Background Presentation and Overview of Alternative Splicing of Adzuki Bean

Bo Wang[1,2], Qinling Wen[3], Jingxuan Wang[1,2], Gulnigar[1,2], Senya Tian[1,2], Yushu Liu[1,2], Jingyu Ma[1,2], Weiyu Li[1,2*]

[1]Beijing Key Laboratory of New Technology in Agricultural Application, Beijing University of Agriculture, Beijing, 102206, China

[2]National Demonstration Center for Experimental Plant Production Education, Beijing University of Agriculture, Beijing, 102206, China

[3]Plant Science College, Tarim University, Tarim, 843300, China

Abstract: Since the completion of the human genome sequencing work in 2003, more and more studies on alternative splicing of animals and plants, but there are little research has been focus on alternative splicing of adzuki bean. Therefore, this article reviews the prospect of alternative splicing of adzuki bean and its influencing factors.

Keywords: Adzuki bean; Alternative splicing; Influencing factors; High-throughput sequencing technology; Gene modification

1. Introduction

China is the birthplace and production power of adzuki bean, and adzuki bean has high nutritional and economic value. But in recent decades, the cultivation and production of adzuki bean has been rapidly developed in America, Canada, Brazil, Argentina, Holland and Australia, where adzuki bean have been exported to Japan and other Asian countries. It seems like that the production of Chinese adzuki bean is facing a new challenge, so how to dig the better adzuki bean gene is very important.

Through a large number of studies, it has been shown that as a mode of gene expression regulation in eukaryotes, alternative splicing plays an important role in animals and plants. It is widely existed in organisms and can regulate the proliferation, differentiation, development and apoptosis of organisms. For animals, alternative splicing plays a major role in the prevention and treatment of diseases; for plants, alternative splicing plays an important role in epigenetic regulation of plants and post-transcriptional fate, and it can increase plant protein diversity. Alternative splicing also plays a key role in plant resistant to stress.

However, the exploration of alternative splicing of adzuki bean has not yet begun. If the alternative splicing technology can be used in the improvement of adzuki bean genes, it will provide new ideas for adzuki bean breeding. Moreover, advances in science and technology will provide great support for Alternative splicing of adzuki beans. For example, advances in high-throughput sequencing technology in recent years have advanced genomics research in many crops. I think that under such premise and background, research on the alternative splicing of adzuki beans can be carried out and will be rewarded. In this paper, the research progress of alternative splicing and the prospect of alternative splicing of adzuki bean were reviewed.

2. Alternative Splicing Overview

Alternative splicing (AS) refers to the process by which pre-mRNAs cleave introns by selecting different splice sites, adding different exons to produce multiple mature mRNA splicing is of orms. Alternative splicing was first discovered in adenovirus in 1977, which is ubiquitous in eukaryotes and is the most important mechanism for expanding protein diversity, and it has a high probability of occurrence (95% of genes have alternative splicing in human). Alternative splicing can also increase the efficiency of genome use and enable gene duplication and exon recruiting. Molecular analyses during the last decade demonstrate that alternative splicing determines the binding properties, intracellular localization, enzymatic activity, protein stability and posttranslational modifications of a large number of proteins. Therefore, alternative splicing plays a crucial role in the evolution of species.

There are four basic types of alternative splicing: 3’-end alternative splicing, 5’-end alternative splicing, cassette splicing (cassette exon/exon skipping) and intron-retained alternative splicing.

3. Analysis of Influencing Factors of Alternative Splicing

3.1. At the chromosomal and genetic levels

Alternative splicing frequency is correlated with intron length, exon number, gene transcriptional level, GC content, and GR rates. And the number of alternative splicing events is positively correlated with intron length, number of exons, gene expression and GR rates;

and it is negatively correlated with GC content. Gene levels are consistent with chromosome levels. The amount of gene expression can affect the alternative splicing of the gene, and the higher the expression level, the more likely the alternative splicing occurs.

