武汉植物学研究 2007,25(3):282~288
Journal of Wuhan Botanical Research

番茄 P56 和部分来源多糖裂解酶家族 I 的其它蛋白的系统演化分析

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摘 要:果胶裂解酶包含果胶酸裂解酶(pectate lyase)和果胶酸酯裂解酶(pectin lyase)二种形式,是一类多糖裂解酶,由细菌、真菌、植物和线虫等生物产生,分布在5个多糖裂解酶家族,果胶酶在食品与饮料、纺织与洗涤、制药等工业中有广泛的应用。利用 NCBI BLASTX 服务器,搜索与番茄 P56 蛋白氨基酸序列相似且都属于多糖裂解酶家族 I 的果胶裂解酶蛋白序列共28条,用 DNAMAN 软件分析其保守区,用 CLUSTALX8.0 软件进行序列比对和 BioEdit 软件进行文件转换,进一步用 PUAP4.0 软件构建系统进化树。构建的 MP 树(phylogenetic trees)和 NJ 树(neighbor-joining)显示:来自植物的果胶酸裂解酶、真菌的果胶酸酯裂解酶、细菌的果胶酸裂解酶分别可聚为一个独立的类群;相对于细菌果胶酸裂解酶和真菌果胶酸酯裂解酶而言,构巢曲霉(*Aspergillus nidulans*)的果胶酸裂解酶 A 和植物的果胶酸裂解酶之间有较近的亲缘关系;细菌 *Pseudomonas syringae* 的果胶酸酯裂解酶和真菌的果胶酸酯裂解酶之间的亲缘关系较近。

关键词:果胶酸裂解酶;果胶酸酯裂解酶;多糖裂解酶家族 I;系统演化树
中图分类号:Q55 **文献标识码**:A **文章编号**:1000-470X(2007)03-0282-07

Phylogenetic Analysis of Tomato P56 and Some Other Proteins from Polysaccharide Lyase Famliy I

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Abstract: Pectate and pectin lyases are a class of polysaccharide lyases produced by a diverse group of organisms including bacteria, fungi, plant and nematodes. Various pectate lyases and pectin lyases are divided into five families. By NCBI BLASTX server, tomato P56 amino-acid sequence was compared with other pectate and pectin lyases belonging to polysaccharide lyase family I 28 proteins, with higher identity of amino acid sequence compared to P56 amino-acid sequence, were obtained. On the base of the constructing phylogenetic tree using PAUP 4.0, MP (phylogenetic trees) and NJ (neighbor-joining) trees of diverse pectate and pectin lyases showed that pectate lyases from plant, pectin lyases from fungi and pectate lyases from bateria belong to different groups, respectively. The pectate lyase A of *Aspergillus nidulans* is more closely related to plant pectate lyases than to pectin lyase from fungi and pectate lyases from bacteria. The pectin lyase from *Pseudomonas syringae* is more closely related to fungi pectin lyases than other lyases.

Key words: Pectate lyase; Pectin lyase; Polysaccharide lyase family I; Phylogenetic tree

果胶酸裂解酶(pectate lyase, EC 4.2.2.2)和果胶酸酯裂解酶(pectin lyase, EC 4.2.2.10)是一类多糖裂解酶,由细菌[1-6]、真菌[7-15]、植物[16-23]和线虫[24,25]等生物产生。真菌来源的果胶裂解酶以果胶酸酯裂解酶为主,果胶酸裂解酶极少[7-15];细菌来源的果胶裂解酶以果胶酸裂解酶为主,果胶酸酯裂解酶极少[1-6];植物来源的果胶裂解酶为果胶酸裂解酶[16-23]。

