTTCGTAGATTTCAGCCGCCAACGTGCATTAGCAGCAGATGGGCGCAGCAAAGCA
	${\tt TTTGGGGGCTGGTGCGGATGGATTAGGTTTTTCCGAAGGGGTTACTTTAGTTCTTTTGGAGCGTTTGTCAGAGGCGCGTCGTAATGGTCATGAGGTGTTAGCAGTGGTT$
	CGCGGCTCTGCCCTGAATCAAGACGGGGCAAGTAACGGCTTGCCTGCC
pLB016_	GTCTTCTTGGGGGGCCAGCTATCATGGTTATGGCCAAGATGCAGTGGTACCTGAGGATTCTGAGGGATATCTGCTTACGGGAAACTCGTCTGCGGTTGTAAGTGGGCCGT
gblock	GTTGCTTATGTACTGGGCTTGGAAGGGCCTGCGGTTACGGTAGATACAGCATGTTCCGCATCTTTGGTAGCTTTGCACAGCGCATGCGGATCTCTTCGTGATGGTGAT
0	TGCGGTCTTGCCGTAGCAGGCGGAGTTAGCGTAATGGCCGGTCCAGAAATCTTTACAGAGTTCTCGCGCCAGGGCGGCCTGGCCGTCGATGGGCGTTGTAAGGCCTTT
	AGCGCAGAGGCGGATGGTTTCGGTTTCGCTGAAGGGGTAGCTGTGGTCTTGTTACAACGCCTTAGTGATGCTCGCCGTGCGGGCCGCCAGGTCTTAGGGGTTGTAGCG
	GGAAGTGCAATTAATCAAGATGGTGCATCAAACGGACTGTCGGCGCCTTCGGGCGTC
pLB017_	GTCTTTTTGGGAGCTTGGTATAGTGGTTATGGGCAAGATGCCGTTGTCCCTGAAGACAGCGAAGGATACTTGCTTACAGGGAACAGTTCCGCCGTCGTGGTCGGGCCGC
gblock	GTTGCTTACGTGTTGGGTTTAGAAGGGCCCGCTGTAACGGTAGATACAGCTTGTTCCGCTAGCCTGGTAGCTTTACATTCCGCCTGTGGCTCACTGCGCGATGGTGAT
C	TGTGGATTAGCAGTGGCAGGAGGGGGGGGGGGGGAATGGCTGGACCCGAGGTTTTTACCGAATTTTCTCGTCAGGGTGGGT
	AGCGCCCGAGGCCGATGGGTTCGGCTTTGCCCGAGGGGTGTTGCAGTAGTTCTTCTTCAGCGTTGTCCGATGCCCGCCGTGCAGGTCGTCAAGTGCTGGGAGTTGTCGCG
	GGTTCTGCGATTAACCAAGATGGCGCGAGCAACGGACTTCGCGCCCCATCGGGCGTC
pLB018_	GTCTTTCTGGGGGGCATGGTATCATGGGTACGGCCAAGATGCCGTGGTGCCAGAGGACTCGGAGGGCTATTTATT
gblock	
	TCCGCTGAAGCAGACGGGTTCGGGTTCGCAGAAGGAGTGGCAGTCGTTTTGCTGCAGCGCTTATCGGATGCTCGTCGCGCCGGACGCCAGGTGTTAGGAGTGGCGCGCGC

gblock	Sequence 5'-3'
	GGCTCCGCGATCAATCAGGATGGCGCTAGCAACGGCCTTACAGCCCCTTCTGGAGTC
pLB019_ gblock	GTCTTTTTAGGTGCCTCATATCATGGATATGGCCAGGATGCTGTCGTTCCCGAGGATTCCGAGGGCTATCTGCTTACGGGTAACTCTTCTGCGGTAGTAAGCGGACGT GTAGCTTATGTCCTTGGCTTAGAAGGACCTGCTGTGACTGTCGACACAGCGTGCTCCGCCAGCTTAGTTGCGCTTCATAGTGCGTGGTAGTTTACGCGACGGAGGAC TGTGGTCTGGCCGTCGCGGGAGGAGTTCATGTAATGGCAGGCCCAGAGGTGTTTACTGAGTTTAGCCGTCAAGGAGGTTTAGCGGTAGACGGACG
