

不同浓度外源IAA处理对杉木茎部基因表达的影响

杨立伟, 施季森

南京林业大学森林资源与环境学院, 林木遗传与生物技术省部共建·教育部重点实验室, 南京 210037

YANG Li-Wei, SHI Ji-Sen

Key Lab of Forest Genetics and Biotechnology of the Ministry of Education of China, College of Forest Resources and Environment, Nanjing Forestry University, Nanjing 210037, China

- 摘要
- 参考文献
- 相关文章

Download: PDF (902KB) [HTML](#) (385KB) Export: BibTeX or EndNote (RIS) [Supplementary data](#)

摘要 为了揭示吲哚-3-乙酸(Indole-3-acetic acid, IAA)参与杉木木材发育调控的遗传机制, 文章分别以0、3 mg.IAA/g.lanolin处理不同阶段的杉木截顶茎秆作为驱动方(Driver)和测试方(Tester), 利用抑制消减杂交技术(Suppression subtractive hybridization, SSH), 对其中差异表达的目的基因进行了分离和克隆。共获得332个Unigenes, 其潜在的功能分别涉及到细胞组织和生物合成、发育进程调控、电子传递、逆境应答以及信号传导等方面; 进一步地表达鉴定发现CIHIRA、CIPGY1和CIARF4等集中于茎部近轴区域表达的基因, 能够积极地响应外源IAA刺激的维管形成层分裂和管胞分化活动; 而CISM1P1、CITCTP1和CITRN2等集中于茎部远轴区域表达的基因, 则在转录水平上对外源IAA的处理水平及近轴次生维管的发育变化表现出负相关的关系。这一结果表明特异性定位的发育基因对木材形成组织中内源IAA水平变化的差异性识别和响应很可能是生长素参与林木维管形成层次生发育调节的重要分子机制。

关键词: 外源IAA 杉木 茎部 差异表达基因的克隆 表达分析

Abstract: To reveal the potential genetic mechanisms of indole-3-acetic acid (IAA) that regulate Chinese fir wood formation, cloned the differentially expressed genes via suppressive subtractive hybridization (SSH) using the truncated stems treated by 0 and 3 mg IAA/g lanolin as the driver and tester, respectively. A total of 332 unigenes that were involved in cell organization and biosynthesis, developmental processes control, electron transport, stress response, and signal transduction. To further test the results from SSH, we selected those unigenes, whose putative encoding proteins showed significantly homologous with HIRA, PGY1, SMP1, TCT, TRN2, and ARF4, and analyzed their expressed specificity in the wood formative tissues and their response to the secondary developmental changes of vascular cambium stimulated by 0, 1, and 3 mg.IAA/g.lanolin treatment. The results showed that CIHIRA, CIPGY1, and CIARF4, which were specifically expressed in the adaxial zone of stem, were positively response to the activities of cell division and tracheid differentiation stimulated by exogenous IAA treatment. However, CISM1P1, CITCTP1, and CITRN2, which were mainly expressed in the abaxial zones of stems, showed negative correlation with the treated levels of exogenous IAA and activities of vascular cambium secondary development at the transcriptional level. This result showed that the differential response of developmental regulatory genes located in different vascular tissues to the level changes of endogenous IAA in stems is likely to be an important molecular mechanism of auxin regulating wood formation.

Keywords: [exogenous IAA](#), [Cunninghamia lanceolata \(Lamb.\) Hook](#), [stems](#), [the cloning of differentially expressed genes](#), [expression analysis](#)

收稿日期: 2011-09-05; 出版日期: 2012-04-25

基金资助:

国家自然科学基金重点项目(编号: 30930077/C0410)资助

通讯作者 施季森 Email: jshi@njfu.edu.cn

引用本文:

杨立伟, 施季森. 不同浓度外源IAA处理对杉木茎部基因表达的影响. 遗传, 2012, 34(4): 472-484.

YANG Li-Wei, SHI Ji-Sen. Cloning and expression analysis of differentially expressed genes in Chinese fir stems treated by different concentrations of exogenous IAA. HEREDITAS, 2012, V34(4): 472-484.

