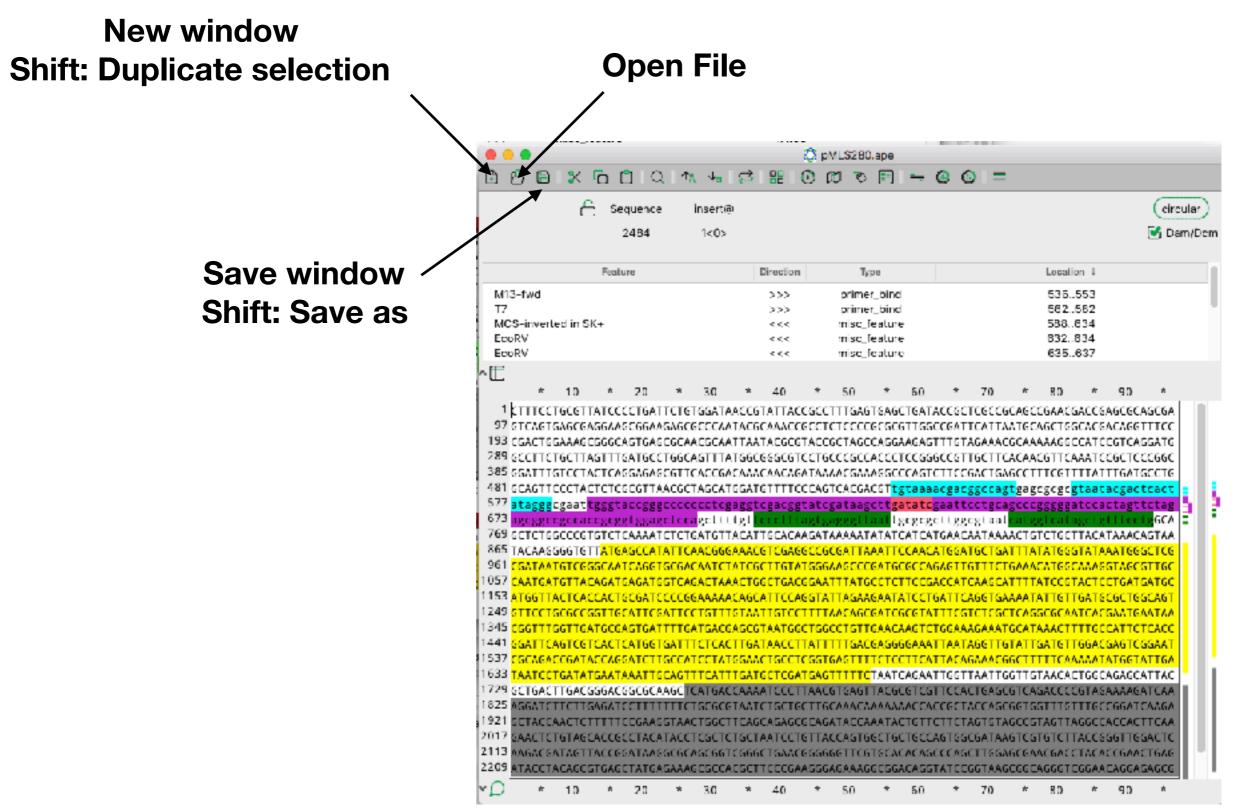
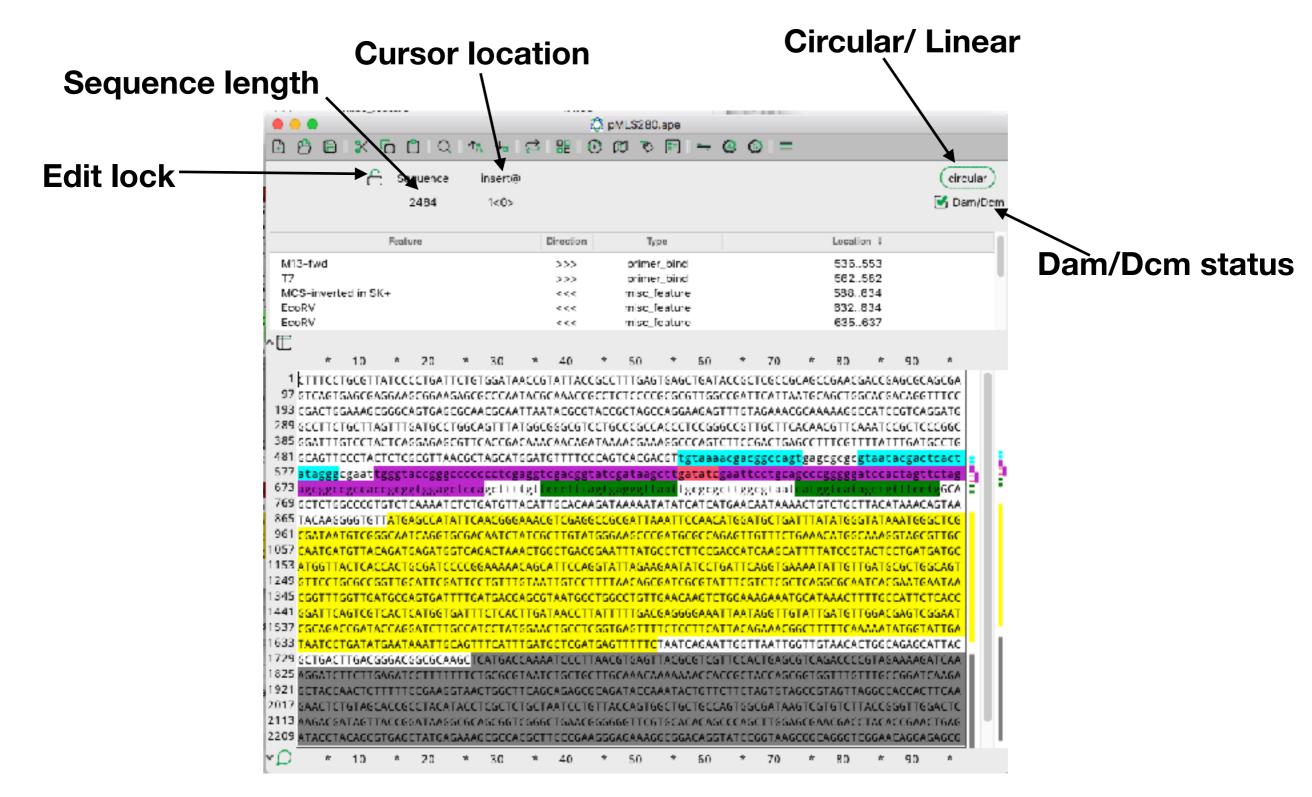
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Shortcuts

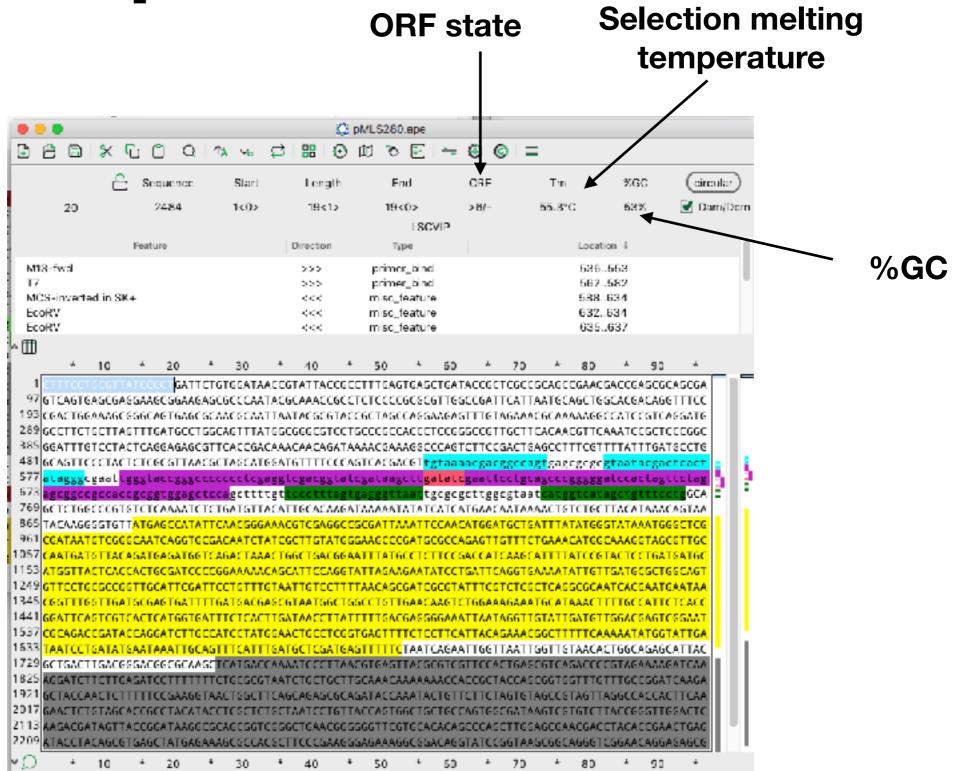
Command	Function
Ο	Open
N	New window
D	Duplicate selected region
Shift-D	Duplicate whole sequence
S	Save sequence
Shift-S	Save sequence as



Selection start Selection length Selection end double click to set

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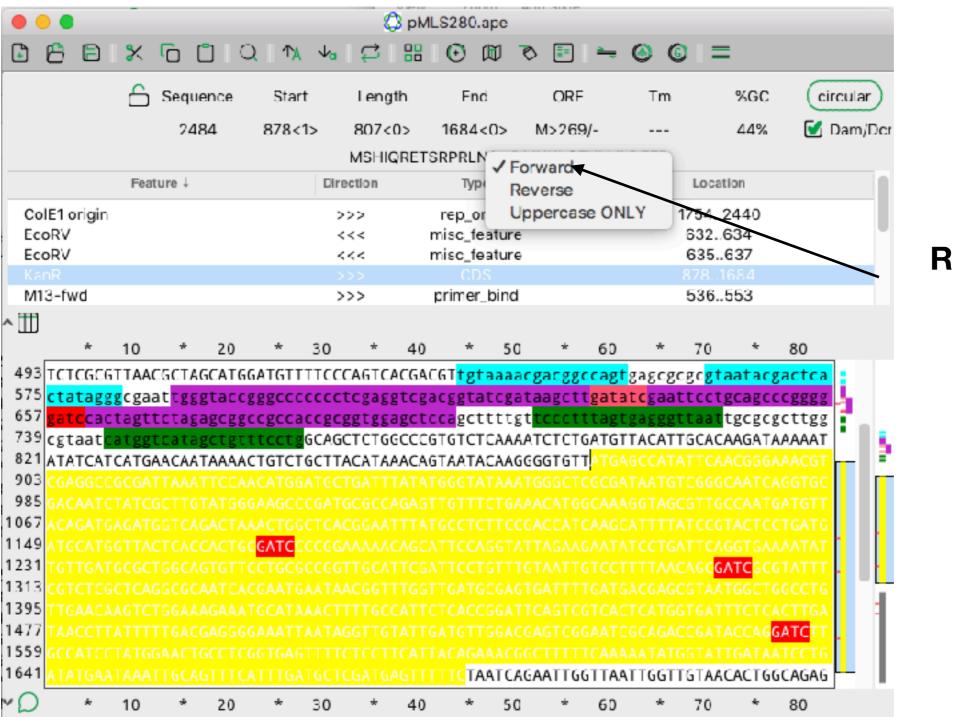




Selection translation

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821 ATATCAT 903 CGASGCC	CATGAACAATAAA GCGATTAAATTCCA	CTGTCTGCTTACATAAA ACATGGATSCTGATTTAT	AGI AATACAAGGG ATGGGTATAAATG	IGGCTCGCGATAAT			
985 <mark>6ACAATC</mark>						ATGTT	
1067 <mark>АСАБАТБ</mark> 1149 <mark>АТБСАТБ</mark>		AACTGGCTGACGGAATTT C <mark>GATC</mark> CCCGGAAAAACAG				AATAT	
1231 <mark>TGTTGAT</mark>		CCTSCGCCSGTTGCATTC			ACAGC <mark>GATC</mark> SCG	TATTT .	
1313 <mark>CGTCTCG</mark> 1395 TTGAACA							
1477 TAACCTT					ACCGATACCAG <mark>G</mark>	АТСТТ .	
1559 <mark>вссатос</mark> 1641 <mark>ататеаа</mark>			TTACAGAAACGGC	ATTGGTTAATTGG	TEGTATTGATAA TTGTAACACTGG	CAGAG	
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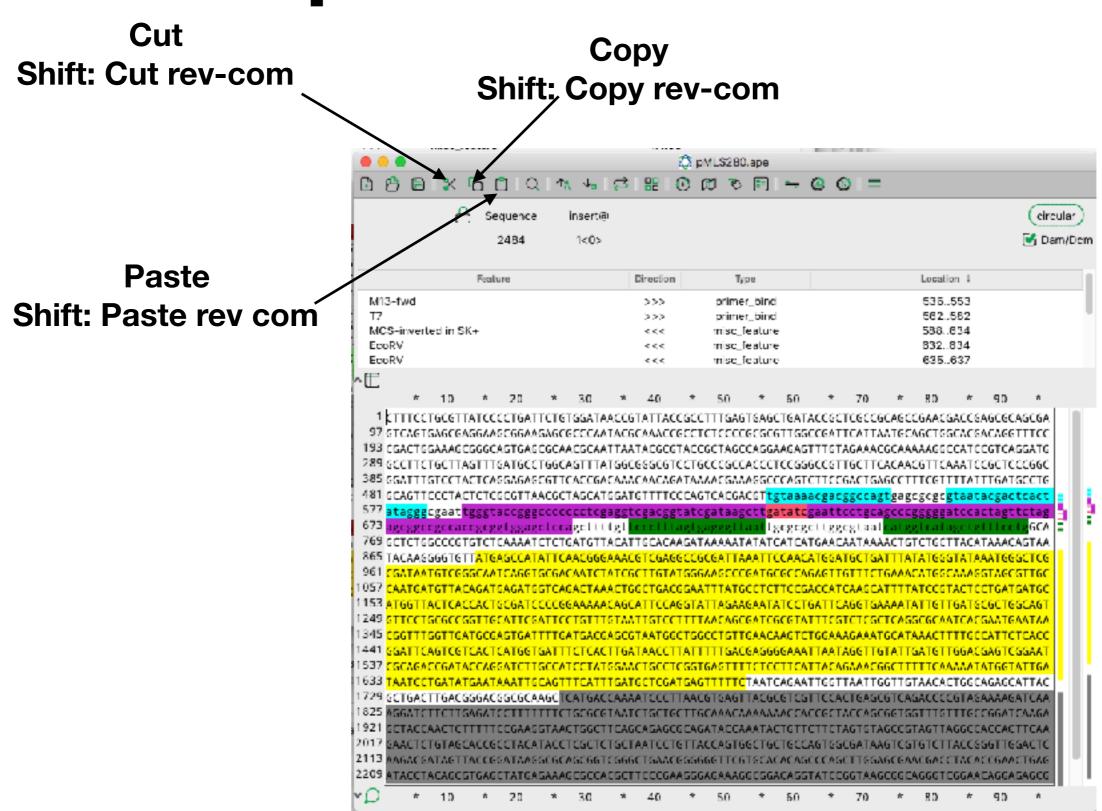
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Right click here to set the translation direction

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Shortcuts

Command	Function
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Shift-X	Cut rev-com
С	Сору
Shift-C	Copy rev-com
V	Paste
Shift-V	Paste rev-com

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⁴⁴¹ SGAFTCAGTCGTCACTCATGGTGATTTCTCACTTGATAACCTTATTTTTGACGAGGGGAAATTAATAGGTTGTATTGATGTTGGACGAGTCGSAAT ⁶³⁷ CSCAGACCGATACCAGSATCTTSCCATCCTATSSAACTSCCTCSSTGASTTTTCTCCTTCATTACAGAAACGGCTTTTTCAAAAATATGGTATTGA ⁶³³ TAATCCTGATATGAATAGATTGCAGTTTCATTTGATGCTCGATGAGTTTTTCTAATCAGAATTGSTTAATTGGTTGTAACACTGGCAGAGCATTAC ⁷²⁹ SCTGACTTGACSSGACSSCGCAAGCTCATGACCAAAATCCCTTAACGTSGTTACGCGTCSTTCCALTGAGCGTCASACCCCGTASAAAGATCAA					
³³⁷ CSCAGACCGATACCAGSATCTISCCATCCTATSSAACTSCCTCSSTGASTITICTCCTTCATTACAGAAACGGCTTTTTCAAAAATATGGTATTGA ³³³ TAATCCTGATATGAATAGATTSCAGTTCATTTGATGCTCGATGAGTTTTTCTAATCAGAATTGGTTAATTGGTTGTAACACTGGCAGAGCATTAC ⁷²⁹ SCTGACTTGACSSGACSSCGCAAGCTCATGACCAAAATCCCTTAACGTSAGTTACGCGTCSTTCCALTGASGGTCASAACCCCCGTASAAAAGATCAA	45 CGGTTTGGTTCATGCCASTGATTTTCATGACC	AGCGTAATGGCT	GGCCTGTTGAACAAGTCTGGAAA	GAAATGCATAAACTTTTGCCAT	TETEACE
⁵³³ <mark>TAATCCTGATATGAATAAATTSCAGTTTCATTTGATGCTCGATGAGTTTTTC</mark> TAATCAGAATTGGTTAATTGGTTGTAACACTGGCAGAGCATTAC ⁷²⁹ SCTGACTTGACSSGACSSCGCAAGC <mark>TCATGACCAAAATCCCTTAACGTSAGTTACGCGTCSTTCCACTGASCGTCASAACCCCCGTASAAAAGATCAA</mark>					
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21 BETACEAACTETTTTEEGAAGGTAACTEGETTEAGCAGAGCGEAGATAECAAATAETGTTETTETAGTGTAGCEGTAGTTAGGEEACCAETTEAA					
¹¹⁷ SAACTCTGTAGCACCGCCTACATACCTCSCTCTSCTAATCCTGTTACCASTGGCTGCCAGTGGCGATAAGTCGTGTCTTACCSGGTTGGACTC	21 SCTACCAACTCTTTTTCCGAAGGTAACTGGCT				
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Shortcuts

Command	Function
F	Find
Shift-F	Clear find highlight
G	Do find again
Shift-G	New feature from find highlight

Find dialog

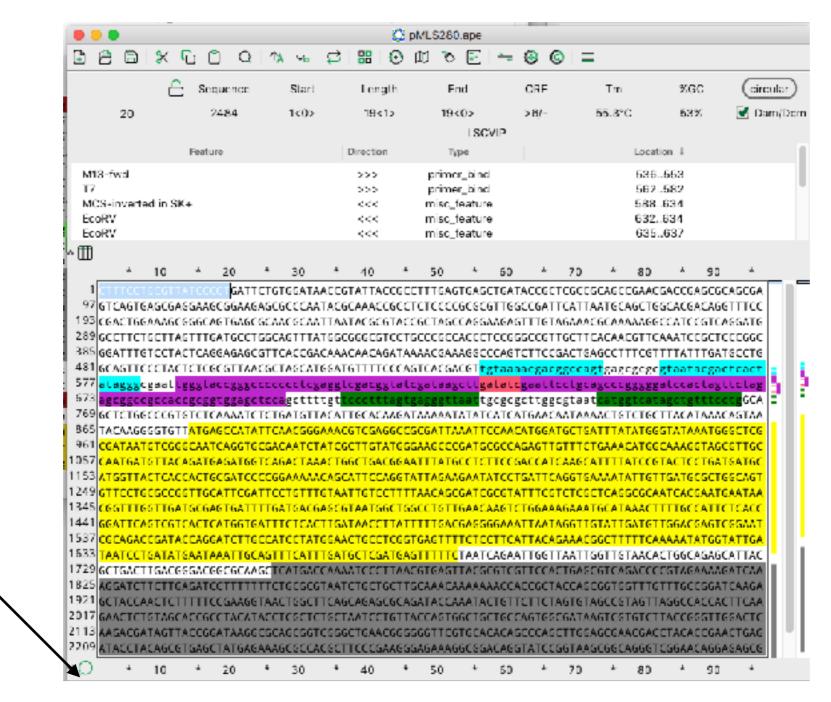
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Shortcuts

Command	Function
-	New Feature
K	Add features using feature library
Shift-K	Clear all features

Enzymes

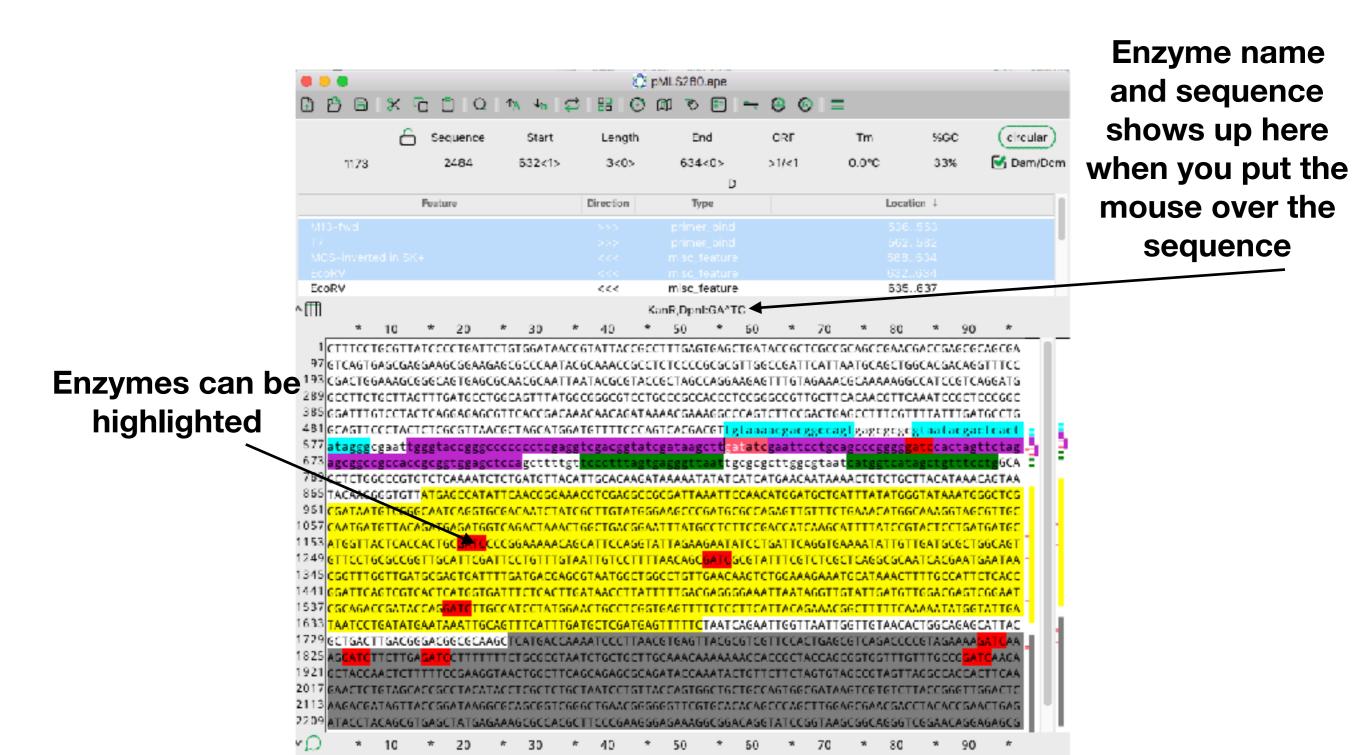
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1 CTTTCCTGCGTT 97 GTCAGTGAGCGA 93 CGACTGGAAAGC 89 GCCTTCTGCTTA 85 GGATTTGTCCTA 81 GCAGTTCCCTAC 77 alaggaCgacac 69 GCTCTGGCCCGT	GGAAGCGGAAG GGGCAGTGAGC GTTTGATGCCT CTCAGGAGAGC TCTCGCGTTAA CCCGCGTTAA CCCGCGTTCAAAATC GTCTCAAAATC	AGCGCCCAA GGCAACGCAA IGGCAGTTTA CGTTCACCGA CGCTAGCAT CCCCCCCCC CCCCCCCCCC	ATACGCAAAC ATTAATACGC ATGGCGGGCG ACAAACAACA IGGATGTTTT GGRTCRACE Ltgt <mark>CCCCT</mark>	CGCCTCTCCC GTACCGCTAG TCCTGCCCGC GATAAAACGA CCCAGTCACGA CCCAGTCACGA STATCSALAR TASTSASSST	CGCGCGTTGGC CCAGGAASAGT CACCCTCCGGC AAGGCCCAGTC ACGT <mark>tgtBabaa STIIggIBICS LBAT</mark> tgCGCGC TATATCATCAT	CGATTCA ITTGTAGA/ CCGTTGC ITTCCGAC CGGaCggCI CGGaCggCI CGGACGGC IGGACAAT/	TTAATGC/ AACGCAA/ TTCACAA/ TGAGCCT Cagtgag(COSTCT) aat <mark>Catg</mark> AAAACTG	AGCTGGCA AAAGGCCA CGTTCAAA ITCGTTTA CGCGC <mark>STA CCGCSCSTA CCGCTA CCGCTTA</mark>	CGACAGGI TCCGTCA(TCCGCTCA ATTTGAT(atacgaci caclagi tgtttcc	ETTEC SGATG COGGC SCCTG CCAC CLAS CCAC CCAC CCAC CCAC CCAC	
1 CTTTCCTGCGTT 97 GTCAGTGAGCGA 93 CGACTGGAAAGC 89 GCCTTCTGCTTA 85 GGATTTGTCCTA 81 GCAGTTCCCTAC 77 alagescesceac 69 GCTCTGGCCCGT 65 TACAAGGGSTGT 61 CGATAATGTCGG	SGAASCSGAAG SGSCAGTGAGG STTTGATGCCT CTCASGAGAGG TCTCSCGTTAA RERIBCCESE STCTCASAATC SCCTCASAATC SCATCAGGTC	AGCGCCCAA CGCAACGCAA IGGCAGTTTA CGTTCACCGA CGCTAGCAA CCCCCCCCA CCCCCCCCCAA CCCCCCCCCAA CCCCCC	ATACGCAAAC ATTAATACGC ATGGCGGGGGG ACAAACAACA IGGATGTTTT GREICRACE TACATTGCAC GAAACGTCGA IATCGCTTGT	CGCCTCTCCC GTACCGCTAG TCCTGCCCGC GATAAAACGA CCCAGTCACG STOLCZOLOG TASTSASSES AGGATAAAAA GGCCGCGATTA ATGGGAAGCC	CGCGCGTTGGC CCAGGAASAGT CACCCTCCGGC AAGGCCCAGTC ACGT <mark>CGTBBBBB SETTGTGTBBBBB SETTGTGTGBCGCCAC CGATGCGCCAC</mark>	CGATTCA ITTGTAGA/ CCGTTGC ITTCCGAC CGGCCGGC CGGCCGGC CGGCCGGC CGGCCGC CGGACAGT/ CGGATGCT CAGTTGTT	TTAATGC/ AACGCAA/ TTCACAA(TGAGCCT GAGCCT GAGCCT GATCAC GATCAC TCTCAAA(TCTCAAA(AGCTGGCA AAAGGCCA CGTTCAAA TTCGTTTA CGCGC <mark>gCa CCGCGCgCa CCTGCTTA CATGGCAA</mark>	CGACAGGI TCCGTCA TCCGCTCA TCCGCTCA ATTTGAT ATTGAT CATAGI CATAGAC AGGTAGC AGGTAGC	ITTEC SGATG CCGGC SCCTG CCACT CCACT CCACT CCACT SCCCG SCTCG CTTGC	
1 CTTTCCTGCGTT 97 GTCAGTGAAGCGA 93 CGACTGGAAAGC 89 GCCTTCTGCTTA 85 GGATTTGTCCTAC 81 GCAGTTCCCTAC 77 aloggacgacac 69 GCTCTGGCCCGT 65 TACAAGGGGTGT 61 CGATAATGTCGG 57 CAATGATGTTAC	GGAAGCGGAAG GGGCAGTGAGG GTTTGATGCCT CTCAGGAGAGG TCTCGCGTTAA CECEGEGTGAGAG GTCTCAGAGATG GCATCAGGTG AGATGAGATG	AGCGCCCAA GGCAACGCAA IGGCAGTTTA CGTTCACCGA CGCTAGCAT CCCCCCCCCA CCCCCCCCCA CCCCCCCCCA CCCCCC	ATACGCAAAC ATTAATACGC ATGGCGGGCG ACAAACAACA IGGATGTTTT CARRIERATC CARRIERATC CARCOTCGA CACCGCCTGA AACTGGCTGA	CGCCTCTCCC GTACCGCTAG TCCTGCCCGC GATAAAACGA CCCAGTCACGA Statcgataa tagtgagggt AAGATAAAAAA GGCCGCGATTA ATGGGAAGCC CGGAATTAA	CGCGCGTTGGC CCAGGCACAGAG CACCCTCCGGC ACGT <mark>tgtBabaa CTTgtBabaa CTTgtBabaa CGTtgtBabaa CGTTgtGCGCGC CGCTCTTCCG/ GCCTCTTCCG/</mark>	CGATTCA ITTGTAGA/ CCGTTGC ITTCCGAC CGaCggC CGACGGC CGACAAT/ CGGATGC CCATCAA	TTAATGC/ AACGCAA/ TTCACAA(TGAGCCT ⁺ Cagtgag(Cagtgag) AAAACTG ⁺ TCTGAAA(SCATTTA	AGCTGGCA AAAGGCCA CGTTCAAA TTCGTTTT CgCgCgCgta CgCgCgta CgCgCgta CgCGCGCgta CGTGCTA TATGGGTA ATCCGTAC	CGACAGGI TCCGTCA TCCGCTCA TCCGCTCA ATTTGAT ATTGAT CATAGI CATAGAC AGGTAGC AGGTAGC	ITTEC SGATG CCGGC SCCTG CCACT CCACT CCACT CCACT SCCCG SCTCG CTTGC	
1 CTTTCCTGCGTT 97 GTCAGTGASCGA 93 CGACTGGAAASC 89 GCCTTCTGCTTA 85 GGATTTGTCCTA 73 GCAGTTCCCTAC 73 accggccgccac 69 GCTCTGGCCCGT 65 TACAAGGGSTST 61 CGATAATGTCSG 57 CAATGATGTTAC 53 ATGGTTACTCAC	GGAAGCGGAAG GGGCAGTGAGC GTTTGATGCCT CTCAGGAGAGC TCTCGCGTTAA CCCGCGTTAA CCCGCGTTAA CCCGCGTCGAGAGC AGATGAGCCATA CCATGAGATGG CACTGCGATCC	AGCGCCCAA GGCAACGCAA IGGCAGTTTA GTTCACCGA CGCTAGCAT CCCCCCCCCC	ATACGCAAAC ATTAATACGC ATGGCGGGGGG ACAAACAACA IGGATGTTTT GARZ LOBALZ IGGATGTTTT GARZ LOBALZ IACATGCCTTG AACGCCTTGT AACAGCATTCC	CGCCTCTCCC GTACCGCTAG TCCTGCCCGC GATAAAACGA CCCAGTCACGA CCCAGTCACGA Lagtgagggt AAGATAAAAA GGCCGCGATTA ATGGGAAGCC CGGAATTTATA	CGCGCGTTGGC CCAGGAASAGT CACCCTCCGGC AAGGCCCAGTC ACGT <mark>tgtBaBB CLTgtBaBB</mark> tgcgcgc TATATCATCAT AAATTCCAACA CGATGCGCCAC GCCTCTTCCGA	CGATTCA ITTGTAGA/ CCGTTGC ITTCCGAC CGCGCGGCGGC CGCGCGGCGGC CGCCGCGCGC CGCCCACAA CCATCAAA ATTCAGG	TTAATGC/ AACGCAA/ TTCACAA(TGAGCCT ⁺ Cagtgag(Cagtgag) Cagtgag Cagtga	AGCTGGCA AAAGGCCA CGTTCAAA ITCGTTTI CgCgCgta CgCgCgta CgCgCgta CgCGCgta CgCGCGTA CGTGCTA ATCGTAC ATCGTAC	CGACAGGI TCCGTCA TCCGCTCA ATTTGAT(atacgaci tgtttcci tgtttcci CATAAAC/ TAAATGG AGGTAGC TCCTGAT(TGCGCTG	ITTCC GGATG CCGGC GCCTG CCACT GGCA GGCA G	
1 CTTTCCTGCGTT 97 GTCAGTGAGCGA 93 CGACTGGAAAGC 89 GCCTTCTGCTTA 85 GGATTTGTCCTA 81 GCAGTTCCCTAC 77 alaggscgaaal 73 agoggocgccac 69 GCTCTGGCCCGT 65 TACAAGGGGTST 61 CGATAATGTCGG 57 CAATGATGTTAC 53 ATGGTTACTCAC 49 GTTCCTGCGCCCG	GGAAGCGGAAG GGGCAGTGAGC GTTTGATGCCT CTCAGGAGAGC TCTCGCGTTAA CCCGCGTTAA CCCGCGTTAA CCCGCGTGGGTGAAAATC TATGAGCCATA GCAATCAGGTG CACTGCGATCGA	AGCGCCCAA GGCAACGCAA IGGCAGTTTA GTTCACCGA CGCTAGCAT CCGCTAGCAT CCGGCAAAC CCGGCAAAAA CCGGGAAAAA CCGGGAAAAA	ATACGCAAAC ATTAATACGC ATGGCGGGGGG ACAAACAACA IGGATGTTTT GARZ LORALS TACATGCCTC GAAACGTCGA GAAACGTCGA ACTGGCTGA ACAGCATTCC IGTAATTGTC	CGCCTCTCCC GTACCGCTAG TCCTGCCCGC GATAAAACGA CCCAGTCACGA CCCAGTCACGA Lagtgagggt AGGATAAAAA GGCCGCGATTA ATGGGAAGCC CGGAATTTACA CTTTTAACAG	CGCGCGTTGGC CCAGGAAGAGT CACCCTCCGGC AAGGCCCAGTC ACGT <mark>tgtbaba CGTtgtbaba CGTtgtbaba CGATCGCCAC CGATCCCCCAC CGATCGCGTAT</mark>	CGATTCA ITTGTAGA/ CCGTTGC ITTCCGAC CGCGCGGC CGCGCGGCGGC CGCGCGGCGGC CGCGCGCGC CCATCAA CCATCAGG ITTCGTCT	TTAATGC/ AACGCAA/ TTCACAA(TGAGCCT ⁺ Cagtgag(Cagtgag) Cagtgag Cagtgag Cagtgag Cagtgag Cagtgag Cagtta Cagtta CGCTCAG CGCTCAG	AGCTGGCA AAAGGCCA CGTTCAAA ITCGTTTA CGCGC <mark>STa CGCGCSTA CATGGCTA ATCGTAC ATCGTAC ATCGTAC</mark>	CGACAGGI TCCGTCA TCCGCTCA ATTTGAT(atacgaci caclag tgtttca CATAAAC/ TAAATGS AGGTAGCO TCCTGAT(TGCGCTGA ACGAATG/	ITTCC GGATG CCGGC GCCTG CCCTG CCCC GCCA GCCA	
1 CTTTCCTGCGTT 97 GTCAGTGAGCGA 93 CGACTGGAAAGC 93 CGACTGGAAAGC 89 GCCTTCTGCTAA 85 GGATTTGTCCTA 81 GCAGTTCCCTAC 73 GCGSCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCC	GGAAGCGGAAG GGGCAGTGAGG GTTTGATGCCT CTCAGGAGAGG TCTCGCGTTAA CTCGCGTTAA CTCGCGTTAA CTCGCGTTAA CTCGCGTGCGTCA GCCAGTGCGATCG CACTGCGATCGA TGCCAGTGATT	AGCGCCCAA GGCAACGCAA IGGCAGTTTA GTTCACCGA CGCTAGCAT CCGCTAGCAT CCGGCAAAC CCGGCAAAAA CCGGGAAAAA CCGGGAAAAA	ATACGCAAAC ATTAATACGC ATGGCGGGGGG ACAAACAACA IGGATGTTTT GGRTGTTTT GGRTGTTTT CARTICCAC GAACGTCGA ACAGCATTCC GAACGTCATTCC GAACGTAATGTC GAACGTAATG	CGCCTCTCCC GTACCGCTAG TCCTGCCCGC GATAAAACGA CCCAGTCACG Stategatag tagtgagggt AAGATAAAAA GGCCGCGATTA ATGGGAAGCC CGGAATTTATA AGGTATTAGA CTTTTAACAG GCTGGCCTGT	CGCGCGTTGGC CCAGGAAGAGT CACCCTCCGGC AAGGCCCAGTC ACGT <mark>tgtBaaa</mark> CTTgtBaaa CTTgtBaaa CGCTCTCCACA CGATCCCCACA CGATCCCCCACA CGATCCCCCACA CGATCCCCCACA CGATCCCCCACA CGATCCCCCACA CGATCCCCCACACACACACACACACACACACACACACACA	CGATTCA ITTGTAGA/ CCGTTGC ITTCCGAC Cgacggc Cgacggc CGACGAC CGACGAT CGATGTT CCATCAAA ATTCAGG ITTCGTCT CGAAAGA/	TTAATGC/ AACGCAA/ TTCACAA(TGAGCCT [*] Cagtgag(Cagtgag(Cagtgag) AAAACTG [*] TGATTTA [*] TGATATA TCTGAAA(SCATTTT/ TGAAAAT/ CGCTCAG(AATGCAT/	AGCTEGCA AAAGECCA CGTTCAAA ITCGTTTA CgCgCgta CgCgCgta CGCGCTA CATGCTA ATCCGTAC ATCCGTAC ACCGCAATC AAACTITI	CGACAGGT TCCGTCA TCCGCTCA ATTTGAT(atacgac1 caclagt Catacac Catacac TAAATGS AGGTAGCC TCCTGATG TCCTGATG ACGAATGS GCCATTC	ITTCC GGATG CCGGC GCCTG CCACT GCACT GCAC CCCG CCCG	
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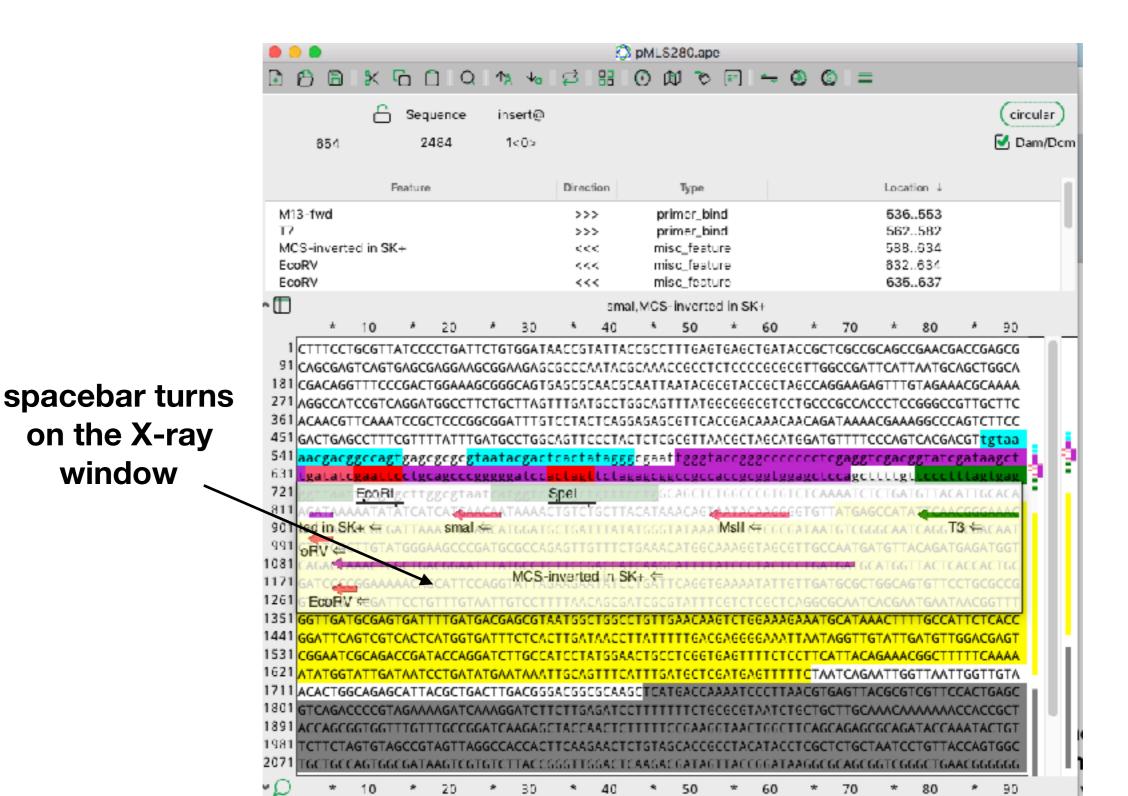
Select an enzyme to highlight

•••			Enzyme	e Selection			
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Aatll (0)	Banll (3)	BspLU11I (1)	Eco47III (0)	Kpnl (1)	NIaIV (5)	Sall (1)	Swal (0)
Absl (1)	Bbel (0)	BsrGI (0)	EcoNI (1)	Mael (5)	Notl (1)	SanDI (0)	Taql (8)
Acc65I (1)	BceAI (5)	BssHII (2)	EcoP15I (3)	Maell (4)	Nrul (1)	Sapl (1)	Tatl (0)
AccB1I (1)	Bcll (0)	BstAPI (0)	EcoRI (1)	MauBI (0)	Nsil (2)	SbfI (0)	TspEl (11)
Accl (1)	Bfml (4)	BstBI (0)	EcoRV (1)	Mbol (0)	Nspl (1)	Scal (0)	TspGWI (3)
Accll (14)	Bgll (0)	BstEll (0)	Fsel (0)	Mfel (0)	Olil (1)	Sdul (4)	Tth1111 (0)
AccIII (0)	BgIII (0)	BstXI (1)	FspAI (0)	Mlul (2)	Pacl (0)	SexAI (0)	Vspl (3)
Acll (1)	Blpl (0)	BstZ17I (0)	Fspl (0)	Mmel (7)	PfIMI (1)	Sfil (0)	Xbal (1)
Acyl (1)	BsaAI (0)	Bsu36I (0)	Haell (2)	Mrel (0)	Pfol (0)	Sfol (0)	Xcml (0)
AfIII (0)	BsaBI (0)	Btrl (0)	Haelll (15)	Mscl (0)	Pmel (0)	Sgfl (1)	Xhol (1)
AfIIII (3)	Bsal (0)	Cac8I (16)	Hhal (18)	Msel (9)	Pmll (0)	SgrAI (0)	Xholl (4)
Agel (0)	BsaWI (3)	Cfr10I (1)	Hincll (2)	Msll (1)	PpuMI (0)	SgrDI (0)	Xmal (1)
Alul (12)	BseRI (0)	Cfrl (3)	HindIII (1)	MspA1I (4)	PshAI (0)	Smal (1)	Xmnl (0)
AlwNI (1)	BseSI (2)	Clal (1)	Hpal (1)	Mwol (11)	Psil (0)	Smll (4)	Zral (0)
Apal (1)	BsiEl (4)	Dpnl (8)	Hpall (11)	Nael (0)	Pstl (1)	SnaBl (0)	
ApaLI (1)	BsiHKAI (2)	Dral (0)	Hpy188III (17)	Narl (0)	Pvul (1)	Spel (1)	
Apol (3)	BsiWI (0)	Drall (1)	Hpy8I (4)	Ncol (0)	Pvull (1)	SphI (0)	
Ascl (0)	BsiYI (10)	Dralll (0)	Hpy99I (6)	Ndel (0)	Rsal (3)	Srfl (0)	
Aval (2)	BsmBI (1)	Drdl (1)	HpyCH4III (7)	NgoMIV (0)	Rsrll (0)	Sspl (1)	
Avrll (0)	Bsp120I (1)	Eagl (1)	HpyCH4V (9)	Nhel (2)	Sacl (1)	Stul (0)	
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Enzyme highlights



X-ray window

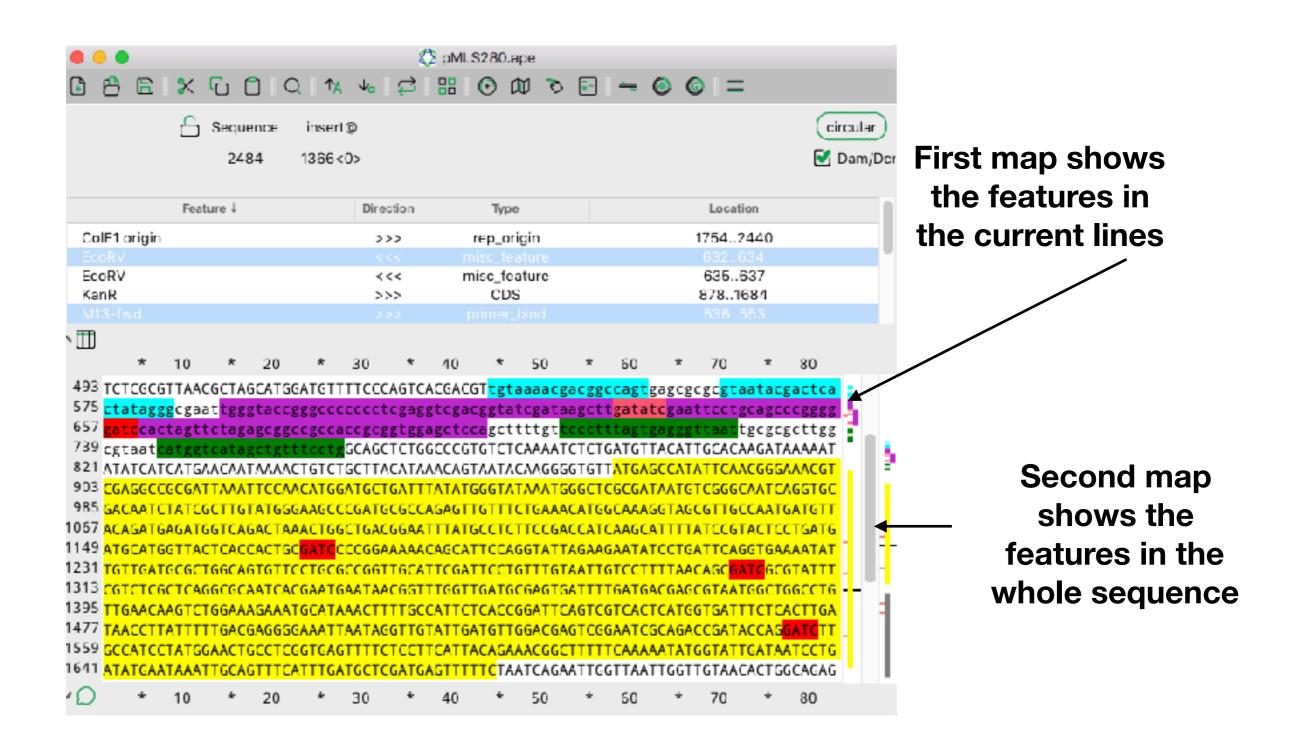


X-ray window

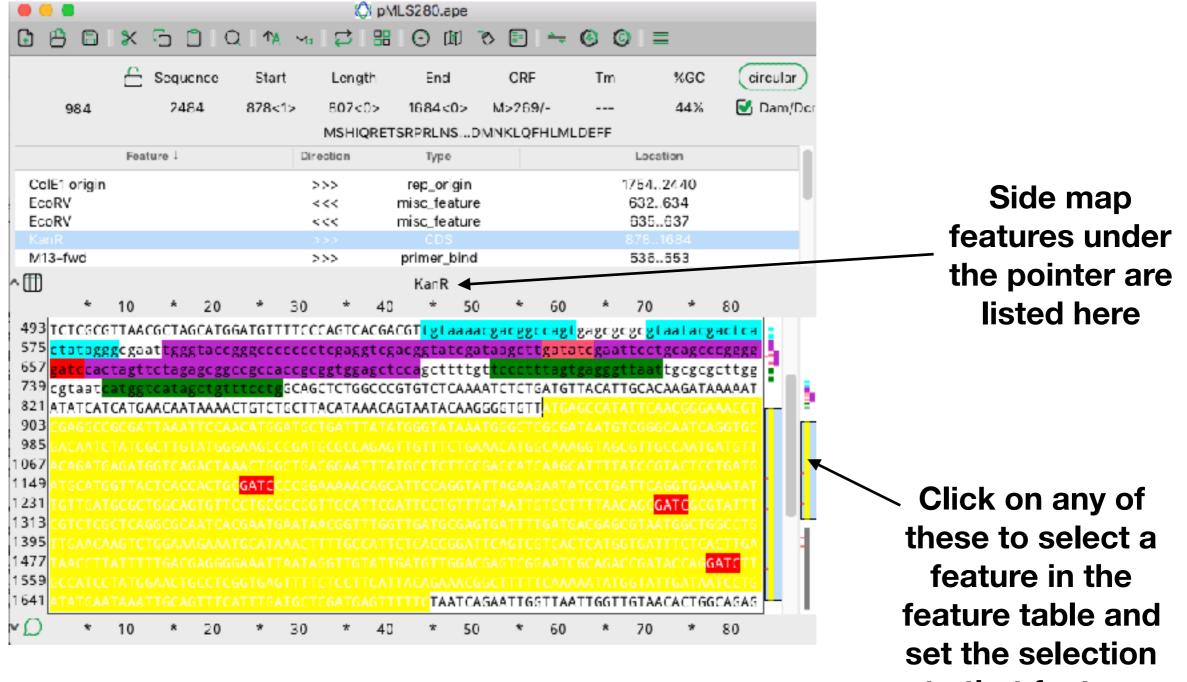
The X-ray window shows all the features and enzyme sites in a row

