Tailoring Papaya Resistant to Papaya Ringspot Virus

Swati Kumari*
Amity University, ICAR-Central Institute for Subtropical Horticulture, Lucknow
Email: scorpion.swati19@gmail.com

It was a stuffy July morning when my sister, Nidhi, returned from her regular morning workout session, baffled with her corpulence. She sat on the couch gulping water. After taking a deep breath, she devoured a sliced papaya hoping to flatten her paunch. Nidhi had read on Google that papaya has an enzyme named papa in which helps in digestion, is rich in fiber, antioxidants, Vitamin A and C. She read that papaya gives a feeling of fullness and eases the bowel movement, culminating in weight loss.

"Didi, what are these green circles I find everyday while peeling off papaya?" Nidhi asked, pointing at the rings on the papaya.

"Ah! These are not circles but rings dear. They are caused by papaya rings potvirus(PRSV) disease", I replied.

"And what does this disease do? Where does this virus come from?" Nidhi asked again.

"Well, this disease affects taste, fruit quality and yield of papaya. If left untreated, it can destroy the papaya plant", I explained.

"How does this virus reach the papaya?" she asked in a daze.

"These viruses are transmitted to the papaya by a bug named aphid, the carrier vector of PRSV", I added.

"If it is such a deadly disease, then why don't papaya growers treat these viruses by spraying something?" she inquired.

---

* Ms. Swati Kumari, Ph.D. Scholar from Amity University, Lucknow, is pursuing her research on "Genetic Engineering of Papaya (Carica Papaya L.) with Tr-Cp Gene for Conferring Resistance against Papaya Ring Spot Virus." Her popular science story entitled "Tailoring Papaya Resistant to Papaya Ringspot Virus" has been selected for AWSAR Award.

"My dear, viruses can't be destroyed so easily. One can kill bacteria using antibiotics and fungus by spraying fungicides but there is no such thing as viricide" I told her but her questions didn't stop. "Then what I am eating is bad? How can we combat this disease? Why doesn't anybody do something about it?" Nidhi shot an array of questions.

Nidhi's inquisitiveness pushed me to collect more information about this disease, its origin and what research has been carried out so far. I wanted to satisfy her curiosity about what is making her favorite fruit so ugly. I went through a number of scholarly articles about papaya and this disease in particular. Being a Post Graduate in Biotechnology, I could conceive the crux of the science behind this disease and research to fight against it. After a thorough reading of about a week, I called Nidhi to answer all her questions.

What is pathogen derived resistance?

Way back in 1984, Prof. Roger Beachy, Washington University, USA, propounded the theory of PDR *i.e.* pathogen derived resistance. Polio vaccination works on the same principle. Prof. Beachy developed tobacco virus resistant plant employing PDR which brought a sea level change across the countries engaged in developing virus resistant plants. In PDR, the pathogen itself is utilized to provide resistance against its attack, thereby, protecting the plant from infection. This technique utilizes a specific gene (*coat protein*), carrying our desired traits, which is taken from the pathogen and mobilized into a transformation vector. This vector acts as a vehicle to deliver the desired gene to the plant by using any of the available direct or indirect gene transfer methods. The plants, thus, produced are called "transgenic" as they have genes from another source.

Devastation of blooming papaya industry in Hawaii

Hawaii comprises many islands and it is surrounded by the Pacific Ocean. Continuous volcanic eruptions from Mauna Lo affect the agroecology of this state. PRSV was first reported in 1940 in the island of Oahu, Hawaii. Kapoho is the most dominant variety of papaya which is sold in the mainland of USA and also exported to Japan. However, after the disease infestation, the entire Kapoho belt had to be shifted from Oahu to Puna is land of the state. Puna grew about 95 per cent of Hawaii papaya covering around 2500 acres of land. Up till 1960s, there were no cases of PRSV in Puna. However, in 1990s, the entire papaya industry was devastated by PRSV. Within a span of six years, Hawaii papaya industry lost 50 per cent of its crop due to the virus.