链接本文:

http://www.chinagene.cn/Jwk_yc/CN/10.3724/SP.J.1005.2012.00472 或 http://www.chinagene.cn/Jwk_yc/CN/Y2012/V34/I4/472

Service

- ▶ 把本文推荐给朋友
- ▶ 加入我的书架
- ▶ 加入引用管理器
- ▶ Email Alert
- ▶ RSS

作者相关文章

- ▶ 杨立伟
- ▶ 施季森

- [2] Sundberg B, Uggla C, Tuominen H. Cambial growth and auxin gradients. In: Savidge R, Barnett J, Napier R, eds. *Cell and Molecular Biology of Wood Formation*. Oxford: BIOS, 2000: 169-188. 
- [3] Savidge RA, Wareing PF. A tracheid-differentiation factor from pine needles. *Planta*, 1981, 153(5): 395-404.
- [4] Savidge RA. The role of plant hormones in higher plant cellular differentiation. II. Experiments with the vascular cambium, and sclereid and tracheid differentiation in the pine, *Pinus contorta*. *Histochem J*, 1983, 15(5): 447-466.
- [5] Little CHA, Savidge RA. The role of plant growth regulators in forest tree cambial growth. *Plant Growth Regul*, 1987, 6(1-2): 137-169. 
- [6] Leitch MA, Savidge RA. Evidence for auxin regulation of bordered-pit positioning during tracheid differentiation in *Larix laricina*. *IAWA J*, 1995, 16(3): 289-297.
- [7] Little CHA, Pharis RP. Hormonal control of radial and longitudinal growth in the tree stem. In: Gartner BL, ed. *Plant Stems: Physiology and Functional Morphology*. San Diego: Academic Press, 1995: 281-319.
- [8] Savidge RA. Intrinsic regulation of cambial growth. *J Plant Growth Regul*, 2000, 20 (1): 52-77.
- [9] Sundberg B, Tuominen H, Little C. Effects of the indole-3-acetic acid (IAA) transport inhibitors N-1-naphthylphthalamic acid and morphactin on endogenous IAA dynamics in relation to compression wood formation in 1-year-old *Pinus sylvestris* (L.) Shoots. *Plant Physiol*, 1994, 106 (2): 469-476.
- [10] Tuominen H, Sitbon F, Jakobsson C, Sandberg G, Olsson O, Sundberg B. Altered growth and wood characteristics in transgenic hybrid aspen expressing *Agrobacterium tumefaciens* T-DNA indoleacetic acid-biosynthesis genes. *Plant Physiol*, 1995, 109(4): 1179-1189.
- [11] Uggla C, Moritz T, Sandberg G, Sundberg B. Auxin as a positional signal in pattern formation in plants. *Proc Natl Acad Sci USA*, 1996, 93(17): 9282-9286. 
- [12] Tuominen H, Puech L, Fink S, Sundberg B. A radial concentration gradient of indole-3-acetic acid is related to secondary xylem development in hybrid aspen. *Plant Physiol*, 1997, 115(2): 577-585.
- [13] Schrader J, Baba K, May ST, Palme K, Bennett M, Bhalerao RP, Sandberg G. Polar auxin transport in the wood-forming tissues of hybrid aspen is under simultaneous control of developmental and environmental signals. *Proc Natl Acad Sci USA*, 2003, 100(17): 10096-100101. 
- [14] Nilsson J, Karlberg A, Antti H, Lopez-Vernaza M, Mellerowicz E, Perrot-Rechenmann C, Sandberg G, Bhalerao RP. Dissecting the molecular basis of the regulation of wood formation by auxin in Hybrid Aspen. *Plant Cell*, 2008, 20(4): 843-855. 