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451 GACTGAGCC 541 aacgacggco 631 Lgalalcean 721 Ep	TTTCGTTTTATTT <mark>ragt</mark> gagegege <mark>g</mark>	GCGGATTTS1 GATGCCTGGC taatacgact	CAGTTCCCTA	GAGAGCGTTC CTCTCGCGTT Cgcgaat <mark>t222</mark>	ACCGACAA AACGCTAG taccgggc	ACAACA CATGGA	GATAAAA TGTTTTC tcgaggt	CGAAA		AGTCTTC CGT <mark>tgta</mark> gataagc	c a t
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97 GTCAGTGA(93 CGACTGGA/ 89 GCCTTCTG(85 GGATTTGT) 81 GCAGTTCC(77 alaxx,cs; 73 agcggccg) 69 GCTCTGGC(65 TACAAGGG)	GCGAGGAA AAGCGGGG CTTAGTTT CCTACTC7 CTACTC7 CTACTC7 CTACTC7 CCACCGC CCGTG7C1	AGCGGAA (AGTGAG) IGATGCC (GGAGAG) (GCGTTA)	GAGCO CGCA/ TGGC/ CGTTO ACGCT	SCCCAA ACGCAA AGTTTA CACCGA TAGCAT	TACSO TTAAT TSGCO CAAAO GGATO	AAACCO ACGCG GGCGTO AACAG/ TTTTCO RALES	GCCTCT TACCGC CCTGCC ATAAAA CCAGTC	CCCCGCG TAGCCAG CGCCACC CGAAAGG ACGACGT Longell	CGTTGG GAAGAG CTCCGG CCCAGT tgtbaaa galalc	CCGATT TITGTA SCCGTT CTTCCG acgacg gaal to	CATTA/ GAAACO GCTTC/ ACTGAO gccagt	ATGCA SCAAA ACAAC SCCTT gago	GCTGG AAGGC GTTCA TCGTT gcgc <mark>g</mark>	CACGA CATCO AATCO TTATT taata	ACAGGT CGTCAG CGCTCC TGATG ACGaCt	GATG CGGC CCTG Cact
97 6TCAGTGA 93 CGACTGGA/ 89 6CCTTCTG 85 6GATTTGT 81 6CAGTTCC 77 aTaxxxc 73 agcsgocg 69 6CTCTGGC 65 TACAAGGG 61 CGATAATGT 57 CAATGATGT 53 ATGGTTAC	GCGAGGAA AAGCGGGG CTTAGTTT CCTACTC7 CTACTC7 CTACTC7 CTACTC7 CCACCGC CCGTG7C1	AGCGGAA (AGTGAG) IGATGCC (GGAGAG) (GCGTTA)	GAGCO CGCA/ TGGC/ CGTTO ACGCT	SCCCAA ACGCAA AGTTTA CACCGA TAGCAT	TACSO TTAAT TSGCO CAAAO GGATO	AAACCO ACGCG GGCGTO AACAG/ TTTTCO RALES	GCCTCT TACCGC CCTGCC ATAAAA CCAGTC	CCCCGCG TAGCCAG CGCCACC CGAAAGG ACGACGT Longell	CGTTGG GAAGAG CTCCGG CCCAGT tgtbaaa galalc	CCGATT TITGTA SCCGTT CTTCCG acgacg gaal to	CATTA/ GAAACO GCTTC/ ACTGAO gccagt	ATGCA SCAAA ACAAC SCCTT gago	GCTGG AAGGC GTTCA TCGTT gcgc <mark>g</mark>	CACGA CATCO AATCO TTATT taata	ACAGGT CGTCAG CGCTCC TGATG ACGaCt	GATG CGGC CCTG Cact
97 6TCAGTGA(93 CGACTGGA/ 89 6CCTTCTG 85 6GATTTGT(81 6CAGTTCC) 77 aloxxxc8; 73 agcsgcc8; 69 6CTCTGGC) 65 TACAAGGG(61 CGATAATG1 53 ATGGTTAC1 49 6TTCCTGC)	GCGAGGAA AAGCGGGG CTTAGTTT CCTACTCT CTACTCTC CTACTCTC CACCCGCS CCGTGTC CCGCGCAA TTACAGAT CCCGGTTC CCCGGTTC CCCGGTTC TGATGCC	AGCGGAA CAGTGAG IGATGCC AGGAGAG CGCGTTA CGCGTTA CAGCGTTA CAGCGTTA CAGCGAT CAGCGAT CAGTGAT CAGTGAT	GAGCO CGCA/ TGGC/ CGTTO ACGCTO ACGCTO ATTC/ GCGAO GTCAO ATTCO TTTG/	GCCCAA ACGCAA AGTTTA CACCGA IAGCAT GACGGA GATGTT GACGGG GAACCT ATGACG	TACSC TTAAT TSGCC CAAAC GGATG ACATT AAACO ATCSC ACTSC CAGCA GTAAT ASCST	AAACCC ACGCG GGCGTC AACAG/ TTTTTCC BUL23 CCCACA/ CCCACA/ CCCACA/ CTCCAG TTCCAC TTCCAC TTCCAC	GCCTCT TACCGC CCTGCC ATAAAA CCAGTC AGATAA GCCGCG TGGGAA GGAATT GGTATT TTTTAA CTGGCC	CCCCGCG TAGCCACG CGCCACC CGAAAGG ACGACGT ACGACGT ACGACGAT TATGCCT AGAAGAA CAGCGAT TGTTGAA	CGTTGG GAASAG CTCCGG CCCAGT tgtbaba tgcgcg tgcgcg TCATCA CCCAAC CCCCAC CCCCCA CAASTC	CCGATT ITTGTA SCCGTT CTTCCG acgacg coalle cttggc IGAACA ATGGAT CACCATC GATTCA ITTCGI IGGAAA	CATTA/ GAAACO GCTTC/ ACTGAO gctagt clgcagt ataat ATAAA/ GCTGAT AGCTAA CTCGCI GAAATO	ATGCA SCAAA SCAAA SCCTT gago SCTT TTAT SAAAC ITTAT SAAAC ITTAT SAAAC	AGCTEG AAGGC GTTCA TCGTT GCGC CCGC CCCGT CCCGT CCCGT CCCCA CCCCA AACTT	CACGA CATCO AATCO TTATT TATAT TATAA AAAGO ACTCO GATGO TTGCO	ACAGGI CGTCAG CGTCAG CGTCAG TGATGA CGCGGC CTGATG CGCTGA CATTCA	ITTEC GATG CCGC CCTG CCTG CCTG CCC CACA CCCG CACA CCCG CACA CACC
97 6TCAGTGA(93 CGACTGGA/ 89 GCCTTCTG(85 GGATTTGT(81 GCAGTTCC) 77 a1a222 c8: 73 a50590058 69 6CTCTGGC(65 TACAAGGG(61 CGATAATG 57 CAATGATG 53 ATGGTTAC) 49 6TTCCTGC(45 CGGTTGG1 41 6GATTCAG	GCGAGGA/ AAGCGGGG CTTAGTTT CCTACTCT CTACTCT CTACTCT CACCQCQ CCGTGTCT GCGCGGCA/ TCACCACT GCGGGCA/ TTACAGAT GCCGGTCC TCGTCACT	AGCGGAA CAGTGAG IGATGCC AGGAGAG GCGTTA GCGCGTTA GCGCGTTA GCGCGTTA GCGCGTTA GCGCGT GCGATG GCGATCG GCGTGGT GCGTGGT	GAGCO CGCA/ TGGC/ CGTT(ACGCT CCCCC CCCCC GCCCC GCCCC GCCCC ATTC/ GCCCCC ATTC/ GCCCCC ATTC/ GCCCCC	GCCCAA ACGCAA AGTTTA CACCGA TAGCAT GACCGA GATGTT AACSGG CAATCT GAAAAA CTGTTT ATGACG CCCAC	TACSC TTAAT TGGCC CAAAC GGATC GGATC ACATT AAACC ATCSC ACTSC CAGCA GTAAT TGAT	AAACCO ACGCG GGCGTU AACAG/ TTTTTCO RECORD COCCTS GCCCA TTGTA CTGACC CAATGGO AACCT	GCCTCT TACCGC CCTGCC ATAAAA CCAGTC LIC20 AGATAA GCCGCG TGGGAA GGAATT GGTATT TTTTAA CTGGCC TATTTT	CCCCGCG TAGCCAG CGCCACC CGANAGG ACGACGT Iaagc11 gsttaat AAATATA ATTAAAT GCCCGAT TATGCCT AGAAGAA CAGCGAT TGACGAG	CGTTGG GAASAG CTCCGG CCCAGT tgtbaa galalo tgcgcg tCATCA CCAAC GCGCCA CTTCCG TATCCT CGCGTA CAASTC GGGAAA	CCGATT TITGTA GCCGTT CTTCCG acgacg gaal fo Cttggo IGAACA ATGGAT GAGTTG ACCATC GATTCA ITGGAAA ITGAAA	CATTA/ GAAACO GCTTC/ ACTGAO gccagi clgcagi gtsat ATAAAA GCTGAI GCTGAI GGTCAA CTCGCI GAAATO GGTTGI		SCTGG AAGGC GTTCA TCGTT CGTCA CGCSC CCGCC CGCCA CGCCA CGCCA CGCCA CGCCA CGCCA CGCCA CGCCA CGCCA	CACGA CATCO AATCO TTATT TATAT COMPANY CACA CACA CACAC CACAC CACAC CACAC	ACAGST CGTCAG CGTCAG CGCTCC TTGATG CGCTCC TGATGS GTAGCC CTGATG CGCTGC GATGA CATTCT GAGTCC	ITTEC GATG CGGC CCTG CCTG CCTG CCTG CCTG CCT
97 6TCAGTGA(93 CGACTGGA/ 89 6CCTTCTG(85 6GATTTGT(81 6CAGTTCC) 77 atoxxxcc; 73 agcsgccc; 69 6CTCTGGC(61 CGATAATG1 53 ATGGTTAC) 49 6TTCCTGC(41 6GATTCAG1 31 CGCAGACC(33 TAATCCTG/	GCGAGGA/ AAGCGGGG CTTAGTTT CCTACTCT CTACTCTC CTACTCTC CTACTCTC CTACTCTC CCGCGGCA/ TCGGGCA/ TCGGGCA/ TCGCGGTTC TCGTCACT GCCGGTTC CTGATCCAC ATATGAA	AGCGGAA CAGTGAG IGATGCC AGGAGAG GGGTTA CGGGTTA CGGGTTA CGGGTA CGGGTA CGGGTA CGGGAT CAGGAT CGGATCG CATTCG CATTCG CATTCG CATTCG CATCGT CATGGT CATGGT CATCGT	GAGCO CGCA2 TGGC2 CGTTO ACGCTO ACGCTO ACTCO ATTCO GCCA0 GCCA0 GATTT GCCA1 GCCA1 CAGTT	GCCCAA ACGCAA ACGCAA AGTTTA CACCGA IAGCAT IGCCAT GACGGG ICCCAC ICCCAC ICCCAT ICCCAT	TACSC TTAAT TSGCC CAAAC GGATC ACATT AAACC ATCSC ACTSC CACCA GTAAT ASCST TTGAT GGAAC TGATC	AAACCC ACGCG GCCCG ACAGA TTTTC CACAGA TTTTC CCCAGA TTCCAG TTCCAG TCCAC TCCAC CACCC CACCC CACCC CCCCCC CCCCCA	GCCTCT TACCGC CCTGCC ATAAAA CCAGTC IIIC23 AGATAA GCCGCG TGGGAA GGAATT GGTATT TTTTAA CTGGCC TATTTT CGGTGA TGAGTT	CCCCGCG TAGCCACG CGCCACC CGAAAGG ACGACGT CGACACGT CGCCGAT TATGCCT AGAAGAA CAGCGAT TGTTGAA TGACGAG GTTTTCT TTTCTAA	CGTTGG GAASAG CTCCGG CCCAGT tgtbaaa tgcgcg tgalatc tgcgcg tcATCA CCTCCG CCTCCG CGCGTA CCGCGTA CCGCGTA CCCTCA CCCTCA TCAGAA	CCGATI ITTGTA SCCGTI CTTCCG acgacg gallo Cttggc IGAACA ATGGAT CAGTTCA ITTCATA ITTACAG ITGGTT	CATTA/ GAAACO GCTTC/ ACTGAO gccag1 clgcag gtaat ATAAAA GCTGAI GCTGAI GGTGAI GGTGAI GGTGAI GAAACO AATTGO	ATGCA SCAAA SCAAA SCAAC SCCTT Bago CCTT CAAAC CATA SCATA SCTTT SCTTT SCTTTS	AGCTEG AAGGC GTTCA TCGTT CGCT CGCS CGCS ATGCC ATGCC CGCAA AGCTT CGCAA CACTT CGCAA CACTT	CACGA CATCO AATCO TTATT taata Cocac gotgi TACAI TACAI TACAI TACAI CACO GATGO TCACO GGACO AAATA TGGCA	ACAGGT CGTCAG CGTCAG CGCTCA TGATG CGCGCCC CGCTGC CGCTGC CGCTGC CATTCT CASTCC ACTCCT CASTCC ACGCTAG	ITTEC GATG GATG CCGGC CCTG CCTG CAGT CAGC CAGT CAGC CAGT CAGC CAGT CAGC CAGT CAGC CAGT CAGC CAGT
97 6TCAGTGA(93 CGACTGGA/ 89 6CCTTCTG(85 GGATTTGT(81 6CAGTTCC) 77 aloxoxcs; 73 agcggccg; 69 6CTCTGGC; 61 CGATAATG1 53 ATGGTTAC1 49 6TTCCTGC; 49 6TTCCTGC; 41 CGATTCAG1 37 CGCAGACC; 33 TAATCCTG/ 29 6CTGACT(GCGAGGAA AAGCGGGG CTTAGTTT CCTACTCT CTACTCT CTACTCT CACCCC CCGTGTC CCGCGCAA TCACCACT CCGCGGTC CCGTCACT CACCACT CACCACT CACCACT CACCACA CCGTCACT CACCACA CATATGAA SACGGGAA	AGCGGAA CAGTGAG IGATGCC AGGAGAG IGAGAGAG IGCGTTA AGCCATA IGAGATG IGAGATG IGAGATG IGAGATG IGAGATG IGAGATG IGAGATG IGAGATG IGAGATG IGAGATG IGAGATG	GAGCO CGCA2 TGGC2 CGTTO ACGCTO ACGCTO ACCCCO ATTC2 GCCA0 GCCA0 GCCA1 GCCA1 GCCA1 CAGTT	GCCCAA ACGCAA AGTTA CACCGA IACCGA IACCGA GATGTT ACCGGS CAATCT ACCGGS ICTCAC ICCTAT ICCTAT	TACSC TTAAT TSGCC CAAAC GGATC ACAT AAACC ACTSC ACTSC ACTSC ACTSC ACTSC CAGCA TSATC CAAAA	AAACCC ACGCG GGCGTC ACAG/ TTTTCC CACAG/ TTGTA CTGACG/ AACCT AATGGC AACCT CACGCC CACGCC CCCCC CCCCC	GCCTCT TACCGC CCTGCC ATAAAA CCAGTC I J C23 AGATAA GCCGCG TGGGAA GGAATT GGTATT TTTTAA CTGGCC TATTTT CGGTGA TGAGTT TAACGT	CCCCGCG TAGCCACG CGCCACC CGANAGG ACGACGT BETTBAT ACGACGAT TATGACGAT TATGCCT AGAAGAA CAGCGAT TGTTGAA TGACGAG GTTTTCTAA GAGTTAC	CGTTGG GAASAG CTCCGG CCCAGT tgtbaba tgcgcg TCATCA TCCAAC GCGCCA CTTCCG CAASTC CGCGTA CCTTCA CGCGAA CCTTCA CCTTCA TCASAA GCGTCG	CCGATI ITTGTA SCCGTT CTTCCG acgacg goal IC Cttggc IGAACA ATGGAT CACCATC GATICA ITACAG ITACAG ITACAG	CATTA/ GAAACO GCTTC/ ACTGAO gctag1 clgcag gtaat ATAAA/ GCTGAI GCTGAI GGTGAI GAAATG GAAATG (AATTGO TGAGCO	ATGCA SCAAA SCAAA SCAAC SCCTT Bago SCTT SAAAC SCTT STTAT SCATA SCTT STTGT	AAGGC AAGGC GTTCA TCGTT SCGC CCGC ATGGC ATGGC ATGGC AGGC AACTT AACAC ACCCC	CACGA CATCO AATCO TTATT TATAT COOR SCORE TACAT TATAA AAAGO AATCO GATGO TCACO GGACO AAATA TGGCA GTAGA	ACAGGI CGTCAG CGTCAG CGTCAG TGATG CGCGC CTGATG CGTGATG CGTGATG CGTGATG CATTCI CATTCI CATCAGA AGAGCA AAAAGA	ITTEC GATG GATG CCGC CCTG CCTG CACC CACT CACC CACT CACC CACT CACC CACT CACC CACT CACC CACT CACC CACT CACC CACT CACCA
97 6TCAGTGA(93 CGACTGGA/ 89 6CCTTCTG(85 6GATTTGT(81 6CAGTTCC) 77 atoxxxcc; 73 agcsgccc; 69 6CTCTGGC(61 CGATAATG1 53 ATGGTTAC) 49 6TTCCTGC(41 6GATTCAG1 31 CGCAGACC(33 TAATCCTG/	GCGAGGAA AAGCGGGG CTTAGTTT CCTACTCT CTACTCT CTACTCTC CTACTCTC CCGCGCG CCGCGCGC CCGCGCGCAA TACCACAC CCGCGGTTC CCGCCACT CACCACAC CCGCGGCAA CTACCACACACACACACACACACACACACACACACA	AGCGGAA CAGTGAG IGATGCC AGGAGAG IGCGTTA IGCGTTA IGCGCTA IGCGATC IGCGATC IGCGATCG IGCGATCT IGAGATGA IGAGATCA IGAGATCA IGAGATCA IGAGATCA IGAGATCA IGAGATCA IGAGATCA IGAGATCA IGAGATCA IGAGATCA IGAGATCA IGAGATCA IGGCGCA	GAGCO CGCA2 TGGC2 CGTTO ACGCTO ACGCTO ACCCCO ATTC2 GCCA0 GCCA0 GCCA1 GCCA1 GCCA1 GCCA1 GCCA1 GCCA1 GCCA1 CAGTTO	GCCCAA ACGCAA ACGCAA AGTTTA CACCGA IAGCAT GCCCAA GATGTT AACSGS CAATGT AACSGS CAATGT ATGACG ICCCAAT ICCCAAT ICCCAAT ICCCAAT ICCCAAT ICCCAAT	TACSC TTAAT TSGCC CAAAC GGATG ACATT AAACG ACCSC ACTSC ACTSC ACTSC CAGCA GTAAT GSAAC TSATC	AAACCC ACGCG GGCGTC ACAG/ TTTTCC BULXX CCCACA/ CCCACA/ CCCACA/ CCCACA/ CCCACA/ CCCACA/ CCCACA/ CCCACA/ CCCCC CACCCC CCCCACA/ CCCCCCCC	GCCTCT TACCGC CCTGCC ATAAAA CCAGTC I J C 2 J AGATAA GCCGCG TGGGAA GGAATT TGGGAA GGAATT TTTTAA CTGGCC TATTTT CGGTGA TGAGTT TAACGT CTTGCA	CCCCGCG TAGCCACG CGCCACC CGAAAGG ACGACGT ACGACGT BETTBAT AAATATA ATTAAAT GCCCGAT TATGCCT AGAAGAA CAGCGAT TGTCGAA GATTAC AACAAAA	CGTTGG GAASAG CTCCGG CCCAGT tgtbaa galal C tgtbaa tgcgcg TCATCA CCACG CCCCA CCTCCG CGCGTA CCCTCA CCCTCA CCCTCA CCCTCA CCCTCA CCCTCA	CCGATT ITTGTA SCCGTT CTTCCG acgacg calle cttggc IGAACA ATGGAT GAGTTCA CATTCA ITACAG ITACAG ITACAG ITACAG ITACAG	CATTA/ GAAACO GCTTC/ ACTGAO gctagt clgcag gtaat ATAAA/ GCTGAI TTTCTC AAGCAI GGTTGI GAAATO GGTTGI AAACO AATTSO CTCACO	ATGCA SCAAA SCAAA SCCTT gago SCTTT SAAAC ITTAT	AAGGO AAGGO GTTCA TCGTT CGTTCA CGCTC CCGCC CGCCC CGCCC CGCCC CCCCC CACCCC CTTCT	CACGA CATCO AATCO TTATT TATAT COM STACAT TACAT TACAT CACC CACCO CA	ACAGGI CGTCAG CGTCAG CGTCAG TGATG TGATG TGATG CGCTGATG CGCTGATG CGCTGATG CGCTGATG CGCTGATG CGCTGATG CGCTGATAG AGAGCA AGAGCA	ITTOC GATG GATG CCGC CCTG CCTG CCCC CACC CCCC CACC CA

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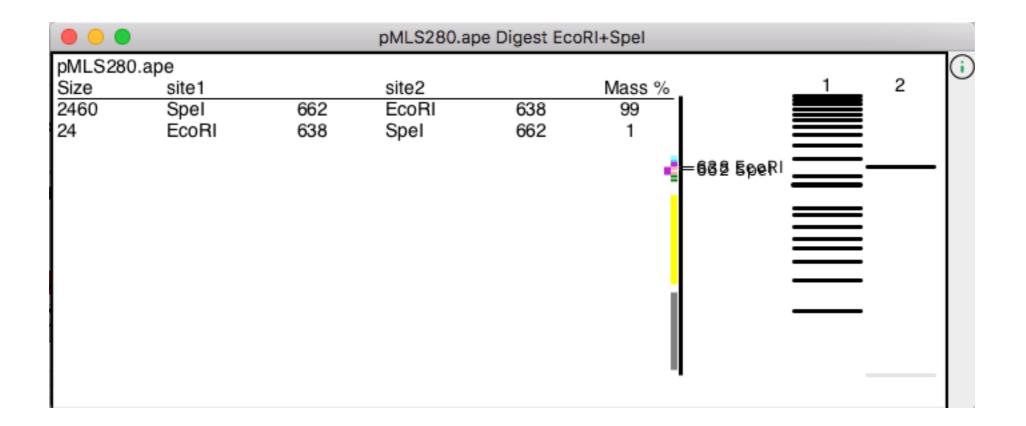
• • •			Enzyme	e Selection			
Window 🤇		pMLS	280.ape		· · · · ·	Selection: 1	- 2484 🗹 Dam/D
Aatll (0)	Banll (3)	BspLU11I (1)	Eco47III (0)	Kpnl (1)	NIalV (5)	Sall (1)	Swal (0)
Absl (1)	Bbel (0)	BsrGI (0)	EcoNI (1)	Mael (5)	Notl (1)	SanDI (0)	Taql (8)
Acc65I (1)	BceAI (5)	BssHII (2)	EcoP15I (3)	Maell (4)	Nrul (1)	Sapl (1)	Tatl (0)
AccB1I (1)	Bcll (0)	BstAPI (0)	EcoRI (1)	MauBI (0)	Nsil (2)	SbfI (0)	TspEl (11)
Accl (1)	Bfml (4)	BstBI (0)	EcoRV (1)	Mbol (0)	Nspl (1)	Scal (0)	TspGWI (3)
Accll (14)	Bgll (0)	BstEll (0)	Fsel (0)	Mfel (0)	Olil (1)	Sdul (4)	Tth1111 (0)
AccIII (0)	BgIII (0)	BstXI (1)	FspAI (0)	Mlul (2)	Pacl (0)	SexAI (0)	Vspl (3)
Acll (1)	Blpl (0)	BstZ17I (0)	Fspl (0)	Mmel (7)	PfIMI (1)	Sfil (0)	Xbal (1)
Acyl (1)	BsaAI (0)	Bsu36I (0)	Haell (2)	Mrel (0)	Pfol (0)	Sfol (0)	Xcml (0)
AfIII (0)	BsaBI (0)	Btrl (0)	HaellI (15)	Mscl (0)	Pmel (0)	Sgfl (1)	Xhol (1)
AfIIII (3)	Bsal (0)	Cac8I (16)	Hhal (18)	Msel (9)	Pmll (0)	SgrAI (0)	Xholl (4)
Agel (0)	BsaWI (3)	Cfr10I (1)	Hincll (2)	MsII (1)	PpuMI (0)	SgrDI (0)	Xmal (1)
Alul (12)	BseRI (0)	Cfrl (3)	HindIII (1)	MspA1I (4)	PshAI (0)	Smal (1)	Xmnl (0)
AlwNI (1)	BseSI (2)	Clal (1)	Hpal (1)	Mwol (11)	Psil (0)	Smll (4)	Zral (0)
Apal (1)	BsiEl (4)	Dpnl (8)	Hpall (11)	Nael (0)	Pstl (1)	SnaBl (0)	
ApaLI (1)	BsiHKAI (2)	Dral (0)	Hpy188III (17)	Narl (0)	Pvul (1)	Spel (1)	
Apol (3)	BsiWI (0)	Drall (1)	Hpy8I (4)	Ncol (0)	Pvull (1)	SphI (0)	
Ascl (0)	BsiYI (10)	Dralll (0)	Hpy99I (6)	Ndel (0)	Rsal (3)	Srfl (0)	
Aval (2)	BsmBI (1)	Drdl (1)	HpyCH4III (7)	NgoMIV (0)	Rsrll (0)	Sspl (1)	
Avrll (0)	Bsp120I (1)	Eagl (1)	HpyCH4V (9)	Nhel (2)	Sacl (1)	Stul (0)	
Select Enzyn	nes unique	(1) 🗸	All 🗸	Select	De-select	AND clear	r all Sel to Mem
Perform Acti	on Graphic M	ap Graphic	Map +U Dig	gest Dige	est with All	Highlight	Text Close
Keep Sele	ctor Dialog Oper	1					
					\mathbf{i}		

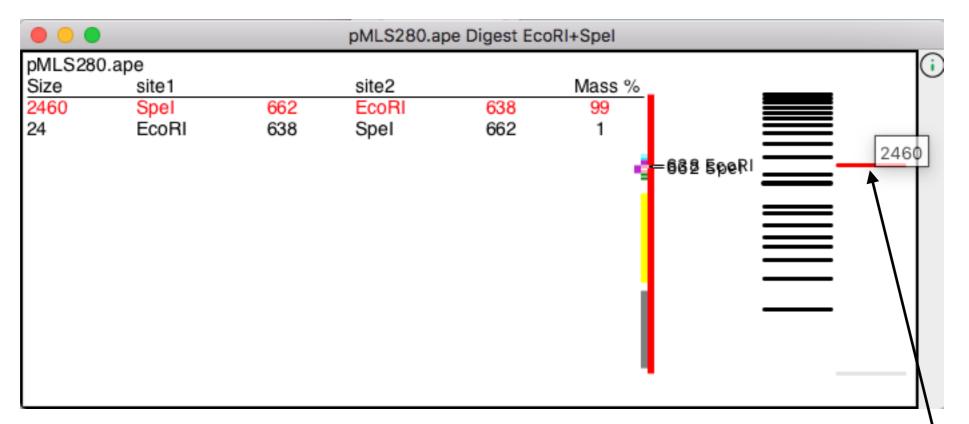
Click here to digest the sequence with each enzyme individually

pMLS280.ape Digest		
Lane 1: 1Kb plus Sizes: 100, 200, 300, 400, 500, 650, 850, 1000, 1650, 2000, 3000, 4000, 5000, 6000, 7000, 8000, 9000, 10000, 11000, 12000 Lane 2: pMLS280 digested with: EcoRI 2484 Lane 3: pMLS280 digested with: SpeI 2484	2	3

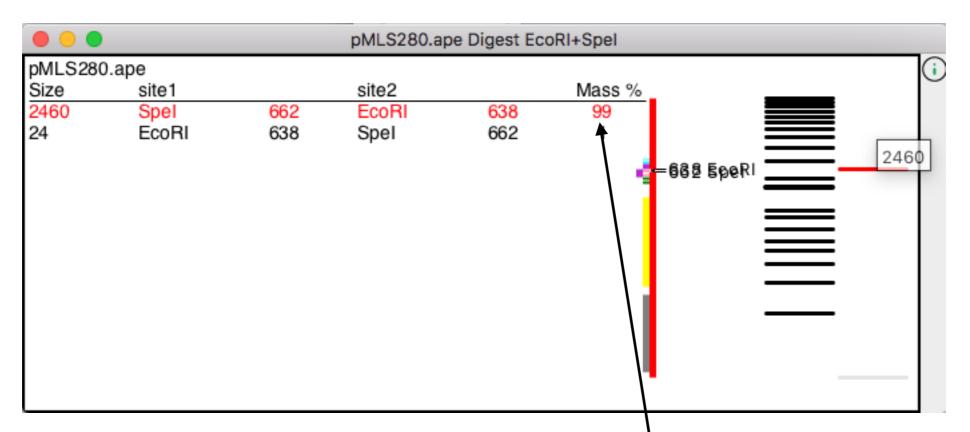
•••			Enzyme	e Selection				
Window 🤇		pMLS	280.ape		· (Selection: 1	- 2484 🗹 Dam/Do	cm
Aatll (0)	Banll (3)	BspLU11I (1)	Eco47III (0)	Kpnl (1)	NIaIV (5)	Sall (1)	Swal (0)	1
Absl (1)	Bbel (0)	BsrGI (0)	EcoNI (1)	Mael (5)	Notl (1)	SanDI (0)	Taql (8)	
Acc65I (1)	BceAI (5)	BssHll (2)	EcoP15I (3)	Maell (4)	Nrul (1)	Sapl (1)	Tatl (0)	
AccB1I (1)	Bcll (0)	BstAPI (0)	EcoRI (1)	MauBI (0)	Nsil (2)	Sbfl (0)	TspEI (11)	
Accl (1)	Bfml (4)	BstBl (0)	EcoRV (1)	Mbol (0)	Nspl (1)	Scal (0)	TspGWI (3)	
Accll (14)	Bgll (0)	BstEll (0)	Fsel (0)	Mfel (0)	Olil (1)	Sdul (4)	Tth1111 (0)	
AccIII (0)	BgIII (0)	BstXI (1)	FspAI (0)	Mlul (2)	Pacl (0)	SexAI (0)	Vspl (3)	
Acll (1)	Blpl (0)	BstZ17I (0)	Fspl (0)	Mmel (7)	PfIMI (1)	Sfil (0)	Xbal (1)	
Acyl (1)	BsaAI (0)	Bsu36I (0)	Haell (2)	Mrel (0)	Pfol (0)	Sfol (0)	Xcml (0)	
AfIII (0)	BsaBI (0)	Btrl (0)	Haelll (15)	Mscl (0)	Pmel (0)	Sgfl (1)	Xhol (1)	
AfIIII (3)	Bsal (0)	Cac8I (16)	Hhal (18)	Msel (9)	Pmll (0)	SgrAl (0)	Xholl (4)	
Agel (0)	BsaWI (3)	Cfr10I (1)	Hincll (2)	MsII (1)	PpuMI (0)	SgrDI (0)	Xmal (1)	
Alul (12)	BseRI (0)	Cfrl (3)	HindIII (1)	MspA1I (4)	PshAI (0)	Smal (1)	Xmnl (0)	
AlwNI (1)	BseSI (2)	Clal (1)	Hpal (1)	Mwol (11)	Psil (0)	Smll (4)	Zral (0)	
Apal (1)	BsiEl (4)	Dpnl (8)	Hpall (11)	Nael (0)	Pstl (1)	SnaBI (0)		
ApaLI (1)	BsiHKAI (2)	Dral (0)	Hpy188III (17)	Narl (0)	Pvul (1)	Spel (1)		
Apol (3)	BsiWI (0)	Drall (1)	Hpy8I (4)	Ncol (0)	Pvull (1)	SphI (0)		
Ascl (0)	BsiYI (10)	Dralll (0)	Hpy99I (6)	Ndel (0)	Rsal (3)	Srfl (0)		
Aval (2)	BsmBl (1)	Drdl (1)	HpyCH4III (7)	NgoMIV (0)	Rsrll (0)	Sspl (1)		
Avrll (0)	Bsp120I (1)	Eagl (1)	HpyCH4V (9)	Nhel (2)	Sacl (1)	Stul (0)		P
Select Enzym	nes unique	(1) •	All 🗸	Select	De-select	AND clear	r all Sel to Mem)
Perform Actio	on Graphic M	ap Graphic	Map +U Dig	gest Dige	est with All	Highlight	Text Close)
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	eter blaidg oper	•		\mathbf{X}				

Click here to digest the sequence with each enzyme at one time





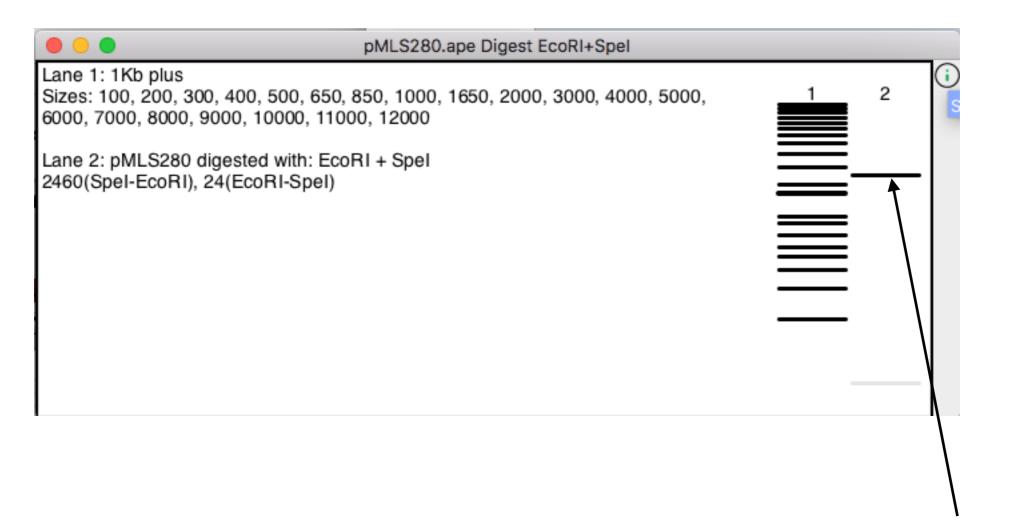
Mouse here to highlight the band Double-click to select that region in the sequence



Mass % shows how much of the total DNA each band represents Use this to decide how much DNA to load on a gel. 50 ng is about the least DNA you can see on a gel.

• • •	pMLS280.ape Digest EcoRI+Spel		
Lane 1: 1Kb plus Sizes: 100, 200, 300, 400 6000, 7000, 8000, 9000,	, 500, 650, 850, 1000, 1650, 2000, 3000, 4000, 5000, 10000, 11000, 12000	1	2
Lane 2: pMLS280 digeste 2460(Spel-EcoRI), 24(Ec			

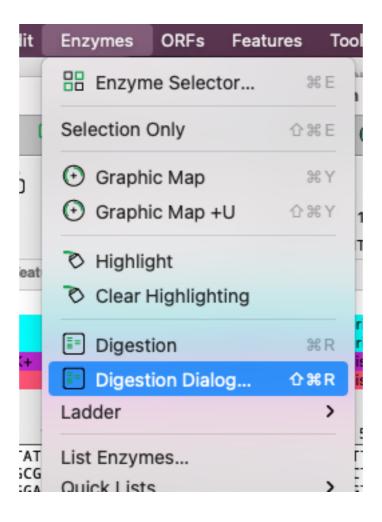
Click here to see a list of digest lanes

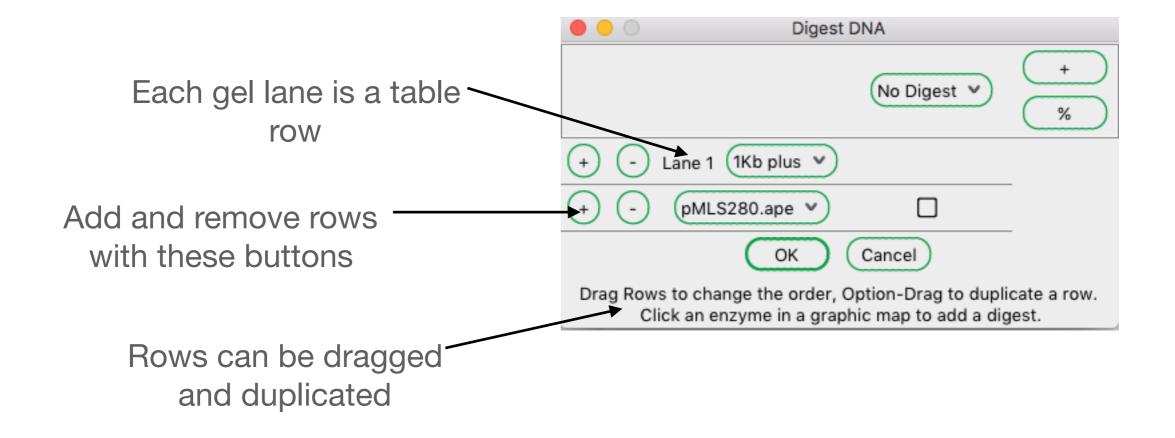


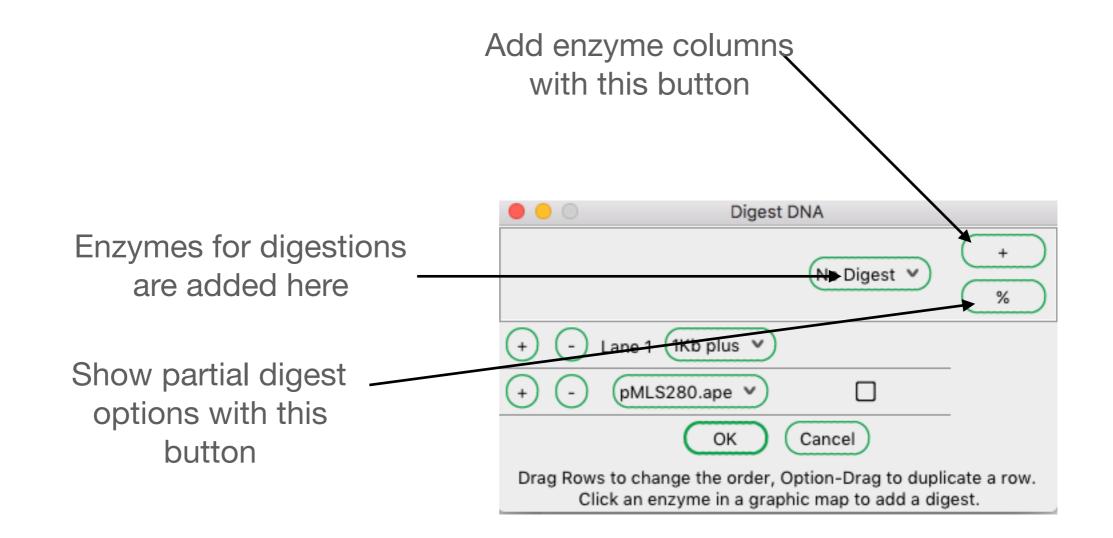
Click and drag lanes to rearrange

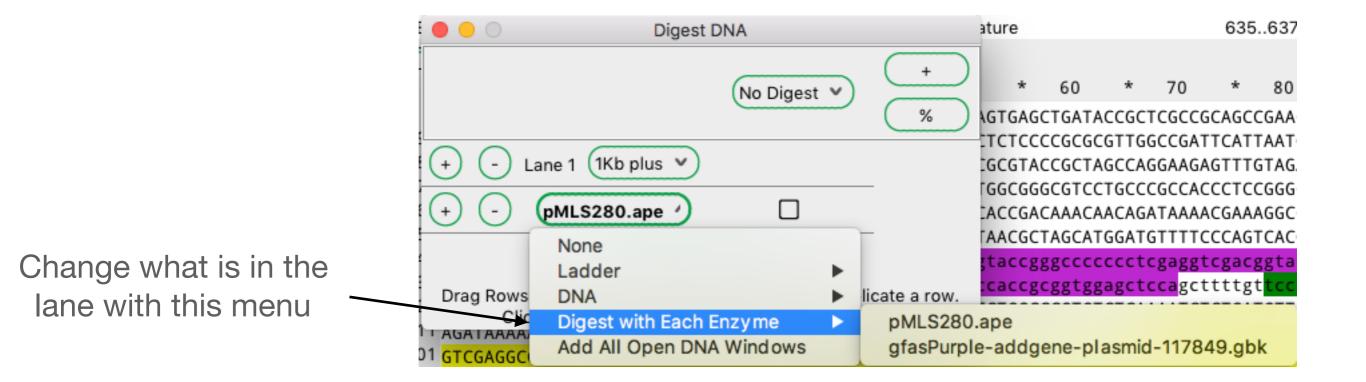
Shortcuts

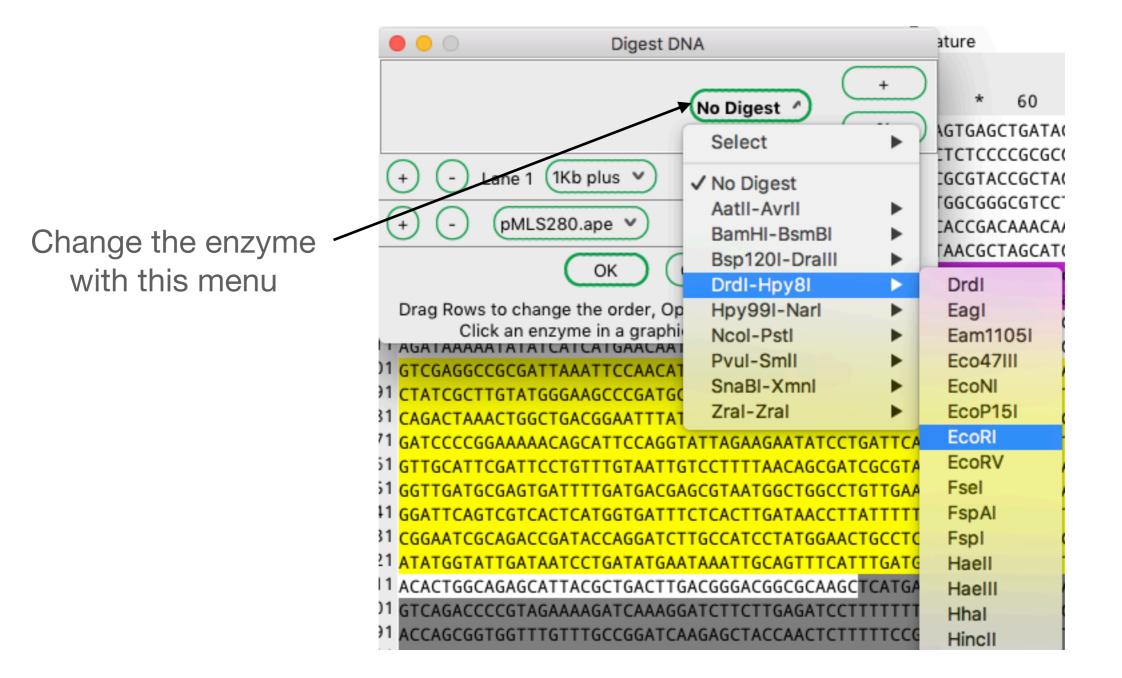
Command	Function
R	Digest with currently selected enzymes
Shift-R	Digest Dialog
E	Enzyme selection dialog
Shift-E	Set all functions to apply to selection

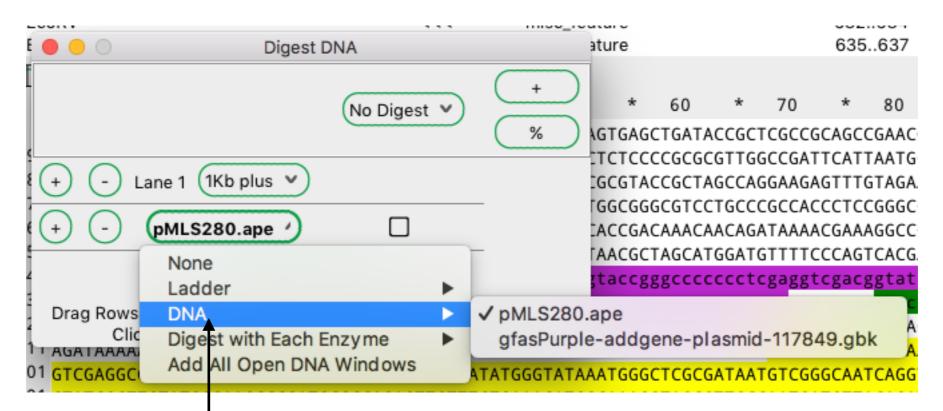




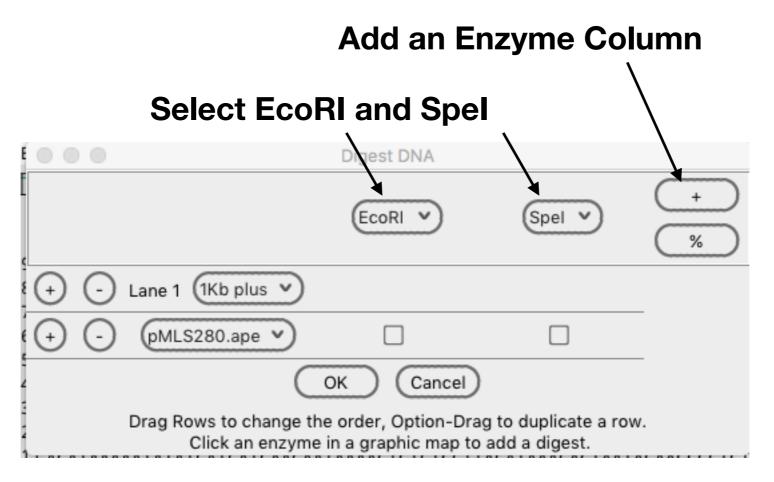








Select the DNA or ladder for each lane



• • •	Digest DNA			
	EcoRI V	Spel	•	+
+ - Lane 1 (1Kb plus 👻			
+ - Lane 2	1Kb plus ^			
Drag Rows to Click 481 GCAGTTCCCTA	None Ladder DNA Digest with Each Enzyme Add All Open DNA Windows	* * *	cate a est. CCAG	row. TCACGACGT
577 ataggegaat	You can also jus	st ci	reat	te a set

You can also just create a set of new lanes with each open DNA

E 🔴 🔴 🔘		Digest DNA				635.	.637
Ī		EcoRI V	Spel V		70 TCGCCGG		
\leftrightarrow \odot	Lane 1 (1Kb plus 👻				GCCGAT1 GGAAGAO CGCCACO	GTTTG	TAG
(+) (-)	pMLS280.ape				ΑΤΑΑΑΑ	GAAA	GGC
	None				GTTTTC		
14	Ladder	►			cgaggto		
2	DNA	►	rag to duplicate a row.		<pre>ccagctt</pre>	tttgt	tcc
1	Digest with Each E	nzyme 🕨	pMLS280.ape				
01 GTCGAGGC	Add All Open DNA	Windows	gfasPurple-addg	ene-plasmi	d-11784	9.gbk	¢ (

For convenience, you can digest a single DNA with all current enzymes

Press this to do partial digests

ı • •	Digest DNA			/	
	EcoRI V	Spel 💙	+		
+ - Lane 1 (1Kb plus 💙)				
+ - pMLS280.ape •			-		
ОК	Cancel		-		
Drag Rows to change the order, Option-Drag to duplicate a row. Click an enzyme in a graphic map to add a digest.					