pLB021_ gblock	GTCTTTTTAGGCGCGTGGTACTCAGGTTACGGCCAGGACGCTGTTGTGCCCGAAGATTCAGAGGGTTACCTTTTAACCGGGAACTCCAGTGCAGTCGTAAGTGGTCGC GTAGCCTACGTTTTAGGTTTAGAGGGTCCCGCCGTCACTGTGGATACGGCGTGTAGCAGTTCCTTAGTAGCGTTGCACAGTGCTTGCGGATCATTACGCGATGGTGAC TGTGGCCTGGCAGTAGCGGGTGGTGTGAGCCGTTATGGCGGGGCCCAGAAGTTTTCACCGAATTTTCTCGTCAAGGGGGTTTAGCTGTTGACGGCCGCTGTAAGGCTTTC TCTGCAGAGGCTGATGGTTTTGGCTTTGCAGAGGGGGTAGCCGTAGTCCTGCTGCAGCGTTTATCGGACGCCCGCC
pLB022_ gblock	GTCTTCTTGGGGGCGGCGTACTCTGGATATGGTCAAGATGCGGTGGTTCCCGAGGACTCTGAGGGGTACTTGTTGACTGGCAATAGTAGCGCTGTAGTCAGCGGTCGT GTCGCGTACGTGCTTGGTCTTGAGGGCCCAGCCGTAACTGTCGATACTGCATGTTCAGCCAGC
pLB023_ gblock	GTCTTTCTTGGCGCGGGGTATCACGGGTACGGACAGGACGCAGTAGTTCCCGAAGATAGCGAAGGTTACCTGTTGACCGGAAACAGCAGTGCCGTGGTCAGCGGACGT GTCGCGTATGTTCTTGGTTTAGAAGGACCTGCGGGGGACTGTTGATACTGCCTGTAGCGCAAGTCTTGTTGCTTTACATAGTGCCTGTGGTTCTTTACGTGATGGAGAC TGTGGGCTGGCGGTGGCCGGAGGCGTGCATGTCATGGCGGGGGCCCGAAGTGTTTACAGAGTTCTCACGCCAAGGAGGCCTTGCGGTAGACGGTCGCTGTAAGGCTTTC TCGGCTGAGGCGGATGGTTTCGGCTTTGCGGAAGGGGTCGCCGTTGTATTACTTCAGCGTCTTTCAGATGCTCGTCGCGCGGCGGTCGTCAGGTCTTGGGCGTGGTAGCG GGATCTGCAATTAATCAGGATGGAGCTAGCAATGGTTTACAGGCTCCGAGCGGCGTC
pLB024_ gblock	GTCTTCCTTGGGGCGTGGTATCAAGGTTACGGTCAGGACGCCGTAGTTCCTGAAGATTCCGAAGGCTATTTGTTAACGGGGAATTCGTCTGCGGTGGTTTCTGGTCGT GTAGCTTACGTCTTAGGGCTTGAGGGACCTGCTGTCACAGTCGATACAGCTTGTAGCGCATCTTTAGTTGCACTGCACTCGGCATGTGGCTCTCTTCGTGATGGAGAC TGCGGATTAGCAGTTGCGGGGGGGCGTCACCGTAATGGCCGGCC
pLB026_ gblock	CGTGCTGGAGTAAGCTCTTTTGGGGTTTCGGGG GTCTTTTTGGGCGCCGCATACCAAGGTTACGGTCAGGATGCCGTTGTTCCCGAAGACTCCGAGGGTTACCTGCTTACTGGTAATAGCAGTGCGGTGGTCTCAGGTCGC GTAGCATACGTGTTGGGTCTGGAGGGGCCTGCCGTCACAGTCGACAGCGGCTTGCCAGTGCGAGGTTTAGTTGCTTTACACAGTGCTTGTGGCAGTCTTCGTGATGGTGGTG TGCGGATTAGCCGTTGCTGGTGGTGTGTGTCTGTGATGGCTGGGCCGGAGCCTTTTTACTGAGTTTAGCCGTCAAGGGGGCCTGGCAGTAGATGGACGTTGCAAGGCGTTT TCCGCCGAGGCAGACGGGTTCGGAATGGCTGAGGGGGTGGCCGTGGCGGTGGTCCTTCTTCAACGCCTTTCAGAGGCGCGCGTCGTGCAAGGGGGCCGAGAGTGGCGGGGGGGG