- [15] Chang SJ, Puryear J, Cairney J. A simple and efficient method for isolating RNA from pine trees. *Plant Mol Biol Rep*, 1993, 11(2): 113-116. 
- [16] Schoof H, Zaccaria P, Gundlach H, Lemcke K, Rudd S, Kolesov G, Arnold R, Mewes HW, Mayer KFX. MIPS *Arabidopsis thaliana* Database (MATDB): an integrated biological knowledge resource based on the first complete plant genome. *Nucleic Acid Res*, 2002, 30(1): 91-93. 
- [17] Fisher K, Turner S. PXY, a receptor-like kinase essential for maintaining polarity during plant vascular-tissue development. *Curr Biol*, 2007, 17(12): 1061-1066. 
- [18] Hirakawa Y, Shinohara H, Kondo Y, Inoue A, Nakanomyo I, Ogawa M, Sawa S, Ohashi-Ito K, Matsubayashi Y, Fu-kuda H. Non-cell-autonomous control of vascular stem cell fate by a CLE peptide/receptor system. *Proc Natl Acad Sci USA*, 2008, 105(39): 15208-15213. 
- [19] Etchells JP, Turner SR. The PXY-CLE41 receptor ligand pair defines a multifunctional pathway that controls the rate and orientation of vascular cell division. *Development*, 2010, 137(5): 767-774.
- [20] Ji JB, Strable J, Shimizu R, Koenig D, Sinha N, Scanlon MJ. WOX4 promotes procambial development. *Plant Physiol*, 2010, 152(3): 1346-1356. 
- [21] Hirakawa Y, Kondo Y, Fukuda H. TDIF peptide signaling regulates vascular stem cell proliferation via the WOX4 homeobox gene in *Arabidopsis*. *Plant Cell*, 2010, 22(8): 2618-2629. 
- [22] Schrader J, Nilsson J, Mellerowicz E, Berglund A, Nilsson P, Hertzberg M, Sandberg G. A high-resolution transcript profile across the wood-forming meristem of poplar identifies potential regulators of cambial stem cell identity. *Plant Cell*, 2004, 16(9): 2278-2292. 
- [23] Mele G, Ori N, Sato Y, Hake S. The knotted1-like homeobox gene *BREVIPEDICELLUS* regulates cell differentiation by modulating metabolic pathways. *Genes Dev*, 2003, 17(17): 2088-2093. 
- [24] Du J, Mansfield SD, Groover AT. The *Populus* homeobox gene *ARBORKNOX2* regulates cell differentiation during secondary growth. *Plant J*, 2009, 60(6): 1000-1014. 
- [25] Schneeberger R, Tsiantis M, Freeling M, Langdale JA. The rough sheath2 gene negatively regulates homeobox gene expression during maize leaf development. *Development*, 1998, 125 (15): 2857-2865.
- [26] Waites R, Selvadurai HRN, Oliver IR, Hudson A. The *PHANTASTICA* gene encodes a MYB transcription factor involved in growth and dorsoventrality of lateral organs in *Antirrhinum*. *Cell*, 1998, 93(5): 779-789.
- [27] Timmermans MCP, Hudson A, Becraft PW, Nelson T. ROUGH SHEATH2: A Myb protein that represses knox homeobox genes in maize lateral organ primordia. *Science*, 1999, 284(5411): 151-153.
- [28] Tsiantis M, Schneeberger R, Golz JF, Freeling M, Langdale JA. The maize *rough sheath2* gene and leaf development programs in monocot and

