青年论坛论文摘要

流感病毒致心脏出生缺陷发生的机制研究

李芳，刘宪楚，袁婺洲，莫小阳，吴秀山

湖南师大心脏发育研究中心，长沙，中国

邮箱

li-evans@hotmail.com（李芳）

摘要

先天性心脏病（先心病）是严重危害人类健康的疾病，新生儿发病率为4-50‰。目前认为心脏结构发育畸形主要由遗传和环境因素及其相互作用所致，而流感病毒是一种重要的致畸环境因素。流行病学临床调查显示孕妇宫内感染流感病毒可以增加胎儿心脏发育畸形的风险，但其分子机制仍不清楚。本研究以孕鼠为模式动物，在胚胎发育早期用流感病毒感染孕母鼠，观察到新生小鼠出现房室间隔缺损等表型缺陷。通过RNA-seq技术筛选流感病毒H1N1感染怀孕母鼠早期胚胎心脏组织的差异表达基因，得到表达下调显著的Nkx2.5等基因；并进一步在小鼠模型中通过qPCR和WB验证了测序筛选的结果。利用全基因组的CHIP-seq分析，筛选到能够与H1N1的NS1蛋白发生结合的141个区域。其中，19个结合区域距离最近的基因为心脏功能基因。接下来，我们采用qPCR检测该19个心脏功能基因在孕鼠胚胎心脏中的表达。实验结果表明流感病毒H1N1感染可能通过调控心脏发育基因表达从而导致胚胎心脏出现发育缺陷。

关键词

流感病毒H1N1，先心病，心脏功能基因

SciencePG
Science Publishing Group

EBP1 Nuclear Accumulation Negatively Feeds Back on FERONIA-Mediated RALF1 Signaling

Chiyu Li[1, *], Xuanming Liu[1, *], Xiaonan Qiang[1], Xiaoyan Li[1], Xiushan Li[1], Sirui Zhu[1], Long Wang[1], Yuan Wang[2], Hongdong Liao[1], Sheng Luan[2], Feng Yu[1, 3]

[1]State Key Laboratory of Chemo/Biosensing and Chemometrics, College of Biology, Hunan Province Key Laboratory of Plant Functional Genomics and Developmental Regulation, Hunan University, Changsha, P.R. China

[2]Department of Plant and Microbial Biology, University of California, Berkeley, USA

[3]State Key Laboratory of Hybrid Rice, Hunan Hybrid Rice Research Center, Changsha, China

Email address:

feng_yu@hnu.edu.cn (Feng Yu)

[*]Corresponding author

Abstract

FERONIA (FER), a plasma membrane receptor-like kinase, is a central regulator of cell growth that integrates environmental and endogenous signals. A peptide ligand RALF1 binds to FER and triggers a series of downstream events, including inhibition of H^+-ATPase activity at the cell surface and regulation of gene expression in the nucleus. We report here that, upon RALF1 binding, FER first promotes *ErbB3 binding protein 1* (*EBP1*) mRNA translation, then interacts with and phosphorylates the EBP1 protein, leading to EBP1 accumulation in the nucleus. There, EBP1 associates with the promoters of previously identified RALF1-regulated genes, such as *CML38*, and regulates gene transcription in response to RALF1 signaling. Interestingly, EBP1 appears to inhibit the RALF1 peptide response, thus forming a transcription-translation feedback loop (TTFL) similar to that found in circadian rhythm control. The plant RALF1-FER-EBP1 axis is reminiscent of animal epidermal growth factor receptor (EGFR) signaling, in which EGF peptide induces EGFR to interact with and phosphorylate EBP1, promoting EBP1 nuclear accumulation to control cell growth. Thus, we suggest that, in response to peptide signals, plant FER and animal EGFR use the conserved key regulator EBP1 to control cell growth in the nucleus.