收稿日期:2006-12-04,修回日期:2007-03-29。
基金项目:国家自然科学基金资助项目(30170005);江苏省教育厅高校自然科学指导性项目(01KJD18005);盐城师范学院博士科研启动基金。
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果胶酸裂解酶和果胶酸酯裂解酶分布于多糖裂解酶家族Ⅰ、Ⅱ、Ⅲ、Ⅸ、Ⅹ等5个家族(polysaccharides family Ⅰ:262 proteins; family Ⅱ:19 proteins; family Ⅲ:72 proteins; family Ⅸ:43 proteins; family Ⅹ:49 proteins),目前报道的植物来源果胶酸裂解酶属于多糖裂解酶家族Ⅰ(http://afmb.cns.fr/CAZY/fam/PL1.html),真菌和细菌果胶裂解酶分别属于不同的家族。果胶酸裂解酶和果胶酸酯裂解酶能够以β-消旋法裂解植物初生细胞壁和植物果胶中果胶酸和果胶酸酯的半乳糖醛羧残基之间的α-1,4-糖苷键,个别果胶酸裂解酶和果胶酸酯裂解酶的空间结构研究已有报道[26]。

1962年,果胶酸裂解酶首次发现是在*Ewinia carotovora*和*Bacillus* sp.[27]的培养基中。至今,研究最为详细的果胶酸裂解酶来自*Erwinia chrysanthemi*[28-36],*Erwinia chrysanthemi*能够分泌5种相互独立调控的果胶酸裂解酶(pelA,pelB,pelC,pelD和pelE)[31,32]。

1989年,来自植物的果胶酸裂解酶类似序列P56首次在蕃茄花粉中被发现[37],自那以后,很多类似序列在其它植物的花粉、花粉管、花柱道等组织中不断被发现[38]。一种存在于日本柚木花粉中的致敏源具有果胶酸裂解酶活性[39];百合维管束分化过程中,有二种果胶酸裂解酶被分离和鉴定[40,41],进一步研究表明酶的表达受到生长素的调控,是在维管束分化的早期表达的[40];在香蕉[42]、草莓[43]和葡萄[44]等水果中,都有果胶酸裂解酶表达的报道。有关研究结果暗示果胶酸裂解酶在花粉细胞壁扩张,花粉管萌发,花柱道组织细胞壁的降解等方面可能起了十分重要的作用[39,45],然而,适当浓度的微生物来源的商品果胶酶也能够激发花粉的萌发和促进花粉管的生长,高浓度的商品果胶酶能够使花柱顶部细胞的细胞壁膨胀,引起花粉管细胞壁硬度下降[36],但商品酶制品中含有很多杂酶,果胶酶作用机理需要进一步确认。

多糖裂解酶家族Ⅰ的系统演化研究可以揭示不同来源的果胶裂解酶相互间的亲缘关系,为果胶裂解酶的如空间结构、酶学性质、催化机理、生理等方面研究奠定基础。多糖裂解酶家族Ⅰ果胶裂解酶成熟蛋白的氨基酸序列之间相似性较低,我们利用多糖裂解酶家族Ⅰ的果胶裂解酶的保守区序列对多糖裂解酶家族Ⅰ的系统演化进行研究。

1　方法

1.1　蛋白序列

根据NCBI(national center for biotechnology information,http://www.ncbi.nlm.nih.gov/)数据库,搜索与番茄P56蛋白具有较近亲缘关系的植物果胶酸裂解酶蛋白序列、真菌果胶裂解酶和细菌果胶裂解酶。

1.2　果胶裂解酶保守区氨基酸序列和系统演化分析

用DNAMAN软件(Lynnon BioSoft, Quebec)对待分析的蛋白氨基酸序列进行多重比对,得到各种酶序列相似性较高的氨基酸序列片段,确定果胶裂解酶保守区氨基酸序列;再用CLUSTALX1.83(http://www-igbmc.u-strasbg.fr/BioInfo/ClustalX/)对各蛋白的保守区氨基酸序列进行序列比对分析[25],用BioEdit(www.mbio.ncsu.edu/BioEdit/bioedit)将CLUSTALX1.83文件转换为PAUP文件,按照最大似然法和邻接法的计算方法,用PAUP4.0b(Swofford. 2002)分别构建MP树(phylogenetic trees)和NJ树(neighbor-joining)[25],来自Globodera rostochiensis Ro-1 Mierenbos的果胶酸裂解酶pectate lyase 1和pectate lyase 2作为外群。