- [29] Byrne ME, Barley R, Curtis M, Arroyo JM, Dunham M, Hudson A, Martienssen RA. Asymmetric leaves1 mediates leaf patterning and stem cell function in *Arabidopsis*. *Nature*, 2000, 408 (6815): 967-971.
- [30] Ori N, Eshed Y, Chuck G, Bowman JL, Hake S. Mechanisms that control *knox* gene expression in the *Arabidopsis* shoot. *Development*, 2000, 127(24): 5523-5532.
- [31] McHale NA, Koning RE. *PHANTASTICA* regulates development of the adaxial mesophyll in *Nicotiana* leaves. *Plant Cell*, 2004, 16(5): 1251-1262. 
- [32] Phelps-Durr TL, Thomas J, Vahab P, Timmermans MCP. Maize rough sheath2 and its *Arabidopsis* orthologue ASYMMETRIC LEAVES1 interact with HIRA, a predicted histone chaperone, to maintain *knox* gene silencing and determinacy during organogenesis. *Plant Cell*, 2005, 17(11): 2886-2898. 
- [33] Spector MS, Raff A, DeSilva H, Lee K, Osley MA. Hir1p and Hir2p function as transcriptional corepressors to regulate histone gene transcription in the *Saccharomyces cerevisiae* cell cycle. *Mol Cell Biol*, 1997, 17(2): 545-552.
- [34] Kaufman PD, Cohen JL, Osley MA. Hir proteins are required for position-dependent gene silencing in *Saccharomyces cerevisiae* in the absence of chromatin assembly factor 1. *Mol Cell Biol*, 1998, 18(8): 4793-4806.
- [35] Magnaghi P, Roberts C, Lorain S, Lipinski M, Scambler PJ. HIRA, a mammalian homologue of *Saccharomyces cerevisiae* transcriptional co-repressors, interacts with Pax3. *Nat Genet*, 1998, 20(1): 74-77. 
- [36] Sharp JA, Fouts ET, Krawitz DC, Kaufman PD. Yeast histone deposition protein Asf1p requires Hir proteins and PCNA for heterochromatic silencing. *Curr Biol*, 2001, 11(7): 463-473. 
- [37] Roberts C, Sutherland HF, Farmer H, Kimber W, Halford S, Carey A, Brickman JM, Wynshaw-Boris A, Scambler PJ. Targeted mutagenesis of the *Hira* gene results in gastrulation defects and patterning abnormalities of mesodermal derivatives prior to early embryonic lethality. *Mol Cell Biol*, 2002, 22(7): 2318-2328. 
- [38] McConnell JR, Emery J, Eshed Y, Bao N, Bowman J, Barton MK. Role of *PHABULOSA* and *PHAVOLUTA* in determining radial patterning in shoots. *Nature*, 2001, 411 (6838): 709-713.
- [39] Kerstetter RA, Bollman K, Taylor RA, Bomblies K, Poethig RS. *KANADI* regulates organ polarity in *Arabidopsis*. *Nature*, 2001, 411(6838): 706-709.
- [40] Emery JF, Floyd SK, Alvarez J, Eshed Y, Hawker NP, Izhaki A, Baum SF, Bowman JL. Radial patterning of *Arabidopsis* shoots by class III HD-ZIP and *KANADI* genes. *Curr Biol*, 2003, 13(20): 1768-1774. 
- [41] Caño-Delgado A, Lee JY, Demura T. Regulatory mechanisms for specification and patterning of plant vascular tissues. *Annu Rev Cell Dev Biol*, 2010, 26: 605-637. 
- [42] Zhong RQ, Ye ZH. Regulation of HD-ZIP III genes by MicroRNA 165. *Plant Signal Behav*, 2007, 2(5): 351-353. 
- [43] Pinon V, Etchells JP, Rossignol P, Collier SA, Arroyo JM, Martienssen RA, Byrne ME. Three *PIGGYBACK* genes that specifically influence leaf patterning encode ribosomal proteins. *Development*, 2008, 135(7): 1315-1324.
- [44] Gagne JM, Downes BP, Shiu SH, Durski AM, Vierstra RD. The F-box subunit of the SCF E3 complex is encoded by a diverse superfamily of genes in *Arabidopsis*. *Proc Natl Acad Sci USA*, 2002, 99(17): 1519-1524.
- [45] Gray WM, Estelle M. Function of the ubiquitin-proteasome pathway in auxin response. *Trends Biochem Sci*, 2000, 25(3): 133-138. 
- [46] Hellmann H, Hobbie L, Chapman A, Dharmasiri S, Dharmasiri N, del Pozo C, Reinhardt D, Estelle M. *Arabidopsis AXR6* encodes CUL1 implicating SCF E3 ligases in auxin regulation of embryogenesis. *EMBO J*, 2003, 22(13): 3314-3325. 
- [47] Hobbie L, McGovern M, Hurwitz LR, Pierro A, Liu NY, Bandyopadhyay A, Estelle M. The *axr6* mutants of *Arabidopsis thaliana* define a gene involved in auxin response and early development. *Development*, 2000, 127(1): 23-32.
- [48] Cope GA, Suh GS, Aravind L, Schwarz SE, Zipursky SL, Koonin EV, Deshaies RJ. Role of predicted metalloprotease motif of Jab1/Csn5 in cleavage of Nedd8 from Cul1. *Science*, 2002, 298(5593): 608-611.
- [49] Worley CK, Zenser N, Ramos J, Rouse D, Leyser O, Theologis A, Callis J. Degradation of Aux/IAA proteins is essential for normal auxin signalling. *Plant J*, 2000, 21(6): 553-562. 
- [50] Karin M, Ben-Neriah Y. Phosphorylation meets ubiquitination: the control of NF-[kappa]B activity. *Annu Rev Immunol*, 2000, 18(8): 621-663. 
- [51] Scarpella E, Meijer AH. Pattern formation in the vascular system of monocot and dicot plant species. *New Phytol*, 2004, 164(2): 209-242. 
- [1] 黄国文 韩玉珍 傅永福.拟南芥SUA41基因的表达和功能分析[J].遗传, 2013,35(1): 93-100
- [2] 焦莎莎, 刘卡, 李刚, 高剑峰, 马润林.绵羊MHC区段3个预测基因的验证与表达分析[J].遗传, 2011,33(12): 1353-1358
- [3] 鲍永美, 刘永惠, 许冬清, 黄骥, 王州飞, 王建飞, 张红生.水稻Qb-SNARE蛋白OsNPSN11多克隆抗体制备、鉴定与应用[J].遗传, 2010,32(9): 961-965
- [4] 陶倩怡, 李征, 何欢乐, 潘俊松, 蔡润.黄瓜单性花决定基因M的表达分析[J].遗传, 2010,32(6): 632-638