Keywords

FERONIA, Peptide, Cell Growth, Signal Transduction

Molecular Mechanism and Inhibitory Targets of Dioscin in HepG2 Cells

Ying-Shuo Zhang, Yi-Long Ma, Kiran Thakur, Sayed Sajid Hussain, Jun Wang, Qi Zhang, Jian-Guo Zhang, Zhao-Jun Wei

Hefei University of Technology, China

Email address:

zjwei@hfut.edu.cn (Zhaojun Wei)

Abstract

Dioscin has been known for its anti-cancer activity; however, its detailed molecular mechanisms have not been studied so far. Herein, we evaluated the anti-cancer activity of dioscin for proliferation inhibition and apoptosis in HepG2 cancer cells. Initially, dioscin was purified and identified from Polygonatum sibiricum by HPLC, MS, and NMR analysis, respectively. Dioscin inhibited the cell multiplication at IC_{50} of 8.34 μM, altered the cell morphology, arrested the cell cycle in G2/M phase and led to considerable programmed cell death. Furthermore, it has efficiently promoted the mitochondrial pathway and death receptor pathway. The inhibition of Caspase-8 and Caspase-9 proteins in these pathways abolished the dioscin induced apoptosis significantly; while dioscin inhibited the PI3K/Akt/mTOR pathway. Moreover, dioscin exposure led to enhanced intracellular ROS generation and the mRNA expression of JNK gene which emphasized their involvement in the apoptosis process in HepG2 cells.

Genomic Analysis of the Relationship of Restorer Line Yazhan

Li Shuang, He Haohua, Peng Xiaosong

Key Laboratory of Crop Physiology, Ecology and Genetic Breeding, Ministry of Education, College of Agronomy, JAU, Nanchang, China

Email address:

Pxs63@163.com (Peng Xiaosong)

Abstract

Huazhan is the most extensive hybrid rice variety in China. Yazhan was selected systematically from Zhanhui 15 (as a female parent) and Huazhan (as a male parent) ,As a derivative restorer line of Huazhan, it is similar to Huazhan in many characters. In this study, 988 microsatellite markers covering 12 chromosomes of rice were first used to screen 103 polymorphic markers, The source map of the Yazhan genome was constructed by the QTL IciMapping. Based on the reference genome of Nipponbare, the genetic laws of parental traits were analyzed through the second generation resequencing of Zhanhui15, Huazhan and Yazhan. Finally, the yield and quality agronomic characters of the three restorer lines were investigated. The results show that among 103 polymorphic markers,the same type has 54 pairs between Huazhan and Yazhan, and 37 between Zhanhui15 and Yazhan, 12 markers of unique type were yachan..Among the three restorer lines, 89.6% was no polymorphism, Yazhan specific segments has 1.21% ,and Polymorphism rate with Huazhan has 3.74%,and 5.47% with Zhanhui15. In the genome of Yazhan, Part of the fragment of chromosome 1 and 4 was mainly derived from Zhanhui 15, while Part of the fragment of chromosome 2, 8, 9, 11 and 12 were mainly derived from Huazhan. The Part of the fragment of chromosome 5, 6 and 7 were derived from both parents, among which chromosome 6 had the highest polymorphism. Zhanhui15, Huazhan, Yazhan SNP density were 5.71, 5.58 and 5.6 SNP/ KB, respectively; the Indel density was 1.06, 1.08 and 1.12 Indel/ KB, respectively; the SNP and Indel density Shared by the three restorer lines were 5.07 SNP/ KB and 0.1 Indel/ KB, respectively. The SNP and Indel density Shared by Yazhan and Huazhan were 0.29, 0.21 SNP/ KB, Yazhan and Zhanhui15 were 0.08 and 0.06 Indel/ KB, indicating that the genetic background of Yazhan and Huazhan were more similar. There were significant differences in plant height, spike length and primary branch number between Yazhan and Zhanhui 15, and only in spike length between Yazhan and Zhanhui15. The percentage of brown rice, white rice and whole rice was lower than that of both parents, while the index of chalkiness was better than that of both parents. Therefore, according to the results of SSR marker genomic scanning and second-generation resequencing, combined with the analysis of phenotypic data, Yazhan, as the descendants of Zhanhui15 and Huazhan, preferred the improved selection of paternal traits in the systematic breeding process, and the genetic basis of the genome was more inclined to Huazhan. The results provide a theoretical reference for elucidating the genetic composition of Yazhan and determining the main genetic source of its important agronomic traits.

Keywords

Yazhan, Genome, SSR

Identification of QTL for Cadmium Tolerance at Seedling Stage of Rice (Oryza Sativa L.)