2　结果

2.1　氨基酸序列的比较

得到的具有较近亲缘关系的28条多糖裂解酶家族Ⅰ的蛋白物种来源和其NCBI蛋白库的蛋白序列号见表1。28条蛋白中有12条来自真菌的果胶酸酯裂解酶、1条来自真菌的果胶酸裂解酶、1条来自细菌的果胶酸酯裂解酶、5条来自细菌的果胶酸裂解酶和10条来自植物的果胶酸裂解酶。表1中多糖裂解酶家族Ⅰ的植物、真菌、细菌等生物来源的裂解酶氨基酸序列相似性不到26%,但其保守区氨基酸序列相似性在96%~29%(图1),保守区序列如Ala-X-Asp-Ile-Lys-Gly-4X-Thr-X-Ser和Val2-X-Arg-X-Pro-2X-Arg-X-Gly-2X-His-3X-Asn等序列其保守性强,相似性高,在酶的催化和构象维持方面可能有重要的作用[46]。

2.2　MP树(Phylogenetic trees)

用PAUP 4.0b软件,以来自*Glomerella cingulata*的2个果胶酸裂解酶作为外群,用28条来自植物、真菌和细菌等生物的果胶裂解酶保守区氨基酸序列构建的MP树见图2。来自植物的果胶酸裂解酶真菌的果胶酸酯裂解酶和细菌的果胶酸裂解酶聚成了3个类群(图2:A_1,A_2)。第一个类群为真菌的果胶酸酯裂解酶,包含10个真菌来源的果胶酸酯裂解酶(*Aspergillus niger*, *A. oryzae*, *Penicillium expansum*, *P. griseoroseum*, *Colletotrichum gloeosporioides* f. sp.),

表1 不同来源的部分果胶酸裂解酶和果胶酸酯裂解酶

Table 1 Proteins used for the construction of the protein sequence alignments

NCBI accession number	Preotein	Organism	Refference
NP-241564	pectate lyase	*Bacillus halodurans* C-125	1
NP-794039	pectin lyase	*Pseudomonas syringae* pv. *tomato* DC3000	2
P18210	Pectate lyase E precursor	*Erwinia chrysanthemi*	3
P29155	Pectate lyase A	*Erwinia chrysanthemi*	4
CAC38815	secreted pectate lyase	*Streptomyces coelicolor* A3(2)	5
JC6502	pectate lyase	*Pseudonocardia* sp.	6
S23573	pectin lyase B	*Aspergillus niger*	7
1IDK	Pectin Lyase A	*Aspergillus niger*	8
P22864	Pectin lyase D precursor (PLD)	*Aspergillus niger*	9
CAD34589	pectine lyase F	*Aspergillus niger*	10
BAB82468	pecyin lyase 2	*Aspergillus oryzae*	11
BAB82467	pecyin lyase 1	*Aspergillus oryzae*	12
Q00645	Pectate lyase A precursor	*Aspergillus nidulans*	13
AAS55382	pectin lyase	*Penicillium expansum*	Unpublished
AAM23008	pectin lyase	*Penicillium expansum*	Unpublished
AAM23009	pectin lyase	*Penicillium griseoroseum*	Unpublished
AAD43565	pectin lyase 2	*Colletotrichum gloeosporioides* f. sp. *malvae*	14
AAF22244	pectin lyase	*Colletotrichum gloeosporioides* f. sp. *malvae*	14
Q00374	Pectin lyase	*Glomerella cingulata*	15
Q9LFP5	Putative pectate lyase 19	*Arabidopsis thaliana*	16
AAB69759	putative pectate lyase	*Arabidopsis thaliana*	17
AAM67091	putative pectate lyase	*Arabidopsis thaliana*	18
S26211	pectate lyase	*Nicotiana taba*	19
E53240	llergen Amb a II precursor	*Ambrosia artemisiifolia*	20
P27759	Pollen allergen Amb a 1.1 precursor	*Ambrosia artemisiifolia* (common ragweed)	21
P15721	Probable pectate lyase P56 precursor	*Lycopersicon esculentum* (tomato)	22
P15722	Probable pectate lyase P59 precursor	*Lycopersicon esculentum* (tomato)	22
Y09541	Zinnia elegans mRNA for pectate lyase	*Zinnia elegans*	23
AAF80746	pectate lyase 1 precursor	*Globodera rostochiensis*	24
AAM21970	pectate lyase 2 precursor	*Globodera rostochiensis*	Unpublished

