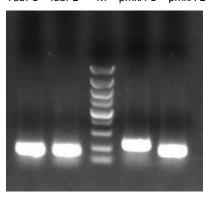


fabl 基因及 pmxA 基因 5'race 实验进展报告

合同编号: PG1-1601001

- 1. 根据 fabl 基因及 pmxA 基因序列,我们分别设计两对引物,依据引物进行 RT-PCR,
- 2. 以提取的 RNA 逆转录后的 cDNA 为模板,进行 RT-PCR,电泳检测结果如下:





3. 并将测序结果进行比对,比对结果如下:

Paenibacillus polymyxa SQR-21, complete genome Sequence ID: gb|CP006872.1| Length: 5828436 Number of Matches: 1

Range 1: 81030 to 81202 GenBank Graphics ▼ Next Match ▲ Previous Match								A Previous Match
Score 283 b	e oits(15)	3)	Expect 7e-73	Identities 169/176(969	Gaps %) 4/176(2%)	Strand Plus/Plus	
Query	16	GGCTGCGT	CTCT-CGTACTT	TTGGCCGCCTTCTGG(CATTCAAGATTTTAACACAA	TGCT 74		
Sbjet	81030	GGCTG-GT	CTATACGTAC-T	TTGGCCGCC-TATGG	CATTAAAGATTTTAACACAA	TGCT 8108	6	
Query	75	AAAGCATA'	TTGAAGAAACTAC	GCCCTACGCAAGAC	GTCGATACGTCTGAAGTCG	GAGA 134		
Sbjet	81087	AAAGCATA	TTGAAGAAACTAC	GCCCTACGCAAGAC	GTCGATACGTCTGAAGTCG	GAGA 8114	6	
Query	135	TACGGCGT	FATTCCTATTTTC	CCATTTATCGAGAGGG	CATTACGGGTGAAGTTATCC	190		
Sbjet	81147	TACGGCGT	TATTCCTATTTTC	CCATTTATCGAGAGG(CATTACGGGTGAAGTTATCC	81202		

Paenibacillus polymyxa SQR-21, complete genome

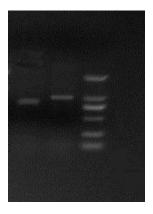
Sequence ID: gb|CP006872.1| Length: 5828436 Number of Matches: 1

Range	e 1: 506	1826 to 5	062012	GenBank	Graphics		▼ Next Match ▲ Previ	ous Match
Score 335 b	e oits(181)		cpect e-88	Identitie: 186/188(Gaps 2/188(1%)	Strand Plus/Plus	
Query	14	GGA-GCGCTGC	CGCCACTTTCC	TCCGAAAGCG	TTCCACCGACGGTTG	TCATAATGGGCGTC	72	
Sbjet	5061826	GGACGCGCTGC	GCCAC-TTCC	TCCGAAAGCG	TTCCACCGACGGTTG	TCATAATGGGCGTC	5061884	
Query	73	AGGGAGCTTGG	GAGCTTTGCTG	TAAGGTAGCC	GCGTCAAGGTATAAT	CGTCGCTACCGAAT	132	
Sbjet	5061885	AGGGAGCTTGG	GAGCTTTGCTG	TAAGGTAGCC	GCGTCAAGGTATAAT	CGTCGCTACCGAAT	5061944	
Query	133	TTTTCGTTCC	AAAACAACGTT	TCTTTTTCAA	AAGCCATCTGGATCG	TTCCCCTCTCTTCA	192	
Sbjet	5061945	TTTTCGTTCC	AAAACAACGTT	TCTTTTTCAA	AAGCCATCTGGATCG	TTCCCCTCTCTCA	5062004	
Query	193	TCGGTAAA 2	200					
Sbjet	5062005	TCGGTAAA 5	5062012					

比对结果正确,根据 RACE 试剂盒进行两个基因 5′端的 PCR 扩增, 扩增结果如下:



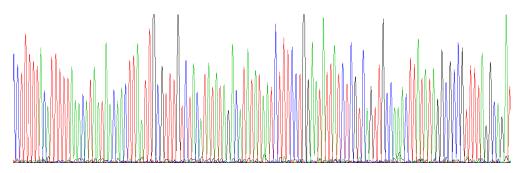
fabl pmxA M



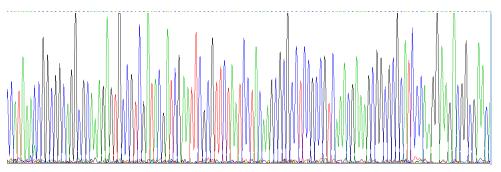
注: Marker 大小采用 DL2000,从上到下大小依次为: 2000、1000、750、500、250、100bp

5. 根据 RACE 扩增结果,凝胶电泳检测结果可以看到扩增出条带大小在 1K 左右的片段,将片段进行切胶 回收送测序,测序峰图结果如下:

FabI:



PmxA:



6. 根据测序结果在 NCBI 上进行比对,比对结果如下:

FabI:

Scor 1203	e bits(6:	51)	Expect 0.0	Identities 666/673(99%)	Gaps 1/673(0%)	Strand Plus/Minus
Query	8	CAGTGCATC	PATTTCTTCATCG	ctctgtacattgcaaggacag	ĄÇĄĄŢĢĢĄĄÇÇÇĢĢĄĄŢ	67
Sbjet	80691	CAGTGCATC	caattetteateg		ACAATGGAACCCGGAAT	80632
Query	68	GGTTTCAGC	PAGCTTTAGCACC	CGTTCTGCAACCCTCTCATTT	rcgtaagtaaaaatgag	127
Sbjet	80631			cottetecaacceteteattt		80572
Query	128	ATTTGCTCC	TTCCGCAGCTAAA	GATTGGGCTGTCCCCCAGGCA.	ATACTCCTGTTATTAGC	187
Sbjet	80571	ATTTGCTCC	rteegeagetaaa	ĠĂŦŦĠĠĠĊŦĠŦĊĊĊĊĊĀĠĠĊĀ.	ATACTCCTGTTATTAGC	80512
Query	188	CACACCCATA	AATCAAAATGTTC	TTTCCACTCAGTAGTACTGCC	ACAGAAACCACCTTTCC	247
Sbjet	80511	ĊÁĊÁĊĊĊÁŤ.	áátcáááátáttc	fffédádfóagfágfádfgód.	ácagaaaccacctttcc	80452
Query	248	TTTTTAGAT	TTTTAAACATAAT	AACAACTTAATTGCGTTTGTC	TACATATATAGACATAT	307
Sbjet	80451	ŤŤŤŤŤÁĊÁŤ	řŤŤŤÁÁÁČÁŤÁÁŤ.	áácáácttááttécétttéte:	racatatatagacatat	80392
Query	308	414415111	CTGGAATATTAT	CTCGTCAGTTGCCAAACTTAT	ATAGGCGCCGTTTTAGG	367
Sbjet	80391	ATAATCTTT	ÉCTGGÁÁTÁTTÁT:	ĊŤĊĠŤĊĂĠŤŤĠĊĊĂĂĂĊŤŤĂŤ.	ÁTÁGGCGCCGTTTÁGG	80332
Query	368	CAGATGGCA	AGAATATTTTAAA	aattgtgaatgtggtttctgt: 	rggtctgtgttcaaatc	427
Sbjet	80331	ĊÁĠÁŤĠĠĊÁ	ágáátáttttááá	ááttgtgáátgtggtttótgt:	főGTCTGTGTTCÁÁÁTC	80272
Query	428			aaagtggtgaggcaacgata 		487
Sbjet	80271	GÁÁÁGGÁGT	CATGCGTGTGAA	aaaagtggtgaggcaacgata.	ágátgáctgtcááágcg	80212
Query	488	ATTTTAATA:	FAAAAATATCATA	CTCTTATAAATATTTCCATTA	FGACGATAGACTCATCC	547
Sbjet	80211	ATTTTAATA	FAAAAATATCATA	CTCTTATAAATATTTCCATTA:	rgacgatagactcatcc	80152
Query	548	CACTATEGA	AAGTAACCACTTC	ATTTCACCGTCAATGACTCTC	CGATTTGAACATCCTG	607
Sbjet	80151			átttóácógtóáátgáctótó:		80092
Query	608	T11111111	\cdots	CTTCCTCTTGGCTAAGCCTT		667
Sbjet	80091	ATTCGTTTC	ÁÁGCTTGÁGGCTC	fcttcctcttggctaagcctt.	átgtítátgtíttttág	80032
Duerr	668	GAG-TGAAG	FATA 679			