Li Weixing[1], Ouyang Linjuan[2], Wen Wen[3]

Key Laboratory of Crop Physiology, Ecology and Genetics and Breeding, Ministry of Education, Jiangxi Agricultural University, Nanchang, China

Email address:

1316376821@qq.com (Li Weixing)

Abstract

For QTL mapping of rice seedling resistance to cadmium stress, the experiment selected in hainan harvest seeds, in indica rice varieties chang 121 as the recurrent parent, japonica rice varieties is more light as the donor parent of chromosome segment substitution (CSSL) group, a total of 97 strains and parents as material, and in July and August in the same test by the same material, respectively. To join 17 mg/L CdCl2 solution as the experimental group, with no CdCl2 solution as the control group, examine its seedling dry weight of rice seedling, MiaoXian weight, seedling length, root dry weight, root fresh weight, root length, such as character, translates into resistance index, used in the evaluation index of rice seedling resistance to cadmium stress resistance. According to the experimental results show that the experiment detected in July 5 QTL associated with resistance to cadmium, at 8, 9, 10 and 12 chromosomes, with long resistance index as an index, detected two QTL associated with resistance to cadmium, are located on chromosome 9, explains respectively 14.26%, 22.5234% of the phenotypic variation; Based on the resistance index of seedlings, a QTL with cadmium resistance was detected, which was located on chromosome 12, which explained the phenotypic variation of 17.1246%. Based on the index of root dry weight resistance index, a QTL related to cadmium resistance was detected, which was located on chromosome 10, explaining the phenotypic variation of 12.30%. In seedling dry weight resistance index as the index, detected a QTL associated with resistance to cadmium, its located on chromosome 8, explained 13.95% of the phenotypic variation, was detected in the experiment of the communist party of China in August 3 QTL associated with milk cadmium, at 1, 4 on chromosome, with long resistance index as an index, detected in chromosome 11 QTL associated with resistance to cadmium, which explained 12.767% of the phenotypic variation; On the fourth chromosome, a QTL related to cadmium resistance was detected on the fourth chromosome, and the phenotypic variation of 12.1927% was explained. The phenotypic variation of 22.7715% was explained in the 4th chromosome, which was detected by the index of root fresh resistance index.

Keywords

Rice, Cadmium Stress, Quantitative Traits

Comparative Genomic Analysis of the IDD Genes in Five Rosaceae Species and Expression Analysis in Chinese White Pear (Pyrus Bretschneideri)

Xueqiang Su†, Yu Zhao†, Guohui Li, Xi Cheng, Muhammad Abdullah, Xu Sun, Dahui Li, Yi Lin, Yongping Cai*

School of Life Science, Anhui Agricultural University, Hefei, China

Email address:

992547404@qq.com (Xueqiang Su)

*Corresponding author

† Xueqiang Su and Yu Zhao are co-first authors.

Abstract

The INDETERMINATE DOMAIN (IDD) gene family encodes hybrid transcription factors with distinct zinc finger motifs and appears to be found in all higher plant genomes. In the model plants Arabidopsis and rice, IDD genes have been identified throughout the genome, and the functions of many members have been studied. However, this gene family has been studied less in Rosaceae species, among which genome-wide identification has only been completed in apple, while there is still no comprehensive research in pear. This study focuses on comparative genomic analysis of the IDD gene family in five Rosaceae species (pear, strawberry, plum, raspberry and cherry). We identified a total of 68 IDD genes: 16 genes in Chinese pear (Pyrus bretschneideri), 14 genes in strawberry (Fragaria vesca), 13 genes in plum (Prunus mume), 14 genes in raspberry (Rubus occidentalis) and 11 genes in cherry (Prunus avium). The evolution of the IDD genes of these five Rosaceae species was revealed by constructing a phylogenetic tree, tracking gene duplication events, sliding window analysis, and conserved microsynteny analysis. The expression analysis of different tissues showed that most of the pear IDD genes had a very high transcription level in fruit, flower, and bud. Combining our results with previous research, we speculate that PbIDD2 and PbIDD8 may participate in plant flowering induction in pear. Temporal expression analysis showed that the expression patterns of PbIDD3 and PbIDD5 were completely opposite to the accumulation pattern of fruit lignin and stone cell contents. Combining the results of the composite phylogenetic tree and expression pattern analysis showed that PbIDD3 and PbIDD5 might be involved in the metabolism of lignin and secondary cell wall (SCW) formation. In summary, we provide basic information about the IDD genes of five Rosaceae species, providing a theoretical basis for deeper study of the functions of these genes.