1个细菌来源的果胶酸酯裂解酶(*Pseudomonas syringae* pv. *tomato*, DC3000)为其并系。第二类群为细菌的果胶酸裂解酶,包含5个细菌来源的果胶酸裂解酶[*Bacillus halodurans* C-125, *Erwinia chrysanthemi*, *Streptomyces coelicolor* A3(2), *Pseudonocardia* sp.]。第三个类群为植物的果胶酸裂解酶,包含9个植物来源的果胶酸裂解酶(*Arabidopsis thaliana*, *Nicotiana taba*, *Ambrosia artemisiifolia*, *Globodera rostochiensis*, *Lycopersicon esculentum*, *Zinnia elegans*),1个真菌来源的果胶酸裂解酶(*Aspergillus nidulans*)为它的并系。

2.3 NJ树(Neighbor-joining)

30种果胶裂解酶NJ树的拓扑结构和其MP树的拓扑结构是类似的(图2:B_1, B_2)。在NJ树中,植物来源的果胶酸裂解酶,真菌来源的果胶酸酯裂解酶和细菌来源的果胶酸裂解酶分别聚为不同的类群,NJ树也显示真菌(*A. nidulans*)来源的果胶酸裂解酶(peLA)为植物的果胶酸裂解酶类群的并系,未和细菌来源的果胶酸裂解酶聚为一类,也未和真菌的果胶酸酯裂解酶聚为一类,细菌(*Pseudomonas syringae* pv. *tomato*, DC3000)来源的果胶酸酯裂解酶(pelA)也为真菌的果胶酸酯裂解酶的并系,未和细菌来源的果胶酸裂解酶聚为一类,也未和植物来源的果胶酸裂解酶聚为一类。与MP树不同的是,在NJ树中,来自细菌的果胶酸裂解酶首先和来自植物的果胶酸裂解酶聚为一类,而在MP树中来自细菌的果胶酸裂解酶首先和来自真菌的果胶酸酯裂解酶聚为一类。

3 讨论

在MP和NJ树中,来自植物的果胶酸裂解酶,真菌的果胶酸酯裂解酶和细菌的果胶酸裂解酶聚成了3个类群(图2),说明植物的果胶酸裂解酶、真菌的果胶酸酯裂解酶和细菌的果胶酸裂解酶分别有相