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PmxA:

Scor 1291	e bits(699	Expe) 0.0		ntities /703(99%)	Ga 1/7	ps '03(0%)	Strand Plus/Minus
Query	14	GGATTATCCATTTC	GAGGAGCTCGTG	GAGCGTTTGAAT	TGAAGCGGGA	gccgggccgct	73
Sbjet	5047487	ggattateca-tte	gaggagetegtg	gagegtttgaat:	stgaageggga	secsssecser	5047429
Query	74	TCCCGCTGTTCGAT	GCCGTTTTCGAC	TTGCAAAATATCG	PAAGAACGAGA	CGCCGAGCTGG	133
Sbjet	5047428	tccccctctccat	dccdttttcdac	ttdcaaaatatc	3AAGAACGAGA	cáccáláctás	5047369
Query	134	AAGGGGTCAGCCTG	*******	97799999777	***	GTTCGATCTGA	193
Sbjet	5047368	AAGGGGTCAGCCTG	aagacttacgag	ċttgaċċattta:	3AAGAAGCGAA	gttegatetga	5047309
Query	194	CACTGTTTATGTAT	GAAAACAACGGG	GCGCTGAGTGGG	PACTICITATA	CGCCACCAAGC	253
Sbjet	5047308	57545444749474	ĠĂĂĂĂĠĂĂĠĠĠĠ	dedetgadtgad	scttetteta	egeékéekkée	5047249
Query	254	TGTTCAAAGAAGCG	ATGATCCGCACC	TTGACCGAGGATT	PACCTGCGGGT	ACTOTCTCAAA	313
Sbjet	5047248	19115	átgátccscácác	ttgáccgággát:	racctdcdddt	áctotctáká	5047189
Query	314	TTGCGGAAAATCCA	544575546574	4955994TT944T	7266676767	GGCGGCAGGCG	373
Sbjet	5047188	ŤŤĠĊĠĠÁÁÁÁŤĊĊÁ	cááctcsásctá	ágccggáttgáá:	řáccátááácc	dddddd Adddd	5047129
Query	374	CAAAGAGTGCCGTC	GATACGATCGAA	TTCGCGTTCTAAT	CCTACAAGCG	CGCCTCCGCCG	433
Sbjet	5047128	5YYYYYYYYYY	gatacgategaa	ttégégítétáá:	rcctacaacc	édééréédééd	5047069
Query	434	TCAAACGAAAGCGG	GGGGGATGGGAA	GGAAGACGGCGA/	ትናትትናናፕናናትት	GCCCGTAAACG	493
Sbjet	5047068	ŤĊÁÁÁĊĠÁÁÁĠĊĠĠ	ċĠċĠċÁŦċċċÁÁ	ġġġġġġġġġġ	ÁCÁÁCGTCCÁÁ	ścccśtaaacs	5047009
Query	494	CGCAGGCCGAAAGC	^^ 95777777699	ACCTGCGCGGG	PEATGEGGTEA	?	553
Sbjet	5047008	égé ággéég á á ágé	áásctttttéss	Acctdccccccc	ścatścśctca	cttcátttttá.	5046949
Greth	554	GGGGAGGTACGAAT	GAAATSTTTGTT	TGAAAAGGAAGA	CGGTACTGGA	GCGGCAAGTTT	613
Sbjet	5046948	ddddaddtacdaat	gaaatetttätt	tgaaaaggaaga	acaatactada	ácáácáááttt	5046889
Query	614	GACGCCGATGACAG	9979 1 9977997	T555T4545T54	ነፕናናፕናናስስስፕ	777566666646	673
Sbjet	5046888	ĠÁĊĠĊĊĠÁŤĠÁĊÁĠ					5046829

7. 下一步进行克隆测序,等待克隆测序结果。