3.2. During the process of splicing

There are four regions on the mRNA precursor that can interact with related proteins (intron splicing enhancer, intron splicing silencer, exon splicing enhancer, exon splicing silencer).

DNA methylation has a preference for exon regions and is an important marker for identifying exons.

The modification of the histones also has an effect on the activity of splicing factors and efficiency of splicing. There are 42 histone modifications and they are not randomly distributed in the genome. The degree of modification in exons is significantly higher than that in introns.

Nucleosome localization can improve the efficiency of mRNA precursor splicing, the nucleosome occupancy rate is positively correlated with the ratio of exons, and the sequence generated by the preferred nucleosome is enriched in the gene's exons. At the same time, the end of the intron sequence is easily enriched in a sequence that can blocks the formation of nucleosome.

In addition, non-coding RNAs including Si RNA, Mi RNA, Inc RNA and circ RNA also affect the splicing process of mRNA precursors. Non-coding RNA not only plays an important regulatory role in biology, it can also be used as a means of biotechnology, disease diagnosis and clinical treatment.

4. The Probability of Alternative Splicing and Its Differential Expression

4.1. The probability of alternative splicing events

In plants, intron retention is the most common form of alternative splicing, and exon skipping rarely occurs, as opposed to in animals. According to previous studies, 33% of intron genes have alternative splicing; in rice, 40% of intron genes have alternative splicing; and 52% of intron genes have alternative splicing in soybeans; whereas 60% of intron genes have alternative splicing in Arabidopsisthaliana. This suggests that in plant, the probability of an intron having an alternative splicing event is very high.

4.2. Differential expression of alternative splicing at different developmental stages

In vivo, different genes with different environment, growth state, tissue, cell type changes will produce different alternative splicing isoforms, which will have different effects, and external stimuli will also affect the alternative splicing. For instance, inplants, the frequency of alternative splicing in the rapidly developing tissues such as shoot meristem and young seeds was significantly higher than other tissues of the same organs. And in mammals, alternative splicing is more likely to occur in tissues with multiple cell morphologies.

5. Alternative Splicing Research Methods

Alternative splicing biological research methods include Expressed Sequence Tag (EST) data, chip microarray technology, and high-throughput sequencing technology. Since EST is not a full-length mRNA and does not contain complete transcription information, it is only the result of one round of sequencing. Therefore, the use of EST for alternative splicing research has certain limitations, and the reliability of the results is low; chip microarray technology acquires more sequence data, but it is often limited by the position and density of the probe on the chip, soit has some limitations for the new alternative splicing events; and the next generation of high-through sequencing technology (NSG), compared with the traditional Sanger method, it has the following advantages: Low experimental cost (in Solexa), only 1/100 of the traditional sequencing method; High efficiency of sample use, even for a small number of samples can be sequenced; Wide range of applications, not subject to species restrictions, without prior knowledge of the pattern species genome sequence and without the need for synthetic probes, can directly carry out genome-wide expression studies. At present, high-throughput sequencing technology has been continuously developed, which has greatly promoted the work of genome-wide analysis.

6. Outlook

Since the completion of the human genome sequencing work in 2003, people have taken a big step in animal and plant genomics, and a large number of research on alternative splicing has promoted animal's and human's disease prevention and control, as well as plant breeding work has opened up new paths. With the development of science and technology, sequencing technology continues to advance, and plant genetic sequencing has been completed more and more, people have gradually found a large number of alternative splicing events in plants. Many plants have obtained research progress in alternative splicing through high-throughput sequencing technology, and the rapid development of high-throughput sequencing technology has also contributed to the genomics research of adzuki bean. However, there is very little research on the alternative splicing of adzuki beans. This problem is still waiting for some further exploration.

7. Acknowledgement

This work was supported by Beijing Municipal Educa-tion Committee Research Project General Plan (KM201610020006); Research Fund for Young Scien-tists of BUA (Project SXQN201805) and Beijing out-stand-ing talent training for young backbone individual projects (Project 2016000020124G049).

