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间伐改形影响密闭苹果园树体花芽激素含量及相关基因

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摘　要:[目的]富士苹果栽培中普遍存在成花困难、大小年结果严重等问题,尤其在密闭果园中更为突出。本文旨在研究间伐改形对甘肃陇东高原密闭苹果园树体花芽激素含量及相关基因的影响,为进一步研究间伐改形对苹果花芽分化提供试验基础。[方法]在甘肃静宁县城川镇,以十五年生树龄阶段的密闭'长富2号'苹果园为研究对象,2022年春季对密闭果园的一半进行间伐改形处理,另一半果园作为对照,不进行间伐改形,同年,分别在花芽生理分化前期和花芽生理分化后期,对间伐改形和不间伐改形树体花芽进行转录组分析和生长相关激素含量测定。[结果]无论在花芽分化生理前期和后期,间伐改形后树体花芽赤霉素、吲哚乙酸和脱落酸含量均明显增加,同时发现,转录组中有580个基因表达发生变化,并从中挑选出5个生长发育相关基因(MD10G1048600(NAC030)、MD01G1090300(CCA1)、MD02G1124900(PIP2－7)、MD01G1016100(ABR1)、MD07G1280300(WRKY27)),对这5个基因进行qRT－PCR,发现在对苹果园进行间伐改形处理后,这5个基因都有明显的上下调变化,且与转录组变化结果一致。[结论]间伐改形对苹果花芽分化有显著作用,可以促进花芽赤霉素、吲哚乙酸和脱落酸的形成,也能促进生长发育相关基因的表达。

关键词:苹果; 间伐改形; 花芽分化; 转录组
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苹果(*Malus pumila* Mill.)是蔷薇科苹果属落叶乔木,喜光、耐寒[1]。中国现存苹果是从美国和日本引进的西洋品种[2]。我国苹果产区分布极广,主要集中在环渤海、西北黄土高原、黄河故道和西南冷凉高地四大产区[3]。'长富2号'是1980年引入我国的着色系富士品种,经过20多年发展,现在已成为我国苹果主栽品种之一,果实综合品质优良。

Askenaly于1867年研究了酸樱桃的花芽分化时期,是果树方面最早的花芽分化研究,其后许多学者从形态、解剖和生理生化等多方面对果树的花芽分化进行了研究,而苹果花芽分化的研究也有100多年的历史。苹果花芽属于混合芽,即展开后可以成为叶芽或花芽[4]。花芽数量、质量是衡量苹果树体结果潜力的关键、核心指标。果树生长包括营养生长和生殖生长,两者既相互促进,又相互抑制。花芽分化是从营养生长到生殖生长的一个重要的变化过程,这个过程是营养生长结束和生殖生长开始的标志。这种变化极大地影响了花期和花的数量[5]。花芽分化的实质是植物经一定时期的营养生长后,由于受到光照、温度等外界因素的影响,产生某些与花芽分化相关的激素,在适宜的温度、光照和营养条件下,茎生长锥不再向叶芽方向转变形成叶原基和腋芽原基,而是发生花原基和花序原基,逐渐形成花芽[6]。果树不同发育阶段的成花能力不同,其枝条生长状况、叶片质量状况都有所差异,这些差异的形成决定了其具有不同的成花能力[7]。有研究发现,温度、光照、水分、矿物质含量等是影响花芽分化的关键因素[5]。

甘肃陇东黄土高原区是我国红富士等晚熟苹果栽培的最佳适宜区,苹果产业已成为当地经济发展和乡村振兴的主导产业。该产区20世纪90年代至21世纪初栽植的苹果园大多数采用乔化密植栽培,虽然乔化密植栽培曾经起到过促进丰产的积极作用,但随着树龄的增长,果园郁闭程度的

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增加,出现枝量繁多、冠层光照条件恶化、病虫害防治难度增大、结果部位外移,年际结果不均衡,产量和果实品质下降等诸多问题[8-10]。严重制约着苹果产业的进一步发展。为此,乔化成龄密闭苹果园的改造和树体结构的合理调整研究已成为当地苹果栽培中的热点课题。

果园间伐是乔化成龄密闭苹果园改造的主要措施,能够在较短时间内改善果园通风透光条件、优化果园群体结构、促进花芽分化、改善果实内外品质[11-12],但是随着时间推移,果园密闭现象将再次显现,既不能持久拥有良好的通风条件,也不能持续具备合理的群体结构[13-14]。缩小树冠、疏除大枝、提高主干、落去主头等改形措施,也可暂时缓解单个树体密闭的问题,进而提高果实内外品质,但也不能永久解决果园整体郁闭问题,不能持久具备优质高产的潜能[15]。目前有研究发现,对老果园进行间伐改形后,花芽分化明显,成花率显著提高,第三年坐果率也大幅提高[16],对密闭苹果园进行间伐改形后发现成花能力和花芽质量明显提高,冠层花芽和果实的分布空间也得到改善[17]。