Keywords

INDETERMINATE DOMAIN (IDD) Genes, Pear, Phylogenetic Analysis, Microsynteny, Lignin Synthesis, SCW Formation

Characterization of the Numts in the Japanese Quail (*Coturnix japonica*) Nuclear Genome and Its Application into Phylogeny of Galliformes

Ding Hengwu[1, 2], Dong Jinxiu[1, 2], Bi De[1, 2], Jiang Lan[1, 2], Wu Xuan[1, 2], Zhu Ran[1, 2], Zeng Juhua[1, 2], Kan Xianzhao[1, 2, *]

[1]The Institute of Bioinformatics, College of Life Sciences, Anhui Normal University, Wuhu, China

[2]The Provincial Key Laboratory of the Conservation and Exploitation Research of Biological Resources in Anhui, Wuhu, China

Email address:

xianzhao@ahnu.edu.cn (Kan Xianzhao)

[*]Corresponding author

Abstract

The nuclear copy of mitochondria DNA fragments (numts) have been detected in the nuclear genome of many eukaryotes. In this study, we used the LAST program to detect the numts in the nuclear genome of Japanese quail (*Coturnix japonica*) and these numts were verified by PCR. The sequence characterization of numts were analyzed using bioinformatic methods. To discuss the phylogenetic implications of numts in Galliformes, the BLASTN program was used to search the orthologous numts in other Galliformes species with nuclear genome and we reconstructed the evolutionary trees using maximum parsimony (MP), maximum likelihood (ML) and bayesian inference (BI) method. Results demonstrated that 25 numt hits and 15 numt regions have been detected and verified in the nuclear genome of Japanese quail. We detected 2 orthologous numts in Galliformes and one numt type can even found in Galloanserae. All numts have been detected in the non-coding regions with high A+T content. The identities between numts and mitochondrial sequences varied from 66.24 to 100%, and the length of numts ranged from 55 to 4348. The molecular phylogenetic analysis indicated that numts had been evolved independently after their insertion. Phylogenetic tree based on orthologous numts have similar topological structure with the tree based on corresponding mitochondrial DNA.

Keywords

Nuclear Copy of Mitochondria DNA Fragments (Numts), Japanese Quail, Galliformes, Phylogeny

Detection and Comparative the miRNA Expression Profiles in the Domestic Gonadal Pigeon (*Columba livia*)

Jiang Lan[1,2], Wu Xuan[1,2], Ding Hengwu[1,2], Zhu Ran[1,2], Zeng Juhua[1,2], Bi De[1,2], Kan Xianzhao[1,2,*]

[1]The Institute of Bioinformatics, College of Life Sciences, Anhui Normal University, Wuhu, China

[2]The Provincial Key Laboratory of the Conservation and Exploitation Research of Biological Resources in Anhui, Wuhu, China

Email address:

xianzhao@ahnu.edu.cn (Kan Xianzhao)

[*]Corresponding author

Abstract

The domestic pigeon (*Columba livia*), is a widespread economic animal in China, which could create substantial economic benefits. Little was known about pigeon miRNAs or their functional profiles from the pigeon genome annotation. Recent studies demonstrated that changes in the expression of miRNAs are associated with sex. In this study, miRNAs were extracted from the gonads of four healthy adult pigeons and sequenced by Illumina next-generation sequencing technology. As determined by miRNA screening, a total of 304 conserved miRNAs and 134 novel putative miRNAs candidates were detected. After differential expression analysis, we noticed that the expressions of 5 miRNAs were significantly up-regulated in young giant pandas compared with that of adults. Moreover, 109 miRNAs were only detected in female pigeons and 36 in the male individuals. Target gene prediction suggested that the miRNAs of giant panda might be relevant to the expressions of 5761 downstream genes. Subsequently, the predicted target genes were conducted to GO and KEGG enrichment analyses, and we found that some miRNAs could target sex-related genes were mainly involved in gonadal development. In conclusion, our results provide the first miRNA profiles, and the predicted functional analyses may open an avenue for the further study of pigeon gonads.