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S2357       AKNVIIN.IAV..TDIN.PKGG.DAITVDDSDLVWIDHVTT...A.R.HIVLGTSADNRVTISYSLIDGR   60
BAB82467    ..KSNVIIQNIAITDIN.PKGGDATITLNEADLVWIDHVTT...A.RQHIVLGTQADNRVTISNSLIDGR   63
1IDK        ..AENIIIQNIAVTDIN.PKGG.DAITLDDCDLVWIDHVTT...A.R.HYVLGTQADNRVSLTNNYIDGV   61
P2286       ..VSNIIIQNIAVTDIN.PKGG.DAITLDEADLVWIDHVTT...A.R.HYVLGTQADSRVSITNNYINGE   61
JC6502      ..ASNVVVRNLTFRN.....WGDDAINVQYSTRVWIDHNSFSN....DGAVDVKRASDYVTVS.WNKFSS   58
BAB8246     ..VENVIIQNIAVTDIN.PKGG.DAITINEADLVWIDHVTT...A.R..YVLGTEADNRVTLSNNYIDGE   60
AAF2224     ..GSNVIIQNIEVSDLN.HPGG.DAITINEADLVWVDHVTTAR...H..YVFGFNPSKRITLSNNFINGE   61
AAD4356     ...AKNVIVNIHITQLN.PQGG.DAVSLDGSDLVWIDHVKTSL.....HIVLGNGANNRVTISNNEIDGS   60
Q00374      ...AKNVIVNIHITNLN.PRGG.DAISLDGTDLVWIDHVKTSLQ....HIVLGNGANNRVTISNNEIDGS   61
CAD345      ..VSNIIIQNIHITELN.PEGG.DAITLDGTNNVWIDHVKINL.....MFVAGYEASHSVTISNSEFDGE   61
CAC38815    ..VDNVIVRNLTFES.....PVYDSAVVYGSTHVWLDHNTFTD....DG.LDIVKGADYVTAS.WNVFTE   57
AAM23008    ...SSSKILTTLLPPKI.KPIRVTPITHPTTAGHDLDRITVTTRAHR...CSAPRASKRVTRSNSFINGA   63
AAM23009    ..VKNVIVQNIAVTDIN.PQGG.DAITINEADLVWLDHITTDLSALR...P.GTEADNRVSITNNYINGE   62
AAS5538     .....RCSRHNRQVQQV.HRWRVTPITHPTTAGHDLDRITVTTRAHR...CSAPRASKRVTPSNSFINGA   61
P15722      ..IKNVIIHGLHI.HDIVEG.DGDGISIFGASYIWIDHVSMQR....DGLIDAVEGSTGITIS.NGHFTD   61
Q9LFP       ..VKNVIIHGLHI.HHISES.SGDGLSIYGSSNIWIDHISMS.....DGLIDAIVGSTGITIS.NSHFTH   60
AAB697      ..VNNVIIHNIYVKH.IVPG.NGDGISLIGATNIWIDHVSMTR....DGMIDAIDGSTAVTIS.NSHFTD   61
P15721      ..ASNIIISNLRI.HNIVPT.AGDAISIFNSHDIWIDHS.MSR....DGLIDAVAGSTNITIS.NCHFTD   60
S26211      ..ASNIIISNLRI.HNIVPTPEGDGISIFSSHDIWIDHISMSR....DGLIDAVAASTNITIS.NCHFTD   62
AAM670      ..VTNIIIHGVNI.HDCKRK.GGDAVSIFGGSHVWVDHCSLSN....DGLIDAIHASTAITIS.NNYLSH   61
Y09541      ..ASNIIIHGIHI.HDCKQA.GGDGISIFASKDIWIDHNSLS.....DGLIDAIHGSTAITISINNYMTH   61
E53240      VKAAN..IHNI.....DVRVLPGDAIHVTGSSDIWIDHCTLS.....DGLVDVNWGSTGVTIS.NCKFTH   57
P27759      VK..NVIIHNINM..HDVKVNPGDAISISGSSQIWIDHCSLS.....DGLVDAKLGTTRLTVS.NSLFTQ   60
NP_241      ..GSNVIVKNLTIRG......TYGIQTGKDAHHIWIDHVTMRK....DGLIDIVNGANYVTIS.NSRFEQ   57
P18210      ..VSNVILRNLYIE.......TPVDAVVIDSTDHVWDHVTISD....DGSLDIKRGSDYVTVS.NSRFEL   56
AAF807      ..KKGAVIKNLIIG.....KNGADGIHCYDSCTLENVWFESVG.............EDATTFRTSKDCGA   50
AAM219      ..KKGAVIKNLIIG.....KNGADGIHCYDSCTLENVWFESVG.............EDATTFRTSKDCGA   50
Q00645      ..VSNVIMRNLKISKVDADN..GDAIGIDASSNVWVDHCDLSG....DGLVDISHGAEWITVSNTYFHDH   62
NP_794      ..AHNVIVRNLTLSDIN..PSGGDALTLNKADGVWIDHNTFARIG.RQMIVTGWGTASHVTISSNEFDGR   65
Consensus