植物激素是调节高等植物发育和环境信号应答的重要分子,其在植物生长的各个阶段发挥作用,花芽分化亦不例外[18]。研究发现生长素、脱落酸、多效唑、赤霉素等激素都能够促进花芽分化[19-20]。有研究表明,高水平含量的脱落酸有利于花器官形成发育;反式玉米素核苷含量升高对菠萝花芽分化的整个过程都有促进作用,与花原基形成的数量正相关;低浓度的生长素和赤霉素有利于藤本月季“安吉拉”的花芽分化[21]。本试验以陇东高原雨养区十五年生‘长富2号’红富士密闭果园为对象,通过间伐与树体改形相结合的改造试验,研究了不同处理对树体花芽分化的影响,通过不同分化时期的生长类相关激素和转录组分析,在激素和分子生物学层面解析间伐改形对乔化成龄密闭苹果园树体成花的影响,为乔化成龄密闭红富士苹果园合理改造和富士苹果品种的改良提供理论依据。

1　材料与方法

1.1　试验区概况和试验材料

试验于2022年在国家苹果产业技术体系平凉苹果综合试验站的静宁示范县城川镇试验园(35°24′16″ N,105° 47′15″ E)进行。试验园海拔1592 m,年降雨量460 mm,春季少雨较为干旱,秋季雨量丰沛且较集中,年无霜期160 d,年日照时数2242 h,平均气温7.21 ℃。土壤为黄绵土,粘性较弱,有机质含量为9.23 g·kg^{-1},pH为8.02。试验园面积为3620 m^2,供试品种为十五年生‘长富2号’红富士苹果树,砧木为山定子(*Malus baccata*),行株距为4 m×3 m,密度为834 株·hm^{-2},改良纺锤形树形,树冠交接率大于140%,树高4.2～4.6 m,主干高度60～75 cm,主枝数6～9个,主枝角度70°～80°,授粉树品种为秦冠和金冠,占比为10%。果园为雨养果园,无灌溉条件,地面自然生草,栽培管理水平中等偏上,果园管理一致,以单个树体为单位进行施肥,即各处理每株树体施肥水平完全一致。

1.2　试验方法

参照牛军强等[5]的方法,于2022年2月上中旬开始对试验园开展间伐改形改造试验,试验采用单因素随机设计的方法,只做2种处理,分别为试验T(间伐改形)和对照CK(不间伐不改形)。间伐改形采用隔株间伐,行株距由原来的4 m×3 m变为4 m×6 m,同时结合缩小树冠、疏除大枝、提高主干、落去主头等改形措施,使树高变为310～340 cm,干高100～110 cm,选留健康无病虫害、大小合理、分布方位均衡、间距适度的枝条4～6个作为永久主枝,开张角度为90°～100°,其他修剪采用富士苹果常用的手法进行修剪,总体修剪量为30%左右。对照(CK)为不间伐改形(既不间伐也不改形),其它修剪手法和修剪量与间伐改形处理完全相同。

1.3　取样时间与方法

每个处理区面积均为20 m×25 m=500 m^2,均有3个重复,每个重复选取树势中庸、无明显病虫害、有代表性的树体5株作为供试树体。于2022年6月22日花芽生理分化前期(CK1和T1)和2022年8月22日花芽生理分化后期(CK2和T2),分别取样1次,取样部位为树体冠层中下部中短枝上的封顶芽体,要求芽体大小、饱满程度一致,以最快速度采取芽体样品,然后用锡箔纸将样品包裹严实并标记清楚,于液氮罐贮藏带回实验室,−80 ℃超低温冰箱保存,等待检测分析。