Keywords

Pigeon, Ovary, Testis, miRNA, Gonad

乌龟血清对非小细胞肺癌的抑制作用

咎嘉伟，江慧，聂刘旺

生命科学学院细胞生物学实验组，安徽师范大学，芜湖，中国

邮箱

lwnie@mail.ahnu.edu.cn（聂刘旺）

摘要

肺癌是一种常见的肺部恶性肿瘤，也是世界上最主要的致死性癌症之一，寻找能够有效靶向诱导肺癌细胞凋亡的药物，已成为肺癌治疗的研究重点。乌龟具有特殊的营养滋补和药用价值，是一种优质的药物来源，充分开发利用乌龟制备抗肿瘤药物具有重要的研究意义。本文以乌龟血清为原料，探讨其抑制非小细胞肺癌的增值、诱导其凋亡的作用及分子机制。在体外实验中，使用乌龟血清含量不同的培液处理H1299和A549细胞，CCK-8法的结果显示，与空白对照组相比，培液中乌龟血清含量为1%、2.5%、5%、7.5%、10%组H1299和A549细胞活力显著降低（$P<0.01$），依据实验结果，后续实验将1%、5%、10%浓度作为作用H1299和A549细胞的实验浓度。DAPI结果显示，空白对照组细胞核的形态为规则卵圆形，染色均匀，为深蓝色；随着乌龟血清含量提高，H1299和A549细胞胞核开始出现浓缩，碎裂，染色加深，视野中可见月牙形细胞核固缩的调往形态。Annexin V-FITC /PI流式细胞术结果显示，随着乌龟血清含量的增加，H1299和A549细胞的凋亡率均有所上升。Western bloting实验结果显示，空白对照组H1299和A549细胞的增殖相关信号通路Akt/mTOR信号通路处于磷酸化活化状态。与空白对照组比较，各个含量的乌龟血清均能抑制这两种细胞的Akt/mTOR信号通路的磷酸化。无论是外源性死亡途径还是内源性死亡途径，都涉及到半胱氨酸蛋白酶家族的切割活化，进而引起其底物PARP的切割，因此我们通过Western bloting实验检测H1299和A549细胞内凋亡相关蛋白的变化。在空白对照组，Cleaved caspase-3基本无表达，未见PARP切割条带。而在乌龟血清处理组，随着乌龟血清含量的增加，Cleaved caspase-3表达量逐渐增加， Pro-caspase-3蛋白水平逐渐减小，PARP切割条带显著上调，进一步证实了乌龟血清对这两种细胞的诱导凋亡作用。因此，我们认为乌龟血清可以通过抑制细胞存活相关信号通路，抑制细胞增殖，诱导细胞凋亡。

关键字

乌龟血清，非小细胞肺癌细胞，细胞凋亡，Akt，mTOR

Coevolution of Positively Selected Izumo1, JUNO and CD9 in Turtles: Implies the Possibility Reasons of Interspecies Hybridization

Jingxiu Dong, Yuantian Bao, Hui Jiang, Lei Xiong, Jiawei Zan, Jianjun Liu, Liuwang Nie*

Life Science College, the Provincial Key Lab of the Conservation and Exploitation Research of Biological Resources in Anhui, Anhui Normal University, Wuhu, Anhui, China

Email address:

lwnie@mail.ahnu.edu.cn (Liuwang Nie)

*Corresponding author

Abstract

The phenomenon of turtle interspecific hybrids is very common, and there is not only close hybridization, but also distant hybridization. Although it is not clear why it happened so frequently, it is certain that a successful recognition and combination of sperm and eggs is a necessary condition for hybridization. In recent years, three proteins have been found are essential for sperm-egg binding in vertebrate, are Izumo1 on sperm surface, JUNO and CD9 on egg surface, respectively. Here we amplified, cloned and sequenced *Izumo1*, *JUNO* and *CD9* in six turtles, which are *Mauremys reevesii*, *M. mutica*, *M. sinensis*, *Cistoclemmys flavomarginata*, *Platysternon megacephalum* and *Chrymeys picta bellii*, then constructed phylogenetic trees and analyzed the evolution of *Izumo1*, *JUNO* and *CD9* in a group of turtles. And through bioinformatic analyses, *Izumo1* and *JUNO* displayed tissue specific expression, and the egg protein CD9 and JUNO coevolve with the sperm protein Izumo1, suggesting a genetic interaction occurs between them. In addition, the results showed that high similarity of amino acid sequence of these three proteins in *Mauremys reevesii*, *M. mutica*, *M. sinensis*, *C. flavomarginata*, and clustered into one branch in the evolutionary tree. However, *P. megacephalum* and *C. picta bellii* can't cross with them, the sequence identity is obviously lower, and the phylogenetic tree is not cluster in a clade. So, a possible reason based on the coevolution can be inferred for why turtles can hybrid whether in closely related species or in distantly related species.