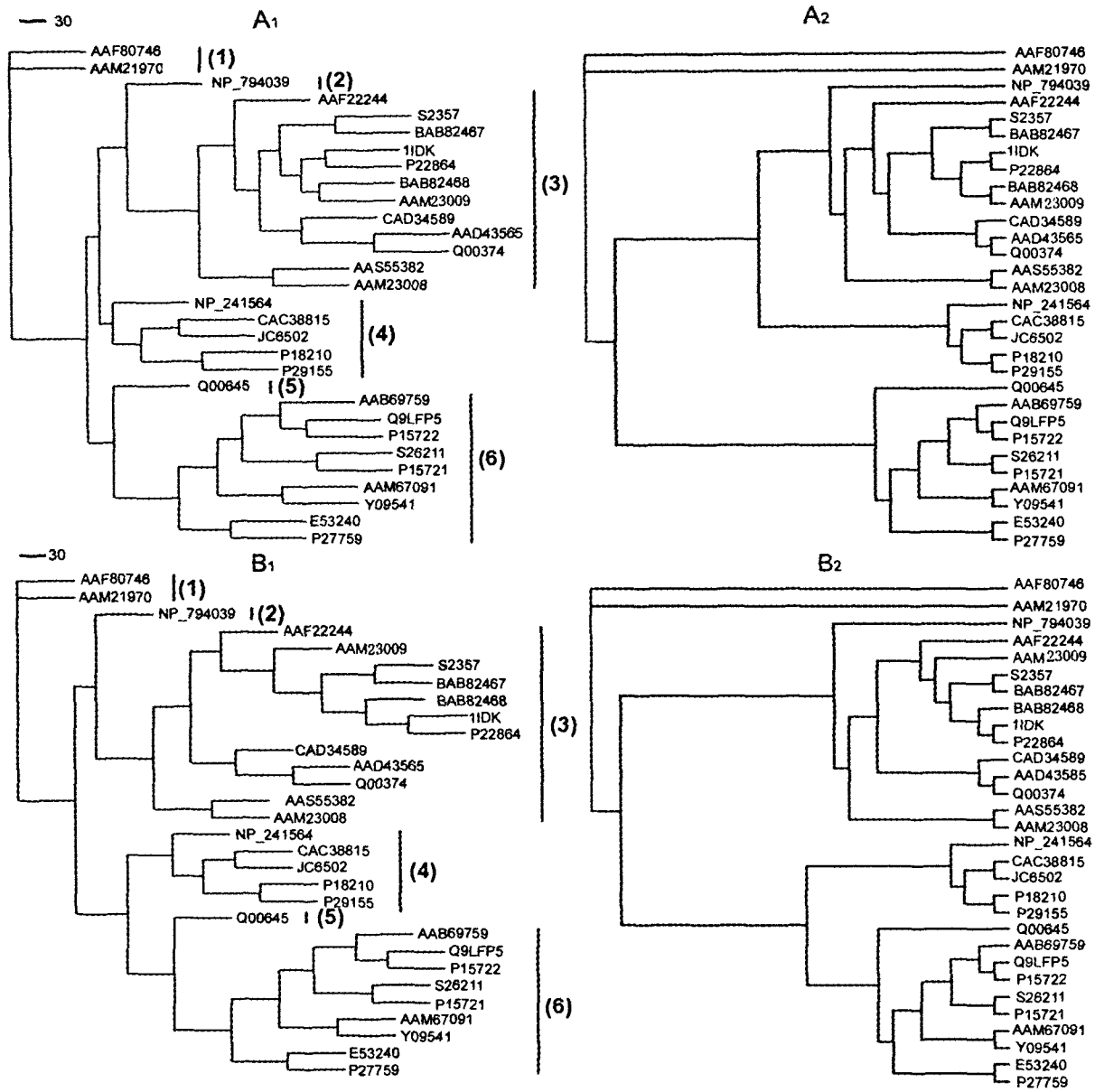
S2357       SDYSATCNGHR.........MPKVQGNTLLHAVNNLFHGYVL.AEGNVF...QDVNI                104
BAB82467    TDYSATCNGH..........MPKVQGNTLLHAVNNLFFGYVL.AEGNAF...QDVDA                106
1IDK        SDYSATCDGYR.........SPKVQDNTLLHAVNNYWYGYVL.AEGNVF...QNVDT                105
P2286       SDYSATCDGH..........APKVQDNTYLHIYNNYWEGYVL.AEGNYF...SNVDT                104
JC6502      HNKTMLLGHSD..R......HPRVRFGNPVHVYNNYYG.VASTMEAGVL...VEGNY                103
BAB8246     SDYSATCDGH..........APKVQGNTYLHAVNNYWNGYVL.AEGNTF...ADVTA                103
AAF2224     STYSTGCNGY..........GPALSGGTLLHAVNNVWEGLSI.ASADAA...SKIKT                104
AAD4356     TSWSATCDNH..........APKIAGNSLVHVVNNYFYAQVVLEGNIFQ...NVKA                 103
Q00374      TSWSATCDNH..........SPKIAGNSLVHISG.YFYANSG                               92
CAD345      TSWSATCDGH..........SPKLEFNSFWHAYNNYWYGRTCVPNTLIS...SGTL                 104
CAC38815    MDKTILGNSDSSER......APRVRFGQ.VDVYNNHFVSFGVGKESALV...AEHN                 103
AAM23008    SDYSATCNGYP.........KVQG..NTLLHAVNNYWFGKNIASASAYT...SVVA                 105
AAM23009    SDYSATCDGH..........APKVQDNTYLHAVNNYWNGETLPLSA.VA...STVPPW               106
AAS5538     SDYSATCNGYV.........PPRFRATPLLHACQQLTCSRTLLLLSPLL...LLD                  104
P15722      HNEVMLF.GASD.R......MPRCRWGY.IHVVNNDYTP......TII....HQGNRFIAPP          104
Q9LFP       HNDVMLL.GAQN.R......MPRIRWGF.VHVVNNDYTP......TILS...H.GNRFIAPP          103