gblock	GTGTTTTGGGGGGGGAAATAGTTCAGGGGGTAGGGGGGTGGGGGGGG

Table bio minino acta bequences of nemy generated substrate admining mouand	Table S10. Amino acid see	quences of newly gen	erated substrate-d	onating module
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Construct	Amino acid sequence
MK178 (5)KS1- AT1-KR2- ACP2-SZ3	MSGDNGMTEEKLRRYLKRTVTELDSVTARLREVEHRAGEPVAVVAMACRLPGGVSTPEEFWELLSEGRDAVAGLPTDRGWDLDSLFH PDPTRSGTAHQRGGGFLTEATAFDPAFFGMSPREALAVDPQQRLMLELSWEVLERAGIPPTSLQASPTGVFVGLIPQEYGPRLAEGG EGVEGYLMTGTTTSVASGRIAYTLGLEGPAISVDTACSSSLVAVHLACQSLRRGESSLAMAGGVTVMPTPGMLVDFSRMNSLAPDGR CKAFSAGANGFGMAEGAGMLLLERLSDARRNGHPVLAVLRGTAVNSDGASNGLSAPNGRAQVRVIQQALAESGLGPADIDAVEAHGT GTRLGDPIEARALFEAYGRDREQPLHLGSVKSNLGHTQAAAGVAGVIKMVLAMRAGTLPRTLHASERSKEIDWSSGAISLLDEPEPW PAGARPRRAGVSSFGISGTNAHAIIEEAPQVVEGERVEAGDVVAPWVLSASSAEGLRAQAARLAAHLREHPGQDPRDIAYSLATGRA ALPHRAAFAPVDESAALRVLDGLATGNADGAAVGTSRAQQRAVFVFPGQGWQWAGMAVDLLDTSPVFAAALRECADALEPHLDFEVI PFLRAEAARREQDAALSTERVDVVQPVMFAVMVSLASMWRAHGVEPAAVIGHSQGEIAAACVAGALSLDDAARVVALRSRVIATMPG NKGMASIAAPAGEVRARIGDRVEIAAVNGPRSVVVAGDSDELDRLVASCTTECIRAKRLAVDYASHSSHVETIRDALHAELGEDFHP LPGFVPFFSTVTGRWTQPDELDAGYWYRNLRRTVRFADAVRALAEQGYRTFLEVSAHPILTAAIEEIGDGSGADLSAIHSLRRGDGS LADFGEALSRAFAAGVAVDWESVHLGTGARRVPLPTYPFQRERVWLLPDRTTPRDELDGWFYRVDWTEVPRSEPAALRCRWLVVVPE GHEEDGWTVEVRSALAEAGAEPEVTRGVGGLVGDCAGVVSLLALEGDGAVQTLVLVRELDAEGIDAPLWTVTFGAVDAGSPVARPDQ AKLWGLGQVASLERGPRWTGLVDLPHMPDPELRGRLTAVLAGSEDQVAVRADAVRARRLSPAHVTATSEYAVPGGTILVTGGTAGLG AEVARWLAGRGAEHLALVSRRGPDTEGVGDLTAELTRLGARVSVHACDVSSREPVRELVHGLIEQGDVVRGVVHAAGLPQQVAINDM DEAAFDEVVAAKAGGAVHLDELCSDAELFLLFSSGAGVWGSARQGAYAAGNAFLDAFARHRRGRGLPATSVAWGLWAAGGMTGDEEA VSFLRERGVRAMPVPRALAALDRVLASGETAVVVTDVDVDPAFAESYTAARPRPLLDRIVTTAPSERAGEPETESLRDRLAGLPRAER TAELVRLVRTSTATVLGHDDPKAVRATTPFKELGFDSLAAVRLRNLLNAATGLRLPSTLVFDHPNASAVAGFLTSELGGGSGGGSGN EVTTLENDAAFIENENAYLEKEIARLRKEKAALRNRLAHKKLEHHHHHH