Press this to select EcoRI in all columns

Digest DNA			
	EcoRI ^	Spel	90_
	Select	•	Select All In Column
 + - Lane 1 (Kb plus v) + - pMLS280.ape v + - gfasPurple-addgene-plasmid-117849.gbk v OK Cance Drag Rows to change the order, Option- 	No Digest AatII-AvrII BamHI-BsmBI Bsp120I-DraIII DrdI-Hpy8I Hpy99I-NarI Ncol-PstI	* * * * *	Deselect All In Column TTC TTC gtaa agct gag ACA AAC
Click an enzyme in a graphic mar CIAICGCIIGIAIGGGAAGCCCGAIGCGCCAGAGIIGIIICIGAAACA CAGACTAAACTGGCTGACGGAATTTATGCCTCTTCCGACCATCAAGCA1	Pvul-Smll SnaBl-Xmnl Zral-Zral	* * *	CAAT TATGTTACAGATGAGATGGT CATGGTTACTCACCACTGC

Oigest DNA			
	EcoRI V	Spel 💙	+ 3
+ - Lane 1 (1Kb plus 💙			
+ - Lane 2 pMLS280.ape V			
+ - Lane 3 (gfasPurple-addgene-plasmid-117849.gbk V			-
OK Cancel			- /
Drag Rows to change the order, Option-Drag to Click an enzyme in a graphic map to add		Ι.	

This will digest pMLS280 and gfasPurple with EcoRI and Spel

O O O 2 Windows Digest				
Lane 1: 1Kb plus Sizes: 100, 200, 300, 400, 500, 650, 850, 1000, 1650, 2000, 3000, 4000, 5000, 6000, 7000, 8000, 9000, 10000, 11000, 12000	1	2	3	i
Lane 2: pMLS280 digested with: EcoRI + Spel 2460(Spel-EcoRI), 24(EcoRI-Spel)				
Lane 3: gfasPurple-addgene-plasmid-117849 digested with: EcoRI + Spel 2047(Spel-EcoRI), 753(EcoRI-Spel)				

This will digest pMLS280 and gfasPurple with EcoRI and Spel

s	Tools	Window Help	
		Sequences 第L Two Sequences	
} g →	Restr	iction-Ligation Assembler	ł

Restriction-Ligation

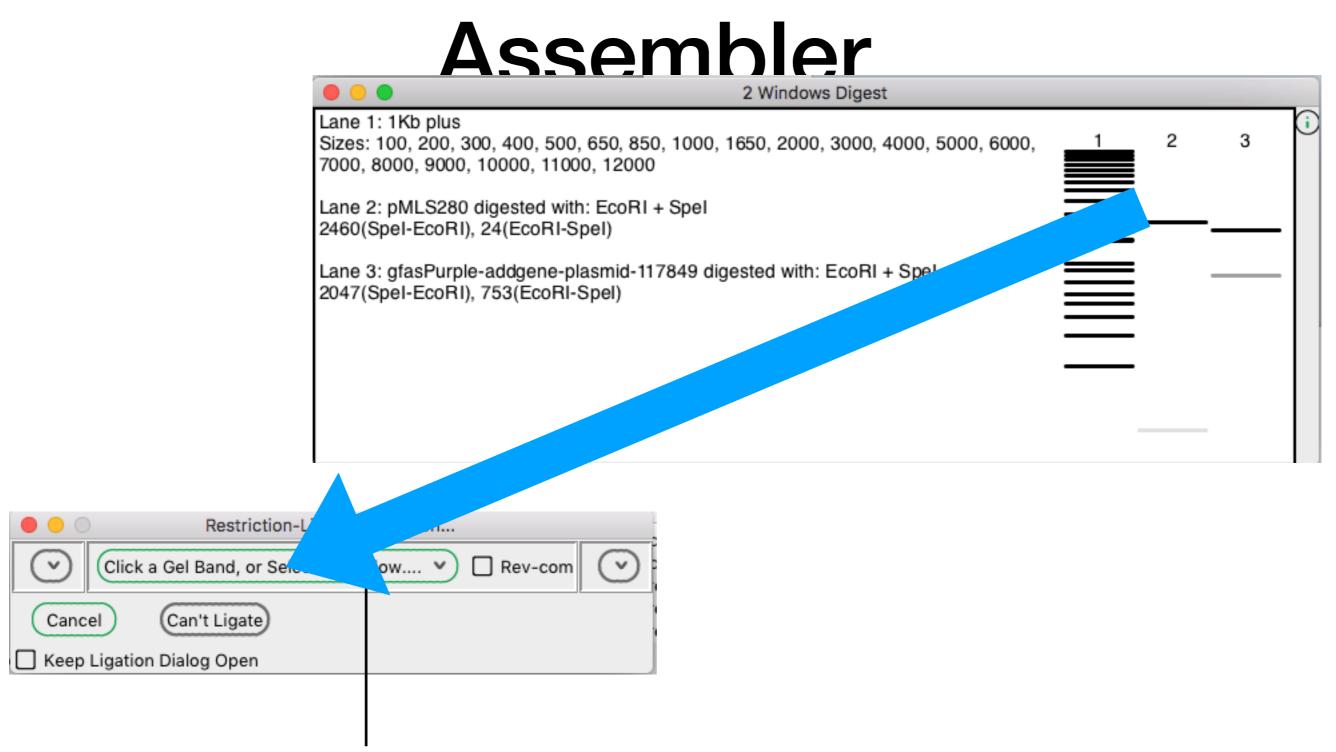
Assembler

2.11	indows bigest			
Lane 1: 1Kb plus Sizes: 100, 200, 300, 400, 500, 650, 850, 1000, 1650, 7000, 8000, 9000, 10000, 11000, 12000	2000, 3000, 4000, 5000, 6000,	1	2	3
ane 2: pMLS280 digested with: EcoRI + Spel 2460(Spel-EcoRI), 24(EcoRI-Spel)				
Lane 3: gfasPurple-addgene-plasmid-117849 digested 2047(Spel-EcoRI), 753(EcoRI-Spel)	with: EcoRI + SpeI			
		-		

Restriction-Ligation Reaction						
Click a Gel Band, or Select a Window 🗸 🗌 Rev-com						
Cancel Can't Ligate						
🗌 Keep	Ligation Dialog Open					

Click and drag a gel lane here

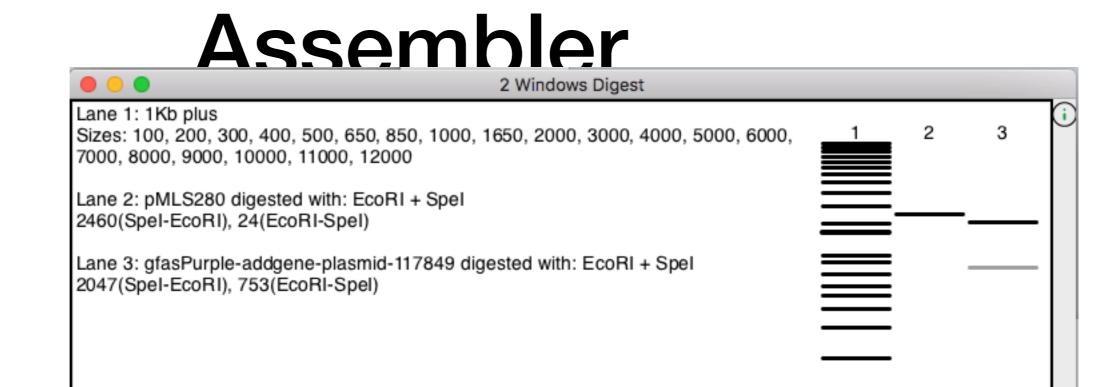
Restriction-Ligation



Click and drag a gel lane here

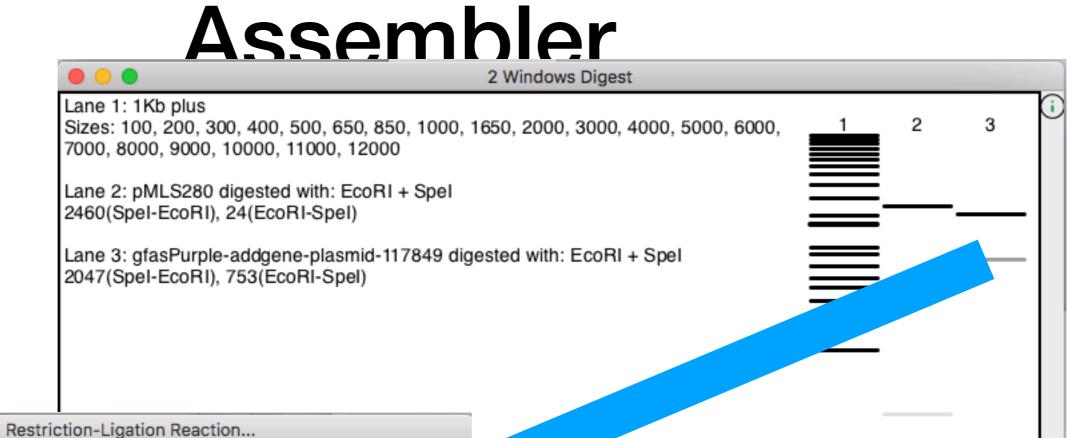
000	Restriction-Ligat	ion Reaction	
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	SpeI ctagttc 💵 aag		
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Restriction-Ligation



	Restriction-Lig	gation Re	action		
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Restriction-Ligation



estriction-Ligation Reaction	
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• • •	Restr	iction-Ligation Re	eaction	
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	Restriction-Ligation R	eaction
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EcoRI None	y gfasPurple-addgene-plasmid-11	7849.gbk 💙 🌠 Rev-com Spel Same 💙
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	ck a Gel Band, or Select a Window	N 🗆 Rev-com
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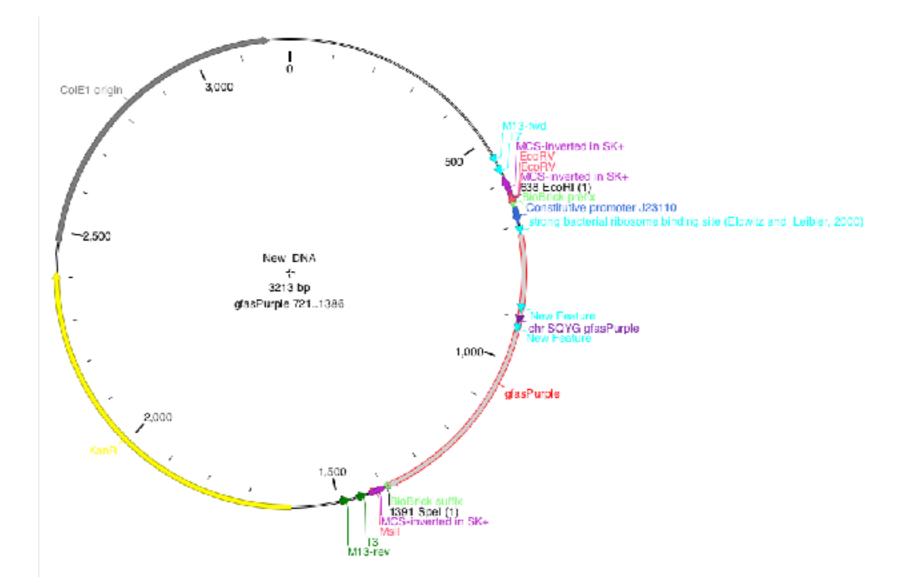
click here to flip a fragment

	Restriction-Ligation Re	eaction
Spel None 🗸	pMLS280.ape	♥
SpeI Spei ata cta tatgatc	I gttc ⊥ ■■ aag	
EcoRI None EcoRI Ecol tcg aat agcttaa	No Linear Windows Found.	117849.gbk 🗌 Rev-com Spel Same 💙
Cancel (Cancel (ck a Gel Band, or Select a Window OK Dialog Open	V Rev-com

click here to delete a fragment

	Restriction-Ligation	Reaction	
Spel None 💙	pMLS280.ape	▼ □ Rev-com EcoRI (Same 💙
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	am/Dcm
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▶ Hildden	_
M13-fwd >>> primer_bind 536553	
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* 10 * 20 * 30 * 40 * 50 * 60 * 70 * 80 * 90 *	
1 CTTTCCTGCGTTATCCCCTGATTCTGTGGATAACCGTATTACCGCCTTTGAGTGAG	
97 GTCAGTGAGCGASGAAGCGGAAGAGCGCCCAATACGCAAACCGCCTCTCCCCGCGCGTTGGCCGATTCATTAATGCAGCTGGCACGACAGGTTTCC	
193 CGACTGGAAAGCGGGCAGTGAGCGCAACGCAATTAATACGCGTACCGCTAGCCAGGAAGAGTTTGTAGAAACGCAAAAAGGCCATCCGTCAGGATG	
289 GCCTTCTGCTTAGTTTGATGCCTGGCAGTTTATGGCGGGCG	
385 GEATTTETCCTACTCAEGAGAGCETTCACCEACAAACAACAACAACAACAAAAGGCCCAGTCTTCCEACTEAGCCTTTCGTTTTATTTEATECCTE	
481 GCAGTTCCCTACTCTCGCGTTAACGCTAGCATGGATGTTTTCCCAGTCACGACGTtgtaaaacgacggccagtgagcgcgcgtaatacgactcact	
577 aragggcgaatrgggtaccgggccccccctcgaggtcgacggtatcgataagcttgatatcgaattcgcggccgcttctagagtttacggctaget	
573 cagirclagglaraalgrlagelarlagag <mark>aaagaggagaaa</mark> larlagAT6TC6GT6ATT6CTAAACA6AT6ACCTACAAA6TCTATAT6TC566T	
769 ACSGTGAACGGCCATTATTTTGAASTTGAAGGTGACGGTAAAGGCAAGCCGTATGAAGGCGAACAGACCGTTAAACTGACCGTCACGAAGGGCGGT	
865 CCSCT6CC6TTTSC <mark>AT6</mark> SGATATTCT6AGTCCSCA6 <mark>TCCCAATAT66C</mark> ASCATCCC6TTC <mark>AC6</mark> AAATATCC6GAA6ATATCCC6GACTAC6T6AA6	1
961 CASTCTTTTCCGSAAGGTTACACCTGGSAACGTATCATGAACTTCGAAGATGSCGCCSTCTGCACCGTGAGTAACGACAGCTCTATTCAAGGTAAT	1
1057 TGTTTCATCTACCATGTCAAGTTCTCASGTCTGAACTTCCCGCCGAATGSCCCGGTGATGCAGAAAAAGACCCAAGGCTGGGAACCGAATACGGAA	
1153 CGTCTGTTTGCACGCGATGGTATGCTGATTGGCAACAATTTCATGGCTCTGAAACTGGAAGCCGGTGGCCACTATCTGTGCGAATTTAAAAGCACC	
1249 TACAAGGCGAAAAAGCCGGTTAAAATGCCGGGCTATCATTACGTGGATCGTAAACTGGACGTTACCAACCA	
1345 CASTGTGAAATTTCAATCGCGCGCGCAAATCGGTGGCTTAAtaa <mark>tactagttetagageggccgccaccgcggtggggctera</mark> getttegtteta 1441 Titagtgagggttaattegggggtttggtgtaat <mark>catggtcatagttgtttectg</mark> GCASCTCTSGCCCGTGTCTCAAAATCTCTGATGTTACATTGC	12
1537 ACAASATAAAAATATATCATCATGAACAATAAAACTGTCTGCTTACATAAACAGTAATACAASGGGTGTT <mark>ATSAGCCATATTCAACGGGAAACGTC</mark>	
1533 GASGCCGCGCGATTAAATTCCAACATSGATGCTGATTTATATGGGTATAAATGGGCTCGCGATAATGTCGGGCAATCAGSTGCGACAATCTATCGCTT	
1729 GTAT566AASCCC6AT6CGCCASASTTSTTTCT6AAACAT66CAAA6STA6CSTT6CCAAT6ATGTTACA6AT6ACGTCA6AT6AT6AT6AT6AT6AT6AT6AT6AT6AT6AT6AT6AT	
1825 GACGGAATTTATSCCTCTTCCGACCATCAAGCATTTTATCCGTACTCCTGATGATGCATGGTTACTCACCACTGCGATCCCCGGAAAAAACAGCATT	
1921 CCACGTATTAGAAGAATATCCTGATTCACGTGAAAATATTGTTGATGCGCTGGCAGTGCTCCTGCGCCGGTTGCATTCGATTCCTGTTGTAATTG	
2017 ICCTTTTAACAGCGATCSCGTATTTCGTCTCGCTCAGSCGCAATCACSAATGAATAACGGTTGGTTGATGCSAGGGATTTTGATGACGAGCGTAA	
2113 TESCIEGCCTETTEAACAASICTESAAAGAAATECATAAACTTTISCCATTCICACCSEATTCASICSTCACTCATESTGATTCICACTTEATAA	
2209 CCTTATTTTT6ACGAGGSGAAATTAATAGSTTSTATTSATGTTGSACGASTCSGAATC6CAGACCGATACCASGATCTTGCCATCCTATGGAACTG	
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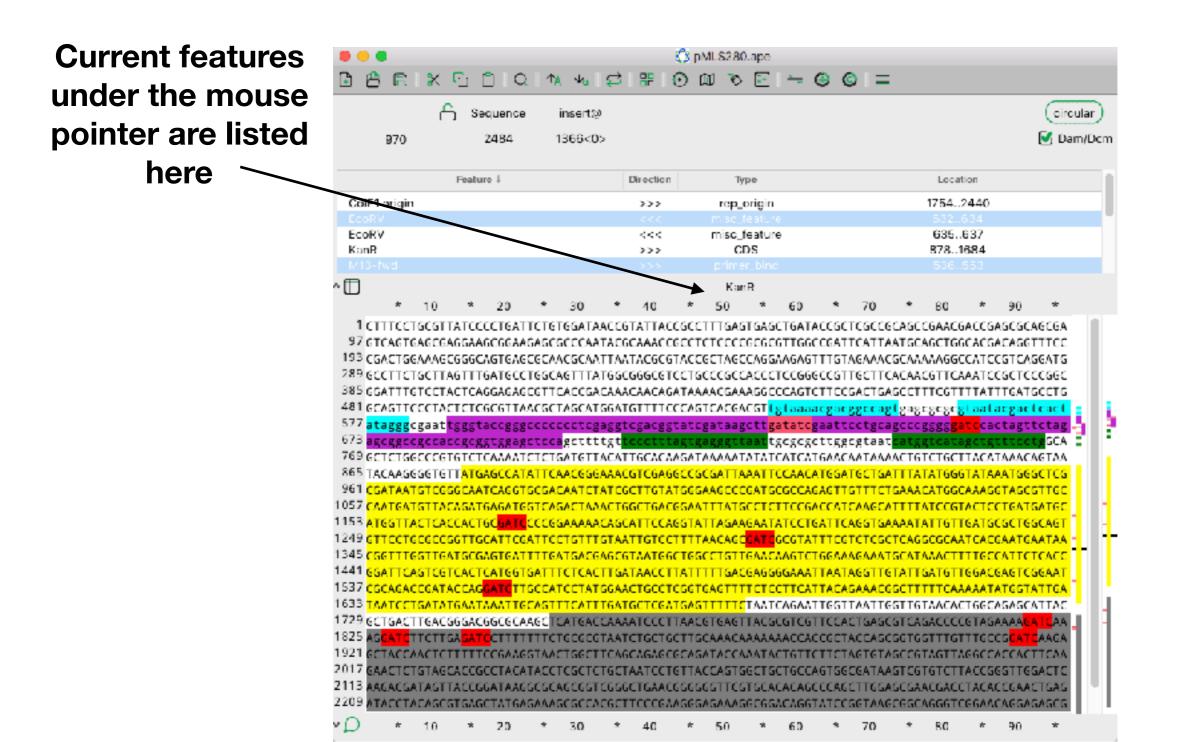


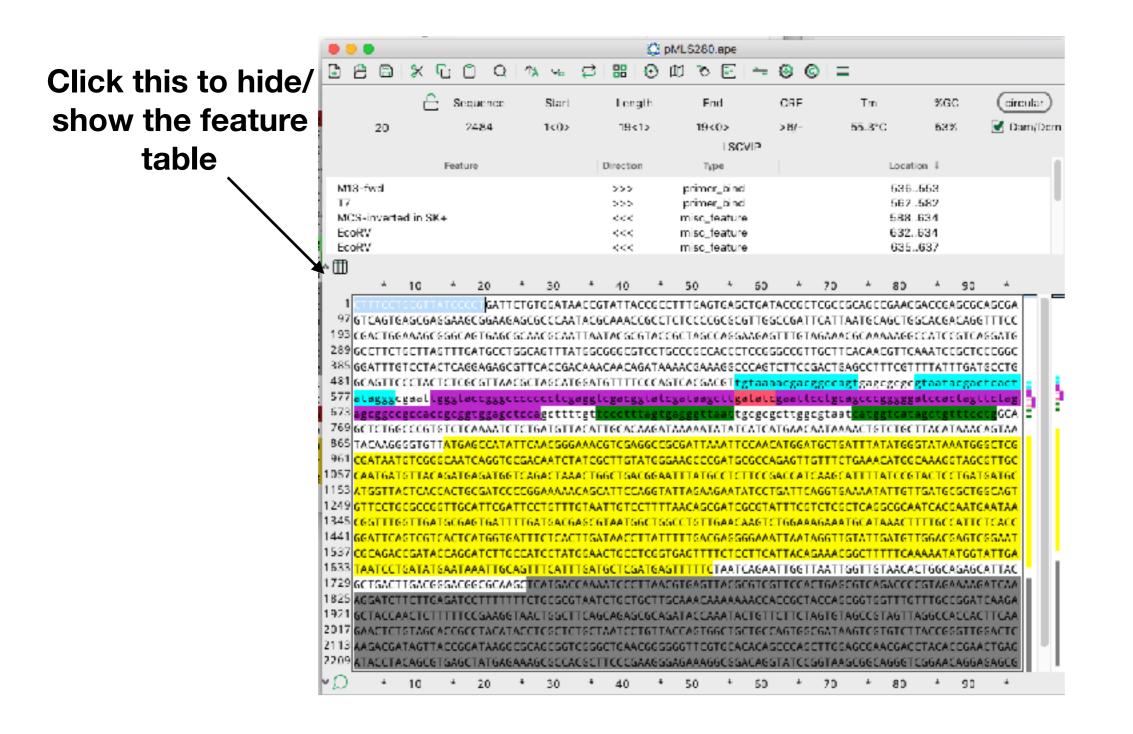
Feature library

	Edit Feature Library
ADH1 promoter	
Cbunc-119	
hGH polyA signal	
SV40 early promoter	
CMV promoter	
SV40 late polyA	
CmR	
TetR	
TetR	
KanR	
Select All Raise	Lower Delete Edit New
Save Changes Save	Changes As Cancel Scan on Close
~/Library/Prefere	ences/ApE/Features/Default_Features.txt

New Feature

Edit Feature
New Feature
119 🗌 Rev-Com
Uppercase Only (Feature Plus Selection) (Feature Minus Selection)
Add Qualifier
Feature type: misc_feature V
Forward color: Reverse color: Same Favorites
Place Directly Above Feature: EcoRV V
OK Make format default for feature type





Right-click here to edit a feature

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193 CGACTO 289 GCCTTO 385 GGATTI 481 GCAGTI 577 <mark>ataggg</mark> 573 <mark>agggg</mark> 769 GCTCTO	GAAAGCGG CTGCTTAGT IGTCCTACT ICCCTACTC CGBBBT CGBCBBC GGCCBGTGT	GCAGTS TTGATS CAGGAS TCGCGT gtaccg cggtgg CTCAAA	CCTGGC AGCGTT TAACGC ggcccc agctcc ATCTCT	AGTTT CACCG TAGCA ccctc agctt GATGT	ATGGO ACAAA IGGAI Baggi ttgt TACAI	CGGGCG ACAACA IGTTTT CGGaCg CCCCCC ITGCAC	GATAA GATAA CCCAG gtatc Tagtg	CCCGCCA AACGAAA TCACGAO gatabgc agggttb AAAAATA	CCCTCC GGCCCA GT <mark>tgta</mark> ttgata at <mark>tgcg</mark> TATCAT	GGGCCGT GTCTTCC abacgac togaatt cgcttgi CATGAAC	GACTG GACTG CCCCC CCCCCC CCCCCC CCCCCC CCCCCC CCCCC	CACAAC AGCCTT gtgagc agcccg tcatgg AACTGT	GTTCAA TCGTTT gcgc <mark>gt ggggat</mark> tCatag CTGCTT	ATCCC TATT taatac tocact tocact tocact	GCTCCCG FGATGCC Cgactca tagttct toctg AACAGT	SC IG IG IG IG IG IG IG IG IG IG IG IG IG
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🕼 pMLS280.ape 88 % 6 🖸 🛍 ര 🗉 🛏 🙆 🙆 🚍 ല $0 \uparrow \sim$ ct. G) **Click this to sort** the feature table Sequence insert@ c rcu ar 🗹 Dam/Dom 2484 1366<0> Feature 4 Direction Туре Location 1754..2440 ColE1 origin rep_origin >>> 635..637 EcoRV misc_feature <<< KonR >>> CDS 878..1684 **^∭** * 10 20 30 40 50 70 80 90 97 GTCAGTGAGCGAGGAAGCGGAAGAGCGCCCAATACGCAAACCGCCTCTCCCCGCGCGTTGGCCGATTCATTAATGCAGCTGGCACGACAGGTTTCC 19BICGACTGGAAAGCGGGCAGTGAGCGCAACGCAATTAATACGCGTACCGCTAGCCAGGAAGAGTTTGTAGAAACGCCAAAAAGGCCATCCGTCAGGATG 385 GGATTIGTCCTACTCAGGAGAGCGTTCACCGACAAACAACAACAACAAAACGAAAGGCCCAGTCTTCCGACTGAGCCTTTCGTTTTATTTGATGCCTG 481 GCAGTTCCCTACTCTCGCGTTAACGCTAGCATGGATGTTTTCCCAGTCACGACGTtgtaaaacgacggccagtgagcgcgcgtaatacgactcact ģ 577 ataggg cgaattgggtaccgggccccccctcgaggtcgacggtatcgataagcttgatatcgaattcctgcagcccggggggtcccccctcgaggtcgacggtatcgatagcttctag 673 agcggccgccaccgcggtggagctccagcttttgt cotttagtgagggttaattgcgcgcttggcgtaat GCA 📑 769 GCTCT6GCCCGT6TCTCAAAATCTCT6AT6TTACATT6CACAA6ATAAAATATATCATCAT6AACAATAAAACT6TCT6CTTACATAAACA6TAA 865 TACAAGGGGTGTT<mark>ATGAGCCATATTCAACGGGAAACGTCGAGGCCGCGATTAAATTCCAACATGGATGCTGATTTATATGGGTATAAATGGGCTCG</mark> 961 CGATAATGTCGGGCAATCAGGTGCGACAATCTATCGCTTGTATGGGAAGCCCGATGCGCCAGAGTTGTT 1057 CAATGATGTTACAGATGAGATGSTCAGACTAAACTGSCTGACGSAATTTATGCCTCTTCCGACCATCAAGCATTTTAT 1153 ATGGTTACTCACCACTGCGATCCCCGGAAAAACAGCATTCCAGGTATTAGAAGAATATCCTGATTCAGGT 1249 GTTCCTGCGCCGGTTGCATTCGATTCCTGTTTGTAATTGTCCTTTTAACAGC<mark>GATC</mark>GCGTATTTCGTCTCGCTCAGGCGCAATCACGAATGAATA 1441 GGATTCAGTCGTCACTCATGGTGATTTCTCACTTGATAACCTTATTTTTGACGAGGGGAAATTAATAGGTTGTATTGATGTTGGACGAGTCGGAAT 1537 CGCAGACCGATACCAG<mark>GATE</mark>TTGCCATCCTATGGAACTGCCTCGGTGAGTTTTCTCCTTCATTACAGAAACGGCTTTTTCAAAAAATATGGTATTGA 1633 <mark>TAATCCTGATATGAATAAATTGCAGTTTCATTTGATGCTCGATGAGTTTTTC</mark>TAATCAGAATTGGTTAATTGGTTGTAACACTGGCAGAGCATTAC 1729 GCTGACTTSACG6GACGGC6CAAGCTCATGACCAAAATCCCTTAACGTGASTTACGCSTCGTTCCACTGAGCGTCAGACCCCGTASAAAA<mark>GATC</mark>AA 1825 AGGATOTTETTGAGATOCTTTTTTTETGEGEGETAATETGETGETGEAAAEAAAAAAECAECGETAECAGEGGTGGTTTGTTGEEG ATCAAGA 1921 GETACCAACTETTTTECGAAGGTAACTGGETTEAGEAGAGEGEAGATACEAAATACTGTTETTAGTGTAGECGTAGTTAGGECACCAETTCAA 2017 GAACTETGTAGCACCGCCTACATACCTCGCTCTGCTAATCCTGTTACCAGTGGCTGCTGCCAGTGGCGATAAGTCGTGTCTTACCGGGTTGGACTC 2113 AAGACGATAGTTACCGGATAAGGCGCAGCGGTCGGGCTGAACGGGGGGTTCGTGCACAGCCCAGCTTGGAGCGAACGACCTACACCGAACTGAG 2209 ATACCTACAGCGTGAGCTATGAGAAAGCGCCACGCTTCCCGAAGGGAGAAAGGCGGACAGGTATCCGGTAAGCGGCAGGGTCGGAACAGGAGAGCG × () 90 20 30 40 50 80 10 60 70

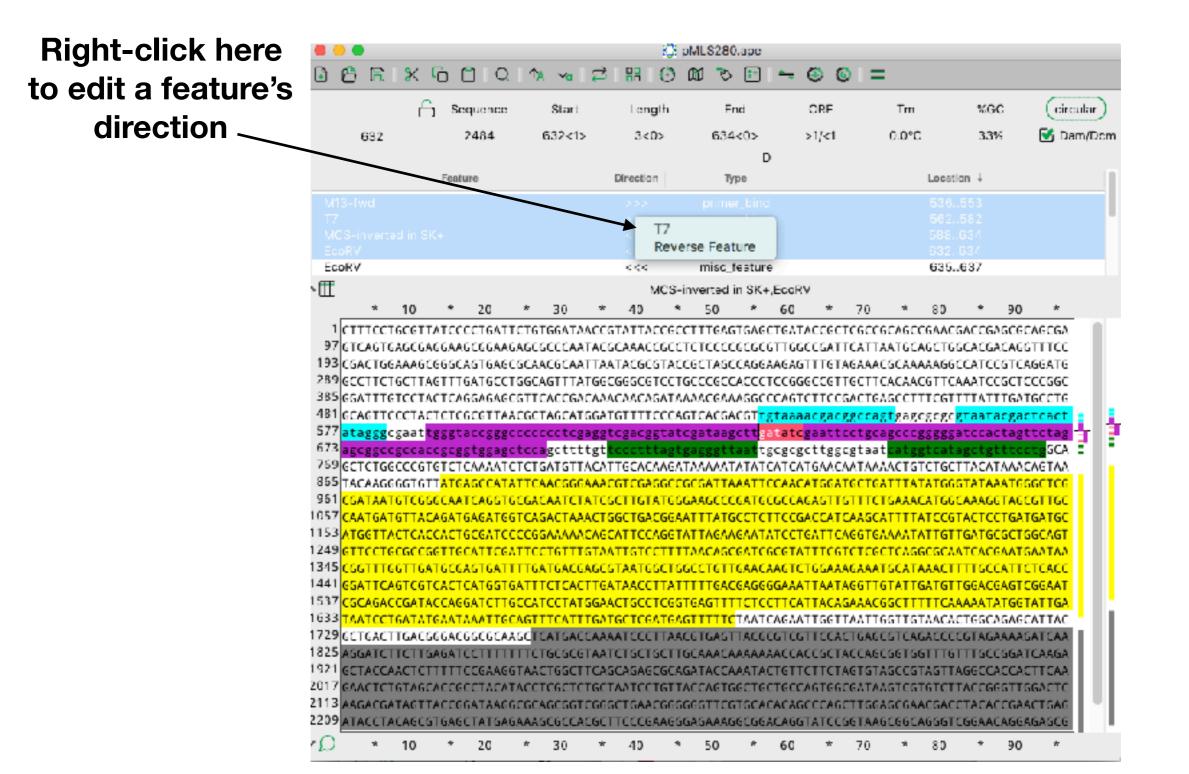
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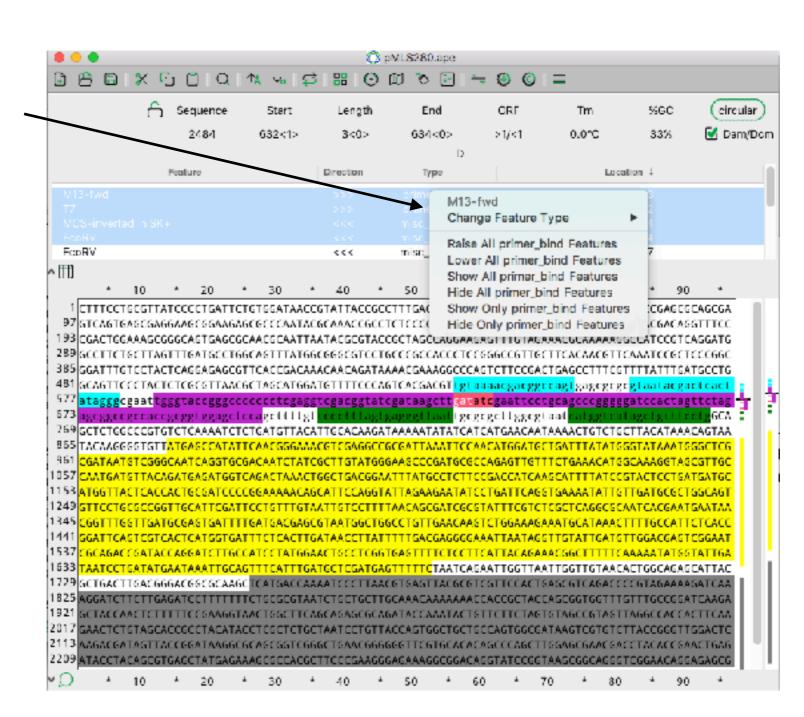
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885 G 181 G 177 B 173 B 173 G 159 G 165 T	GATTTGTCC CASTTCCCT CBEEECEBB BCEECEBCC CTCTGGCCC ACAASSSST	TACTCAGGAGAGCG ACTCTCGCGTTAAC t <mark>tegetaccegecc accgcggtggagct</mark> GTGTCTCAAAATCT GTT <mark>ATSASCCATAT</mark>	TTCACCGACAA SCTASCATSSA CCCCCCCGBgg CCBgCttttgt CTGATGTTACA TCAACGSSAAA	ACAACAGATAA ATSTITICCCA6 tegaeggtate tecctttagtg ATTGCACAAGAT CSTCGA66CC6	AACGAAAGGCCCA TCACGACGT <mark>tgta</mark> gataagcttgata (agggttaattgs TAAAATATATAT CGATTAAATTCCA	GTCTTCCGAC DODESDESSE tcgaattcct cgcttggegt CATGAACAAI ACATGGATGC	TGAGCCTTT COSTSOSCS SCASCCCSS SAATSATSST MAMACTGTC TGALITATA	CGTTTTATTTG/ CGCC <mark>ALDOLOCZO</mark> EggalCCaClag CalagCLgLLCC IGCTTACATAA/ IGGGLATAAALC	ATGCCTG CTCGCT CTCCTG CTCCTG CCTG CCCG CCCG CCCG CCCG CCCG CCCG CCCG CCCG CCCG CCCG CCCG CCCG CCCG CCCG CCCG CCCG CCCG CCCGCT CCCCGCT CCCGCCT CCCGCT CCCCCCCC
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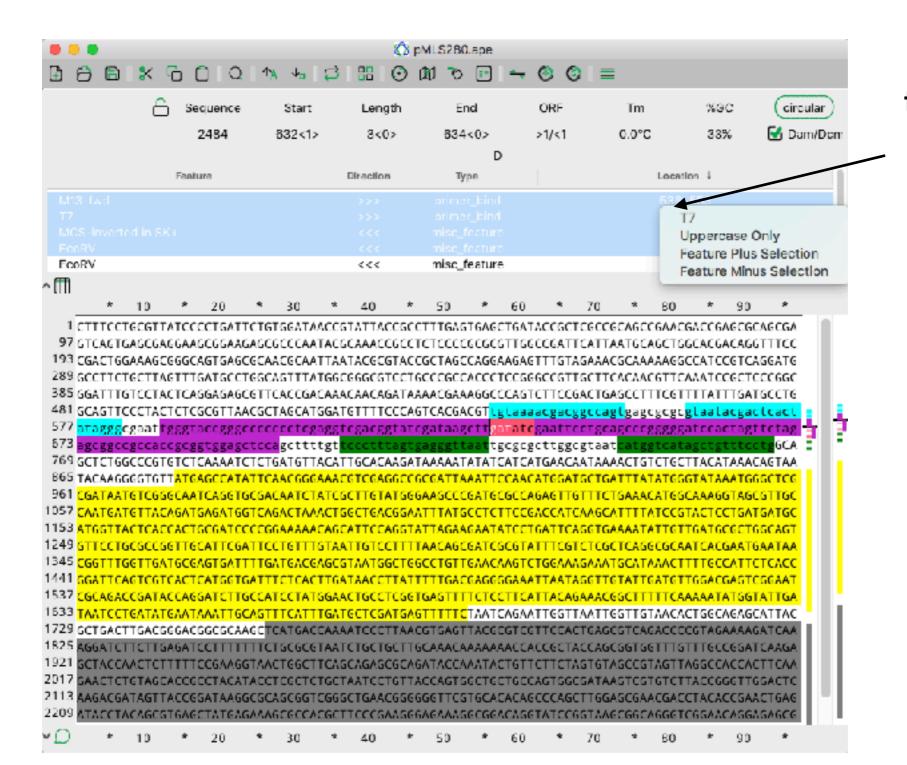
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577 673 769 865 961 057 0 153 A 153 A 1441 6 1441 6 1537 0	Lower 4 Sel Hide 4 Sele Show 4 Sele SATAATGTCGS AATGATGTTCGS TGGTTACTCAC TTCCTSCSCCS GGTTTGGTTCA GATTCACTCGT GCAGACCGATA	lected Features oted Features octed Features octed Features octocogetoco AGATGAGAGAGAGA octocogetoco coccogetocogetoco octocogetocogetoco occogetocogetocogetocogetoco occogetocogetocogetocogetocogetocogetocogetoco occogeto	S to Bottom S GACAATCTATO CAGACTAAAC CGGAAAAACAO TCCTGTTTGT/ TGATGACGAGO CTTTCTCACTTO CATCCTATGG/	Cgacggtat CCCTTGAGGCC GCCGAGGCC CSCTTGTATGG IGGCTGACGGA SCATTCCAGGT AATTGTCCTTT CGTAATGGCTG CATAACCTTAT AACTGCCTCGG	cgataagetter gagggttert SGGATTAAATA SAASCCCGATG ATTATGCCTCT ATTAGAAGAATA TAACAGCGATC SCCTGTTCAACA TTTTGACCAGG TGAGTTTTCCC	TELCGEELT CORCATEGA CATCATEGA CORCASACTI COCESACTI COCESACTI COCESACTI COCESACTICE COCESAC	COURCERCES CETERT CETERTIAN CATEGATIAN STITCIGAAAC CAASCATITTA AGGTGAAAATA FCTCGCTCAGG AGAAATGCATA GGAAATGCATTTC GAAACGGCTTT	Egggatceact ECATAgetgti ICTGCTTACAT/ IATGGCAAAGGI ATGCGTACTCCT ATGCTACTCCT ATGCTSATGCC ICGCAATCACG/ ICGCAATCACG/ ITTCAAAAATA1	tagttetag MACAGTAA MEGGETEC IACCETEC IGATGATEC GETEGEAEE AATGAATAA ATTETEAEE AGTEGEAAT IGGTATTEA
577 673 769 855 951 057 0 153 749 345 941 537 0 633 7	Lower 4 Sele Hide 4 Sele Show 4 Sele GATAATGTCGS AATGATGTTAC TGGTTACTCAC TCCTSCGCCS GGTTTGGTTCA GATTCACTCGT GCAGACCGATA AATCCTGATAT	lected Features oted Features octed Features octed Features octocosofic AGATGAGATGG octocosofic cattocosofic cattocosofic cattocosofic gaataaattoco	S to Bottom SACAATCTATO CAGACTAAAC CGGAAAAACAO TCCTGTTTGT/ TGATGACGAGO ATTTCTCACTTO CATCCTATGG/ AGTTTCATTG/	CGACGSTAT CCCTTGAGG GTCGAGGCC CSCTTGTATGG IGGCTGACGGA SCATTCCAGGT AATTGTCCTTT CGTAATGGCTG GATAACCTTAT AACTGCCTCGATGA	CGATAAGCTTE BABESTTAAT TAAAAATATAT SAASCCCGATG ATTATGCCTCT ATTAGAAGAAT/ TAACAGCGATC GCCTGTTGAAC/ TTTTGACCAGG TGAGTTTTCTC STTTTCTAAT	HELCGART CGCCGCCTTGAAC CAACATGAAC CCAACATGAAC CCCCGAACAT CCCGACCAT CCCGACCAT CCGTATTCG CGTATTCG CGTATTCG CGAATTACA CGAATTGGT	CCTGCAGCCCG CGTBAT AATAAAACTGT IGCTGATTTAT STITCTGAAAC CAAGCATTTTA AGGTGAAAATA ICTCGCTCAGC AGAAATGCATA AGGTTCTATTC GAAACGGCTTT IAATTGGTTGT	Egggatceact Eccatagetgt ICTGCTTACAT/ IATGGGTATAA/ IATGGGAAAGGT ATCCGTACTCCT ATGTTSATSCC CGCAATCACG/ IAACTTTGGCC/ ITTCAAAAATAT IAACACTGGCAC	tagttetag TreetgSCA AAACAGTAA ATGSGETEC FGATGATGC GGTGGCAGT ATGAATAA ATTETCACC AGTEGCAAT FGGTATTGA GAGCATTAC
577 673 769 855 951 057 0 153 4 249 345 0 441 537 0 633 1 729 5	Lower 4 Sel Hide 4 Sele Show 4 Sele AATGATGTCGS AATGATGTTCG ITCCTSCGCCS GGTTTGCTTCA GATTCAGTCGT GCAGACCGATA AATCCTGATAT CTSACTTGACS	lected Features oted Features acted Features accented Features acc	S to Bottom SACAATCTATO CAGACTAAACA CGGAAAAAACA TCCTGTTTGT/ TGATGACGAG CATCCTATGG/ GTTTCATTG/ CATCCTATGG/ GTTCATGACCA/	CGACGSTAT CCCTTGAGGCC GCCTGAGGCC CCCTTGTATGG IGGCTGACGGA SCATTCCAGGT AATTGTCCTTT CGTAATGGCTG GATAACCTTAT AACTGCCTCGATGA AATTCCCTTAA	CGATAAGCTER BABESTIAAT TAAAAATATAT SAASCCCGATG ATTATGCCTCT ATTAGAAGAATA TAACAGCGATC SCCTGTTGAACA TTTTGACCAGG TGAGTTTTCTC STTTTCTAAT CGTSAGTTACG	TELCGEATT CORCATEGAAC CAACATEGAA COCASAGTT COCASAGTT COCASAGTT COCASAGTT COCASACTA COCASACTACA CAAATTACA CASAATTGC COCCASACTACA	CCTGCAGCCCG CGTBAT CGTBAT IGCTGATITAT STITCTGAAAC CAAGCATITTA AGGTGAAAATA CTCGCTCAGC AGAAATGCATA AGGTTCTATTC GAAACGGCTTT TAATTGGTTGT	ESSERENCE STATEMENT ECAL ASSERED ELECTION AND ELECTION AND AND ELECTION AND AND ELECTION AND AND AND ELECTION AND AND AND ELECTION AND AND AND AND ELECTION AND	tagttetag treetgSCA AAACAGTAA ATGSGETEC FGATGATGE GATGATGE GATGATAA ATTETEAC AGTEGEAAT GGETATTEA GAGCATTAC AAASATEAA
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Features