Keywords

Sperm-egg Fusion, Izumo1, JUNO, CD9, Hybridization

Comparative Sequencing and Analysis of Intestinal Bacteria Between the Silkworm *Bombyx mori* and *Diaphania pyloalis* Walker

Tongtong Sun, Liangli Yang, Dandan Xia, Xinwei Zhang, Ting Xue, Yan Meng*

School of Life Sciences, Anhui Agricultural University, Hefei, China

Email address:

mengyan@ahau.edu.cn (Yan Meng)

*Corresponding author

Abstract

The alkaloids of mulberry leaves have a toxic effect on non-mulberry feeding insects, but have no effect on mulberry-feeding insects such as the silkworm *Bombyx mori* and *Diaphania pyloalis* walker. It is speculated that *B. mori* and *D. pyloalis* may use the β-fructofuranosidase (β-FFase) expressed in larval intestine to avoid the toxic effects of mulberry sugar-mimic alkaloids. BmSUC1 (β-FFase of *B. mori*) showed enzymatic activity both *in vivo* and *in vitro*. DpSUC1a was similarly identified in *D. pyloalis* larval midgut where displayed marked β-FFase activity, however, recombinant DpSUC1a had no activity *in vitro*, obviously different from BmSUC1. More and more intestinal microorganisms have recently been identified and play an important role in insect growth and development, such as insect digestion, resistance to disease and toxic substances from plant. In fact, β-FFases exist widely in bacteria, fungi and plants, but rarely in animals. We hypothesize that the defense against mulberry alkaloids in *D. pyloalis* might be associated with the intestinal microbes by their β-FFases.

In this study, larvae of *B. mori* (wild type and transgenic RNAi-*BmSuc1* strains) and *D. pyloalis* were used to explore the difference of bacterial population in intestines by 16S rDNA diversity sequencing. Our primary analyses showed that dominant bacteria phylum in *D. pyloalis* was *Firmicutes*, the same as RNAi-*BmSuc1* silkworm, while that in the wild-type silkworm was *Proteobacteria*. From the level of genus, dominant bacteria in *D. pyloalis* and RNAi-*BmSuc1* silkworm were *Enterococcus* and *Tyzzerella*, respectively (both belonging to *Firmicutes*), while that in the wild-type silkworm was *Methylobacterium* (*Proteobacteria*). The results suggest that the intestinal bacterial flora in *D. pyloalis* is naturally diverse from *B. mori* and down regulation of *BmSuc1* has changed its bacterial dominance to be similar to *D. pyloalis*. More comparative analyses are in progress.

Keywords

β-Fructofuranosidase, Intestinal Microorganisms, 16S rDNA Sequencing, *Diaphania pyloalis*

nm23-H1与MMP-2/MMP-9、β-catenin在子宫内膜异位症中的表达及其意义

曹慧茹[1]，郭耀省[2]，王玮[2]，李铁臣[2]，孙青[*]