MK179 (5)KS1- AT1-KR5- ACP5-SZ3	MSGDNGMTEEKLRRYLKRTVTELDSVTARLREVEHRAGEPVAVVAMACRLPGGVSTPEEFWELLSEGRDAVAGLPTDRGWDLDSLFH PDPTRSGTAHQRGGGFLTEATAFDPAFFGMS PREALAVDPQQRLMLELSWEVLERAGIPPTSLQAS PTGVFVGLIPQEYGPRLAEGG EGVEGYLMTGTTTSVASGRIAYTLGLEGPAISVDTACSSSLVAVHLACQSLRRGESSLAMAGGVTVMPTPGMLVDFSRMNSLAPDGR CKAFSAGANGFGMAEGAGMLLLERLSDARRNGHPVLAVLRGTAVNSDGASNGLSAPNGRAQVRVIQQALAESGLGPADIDAVEAHGT GTRLGDPIEARALFEAYGRDREQPLHLGSVKSNLGHTQAAAGVAGVIKMVLAMRAGTLPRTLHASERSKEIDWSSGAISLLDEPEPW PAGARPRRAGVSSFGISGTNAHAIIEEAPQVVEGERVEAGDVVAPWVLSASSAEGLRAQAARLAAHLREHPGQDPRDIAYSLATGRA ALPHRAAFAPVDESAALRVLDGLATGNADGAAVGTSRAQQRAVFVFPGQGWQWAGMAVDLLDTSPVFAAALRECADALEPHLDFEVI PFLRAEAARREQDAALSTERVDVVQPVMFAVMVSLASMWRAHGVEPAAVIGHSQGEIAAACVAGALSLDDAARVVALRSRVIATMPG NKGMASIAAPAGEVRARIGDRVEIAAVNGPRSVVVAGDSDELDRLVASCTTECIRAKRLAVDYASHSSHVETIRDALHAELGEDFHP LPGFVPFFSTVTGRWTQPDELDAGYWYRNLRRTVRFADAVRALAEQGYRTFLEVSAHPILTAAIEEIGDGSGADLSAIHSLRRGDGS LADFGEALSRAFAAGVAVDWESVHLGTGARRVPLPTYPFQRERVWLPIPTGGRARDEDDDWRYQVVWREAEWESASLAGRVLLVTGP GVPSELSDAIRSGLEQSGATVLTCDVESRSTIGTALEAADTDALSTVVSLLSRDGEAVDPSLDALALVQALGAAGVEAPLWVLTRNA VQVADGELVDPAQAMVGGLGRVVGIEQPGRWGGLVDLVDADAASIRSLAAVLADPRGEEQVAIRADGIKVARLVPAPARAARTRWSP RGTVLVTGGTGGIGAHVARNLARSGAEHLVLLGRRGADAPGASSLREELTALGTGVTIAACDVADRARLEAVLAAERAEGRTVSAVM HAAGVSTSTPLDDLTEAEFTEIADVKVRGTVNLDELCPDLDAFVLFSSNAGVWGSPGLASYAAANAFLDGFARRRSEGAPVTSIAW GLWAGQNMAGDEGGEYLRSQGLRAMDPDRAVEELHITLDHGQTSVSVVDMDRRFVELFTAARHRPLFDEIAGARAEARQSEEGPAL AQRLAALSTAERREHLAHLIRAEVAAVLGHGDDAAIDRAFRDLGFDSMTAVDLRNRLAAVTGVREAATVVFDHPTITRLADHYLE RLVGGSGGGSGNEVTTLENDAAFIENENAYLEKEIARLRKEKAALRNRLAHKKLEHHHHH