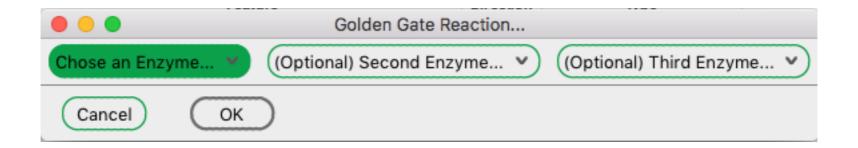
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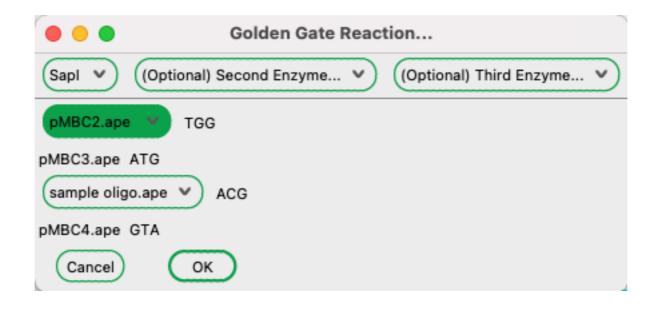
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97 GTCAGTO 193 CGACTGO 289 GCCTTCT 385 GGATTO 481 GCAGTTO 577 atagggo 573 against 769 GCTCTGO 865 TACAAGO 961 CGATAAT	AGCGAGGA AAASCGGGG GCTTAGTTT TCCTACTCA CCTACTCA CCTACTCA CCCACTCA CCCCGTGTCT GCCGGGGTATCA GCCGGGCA	AGCGCAAGA CAGTGAGCC IGATGCCTG AGGCCAGCCCC CGCGTTAAC CGCCGGGCCC CGCCGGGCC IGAAAATCT AGCCATAT	AGEGEECEAATA SCAACGEAATT SGCAGTITATG STTCACEGACA CGCTAGEATGG CCCCCTEGAG TEGECTTEG TEGECTTEG TEGATGTTAC TCAACGGGAA	CGCAAACCGCC IAATACGCGTAC GCGGGCGTCCT AACAACAGATA ATGTTTTCCCA CGCGGCGGCGTCC ATGCACGAGAC ACGTCGAGGCC CGCTTGTATSG	TCTCCCCGCGCGCTT CGCTAGCCAGGAAG GCCCGCCACCCTCC AAACGAAAGGCCCA GTCACGACGT <mark>tgta CgataagcttgtaTa</mark> CgataagcttgtaTa CgataagcttgtaTa GCGATTAAATTCCA GAAGCCCGATGCSC	GGCCGATTC AGTTTGTAG GGGCCGTTG GTCTTCCGA BBBCGBBCCG CCCGBBCCG CATGAACAA ACATGGATG CAGAGTTGT	ATTAATGCA AAACGCAAA CTTCACAAC CTGAGCCTT CCagtgagc tgcagcccg Laatgcagcccg TAAAACTGT CTGATTTAL TTCTGAAAC	SCTESCACEACA AAGECCATCEGT STTCAAATCEGE TEGTTTTATTTG gegegtatecacta Ferragety Egggatecacta Ferragety Egggatecacta AGECTACATAA ATGECAAAESTA	GGTTTCC CAGGATG TCCCGGC ATSCCTS actcact gttctag TLSGCA ACAGTAA GSGCTCG GCGTTGC
97 GTCAGTO 193 CGACTGO 289 GCCTTCT 385 GGATTO 481 GCAGTTO 577 atagge 573 arrspor 769 GCTCTGO 865 TACAAGO 961 CGATAAT 1057 CAATGAT	AGCGAGGA AAASCGGGG GCTTAGTTI TCCTACTCA CCTACTCA CCTACTCA SCCTACTCA SCCTACTCA CCCGTGTCA GCCGGTGTTATS GCCGGGCA GTTACAGA	AGCGCAAGA CAGTGAGCG ISATGCCTG AGGCAGCG CGCGTTAAC CACAGGCG AGCCATAT ICCAGGTGG ISAGATGGT	AGEGECCEAATA SCAACGEAATT SCAACGEAATT SCAGETTATG ETTCACEGACA CECCCCCEGAG TEGACETTAC TEGACECTAA SACAATETAT TEAACSGGAA	CGCAAACCGCC IAATACGCGTAC GCGGGCCGTCCT AACAACAGATA ATGTTTTCCCA CTCGACGGGGC CGCTCGACGCC CGCTCGACGCCA CGCTCGACGCA CGCTCGACGCA	TCTCCCCGCGCGCTT CGCTAGCCAGGAAG GCCCGCCACCCTCC AAACGAAAGGCCCA GTCACGACGT <mark>tgtB cgatBagcttgtB cgatBagcttgtB CgatBagcttgtB CgatBagcttgtB GCGATTAAATTCCA GAAGCCCGATGCSC ATTTATGCCTCTTC</mark>	GGCCGATTC AGTTTGTAG GGGCCGTTG GTCTTCCCA GTCTTCCCA GTCTTCCCA CATGAACAA ACATGGATG CAGACTCA CAGACATCA	ATTAATGCA AAACGCAAA CTTCACAAC CTGAGCCTT CCagtgagc tgcagcccg Laat DJgg TAAAACTGT CTGATTTAT TTCTGAAAC AGCATTTA	SCTESCACEACA AAGECCATCEGT STICAAATCEGE TEGTTITATTIG gege <mark>gtaataeg</mark> ggggatecacta ICAT 25/19111 CIGCTTACATAA AIGESTATAAAT AIGESTATAAAT AIGESTATACAESTA	GGTTTCC CAGGATG TCCCGGC ATSCCTS actcact gttctag Gttctag ACAGTAA GSSCTCG GCSTTGC ATSATGC
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97 GTCAGTO 193 CGACTGO 289 GCCTTCT 385 GGATTTO 481 GCAGTTO 577 atagggo 673 agr 381 769 GCTCTGO 865 TACAAGO 961 CGATAAT 1057 CAATGAT 1153 ATGGTTA 1249 GTTCCTO 1345 CGSTTTO	AGCGAGGA/ GCTTAGTT CCTACTC/ CCTACTC/ CCTACTC/ GCCGCGTGTC GCCGCGTGTC GCCGGGGA/ GTTACAGA1 CCCCGGTC GTTACAGA1 CCCCGGTC GTTACAGA1	AGCGCAAGA CAGTGAGCCTG AGGACAGCCTG AGGACAGCC CSCGTTAAC CSCGTTAAC CSCGTTAAC CAGACATAT AGCCATAT AGCCATAT CSCGATCCC CATTCGAT CAGTGATCT	AGEGEECEAATA GEAACGEAATT GECAGETTATG GETAGEATG GECTAGEATG CECCETEGA CECCETEGA CECCETEGA CECCETEGA CAACAATETAT CAAACGEGAAAAACA CECCETTTG CECCETTTG CECCETTG	CGCAAACCGCC AATACGCGTAC GGGGGCGTCCT AACAACAGATA ATGTTTTCCCA CTGCTGCACAGAC CGCTGCACAGAC GGCTGCACAGAC GGCTGCCAGAC GGCTGCCGAGGC GGCTGCCGCG CGCATGCCTG CGTAATGGCTG	TCTCCCCGCGCGCTT CGCTAGCCAGGAAG GCCCGCCACCCTCC AAACGAAAGGCCCA GTCACGACGT <mark>tgta cgataagcttgta</mark> cgataagcttgta cgataagcttgta CGATTAAATTCCA GAAGCCCGATGCSC ATTTATGCCTCTC ATTAGAAGAATATCC TAACAGCGATCGCG GCCTGTTGAACAAG	GGCCGATTC AGTTTGTAG GGGCCGTTG GTCTTCCCA GTCTTCCCA GTCTTCCCA CAGACCACA CAGACTGAACAA ACATGGATG CAGACTCA CAGACTCA CTGATTCAG GTCTGGAAAG	ATTAATGCA AAACGCAAAC CTTCACAAC CTGAGCCTT CCagtgagc tgcagcocc laataly TAAAACTGT CTGATTTAL TTCTGAAAC GTGAAAATA TCGCTCAGS AAATGCATA	SCTESCACEACA AAGECCATCOGT STICAAATCOGC TCGTTTTATTTG gogo <mark>gtaatacg</mark> ggggatocacta for portyfff CTGCTTACATAA ATGECAAAESTA TCCGTACTCCTG TTGTTGATECCA AACTTTTGCCAT	GGTTTCC CAGGATG TCCCGGC ATGCCTG actcact gttctag gttctag GCTTGC ACAGTAA GSGCTCG GCGTTGC ATGATGC TGGCAGT TSAATAA TCTCACC
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97 GTCAGTG 193 CGACTGG 289 GCCTTCT 385 GGATTG 481 GCAGTTG 577 ataggg 673 apresent 769 GCTCTGG 865 TACAAGG 961 CGATAAT 1057 CAATGAT 1057 CAATGAT 1153 ATGGTTA 1249 GTTCCTG 1345 CGSTTG 1441 GGATTCA 1537 CGCAGAG 1533 TAATCCT 1729 GCTGACT 1825 AGGATCT	AGCGAGGA AAASCGGGG GCTTAGTTI TCCTACTCA CCTACTCA CCTACTCA CCTACTCA CCCACTACTCA GCCGCGGGCA GCCGCGGGCA GCCCGGGCA GCCGCGGGCA GCCGCGGGCA GCCGCGGCA GCCGCGGCA GCCGCGGCA GCCGCGGCA GCCGCGGCA GCCGCGGCA GCCGCGGCA GCCGCGGCA GCCGCGGCA GCCGGGGA GCCGGGGA TCTTGAGA	AGCGCAAGA CAGTGAGCC CGATGCCTG AGGGCAGCC CGCGTTAAC CGCGRCCC CGCGRCCC CGCGRCCCTG CAAAATCT CAGCCATAT CAGCCATAT CGCGCCATCG CATTCGAT CGCGCCAAG CGTTCTTGC CAATTGCA CGCGCCAAG CCTTTTT	AGEGEECEAATA GEAACGEAATT GECAGETTATG GETTEACEGACA GECAECEACA GECAECEACA CECECETEGA CECECETEGA CECECETEGA CECECETEGA CECECETATGA CATECTATGACCA AGETTEATCA CECECETATGACCA CECECETATGACCA CECECETATGACCA CECECETATGACCA	CGCAAACCGCC AATACGCGTAC GCGGGCCGTCCT AACAACAGATA ATGTTTTCCCA CATGTTTTCCCA CATGCACAAGA CGTCGACAAGA CGCTCGAGGCC CGCTTGTATGG CGCTTGTACGCT CGCTCGACGAGA GATTGTCCTTT CGTAATGCCTCG GATAACCTTCA AACTGCCTCGATGA AACTGCCTCGATGA	TCTCCCCGCGCGCTT CGCTAGCCAGGAAG GCCCGCCACCCTCC AAACGAAAGGCCCA GTCACGACGT <mark>tgtB CgatBagcttgtB CgatBagcttgtB CgatBagcttgtB CgatBagcttgtB CgatBagcttgtB CgatBagcttgtCA GCGATTAGCCATGCSC ATTAGACGAACAAC ATTAGACGAGCGATCCCS GCCTGTTGACCAGGCA TGAGTTTCCCTT GTTTTCTAATCAG CGTGAGTTACGCST GCAACAAAAAAAC</mark>	GGCCGATTC AGTTTGTAG GGGCCGTTG GTCTTCCCA GTCTTCCCA CACATGACCA CACATGACCA CACATGACCA CACATGACCA CACATGACCA CACATGACA CACATGACA CACTACACA CATTACACA AATTGGTTA CACCGCTAC	ATTAATGCA AAACGCAAAA CTTCACAAC CTCAGCCTT CCOBC gag taaa CTGAGCCCR TAAAACTGT CTGATTTAL TTCTSAAAC AGCATTTA GTCAAAATA GTCAAAATA GTCAAAATA GTCAAAATA AACGGCTTT ATTSGTTCAS CAGCGGTCAS CAGCGGTCG	SCTESCACEACA AAGECCATCEGT STICAAATCEGE TEGTITTATTTG gegegtaataeg gggatecacta ICAT 201711 CTECTACATAA ATEECAAAESTA TECETACACEGE CECAATCACEAA AACTTTESCACEAA AACTTTESCACEAA AACACTESCAEAA ACCCESTAEAAA TTECTTECCEG	GGTTTCC CAGGATG TCCCGGC ATGCCTG actcact gttctag gttctag GCTCG GCSTTGC ACAGTAA GSSCTCG GCSTTGC TCSGAAT ISAATAA TCTCACC TCSGAAT GTATTCA GCATTAC ASATCAA ATCAACA
97 GTCAGTG 193 CGACTGG 289 GCCTTCT 385 GGATTG 481 CCAGTTG 577 atagge 673 aprovement 769 GCTCTGG 865 TACAAGG 961 CGATAAT 1057 CAATGAT 1057 CAATGAT 1153 ATGGTTA 1249 GTTCCTG 1345 CGSTTTG 1441 CGATTCA 1533 TAATCCT 1533 TAATCCT 1533 TAATCCT 1825 AGGATCT 1921 GCTACCA	AGCGAGGAG GCTTAGTTI TCCTACTCA CCTACTCA CCTACTCA CCTACTCA CCCACTACTCA GCCCSGGTCA GCCCSGGTCA GTCACCACA GTCACCACA GTCACCACA GTCACCACA GTCACCACA GTCACCACA GTCACCACA GTCACCACA GTCACCACA GTCACCACA GTCACCACA GTCACCACA GTCACCACA GTCACCACA GTCACCACA GTCACCACA GTCACCACA GTCACCACACA GTCACCACACACACACACACACACACACACACACACACAC	AGCGCAAGA CAGTGAGCC CGATGCCTG AGGGCAGCC CGCGTTAAC CGCGRCCC CGCGRCCC CAGCGRCCC CAGCGCAAG CGTTCGAT CGCGCCAAG CGCTTTTT CCGCGCAAG CCTTTTTT CCGAAGG		CGCAAACCGCC AATACGCGTAC GCGGGCCGTCCT AACAACAGATA ATGTTTTCCCA CATGTTTTCCCA CATGTCGACAGAC CGCCGACGACAGA CGCCGACGACAGA CGCTCGACGCA CGCATGCCTCGA CGCACGCCCGACGA AATTGCCCTCGATGA AATTGCCCTCAA AATCCCCTTAA AATCCCCTTAA	TCTCCCCGCGCGCTT CGCTAGCCAGGAAG GCCCGCCACCCTCC AAACGAAAGGCCCA GTCACGACGT <mark>tgtB CgatBagcttgtB CgatBagcttgtB CgatBagcttgtB CgatBagcttgtB CgatCaCGATGCSC ATTAGCCCGATGCSC ATTAGAACAATACTC GCCTGTTGACCAGGCA TGAGTTTCCCTT GTTTTGACCAGGCCA GGTGTTTCCCTT GCAACAAAAAAAAC GATACCAAATACTG</mark>	GGCCGATTC AGTTTGTAG GGGCCGTTG GGGCCGTTG GTCTTCCCA GTCTTCCCA CAGACGACGAC CATGAACAA ACATGGATG CAGACTTCAC CAGACTTCAC CAGACTTCACACA CATTACACA AATTGGTTA CATTCCACT CACCGCTAC GTCTCCACT	ATTAATGCA AAACGCAAAC CTTCACAAC CTTCACAAC CTGAGCCTT CCOULT CCOULT CCOULT CTGATTTAL TAAAACTGT CTGATTTAL CTGATTTAL GTCAAAACATA GTCAAAACATA GTCAAAATA CGCTCAGG AAATGCATAL AACGGCTTT ATTSGTTGT CACCGGTCG CACCGGTCG TGTAGCCGT	SCTESCACEACA AAGECCATCEGT STICAAATCEGE TEGTTITATTIG gegegtaataeg ggggatecacta ICAT 95 TYTH CTECTTACATAA ATEGCAAAESTA TEGCAACAESTA AACTTITECCT AACTTIGECAA AACACTEGCAAA AACCTEGCAAA AACACTEGCAAA AACACTEGCAAA AACACTEGCAAA AACACTEGCAAA AACACTEGCAAA	GGTTTCC CAGGATG TCCCGGC ATGCCTG actcact gttctag gttctag GGCTCG GGGTTGC ACAGTAA GGGTTGC TCGGAAT TCTCACC TCGGAAT GTATTCA GCATTAC ASATCAA ATCAAGA ACTTCAA
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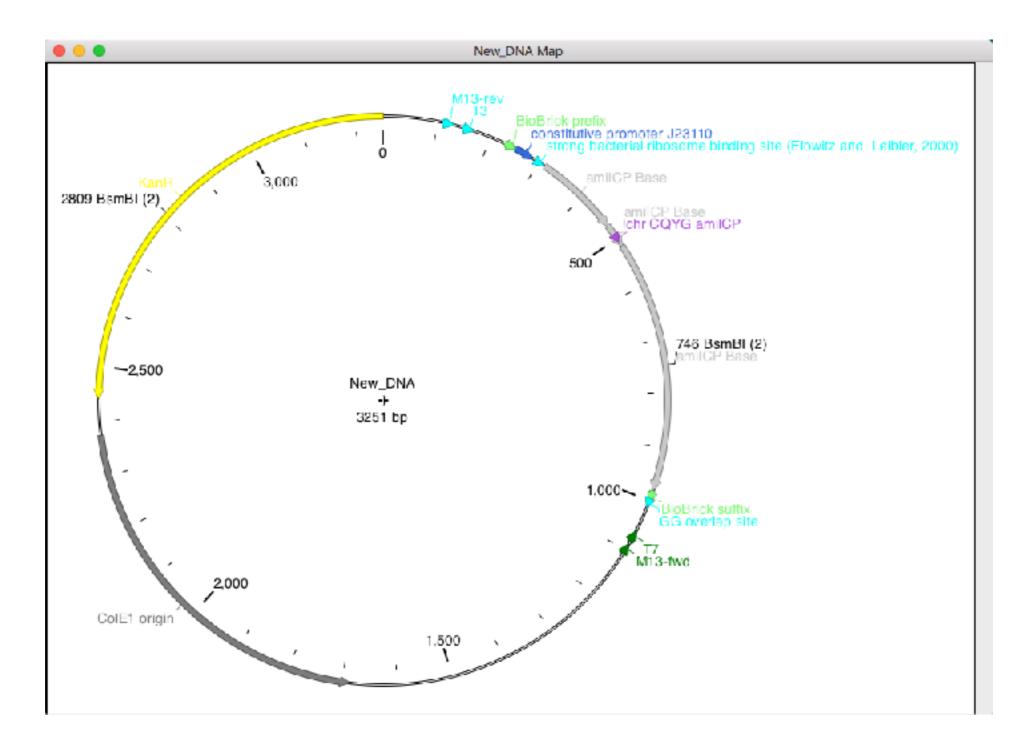


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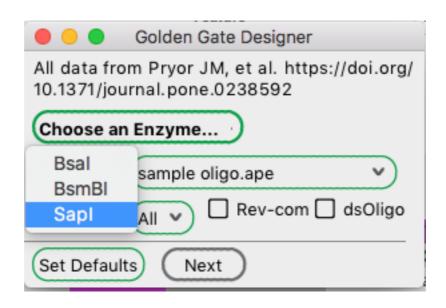
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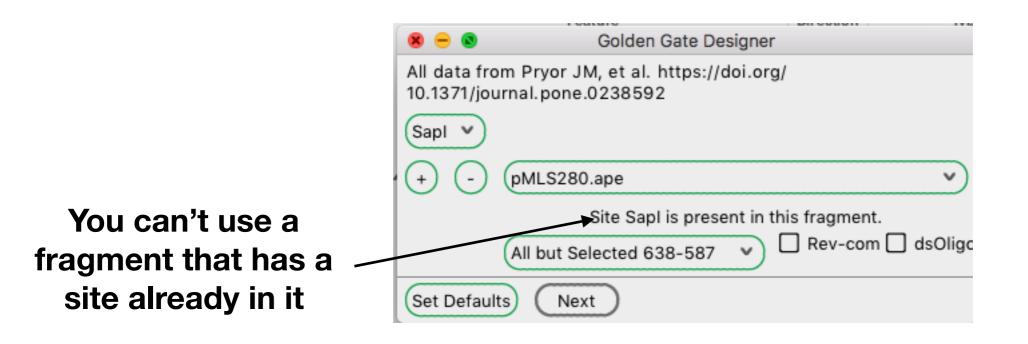
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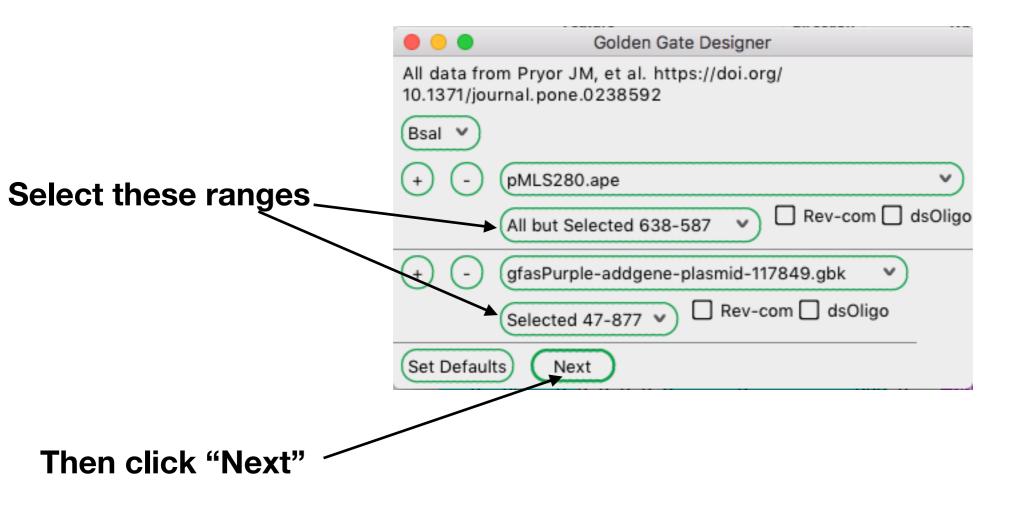






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Calculates the total	🔴 😑 🧲 Golden Gate Designer
	Total efficiency: 100.0%
efficiency (on-target to	pMLS280 Left overhang: gaat, 100.0% efficiency.
off target)	Right overhang: toat, 100.0% efficiency.
	fwd primer (Tm1=59.4°C, Tm2= 74.7°C): GCGCggteteG gaatteetgeageeeggg rev primer (Tm1=59.4°C, Tm2= 70.9°C: GCGCggteteGte attegeeetalagtgagtegtattaeg
Each frommont and the	gfasPurple-addgene-plasmid-117849 Ipft overhang: atga, 100.0% efficiency.
Each fragment and the ——	Right overhang: attc, 100.0% efficency. fwd primer (Tm1=59.4°C, Tm2= 73.6°C): GCGCggteteGat gaattegeggeegettet
primers needed	rev primer (Tm1=60.4°C, Tm2=70.7°C: GCGCggtctcGatte tottttcggttttaaagaaaagggcagg
You can set default ——	(Set Defaults) Back (Calculate New) (Recalculate) (Generate Product) (Done)
	(Set Defaults) Back (Calculate New) (Recalculate) (Generate Product) (Done)
values here bases	

	Golden Gate Designer
Default primer Tm	59
Default search time	1.0
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Default site linker sequence G	
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	🔴 🕘 Golden Gate Designer
	Total efficiency: 100.0%
	pMLS280 Left overhang: gaat, 100.0% efficiency. Right overhang: toat. 100.0% efficiency. fwd primer (Tm1=59.4°C, Tm2= 74.7°C): GCGCggteteG gaatteetgeageeeggg rev primer (Tm1=59.4°C, Tm2= 70.9°C: GCGCggteteGte attegeeelalagtgagtegtattaeg
	gfasPurple-addgene-plasmid-117849 Left overhang: atga, 100.0% efficiency. Right overhang: attc, 100.0% efficency. fwd primer (Tm1=59.4°C, Tm2= 73.6°C): GCGCggteteGat gaattegeggeegettet rev primer (Tm1=60.4°C, Tm2= 70.7°C: GCGCggteteGatte tetttteggttttaaagaaaaagggeagg
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Colden Gate Designer
otal efficiency: 100.0%
MLS280 eft overhang: gaat, 100.0% efficiency. ight overhang: toat, 100.0% efficiency. wd primer (Tm1=59.4°C, Tm2= 74.7°C): GCGCggteteG gaatteetgeageeeggg ev primer (Tm1=59.4°C, Tm2= 70.9°C: GCGCggteteGte attegeeetatagIgagtegtattaeg
fasPurple-addgene-plasmid-117849 eft overhang: atga, 100.0% efficiency. light overhang: attc, 100.0% efficency. wd primer (Tm1=59.4°C, Tm2= 73.6°C): GCGCggteteGat gaattegeggeegettet ev primer (Tm1=60.4°C, Tm2= 70.7°C: GCGCggteteGatte tetttteggttttaaagaaaaagggeagg
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77 ΤΑΑΑΑΑΤΑΤΑΤΑΤΟΙ	ICATGAACAAI	TAAAACTGT	CEGCETACAL	TAAACAGTAATACAAG	AGGETATI	ATGAGCCA	TATTCAAC656/	MACG
265 TCGAGGCCGCGATT	TAAATTCCAA	CATGGATGC	IGATTIATA	IGEG TATAAATGE G CI	ICECEATA	ATGTCGGG	CAATCAGGTGCO	SACSA
53 TCTATCGCTTGTAT	GGGAAGCCC	GATGEGEEA	GAGETIGETER	LI GAAACA I GGCAAAG	99159561	TECOATE	ATGETACAGATO	aAGAT
41 GETCAGACTAAACT	1 (a CaC - 1 4a CaC - CaC - A							TCLCC
					TCCGTACT		TGCATGGTTACT	TCACC
29 ACTGCGATCCCCGG			ATTAGAAGAA	VTATCCTGATTCAGG	ΓΟΟΘΤΑΟΤ ΓΘΑΛΛΑΤΑ	TIGTTGAT	GCGCTGGCAGT	STTCC
29 ACTGCGATCCCCG6 517 TGCGCCGGTTGCAT		ATTCCAGGT GTTTGTAAT	ATTAGAAGA/ TGTCCTTTTA			TIGTTGAT	TGCATGGTTACT GCGCTGGCAGTC CGCAATCACGAA	STTCC
529 <mark>ACTGCGATCCCCGG</mark> 517 TGCGCCGGTTGCAT 705 TAACGGTTTGGTTG		ATTCCAGGT GTTTGTAAT	ATTAGAAGAA	VTATCCTGATTCAGG		TIGTTGAT	GCGCTGGCAGT	STTCC
29 ACTGCGATCCCCGG 17 TGCGCCGGTTGCAT 705 TAACGGTTTGGTTG 793 CCATTCTCACCGGA		ATTCCAGGT GTTTGTAAT	ATTAGAAGA/ TGTCCTTTTA	VTATCCTGATTCAGG		TIGTTGAT	GCGCTGGCAGTC CGCAATCACGAA AATGCATAAACT TTAATAGGTTG1	ATTCC ATGAA FTTTG FATTG
29 ACTSCGATCCCC66 17 TECSCCGGTTGCAT 05 TAACSGTTTSGTT6 793 CCATTCTCACCG6A 81 ATGTTGGACGAGTC	SAAAAACAGC/ ITCGATTCCT(SATGCGAGTG/ ATTCAGTCGT(ISGAATCGCA(ATTCCAGGT GTTTGTAAT ATTTTGATG CACTCATGG GACCGATAC	ATTAGAASAA TGTCCTTTTA ACGAGCGTAA TGATTTCTCA CASSATCTTC	VTATECTGATTCAGG ACAGCGATCGCGTAT ATGGCTGGCCTGTTG/ ACTTGATAACCTTATT SCCATCCTATGGAACT	EGAAAATA ETTEGTET AACAAGTE ETTEGACE EGEETEGE	TTGTTGAT(CGCTCAGG) TGGAAAGA AGGGGAAA TGAGTTTT(GCGCTGGCAGTO CGCAATCACGAA AATGCATAAACT TTAATAGGTTGT CTCCTTCATTAC	STTCC ATGAA FTTTG FATTG CAGAA
29 ACTGCGATCCCCGG 17 TGCGCCGGTTGCAT 105 TAACGGTTGGTTG 193 CCATTCTCACCGGA 181 ATGTTGGACGAGTC 195 ACGSCTTTTTCAAA	SAAAAACAGCA TTCGATTCCT(SATGCGAGTGA ATTCAGTCGT(CGGAATCGCA(AATATSGTA)	ATTCCASST GTTTGTAAT ATTTTGATG CACTCATSG GACCGATAC TTGATAATC	ATTAGAAGA/ TGTCCTTTTA ACSAGCGTAA TGATTTCTCA CASSATCTTC CTSATATSAA	ATATECTEATTCAEGT ACASCEATCECETAT ATEECTEECCTETTEA ACTTEATAACCTTATT SCCATCCTATEEAACT	IGAAAATA ITTEGTET AACAAGTE ITTTGACE IGEETEGE ATTTGATE	CECTCASS CGCTCASS TGSAAASAA ASSGGAAA TSAGTTTT CTCGATSA	GCGCTGGCAGTO CGCAATCACGA/ AATGCATAAACT TTAATAGGTTG CTCCTTCATTAO GTTTTTCTAATO	GTTCC ATGAA FTTTG FATTG CAGAA CAGAA
29 ACTSCGATCCCCGG 17 TECSCCGGTTGCAT 105 TAACSGTTTGGTTG 193 CCATTCTCACCGGA 181 ATGTTGGACSAGTC 169 ACGSCTTTTTCAAA 157 TTGGTTAATTGGTT	SAAAAACAGC ITCGATTCCT SATGCGAGTG/ STTCAGTCGT(ISGAATCGCA(AATATSGTA) IGTAACACTG	ATTCCAGGT GTTTGTAAT ATTTTGATG CACTCATGG GACCGATAC FTGATAATC GCAGAGCAT	ATTAGAAGA/ TGTCCTTTTA ACGAGCGTAA TGATTTCTCA CASSATCTTC CTSATATSAA TACGCTGACT	ATATECTGATTCAGG ACAGCGATCGCGTAT ATGGCTGGCCTGTTG/ ACTTGATAACCTTAT GCCATCCTATGGAACT ATAAATTGCAGTTTC/ ITGACGGGACGGCGC	IGAAAATA ITTCGTCT AACAAGTC ITTTGACG IGCCTCGG ATTTGATG AAGC <mark>TCA</mark> T	CECTCASS CGCTCASS TGSAAASAA ASSGGAAA TSAGTTTT CTCGATSA	GCGCTGGCAGTO CGCAATCACGAA AATGCATAAACT TTAATAGGTTGT CTCCTTCATTAC GTTTTTCTAATG TCCCTTAACGTO	STTCC ATGAA FTTTG FATTG CAGAA CAGAA SAGTT
29 ACTSCGATCCCCGG 17 TECSCCGGTTGCAT 05 TAACSGTTTSGTTG 93 CCATTCTCACCGGA 81 ATGTTGGACSAGTC 959 ACGSCTTTTTCAAA 957 TTGGTTAATTGGTT 145 ACGCSTCGTTCCAC	SAAAAACAGCA ITCGATTCCT(SATGCGAGTGA ATTCAGTCGT(CGGAATCGCA(AATATSGTAT IGTAACACTG(CTGAGCGTCA)	ATTCCAGGT GTTTGTAAT ATTTTGATG CACTCATGG GACCGATAC FTGATAATC GCAGAGCAT	ATTAGAASA/ TGTCCTTTTA ACSAGCGTAA TGATTTCTCA CASSATCTTC CTSATATSAA TACSCTGACT GAAAAGATCA	ATATECTEATTCAEGT ACAGCGATCGCETAT ATEGCTGGCCTGTTGA ACTTGATAACCTTATT SCCATCCTATGGAACT ATAAATTGCAGTTTCA TTGACGGGACGG	IGAAAATA ITTCGTCT AACAAGTC ITTTGACG IGCCTCGG ATTTGATG AAGCICAT GATCCTTT	CECTCASS CGCTCASS TGSAAASAA ASSGGAAA TSAGTTTT CTCGATSA	GCGCTGGCAGTO CGCAATCACGA/ AATGCATAAACT TTAATAGGTTGT CTCCTTCATTAC GTTTTTCTAATC GCCTTAACSTC GCGTAATCTSCT	STTCC ATGAA FTTTG FATTG CAGAA CAGAA SAGTT FGCTT
29 ACTGCGATCCCCGG 517 TGCGCCGGTTGCAT 505 TAACGGTTTGGTTG 531 CCATTCTCACCGGA 381 ATGTTGGACSAGTC 359 ACGGCTTTTTCAAA 357 TTGGTTAATTGGTT 445 ACGCGTCGTTCCAC 333 GCAAACAAAAAAA	SAAAAACAGCA ITCGATTCCT(SATGCGAGTGA ATTCAGTCGT(CGGAATCGCA(AATATSGTAT IGTAACACTG(CTGAGCGTCA)	ATTCCAGGT GTTTGTAAT ATTTTGATG CACTCATGG GACCGATAC FTGATAATC GCAGAGCAT	ATTAGAASA/ TGTCCTTTTA ACSAGCGTAA TGATTTCTCA CASSATCTTC CTSATATSAA TACSCTGACT GAAAAGATCA	ATATECTGATTCAGG ACAGCGATCGCGTAT ATGGCTGGCCTGTTG/ ACTTGATAACCTTAT GCCATCCTATGGAACT ATAAATTGCAGTTTC/ ITGACGGGACGGCGC	IGAAAATA ITTCGTCT AACAAGTC ITTTGACG IGCCTCGG ATTTGATG AAGCICAT GATCCTTT	CECTCASS CGCTCASS TGSAAASAA ASSGGAAA TSAGTTTT CTCGATSA	GCGCTGGCAGTO CGCAATCACGA/ AATGCATAAACT TTAATAGGTTG CTCCTTCATTAC GTTTTTCTAATG GCGTAATCTGCT GGTAACTGGCT	STTCC ATGAA FTTTG FATTG CAGAA CAGAA SAGTT FGCTT FCAGC
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29 ACTGCGATCCCCGG 517 TGCGCCGGTTGCAT 705 TAACGGTTTGGTTGGTTGGTTGGTTGGACGGAGTG 793 CCATTCTCACCGGA 793 CCATTCTCACCGGA 793 CCATTCTCACCGGA 794 ATGTTGGACGAGTG 795 ACGGCTTTTCAACGGA 796 ACGGCTTTTCAACGGA 797 TTGGTTAATTGGTT 145 ACGCGTCGTTCCAC 233 GCAAACAAAAAAAAAAA 221 ACAGCGCAGATACCG 199 CTCTGCTAATCCTG	GAAAAACAGC ITCGATTCCT GATGCGAGTG/ GTCAGTCGTCGT GGAATCGCA GGAATCGCA IGTAACACTG CTGAGCGTCA CAACACTGT GTTACCAGTC	ATTCCASST GTTTGTAAT ATTTTGATG CACTCATSG GACCGATAC TTGATAATC GCAGAGCAT GACCCCGTA GACCCCGTA CAGCCGTSG FCTTCTAST	ATTAGAASAA TGTCCTTTA ACSAGCGTAA TGATTTCTCA CASSATCTTC CASSATCTTC CTSATATSAA TACSCTGACT GAAAAGATCA TTTSTTTSCC GTASCCGTAC	ACASCGATCGCGTAT ACASCGATCGCGTAT ATGGCTGGCCTGTTG/ ACTTGATAACCTTAT GCCATCCTATGGAACT ATAAATTGCAGTTTC/ ITGACGGGACGGCGC/ AAAGGATCTTCTTGAC GGCATCAAGAGCTACC AAGTCGTGTCTTACCC	IGAAAATA ITTCGTCT AACAAGTC ITTTGACG IGCCTCGG ATTTGATG AAGCTCAT GAACTCTTT GAACTCTTT GAAGAACT GGGTTGGA	CITISTTEATI COCTCASS TIGGAAAGA/ ASSGGAAA TIGAGTTTT CITISTAGAA CITISTAGCAI CITISTAGCAI	GCGCTGGCAGTO CGCAATCACGA/ AATGCATAAACT TTAATAGGTTGT CTCCTTCATTAC GTTTTTCTAATG GCGTAATCTGCT GGTAACTGGCT GGTAACTGGCT GGTAACTGGCT GGTAACTGGCT	STTCC ATGAA FTTTG FATTG CAGAA CAGAA SAGTT FGCTT FCASC CCTCG SATAA
29 ACTGCGATCCCCGG 517 TGCGCCGGTTGCAT 705 TAACGGTTTGGTTGGTTGGTTGGT 793 CCATTCTCACCGGA 881 ATGTTGGACSAGTC 959 ACGSCTTTTTCAAA 957 TTGGTTAATTGGTT 145 ACGSCTCGTTCCAC 833 GCAAACAAAAAAAA 841 ACGSCGCAGCAGATACC 859 ACGSCGCAGCGGTCCAC 829 ACGSCGCAGCAGATACC 821 ACASCGCAGATACCCC 821 ACASCGCAGATACCCCC 822 ACASCGCAGCAGATACCCCCC 827 GCCGCAGCGGTCCCCCCCCCCCCCCCCCCCCCCCCCCCC	SAAAAACAGC ITCGATTCCT(SATGCGAGTG/ ATTCAGTCGT(CGGAATCGCA(AATATSGTAT ISTAACACTG(CACCGCTAC) SAATACTGTT STTACCAGTG(SCTCAACGC)	ATTCCAGGT GTTTGTAAT ATTTTGATG CACTCATGG GACCGATAC TTGATAATC GCAGAGCAT GACCCCGTA CAGCGCTGG FCTTCTAGT GCTGCTGCC GGGGTTCGT	ATTAGAASA/ TGTCCTTTTA ACSAGCGTAA TGATTTCTCA CASSATCTTC CTSATATSAA TACSCTGACT GAAAAGATCA TTTSTTTSCC GTASCCGTAC AGTSGCGATA	ACAGCGATCGCGTAT ACAGCGATCGCGTAT ATGGCTGGCCTGTTG/ ACTTGATAACCTTATT SCCATCCTATGGAACT ATAAATTGCAGTTTC/ TGACGGGACGGCGC/ AAAGGATCTTCTTGAC GGATCAAGAGCTACC ATTAGGCCACCACTTC AAGTCGTGTCTTACCC	IGAAAATA ITTCGTCT AACAAGTC ITTTGACG IGCCTCGG ATTTGACG AAGCTCAT GACCTCT GAGGATCGA CGACCTAC	CECECAGO CONTRACTOR CO	GCGCTGGCAGTO CGCAATCACGA/ AATGCATAAACT TTAATAGGTTGT CTCCTTCATTAC GTTTTTCTAATG GCGTAATCTGCT GGTAACTGGCT GGTAACTGGCT GGTAACTGGCT GGTAACTGGCT GAGATACCTAC/	ATGAA TTTTG TATTG TATTG CAGAA CAGAA CAGAA GAGTT TGCTT TCAGC CCTCG GATAA AGCGT
529 ACTGCGATCCCCGG 617 TGCGCCGGTTGCAT 705 TAACGGTTTGGTTGGTTG 703 CCATTCTCACCGGA 881 ATGTTGGACSAGTC 969 ACGSCTTTTTCAAA 957 TTGGTTAATTGGTT 145 ACGSSTCGTTCCAC 233 GCAAACAAAAAAAAA 321 ACASEGCASATACCTO 199 CTCTSCTAATCCTO 197 GGCSCAGCSSTCGO 585 GAGCTATGASAAAA	SAAAAACAGC/ ITCGATTCCT(SATGCGAGTG/ ATTCAGTCGT(CGGAATCGCA(GGGAATCGCACGCACGCACGCACGCACGCACGCACGCTA SCTCGACGCTACGCACGCTACGCACGCTACGCACGCTACGCACGC	ATTCCASST GTTTGTAAT ATTTTGATG CACTCATSG GACCGATAC ITGATAATC GCAGAGCAT GACCCCSTA CAGCGCTSG ICTTCTAST GCTGCTSCC GSGGTTCST ICCCGAASG	ATTAGAASAA TGTCCTTTA ACSAGCGTAA TGATTTCTCA CASSATCTTC CTSATATSAA TACSCTGACT GAAAAGATCA TTTSTTTSCC GTASCCGTAC GCACACASCC GASAAAGSCC	ACAGCATCGATCAGGT ACAGCGATCGCGTAT ACGCTGGCCTGTTG/ ACTTGATAACCTTATT SCCATCCTATGGAACT ATAAATTGCAGTTTC/ TTGACGGGACGGCGCA AAAGGATCTTCTTGAC GGATCGTGTCTTACCC CCAGCTTGGAGCGAAC GGACAGGTATCCGGT/	IGAAAATA ITTCGTCT AACAAGTC ITTTGACG IGCCTCGG ATTTGATG AAGCTCAT GAACTCTT CAAGAACT GGGCTTGGA CGACCTAC	CITISTTEATO COCTCASSO TIGSAAASAA ASSOGAAA TIGAGTITTO CITISCAAAA TITTCISCO TITTCISAA CITISTAGCAO CITISTAGCAO CICISTAGCAO CICISAACACO ACCGAACTO	GCGCTGGCAGTO CGCAATCACGA/ AATGCATAAACT TTAATAGGTTG TCCCTTCATTAC GTTTTTCTAATG GCGTAATCTGCT GGTAACTGGCTT CCGCCTACATAC GAGATACCTAC/ ACAGGAGAGCGC	ATGAA TTTTG TATTG CAGAA CAGAA CAGAA GAGTT TCAGC CCTCG GATAA AGCGT CACGA
529 ACTSCGATCCCCGG 517 TGCGCCGGTTGCAT 705 TAACSGTTTSGTTG 705 TAACSGTTTSGTTG 705 CCATTCTCACCGGA 881 ATGTTGGACSAGTC 969 ACGSCTTTTCAAA 957 TTGSTTAATTGGTT 145 ACGSCTCGTTCCAC 821 ACGSCTCGTTCCAC 823 GCAAACAAAAAAAAC 821 ACGSCGCAGATACCC 821 ACGSCGCAGAAAAAAAAC 821 ACGSCGCAGAAAAAAAAC 821 ACGSCGCAGAAAAAAAAC 821 ACGSCGCAGAAAAAAAAC 821 ACGSCGCAGAAAAAAAAC 821 ACGSCGCAGAAAAAAAAC 821 ACGSCGCAAAAAAAAAAC 821 ACGSCGCAGAAAAAAAAAAAAAAAAAAAAAAAAAAAAAA	GAAAAACAGC TCGATTCCT GATGCGAGTGA ATTCAGTCGTG CGGAATCGCA GGAATCGCA CTGAGCGTCA CTGAGCGTCA CAACTACTGT GTTACCAGTG GGCCACGCT GGCCACGCT GGCAACGCC	ATTCCAGGT GTTTGTAAT ATTTTGATG CACTCATGG GACCGATAC TTGATAATC GCAGAGCAT GCCGGTAG GCCGCGTAG GCCGCGAGG TGGTATCTT	ATTAGAASA/ TGTCCTTTA ACSAGCGTAA TGATTTCTCA CASSATCTTC CASSATCTTC CASSATCTCA CASSAAAGATCA TACSCTGACT GCACACASCC GCACACASCC TATAGTCCTC	ACAGCAGGTTCCGGT/ ACAGCGATCGCGTAT ACGCTGGCTGGCCTGTTG/ ACTTGATAACCTTAT GCCATCCTATGGAACT ATAAATTGCAGTTTC/ TTGACGGGACGGCGCGC AAAGGATCTTCTTGAC GGGATCAAGAGCTACC GTTAGGCCACCACTTC AGTCGTGTCTTACCC GGACGGGTTTCGCCAC	IGAAAATA ITTCGTCT AACAAGTC ITTTGACG IGCCTCGG ATTTGACG AAGCTCAT GACCTCAT GACCTCAT GACCTAC AAGCGGCCAC	CITISTTEATI COCTCASS TIGSAAAGA/ ASSGGAAA TIGAGTTTTC CTCGATGA(GACCAAAA TITTCTSC) TITTCCGAA(CTGTAGCAC) CTGTAGCAC CTCAAGACC(ACCGAACTC) GGGTCGGA/ TIGAGCGTC)	GCGCTGGCAGTO CGCAATCACGA/ AATGCATAAACT TTAATAGGTTG CTCCTTCATTAC GTTTTTCTAATG GCGTAATCTGCT GGTAACTGGCT GGTAACTGGCT GGTAACTGGCT GAGATACTACCGC GAGATACCTAC/ ACAGGAGAGGGCGC	STTCC ATGAA FTTTG FATTG CAGAA CAGAA SAGTT FGCTT FGCTT FGCTC GATAA AGCST CACGA FGCTC
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529 ACTSCGATCCCCGG 517 TGCGCCGGTTGCAT 705 TAACGGTTTGGTTGGTTG 705 TAACGGTTTGGTGGTG 706 CCATTCTCACCGGA 881 ATGTTGGACSAGTC 969 ACGSCTTTTTCAAA 957 TTGGTTAATTGGTT 145 ACGCSTCGTTCCAC 233 GCAAACAAAAAAAAA 321 AGASCGCAGATACCCO 199 CTCTSCTAATCCTC 197 GCCSCAGCSSTCGC 585 GAGCTATGASAAAA 573 GGGASCTTCCAGGG 573 GGGASCTTCCAGGG 574 GTCASGGGSSCGGA 575 GTCASGGGSSCGGA	SAAAAACAGC ITCGATTCCT(SATGCGAGTGA ATTCAGTCGT(CGGAATCGCA(CGGAATCGCA CACCGCTAC CACCGCTAC CACCGCTAC CACCGCTAC CACCGCTACGCA CGCCACGCT CGCCACGCT CGCCACGCT CGCCACGCT CGCCACGCT CCCCGATTC	ATTCCAGGT GTTTGTAAT ATTTTGATG CACTCATGG GACCGATAC ITGATAATC GCAGAGCAT GACCCCGTA GACCCCGTA GACCCCGTA GACCCCGTA GCTGCTGCC GGGGTTCGT ICCCGAAGG TGGTATCTT AAAACGCCA IGTGGATAA	ATTAGAASA/ TGTCCTTTTA ACSAGCGTAA TGATTTCTCA CASSATCTTC CASSATCTTC CTSATATSAA TACSCTGACT GAAAAGATCA TTTSTTTSCC GASACAGACA GCACACAGCG GASAAAGSCC TATAGTCCTC GCACACGCSSC ACCGTATTACC	ACAGCGATCGCGTAT ACAGCGATCGCGTAT ACGCTGGCCTGTTG/ ACTTGATAACCTTAT SCCATCCTATGGAACT ATAAATTGCAGTTTC/ TTGACGGGACGGCGC/ AAAGGATCTTCTTGAC GGATCAAGAGCTACCG GGACCAGGTATCCGGT/ GGACCAGGTATCCGGT/ GCCTTTGAGTGAGCCACC CCCTTTGAGTGAGCCACC	IGAAAATA ITTCGTCT AACAAGTC ITTTGACG IGCCTCGG ATTTGATG AAGCTCAT GATCCTTT CAAGAACT GGCCTTAC AAGCGGCCAA IGGCCTTT IGGCCTTT	CTCAAGAC CGCTCAGG TGGAAAGA/ AGSGGAAA TGAGTTTTC CTCGATGA/ GACCAAAA TTTTCTGC TTTCCGAA CTCAAGAC CTCAAGAC CTCAAGAC CTCAAGAC GGGTCGGA/ TGAGCGTC CTCGCCCC	GCGCTGGCAGTO CGCAATCACGA/ AATGCATAAACT TTAATAGGTTGT CTCCTTCATTAC GTTTTTCTAACSTC GCGTAATCTGCT GGTAACTGGCT GGTAACTGGCT GGTAACTGGCT GAGATACCTACA GAGATACCTACA ACAGGAGAGCGC GATTTTTGCTCACA AGCCGAACGACC	STTCC ATGAA TTTG TATTG CAGAA CAGAA CAGAA CAGAA GAGTT FCAGC CCTCG GATAA AGCGT CACGA FGCTC CGTCC CGAGC
529 ACTSCGATCCCCGG 517 TGCGCCGGTTGCAT 705 TAACSGTTTSGTTG 705 TAACSGTTTSGTTG 705 TAACSGTTTSGTTG 707 CCATTCTCACCGGA 881 ATGTTGGACSAGTC 969 ACGSCTTTTCAAA 957 TTGSTTAATTGGTT 145 ACGSCTCGTTCCAC 323 GCAAACAAAAAAAAA 321 ACASCGCAGATACCCO 409 CTCTSCTAATCCTCO 525 GAGCTATGASAAAAA 673 GGCASCTTCCAGGGGGGGGGGGGGGGGGGGGGGGGGGGGG	GAAAAACAGC TCGATTCCT GATGCGAGTGA ATCAGTCGTG CGGAATCGCA AATATSGTA IGTAACACTG CACCGCTAC CACCGCTAC GGCCACGCT GGCCACGCT GGCCACGCT GGCCACGCT GGCCACGCT GGCCACGCT GGCCACGCT GGCCACGCT GGCCACGCACGCT GGCCACGCT GGCCACGCACGCA GCCCTGATTC GGCGAGGAGGAA	ATTCCASST GTTTGTAAT ATTTTGATG CACTCATSG GACCGATAC TGATAATC GCAGAGCAT GCAGAGCAT GACCCCSTA CAGCCCCSTA CAGCCCCSTA CAGCCCCSTA CAGCCCCSTA CAGCCCCAASG TGCTATCTT AAAACGCCA GCGGAASAG	ATTAGAASA/ TGTCCTTTA ACSAGCGTAA TGATTTCTCA CASSATCTTC CASSATCTTC CASSATCTCA CASSATCTA CASSATATSAA TACSCTGACTA CASAGAGACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACA	ACAGCATCGATTCAGGT ACAGCGATCGCGTAT ACGCTGGCCTGTTGA ACTTGATAACCTTAT GCCATCCTATGGAACT ATAAATTGCAGTTTCA TTGACGGGACGG	IGAAAATA ITTCGTCT AACAAGTC ITTTGACG IGCCTCGG ATTTGACG IGCCTCGG AAGCTCAT CAAGCAACT CAAGCAACT CGACCTAC AAGCSSCA CTCTGACT IGGCCTTT IGGCCTTT IGGCCGCGCGCG	CITISTTEATO COCTCASSO TGSAAASAA ASSGGAAA TSAGTTTTO CTCGATSAO GACCAAAA TTTTCTSCO TTTCCGAA CTSTAGCAO CTCAAGACO ACCGAACTO GSSTCGSAA TGAGCGTCO TGCTGGCCC CTCGCCGCA	GCGCTGGCAGTO CGCAATCACGA/ AATGCATCACGA/ AATGCATCACGA TTAATAGGTTG CTCCTTCATTAC GTTTTTCTAACGT GCGTAATCTGCT GGTAACTGGCT GGTAACTGGCT GGTAACTGGCT GAGATACCTAC/ ACAGGAGAGCGC GATATTTGTGAT TTTTGCTCACAT AGCCGAACGACC TTCATTAATGC/	ATGAA TTTTG TATTG TATTG TATTG TATTG TAGAA CAGAA CAGAA CAGAA CAGAT TGCTT TCAGC CCTCG CATGA CACGA TGCTC TGTTC CGAGC AGCTG
529 ACTGCGATCCCCGG 517 TGCGCCGGTTGCAT 705 TAACGGTTTGGTTGGAT 705 TAACGGTTTGGTTGGAT 708 CCATTCTCACCGGA 881 ATGTTGGACSAGTC 969 ACGGCTTTTTCAAA 957 TTGGTTAATTGGTT 145 ACGCGTCGTTCCAC 233 GCAAACAAAAAAAAA 321 ACAGCGCAGATACCCAC 199 CTCTGCTAATCCTCAC 197 GCCGCAGCGGTCGCG 585 GAGCTATGASAAAA 673 GGGAGCTTCCAGGG 673 GGGAGCTTCCAGGGGGCGGA 674 GTCAGGGGGGCGGA 674 GTCAGGGGGGCGGA	GAAAAACAGC TCGATTCCT GATGCGAGTGA ATCAGTCGTG CGGAATCGCA AATATSGTA IGTAACACTG CACCGCTAC CACCGCTAC GGCCACGCT GGCCACGCT GGCCACGCT GGCCACGCT GGCCACGCT GGCCACGCT GGCCACGCT GGCCACGCT GGCCACGCACGCT GGCCACGCT GGCCACGCACGCA GCCCTGATTC GGCGAGGAGGAA	ATTCCASST GTTTGTAAT ATTTTGATG CACTCATSG GACCGATAC TGATAATC GCAGAGCAT GCAGAGCAT GACCCCSTA CAGCCCCSTA CAGCCCCSTA CAGCCCCSTA CAGCCCCSTA CAGCCCCAASG TGCTATCTT AAAACGCCA GCGGAASAG	ATTAGAASA/ TGTCCTTTA ACSAGCGTAA TGATTTCTCA CASSATCTTC CASSATCTTC CASSATCTCA CASSATCTA CASSATATSAA TACSCTGACTA CASAGAGACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACACASCC CASACA	ACAGCATCGATTCAGGT ACAGCGATCGCGTAT ACGCTGGCCTGTTGA ACTTGATAACCTTAT GCCATCCTATGGAACT ATAAATTGCAGTTTCA TTGACGGGACGG	IGAAAATA ITTCGTCT AACAAGTC ITTTGACG IGCCTCGG ATTTGACG IGCCTCGG AAGCTCAT CAAGCAACT CAAGCAACT CGACCTAC AAGCSSCA CTCTGACT IGGCCTTT IGGCCTTT IGGCCGCGCGCG	CITISTTEATO COCTCASSO TGSAAASAA ASSGGAAA TSAGTTTTO CTCGATSAO GACCAAAA TTTTCTSCO TTTCCGAA CTSTAGCAO CTCAAGACO ACCGAACTO GSSTCGSAA TGAGCGTCO TGCTGGCCC CTCGCCGCA	GCGCTGGCAGTO CGCAATCACGA/ AATGCATCACGA/ AATGCATCACGA TTAATAGGTTG CTCCTTCATTAC GTTTTTCTAACGT GCGTAATCTGCT GGTAACTGGCT GGTAACTGGCT GGTAACTGGCT GAGATACCTAC/ ACAGGAGAGCGC GATATTTGTGAT TTTTGCTCACAT AGCCGAACGACC TTCATTAATGC/	ATGAA TTTTG TATTG TATTG TATTG TATTG TAGAA CAGAA CAGAA CAGAA CAGAT TGCTT TCAGC CCTCG CATAA AGCGT TGCTC TGTTC CGAGC AGCTG