1.4 转录组学测定方法

用TRIzol (thermofisher, 15596018),根据厂商提供的操作方案对总样品的RNA进行分离和纯化。然后用NanoDrop ND-1000(NanoDrop,美国)对总RNA的量与纯度进行质控并通过Bioanalyzer 2100(Agilent,美国)对RNA的完整性进行检测。使用oligo(dT)磁珠(Dynabeads Oligo (dT), cat. 25-61005, Thermo Fisher,美国)通过2轮纯化对其中的带有PolyA(多聚腺苷酸)的mRNA进行特异性捕获。将捕获到的mRNA在高温条件下利用镁离子打断试剂盒(NEBNext Magnesium RNA Fragmentation Module, cat. E6150S,美国)进行片段化,94 ℃保持5~7 min。将片段化的RNA在逆转录酶(Invitrogen SuperScript™ II Reverse Transcriptase, cat. 1896649,美国)的作用下合成cDNA。然后使用*E. coli* DNA polymerase I (NEB, cat. m0209,美国)与RNase H(NEB, cat. m0297,美国)进行二链合成,将这些DNA与RNA的复合双链转化成DNA双链,同时在二链中掺入dUTP Solution (Thermo Fisher, cat. R0133, 美国),将双链DNA的末端补齐为平末端。再在其两端各加上1个A碱基,使其能与末端带有T碱基的接头进行连接,并利用磁珠对其片段大小进行筛选和纯化。以UDG酶(NEB, cat. m0280,美国)消化二链,再通过PCR——预变性95 ℃保持3 min,98 ℃变性总计8个循环、每次15 s,退火到60 ℃保持15 s,72 ℃下延伸30 s,最后72 ℃保留5 min,使其形成片段大小为300 bp±50 bp的文库。最后,使用illumina Novaseq™ 6000按照标准操作对其进行双端测序,测序模式为PE150。

1.5 激素含量测定

通过ELISA试剂盒测定赤霉素、生长素、脱落酸含量。

1.6 RNA提取和基因表达分析

根据制造商的说明,使用TRIzol(Vazyme,中国)方法从叶中提取总RNA。使用反转录试盒(TransGen,中国)将总RNA逆转录成cDNA。使用TransStart® Tip Green qPCR SuperMix(TaKaRa,中国)和引物进行qRT-PCR。反应的总体积为20 μL,其中cDNA为200 ng,正向引物、反向引物和ROX Reference Dye均为0.4 μL, TB Green *Premix Ex Taq* (Tli RnaseH Plus)(2×) qPCR super mix为10 μL,少于20 μL的部分用双蒸水补充。PCR循环条件设定如下:在94℃下进行30 s的预变性,接着94 ℃下5 s、60 ℃下30 s进行40个循环,最后使用溶解阶段检测荧光信号以验证目标产物是否是单一。qRT-PCR分析中使用的引物如表1所示,*Actin*为内参。

1.7 数据处理

数据采用Excel 2013软件进行统计处理,差异显著性分析使用SPSS 26统计分析软件。

表1 荧光定量引物名称和序列

Table 1 Name and sequence of fluorescent quantitative primers

基因ID Gene ID	上游引物(5'-3') Forward primer	下游引物(5'-3') Reverse primer
MD10G1048600	GCTTCAGATTTCACCCGACAG	GCTCTATTTGTTCGCGTTCC
MD01G1090300	CCATGGTTCGCTCACAAG	CCTTCTGTCCATCGCTCTC
MD02G1124900	GTGGCCGTTCTTCAGAGCT	GACGAAGATCATACCGCC
MD01G1016100	TCCCTTGTTCGAGTCGGT	GGGAACGCTGTTTGTGTAGG
MD07G1280300	CGCCAGCTCCAAACTCAG	CTGCTGCATCTGTAGTAGTTCC
Actin	ATTCACGAGACCACCTACA	TGCCACAACCTTAATCTTCAT

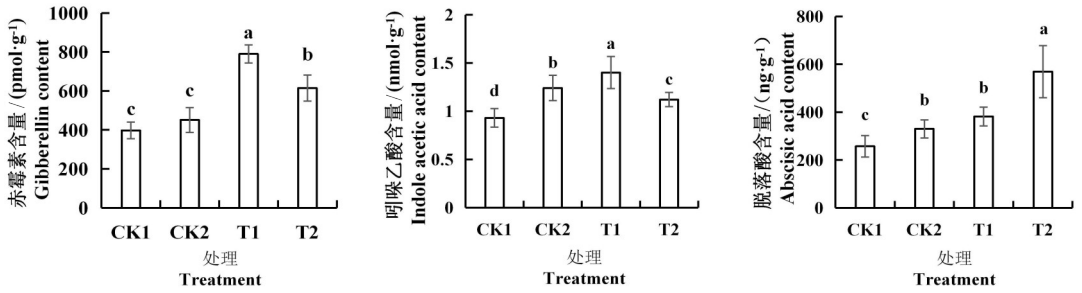
注:表中*Actin*代表本次荧光定量所用的内参序列

Note: *Actin* in the table represents the internal reference sequence used in this fluorescence quantification

2 结果与分析

2.1 激素含量测定

对T1、T2、CK1、CK2进行赤霉素、吲哚乙酸、植物脱落酸含量测定,结果如图1所示,T组的激素含量基本都高于CK组的激素含量。与CK1相比,T1的赤霉素含量、吲哚乙酸含量和脱落酸含量明显增加,与CK2相比,T2赤霉素含量和脱落酸含量显著增加,吲哚乙酸含量稍有下降。这说明,间伐改形能显著提高树体花芽赤霉素、吲哚乙酸