MK183 (5)KS1- AT1-KR2- ACP1-SZ3	MSGDNGMTEEKLRRYLKRTVTELDSVTARLREVEHRAGE PVAVVAMACRLPGGVSTPEEFWELLSEGRDAVAGLPTDRGWDLDSLFH PDPTRSGTAHQRGGGFLTEATAFDPAFFGMSPREALAVDPQQRLMLELSWEVLERAGIPPTSLQASPTGVFVGLIPQEYGPRLAEGG EGVEGYLMTGTTTSVASGRIAYTLGLEGPAISVDTACSSSLVAVHLACQSLRRGESSLAMAGGVTVMPTPGMLVDFSRNNSLAPDGR CKAFSAGANGFGMAEGAGMLLLERLSDARRNGHPVLAVLRGTAVNSDGASNGLSAPNGRAQVRVIQQALAESGLGPADIDAVEAHGT GTRLGDPIEARALFEAYGRDREQPLHLGSVKSNLGHTQAAAGVAGVIKMVLAMRAGTLPRTLHASERSKEIDWSSGAISLLDEPEPW PAGARPRRAGVSSFGISGTNAHAIIEEAPQVVEGERVEAGDVVAPWLSASSAEGLRAQAARLAAHLREHPGQDPRDIAYSLATGRA ALPHRAFFAPVDESAALRVLDGLATGNADGAAVGTSRAQQRAVFVFPGQGWQWAGMAVDLLDTSPVFAAALRECADALEPHLDFEVI PFLRAEAARREQDAALSTERVDVQPVMFAVMVSLASMWRAHGVEPAAVIGHSQGEIAAACVAGALSLDDAARVVALRSRVIATMPG NKGMASIAAPAGEVRARIGDRVEIAAVNGPRSVVVAGDSDELDRLVASCTTECIRAKRLAVDYASHSSHVETIRDALHAELGEDFHP LPGFVPFFSTVTGRWTQPDELDAGYWYRNLRRTVRFADAVRALAEQGYRTFLEVSAHPILTAAIEEIGDGSGADLSAIHSLRRGDGS LADFGEALSRAFAAGVAVDWESVHLGTGARRVPLPTYPFQRERVWLLPDRTTPRDELDGWFYRVDWTEVPRSEPAALRGRWLVVVPE GHEEDGWTVEVRSALAEAGAEPEVTRGVGGLVGDCAGVVSLLALEGDGAVQTLVLVRELDAEGIDAPLWTVTFGAVDAGSPVARPDQ AKLWGLGQVASLERGPRWTGLVDLPHMPDPELRGRLTAVLAGSEDQVAVRADAVRARRLSPAHVTATSEYAVPGGTILVTGGTAGLG AEVARWLAGRGAEHLALVSRRGPDTEGVGDLTAELTRLGARVSVHACDVSSREPVRELVHGLIEQGDVVRGVHAAGLPQQVAINDM DEAAFDEVVAAKAGGAVHLDELCSDAELFLLFSSGAGVWGSARQGAYAAGNAFLDAFAHRRGRGLPATSVAWGLWAAGGMTGDEEA VSFLRERGVRAMPVPRALAALDRVLASGETAVVVTDVDWPAFAESYTAARPRPLLDRIVTTFDALAAELGGGSGGGSGN EVTTLENDAAFIENENAYLEKEIARLRKEKAALRNRLAHKKLEHHHHH

Construct	Amino acid sequence
MK184 (5)KS1- AT1-KR1- ACP2-SZ3	MSGDNGMTEEKLRRYLKRTVTELDSVTARLREVEHRAGEPVAVVAMACRLPGGVSTPEEFWELLSEGRDAVAGLPTDRGWDLDSLFH PDPTRSGTAHQRGGGFLTEATAFDPAFFGMSPREALAVDPQQRLMLELSWEVLERAGIPPTSLQASPTGVFVGLIPQEYGPRLAEGG EGVEGYLMTGTTTSVASGRIAYTLGLEGPAISVDTACSSSLVAVHLACQSLRRGESSLAMAGGVTVMPTPGMLVDFSRMNSLAPDGR CKAFSAGANGFGMAEGAGMLLLERLSDARRNGHPVLAVLRGTAVNSDGASNGLSAPNGRAQVRVIQQALAESGLGPADIDAVEAHGT GTRLGDPIEARALFEAYGRDREQPLHLGSVKSNLGHTQAAAGVAGVIKMVLAMRAGTLPRTLHASERSKEIDWSSGAISLLDEPEPW PAGARPRRAGVSSFGISGTNAHAIIEEAPQVVEGERVEAGDVVAPWVLSASSAEGLRAQAARLAAHLREHPGQDPRDIAYSLATGRA ALPHRAAFAPVDESAALRVLDGLATGNADGAAVGTSRAQQRAVFVFPGQGWQWAGMAVDLLDTSPVFAAALRECADALEPHLDFEVI