* 10 * 20 * 30 * 40 * 50 * 60 * 70 * 80

Golden Gate reaction: Bsal PCR: pMLS280 Fwd_primer_1 GCGCggtctcGgaattcctgcagcccggg 59.4, 74.7 Rev_primer_1 GCGCggtctcGtcattcgccctatagtgagtcgtattacg 59.4, 70.9 Product length : 2458

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The primers and PCR conditions are in the comments

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* 10 * 20 *	30 * 40	* 50 * 6	50 * 70 * 80 [,]	e ç

The primers are new features in the feature table

The primers are new features in the sequence

Text map dialog

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Text map dialog

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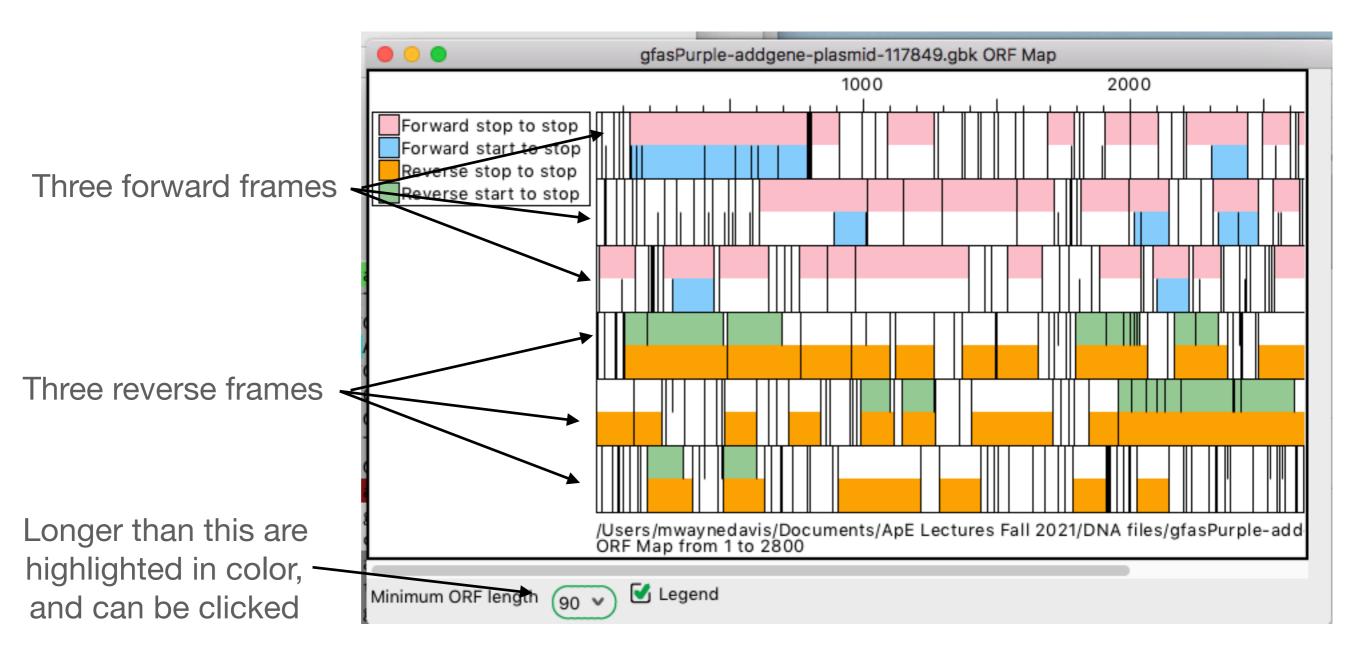
Text map dialog

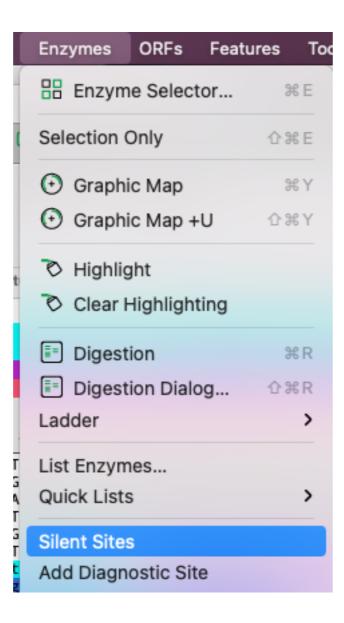
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ORF map dialog

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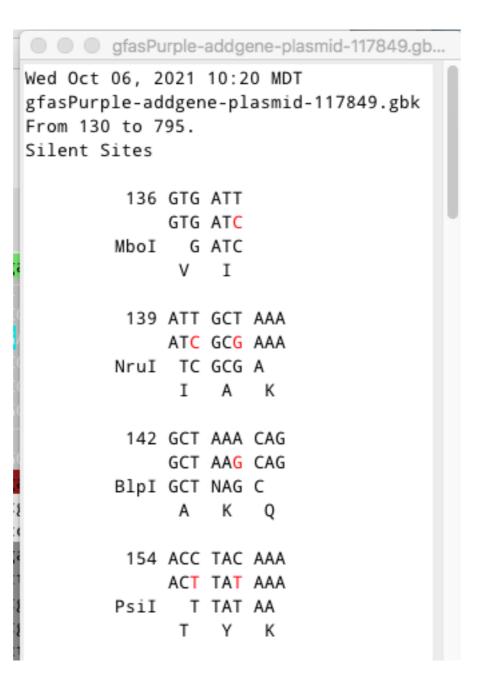
ORF map dialog





	Enzymes ORFs Featu	ures Too
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r	List Enzymes	
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G	Silent Sites	
t	Add Diagnostic Site	

***Select a forward ORF and a set of restriction enzymes before selecting the menu item

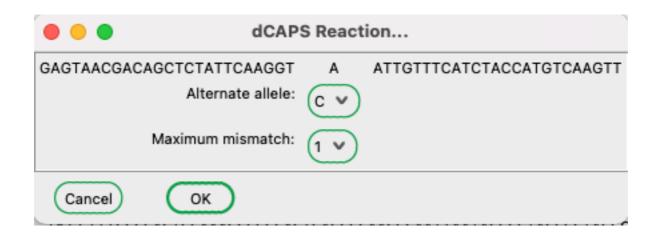


it	Enzymes	ORFs	Features	Тос
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Select diagnostic site if you are adding a site that is NOT in an ORF

Add Diagnostic Sites
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Mismatches: 🔘 1 🔿 2 🔿 3
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dCAPS dialog



dCAPS dialog



dCAPS dialog



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89 GCCTTCTGC 81 GCAGTTCCC 77 atagggcga 73 hgt gate ga 69 GCTCTGGCC 65 TACAAGGG	TTAGTITGATGO CTACTCAGGAGAG TACTCTCGGCCT attgggtaccgg caccgcgglgga CGTGTCTCAAAA	CTEGCAGTTTATEC SCETTCACCEACA/ AACSCTAGCATGE/ SCECCCCCCCEAGA SCECCCCCCCAGAGA	AATACSCSTAC SCGGGCCGTCCT AACAACASATA ATGTTTTCCCA gtcgac <mark>ggtat</mark> I cccltLagt	CGCTAGCCAGGAAG GCCCGCCACCCTCC AAACGAAAGGCCCA GTCACGACGT <mark>TgTa Cgataagcttgata</mark> guggglfaat	AGTITGT/ GGGCCGTT GTCTTCCC aaacgacy tcgaattc cgcUlggo	GAAACGCAAA GCTTCACAAC ACTGAGCCTT gccagtgagc ctgcagcccg glaat <mark>catyg</mark>	AAGGCCATCC GTTCAAATCC TCGTTTTATT gcgc <mark>gtaata ggggatccac</mark> Icala <u>s</u> cipt	GTCAGGATS GCTCCCGGC TGATGCCTG Cgactcact tagttctag
89 GCCTTCTGC 81 GCAGTTCCC 77 atagggcga 73 BREXECCE 69 GCTCTGGCC 65 TACAAGGGG 61 CGATAATGT	TTAGTITGATGO CTACTCAGGAGAG TACTCTCGGCCT attgggtaccgg caccgcgglgga CGTGTCTCAAAA	CTEGCAGTTTATEC SCETTCACCEACA/ AACSCTAGCATGE/ SCECCCCCCCEAGA SCECCCCCCCAGAGA	AATACSCSTAC SCGGGCCGTCCT AACAACASATA ATGTTTTCCCA gtcgac <mark>ggtat</mark> I cccltLagt	CGCTAGCCAGGAAG GCCCGCCACCCTCC AAACGAAAGGCCCA GTCACGACGT <mark>TgTa Cgataagcttgata</mark> guggglfaat	AGTITGT/ GGGCCGTT GTCTTCCC aaacgacy tcgaattc cgcUlggo	GAAACGCAAA GCTTCACAAC ACTGAGCCTT gccagtgagc ctgcagcccg glaat <mark>catyg</mark>	AAGGCCATCC GTTCAAATCC TCGTTTTATT gcgc <mark>gtaata ggggatccac</mark> Icala <u>s</u> cipt	GTCAGGATS GCTCCCGGC TGATGCCTG Cgactcact tagttctag
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89 GCCTTCTGC 81 GCAGTTCCC 77 atagggcga 73 agegsecga 769 GCTCTGGCC 65 TACAAGGGG 661 CGATAATGT 53 ATGGTTACT	TTAGTITGATGO CTACTCAGGAGAG TACTCTCGGCCT attgggtaccgg caccgcgglgga CGTGTCTCAAAA	CTEGCAGTTTATEC SCETTCACCEACA/ AACSCTAGCATGE/ SCECCCCCCCEAGA SCECCCCCCCAGAGA	AATACSCSTAC SCGGGCCGTCCT AACAACASATA ATGTTTTCCCA gtcgac <mark>ggtat</mark> I cccltLagt	CGCTAGCCAGGAAG GCCCGCCACCCTCC AAACGAAAGGCCCA GTCACGACGT <mark>TgTa Cgataagcttgata</mark> guggglfaat	AGTITGT/ GGGCCGTT GTCTTCCC aaacgacy tcgaattc cgcUlggo	GAAACGCAAA GCTTCACAAC ACTGAGCCTT gccagtgagc ctgcagcccg glaat <mark>catyg</mark>	AAGGCCATCC GTTCAAATCC TCGTTTTATT gcgc <mark>gtaata ggggatccac</mark> Icala <u>s</u> cipt	GTCAGGATS GCTCCCGGC TGATGCCTG Cgactcact tagttctag
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DMLS280.ape Q 1\ 🗤 😅 🔛 🖸 10 10 10 🖃 🛏 🚳 🌀 🚍 Sequence ORF %90 circular Start Length End Tm. 59.2°C 2484 596<1> 21<0> 616<0> >7/<7 🛃 Dam/Dem 617 81% GPPL EVD Location 4 Feature Direction Type M13-fwd 536..553 >>> primer TZ 562..582 222 bind MCS-inverted in SK+ miso_feature 588..**6**34 EcoRV miso feature 632..634 666 EcoRV mise_feature 835..637 <<< **^⊞** MCS-inverted in SK+ 203040 50 * 60 70 80 * Try to keep the %GC AGEGAGGAAGEGGAAGAGEGEECAATAEGEAAACEGEETETEEEEGEGETIGGEEGATTEATTEATGEAGETIGGEACGAEAGGTITEE 193 CGACTGGAAAGCGGGCAGTGAGCGCAACGCAATTAATACGCGTACCGCTAGCCAGGAAGAGTTTGTAGAAAACGCAAAAAGGCCATCCGTCAGGATG near 50%, if possible 289 TAGTTTGATGCCT6GCAGTTTAT6GC6G6C6TCCT6CC6GCCACCCTCC6G6CCGTT6CTTCACAAC6TTCAAATCC6CTCCC6GC 385 ICCTACTCAGGAGAGCGTTCACCGACAAACAACAACAACAAACGAAAAGGCCCAGTCTTCCGACTGAGCCTTTCGTTTTATTTGATGCCTG 481 577 atagggcgaattgggtaccgggccccccctcgaggtcgacggtatcgataagcttgatatcgaattcctgcagcccgggggatccactagttcta 673 pressure cache cast grass i chagoi i ligi Lecaratilescelaat 769 GCTCTGGCCCGTGTCTCAAAATCTCTGATGTTACATTGCACAAGATAAAATATATCATCATGAACAATAAAACTGTCTGCTTACATAAACAGTAA 865 TACAAGGGGTGTTATGAGCCATATTCAACGGGAAACGTCGAGGCCGCGATTAAATTCCAACATGGATGC 961 1057 1153 1249 STICCISC SCCSSTISCATICS ATTCCTSTITSTAATTSTCCTTTTAACASC SATCSCSTATTCSTCTCSCTCASSCSCAATCACSAA 1345 GSTITESTIGATECGASIGATTTIGATEACGAGCGTAATGSCIGCCIGITGAACAAGTCTGGAAAGAAATGCATAAACTTITGCCATT 1441 CASTCGTCACTCATGGTGATTTCTCACTTGATAACCTTATTTTTGACGAGGGGAAATTAATAGGTTGTATTGATGTTGGACGA 1537 IGCAGACCGATACCAGGATCTTGCCATCCTATGGAACTGCCTCGSTGAGTTTTCTCCTTCATTACAGAAACGG 1533 <mark>FAATCCTGATATGAATAAATTGCAGTTTCATTTGATGCTCGATGAGTTTTTC</mark>TAATCAGAATTGGTTAATTGGTTGTAACACTGGCAGAGCATTAC 1729 SCTGACTTGACG5GACG5C5CAAGCTCAT5ACCAAAATCCCTTAACSTGAGTTACGCGTCGTTCCACT5AGCGTCAGACCCCGTA5/ 1825 1921 SCIACCAACICITIIICCESAEGISACIEGCIICAECAEAECECAEATACIEAEATACIE 2017 SAACTCTSTAGCACCGCCTACATACCTCGCTAATCCTGCTACCASTSGCTGCTGCCAGTGGCGATAAGTCGTGTC 2113 AASACSATAGTTACCGCATAAGSCSCASCSCTCCSCCTGAACGGSCSGTTCSTGCACAGCCCAGCTTGGAGCGAACSACCT 2209 ATACCTACAGCGTGAGCTATGAGAAAGCGCCACGCTTCCCGAAGGGAGAAAGGCGGACAGGTATCCGGTAAGCGGCAGGGTCGGAACAGGAGAGCC 30 4050 60 70 90

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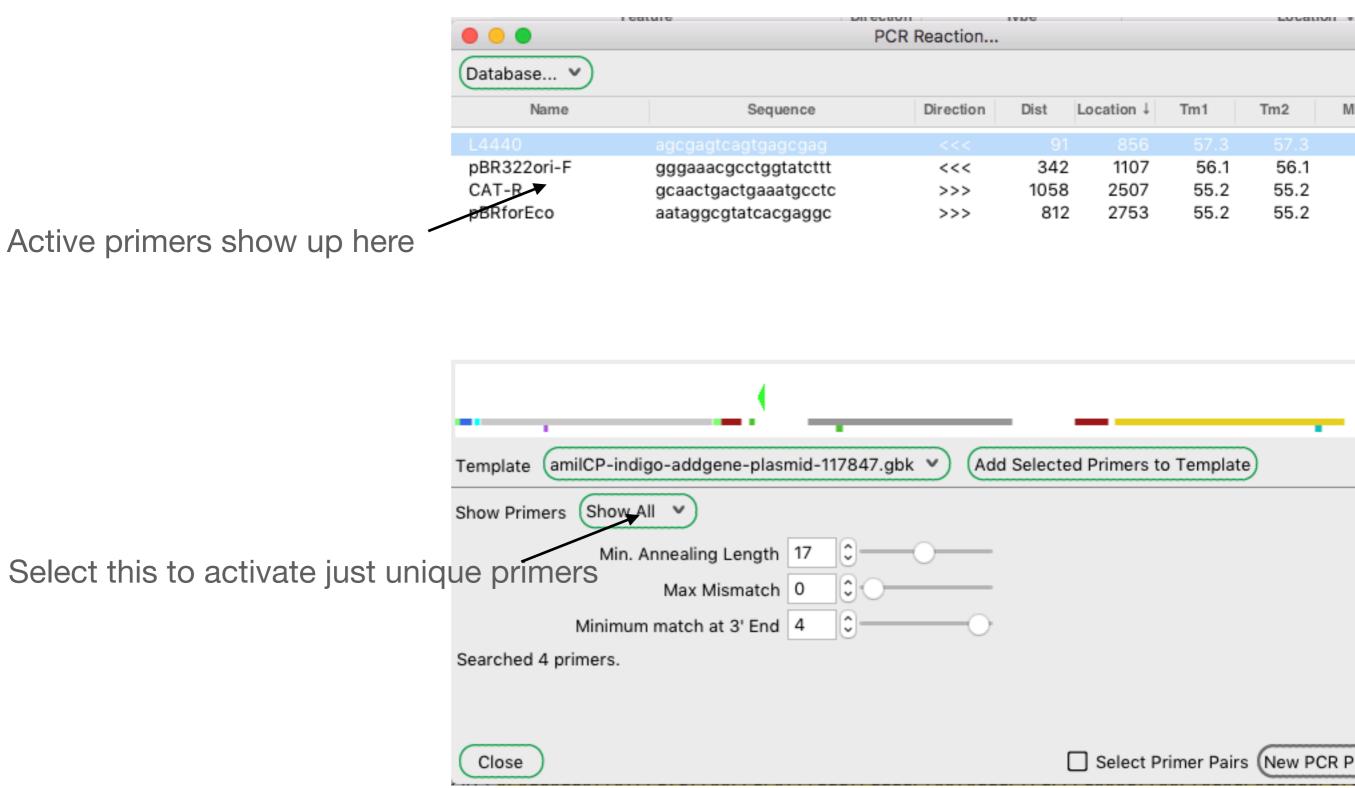
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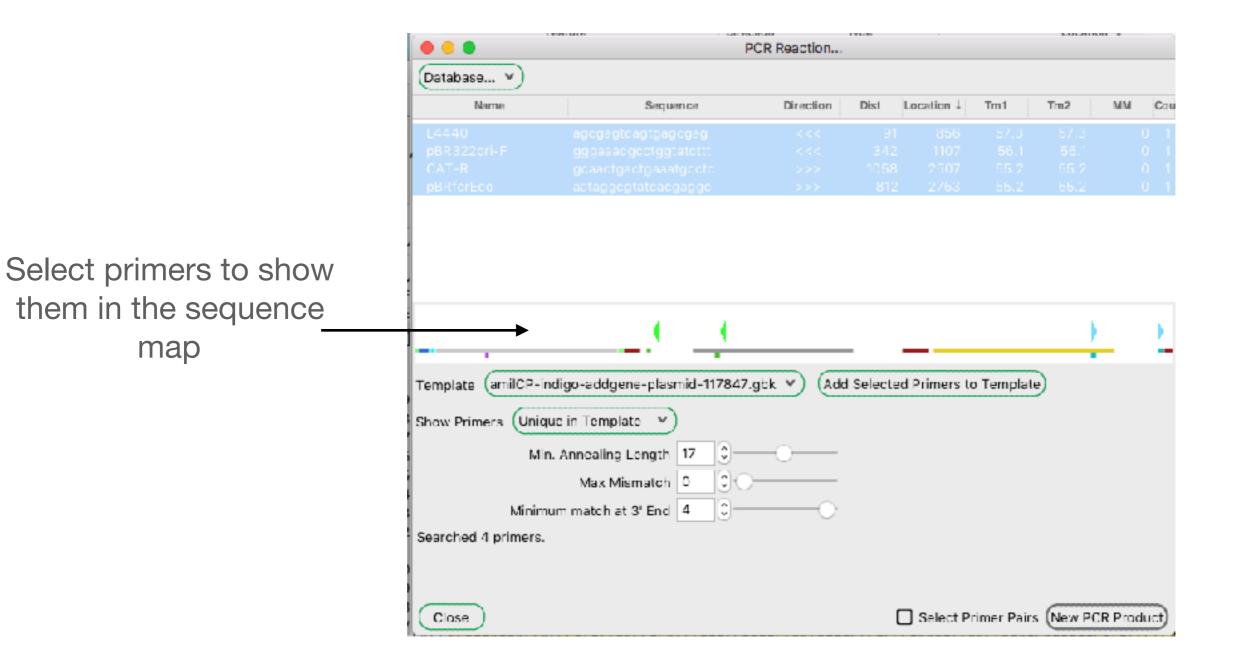
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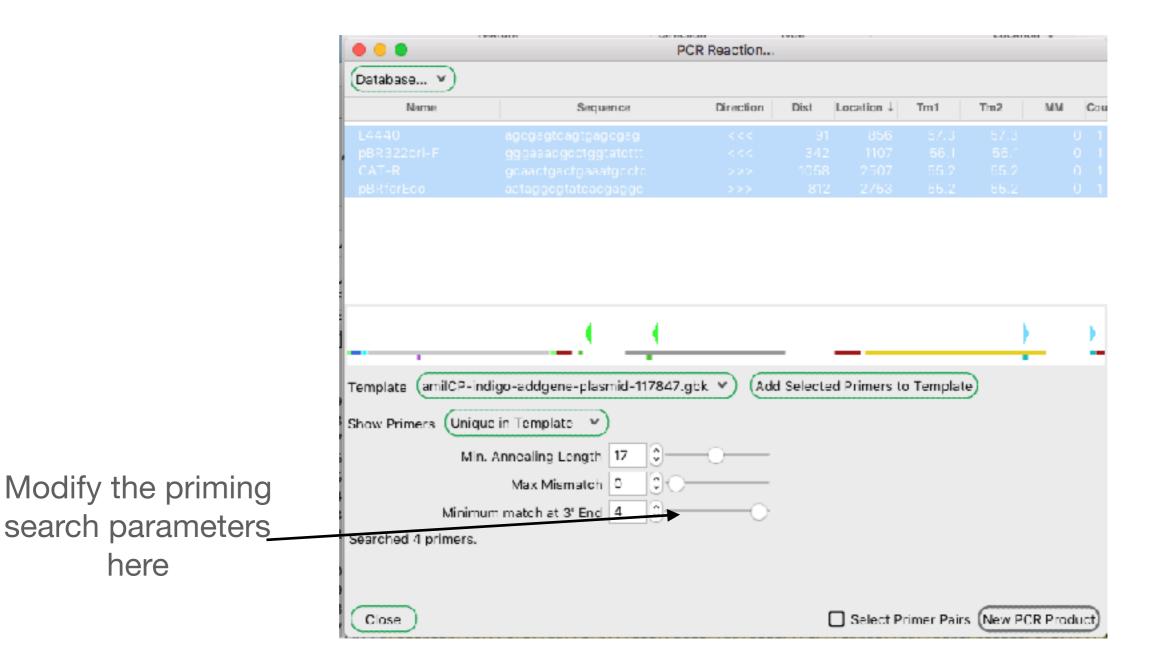
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Select all of the class primers and sequences then copy

	A	В	G	D
1	oMBC1	gaattogoggoog	cttctagag	
2	oMBC2	agcgagtcagtga		
3	aMBC3		tacctaggad	ctgagetag
4	oMBC4		agoggeeget	
5	oMBC5		agaggagaaa	
5	oMBC5		ACCACAGGT	
7	oMBC7	gtgGCTCTTC	gTGGgaatte	sgoggoogo
8	oMBC8	gtgGCTCTTC	gCALGCAAA	radcadage
9	oMBC9		GACGAAGTA	
10	oMBC10	gtgGCTCTTC	gTACctgcag	goggooget
11	oMBC11		GCGTGAATCO	
12	oMBC12	gtgGCTCTTC	GATGGGATA	TTTATCAC
13	oMBC13	gtgGCTCTTC	GATGGGATA!	PITTATCAC
14	oMBC14	gEgectette	gategeatai	TTTATCAC
15	oMBC15	gtgGCTCTTC	gATGGGATA:	TTTATCAC
16	oMBC16	gtgGCTCTTC	GATGGGATA!	PITTATCAC
17	oMBC17	gtgGCTCTTC	gateggata:	TTTATCAC
18	oMBC18	gtgGCTCTTC	gATGGGATA:	TTTATCAC
19	oMBC19	gtgGCTCTTC	GATGGGATA	PITTATCAC
20	aMBC20	gtgGCTCTTC	gateggata:	PTTTATCAC
21	oMBC21	gtgGCTCTTC	GATGGGATA:	TTTATCAC
22	oMBC22	/phos/CTG1	GGTGATAAAA	ATATCCCAA
23	aMBC23	/phos/NNKX	INKTACIGAAC	CATACCAT
24	oMBC24	ggggacaagt	ttgtacaaaa	aaagcaggc
25	oMBC25	ggggaccact	ttgtacaaga	aagetggg
25	aMBC26	actggccgtcgttt	taca	
27	oMBC27	catggtcatagetg	ttteetg	
28	oMBC28	tgtaaaacgacgg	ccagtgaattogog	gccgcttctagaį
29	aMBC29	CAGGGTACTTO	GTGAATGGTA	IGCT
30	oMBC30	AGCATACCATT	CACCAAGTACC	CTG
31	oMBC31	caggaaacagcta	tgaccatgctgcag	cggccgctacta
32	aMBC32	caggaaacagcta	tgaccatg	
33	oMBC33	tgtasaacgacgg	ccagt	
34	oMBC22	CTGTGGTGAT	AAAATATCCC	CAAGCAAAT
35	oMBC23	NNKNNKTACO	GAAGCATAC	CATTCACCA
38				
37				

Primers can be:

sequence name (tab) sequence name (tab) sequence (tab) note feature library format

		Р	CR Reaction						
Database									
Add Primers from Clipboard	æ٧		Direction	Dist	Location	Tm1	Tm2	MM	Co
Add Primers from File	жo	c	>>>	1058	2507	55.2	55.2		0 1
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Copy Selected Primers	жc	:	<<<	342	1107	56.1	56.1		01
Save Selected Primers	жs	jc	>>>	812	2753	55.2	55.2	,	01
Select All	₩A	1							
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Template amilCP-indigo-addgene	-plasmid-1	17847.	gbk 🗸 🗛	d Selecte	d Primers t	o Templat	e		
Show Primers Unique in Template	v)								
Min. Annealing Le	ngth 17	0 —	-0						
Max Mism	atch 0								
Minimum match at 3'		1 0-							
Forward primer: pBRforEco Reverse primer: L4440									
Close					🖌 Select P	rimer Pair:	s New PC	R Prod	uct

	PC	R Reaction	1					
Database 🗸								
Name ↓	Sequence	Direction	Dist	Location	Tm1	Tm2	MM	Соц
CAT-R	gcaactgactgaaatgcctc	>>>	1058	2507	55.2	55.2	0	1
L4440	agcgagtcagtgagcgag	<<<	91	856	57.3	57.3	0	1
oMBC1	gaattcgcggccgcttctagag	>>>	741	24	61.4	61.4	0	1
oMBC2	agcgagtcagtgagcgaggaagc	<<<	86	851	64.4	64.4	0	1
oMBC3	gctagcattgtacctaggactgagctag	<<<	2066	31	61.4	61.4	0	1
oMBC4	GCTtactagtagcggccgctgcag	>>>	2790	775	62.4	65.0	0	1
oMBC5	tactagagaaagaggagaaatactag/	>>>	671	94	60.1	60.1	0	1
oMBC6	ttaTTAGGCGACCACAGGTTTG	<<<	2765	730	61.3	61.3	0	1
oMBC7	gtgGCTCTTCgTGGgaattcgcgg	>>>	741	24	65.1	73.0	0	1
oMBC8	gtgGCTCTTCgCAtGCAAATGG	<<<	2261	226	56.2	69.8	0) 1
emplate amilCF	-indigo-addgene-plasmid-117847.g	bk 🗸 (A	dd Selec	ted Primer	s to Temp	late		
how Primers	now All 💙							
	Min. Annealing Length 17	0	_					
	Max Mismatch 0		_					
Mir	imum match at 3' End 4)					
orward primer: p everse primer: L								
Close				Select	Primer P	airs (New	PCP Pro	du

PCR Reaction... Database... 🗸 Name ↓ Direction Dist Location Tm₂ MM Sequence Tm1 Cou oMBC16 gtgGCTCTTCgATGGGATATTT 0 oMBC17 gtgGCTCTTCgATGGGATATTT 0 oMBC18 gtgGCTCTTCgATGGGATATTT 0 oMBC19 gtgGCTCTTCgATGGGATATTT 0 Select oMBC22 and oMBC20 gtgGCTCTTCgATGGGATATTT 0 23 primers. gtgGCTCTTCgATGGGATATTT oMBC21 0 oMBC22 CTGTGGTGATAAAATATCCC 2270 60.3 60.3 235 0 1 <<< These are the around NNKNNKTACGGAAGCATAC(65.2 oMBC24 ggggacaagtttgtacaaaaaagcag 75.3 0 741 24 1 >>> the horn mutagenic oMBC25 ggggaccactttgtacaagaaagctgg 852 62.4 75.6 0 87 1 <<< primers. (amilCP-indigo-addgene-plasmid-117847.gbk ♥ Add Selected Primers to Template Template Show Primers Show All 🛛 🗸 0 Min. Annealing Length 17 0 Max Mismatch 0 \$ Minimum match at 3' End 4 Forward primer: oMBC23 Reverse primer: oMBC22 Select Primer Pairs (New PCR Product) Close

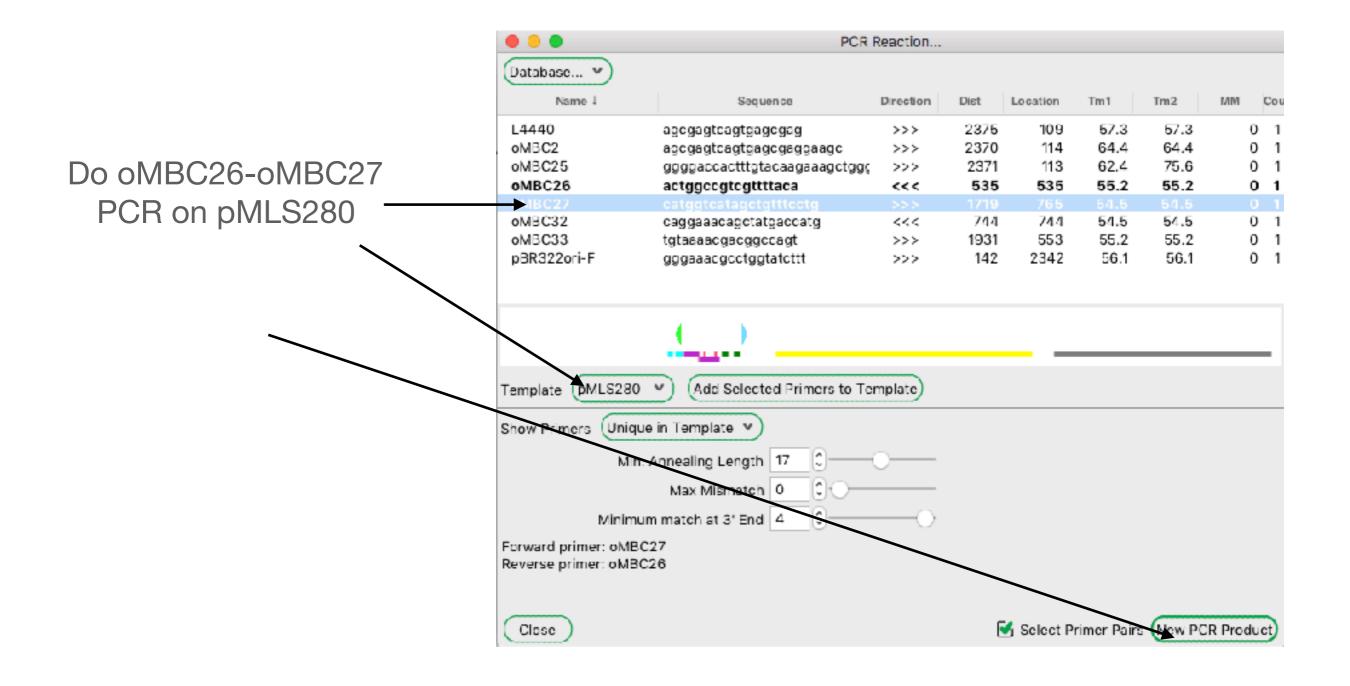
		PC	R Reaction					ATTUN -	
	Database 💙								
	Name ↓	Sequence	Direction	Dist	Location	Tm1	Tm2	MM	Cou
	oMBC16	gtgGCTCTTCgATGGGATATTT							0
	oMBC17	gtgGCTCTTCgATGGGATATTT							0
	oMBC18	gtgGCTCTTCgATGGGATATTT							0
	oMBC19	gtgGCTCTTCgATGGGATATTT							0
	oMBC20	gtgGCTCTTCgATGGGATATTT							0
	oMBC21	gtgGCTCTTCgATGGGATATTT							0
	oMBC22	CTGTGGTGATAAAATATCCC	<<<	2270	235	60.3	60.3		01
	oMBC23			463	302	64.9	65.2		0 1
	oMBC24	ggggacaagtttgtacaaaaaagcag		741	24	65.2	75.3	(01
	oMBC25	ggggaccactttgtacaagaaagctgg		87	852	62.4	75.6	(0 1
	Template amilC	P-indigo-addgene-plasmid-117847.g	bk 💙 (4	Add Selec	ted Primers	s to Temp	ate	•	I
	Template amilC	P-indigo-addgene-plasmid-117847.g	bk 🔻 🦨	Add Selec	ted Primer:	s to Temp	ate	•	
	Template amilC		bk 💙 (4	Add Selec	ted Primer:	s to Temp	ate	•	-
	Template amilC	Min. Annealing Length 17 0 Max Mismatch 0	bk 💙 (4	Add Selec	ted Primer:	s to Temp	ate	•	
	Template amilC Show Primers	Min. Annealing Length 17	bk 💙 (4	Add Selec	ted Primer:	s to Temp	ate	•	
	Template amilC Show Primers	Nin. Annealing Length 17 Max Mismatch 0 nimum match at 3' End 4	bk 💙 (4	Add Selec	ted Primer	s to Temp	ate		
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	Template amilC Show Primers S Mi Forward primer: o Reverse primer: o	Nin. Annealing Length 17 Max Mismatch 0 nimum match at 3' End 4	bk 💙 🥢	-	ted Primer			PCR Pr	oduc
is to do a	Template amilC Show Primers S Mi Forward primer: o Reverse primer: o	Nin. Annealing Length 17 Max Mismatch 0 nimum match at 3' End 4	bk V	-				PCR Pr	oduc
is to do a { reaction	Template amilC Show Primers S Mi Forward primer: o Reverse primer: o	Nin. Annealing Length 17 Max Mismatch 0 nimum match at 3' End 4	bk V (-				PCR Pr	oduc

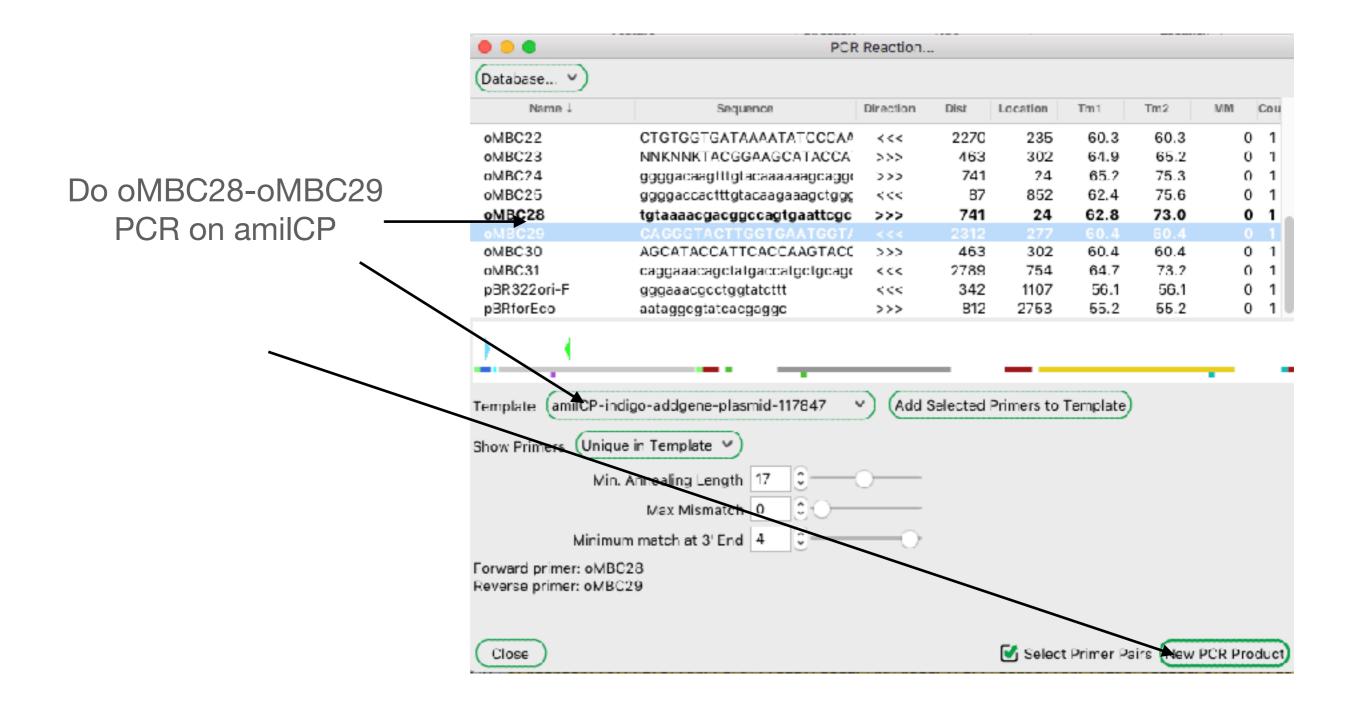
In primer pair mode, only one forward and one reverse primer can be selected. Selected primers are in bold.

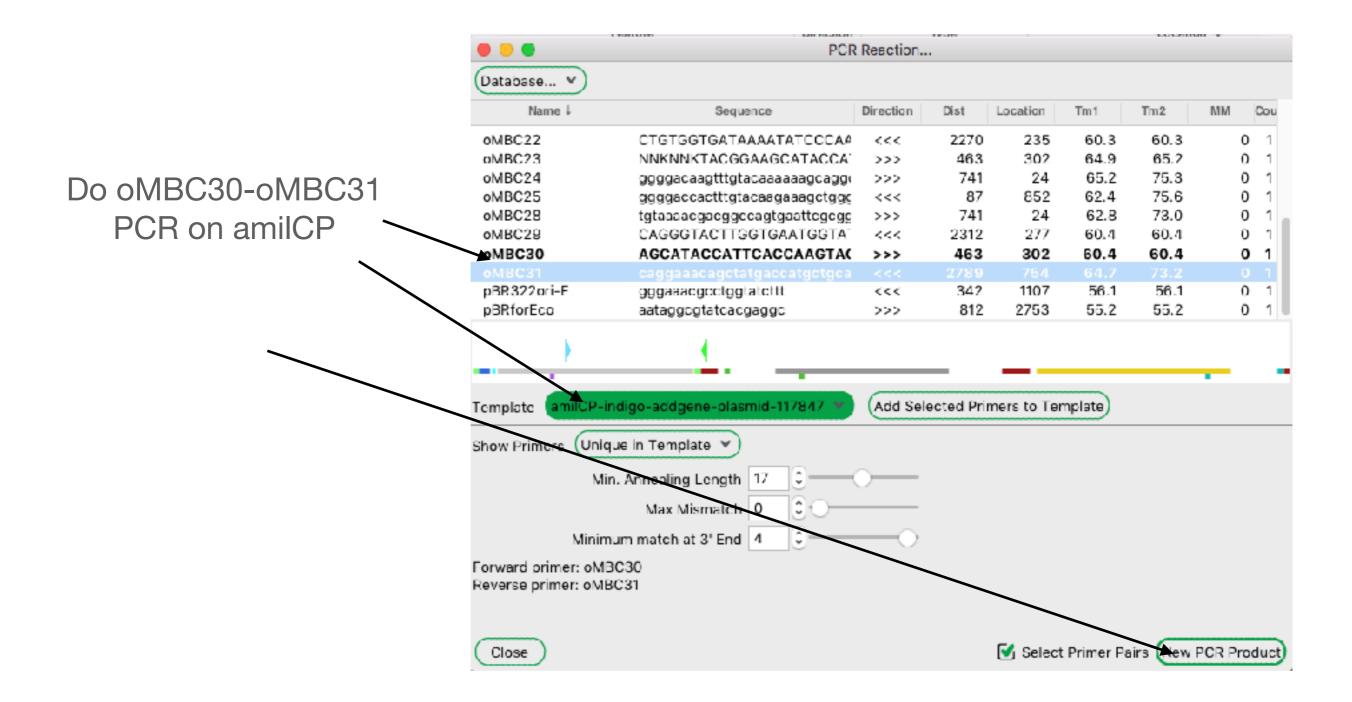
> New PCR will become active when a pair of primers is selected.

