

Manipulation of Genes: A Promising Avenue for Improved Rice Productivity

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Why rice has been in focus for the crop improvement plans?

Rice is the world's second most important cereal crop which is eaten by almost 40% of the world's total population. Elevated temperature and scarcity of water for irrigation both have a negative impact on rice productivity. As per FAO (Food and Agriculture Organization) estimates, rice yield needs to be increased by 60% to keep pace with the exponentially increasing population. Like most other crops in India, rice cultivation is also facing a similar loss of productivity due to unfavourable environmental conditions, i.e, abiotic stress (for example, drought stress caused by lack of sufficient rainfall and irrigation) and devastating diseases caused by bacterial and fungal pathogens. It is of utmost importance for the crops to be able to withstand these adverse conditions and survive without compromising with the yield.

Role of molecular biology in attaining the target of crop improvement

In the present scenario, plant molecular biology plays a pivotal role in looking out for alternative solutions to ensure sustainable rice production and food security by manipulating the suitable genes that can mitigate the unfavourable conditions. The complete set of genes or genetic material present in an organism is called 'genome'. The number of functional genes in rice varies from

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between 30,000 to 50,000. Genes are the basic physical and functional units of heredity and are made up of DNA (deoxyribonucleic acid) which contains the information as to how an organism should function and develop. The expression of a gene is the process by which the information from a specific gene is used in synthesizing a specific functional gene product such as a protein. A single protein is encoded by a single gene. These gene products (proteins) are the building blocks of life and have specific roles in maintaining the normal functionality of the organism. A huge proportion of these genes also play significant roles in withstanding the harsh environmental changes and thus maintaining the health and yield of the plant.

By using various tools of plant molecular biology, we can manipulate the level of expression of these genes. This means that if we want a desirable gene to express more than their normal state in order to produce more copies of that gene product, we can achieve it. If we can make a suitable gene to express more than its basal expression level, we can get amplified benefits from that particular gene which is advantageous for the plant. But, not all genes are directly related in conferring resistance to environmental adversities. Hence, it is of great importance to find genes in plants that might have a role in combating stress conditions so that they can be utilized wisely to enhance the genetic make-up of the plants.

How mining novel genes is important

To address this issue, a collaborative research project was sanctioned by the Department of Biotechnology, Government of India (DBT) entitled as 'Identification of candidate genes for enhanced water use efficiency in rice through activation tagging'. The author's team under the guidance of Prof PB Kirti at University of Hyderabad was directly involved in this project. This project was aimed at mining the uncharacterized or novel genes in rice that can be utilized to tackle the negative impact of water scarcity on rice productivity using molecular biology techniques.

Activation tagging is an advanced strategy and its success in identifying novel genes in rice has proven it to be a goldmine for agronomic applications. The strategy involves a complicated procedure in which a DNA segment from a source other than rice is randomly introduced into the rice genome. This DNA segment is known as 'enhancer element' which, when introduced into the plant genome, has the ability to locate itself beside any particular genes and subsequently increases the level of expression to a certain extent. The information regarding this DNA segment is known by the researchers so that its presence can be easily tracked in the plant system.

As we have mentioned earlier, an increase in the expression is directly correlated with production of more copies of that specific gene product or protein. Now, if this protein is beneficial for the plant to combat adverse climatic conditions, elevated production of the same should give the plants the desired benefits under such climatic conditions. We can presume that after introducing these DNA elements (enhancers) in rice plants, a considerable number of genes have been over-expressed. Consequently, when water-limited environment was mimicked under greenhouse condition, we noticed that these plants (with the introduction of DNA elements) were more resistant to wilting and exhibited better vigour than the normal plants (with no DNA elements being introduced).

As these DNA elements can be tracked in the plant genome by molecular biology techniques, we could get complete information regarding the particular genes that have been over-expressing.

Hence, we can conclude that these particular genes engage, at least to some extent, in providing resistance to abiotic stress. Thus, activation tagging helps in identifying novel genes and their significance in a single step. Finally, this project led our research team to identify numerous such genes which might have immense agronomic importance in terms of abiotic stress tolerance and some of them were not reported earlier.

Ribosomal protein genes emerging as potential players to safeguard plants from harsh climatic conditions

Among other genes that were identified by this approach, two were members of a gene family that codes for ribosomal proteins large subunits. Ribosomes are small organelles present in a cell which are involved in the process of synthesizing proteins. They have mainly two subunits (small and large subunits) which come together along with other genetic elements to form a functional protein synthesizing machinery. Each of the ribosomal subunits is made up of proteins (ribosomal proteins) and Ribonucleic Acids (RNA). There are numerous ribosomal proteins associated with the two subunits of ribosomes, and the number of ribosomal proteins varies greatly in different organisms. Since each protein in a cell is encoded by a specific functional gene, it is important to understand that since there are numerous ribosomal proteins in rice, a large family encompassing a huge number of genes is responsible for the synthesis of these ribosomal proteins. Ribosomal proteins have long been considered to be essential in ribosomal synthesis and in maintaining the structural integrity of both the subunits of ribosomes. But with our previous study involving activation tagging, we could identify two genes of this family with a novel role in abiotic stress amelioration. This inspired us to find out if there are some other genes belonging to this family which could be utilized for manipulating rice genome for obtaining better traits. After going through extensive experimental analysis, we could shortlist few such genes coding for ribosomal proteins that have the potential to induce stress resistance to rice plants.

We then attempted to elevate the expression of these ribosomal protein genes in a very widely cultivated Indian rice variety, BPT-5204 (also called Samba Mahsuri) to validate their functions because the ability to over-express a particular gene allows researchers to explore the functional characteristics of the same. Consequently, engineering the plant genome by over-expressing one of these shortlisted genes, our research group has experimentally demonstrated improved yield and less susceptibility of the modified plants to drought conditions. Further, we are interested in confirming and characterizing the functions of the rest of the shortlisted ribosomal protein genes by increasing the level of expression of these genes.

Significance of the research

Rice is an essential crop having national economic importance because food security is essentially a reflection of rice security in Asia. In India, rice is more than just a food crop; it is rather an integral part of the history, culture, and lifestyle in various ways. Although rice yield has exhibited

comparative improvement over the years, recently rice yield in India has dropped down compared to other countries in South Asia. Additionally, the efficiency of rice production has dwindled by extreme climatic changes. Rain-fed rice growing areas are predicted to be wrecked by frequent droughts. Under these circumstances, scientists should necessarily be equipped with approaches to feed the growing world population. Identifying genes that could be possible targets to manipulate the genetic make-up of the plants, in order to enhance the beneficial agronomic traits or decrease susceptibility to diseases, is of great significance. The research discussed in this article is important as it identifies few ribosomal protein genes to have fair prospects to be exploited for crop improvement and sustainability. As rice has a close evolutionary relationship with other cereal crops, genes identified and characterized for improvement of rice cultivation can extensively be used for other important crops as well.