CK1:对照组花芽分化前期;CK2:对照组花芽分化后期;T1:间伐改形处理后花芽分化前期;T2:间伐改形处理后花芽分化后期。表2与图3、图4、图6同

CK1: the prophase of flower bud differentiation in control group; CK2: the later stage of flower bud differentiation in control group; T1: the prophase of flower bud differentiation after thinning; T2: the later stage of flower bud differentiation after thinning. The same as Table 2, Fig. 3, Fig. 4, and Fig. 6

图1　赤霉素、吲哚乙酸、植物脱落酸含量测定

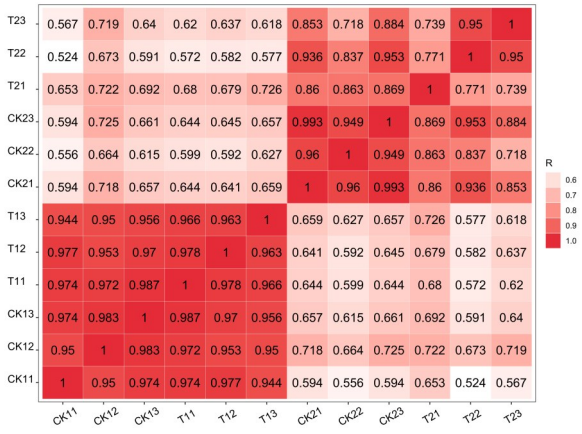
Fig.1　Determination of gibberellin, indoleacetic acid, and abscisic acid content

和脱落酸等生长相关激素的含量,进而说明间伐改形对花芽分化有一定促进作用。

2.2　转录组学分析

2.2.1　相关性分析

为探索间伐改形对花芽分化的影响,对T1(间伐改形)和CK(不间伐改形)花芽进行转录组测序。样本之间的相关系数越大,样本聚类越好。Pearson相关系数可直观看出样本之间的可重复性,并有助于发现数据异常样本。图2可见,本研究中样本相关性结果很好,足以支撑后续研究。



CK11～CK13:对照组花芽分化前期;CK21～CK23:对照组花芽分化后期;T11～T13:间伐改形处理后花芽分化前期;T21～T23:间伐改形处理后花芽分化后期

CK11～CK13: the prophase of flower bud differentiation in control group; CK21～CK23: the later stage of flower bud differentiation in control group; TT11～T13: the prophase of flower bud differentiation after thinning; T21～T23: the later stage of flower bud differentiation after thinning.

图2　样本间的皮尔逊相关性分析

Fig.2　Pearson correlation analysis among samples

2.2.2　质量评估

T1与CK1相比,总共检测48个基因表达上调,54个基因表达下调,T2与CK2相比,总共检测353个基因表达上调,227个基因表达下调,如图3所示。为了解差异表达基因的整体分布情况,以log(fc)为横坐标,-log(q value)为纵坐标,对差异表达分析中的所有基因绘制火山图,如图4所示。