• • •	PC	R Reaction	ı						
Database 💙									
Name ↓	Sequence	Direction	Dist	Location	Tm1	Tm2	MM	0	Cou
oMBC16 oMBC17 oMBC18 oMBC19 oMBC20 MBC21	gtgGCTCTTCgATGGGATATTT gtgGCTCTTCgATGGGATATTT gtgGCTCTTCgATGGGATATTT gtgGCTCTTCgATGGGATATTT gtgGCTCTTCgATGGGATATTT gtgGCTCTTCgATGGGATATTT								0 0 0 0 0 0
oMBC22	CTGTGGTGATAAAATATCCC	<<<	2270	235	60.3	60.3		0	1
oMBC23	NNKNNKTACGGAAGCATAC(463	302	64.9	65.2		0	
oMBC24	ggggacaagtttgtacaaaaaagcag	>>>	741	24	65.2	75.3		0	1
oMBC25	ggggaccactttgtacaagaaagctgg	<<<	87	852	62.4	75.6		0	1
	P-indigo-addgene-plasmid-117847.gl	bk 🔻 🥢	Add Selec	ted Primers	s to Temp	late			
	Min. Annealing Length 17	-0	_						
	Max Mismatch 0		_						
Mir	nimum match at 3' End 4)						
Forward primer: o Reverse primer: o									
Close				Select	Primer Pa	irs New	PCR Pr	od	lue

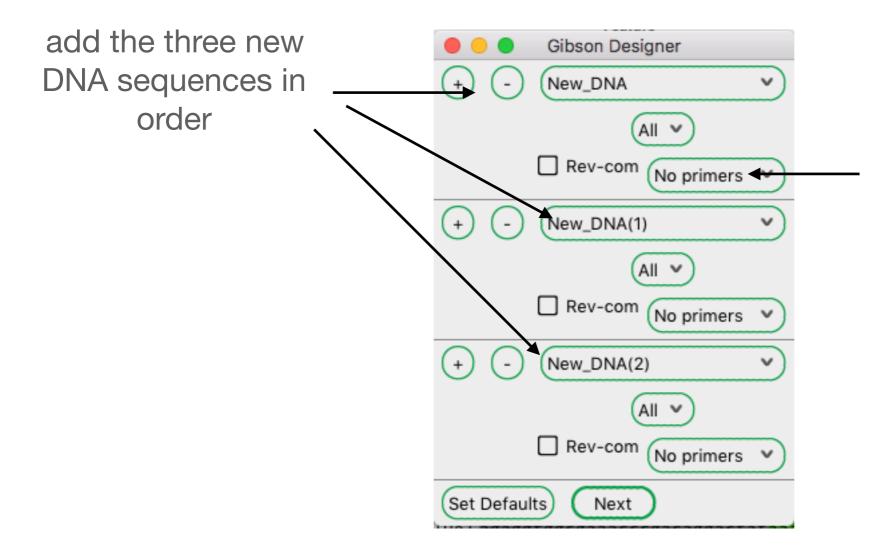
							N	ew_DNA										
3 8 E) 🔀	6(ή γ	√ ⊌a	28	8 📀	D T	6 🖬	-	٩	٢	=					
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		Feature				Direction	n	Тур	0				1	Locatio	n↓			
Hidden																		
oMBC23						>>>		primer,	bind					137	7			
chr CQYG	amilCP					>>>		misc_fe						812	2			
amilCP Ba	se					>>>		misc_fe						848	6			
BioBrick s	uffix					>>>		misc_fe	alure					4905	10			
TT.								oMBC:	23									
···· *	10	*	20	-	30	* 4	10	* 50		60	-	70		*	30	*	90	
						ACCCTGA/												
						CTCTGAAC TATGTTGA												
857 AAASA 446 AGTGT 535 BCCBC 524 BBCCB 713 DDDGB 802 CCBBC 891 LLCCB 980 BGTTC	AGCCTGT GAAATTT cotgoog cggcgag ccagcaa gctcaag accotgo ggtgtag	GAAGA CCATT CCATT CggLa CggLa CggLa CggLa CggLa CggLa CggLa CggLa	TGCCAG GCACGG CTTTAP (Cage) Cages access taccess	SGGTA CAAAC Loact Loact Cgaaa atacc ccaag	ATCACI CCTGTC CCTGTC Caaaa Laaaaa Laaaaa accega Lgtcc CCTggg	TATGTT5/ SGTCGCC1 agattact ggcgglaa Aggccgcg acaggact gcclllo gctgtgtgtg	ACCGCA IAAtaa Itogog atacgg atacgg sttgot tat <mark>aaa</mark> stocol goacga	AACTGG tactag ttatgc llatcc ggcgtt gatacc lcggga accccc	ATGTAA tagcgg aggctt acagaa tttcca agcgtg cgttca	CCAAT cogot cotog coggo	CACA gcag ctca ggal tccg cctg LLCL accg	ACAAG CCCCCC aacgo cccccc gaago calag cfgcg	GATI Coast C	TACAC about ab	TTCG ggca ctcg acal catc gcgc glag ggta	GTTG aggt; gtcg gLga; acaa; tctc; gLat; acta;	AGC gtc gca aaa ctg ctc tcg	
57 AAAGA 46 AGTGT 35 Eccac 524 BBC(B 13 DDDGG 14 DDDGG 14 DDDGG 14 DDDGG 14 DDDGG 14 DDDGG 14 DDDGG 14 DDDGG 14 DDGG 14 DDGGG 14 DDGGG 14 DDGGG 14 DDGGG 14 DDGGG 14 DDGGG 14 DDGGG 14 DDGGG 14 DDGGG 14 DDGGGG 14 DDGGGGG 14 DDGGGGGGGGGGGGGGGGGGGGGGGGGGGGGGGGGGG	AGCCTGT GAAATTT Cotgood Cogoogoag Coogoag Coogoag gotcaag accolgo ggtgtag agtocaa gagtcoa	GAAG/ CCATT CCATT CCATT CCATT CCATC CCCAT CCCAT CCCAT CCCAT CCCAT CCCAT CCCAT CCCAT CCCAT CCCAT CCCAT CCCATT	ATGCCAG GCACGG CTTTA COGGOC COGOC COGGOC COC C	SGGTA CAAAC Loact Locot Cgaaa Lacc ccaag cacga goota	ATCACI CCTGTC Igaaaa Loaaaa accoga accoga Lguco gotggg acttat acctao	TATGTTS/ GGTCGCCT agattact ggcgglaa AggcCgCg acaggact cgcclllc gctgtgtg tcgccact	ACCGCA IAAtaa Itegeg stacgg sttget stataaa steeel staega stggcag actoga	AACTGG tactag ttatgc llatcc ggcgtt gatacc lcggga accccc cagcca agaaca	ATGTAA tagcgg aggctt acagaa tttcca aggcgt agcgtg cgttca ctggta gtattt	CCAAT ccgct rtrg lcagg coggc ttccc gcggc gcggc gcggt ggtot	CACA gcag gctca ggal tccg cctg cctg tcctg actg actg	ACAAG teegg tegg aacgo coccoo gaago catago catago gcaga gcaga	GATI Caaga Cagga Cotga C	TACAC aboog ctgrg abog abog ctcgt acgcl tatrc aggta gobgc	TTCG ggca ctcg acal catc; gcgc gcgc ggta; ggta; tgta; cagt	GTTG aggt gtcg gtcg gtga gtcta gtata gtata gca tacc	AGC gtc gca aaaa ctg clc tcg gtg gtg ttc	
57 AAASA 46 ASTST 535 BCCBC 524 BBCLB 524 BBCLB 524 BBCLB 524 BBCLB 525 BCCB 525 BCCB 525 CCBCB 525 CCCB 525 CC	AGCCTGT GAAATTT Cotgoog Coggogag Coogoaa gotcaag accolgo ggtgtag agtocaa gagtoca gagtoca	GAAG/ CCATT CCQg La aaggo tcaga cgc Ll gt cgt cgc gt cgc g	TGCCAG GCACGG CTTTAF (Cage) Cages access taccess taccess taccess taccess taccess taccess taccess taccess taccess taccess	SGGTA CAAAC Loact Loact Sgaaa Lacc Coaag Lacc Coaag Lacc Scaag	ATCACI CCTGTC Igaaaa Lcaaaa Lcaaaa accoga LgLcc gctggg acttat acctac gctac	TATGTTS/ SGTCGCC1 agattact ggcgglaa Aggccgcg acaggact gccllla gctgtgtgtg tcgccact cggctacc aacaaard	ACCSCA IAAtaa Itogog atacgg atacgg atacgg atacgg atacgg atacgg accel aco	AACTGG tactag ttatgc ggcgtt gatacc Loggga accccc cagcca agaaca tggtag	ATGTAA tagcgg aggctt acagaa tttcca agcgtg cgttca ctggta gtattt cggtgg	CCAAT ccgct ccgct ccgct caggo caggo ttocc gcgct gcgct gcgct cggtat	CACA gcag ctca ggal tccg cctg cctg accg accg atta ctgc ttgt	ACAAG toogg ctgac aacgo coccoo gaago calag ctgcaga gotot ttgca	GATI Content agga Content Cont	TACAC DODOG ot gog asga acgog ctogt acgot tatoc aggta googo agong	TTCG ggca ctcg acal catc gcgc gcgc ggta ggta tgta tgta tgta	GTTG/ aggt) gtcg gtgaj acaa toto gtato gtato acta gccg gcg/ gcg/ gcg/ gcg/ gcg/ gcg/ gcg	AGC gtc ttc gca aaaa ctg ctc tcg gtg ttc cag	
357 AAASA 146 AGTGT 358 ACCAC 359 ACCAC 351 ACCAC 352 BECLE 352 ECCAC 353 ACCAC 354 BECLE 352 ECCAC 360 ECCAC 360 AGTTC 360 AGTTC 360 AGTTC 360 ECCAC 376 ECCAC	AGCCTGT GAAATTT Cotgoogag Cogogag Cogogag gotcaag accolgo gotcaag agtocaa gagtoca gagtoca aagagttot aaggalo	GAAG/ CCATT ttttt cggta aaggo tcagat gtcgt cgctl cccgg tgaag ggtag	ATGCCAG TGCACGG TCCAGGO ACCAGGO AGREG	SGGTA CAAAC Loact Loact Scogt Sgaaa alaco coag alaco coag acacga gotta gatco colll	ATCACI CCTGTC Igaaaa Laaaaa accega Lgtcc 2CTgg acttat acttat acttat Iggtcl	TATGTT5/ GGTCGCC1 ggcgglaa ggcgglaa ggccgcg acaggact gcclllc gctgtgtg tcgccact ggctacc aacaaacc	ACCGCA IAAtaa Itegeg ataegg ataegg ataegg ataegg acteel acteel acteel acteel actege actege actegel	AACTGG tactag ttatgc ggcgtt gatacc loggga accccc cagcca agaaca tggtag clgacg	ATGTAA tagcgg aggett acagaa tttcca aggcgtg agcgtg cgttca ctggta gtattt cgg†gg clcagt	CCAAT retege loage ttoo geoge geoge geoge geoge geoge ttoo geoge geoge ttoo geoge geoge ttoo	CACA gcag ctca ggal tccg cctg cctg accg atta ctgc ttgt	ACAAG CCCCC CCCCCC CCCCCC CCCCCCC CCCCCC	GATI Coast C	TACAC DODOG CTORE ABB	TTCG gca ctcg acal catc gcgc gcac glag ggta tgta cagt atta gall	GTTG/ aggt glgag dcaa tctco glato glato acta gCG/ tacc cgcg/ llgg	AGC gtc gca aaa ctg ctc tcg gtg ttc cag Lca	
357 AAASA 146 AGTGT 535 AFFAC 524 BBCLB 525 LCCBB 526 ABASA 527 LCCBB 528 LCCCD 529 LCCCD 540 ABTTC 558 CCDCD 547 SBAAA 548 LCCD 549 LCCD 540 ABTTC	AGCCTGT GAAATTT Cotgood Cogogogag Coogoaa gotcaag accolgo gotcaag agtocaa gagtocaa gagtocaa gagtocaa gagtocaa gagtocaa gagtocaa	GAAG/ CCATT CCATT CCATT CCAT CCAT CCAT CCAT	TGCCAG GCACGG CTTTAR (LCago) Coggos (accggos (accggos (tcgctg) (tc	SGGTA CAAAC Loact Loact Caaaa Laco Caaaa Cacga Gata Gata Cacct Cacct	ATCACI (CTGTC (gaaa) (Laaa) (Laaa) (Lgaaa) (Lgaaa) (Lgaaa) (Lgaaa) (Lgaaa) (Lgaaa) (Lgaaa) (Lgaaa) (Lgaaa)	TATGTTS/ GGTCGCCT agattact ggcgglaa Aggccgcg acaggact cgccllc gctgtgtg tcgccact cggctact acaaac Lllclac	ACCGCA IAAtaa Itogog atacgg sttgot tataaa itocol scarga actoga actoga actoga actoga actoga actoga	AACTGG tactag ttatgc llatcc ggcgtt gatacc lcggga accccc cagcca agaaca tggtag clgacg aaatga	ATGTAA tagcgg aggctt acagaa tttcca agcgtg cgttca ctggta gtattt cggtgg cgtttt	CCAAT ccgct rtrg lcagg ccggc ttccc gcccg gcccg gcccg gcccg gcccg gcccg gcccg gcccg gcccg gcccg gcccg gcccg gcccg ttttt tttt tttt	CACA gcag ctca ggal tccg cctg; tcctg; accg accg acta; ctgc; ttgt gaaa; aatc	ACAAG toogg taacgo coccco gaago catag ctgcaga gctot ttgca actca taaag	GATI COM Cagga Cong Cong Cong Cong Cong Cong Cong Cong	TACAC aboog ctgrg abogog ctcgt acgcl tatrc aggta gaogc agrag labgg atotg	TTCG ggco ctcg; acal; catc; gcgc; glag; ggta; tgta; cagt; atta; gall agta;	GTTG/ aggt; gtcg; gtga; acaa; tete; gtat; acta; gcg; tacc; cgcg; tacc; cgcg; tacc;	AGC gtc gca aaaa ctg clc tcg gtg ttc cag Lca tgg	
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357 AAASA 446 AGTGT 535 ACCAC 624 BBCLB 624 BBCLB 624 BBCLB 624 BBCLB 624 BBCLB 802 tcgac 801 LLCCB 980 Agttc 980	AGCCTGT GAAATTT Cotgocgag Coagcaa gotcaag accolgo gotcaag agtocaa gagtoca gagtoca gagtoca gagtoca cagotog ctggato cagotog ctggato cattotg lgoccat	GAAG/ CCATT CCATT Cgg La aaggo tcaga cgc Ll gt cgt cgg tag cgg tag cccgg tgaag tgaag aaaggo ttaag aaaggo ttaag aaaggo tatca gg La aaaggo tagaag aaaggo taag taag	ATGCCAG GCACGG CTTTAF ALCABCI COGGOS AGREGOS A	SGGTA CAAAC Loact Loact Loact Cgaaa alacc cacga gata cacga gata cacct totca agto agto agto agco agto agto agco agco agco agto agco agco agco agco agco agco agco agc	ATCACI CCTGTC Caaaaa Laaaaa Laaaaa acccga Lgtcc Cfgg acttat Dactaca Cggcaa Lggtcl Lagato Caaacaa Lggtcl Lagato Caaacaa Lggtcl Lagato Caaaaa Lggtcl Lagato Caaaaaaaaaa Ccaaaaaaaaaaaaaaaaaaaaaaaa	TATGTTS/ SGTCGCC1 agattact ggcgglaa Aggccgcg acaggact ggclltc gctgtgtgt tcgccact tggctacc aacaaac Llllclac ccttttbc gagctcga aacggca aagllgtg	ACCSCA IAAtaa Itogog ataogg ataogg ataogg ataogg actoga ac	AACTGG tactag ttatgc ggcgtt gatacc lcggga accccc cagcca agaaca tggtag clgacg aattac aactg lggcca gccagg	ATGTAA tagcgg aggett acagaa ttteca agcgtg cgttea ctggta gtattt cggtgg cleagt cleagt accgag gccccg aatcgc cgttta ttttca	CCAAT ccgct r treg lcagge ttccc gcgct gcgct gcgct ggtat ttfff ggaac cage gctt ccgta ccgta ccgta ccgta ccgta ccgta ccgca cc	CACA gcag ctca ggal tccg cctg cctg accg accg ctgc ttgt gaaa ccac gcat accac gcat	ACAAG CCCCC CCCCCC Gaago CCCCCC Gaago CCCCCC Gaago CCCCCC Gaago CCCCCCC Gaago CCCCCCC CCCCCCCCCCCCCCCCCCCCCCCCCCC	GATI Coase Coa	TACAC DODDE ct grg aaaga acgag ctcgt acgcl tatro aggta gabge aggtag aaggta tatgg aggtagt tgtcg ctcac tgtcgc	TTCG ggca ctcg acal catc gcgc gcgc ggta tgta ggta tgta ggta tgta gtt gall agtt gall catt gall agtt gall agta agtt gala gala	GTTG/ aggt; glga; glga; acta; glat; glat; acta; glat; acta; glat; acta; glat; acta; glat; acta; glat; acta; glgat; glgat; glgat; glgat; tatg;	AGC gtc gca aaa ctg ctc tcg gtg ttc tca tgg ttca tca tgg ttca tgg ttca tgg ttca	
357 AAASA 446 AGTGT 535 ACCAC 524 BBCLB 502 tcgac 801 LLCCB 920 BCTGB 921 LLCCB 920 BCTGB 921 LLCCB 922 TggBB 924 tctgB 925 ttaB 926 ttaB 927 ttaB 928 ttaB 929 ttaB 921 ttaB 922 ttaB 921 ttaB 922 ttaB 921 ttaB 922 ttaB 924 ttaB	AGCCTGT GAAATTT Cotgocgag Coagcaa gotcaag accolgo gotcaag accolgo gotcaag agtocaa gagtoca gagtoca aagagtot tatocaa cagotog ctggato cattotg Lgoccat acgaaaa	GAAG/ CCATT CCATT Cgg La aaggo tcaga cgc Ll gt cgt cgg tag cgg tag cccgg tgaag tgaag aaaggo ttaag aaaggo ttaag aaaggo tatca gg La aaaggo tagaag aaaggo taag taag	ATGCCAG GCACGG CTTTAF ALCABCI COGGOS AGREGOS A	SGGTA CAAAC Loact Loact Loact Cgaaa alacc cacga gata cacga gata cacca gata gat	ATCACI CCTGTC Caaaaa Laaaaa Laaaaa acccga Lgtcc Cfgg acttat Dactaca Cggcaa Lggtcl Lagato Caaacaa Lggtcl Lagato Caaacaa Lggtcl Lagato Caaaaa Lggtcl Lagato Caaaaaaaaaa Ccaaaaaaaaaaaaaaaaaaaaaaaa	TATGTTS/ SGTCGCC1 agattact ggcgglaa Aggccgcg acaggact ggclltc gctgtgtgt tcgccact tggctacc aacaaac Llllclac ccttttbc gagctcga aacggca aagllgtg	ACCSCA IAAtaa Itogog ataogg ataogg ataogg ataogg actoga ac	AACTGG tactag ttatgc ggcgtt gatacc lcggga accccc cagcca agaaca tggtag clgacg aattac aactg lggcca gccagg	ATGTAA tagcgg aggett acagaa ttteca agcgtg cgttea ctggta gtattt cggtgg cleagt cleagt accgag gccccg aatcgc cgttta ttttca	CCAAT ccgct r treg lcagge ttccc gcgct gcgct gcgct ggtat ttfff ggaac cage gctt ccgta ccgta ccgta ccgta ccgta ccgta ccgca cc	CACA gcag ctca ggal tccg cctg cctg accg accg ctgc ttgt gaaa ccac gcat accac gcat	ACAAG CCCCC CCCCCC Gaago CCCCCC Gaago CCCCCC Gaago CCCCCC Gaago CCCCCCC Gaago CCCCCCC CCCCCCCCCCCCCCCCCCCCCCCCCCC	GATI Coase Coa	TACAC DODDE ct grg aaaga acgag ctcgt acgcl tatro aggta gabge aggtag aaggta tatgg aggtagt tgtcg ctcac tgtcgc	TTCG ggca ctcg acal catc gcgc gcgc ggta tgta ggta tgta ggta tgta gtt gall agtt gall catt gall agtt gall agta agtt gala gall	GTTG/ aggt; glga; glga; acta; glat; glat; acta; glat; acta; glat; acta; glat; acta; glat; acta; glat; acta; glgat; glgat; glgat; glgat; tatg;	AGC gtc gca aaa ctg ctc tcg gtg ttc tca tgg ttca tca tgg ttca tgg ttca tgg ttca	
357 AAASA 446 AGTGT 535 AFFAF 535 AFFAF 534 BBCLB 524 BBCLB 524 BBCLB 524 BBCLB 524 BBCLB 524 BBCLB 502 tcgac 801 Ltccg 980 Agttc 981 Agttc 982 Agttc 982 Agttc 982	AGCCTGT GAAATTT Cotgood Cogogag Coogoaa gotcaag accolgo gotcaag agtocaa gagtocaa gagtocaa gagtocaa gagtoca cagato cagotog cotgoaaa ctgoaaaa ctgoaaaa	GAAG/ CCATT TTTTT cggta aaggo tcaga cgctl gtcgt gtcgt ggtag cggtag aaaggo tatca ggtag aaaggo ccgao ggtag aaato	ATGCCAG GCACGG CTTTAR (LCago) COGGO CAGO CA	SGGTA CAAAC LCaCl LCaCl LCaCl Caccat Cacaa Cacga	ATCACI CCTGTC Igaaaa Lcaaag Lcaaag Lcaaag Lcaaag Code	TATGTTS/ GGTCGCCT agattact ggcgglaa Aggccgcg acaggact ggccllt ggctlt ggctact acaaaaa Llltclaa cottttac aaaaaaaa gagctcga aaacggca aaacggca	ACCGCA IAAtaa Itogog atacgg atacgg atacgg atacaa Itocol Isarga Itocol Isarga Isagoag Isarga Isagoag Isago	AACTGG tactag ttatgc llatcc ggcgtt gatacc lcggga accccc cagcca agaaca tggtag clgacg aaattac aacctg lggcca gccagg aaacgt	ATGTAA tagcgg aggctt acagaa tttcca aggcgt agcgtg cgttca ctggta gtattt cggtgg clcagt agtttt accgag gccccg aatcgc cgllla ttttcagt	CCAAT CCGCC CCGCC CCGCC CCCCC CCCCCCCCCC	CACA gcag gcag gctca ggal tccg tccg actg actg actg atta gaaa ctgaa gcat aaac acac gcat acac gcat	ACAAG togg trgac aacgo cocco gaago catago catago catago gcaga gcaga toato cagca tggts gccao gcaaa	GATI Coase Coa	TACAC DODOS CT SCS ABBS ABSS ACSOS ACSOS ASSA	TTCG ggca ctcg; acal; catc; gcgc; gcgc; glag; gtgta; cagt; atta; atta; gall gttg gttg ctt; cag; catc; gcgc; cag; atta; gall; gtta; catc; gcgc; catc; ca	GTTG/ aggt; gtcg; gtga; acaa; toto; gtat; acta; gcgc; tacc; cgcg; tacc; ctga; tact; ggat; tatg; ggat;	AGC gtc gca aaaa ctg ctc tcg gtg ttc cag tca tca tca tca tca tca tca tca tca tca	



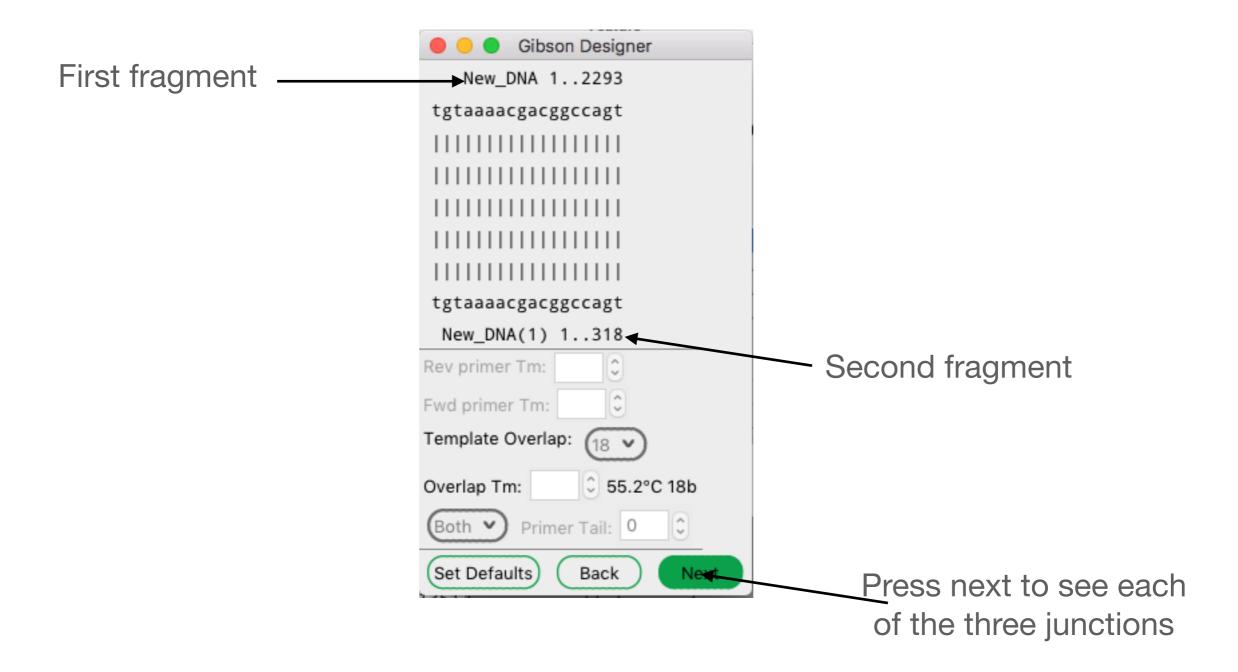


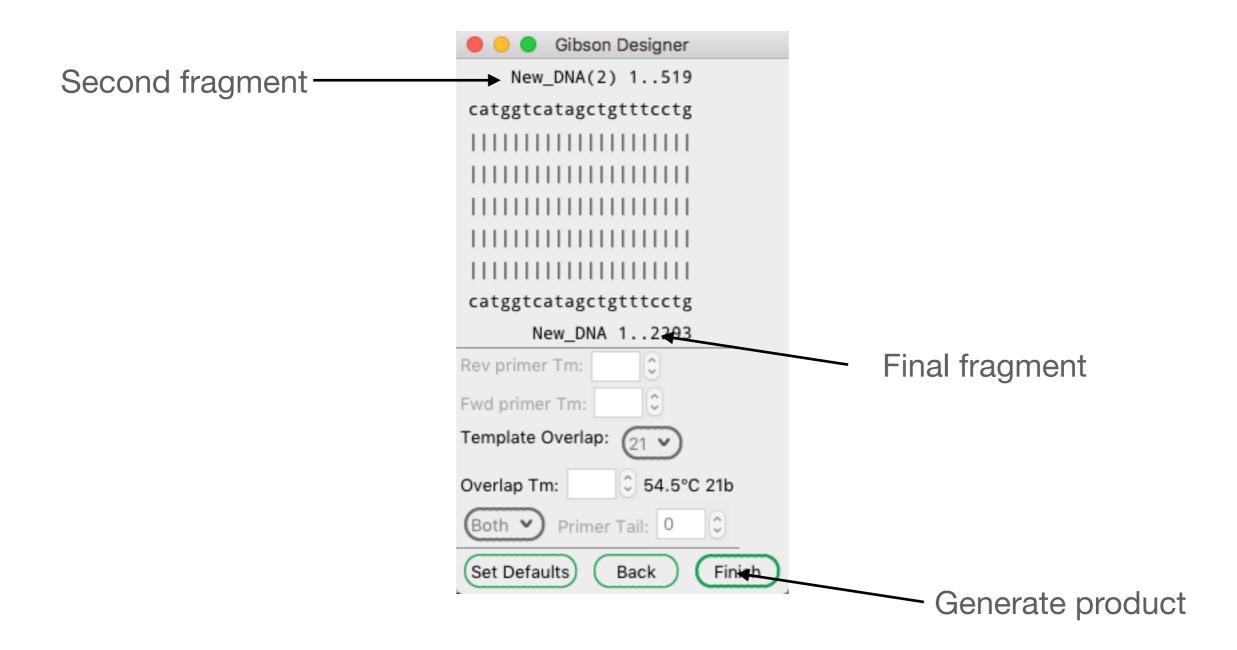


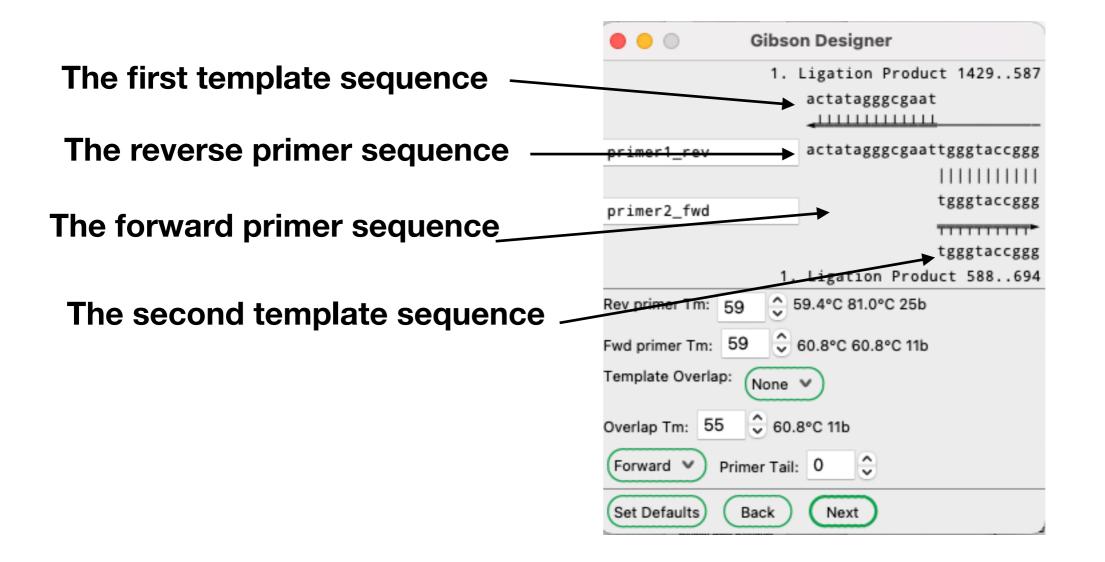
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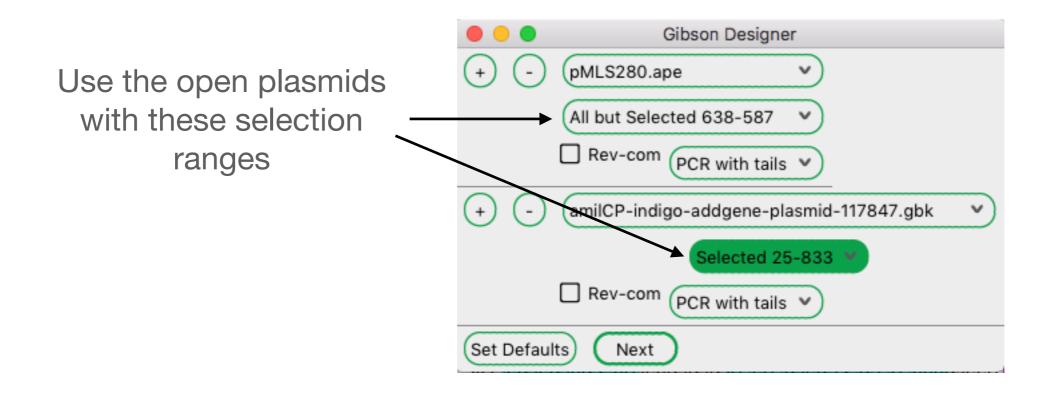
In this case, we don't need new primers. We are doing a Gibson using someone else's design

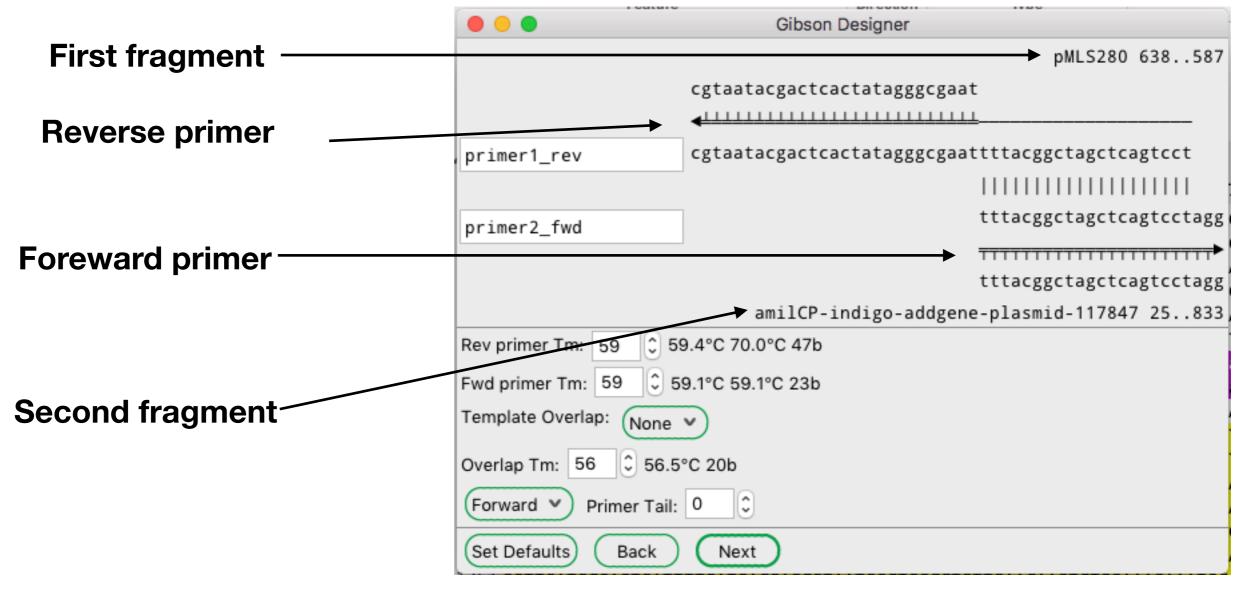


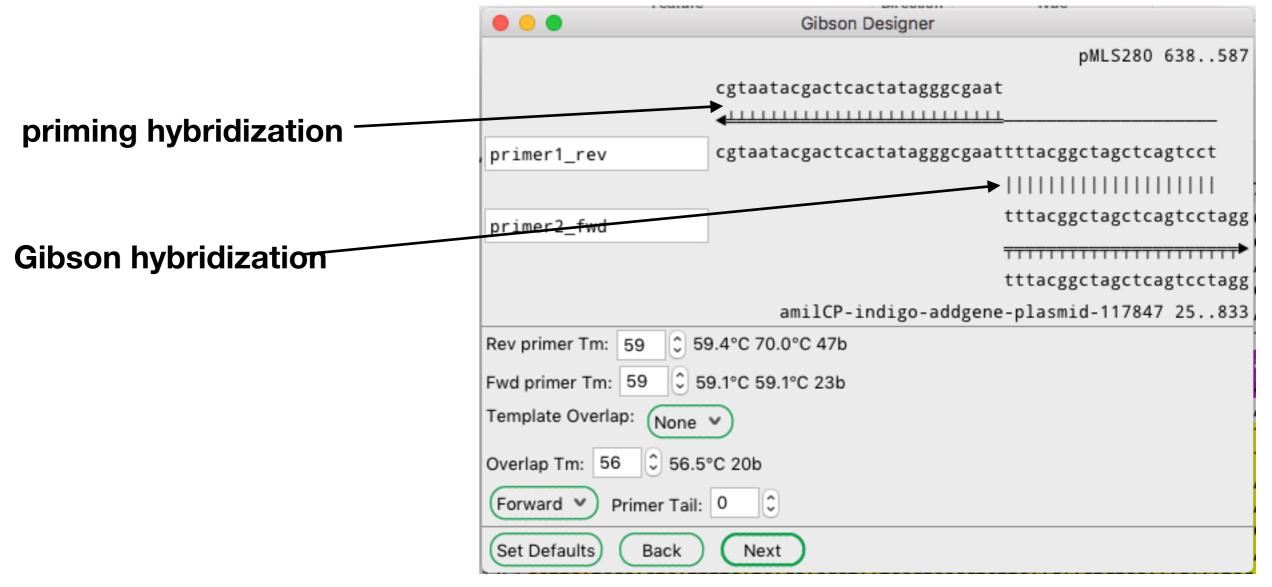


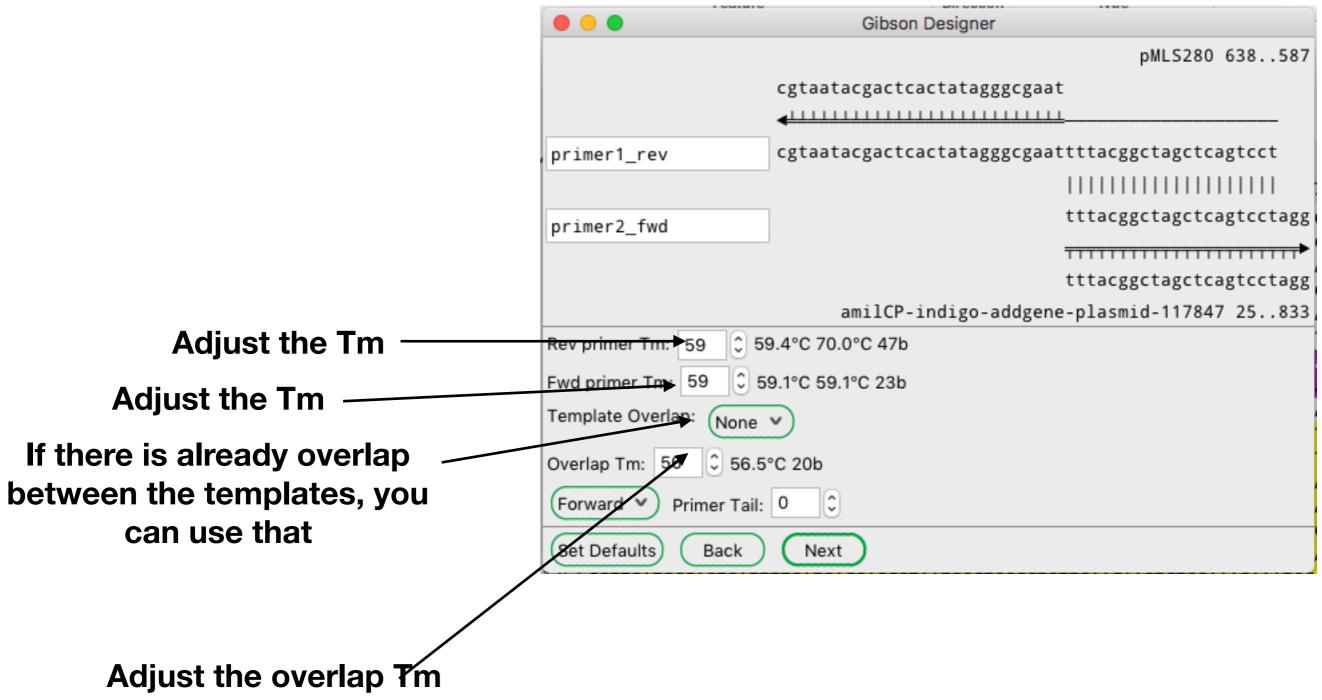


Design a new Gibson reaction









	• • •	Gibson Designer
		pMLS280 638587
		cgtaatacgactcactatagggcgaat
		IIIIIIIIIIIIIIIIIIIIIIIIIIIIIIIIIIII
	primer1_rev	cgtaatacgactcactatagggcgaattttacggctagctcagtcct
	primer2_fwd	tttacggctagctcagtcctagg
		++++++++++++++++++++++++++++++++++++++
		tttacggctagctcagtcctagg
		amilCP-indigo-addgene-plasmid-117847 25833
	Rev primer Tm: 59 🗘 5	59.4°C 70.0°C 47b
	Fwd primer Tm: 59 3 g	59.1°C 59.1°C 23b
	Template Overlap: None	 ✓
You can also add non-	Overlap Tm: 56 \$ 56.5	5°C 20b
templated sequence here	Forwarz V Primer Tail:	: 0
	Set Defaults Back	Next
Set which fragment gets the overlap	Add overla	p to the other side

junction

		Gibson Designer	
			pMLS280 638587
		cgtaatacgactcactatagggcgaa	t
		<pre>4111111111111111111111111111111111111</pre>	<u> </u>
	primer1_rev	cgtaatacgactcactatagggcgaa	ttttacggctagctcagtcct
1			
ſ	primer2_fwd		tttacggctagctcagtcctagg
	p1 1m01 2_1m0		,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
			tttacggctagctcagtcctagg
		amilCP-indigo-addgen	ne-plasmid-117847 25833
1	Rev primer Tm: 59 🗘 5	9.4°C 70.0°C 47b	
	Fwd primer Tm: 59 🗘 5	9.1°C 59.1°C 23b	
	Template Overlap: None	v	
	Overlap Tm: 56 🗘 56.5	°C 20b	
	Forward V Primer Tail:	0	
-	Set Defaults Back	Next	
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smal					<<<	misc_fr				1318		
Msl					<<<	misc_f				4554		
T						amilCP	Base					
	10	* 20		30	* 40			60	* 70	* 80		
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45 ACGCC	STEGTTECA	CTGAGCG	TCAGAC	CCCGTA	GAAAAGAT	TCAAAGGATCI	TTCTTGA	GATCCTT	TTTTTCT	GCGCGTAATCT	GCTGCTT	
33 GCAAA	асалалала	CCACCGC	TACCAG	CEGTGE	TITETITE	GCCGGATCAA	GAGCTAC	CAACTEE	TTTCCG	AAGGTAACTGS	ICTTCAGC.	
21 AGAGO	CGCAGATAC	CAAATAC	TGTTCT	TCTAGT	GTAGCCG	TAGTTAGGCC	ACCACTT	CAAGAAC	TCTGTAG	CACCECCTACA	TACCTCG	
09 CTCT0	GCTAATCCT	GTTACCA	GTGGCT	бствес	AGTEECGA	ATAAGTCGTGT	ICTTACO	GEGTTEG/	ACTEAAG	ACGATAGTTAC	CEGATAA	
						GCCCAGCTTG						
									CHCC3HA(LIGNONIACCI	Provide to a	
85 GAULI	IAIGAGAAA	IGCGCCAC	GCTTCC	CGAAGG								
					GAGAAAG	GCGGACAGGTJ	ATCCGGT	AAGEGGE	AGEGTEE	GAACAGGAGAG	GCGCACGA	
73 GGGAG	SCTTCCAGE	GGGAAAC	SCCTGG	TATCTT	GAGAAAGG	SCGGACAGGT/ CTGTCGGGTTI	ATCCGGT/ FCGCCAC	AAGEGGE() CTETGACI	AGEGTEEL FTEAGEET	GAACAGGAGAG ICGATTTTTGT	GATGCTC	
73 GGGAC 51 GTCAC	SCTTCCAGS SGGSSGCGS	IGGGAAAC IAGCCTAT	GCCTGG GGAAAA	TATCTT ACGCCA	GAGAAAGO TATAGTCO GCAACGCO	GCGGACAGGT/ CTGTCGGGTTI GGCC <mark>TTTTTA</mark> (ATCCGGT TCGCCAC CGGTTCC	AAGOGGO CTCTGAC TGGCCTT	AGGGTCGI FTGAGCGI FTGCTGGI	GAACAGGAGAG ICGATTTTGT CCTTTTGCTCA	GATSCTC ACATSTTC	
73 GCCAC 61 GTCAC 49 TTTCC	SCTTCCAGS SGGSSGCGS CTGCGTTAT	GGGAAAC AGCCTAT CCCCTGA	GCCTGG GGAAAA TTCTGT	TATCTT ACGCCA GGATAA	GAGAAAGO TATAGTCO GCAACGCO CCGTATTA	GCGSACAGGT/ CTGTCCGGTTT GGCCTTTTA(ACCSCCTTTG/	ATCCEST ICGCCAC CSGTTCC ASTEASC	AAGEGGO CTETGAC TGGECTT TGATACC	AGGGTCGI ITGAGCGI ITGCTGGI GCTCGCCI	GAACAGGAGAG ICGATTITTGT CCTTTTGCTCA GCAGCCGAACG	COCACGA GATGCTC ACATGTTC GACCGAGC	
73 GCCAC 61 GTCAC 49 TTTCC 37 GCAGC	SCTECCAGS SGGS SGCGS CTGCGTTAT CGASTCAGT	GGGAAAC AGCCTAT CCCCTGA GAGCGAG	GCCTGG GGAAAA TTCTGT GAAGCG	TATCTT ACGCCA GGATAA GAAGAG	GAGAAAGO TATAGTCO GCAACGCO CCGTATTA CGCCCAAT	SCSSACASGT/ CESECCESTE SGCCETETTA ACCSCCETES/ TACSCAAACCO	ATCCOST FLGCCAC CGGTTCC AGTGASC GCCTCTC	AASCGGO CTCTGAC TGGCCTT TGGTACCO CCCGCGCO	AGEGTEE ITEASEC ITEETGE SETESEC STTESEC	CAACAGGAGAG ICGATTTTGT CCTTTTGCTCA SCAGCCGAACG SATTCATTAAT	COCACGA IGATGCTC ACATGTTC ACCGAGC IGCAGCTG	
73 GGCAC 61 GTCAC 49 TTTCC 37 GCASC 25 GCACC	SCTTCCAGS SGGSSGCGS CTGCGTTAT CGASTCAGT SACASGTTT	IGGGAAAC IAGCCTAT CCCCTGA GAGCGAG CCCGACT	GCCTGG GGAAAA (TTCTGT) GAAGCG GGAAAG	TATCTT ACGCCA GGATAA GAAGAG CGGGCA	GAGAAAGO TATAGTCO GCAACGCO CCGTATTA CGCCCAAT GTGAGCGO	SCSSACASGT/ CESECCETETTA SGCCETETTA ACCSCCETEG/ FACSCAAACCO CAACGCAATT/	ATCCOST FLGCCAC CGGTTCC AGTGAGC GCCTCTC AATACGC	AASCGGO CTCTGAC TGGCCTT TGATACCO CCCGCGCO GTACCGC	AGEGTEE ITEASEC ITEETGE SETESECE STTESECE IAGEEAGE	EAACAGGAGAG ICGATTTTGT CCTTTTGCTCA SCAGCCGAACG SATTCATTAAT SAAGAGTTTGT	CGCACGA IGATGCTC ACATGTTC BACCGAGC IGCAGCTG IAGAAACG	
73 GCCAC 61 GTCAC 49 TTTCC 37 GCASC 25 GCASC 13 CAAAA	SCEECAGS SGGS SGC 65 CEGCGEEAT CGAGECAGE SACASGEET AAGGCCAEC	GCGAAAC AGCCTAT CCCCTGA GAGCGAG CCCGACT CCGTCAGG	SCCTSS SGAAAA TTCTST SAAGCS SGAAAS ATGGCC	TATCTT ACGCCA GGATAA GAAGAG CGGGCA TTCTGC	GAGAAAGO TATAGTCO GCAACSCO CCGTATTA CGCCCAAT GTGAGCGO TTAGTTTO	GESSACASGT/ CEGECETTER/ GECETTER/ ACCSCCTTE/ TACSCAAACCO CAACGCAATT/ SATSCCTSGC/	ATCCGST TCGCCAC CSGTTCC AGTGASC SCCTCTC AATACSC ASTTTAT	AAGEGGE CTETGAE TGGEETT TGATACCO CEESCGEO GTAECGE GGESGGEO	AGGGTCG ITGASCG ITGCTGGC GCTCGCCC GTTGSCCC IAGCCAG GTCCTGCC	CAACAGGAGAG ICGATTTTGT CCTTTTGCTCA GCAGCCGAACG GATTCATTAAT GAAGAGTTTGT CCGCCACCCTC	ACCACCA TGATGCTC ACATGTTC ACCGAGC TGCAGCTG TAGAAACG CCGGGCCG	
73 GCCAC 61 GTCAC 49 TTTCC 37 GCAGC 25 GCACC 13 CAAAA 01 TTGCT	SCITICAGS SGGS SGC SS CTGC STTAT CGAGTCAGT SACAGGTCAGT SACAGGCCATC TTCACAACS	GCGAAAC AGCCTAT CCCCTGA GAGCGAG CCCGACGAG CCCGACT CCGTCAGG	SCCTSS SGAAAA TTCTST SGAAGCG SGAAAG ATGGCC CCGCTC	TATCTT ACGCCA GGATAA GAAGAG CGGGCA TTCTGC CCGGCG	GAGAAAGO TATAGTCO GCAACGCO CCGTATTA CGCCCAAT GTGAGCGO TTAGTTTO GATTTSTO	SCSSACASGT CESECCETTEA SGCCETTEA ACCSCCETTEA TACSCAAACCO CAACSCAAACCO CAACSCAAACCO SAESCCESSCA CCEACECAGGA	ATCCGST TEGECACI EGGTTECE AGTGAGE GECTETCI AATACSCI AGTTTATI ASAGEGT	AAGEGGE CTETGAE TGGEETT TGATACCO CEEGEGGE GTAECGE GGEEGGE TEAECGA	AGGGTCG ITGASCGT ITGCTGGC SCTCSCCC STTGGCCC IAGCCAG STCCTGCC CAAACAA	CAACAGGAGAG ICGATTITIGT CCTTTIGCTCA SCAGCCGAACG SATTCATTAAT SAAGAGTTIGT CCGCCACCCTC CAGATAAAACG	ACCCACCA IGATGCTC ACCCGAGC IGCAGCTG IGCAGCTG IAGAAACG ICCGGSCCG GAAASGCC	
73 GCCAC 61 GTCAC 49 TTTCC 37 GCAGC 25 GCACC 13 CAAAA 01 TTGCT 69 CAGTC	SCEECAGS SGGS SGC SS CEGC SEECAGE CGASTCAGE SACASGEETE AAGS CCAEC ETEC CGACE	GCGAAAC AGCCTAT CCCCTGA GAGCGAG CCCGACT CCTCAGG TTCAAAT GAGCCTT	SCCTSS SGAAGA TTCTGT SGAAGCS GGAAGS ATGGCC CCGCTC TCGTTT	TATCTT ACGCCA GGATAA GAAGAG CGGGCA TTCTGC CCGGCG TATTTG	GAGAAAGO TATAGTCO GCAACGCO CCGTATTA CGCCCAAT GTGAGCGO TTAGTTTO GATTTGTO ATGCCTGO	SCSSACASGT/ SGCCTTTTA(ACCSCCTTTG/ TACSCAAACC(CAACGCAAACC) SATSCCTSGC/ SCTACTCAGG/ SCASTTCCCT/	ATCCGST TEGECACI EGGTTCC AGTGAGC GECTETCI AGTACGCI AGTACGCI AGTGCGT ACTCTCG	AAGEGGE CTETGAC TGGECTT TGATACCC CCEGEGGE GTACCGE GGEGGGE TCACCGA CGTTAAC	AGGGTCG ITGASCGT ITGCTGGC SCTCGCCC STTGSCCC IAGCCAGC STCCTGCC CAAACAAC SCTASCAT	CAACAGGAGAGA CCTTTTGCTCA SCAGCCGAACG SATTCATTAAT SAAGAGTTTGT CCGCCACCCTC CAGATAAAACG IGGATGTTTTC	ACCCACCA IGATGCTC ACATGTTC ACCCGAGC IGCAGCTG IGCAGCTG AGAAACG CCGGSCCG AAASGCC CCCAGTCA	
73 GCCAC 61 GTCAC 49 TTTCC 37 GCAGC 25 GCACC 13 CAAAA 01 TTGCT 69 CAGTC 77 CGACC	SCITICAGS SGGSSGCGS CTGCGTTAT CGAGTCAGT SACASGTTT AAGSCCATC FTCACAACS CTTCCGACT ST <mark>tgtaaaa</mark>	GCGAAAC AGCCTAT CCCCTGA GAGCGAG CCCGACT CCGTCAGG TTCAAAT GAGCCTT	SCCTSS SGAAGA TTCTST SGAAGCG GGAAGS ATGGCC CCGCTC TCGTTT cagtga	TATCTT ACGCCA GGATAA GAAGAG CGGGCA TTCTGC CCGGCG TATTTG gCgCgCg	GAGAAAGG TATAGTCO GCAACGCO CCGTATTA CGCCCAAT GTGAGCGO TTAGTTTO GATTTGTO ATGCCTGO gtaatacg	SCSSACASGT/ SGCCTTTTA(ACCSCCTTTG/ TACSGCAAACC(CAACGCAAACC) SATSCCTSGC/ SCTACTCAGG/ SCASTTCCCT/ gattactata	ATCCGST TEGECACI EGGTTCC AGTGAGC GECTCTCI AATACGC AGTTTATI AGAGCGT AGTCTCG agggcga	AAGEGGE CTETGAE TGGECTT TGATACCC CCEGEGEC GTAECGEC GGEGGGGC TEAECGA CGTTAAC atttae	AGGGTCGC ITGASCGT ITGCTGGC SCTCSCCC STTGSCCC IAGCCAGC STCCTGCC SCCASCAI SCCASCAI SCCASCAI	CAACAGGAGAGAG ICGATTITIGT CCTTTIGCTCA SCAGCCGAACG GATTCATTAAT GAAGAGTTTGT CCGCCACCCTC CAGATAAAACG ISGATGTTTTC CCGCCCCCCCCC	CGCACGA GATGCTC ACATGTTC GACCGAGC GGCAGCTG TAGAAACG CGGGSCCG GAAAGGCC CCCAGTCA GCCAGTCA	
73 GCCAC 61 GTCAC 49 TTTCC 37 GCAGC 25 GCACC 13 CAAAA 01 TTGC1 69 CAGTC 77 CGACC 55 CTage	SCITCCAGS SGGSSGCGS CTGCGTTAT CGAGTCAGT SACASGTTT AAGGCCATC ITCACAACS CTTCCGACT ST <mark>tgtaaaa</mark> tactagag	GCGAAAC AGCCTAT CCCCTGA GAGCGAG CCCGACT CCCCGACT CCTCAGG TTCAAAT GAGCCTT GAGCCTT	GCTGG GGAAAA TTCTGT GAAGCG GGAAAG ATGGCC CCGCTC TCGTTT Cagtga agaaat	TATCTT ACGCCA GGATAA GAAGAG CGGGCA TTCTGC CCGGCG TATTTG gcgcg <mark>C</mark> actagA	GAGAAAGG TATAGTCO GCAACGCO CCGTATTA CGCCCAAT GTGAGCGO TTAGTTTO GATTTSTO ATGCCTGO gtaatacg TSAGTSTO	GEGGACAGGT GEGETTTTA GEGETTTTA ACEGECATTTG/ TAEGEAAACCO CAAEGEAAACCO CAAEGEAAACCO SATSCETGEC/ GETACTEAGG/ GETCACTCAGG/ GAETEGETAAA	ATCCGSTA TCGCCAC CGGTTCC AGTGAGC GCCTCTC ACTCTCC AGTATAC AGAGCGT ACTCTCG BggcGa CAAATSA	AAGCGGC CTCTGAC TGGCCTT TGATACCC CCCGCGCG GTACCGC GGCGGGGC GGCGGGGC TCACCGA CGTTAACC GTTAACC GCTACAA	AGGGTCG ITGAGCG GCTCGCCG GTTGGCCG GTTGGCCAG GTCCTGCC CAAACAA(GCTAGCAG GCTAGCAG GCTAGCAG GCTAGCAG GCTAGCAG	CAACAGGAGAGAG ILGATTITIGT CCTTTIGCTCA GCAGCCGAACG GATTCATTAAT GAAGAGTTTGT CCGCCACCCTC CAGATAAAACG ISGATGTTTTC LCAGTCAGGCAC	GCCACCA GATGCTC ACATGTTC GACCGAGC GCAGCTG GCAGCTG GCAGCCG GCAGGCCG GAAAGGCC CCCAGTCA GGGTCAAT	
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2729 TGTAAAGCAGTCATTCCCGGAGGGCTATACATGGGAGAGGATCATGAACTTTGAAGATGGTGCAGTGTGTACTGTCAGCAATGATTCC 2817 AGCATCCAAGGCAACTGTTTCATCTACCATGTCAAGTTCTCTGGTTTGAACTTTCCTCCCAATGGACCTGTCATGCAGAAGAAGAAGACAC 2905 AGGGCTGGGAACCCAACACTGAGCGTCTCTTTGCACGAGATGGAATGCTGCTAGGAAACAACTTTATGGCTCTGAAGTTAGAAGGAGG 2993 CGGTCACTATTTGTGTGAATTTAAAACTACTTACAAGGCAAAGAAGCCTGTGAAGATGCCAGGGTATCACTATGTTGACCGCAAACTG GATGTAACCAATCACAACAAGGATTACACTTCGGTTGAGCAGTGTGAAATTTCCATTGCACGCAAACCTGTGGTCGCCTAAtaataatact 3081 ccggcaaaaaagggcaaggtgtcaccacctgccctttttctttaaaaccgaaaaga 3169 agtagcggccgctgcag 10 20 30 40 60 80 50 70 Gibson reaction: PCR: pMLS280 primer1_fwd gaatteetgeageeegg 59.4, 59.4 primer1_rev aggactgagctagccgtaaaattcgccctatagtgagtcgtattacg 59.4, 70.0 Product length : 2454