1弋矶山医院妇产科，皖南医学院，芜湖，中国
2分子生物学研究室，皖南医学院，芜湖，中国

邮箱
469970694@qq.com（曹慧茹），sunqingl@yahoo.com.cn（孙青）
[*]通讯作者

摘要

目的：探讨*nm23-H1*及*MMP-2/MMP-9*、*β-catenin*在子宫内膜异位症（EMs）组织、上皮细胞及基质细胞中的表达及其意义。

方法：收集51例正常子宫内膜和45例EMs异位组织。1. 采用qRT-PCR检测*nm23-H1*基因的mRNA表达水平以及Western Blot检测*nm23-H1*的蛋白表达水平；2. 采用激光捕获显微切割技术（LCM）捕获子宫内膜上皮细胞，qRT-PCR检测*nm23-H1*基因的mRNA表达水平。3. 将正常和异位子宫内膜组织进行原代培养，（1）qRT-PCR测定*nm23-H1*、*MMP-2/MMP-9*以及*β-catenin*的表达；（2）用17β-雌二醇（终浓度10^{-6} mol/L）刺激48h后，qRT-PCR测定*nm23-H1*及*MMP-2/MMP-9*、*β-catenin*的表达；（3）*nm23-H1* siRNA（50nM）转染原代细胞，培养48h后，qRT-PCR测定*nm23-H1*及*MMP-2/MMP-9*、*β-catenin*的表达。

结果：1. EMs中*nm23-H1*的mRNA水平显著低于正常子宫内膜组织（$P < 0.01$），WB检测结果进一步证实*nm23-H1*在EMs中表达下调；2. 与正常子宫内膜上皮细胞相比，*nm23-H1*在异位子宫内膜上皮细胞中的表达亦显著降低（$P < 0.05$）；3. 原代培养的细胞中，*nm23-H1*在EMs中的表达明显低于正常组（$P < 0.05$），而*MMP-9*和*β-catenin*的表达则均显著升高（P均< 0.05），但*MMP-2*的表达水平未见明显改变（$P > 0.05$）。17β-雌二醇刺激或干扰*nm23-H1*后发现，与未刺激组相比，基质细胞中*nm23-H1*的表达水平均下降（P均< 0.05），而*β-catenin*的表达则上调（$P < 0.05$），但*MMP-2/MMP-9*的表达未受影响。

结论：1. *nm23-H1*在EMs组织、上皮细胞及基质细胞中表达均下调，且受雌激素影响；2.*β-catenin*的表达受雌激素刺激和*nm23-H1*沉默的影响。

关键词

子宫内膜异位症，nm23-H1基因，MMP-2/MMP-9，β-catenin

Genome Sequencing of Rice Subspecies and High-Resolution QTL Mapping Reveals Agronomic Traits-Associated Loci on Recombinant Inbred Lines

Hua Zhong, Xiaoxiao Deng, Weilong Kong, Jing Yang, Yangsheng Li*

State Key Laboratory of Hybrid Rice, Key Laboratory for Research and Utilization of Heterosis in Indica Rice, Ministry of Agriculture, College of Life Sciences, Wuhan University, Wuhan, P.R. China

Email address:

lysh2001@whu.edu.com (Yangsheng Li)

*Corresponding author

Abstract

Rice (*Oryza sativa* L.) is a model organism of the monocotyledon *Gramineae*, and plays an important role in the world food production and the diet structure of Asian residents. Rice is divided into *Oryza sativa* L. ssp. *indica* and *japonica* subspecies, and they are highly distinct in terms of geographical distribution, morphological traits and genetic basis. After rice dwarf breeding, three-line and two-line hybrid rice breeding, the heterosis utilization of *indica* and *japonica* subspecies with an ideal plant type will have become an important approach for new breakthrough in rice production. However, the relationship among genetic background, ecological conditions, and agronomic traits is still unclear. In this study, we performed the de novo assembly of two high-quality genomes of *indica* cv. Luohui9 and *japonica* cv. RPY using Pacbio sequencing. A RIL population derived from an inter-subspecific cross between Luohui9 and RPY was generated. By combining the resequencing-based bin-map, QTL identification was conducted on tillering number (TN), panicle length (PL), spikelet per panicle (SPP), spikelet density (SD), 1000-grain weight (KGW) and plant height (PH). A total of 76 QTLs were identified: 13 for TN, 11 for PL, 12 for SPP, 9 for SD, 11 for KGW, and 20 for PH. Phenotypic effect variance explained by these QTLs ranged from 0.0846% to 49.38%. Combining with genome-wide association studies (GWAS) of plant height, we revealed some additional secondary significantly associated loci interact with SD1. This study provides information on genetic background for functional genes in rice breeding. Moreover, the public availability of the high-quality reference genomes of Luohui9 and RPY not only facilitates the identification of genes corresponding to agronomic traits but also provides a range of implications for plant biology and crop genetic improvement.

Keywords

Oryza sativa, De Novo Assembly, Bin Map, Plant Height, Yield