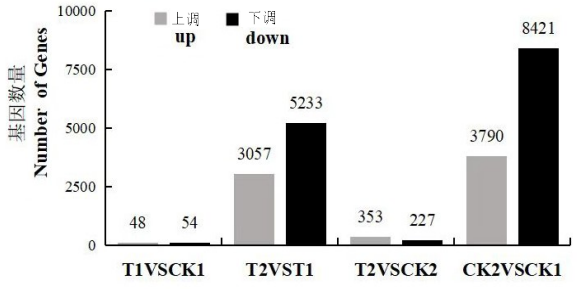


图3　差异表达基因上下调频数统计

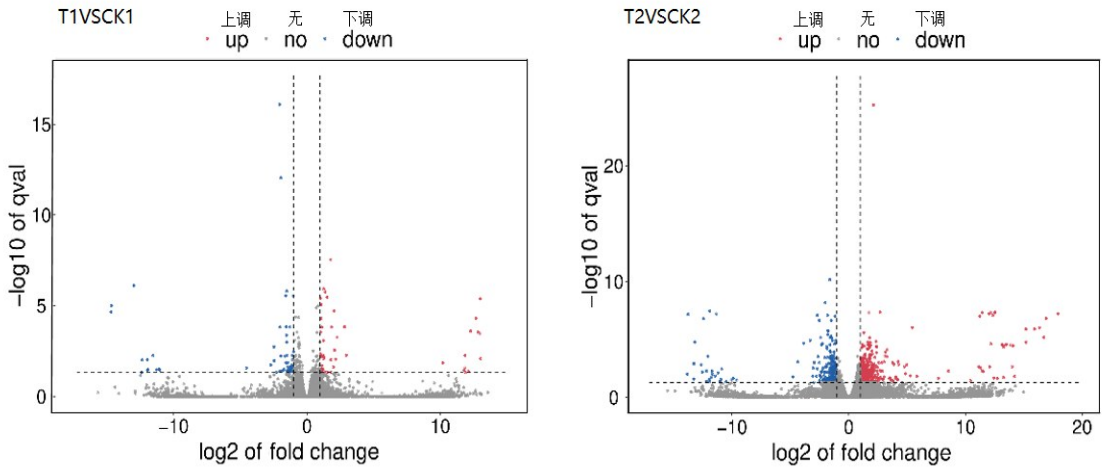
Fig.3　Frequency distribution chart of upgraded and downregulated differentially expressed genes

2.2.3　DEGs GO富集分析

GO富集分析结果以气泡图的形式呈现。根据Unigene在GO数据库中的比对结果,选取每组样品中差异表达量最高的20个基因进行泡绘,如图5所示。

2.2.4　生长发育相关基因的挑选

根据GO富集结果,从3 diffExpression文件夹的各比较组中的文件-Gene differential expression中找到如下57个生长发育相关基因(表2)。由表2可以看出,gene:MD10G1048600、gene:MD01G1090300、gene:MD09G1176100、gene:MD16G1218900、gene:MD15G1393600等基因在花芽分化前期和后期在生长素相关GO富集通路检测到明显变化;gene:MD10G1048600、gene:



红色代表上调的显著差异表达基因,蓝色代表下调的显著差异表达基因,灰色的点代表非显著性差异表达基因

Red represents up−regulated significantly differentially expressed genes, blue represents down−regulated significantly differentially expressed genes, and gray dots represent non−significantly differentially expressed genes

图4 差异基因火山图

Fig. 4 Volcanic map of differential genes

MD01G1090300、gene: MD06G1003800、gene: MD15G1012500、gene: MD02G1124900、gene: MD01G1016100等基因在花芽分化前期和后期在脱落酸相关GO富集通路检测到明显变化;gene: MD01G1090300、gene: MD07G1280300、gene: MD15G1012500、gene:MD15G1109300等基因在花芽分化前期和后期在赤霉素相关GO富集通路检测到明显变化。

综合分析后挑选出表达量变化明显的基因5个,分别是:gene:MD10G1048600(生长素、脱落酸相关基因)、gene:MD01G1090300(生长素、脱落酸、赤霉素相关基因)、gene:MD02G1124900(脱落酸相关基因)、gene:MD01G1016100(脱落酸相关基因)、gene:MD07G1280300(赤霉素相关基因)。

表2 生长发育相关基因

Table 2 Growth and development related genes

基因ID Gene_ID	log2(FC)			基因ID Gene_ID	log2(FC)		
	T1VSCK1	T2VSCK2	T2VST1		T1VSCK1	T2VSCK2	T2VST1
gene:MD16G1142800 GO:0009733(response to auxin)	−9.57	0	0	gene:MD15G1246800	−8.64	0	0
gene:MD05G1052300	−11.31	0	0	gene:MD00G1016500	8.51	0	−8.51
gene:MD10G1048600	10.54	−10.77	−10.54	gene:MD16G1142800 GO:0010252(auxin homeostasis)	−9.57	0	0
gene:MD01G1090300	−10.73	0.71	12.56	gene:MD04G1243800	−9.77	0	0
gene:MD07G1260700	8.19	−1.42	2.36	gene:MD10G1048600 GO:0009737(response to abscisic acid)	10.54	−10.77	−10.54
gene:MD07G1110600	8.24	0	−8.24	gene:MD07G1307600	9.42	−0.13	−1.66
gene:MD05G1181100	8.15	0	−8.15	gene:MD07G1285400	8.94	0.61	6.96
gene:MD09G1176100	−10.37	10.27	10.27	gene:MD01G1090300	−10.73	0.71	12.56
gene:MD04G1095800	9.97	5.30	0.96	gene:MD05G1181100	8.15	0	−8.15
gene:MD04G1093700	−9.63	0	0	gene:MD06G1003800	−9.25	−9.35	0