	PFLRAEAARREQDAALSTERVDVVQPVMFAVMVSLASMWRAHGVEPAAVIGHSQGEIAAACVAGALSLDDAARVVALRSRVIATMPG NKGMASIAAPAGEVRARIGDRVEIAAVNGPRSVVVAGDSDELDRLVASCTTECIRAKRLAVDYASHSSHVETIRDALHAELGEDFHP LPGFVPFFSTVTGRWTQPDELDAGYWYRNLRRTVRFADAVRALAEQGYRTFLEVSAHPILTAAIEEIGDGSGADLSAIHSLRRGDGS LADFGEALSRAFAAGVAVDWESVHLGTGARRVPLPTYPFQRERVWLEPKPVARRSTEVDEVSALRYRIEWRPTGAGEPARLDGTWLV AKYAGTADETSTAAREALESAGARVRELVVDARCGRDELAERLRSVGEVAGVLSLLAVDEAEPEEAPLALASLADTLSLVQAMVSAE LGCPLWTVTESAVATGPFERVRNAAHGALWGVGRVIALENPAVWGGLVDVPAGSVAELARHLAAVVSGGAGEDQLALRADGVYGRRW VRAAAPATDDEWKPTGTVLVTGGTGGVGGQIARWLARRGAPHLLLVSRSGPDADGAGELVAELEALGARTTVAACDVTDRESVRELL GGIGDDVPLSAVFHAAATLDDGTVDTLTGERIERASRAKVLGARNLHELTRELDLTAFVLFSSFASAFGAPGLGGYAPGNAYLDGLA QQRRSDGLPATAVAWGTWAGSGMAEGPVADRFRRHGVIEMPPETACRALQNALDRAEVCPIVIDVRWDRFLLAYTAQRPTRLFDEID DARRAAPQAAAEPRVGALAGLPRAERTAELVRLVRTSTATVLGHDDFKAVRATTPFKELGFDSLAAVRLRNLLNAATGLRLPSTLVF DHPNASAVAGFLTSELGGGSGGGSGNEVTTLENDAAFIENENAYLEKEIARLRKEKAALRNRLAHKKLEHHHHHH
MK185 (5)KS1- AT1-KR1- ACP5-SZ3	MSGDNGMTEEKLRRYLKRTVTELDSVTARLREVEHRAGEPVAVVAMACRLPGGVSTPEEFWELLSEGRDAVAGLPTDRGWDLDSLFH PDPTRSGTAHQRGGGFLTEATAFDPAFFGMS PREALAVDPQQRLMLELSWEVLERAGIPPTSLQAS PTGVFVGLIPQEYGPRLAEGG EGVEGYLMTGTTTSVASGRIAYTLGLEGPAISVDTACSSSLVAVHLACQSLRRGESSLAMAGGVTVMPTPGMLVDFSRMNSLAPDGR CKAFSAGANGFGMAEGAGMLLLERLSDARRNGHPVLAVLRGTAVNSDGASNGLSAPNGRAQVRVIQQALAESGLGPADIDAVEAHGT GTRLGDPIEARALFEAYGRDREQPLHLGSVKSNLGHTQAAAGVAGVIKMVLAMRAGTLPRTLHASERSKEIDWSSGAISLLDEPEPW PAGARPRRACVSSFGISGTNAHAIIEEAPQVVEGERVEAGDVVAPWVLSASSAEGLRAQAARLAAHLREHPGQDPRDIAYSLATGRA ALPHRAAFAPVDESAALRVLDGLATGNADGAAVGTSRAQQRAVFVFPGQGWQWAGMAVDLLDTSPVFAAALRECADALEPHLDFEVI