AAB697      HQEVMLF.GARD.R......MPRCRYGT.IHVVNNDYT.......TIIS...Q.GNRFIAPP          103
P15721      HEKVMLF.GAND.Q......MPRCRFG..FHLVNNDYTA......TIIS...Q.GNRFIAED          102
S26211      HEKVMLF.GAND.R......MPRCRFGF.FHLVNNDYTA......TIIS...Q.GNRFIAED          105
AAM670      HNKVHLL.GHSD.R......MPRCRHGY.FHVVNNDYTP......TIIT...Q.GNRFIAP           103
Y09541      HDKVHLL.GHSD.R......MPRCRHGY.FHVVNNDYTH......TIYS...Q.GNRFLAPN          104
E53240      HEKAVLL.GASD.R......MPRCRFGF.FQIVNNFYDRP.....TILS...Q.GNKFVAPD          101
P27759      HQ.FVLLFGAGDER......MPRCRHGF.FQVVNNNYDKP.....TILS...Q.GNRFCAPD          105
NP_241      HNKSITISGND..R......NPRVRFGM.VHLYNNYYSAIGVGVSAKIY...SENNS               102
P18210      HDKTILIGHSDNNR......TPRVRFGS.VHAYNNVYS......SGSLL...SESNAFTIDNM         103
AAF807      GSFVVTGGGARNAH......DKVFQMNGGLTIRNYVGENIGKLARSCGNCEVQCKNRKFVVENV        108
AAM219      GSFVVTGGGARNAH......DKVFQMNGGLTIRNYVGENIGKLARSCGN.EVQCKN.KFVVENV        106
Q00645      HWKGSLIGHSDR........TPLIRFAT.VHIINNYWDAQVLIQSSAPHNCPD                    106
NP_794      TPYSSTCGHRAPHSGGMKNAQIRAQLVSQLLVEGNDFENVAH                               107
Consensus
```

图 1　植物、真菌、细菌和线虫等生物部分的果胶酸裂解酶和果胶酸酯裂解酶保守区氨基酸序列比对

Fig. 1　Alignment of conserved regions of different pectate and pectin lyases from plant, fungi, bacteria, and nematodes

对独立的演化进程。图 2 中系统进化树虽然仅是用来自动物、植物、真菌和细菌等 4 类不同生物的果胶裂解酶保守区的氨基酸序列构建的蛋白树,但该树一定程度上也反映了动物、植物、真菌和细菌等 4 类生物之间的相对独立的演化关系,提示该蛋白可以作为物种亲缘关系研究的一个标记基因[47,48]。