Technology led revival of papaya industry in Hawaii

To strive against PRSV disease Dr. Dennis Gonsalves, who was a musician by chance and scientist by choice, adopted the method of PDR which was discovered by Prof. Beachy. Dr. Gonsalves not only developed two transgenic varieties, namely 'Rainbow' and 'Sun Up', but also successfully commercialized it. Transgenic papaya covered about 85 per cent of total sold papaya available in supermarket, besides other non-transgenic fruit crops. The virus free 'Rainbow' yielded about

125,000 pounds of marketable fruit per acre per year, whereas non-transgenic 'Sunrise' (PRSV infected) produce was 5,000 pounds of fruit per acre per year.

After the sale of transgenic papaya in 1998, Puna received 53 million pounds whereas non-transgenic papaya grossed only 26 million pounds in 1992. Hence, it can easily be concluded that transgenic papaya gave around twice the profit to papaya growers in comparison to non-transgenic papaya. As transgenic papaya fields expanded in Puna, the PRSV inoculum also reduced due to replacement of non-transgenic fields with the transgenic ones. Rainbow accounted for around 50 per cent of the fresh fruit production in Puna. The bearing acres of transgenic Rainbow were about 595 whereas that of non-transgenic Kapoho was recorded to be 380 acres only. Despite being one of the most developed countries with highly stringent and strict food safety norms and regulations, Japan and Canada signed the treaty and deregulated the import of transgenic papaya varieties to their countries. Japan held 20 per cent while Canada had about 11 per cent of Hawaii's export market. This signifies that transgenic papaya is safe and it should be adopted more widely across the globe.

In the process of transgenic papaya commercialization, on the one hand, Dr Gonsalves was admired by the farmers for his endless efforts in saving the papaya industry as he was helping farmers to earn their livelihood from papaya again. On the other hand, he was denounced by the anti-GMO activists. However, his approach was always pro-farmer and he only focussed on end goal without getting disoriented. Dr Gonsalves said, "Don't just be a test tube scientist, and do something to help people. This kind of work is not for the faint-hearted. When you put the human part of Biotechnology into the equation, it gets easier to continue the work. Science and Technology is the only move to feed 9 million people".

Transgenic papaya research in India

Hawaii, Thailand, Jamaica, Brazil and Venezuela have also developed transgenic papaya. However, India is quite new in this fray. Indian PRSV isolates exhibit about 11 per cent variability as we move from North to South. So, a transgenic papaya developed in North India will not show resistance in Southern India due to different serotypes of the virus. Hence, a biological process called RNAi has proven to be the only approach which can help in combating the disease in India. It kills the virus irrespective of its serotype variability.

Research on transgenic papaya using *coat protein* gene was started in Southern parts of India like Tamil Nadu Agriculture University, Coimbatore and ICAR- Indian Institute of Horticultural Research, Bengaluru, but with scanty success. Later, a vast networking project of ICAR with its two premier Institutes, ICAR-Indian Agricultural Research Institute (IARI), New Delhi, and ICAR-Central Institute for Subtropical Horticulture (CISH), Lucknow, came into the picture to solve this problem. ICAR-IARI developed the gene construct for this program while ICAR-CISH designed a successful delivery system for transferring the gene construct into the papaya plant.

As per biosafety regulations in India, scoreable and selectable markers cannot be a part of the transgene after integration into the plant. So, the gene construct used in the study has been

made marker free. It is to be noted that marker removal from the gene construct makes the job of screening transgenic papaya very cumbersome because then each and every plant needs to be tested whereas the plants having gene construct with markers need not be screened individually as the non-transformed plants get killed in the selection process. After the transgenic plants are screened for stable transgene integration, the plants are acclimatized and hardened. About 60 per cent of transgenic plants die during the course of acclimatization as they are unable to cope with the natural climatic conditions. To increase the successful acclimatization of transformants, biotization technology is practised. In this technology, the endogenous bacteria are utilized to ease out the process of rooting and acclimatization. A few transgenic lines have been identified and are under advance stage of evaluation. So far, three generation advancement has been achieved.

"Oh! This means India will also be equivalent to Hawaii, not only in terms of producing the PRSV resistant papaya but also in exporting its transgenic papaya varieties to countries abroad. This is really awesome, Didi!", exclaimed Nidhi with enigmatic pride, who was engrossed in listening to the long story about her favorite fruit till now.

"Yes, my dear, why not! But we have a long way to go in terms of regulatory mechanism to achieve our goal with glorious success. Amen!" I replied.