- [5] 杜金芳, 曾勇庆, 陈伟, 崔景香, 陈其美, 杨伦, 胡艳霞. 猪CuZnSOD基因的克隆、表达及功能分析[J]. 遗传, 2010, 32(10): 1037-1042
- [6] 柳展基, 邵凤霞, 唐桂英, 单雷, 毕玉平. 一个新的玉米NAC类基因(*ZmNAC1*)的克隆与分析[J]. 遗传, 2009, 31(2): 199-205
- [7] 喻达时, 赵琼, 邓克勤, 郭新红. 拟南芥CK1A基因功能初步研究[J]. 遗传, 2009, 31(10): 1037-1042
- [8] 任洪林, 徐丹丹, 乔琨, 蔡灵, 黄伟滨, 张鼐, 王克坚. 细菌攻毒杂色鲍血淋巴细胞抑制性差减杂交文库构建及巨噬细胞表达蛋白cDNA的克隆与差异表达[J]. 遗传, 2008, 30(8): 1043-1050
- [9] 何建锋, 张彦明, 段会娟, 曹金锁, 张德礼. 人天然免疫基因*BCL10*的猪同源物的识别、克隆与初步表达分析[J]. 遗传, 2008, 30(6): 747-754
- [10] 林凡云, 陆琼娴, 徐剑宏, 史建荣. 两个与盐和赤霉病菌胁迫相关的小麦糖基转移酶基因的克隆与表达[J]. 遗传, 2008, 30(12): 1608-1614
- [11] 刘明坤, 刘关君, 魏志刚, 阎秀峰, 曲春浦, 王垠, 刘桂丰, 杨传平. 西伯利亚蓼半胱氨酸合成酶基因的克隆与表达[J]. 遗传, 2008, 30(10): 1363-1371
- [12] 郭书巧, 黄骥, 江燕, 张红生. 水稻C2H2型锌指蛋白基因*RZF71*的克隆与表达分析[J]. 遗传, 2007, 29(5): 607-607-613
- [13] 王桂凤, 高燕, 杨立伟, 施季森. 杉木木材形成过程特异表达基因的鉴定与分析[J]. 遗传, 2007, 29(4): 483-489
- [14] 周洲, 张德强, 卢孟柱. 毛白杨乙酰-乙酰载体蛋白硫脂酶基因(PtFATB)的克隆与表达分析[J]. 遗传, 2007, 34(3): 267-274
- [15] 侯夫云, 黄骥, 陆驹飞, 王州飞, 张红生. 水稻质体葡萄糖-6-磷酸脱氢酶基因的克隆与表达研究[J]. 遗传, 2006, 33(5): 441-448