PFLRAEAARREQDAALSTERVDVVQPVMFAVMVSLASMWRAHGVEPAAVIGHSQGEIAAACVAGALSLDDAARVVALRSRVIATMPG NKGMASIAAPAGEVRARIGDRVEIAAVNGPRSVVVAGDSDELDRLVASCTTECIRAKRLAVDYASHSSHVETIRDALHAELGEDFHP LPGFVPFFSTVTGRWTQPDELDAGYWYRNLRRTVRFADAVRALAEQGYRTFLEVSAHPILTAAIEEIGDGSGADLSAIHSLRRGDGS LADFGEALSRAFAAGVAVDWESVHLGTGARRVPLPTYPFQRERVWLEPKPVARRSTEVDEVSALRYRIEWRPTGAGEPARLDGTWLV AKYAGTADETSTAAREALESAGARVRELVVDARCGRDELAERLRSVGEVAGVLSLLAVDEAEPEEAPLALASLADTLSLVQAMVSAE LGCPLWTVTESAVATGPFERVRNAAHGALWGVGRVIALENPAVWGGLVDVPAGSVAELARHLAAVVSGGAGEDQLALRADGVYGRRW VRAAAPATDDEWKPTGTVLVTGGTGGVGGQIARWLARRGAPHLLLVSRSGPDADGAGELVAELEALGARTTVAACDVTDRESVRELL GGIGDDVPLSAVFHAAATLDDGTVDTLTGERIERASRAKVLGARNLHELTRELDLTAFVLFSSFASAFGAPGLGGYAPGNAYLDGLA QQRRSDGLPATAVAWGTWAGSGMAEGPVADRFRRHGVIEMPETACRALQNALDRAEVCPIVIDVRWDRFLLAYTAQRPTRLFDEID DARRAAPQAAAEPRVGALAALSTAERREHLAHLIRAEVAAVLGHGDDAAIDRDRAFRDLGFDSMTAVDLRNRLAAVTGVREAATVVF DHPTITRLADHYLERLGGGSGGGSGNEVTTLENDAAFIENENAYLEKEIARLRKEKAALRNRLAHKKLEHHHHHH

References:

- (1) Dutta, S.; Whicher, J. R.; Hansen, D. a; Hale, W. a; Chemler, J. a; Congdon, G. R.; Narayan, A. R. H.; Håkansson, K.; Sherman, D. H.; Smith, J. L.; et al. Structure of a Modular Polyketide Synthase. *Nature* **2014**, *510* (7506), 512–517.
- (2) Kapur, S.; Chen, A. J.; Cane D. E.; Khosla C.; Molecular Recognition between Ketosynthase and Acyl Carrier Protein Domains of the 6-Deoxyerythronolide B Synthase. *Proc. Natl. Acad. Sci.* **2010**, *107* (51), 22066-22071.
- (3) Lowry, B.; Robbins, T.; Weng, C.; Brien, R. V. O.; Cane, D. E.; Khosla, C. In Vitro Reconstitution and Analysis of the 6-Deoxyerythronolide B Synthase. *J. Am. Chem. Soc.* **2013**, *135*, 16809–16812.
- (4) Klaus, M.; D'Souza, A. D.; Nivina, A.; Khosla, C.; Grininger, M. Engineering of Chimeric Polyketide Synthases Using SYNZIP Docking Domains. *ACS Chem. Biol.* **2019**, *14* (3), 426–433.