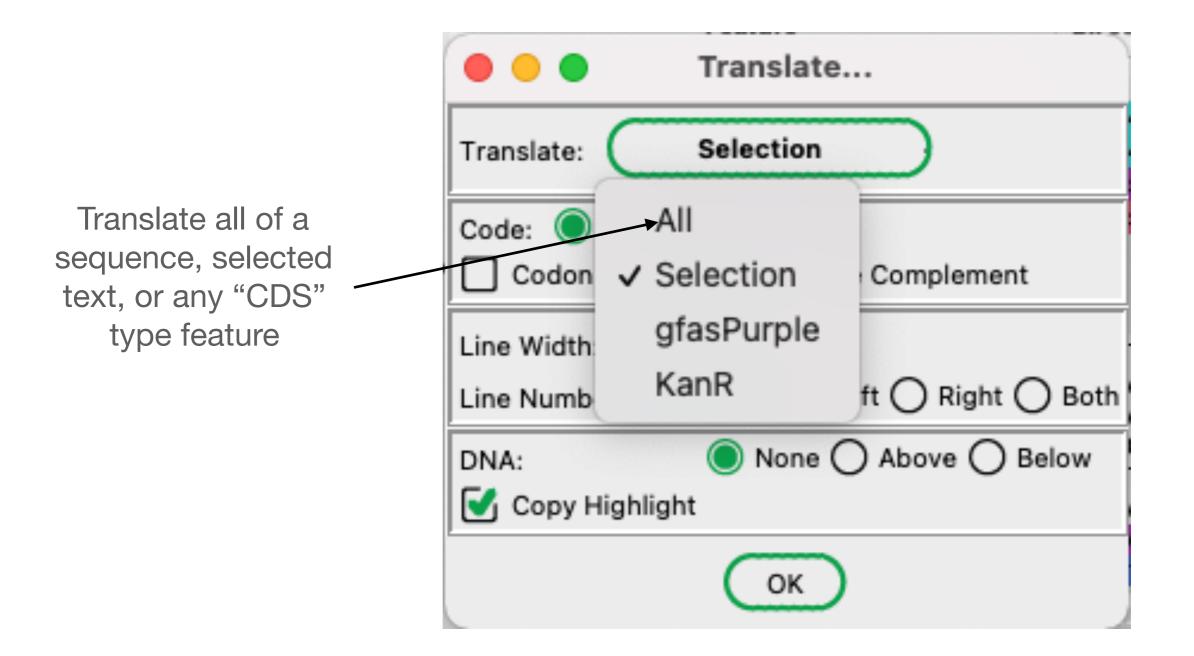
Gibson PCR products, primers etc. are in the file comment

		EFLQPG	
Feature	Direction	Туре	Location ↓
▶ Hidden			
▼ primer1_1wd			
		PCR_conditions	primer sequence:gaattootgoagcooggg
MCS-inverted in SK+	<<<	misc_feature	1.62
smal	<<<	misc_feature	1318
^ [[]	MCS-inverted	in SK+,primer1_fwd,pr	imer2_rev

2113 CAAAAAGGCCATCCGTCAGGATGSCCTTCTGCTTAGTTTGATSCCTGGCAGTTTATSGCGGGCGTCCTGCCCGCCACCCTCCGGSCCG 2289 CAGTCTTCCSACTGAGCCTTTCGTTTTATTTSATSCCTGGCASTTCCCTACTCTCGCGTTAACGCTASCATGGATGTTTTCCCASTCA GACGTtgtaaaacgacggccagtgagcgcgcgtaatacgactcactataggg 2377 2465 M13-fwd -77-⇒ primer2_fwd ⇒ 2553 primer 1 rev 🚝 2641 ٠. 2729 constitutive prom 281 2905 2993 3081 GATGTAACCAATCACAACAAGGATTACACTTCGGTTGAGCAGTGTGA GCACGCAAACCTGTGCTCGCCTAALaaLac

Gibson PCR primers are in the file features

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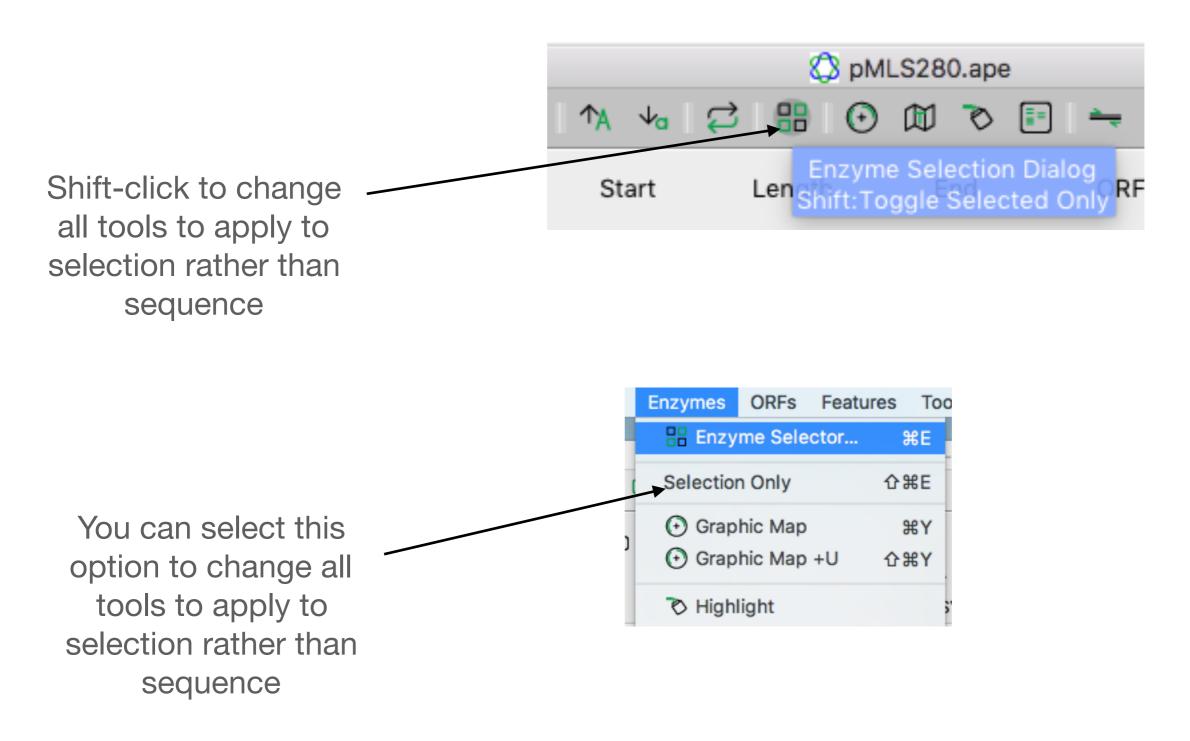


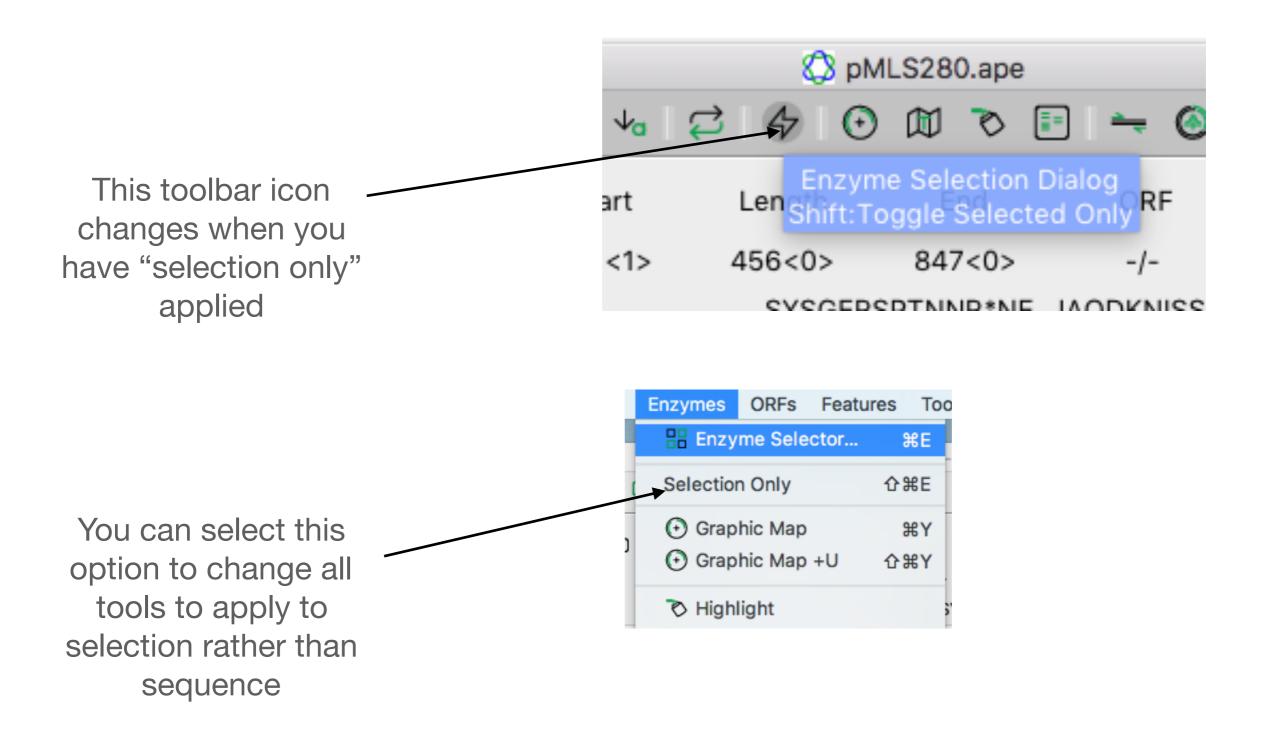
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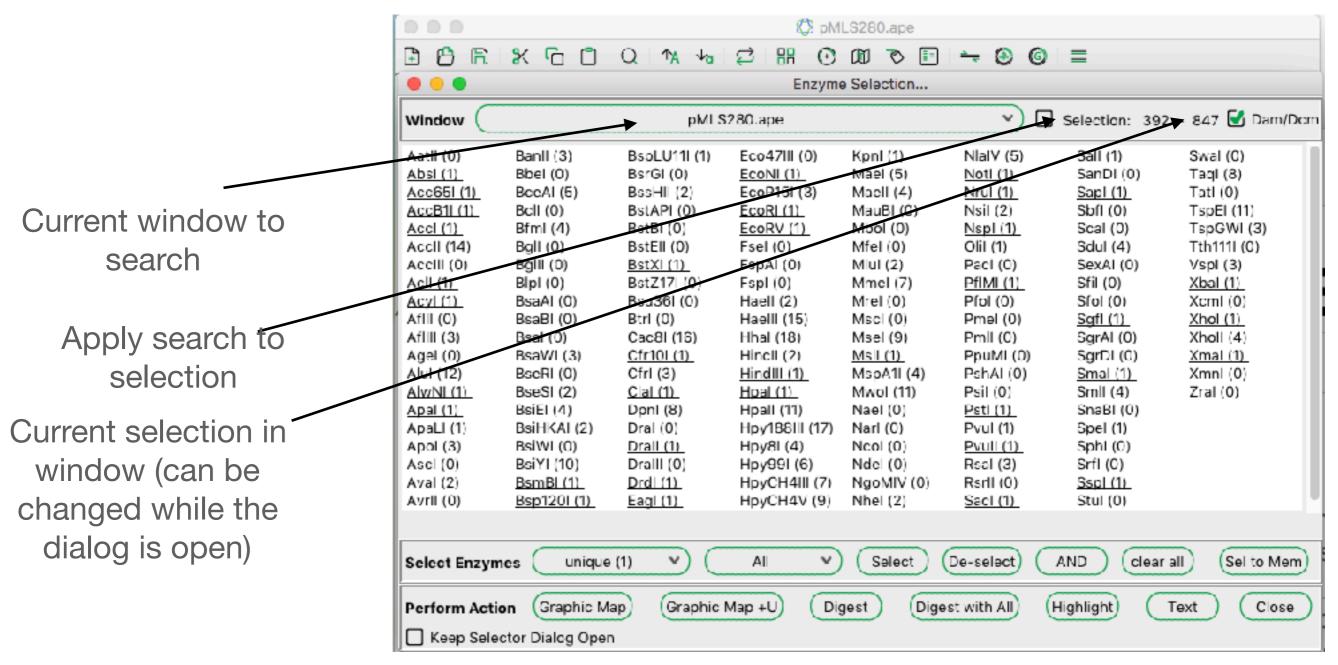
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Tue Jul 27, 2021 20:49 MDT 1. Ligation Product.ape /Users/waynedavis/Downloads/1. Ligation Product.ape gfasPurple Translation 221 a.a. MW=24947.95

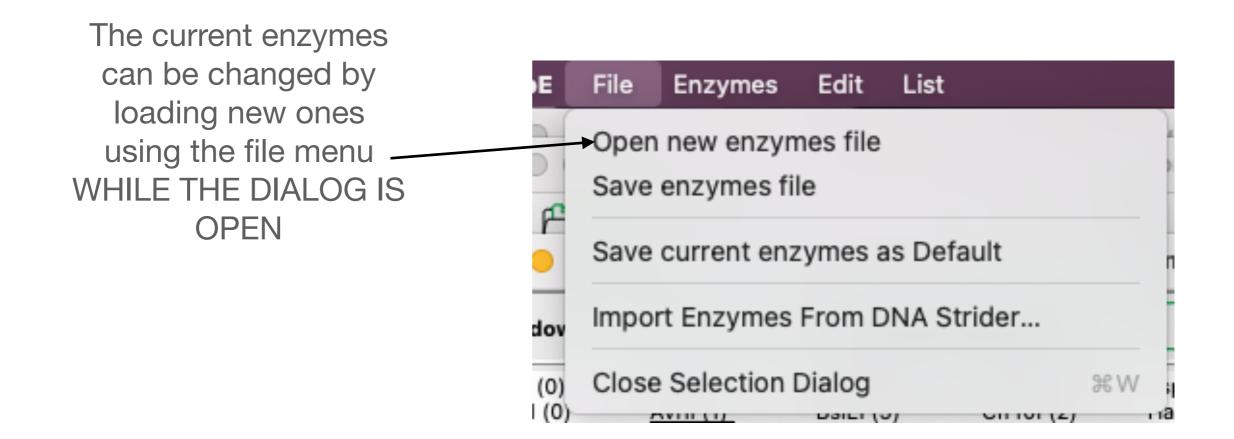
MSVIAKQMTYKVYMSGTVNG HYFEVEGDGKGKPYEGEQTV KLTVTKGGPLPFAWDILSPQ SQYGSIPFTKYPEDIPDYVK QSFPEGYTWERIMNFEDGAV CTVSNDSSIQGNCFIYHVKF SGLNFPPNGPVMQKKTQGWE PNTERLFARDGMLIGNNFMA LKLEGGGHYLCEFKSTYKAK KPVKMPGYHYVDRKLDVTNH NKDYTSVEQCEISIARKSVV A*







				Mq 🖏	LS280.ape			
	🕒 🖰 🖻	8 G O	Q 1/A to	2 H O	00 🔊 🗈	🗕 🕢 🔶	=	
				Enzyme	Selection			
	Window (pMLS	280.ape		~) 🗆	Selection: 392	2 - 847 🛃 Dam/Dom
All enzymes are shown here	Aatil (0) <u>Absi (1)</u> <u>Acc65i (1)</u> <u>AccB1i (1)</u> <u>Accl (1)</u> <u>Accli (14)</u> <u>Acclii (0)</u>	Banll (3) Bbel (0) BccAl (5) Bcll (0) Bfml (4) Bgll (0) Bglll (0)	BsoLU11I (1) BsrGI (0) BssHII (2) BstAPI (0) BstBI (0) BstEII (0) BstEII (0)	Eco47III (0) <u>EcoNI (1)</u> EcoP15I (3) <u>EcoRI (1)</u> <u>EcoRV (1)</u> Fsel (0) FspAI (0)	Kpnl (1) Mael (5) Maell (4) MauBl (0) Mool (0) Mfel (0) Mlul (2)	NlalV (5) <u>Notl (1)</u> Nsil (2) <u>Nspl (1)</u> Olil (1) Pacl (0)	Sall (1) SanDI (0) <u>Sapl (1)</u> Sbfl (0) Scal (0) Sdul (4) SexAl (0)	Swal (0) Taqi (8) Tati (0) TspEl (11) TspGWI (3) Tth1111 (0) VspI (3)
	Acil (1) Acyl (1) Afili (0) Afilii (3) Agel (0) Alul (12)	Bipi (0) BsaAi (0) BsaBi (0) Bsal (0) BsaWi (3) BscRi (0)	BstZ17I (0) Bsu36I (0) Btrl (0) Cac8I (16) <u>Cfr10I (1)</u> Cfrl (3)	Fspl (0) Haell (2) Haelli (15) Hhai (18) Hincii (2) <u>Hindiii (1)</u>	Mmel (7) Mrel (0) Msel (0) <u>Msel (9)</u> MspA1I (4) MspA1I (4)	<u>PfIMI (1)</u> Pfol (0) Pmel (0) Pmll (0) PpuMI (0) PshAI (0)	Sfil (0) Sfol (0) <u>Sgfl (1)</u> SgrAl (0) SgrDl (0) <u>Smal (1)</u>	<u>Xbal (1)</u> Xcml (0) <u>Xhol (1)</u> Xholl (4) <u>Xmal (1)</u> Xmnl (0) Zcml (0)
	<u>AlwNI (1)</u> <u>Apal (1)</u> ApaLI (1) Apol (3) Ascl (0) Aval (2) Avril (0)	BseSI (2) BsiEI (4) BsiHKAI (2) BsiWI (0) BsiYI (10) BsmBI (1) Bsp1201 (1)	<u>Clal (1)</u> DpnI (8) Dral (0) <u>DralI (1)</u> DralII (0) <u>DrdI (1)</u> Eagl (1)	<u>Hpal (1)</u> Hpall (11) Hpy18811 (17) Hpy81 (4) Hpy991 (6) HpyCH411 (7) HpyCH4V (9)	Mwol (11) Nael (0) Naol (0) Ndol (0) Ndol (0) NgoMIV (0) Nhel (2)	Psil (0) <u>Pstl (1)</u> Pvul (1) <u>Pvuli (1)</u> Rsal (3) Rsal (0) Saal (1)	Smll (4) SnaBl (0) Spel (1) Sphl (0) Srfl (0) <u>Sspl (1)</u>	Zral (0)
	Select Enzym				Salect		AND clear	all Sel to Mem
		ctor Dialog Oper						



		Enzyme Selection							
	Window 🤇		pMLS	280.ape		💙 🗌 Selection: 1 - 2484 🗹 Dar			
Click on any enzyme to select it Shift-click on any enzyme to select only that enzyme The enzyme currently	Aatll (0) <u>Absl (1)</u> <u>Acc65I (1)</u> <u>AccB1I (1)</u> <u>AccII (1)</u> AccII (14) AccIII (0) <u>AcII (1)</u> <u>AccIII (0)</u> <u>AfIII (0)</u> AfIII (0) <u>AfIII (1)</u> <u>AgeI (0)</u> <u>Alul (12)</u> <u>Alul (12)</u> <u>ApaI (1)</u> <u>ApaI (1)</u> <u>ApoI (3)</u>	Banli (3) Bbel (0) BceAl (5) Bcli (0) Bfml (4) Bgli (0) Bgli (0) BsaAl (0) BsaBl (0) BsaBl (0) BsaWl (3) BseRl (0) BseSl (2) BsiEl (4) BsiHKAI (2) BsiWl (0)	Bspl U111 (1) BsrGI (0) BssHI (2) BstAPI (0) BstEff (0) BstZ17I (0) BstZ17I (0) Bsu36I (0) Btrl (0) Cac8I (16) Cfr10I (1) CfrI (3) Clal (1) Dpnl (8) Drall (0)	Fco47III (0) <u>EcoNI (1)</u> FcoP15I (3) <u>FcoRV (1)</u> Fsel (0) FspI (0) Haell (2) Haell (15) Hhal (18) HincII (2) <u>HindIII (1)</u> <u>HpaI (1)</u> HpaII (11) Hpy188III (17) Hpy8I (4)	Kpnl (1) Mael (5) Maell (4) MauBl (0) Mbol (0) Mfel (0) Mfel (0) Mrel (2) Mrel (7) Mrel (7) Mrel (0) Msel (9) Msel (9) Msel (9) Msel (9) Msel (1) Nael (0) Narl (0) Ncol (0)	NlaIV (5) <u>Notl (1)</u> <u>Nrul (1)</u> Nsil (2) <u>Nspl (1)</u> <u>Olil (1)</u> Pacl (0) <u>PfIMI (1)</u> Pfol (0) Pmel (0) Pmel (0) Pmll (0) PshAI (0) Psil (0) <u>Pstl (1)</u> <u>Pvul (1)</u>	<u>Sall (1)</u> SanDI (0) <u>Sapl (1)</u> Sbfl (0) Scal (0) Sdul (4) SexAI (0) Sfol (0) Sfol (0) SgrAI (0) SgrAI (0) SgrAI (0) Smal (1) Smal (1) Small (4) SnaBI (0)	Swal (0) Taql (8) Tatl (0) TspEl (11) TspGWI (3) Tth1111 (0) Vspl (3) <u>Xbal (1)</u> Xcml (0) <u>Xholl (1)</u> Xholl (4) <u>Xmal (1)</u> Xmnl (0) Zral (0)	
under the mouse is shown here	Ascl (0) Aval (2) Avril (0) Select Enzym	BsiYi (10) BsmBI (1) <u>Bsp120I (1)</u> es unique	DrallI (0) Drdl (1) Eagl (1) (1) Graphic N	Hpy99I (6) HpyCH4III (7) HpyCH4V (9) el: A^CTAGT (con All	Ndel (0) NgoMIV (0) Nhel (2) mpat w/: AvrII N Select	Rsal (3) RsrII (0) <u>SacI (1)</u> Ihel Xbal) De-select	Srfl (0) Sspl (1) Stul (0)	all Sel to Mem Text Close	

				Enzyme	e Selection			
	Window 🤇		pMLS	280.ape		v [Selection: 1	- 2484 🗹 Dam/Dcm
	Aatll (0) Absl (1)	Banli (3) Bbel (0)	<u>Bspl U11I (1)</u> BsrGI (C)	Eco47III (0) EcoNII (1)	<u>Kpnl (1)</u> Mael (5)	NIaIV (5) Notl (1)	<u>Sall (1)</u> SanDI (0)	Swal (0) Tagl (8)
	Acc65L(1)	BceAl (5)	BssHI (2)	<u>EcoNI (1)</u> EcoP15I (3)	Maell (4)	Nrul (1)	Sapl (1)	Tatl (0)
	AccB1I (1)	Bcll (0)	BstAPI (0)	EcoRI (1)	MauBI (C)	Nsil (2)	Sbfl (0)	TspEl (11)
	Accl (1)	Efml (4)	BstBI (0)	EcoRV (1)	Mbol (0)	Nspl (1)	Scal (0)	TspGWI (3)
	AccII (14)	Bgll (0)	BstEll (0)	Fsel (0)	Mfel (0)	Olil (1)	Sdul (4)	Tth1111 (C)
	AccIII (0)	BgII (0)	BstXI (1)	FspAI (0)	Miul (2)	Pacl (0)	SexAI (0)	Vspl (3)
	Acll (1)	Bipl (0)	BstZ17I (0)	Fspl (0)	Mmel (7)	PfIMI (1)	Sfil (O)	Xbal (1)
	Acyl (1)	BsaAl (0)	Bsu36I (0)	Haell (2)	Mrel (0)	Pfol (0)	Sfol (0)	Xcml (C)
	AfIII (C)	BsaBI (0)	Btrl (0)	HaellI (15)	Mscl (0)	Pmel (0)	Sgfl (1)	Xhol (1)
	AfIIII (3)	Bsal (0)	Cac8I (16)	Hhal (18)	Msel (9)	Pmll (0)	SgrAI (0)	Xholl (4)
	Agel (0)	BsaWI(3)	Cfr10I (1)	Hincll (2)	<u>Msll (1)</u>	PpuMI (0)	SgrDI (C)	Xmal (1)
	Alul (12)	BseRI (0)	Cfrl (3)	HindIII (1)	MspA1 (4)	PshAI (0)	Smal (1)	Xmnl (0)
	AlwNI (1)	BseSI (2)	Clal (1)	<u>Hpal (1)</u> Heall (11)	Mwol (11)	Psil (0)	Smll (4)	Zral (0)
	Apal (1)	BsiEl (4)	Dpnl (8)	Hpall (11)	Nael (0)	<u>Pstl (1)</u>	SnaBl (0) Spel (1)	
	ApaLI(1) Apol(3)	BsiHKAI (2) BsiWI (0)	Dral (0) Drall (1)	Hpy188III (17) Hpy8I (4)	Narl (0) Ncol (0)	<u>Pvul (1)</u> Pvull (1)	SphI (0)	
The sector sticks with suite		BsiYI (10)	DrallI (0)	Hpy991 (6)	Ndel (0)	Rsal (3)	Srfl (0)	
The selection criteria	Aval (2)	EsmBI (1)	Drdl (1)	HpyCH4III (7)	NgoMIV (0)	RsrII (0)	Sspl (1)	
are shown here —	Avrll (0)	Bsp120I (1)	Eagl (1)	HpyCH4V (9)	Nhel (2)	Sacl (1)	Stul (0)	
			Spe	el: A^CTAGT (cor	mpat w/: Avril N	hel Xbal}		
	Select Enzym	es 🕞 unique	(1)	All 💙	Select (De-select	AND clear	all Sel to Mem
	Perform Actio	on Graphic Ma	ap Graphic M	Map +U Dig	jest Dige	st with All	lighlight	Text Close
	Keep Selec	ctor Dialog Open	I					

				Enzyme	e Selection			
	Window 🤇		pMLS	280.ape		v	Selection: 1	- 2484 🗹 Dam/Dom
Any enzyme matching both the count AND the group are underlined	Aatli (0) <u>Absi (1)</u> <u>Acc65I (1)</u> <u>AccB1I (1)</u> <u>AccII (1)</u> <u>AccII (1)</u> <u>AccII (1)</u> <u>AccII (1)</u> <u>AccII (1)</u> <u>ActII (1)</u> <u>AfIII (0)</u> <u>AfIII (3)</u> <u>AgeI (0)</u> <u>Alul (12)</u> <u>Alul (12)</u> <u>ApaLI (1)</u> <u>ApaLI (1)</u> <u>ApaLI (1)</u> <u>ApoI (3)</u> <u>AscI (0)</u> <u>AvaI (2)</u> <u>AvrII (0)</u>	Banli (3) Bbel (0) BceAI (5) Bcll (0) Bfml (4) Bgll (0) Bgll (0) BsaAI (0) BsaAI (0) BsaBI (0) BsaBI (0) BsaBI (0) BseBI (0) BseBI (2) BsiEI (4) BsiHKAI (2) BsiHKAI (2) BsiYI (10) BsmBI (1) Bsp120I (1)	Bspl U11I (1) BsrGI (0) BssHI (2) BstAPI (0) BstBI (0) BstEII (0) BstEII (0) BstZ17I (0) Bsu36I (0) Btrl (0) Cac8I (16) Cfr10I (1) CfrI (3) Clal (1) Dpnl (8) Drall (0) Drall (1) Eagl (1)	Eco47III (0) EcoNI (1) EcoP15I (3) EcoRI (1) EcoRV (1) Esel (0) EspAI (0) EspI (0) HaeII (2) HaeIII (15) Hhal (18) HincII (2) HindIII (11) HpaI (1) Hpy188III (17) Hpy8I (4) Hpy99I (6) HpyCH4VI (9)	Kpnl (1) Mael (5) Maell (4) MauBl (0) Mbol (0) Mfel (0) Mfel (0) Mrel (2) Mrel (2) Mrel (0) Msel (9) Msel (9) Msel (9) Msel (9) Msel (1) MspA1i (4) Mwol (11) Nael (0) Narl (0) Ncol (0) Ndel (0) Ndel (0) Nhel (2)	NlaIV (5) <u>Notl (1)</u> <u>Nrul (1)</u> Nsil (2) <u>Nspl (1)</u> <u>Olil (1)</u> Pacl (0) <u>PfIMI (1)</u> Pfol (0) Pmel (0) Pmll (0) PshAl (0) Psil (0) <u>Pstl (1)</u> <u>Pvul (1)</u> <u>Rsal (3)</u> <u>Rsrll (0)</u> <u>Sacl (1)</u>	<u>Sall (1)</u> SanDI (0) <u>Sapl (1)</u> SbfI (0) Scal (0) Sdul (4) SexAI (0) Sfil (0) Sfol (0) SgfI (1) SgrAI (0) SgrAI (0) SgrDI (0) Small (1) SnaBI (0) Spel (1) SphI (0) SrfI (0) SspI (1) Stul (0)	Swal (0) Taqi (8) Tati (0) TspEl (11) TspGWI (3) Tth1111 (0) Vspi (3) <u>Xbal (1)</u> Xcml (0) <u>Xholi (1)</u> Xholi (4) <u>Xmal (1)</u> Xmni (0) Zral (0)
	·		Sp	el: A^CTAGT (co	mpat w/: AvrILN	lhel Xbal)		
	Select Enzyme Perform Actio	n Graphic Ma	p Graphic I	All V Map +U Dig		De-select	AND clear	Text Close
	C Keep Selec	tor Dialog Open						
This is the group								

1	•••			Enzym	e Selection				
	Window 🤇		pMLS	280.ape		🗸 🗌 Selection: 1 - 2484 🗹 Dam/Dom			
	Aatll (0) <u>Absl (1)</u> <u>Acc65l (1)</u> <u>Acc65l (1)</u> <u>Acc1 (1)</u> <u>Accll (1)</u> <u>Accll (14)</u> <u>Accll (10)</u> <u>Accll (1)</u> <u>Accll (1)</u> <u>AflII (0)</u> <u>AflII (3)</u> <u>Agel (0)</u> <u>Alul (12)</u> <u>Alul (12)</u> <u>Alul (11)</u> <u>Apal (1)</u> <u>Apal (1)</u> <u>Apol (3)</u> <u>Ascl (0)</u> <u>Aval (2)</u> <u>Avrll (0)</u>	Banll (3) Bbel (0) BceAl (5) Bcll (0) Bfml (4) Bgll (0) Bgll (0) Bgll (0) BsaAl (0) BsaAl (0) BsaBl (0) BsaBl (0) BsaBl (0) BseRl (0) BseRl (2) BsiEl (4) BsiHKAI (2) BsiHKAI (2) BsiYI (10) BsmBl (1) Bsp120I (1)	Bspl U11I (1) BsrGI (0) BssHI (2) BstAPI (0) BstBI (0) BstEII (0) BstEII (0) BstEII (0) BstZ17I (0) Bsu36I (0) BtrI (0) Cac8I (16) Cfr10I (1) CfrI (3) Clal (1) DpnI (8) Drall (0) Drall (1) DrdI (1) Eagl (1)	Eco47III (0) EcoNI (1) EcoP15I (3) EcoRI (1) EcoRV (1) Esel (0) EspAI (0) EspI (0) HaeII (2) HaeIII (15) Hhal (18) HincII (2) HindIII (11) HpaI (1) Hpy188III (17) Hpy8I (4) Hpy99I (6) HpyCH4VI (9)	Kpnl (1) Mael (5) Maell (4) MauBl (0) Mbol (0) Mfel (0) Mfel (0) Mrel (2) Mrel (2) Mrel (0) Mscl (0) Msel (9) <u>Msll (1)</u> MspA1l (4) Mwol (11) Nael (0) Narl (0) Ndel (0) Ndel (0) Ndel (0) Ndel (2)	NlaIV (5) <u>Notl (1)</u> <u>Nsil (2)</u> <u>Nsil (2)</u> <u>Nsil (2)</u> <u>Olil (1)</u> Pacl (0) <u>PfIMI (1)</u> Pfol (0) <u>Pmel (0)</u> <u>Pmel (0)</u> <u>Pmil (0)</u> <u>PshAI (0)</u> <u>Psil (0)</u> <u>Pstl (1)</u> <u>Pvul (1)</u> <u>Pvul (1)</u> <u>Rsal (3)</u> <u>RsrII (0)</u> <u>Sacl (1)</u>	<u>Sall (1)</u> SanDI (0) <u>Sapl (1)</u> SbfI (0) Scal (0) Sdul (4) SexAI (0) Sfil (0) Sfol (0) <u>SgfI (1)</u> SgrAI (0) SgrDI (0) <u>Smal (1)</u> SmII (4) SnaBI (0) <u>Spel (1)</u> SphI (0) SrfI (0) Sspl (1) Stul (0)	Swal (0) Taqi (8) Tati (0) TspEl (11) TspGWI (3) Tth1111 (0) Vspi (3) <u>Xbal (1)</u> XcmI (0) <u>Xholi (1)</u> Xholi (4) <u>Xmal (1)</u> XmnI (0) Zral (0)	
			Sp	el: A^CTAGT (co	mpat w/: AvrII N	/: AvrII Nhel Xbal)			
	Select Enzym	ies Unique	(1) ~) (All 👻	Select	Da-select)	AND clear	all Sel to Mem	
	Perform Acti	on Graphic Ma ctor Dialog Open		Man +0 Di	Dige	est with All	Highlight	Text Close	
Click here to select underlined enzym	es C		to DE-se ned enzy			nere to c LL enzyr	le-select nes		

•••			Enzyme	e Selection			
Window 🤇		pMLS	280.ape		v (Selection: 1	- 2484 🗹 Dam/Dom
Aatll (0)	Banli (3)	Bspl.U111 (1)	Eco47III (0)	Kpnl (1)	NIalV (5)	Sall (1)	Swal (0)
Absl (1)	Bbel (0)	BsrGI (C)	EcoNI (1)	Mael (5)	Notl (1)	SanDI (0)	Taqi (8)
Acc65I (1)	BceAl (5)	BssHI (2)	EcoP15I (3)	Maell (4)	Nrul (1)	Sapl (1)	Tatl (0)
AccB1I (1)	Bell (0)	BstAPI (0)	EcoRI (1)	MauBI (C)	Nsil (2)	Sbfl (0)	TspEl (11)
Accl (1)	Efml (4)	BstBI (0)	EcoRV (1)	Mbol (0)	<u>Nspl (1)</u>	Scal (0)	TspGWL(3)
Accll (14)	Bgll (0)	BstEll (0)	Fsel (0)	Mfel (0)	Olil (1)	Sdul (4)	Tth1111 (C)
AccIII (0)	BgII (0)	BstXI (1)	EspAI (0)	Mlul (2)	Pacl (0)	SexAI (0)	Vspl (3)
Acll (1)	Blpl (0)	BstZ17I (0)	Fspl (0)	Mmel (7)	PfIMI (1)	Sfil (O)	Xbal (1)
Acyl (1)	BsaAI (0)	Bsu36I (0)	Haell (2)	Mrel (0)	Pfol (0)	Sfol (0)	Xcml (C)
AfIII (C)	BsaBI (0)	Btrl (0)	Haelli (15)	Mscl (0)	Pmel (0)	Sgfl (1)	Xhol (1)
AfIIII (3)	Bsal (0)	Cac8I (16)	Hhal (18)	Msel (9)	Pmll (0)	SgrAI (0)	Xholl (4)
Agel (0)	BsaWI(3)	Cfr10I (1)	Hincll (2)	<u>Msll (1)</u>	PpuMI (0)	SgrDI (C)	Xmal (1)
Alul (12)	BseRI (0)	Cfrl (3)	HindIII (1)	MspA1 (4)	PshAI (0)	Smal (1)	XmnI (0)
AlwNI (1)	BseSI (2)	Clal (1)	<u>Hpal (1)</u> He all (11)	Mwol (11)	Psil (0)	Smll (4)	Zral (0)
Apal (1)	BsiEl (4)	Dpnl (8)	Hpall (11)	Nael (0)	Pstl (1)	SnaBI (0)	
ApaLI(1)	BsiHKAI (2)	Dral (0)	Hpy188III (17)	Narl (0)	Pvul (1)	Spel (1)	
Apol (3)	BsiWI (0)	Drall (1)	Hpy8I (4)	Ncol (C)	Pvull (1)	SphI (0)	
Ascl (0)	BsiYI (10) RemPL(1)	Dralli (0)	Hpy991 (6)	Ndel (0)	Rsal (3)	Srfl (0) Scol (1)	
Aval (2) Avrll (0)	BsmBI (1) <u>Bsp120I (1)</u>	Drdl (1) Eagl (1)	HpyCH4III (7) HpyCH4V (9)	NgoMIV (0) Nhel (2)	Rsrll (0) Sacl (1)	Sspl (1) Stul (0)	
Aviii (0)	<u>bsp1201(1)</u>		el: A^CTAGT (co			5(0)(0)	
Select Enzyr	mes unique		All 💙	(Select)	De-select	AND clear	rall (Sel to Mem)
Perform Act	ion Graphic M	ap) (Graphic	Map +U) Dir	gest) Dige	est with All)	Highlight)	Text Close
_	ector Dialog Oper			Jest Page	-se with All		Text Close
	ceto blang oper						
Clic	k here to	select -					
only	enzymes	inat are					
currer	nuy selec	cted AND	1				
	undarlin						
	underlin	IEU					