References

[1] Liu L., Bestel S., Shi J., et al. Paleolithic human exploitation of plant foods during the last glacial maximum in North China. Proceedings of the National Academy of Sciences. 2013, 110(14), 5380-5385.

[2] Ning X.U., Xu Z.C., Su H.W., et al. Establishment of an adzuki bean core collection based on geographical distribution and phenotypic data in China. Acta Agronomica Sinica. 2008, 34(8), 1366-1373.

[3] Yamaguchi H., Isigami M., Yasuda K. The sequence variations of intron-3 of the α-amylase gene in adzuki bean. Agricultural Sciences in China. 2003, 2(10), 18-24.

[4] Chow L.T., Gelinas R.E. Broker T.R., et al. An amazing sequence arrangement at the 5′ ends of adenovirus 2 messenger RNA. Cell. 1977, 12(1), 1-8.

[5] Berget S.M., Moore C., Sharp P.A. Spliced segments at the 5' terminus of adenovirus 2 late mrna. Proc Natl AcadSci USA. 1977, 74(8), 3171-3175.

[6] Pan Q., Shai O., Lee L.J., et al. Deep surveying of alternative splicing complexity in the human transcriptome by high-throughput sequencing. Nature Genetics. 2008, 40(12), 1413-1415.

[7] Stamm S., Benari S., Rafalska I., et al. Function of alternative splicing. Gene. 2013, 514(1), 1-30.

[8] Shen Y., Zhou Z., Wang Z., et al. Global dissection of alternative splicing in paleopolyploid soybean. The Plant Cell. 2014, 26(3), 996-1008.

[9] Chodavarapu R.K., Feng S., Bernatavichute Y.V., et al. Relationship between nucleosome positioning and DNA methylation. Nature. 2010, 466(7304), 388-392.

[10] Spies N.W.B., Nielsen C.,B., Padgett R.A., et al. Biased chromatin signatures around polyadenylation sites and exons. Molecular cell. 2009, 36(2), 245-254.

[11] Andersson R., Enroth S., Radaiglesias A., et al. Nucleosomes are well positioned in exons and carry characteristic histone modifications. Genome Research. 2009, 19(10), 1732.

[12] Schwartz S., Meshorer E., Ast G. Chromatin organization marks exon-intron structure. Nature Structural & Molecular Biology. 2009, 16(9), 990-995.

[13] Chen L., Shan G. Research on non-coding rna function and functional mechanism. Chinese Science Life Science. 2017, 47(01), 36-42.

[14] Zhang G., Guo G., Hu X., et al. Deep rna sequencing at single base-pair resolution reveals high complexity of the rice transcriptome. Genome Research. 2010, 20(5), 646-654.

[15] Thatcher S.R., Zhou W., Leonard A., et al. Genome-wide analysis of alternative splicing in zea mays: landscape and genetic regulation. The Plant Cell. 2014, 26(9), 3472-3487.

[16] Filichkin S.A., Priest H.D., Givan S.A., et al. Genome-wide mapping of alternative splicing in Arabidopsis thaliana. Genome Research. 2010, 20(1), 45-58.

[17] Modrek B., Resch A., Grasso C., et al. Genome-wide detection of alternative splicing in expressed sequences of human genes. Pac Symp Biocomput. 2001, 29(13), 29-41.

[18] Yeo G., Holste D., Kreiman G., et al. Variation in alternative splicing across human tissues. Genome Biology. 2004, 5(10), 1-15.

[19] Johnson J.M., Castle J., Garrettengele P., et al. Genome-wide survey of human alternative pre-mrna splicing with exon junction microarray. Science. 2003, 302(5653), 2141-2144.

[20] Wang S.Y. Next generation of high-through sequencing technologyand its clinical application prospects. Guangdong Medical Journal. 2010, 31(3), 269-272.