续表

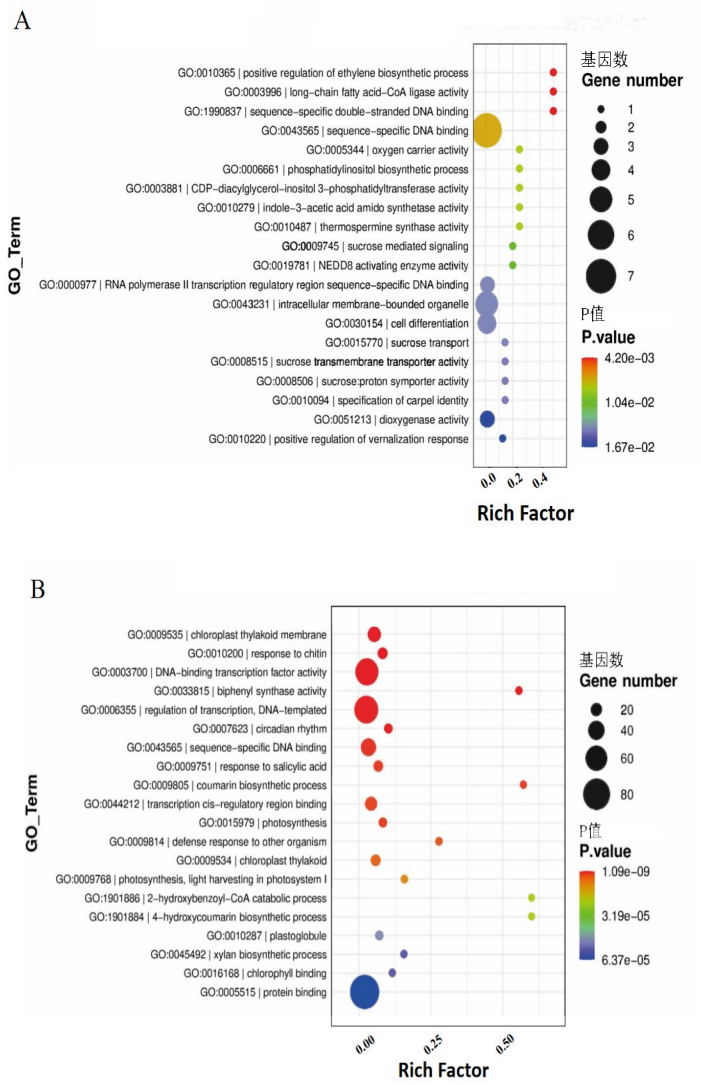
基因ID Gene_ID	log2(FC)			基因ID Gene_ID	log2(FC)		
	T1VSCK1	T2VSCK2	T2VST1		T1VSCK1	T2VSCK2	T2VST1
gene:MD16G1218900	10.08	11.32	0	gene:MD08G1068100	−8.57	0	0
gene:MD10G131230	8.41	0	−8.41	gene:MD15G1012500	9.35	0	−9.35
gene:MD06G1137300	8.40	0	−8.40	gene:MD02G1124900	10.10	−11.30	−10.10
gene:MD07G1220700	8.56	0	−8.56	gene:MD10G1317300	8.62	10.64	2.02
gene:MD12G1115800	−8.71	0	0	gene:MD01G1088500	−8.31	−7.69	0
gene:MD15G1246800	−8.64	0	0	gene:MD01G1016100	−8.51	−2.34	11.37
gene:MD00G1016500	8.51	0	−8.51	gene:MD01G1191300	9.49	0.85	2.05
gene:MD10G1059100	−8.26	0	0	gene:MD01G1090300 GO:0009739(response to gibberellin)	−10.73	0.71	12.56
gene:MD10G1059400	−8.34	0	0	gene:MD07G1280300	−9.01	−0.71	7.69
gene:MD00G1147100 GO:0009851(auxin biosynthetic process)	−1.51	1.35	2.05	gene:MD12G1143800	−7.90	10.40	10.40
gene:MD06G1147500	1.69	−1.29	−1.22	gene:MD15G1012500	9.35	0	−9.35
gene:MD16G1098400 GO:0009734(auxin−activated signaling pathway)	−8.39	0	0	gene:MD15G1109300	−9.66	10.55	10.55
gene:MD15G1393600	10.43	0	−10.43	gene:MD08G1131700	−9.45	0	0
gene:MD09G1176100	−10.37	10.27	10.27	gene:MD13G1169600	8.84	10.55	1.71
gene:MD15G1031600	−8.73	−7.36	0	gene:MD00G1012500	−8.76	0	0
gene:MD10G1312300	8.41	0	−8.41	gene:MD13G1086500	8.44	−0.37	1.53
gene:MD15G1305900	8.43	0	−8.43	gene:MD16G1142800 GO:0010279(indole−3−acetic acid amido synthetase activity)	−9.57	0	0
gene:MD08G1130800	−8.43	0	0	gene:MD16G1098400 GO:0009684(indole-acetic acid biosynthetic process)	−8.39	0	0