在 MP 和 NJ 树中,真菌来源的果胶酸裂解酶(PeLA)为植物的果胶酸裂解酶类群的并系,未和细菌来源的果胶酸裂解酶聚为一类,说明该果胶酸裂解酶和植物来源的果胶酸裂解酶的亲缘关系要近于细菌来源的果胶酸裂解酶和植物来源的果胶酸裂解酶的亲缘关系。细菌(*Pseudomonas syringae* pv. *tomato*, DC3000)来源的果胶酸酯裂解酶(PelA)为真菌的果胶酸酯裂解酶的并系,说明该细菌细菌来源



A_1, MP tree; A_2, consensus tree from MP tree; B_1, NJ tree; B_2, consensus tree from NJ tree. (1) nematode; (2) pectin lyase of bacteria; (3) pectin lyase of fungi; (4) pectate lyase of bacteria; (5) pectate lyase of fungi; (6) pectate lyase of plant

图 2 植物、真菌、细菌和线虫等生物部分的果胶酸裂解酶和果胶酸酯裂解酶系统演化分析

Fig. 2 Phylogenetic trees of pectate lyases and pectin lyases from plant, fungi, bacteria, and nematode

的果胶酸酯裂解酶和真菌来源的果胶酸裂解酶的亲缘关系要近于细菌来源的果胶酸酯裂解酶和细菌来源的果胶酸裂解酶的亲缘关系。MP 和 NJ 树结果还显示,在真菌的果胶裂解酶中,来自 *A. nidulans* 的果胶酸裂解酶与来自植物的果胶酸裂解酶亲缘关系一方面要近于 *A. nidulans* 果胶酸裂解酶与来自真菌的果胶酸酯裂解酶的亲缘关系,另一方面要近于真菌果胶酸酯裂解酶与植物的果胶酸裂解酶的亲缘关系。

本文中 MP 树和 NJ 树的结构有一定的差异,如在 NJ 树中来自细菌的果胶酸裂解酶首先和来自植物的果胶酸裂解酶聚为一类,而在 MP 树中来自细菌的果胶酸裂解酶首先和来自真菌的果胶酸酯裂解酶聚为一类,导致这种差异的原因是源于 MP 树和 NJ 树的计算方法上的差异,但是本文中 MP 树和 NJ 树的拓扑结构未有明显的差异。

果胶酸酯裂解酶和细菌果胶酸裂解酶催化方式虽然都是 β 消旋法,果胶酸酯裂解酶催化的底物是甲基化的果胶酸,果胶酸裂解酶催化的底物是未甲基化的果胶酸,底物的差异可能会导致关键部位氨

基酸发生差异,这可能是真菌果胶酸裂解酶和植物果胶酸裂解酶亲缘关系比较近,细菌果胶酸酯裂解酶和真菌果胶酸酯裂解酶亲缘关系比较近的原因之一。

有研究表明,花粉表达的果胶酸裂解酶序列和来自 Erwinia 的果胶酸裂解酶序列相似性较高[46],其它一些研究[13]报道细菌的果胶酸裂解酶的保守区明显不同于植物和真菌的果胶酸裂解酶,这一点与本研究结果相似。在本文中 MP 树和 NJ 树都显示真菌的果胶酸裂解酶是植物果胶酸裂解酶类群的一个并系,真菌果胶酸裂解酶和细菌果胶酸裂解酶有较远的亲缘关系。

植物的果胶酸裂解酶异源表达一直未能成功,使得植物果胶酸裂解酶在花粉萌发,维管束诱导分化和果实成熟等方面的机理研究一直难以得到确切的证据,从真菌来源发现与植物亲缘关系近的果胶酸裂解酶,来模拟研究果胶酸裂解酶在相关植物生理过程中的作用机制,可能是值得尝试的方法之一,该方面的研究成果以后将在相关刊物上发表。

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