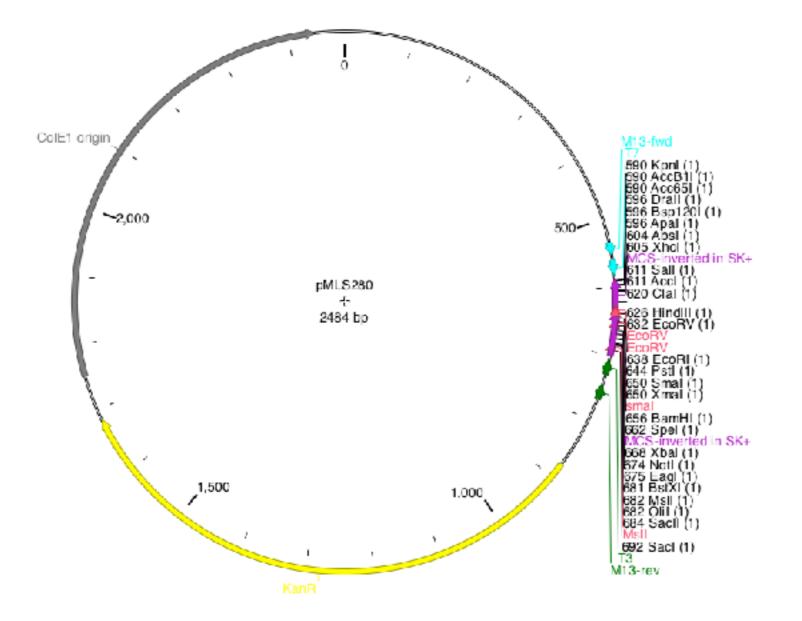
Vindow 🤇		pMLS	280.ape	·) [Selection: 1	- 2484 🗹 Dam/D	
Aatll (0)	Banll (3)	BspLU11I (1)	Eco47III (0)	Kpnl (1)	NIalV (5)	Sall (1)	Swal (0)
	Bbel (0)	BsrGI (0)	EcoNI (1)	Mael (5)	Notl (1)	SanDI (0)	Taql (8)
Acc65I (1)	BceAl (5)	BssHII (2)	EcoP15I (3)	Maell (4)	Nrul (1)	Sapl (1)	Tatl (0)
AccB1I (1)	Bcll (0)	BstAPI (0)	EcoRI (1)	MauBI (0)	Nsil (2)	Sbfl (0)	TspEl (11)
Accl (1)	Bfml (4)	BstBl (0)	EcoRV (1)	Mbol (0)	Nspl (1)	Scal (0)	TspGWI (3)
Accll (14)	Bgll (0)	BstEll (0)	Fsel (0)	Mfel (0)	Olil (1)	Sdul (4)	Tth1111 (0)
AccIII (0)	Bglll (0)	BstXI (1)	FspAI (0)	Mlul (2)	Pacl (0)	SexAI (0)	Vspl (3)
Acll (1)	Blpl (0)	BstZ17I (0)	Fspl (0)	Mmel (7)	PfIMI (1)	Sfil (0)	Xbal (1)
	BsaAI (0)	Bsu36I (0)	Haell (2)	Mrel (0)	Pfol (0)	Sfol (0)	Xcml (0)
Afili (0)	BsaBI (0)	Btrl (0)	Haelli (15)	Mscl (0)	Pmel (0)	Sqfl (1)	Xhol (1)
AfIIII (3)	Bsal (0)	Cac8l (16)	Haelii (13) Hhal (18)	Msel (9)	Pmll (0)	SgrAI (0)	Xholl (4)
Agel (0)	BsaWI (3)	Cfr10I (1)	Hincll (2)	Msll (1)	PpuMI (0)	SgrDI (0)	Xmal (1)
Alul (12)	BseRI (0)	-		1 (4)	PshAI (0)	Smal (1)	Xmnl (0)
AlwNI (1)	BseSI (2)		enzyme search		Psil (0)	Smll (4)	Zral (0)
Apal (1)	BsiEl (4)		soschizomer se	arch	Pstl (1)	SnaBl (0)	2141(0)
ApaLI (1)	BsiHKAI (2)	REBASE: 9	supplier search	S.	Pvul (1)	Spel (1)	
Apol (3)	BsiWI (0)	Drzil (1)	Hpy8I (4)	Ncol (0)	Pvull (1)	SphI (0)	
Ascl (0)	BsiYI (10)	Dralll (0)	Hpy99I (6)	Ndel (0)	Rsal (3)	Srfl (0)	
Aval (2)	BsmBl (1)	<u>Drdl (1)</u>	HpyCH4III (7)	NgoMIV (0)	Rsrll (0)	Sspl (1)	
Avrll (0)	Bsp120I (1)	Eagl (1)	HpyCH4V (9)	Nhel (2)	Sacl (1)	Stul (0)	
(0)		cage (17		I: A^AGCTT	Justin	0(0)	
Select Enzyn	nes unique	e (1) V	All 🗸	Select	De-select	AND clear	r all Sel to Mem
Perform Acti	on Graphic M	lap Graphic	Map +U Dig	gest Dige	st with All	Highlight	Text Close
Keep Sele	ctor Dialog Ope	n					
- /							

Right-click to search for enzyme info

8 -		Enzyme	e Selection			
Window	pMLS	5280.ape	0.ape			- 2484 🗹 Dam/Dcm
Aatll (0) Banll (3) Absl (1) Bbel (0) Acc65I (1) BceAI (5) AccB1I (1) Bcll (0) Accl (1) Bfml (4) Accll (14) Bgll (0) Accll (1) Bfpl (0) Accll (1) Blpl (0) Accll (1) Blpl (0) Accll (1) BsaAl (0) Afill (0) BsaBl (0) Afill (3) Bsal (0) Agel (0) BsaVI (3) Alul (12) BseRI (0) Alul (12) BsiEI (4) Apal (1) BsiHKAI (2) Apol (3) BsiWI (0) Aval (2) BsmBl (1) Avril (0) Bsp120I (1)	Drall (1) Dralll (0) Drdl (1)	Eco47III (0) <u>EcoNI (1)</u> EcoP15I (3) <u>EcoRV (1)</u> Fsel (0) FspAI (0) FspI (0) HaeIII (2) HaeIII (15) Hhal (18) HincII (2) <u>HindIII (1)</u> <u>HpaI (1)</u> HpaII (11) <u>Hpy188III (17)</u> Hpy8I (4) Hpy99I (6) HpyCH4VI (9)	Kpnl (1) Mael (5) Maell (4) MauBl (0) Mbol (0) Mfel (0) Miul (2) Mmel (7) Mrel (0) Mscl (0) Mscl (0) Msel (9) Msll (1) MspA1I (4) Mwol (11) Nael (0) Narl (0) Ncol (0) Ndel (0) NgoMIV (0) Nhel (2)	NlalV (5) Notl (1) Nrul (1) Nsil (2) Nspl (1) Oliil (1) Pacl (0) PfIMI (1) Pfol (0) Pmel (0) Pmel (0) Pmll (0) PshAl (0) PshAl (0) Psil (0) Pstl (1) Pvull (1) Rsal (3) Rsrll (0) Sacl (1)	Sall (1) SanDI (0) Sapl (1) SbfI (0) Scal (0) Sdul (4) SexAl (0) Sfil (0) Sfol (0) SgrI (1) SgrAl (0) SgrDI (0) Smal (1) Small (4) SnaBl (0) Spel (1) Sphl (0) Srfl (0) Spel (1) Sphl (0) Srfl (0) Stol (0)	Swal (0) Taql (8) Tatl (0) TspEl (11) TspGWI (3) Tth1111 (0) Vspl (3) Xbal (1) Xcml (0) Xhol (1) Xholl (4) Xmal (1) Xmnl (0) Zral (0)
Select Enzymes union Perform Action Graphic Keep Selector Dialog O here to do is functions		All V Map +U Dig	gest Dige	De-select	AND clear	r all Sel to Mem Text Close

	Enzyme Selection							
Window 🤇		pML	S280.ape	· □	Selection: 58	7 - 726 🗹 Dam/		
Aatll (0)	Banll (3)	BspLU11I (1)	Eco47III (0)	Kpnl (1)	NIaIV (5)	Sall (1)	Swal (0)	
Absl (1)	Bbel (0)	BsrGI (0)	EcoNI (1)	Mael (5)	Notl (1)	SanDI (0)	Taql (8)	
Acc65I (1)	BceAI (5)	BssHII (2)	EcoP15I (3)	Maell (4)	Nrul (1)	Sapl (1)	Tatl (0)	
AccB1I (1)	Bcll (0)	BstAPI (0)	EcoRI (1)	MauBI (0)	Nsil (2)	Sbfl (0)	TspEI (11)	
Accl (1)	Bfml (4)	BstBI (0)	EcoRV (1)	Mbol (0)	Nspl (1)	Scal (0)	TspGWI (3)	
Accll (14)	Bgll (0)	BstEll (0)	Fsel (0)	Mfel (0)	Olil (1)	Sdul (4)	Tth1111 (0)	
AccIII (0)	BgIII (0)	BstXI (1)	FspAI (0)	Mlul (2)	Pacl (0)	SexAI (0)	Vspl (3)	
Acll (1)	Blpl (0)	BstZ17I (0)	Fspl (0)	Mmel (7)	PfIMI (1)	Sfil (0)	Xbal (1)	
Acyl (1)	BsaAI (0)	Bsu36I (0)	Haell (2)	Mrel (0)	Pfol (0)	Sfol (0)	Xcml (0)	
AfIII (0)	BsaBI (0)	Btrl (0)	Haelll (15)	Mscl (0)	Pmel (0)	Sgfl (1)	Xhol (1)	
AfIIII (3)	Bsal (0)	Cac8I (16)	Hhal (18)	Msel (9)	Pmll (0)	SgrAI (0)	Xholl (4)	
Agel (0)	BsaWI (3)	Cfr10I (1)	Hincll (2)	Msll (1)	PpuMI (0)	SgrDI (0)	Xmal (1)	
Alul (12)	BseRI (0)	Cfrl (3)	HindIII (1)	MspA1I (4)	PshAI (0)	Smal (1)	Xmnl (0)	
AlwNI (1)	BseSI (2)	Clal (1)	Hpal (1)	Mwol (11)	Psil (0)	Smll (4)	Zral (0)	
Apal (1)	BsiEI (4)	Dpnl (8)	Hpall (11)	Nael (0)	Pstl (1)	SnaBl (0)		
ApaLI (1)	BsiHKAI (2)	Dral (0)	Hpy188III (17)	Narl (0)	Pvul (1)	Spel (1)		
Apol (3)	BsiWI (0)	Drall (1)	Hpy8I (4)	Ncol (0)	Pvull (1)	SphI (0)		
Ascl (0)	BsiYI (10)	Dralll (0)	Hpy99I (6)	Ndel (0)	Rsal (3)	Srfl (0)		
Aval (2)	BsmBl (1)	Drdl (1)	HpyCH4III (7)	NgoMIV (0)	Rsrll (0)	Sspl (1)		
Avrll (0)	Bsp120I (1)	Eagl (1)	HpyCH4V (9)	Nhel (2)	Sacl (1)	Stul (0)		
Diff Window	pMLS280.ape	🗸 🗹 Selecti	ion Only					
Select Enzyr	nes unique		Difference ^	Select	De-select	AND clear	r all Sel to Me	
Perform Act	Perform Action Graphic Map Grap Grap All Mem Recall t Digest with All Highlight Text Close							
Keep Sele	ector Dialog Ope	n 🤒	Difference					
ce group	IS							
Jai								
cial	13							

- Select the MCS in pMLS280
- Open the enzyme selector
- Select all unique enzymes in pMLS280
- Chose the difference group and pMLS280+selection within the difference selector
- Deselect the different enzymes
- Do a graphic map
- What do you see?



Find enzymes to clone amilCP into pMLS280

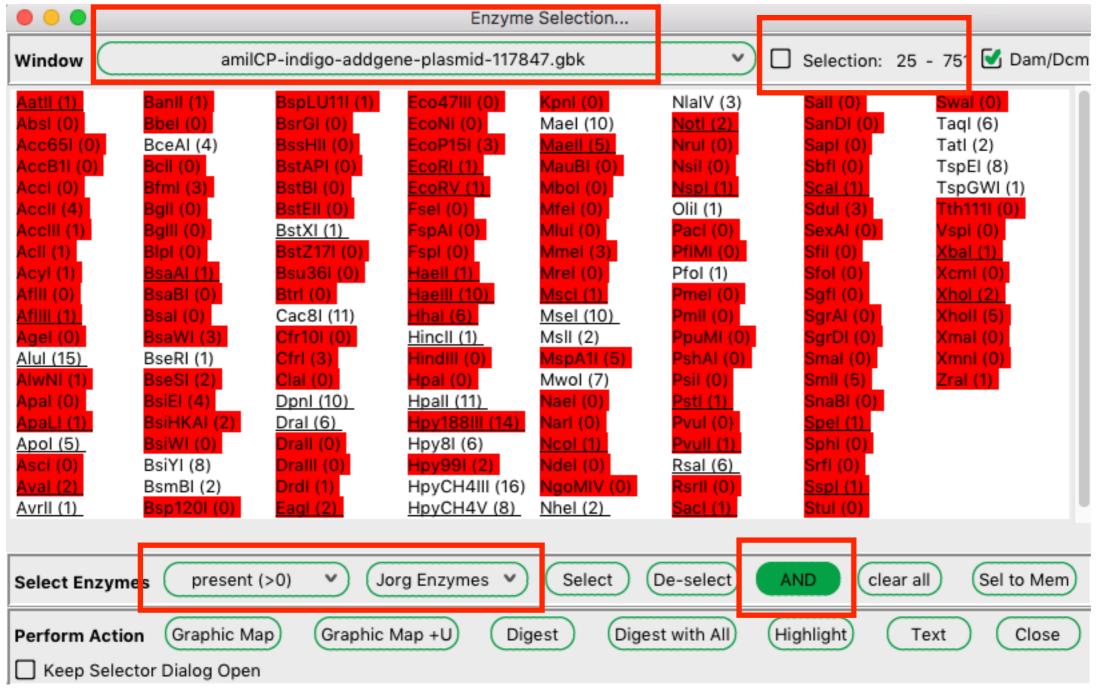
Select from 25 to 751 in amilCP

		ĥ	Sequence	e Start	Length	End	ORF	Tm	%GC	circular
	753		2800	25<0>	727<1>	751<0>	-/-		46%	🗹 Dam/Dcr
					FTASS	ULGTMLATRE	.*AV*NFHCTQ	TCGRL		
		F	eature		Direction	Туре		Lo	cation ↓	
▶ Hid	den									
▶ Bioł	Brick pre	efix			>>>	misc_featur	e		324	
con	stitutive	promot	er J23110		>>>	promoter		2	2559	
	-		osome bind	ling site (Elowitz	>>>	RBS		e	6879	
ami	ICP Base	Э			>>>	misc_featur	e	8	6751	
<u>۲</u>										
ш	*	10	* 20	* 30	* 40	* 50	* 60 *	70 *	80 *	90
1	taaatt									
91				agag <mark>tttacggct</mark> CCTACAAGGTTTA				ag <mark>aaagagga</mark>		
181										
361		•								
451										
541										
631				ACTATGTTGACCO						
721				IGGTCGCCTAA <mark>t</mark> a						
811				agattacttcgcg						
	cggtato	cagetea	actcaaagg	gcggtaatacggt	tatccacag	aatcaggggat	aacgcaggaaag	gaacatgtgag	caaaaggcca	agcaaa
991		772266	+	*****	********	acaggeteege	eccetaseas	restenciona	ateracete	coorte

 select all enzymes that are absent from the selection in amilCP

			Enzyme	e Selection					
Window	amil	CP-indigo-addge	ne-plasmid-1178	47.gbk	~	Selection:	25 - 751 🗹 Dam/Dcm		
Aatll (0)	Banll (0)	BspLU11I (0)	Eco47III (0)	Kpnl (0)	NlalV (1)	Sall (0)	Swal (0)		
Absl (0)	Bbel (0)	BsrGI (0)	EcoNI (0)	Mael (6)	Notl (0)	SanDI (0)	Taql (1)		
Acc65I (0)	BceAI (1)	BssHII (0)	EcoP15I (0)	Maell (0)	Nrul (0)	Sapl (0)	Tatl (1)		
AccB1I (0)	Bcll (0)	BstAPI (0)	EcoRI (0)	MauBI (0)	Nsil (0)	Sbfl (0)	TspEI (2)		
Accl (0)	Bfml (0)	BstBI (0)	EcoRV (0)	Mbol (0)	Nspl (0)	Scal (0)	TspGWI (1)		
Accll (0)	Bgll (0)	BstEll (0)	Fsel (0)	Mfel (0)	Olil (1)	Sdul (0)	Tth1111 (0)		
AccIII (0)	BgIII (0)	BstXI (1)	FspAl (0)	<u>Mlul (0)</u>	Pacl (0)	SexAl (0)	Vspl (0)		
Acll (0)	Blpl (0)	BstZ17I (0)	Fspl (0)	Mmel (0)	PfIMI (0)	Sfil (0)	Xbal (0)		
Acyl (0)	BsaAI (0)	Bsu36l (0)	Haell (0)	Mrel (0)	Pfol (1)	Sfol (0)	Xcml (0)		
AfIII (0)	BsaBI (0)	Btrl (0)	Haelll (0)	Mscl (0)	Pmel (0)	Sgfl (0)	Xhol (0)		
AfIIII (0)	Bsal (0)	Cac8I (3)	Hhal (0)	Msel (1)	Pmll (0)	SgrAl (0)	Xholl (0)		
Agel (0)	BsaWI (0)	Cfr10I (0)	Hincll (1)	MsII (1)	PpuMI (0)	SgrDI (0)	Xmal (0)		
Alul (3)	BseRI (1)	Cfrl (0)	HindIII (0)	MspA1I (0)	PshAI (0)	Smal (0)	XmnI (0)		
AlwNI (0)	BseSI (0)	Clal (0)	Hpal (0)	Mwol (1)	Psil (0)	Smll (0)	Zral (0)		
Apal (0)	BsiEI (0)	Dpnl (2)	Hpall (1)	Nael (0)	Pstl (0)	SnaBl (0)			
ApaLI (0)	BsiHKAI (0)	Dral (1)	Hpy188III (0)	Narl (0)	Pvul (0)	Spel (0)			
Apol (2)	BsiWI (0)	Drall (0)	Hpy8I (2)	Ncol (0)	Pvull (0)	SphI (0)			
Ascl (0)	BsiYI (3)	Dralll (0)	Hpy99I (0)	Ndel (0)	Rsal (4)	Srfl (0)			
Aval (0)	BsmBI (1)	Drdl (0)	HpyCH4III (6)	NgoMIV (0)	Rsrll (0)	Sspl (0)			
Avrll (1)	Bsp120I (0)	Eagl (0)	HpyCH4V (4)	Nhel (2)	Sacl (0)	Stul (0)			
Select Enzym	es absent	(0) 👻 🤇	All 🗸	Select	De-select	AND clea	ar all Sel to Mem		
Perform Actio	Perform Action Graphic Map Graphic Map +U Digest Digest with All Highlight Text Close								
Keep Selec	ctor Dialog Open	1							

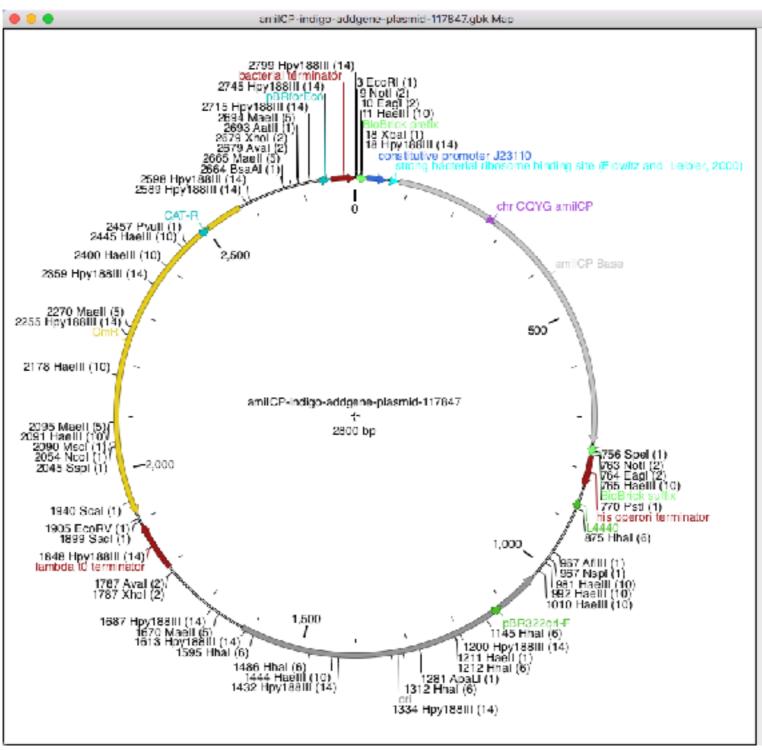
 select all enzymes that are ALSO present somewhere in the plasmid, and in the Jorg collection



• do a graphic map

•••			Enzyme	e Selection					
Window 🤇	amil	ne-plasmid-1178	v C] Selection: 2	25 - 751 🗹 Dam/Dcm				
Aatii (1)	Banll (1)	BspLU11I (1)	Eco47III (0)	Kpnl (0)	NlaIV (3)	Sall (0)	Swal (0)		
Absl (0)	Bbel (0)	BsrGI (0)	EcoNI (0)	Mael (10)	Notl (2)	SanDI (0)	Taql (6)		
Acc65I (0)	BceAI (4)	BssHII (0)	EcoP15I (3)	Maell (5)	Nrul (0)	Sapl (0)	Tatl (2)		
AccB1I (0)	Bcll (0)	BstAPI (0)	EcoRI (1)	MauBI (0)	Nsil (0)	SbfI (0)	TspEl (8)		
Accl (0)	Bfml (3)	BstBI (0)	EcoRV (1)	Mbol (0)	Nspl (1)	Scal (1)	TspGWI (1)		
AccII (4)	Bgll (0)	BstEll (0)	Fsel (0)	Mfel (0)	Olil (1)	Sdul (3)	Tth1111 (0)		
AccIII (1)	BgIII (0)	BstXI (1)	FspAI (0)	Mlul (0)	Pacl (0)	SexAI (0)	Vspl (0)		
Acll (1)	Blpl (0)	BstZ17I (0)	Fspl (0)	Mmel (3)	PfIMI (0)	Sfil (0)	Xbal (1)		
Acyl (1)	BsaAI (1)	Bsu36I (0)	Haell (1)	Mrel (0)	Pfol (1)	Sfol (0)	Xcml (0)		
AfIII (0)	BsaBI (0)	Btrl (0)	Haelll (10)	Mscl (1)	Pmel (0)	Sgfl (0)	Xhol (2)		
Afilii (1)	Bsal (0)	Cac8I (11)	Hhal (6)	Msel (10)	Pmll (0)	SgrAI (0)	Xholl (5)		
Agel (0)	BsaWI (3)	Cfr10I (0)	Hincll (1)	Msll (2)	PpuMI (0)	SgrDI (0)	Xmal (0)		
<u>Alul (15)</u>	BseRI (1)	Cfrl (3)	HindIII (0)	MspA1I (5)	PshAI (0)	Smal (0)	XmnI (0)		
AlwNI (1)	BseSI (2)	Clal (0)	Hpal (0)	Mwol (7)	Psil (0)	Smll (5)	Zral (1)		
Apal (0)	BsiEl (4)	Dpnl (10)	<u>Hpall (11)</u>	Nael (0)	Pstl (1)	SnaBl (0)			
ApaLI (1)	BsiHKAI (2)	Dral (6)	Hpy188III (14)	Narl (0)	Pvul (0)	Spel (1)			
Apol (5)	BsiWI (0)	Drall (0)	Hpy8I (6)	Ncol (1)	Pvull (1)	SphI (0)			
Ascl (0)	BsiYI (8)	Dralll (0)	Hpy99I (2)	Ndel (0)	Rsal (6)	Srfl (0)			
Aval (2)	BsmBl (2)	Drdl (1)	HpyCH4III (16)	NgoMIV (0)	Rsrll (0)	Sspl (1)			
Avrll (1)	Bsp120I (0)	Eagl (2)	HpyCH4V (8)	Nhel (2)	Sacl (1)	Stul (0)			
Select Enzymes present (>0) V Jorg Enzymes V Select De-select AND clear all Sel to Mem									
Perform Action Graphic Map Graphic Map +U Digest Digest with All Highlight Text Close									
Keep Selec	ctor Dialog Oper	n							

• do a graphic map

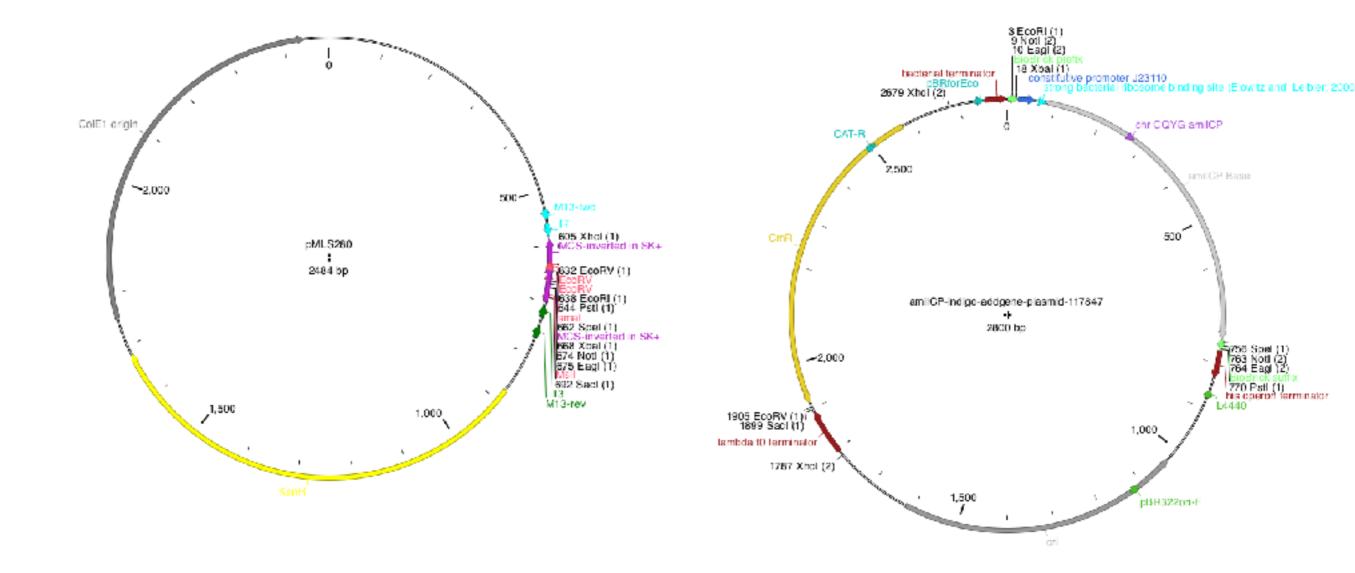


 select all enzymes that are ALSO unique in the pMLS MCS

			Enzyme	Selection					
Window 🤇		pMLS		~	🗹 Selection: 587	′ - 699 🗹 Dam/Dcm			
Aatil (0)	Banll (2)	BspLU11I (0)	Eco47III (0)	Kpnl (1)	NlalV (4)	Sall (1)	Swal (0)		
Absl (1)	Bbel (0)	BsrGI (0)	EcoNI (0)	Mael (2)	Notl (1)	SanDI (0)	Taql (4)		
Acc65I (1)	BceAI (0)	BssHll (0)	EcoP15I (0)	Maell (0)	Nrul (0)	Sapl (0)	Tatl (0)		
AccB1I (1)	Bcll (0)	BstAPI (0)	EcoRI (1)	MauBI (0)	Nsil (0)	Sbfl (0)	TspEl (1)		
Accl (1)	<u>Bfml (1)</u>	BstBI (0)	EcoRV (1)	Mbol (0)	Nspl (0)	Scal (0)	TspGWI (0)		
Accll (1)	Bgll (0)	BstEll (0)	Fsel (0)	Mfel (0)	Olil (1)	Sdul (2)	Tth1111 (0)		
AccIII (0)	BgIII (0)	BstXI (1)	FspAI (0)	Mlul (0)	Pacl (0)	SexAI (0)	Vspl (0)		
Acll (0)	Blpl (0)	BstZ17I (0)	Fspl (0)	Mmel (0)	PfIMI (0)	Sfil (0)	Xbal (1)		
Acyl (0)	BsaAI (0)	Bsu36I (0)	Haell (0)	Mrel (0)	Pfol (0)	Sfol (0)	Xcml (0)		
AfIII (0)	BsaBI (0)	Btrl (0)	Haelll (2)	Mscl (0)	Pmel (0)	Sgfl (0)	Xhol (1)		
Aflill (0)	Bsal (0)	Cac8I (0)	Hhal (0)	Msel (0)	Pmll (0)	SgrAI (0)	Xholl (1)		
Agel (0)	BsaWI (0)	Cfr10I (0)	Hincll (1)	Msll (1)	PpuMI (0)	SgrDI (0)	Xmal (1)		
Alul (3)	BseRI (0)	<u>Cfrl (1)</u>	HindIII (1)	MspA1I (1)	PshAI (0)	Smal (1)	XmnI (0)		
AlwNI (0)	BseSI (1)	<u>Clal (1)</u>	Hpal (0)	Mwol (1)	Psil (0)	Smll (1)	Zral (0)		
<u>Apal (1)</u>	BsiEl (1)	Dpnl (1)	Hpall (2)	Nael (0)	Pstl (1)	SnaBl (0)			
ApaLI (0)	BsiHKAI (1)	Dral (0)	Hpy188III (1)	Narl (0)	Pvul (0)	Spel (1)			
Apol (1)	BsiWI (0)	Drall (1)	Hpy8I (1)	Ncol (0)	Pvull (0)	SphI (0)			
Ascl (0)	BsiYI (1)	Dralll (0)	Hpy99I (1)	Ndel (0)	<u>Rsal (1)</u>	Srfl (0)			
Aval (2)	BsmBI (0)	Drdl (0)	HpyCH4III (1)	NgoMIV (0)	Rsrll (0)	Sspl (0)			
Avrll (0)	Bsp120I (1)	Eagl (1)	HpyCH4V (1)	Nhel (0)	Sacl (1)	Stul (0)			
				-					
Select Enzym	es uniqu	e (1) 🔹	All 🗸	Select	De-select	AND clear	all Sel to Mem		
Perform Action Graphic Map Graphic Map +U Digest Digest with All Highlight Text Close									
Keep Selec	tor Dialog Ope	n							

select all enzymes that are ALSO unique in pMLS

			Enzyme	e Selection					
Window C		pMLS	280.ape		~	Selection: 587	7 - 699 🗹 Dam/Dcm		
Aatll (0)	Banll (3)	BspLU11I (1)	Eco47III (0)	Kpnl (1)	NlalV (5)	Sall (1)	Swal (0)		
Absl (1)	Bbel (0)	BsrGI (0)	EcoNI (1)	Mael (5)	Notl (1)	SanDI (0)	Taql (8)		
Acc65I (1)	BceAI (5)	BssHII (2)	EcoP15I (3)	Maell (4)	Nrul (1)	Sapl (1)	Tatl (0)		
AccB1I (1)	Bcll (0)	BstAPI (0)	EcoRI (1)	MauBI (0)	Nsil (2)	SbfI (0)	TspEI (11)		
Accl (1)	Bfml (4)	BstBI (0)	EcoRV (1)	Mbol (0)	Nspl (1)	Scal (0)	TspGWI (3)		
Accll (14)	Bgll (0)	BstEll (0)	Fsel (0)	Mfel (0)	Olil (1)	Sdul (4)	Tth1111 (0)		
AccIII (0)	BgIII (0)	BstXI (1)	FspAI (0)	Mlul (2)	Pacl (0)	SexAI (0)	Vspl (3)		
Acll (1)	Blpl (0)	BstZ17I (0)	Fspl (0)	Mmel (7)	PfIMI (1)	Sfil (0)	Xbal (1)		
Acyl (1)	BsaAI (0)	Bsu36I (0)	Haell (2)	Mrel (0)	Pfol (0)	Sfol (0)	Xcml (0)		
AfIII (0)	BsaBI (0)	Btrl (0)	Haelll (15)	Mscl (0)	Pmel (0)	Sgfl (1)	Xhol (1)		
AfIIII (3)	Bsal (0)	Cac8I (16)	Hhal (18)	Msel (9)	Pmll (0)	SgrAI (0)	Xholl (4)		
Agel (0)	BsaWI (3)	Cfr10I (1)	Hincll (2)	MsII (1)	PpuMI (0)	SgrDI (0)	<u>Xmal (1)</u>		
Alul (12)	BseRI (0)	Cfrl (3)	HindIII (1)	MspA1I (4)	PshAI (0)	Smal (1)	XmnI (0)		
AlwNI (1)	BseSI (2)	<u>Clal (1)</u>	<u>Hpal (1)</u>	Mwol (11)	Psil (0)	Smll (4)	Zral (0)		
<u>Apal (1)</u>	BsiEl (4)	Dpnl (8)	Hpall (11)	Nael (0)	Pstl (1)	SnaBl (0)			
ApaLI (1)	BsiHKAI (2)	Dral (0)	Hpy188III (17)	Narl (0)	Pvul (1)	Spel (1)			
Apol (3)	BsiWI (0)	Drall (1)	Hpy8I (4)	Ncol (0)	Pvull (1)	SphI (0)			
Ascl (0)	BsiYI (10)	Dralll (0)	Hpy99I (6)	Ndel (0)	Rsal (3)	Srfl (0)			
Aval (2)	BsmBl (1)	<u>Drdl (1)</u>	HpyCH4III (7)	NgoMIV (0)	Rsrll (0)	Sspl (1)			
Avrll (0)	Bsp120I (1)	Eagl (1)	HpyCH4V (9)	Nhel (2)	Sacl (1)	Stul (0)			
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Preferences dialog

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Preferences dialog

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Preterences
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Change Directory)
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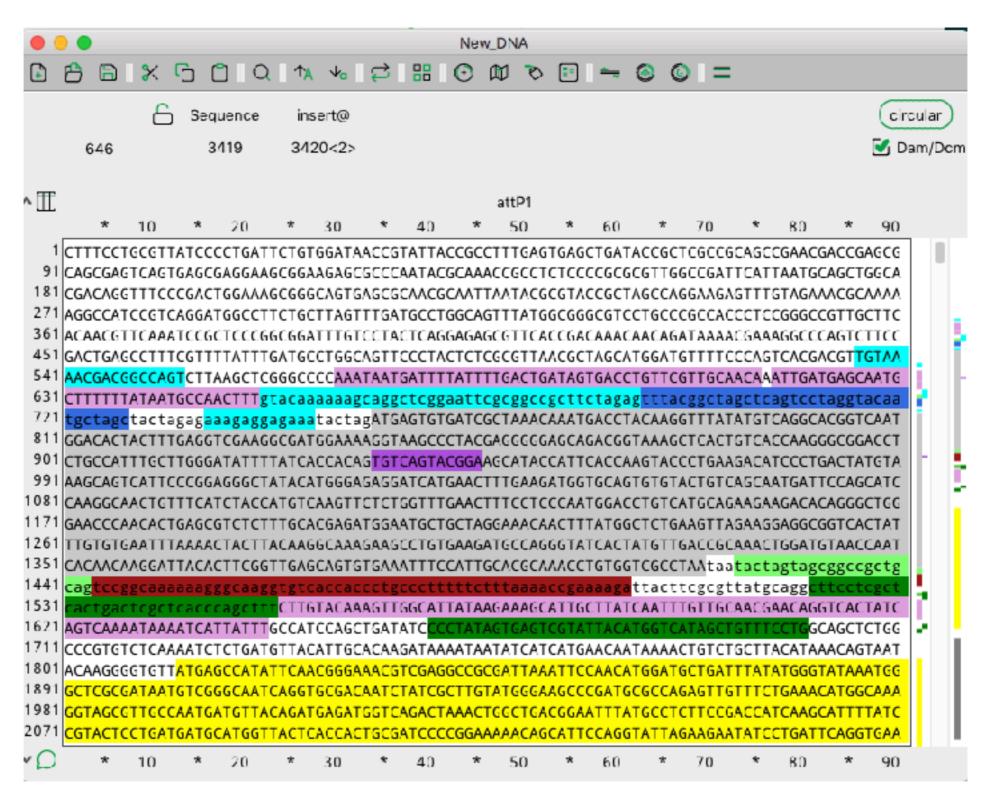
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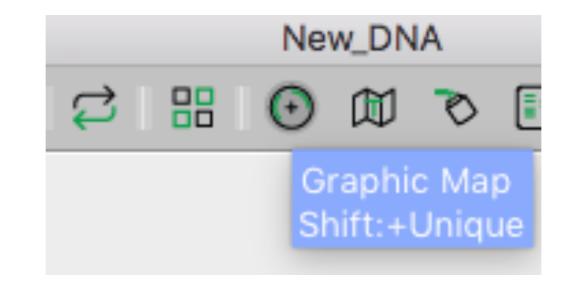
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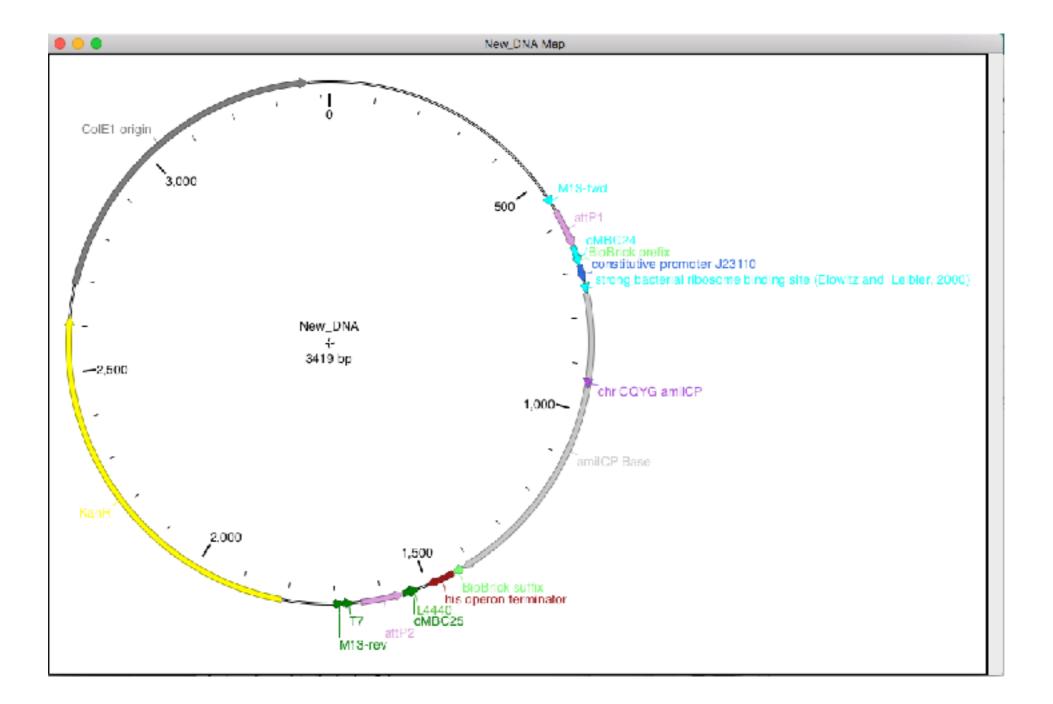
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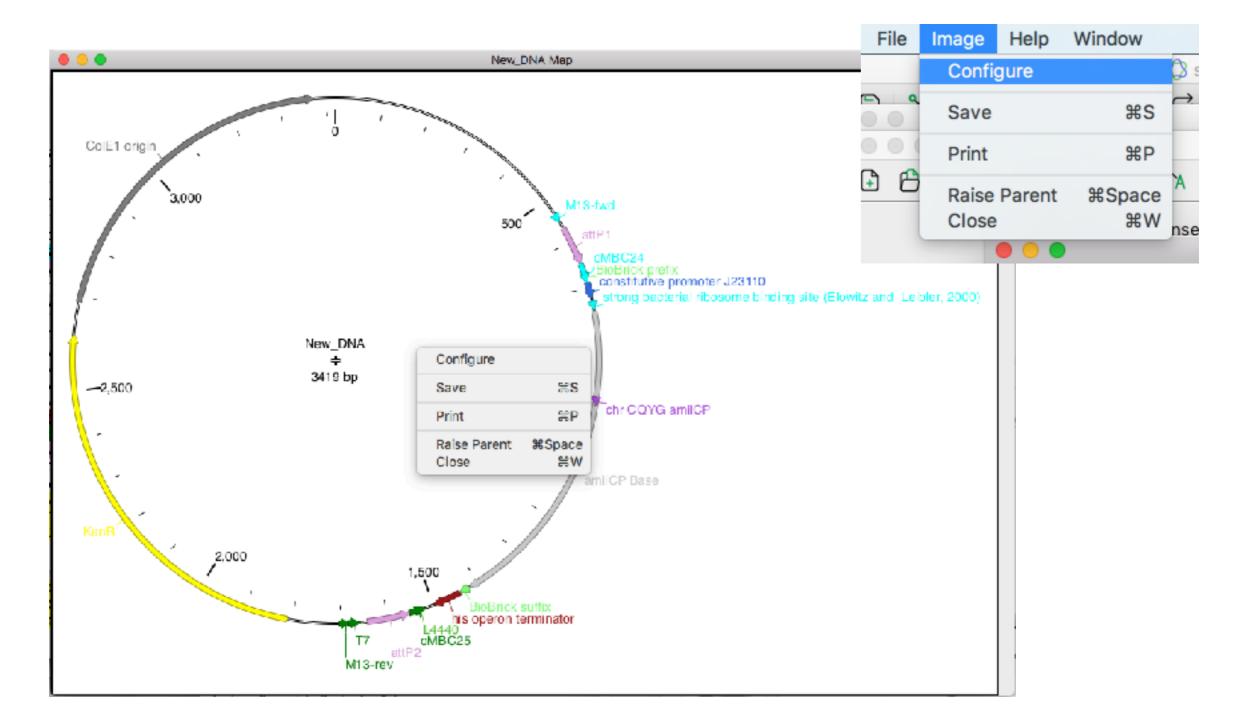


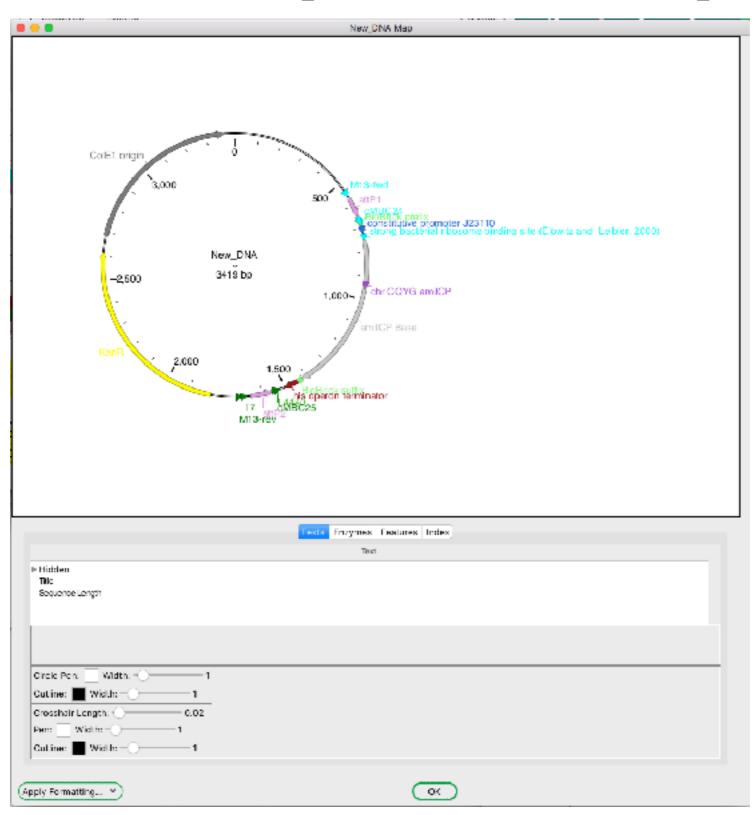
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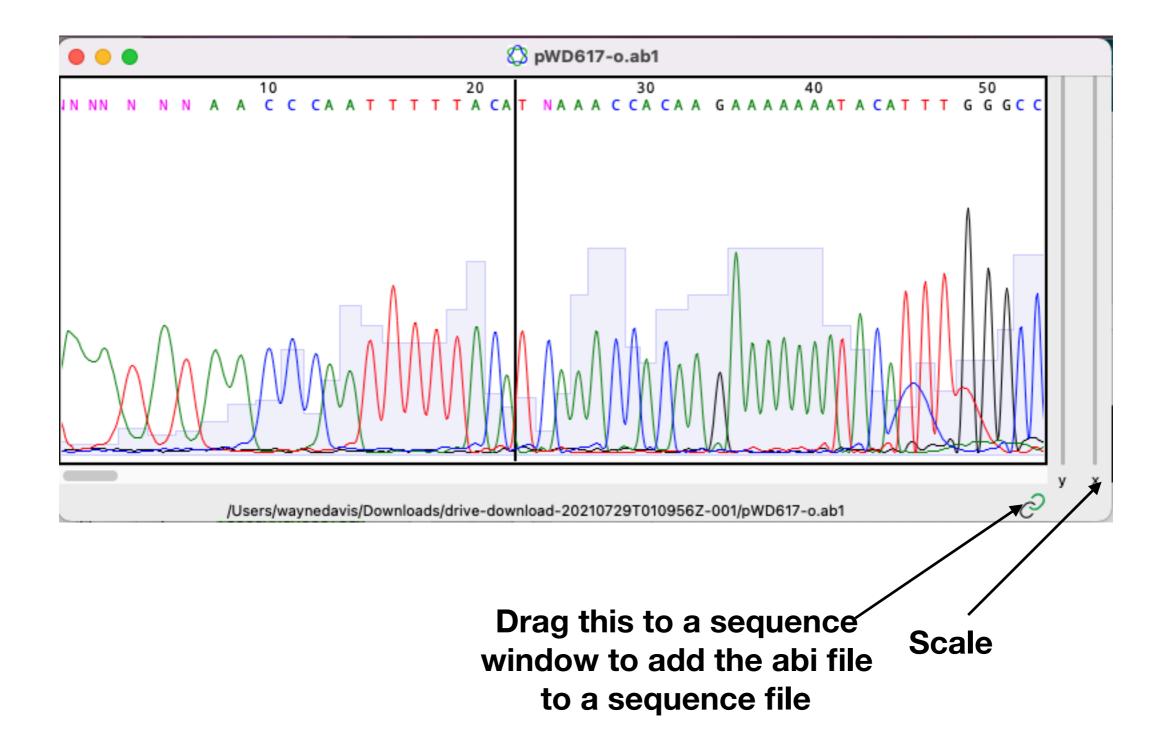


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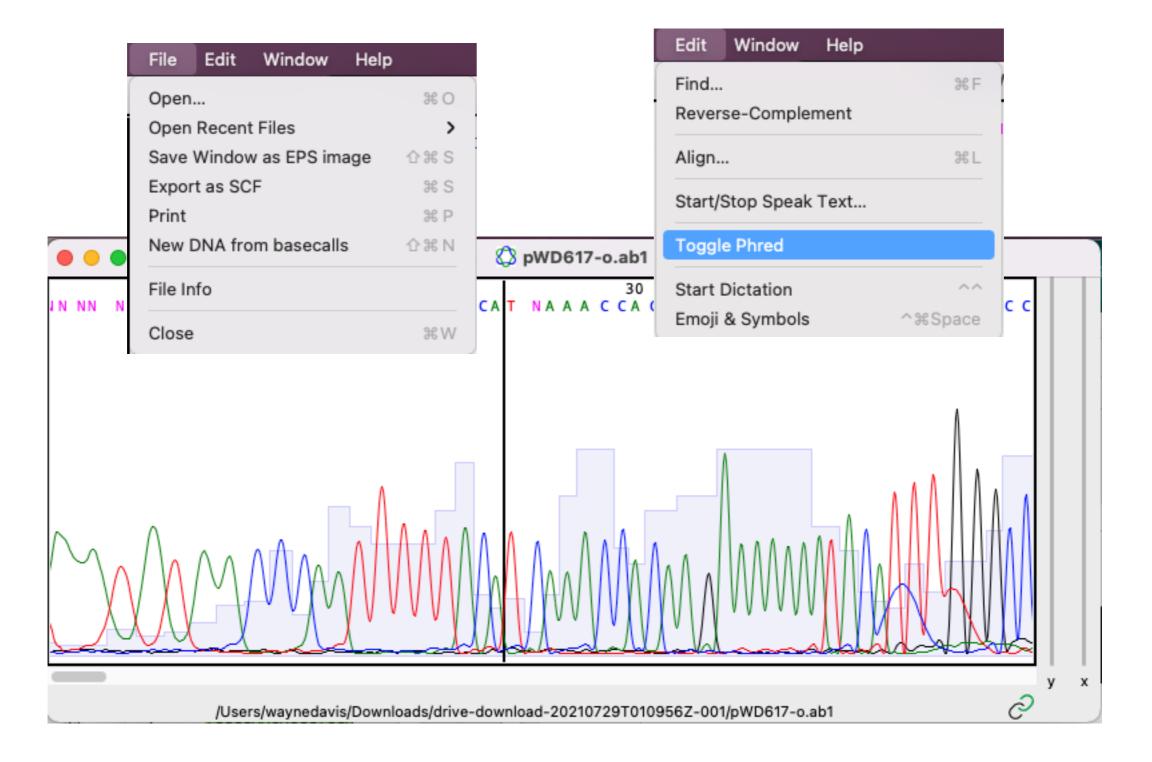
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ABI files



ABI files



Tools	Window Help	
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e e e Align	DNA		
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pMLS280.ape	All	Forward	
pCFJ104 - EXP[pMYO-3_wormmCherry_unc-54].str	All	Forward	
pWD602 miniMos Pvit-2cyOFP Phsp_mosase_SL2GFP.ape	All	Forward	
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