2.3　差异表达基因qRT−PCR验证

为了验证转录组测序数据的可信度,将上述挑选出的5个基因:gene:MD10G1048600(NAC030)、gene:MD01G1090300(CCA1)、gene:MD02G1124900(PIP2-7)、gene:MD01G1016100(ABR1)、gene:MD07G1280300(WRKY27),进行qRT-PCR分析。qRT-PCR结果与转录组结果趋势基本一致,表明转录组数据的可信度。与CK1相比,T1处理的MD02G1124900表达量上调,MD10G1048600、MD01G1090300、MD01G1016100、MD07G1280300这4个基因的表达量都下调;与CK2相比,T2处理的MD01G1090300表达量上调,MD10G1048600、MD02G1124900、MD01G1016100、MD07G1280300这4个基因表达量都下调;与T1相比,T2处理的MD02G1124900表达量下调,MD10G1048600、MD01G1090300、MD01G1016100、MD07G1280300这4个基因表达量都上调。其中MD02G1124900上调比较明显。

3　讨论

很多苹果果园目前都存在果园郁闭的问题,果园郁闭会影响通风透光性、降低花芽质量、加大



(A)T1VSCK1气泡图;(B)T2VSCK2气泡图。横坐标Rich Factor表示位于该GO的差异基因数占位于该GO的总基因数的比例(Rich. Factor = S / B),Rich factor越大,GO富集程度越高;纵坐标是GO Term,即GO功能注释;气泡图中,气泡的大小代表S数目,气泡的颜色代表富集分析的P值(或Q值),即富集的显著性,P值(或Q值)越小,富集越显著

(A) is T1VSCK1 Bubble Chart and (B) is T2VSCK2 Bubble Chart. The abscissa Rich Factor indicates the ratio of the number of differential genes located in this GO to the total number of genes located in this GO (Rich. Factor = S/B). The greater the Rich factor, the higher the enrichment degree of GO. The ordinate is GO TERM, that is, go function annotation; In the bubble diagram, the size of bubbles represents the number of S, and the color of bubbles represents the P value (or Q value) of enrichment analysis, that is, the significance of enrichment. The smaller the P value (or Q value), the more significant the enrichment is

图5 GO富集气泡图

Fig.5 GO enrichment bubble chart

营养消耗、导致大小年现象严重与树势衰弱等[22-23]。为了改变苹果果园郁闭的现象,通常采用间伐的方式缓解。前人研究表明,间伐可以提高植株叶片的光合速率、促进新梢生长和短枝的成枝、改善果树的枝类组成比例、促进果实着色,提高果面的光洁程度,提高单果重和可溶性固形物含量,可以解决果园郁闭的问题[24-25]。

植物激素的研究于20世纪30年代因生长素的发现而拉开序幕,目前已基本明确植物激素的生物合成途径[26]。植物激素是调节植物生长、发育和抗逆性的重要物质,其几乎参与了植物生长发育所有的生理过程[27]。研究发现低水平生长素、赤霉素(GA3)、茉莉酸甲酯(MeJA)和高水平脱落酸(ABA)有利于花芽分化,且较高的mABA/mGA3和mABA/mMeJA比值有利于花芽分化;吲哚乙酸可以激活花原基发育,有利于花器官的

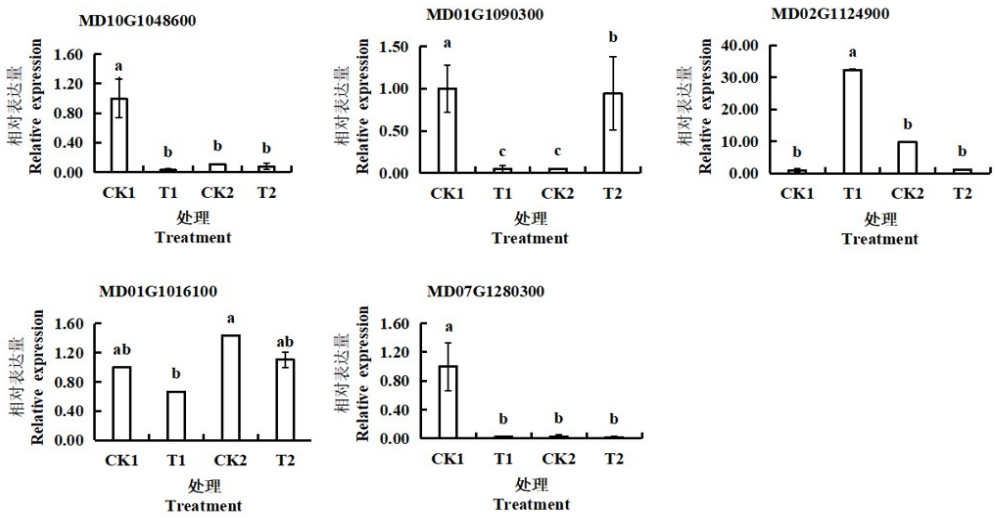


图6　5个生长发育相关基因在苹果花芽中的表达

Fig.6　Expression of five growth and development related genes in apple flower buds

形成;低水平含量的脱落酸有利于花原基形成,促进菠萝进入花芽分化[28-29]。本试验中对苹果园进行间伐改形后,与对照相比花芽分化后期吲哚乙酸含量降低,花芽分化前期和后期的赤霉素和脱落酸含量及花芽分化前期吲哚乙酸含量均明显提高,说明间伐改形能显著提高生长相关激素的含量,这一结果与前人研究中生长素、赤霉素、脱落酸等激素有利于花芽分化的结果一致,进而说明间伐改形对花芽分化有一定促进作用。

转录组学是从RNA水平研究基因表达的情况,即一个活细胞所能转录出来的所有RNA的总和,是一种重要的生命科学研究手段,被广泛应用到多个物种的研究中[30]。本试验中苹果园进行间伐改形后进行花芽转录组学分析,以期找到间伐改形促进苹果树体花芽分化的关键基因,通过转录组分析,在苹果花芽分化2个阶段的GO富集分析中找到激素介导的信号通路,包括生长素相关信号通路、对脱落酸的反应、对赤霉素的反应、吲哚乙酸生物合成过程等通路。在上述激素介导的信号通路中发现大量与吲哚乙酸、赤霉素和脱落酸等信号通路的相关基因,从中挑选出5个表达量变化明显的基因,分别是:gene:MD10G1048600(NAC030)、gene:MD01G1090300(CCA1)、gene:MD02G1124900(PIP2-7)、gene:MD01G1016100(ABR1)、gene:MD07G1280300(WRKY27)。对这5个基因进行qRT-PCR分析,其结果与转录组结果趋势基本一致,进一步证实转录组数据的可信度。

间伐改形是常用的提高苹果果实品质的物理方法,但是间伐改形通过哪些基因发挥作用仍未可知。本研究通过转录组分析获得的5个生长发育相关基因,为以后进一步研究如何促进花芽分化,提高苹果果实坐果率,提高果实品质提供理论基础。转基因已经成为现代育种的重要方向,我们下一步将通过对选出的5个基因进行转基因等领域研究,为培育高质量苹果品种做出贡献。

4　结论

综上所述,郁闭苹果园通过隔间伐改形可以有效提高赤霉素、吲哚乙酸和脱落酸等生长相关激素的含量,进而说明间伐改形对花芽分化有一定促进作用。这种作用可能是通过gene:MD10G1048600(NAC030)、gene:MD01G1090300(CCA1)、gene:MD02G1124900(PIP2-7)、gene:MD01G1016100(ABR1)、gene:MD07G1280300(WRKY27)等基因来调控的。

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Effects of thinning and reshaping on hormone content and related genes in flower buds of a closed apple orchard in eastern Gansu Province

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Abstract: [Objective] In the cultivation of Fuji apples, problems such as difficulty in flower formation , the yield varies greatly in different years are common, and other serious problems especially in confined orchards. This study aimed to investigate the effect of thinning reshaping on the hormone content and related genes in the flower bud differentiation of a closed apple orchards in the Longdong Plateau of Gansu, and provides an experimental basis for further study on the effect of thinning on apple flower bud differentiation in the future. [Methods] In the spring of 2022, this study was conducted on a closed 'Changfu No. 2' apple orchard with 15-year-old trees located in Chengchuan Town, Jingning County, Gansu. Half of the orchard underwent thinning and reshaping, while the other half served as a control and did not undergo thinning and reshaping. Transcriptome analysis and growth-related hormone content determination were performed on flower buds from trees that underwent thinning and reshaping and those that did not. [Results] After thinning and reshaping, the content of gibberellin, indoleacetic acid, and abscisic acid in the flower buds significantly increased. Additionally, a total of 580 genes showed differential expression in the transcriptome analysis. Five growth-related genes were selected for qRT-PCR analysis, and significant upregulation and downregulation were consistent with transcriptome results. [Conclusion] Thinning and reshaping have a significant effect on apple flower bud differentiation, which promote the formation of gibberellin, indoleacetic acid and abscisic acid in flower buds, as well as the expression of genes related to growth and development.

Keywords: Apple, Thinning and reshaping, Flower bud, Transcriptome

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