

## The long-lasting tomato

09 November 2006 | News

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*Using genetic engineering tools, Indian scientists have increased the shelf life of the tomato crop.*

Looking at the shooting prices of vegetables and fruits in the market, it is difficult to believe that India is the largest producer of fruits and the second largest producer of vegetables globally, after China. Annually the country produces 32 million MT of fruits and over 71 million MT of vegetables, which accounts for about 15 percent of the world's production of vegetables. However, it is estimated that about 30-40 percent of the total production of fruits and vegetables is lost due to spoilage at various post harvest stages.

One way of reducing the post harvest losses is by increasing the shelf life of the fruits and vegetables. Worldwide scientists have been adopting this approach to tackle the problem of post harvest losses. In India, Dr KC Bansal, principal scientist, National Research Centre on Plant Biotechnology, (NRCPB), IARI, along with his group has been working on extending the shelf life of tomatoes. He said, "Tomato is a very important crop for the country. It remains metabolically active even after harvest and under storage at room temperature leading to huge losses: spoilage due to over ripening, attack by bacterial and fungal pathogens. We cannot prevent this unless we have refrigerated storage facility, which India can ill afford at the moment. Instead, we can increase the shelf life of the tomatoes by using the biotechnology tools or delay the ripening process."

Ethylene gas is the ripening hormone and is responsible for inducing ripening in most of these climacteric fruits including

tomato. For ethylene biosynthesis, four enzymes are required and if the expression of one of these enzymes is knocked off, the pathway is broken and there is no ethylene production. So the global idea has been to curtail the production of ethylene by the antisense RNA technology where a gene responsible for one of the enzymes is put in the reverse/antisense orientation. Flavr-Savr tomato was the first product in the world based on this technology and the idea was to increase the shelf life.

The Indian effort has been also on the same lines but with a significant difference. Elaborating on the technique used, Dr Bansal explained, "The idea in the Indian effort is to cut down the ethylene production not throughout the whole plant body but only in the fruit where ethylene is induced with the onset of ripening. If we use the CaMV 35S promoter, which most of the scientists have used, then the whole ethylene pathway is disturbed in different parts of the plant body as CaMV 35S is a constitutive promoter."

So in order to curtail the amount of ethylene produced which starts picking up at breaker stage (the point at which 25 percent of the fruit begins to turn red) onwards, we used fruit-specific promoters. Also, when one uses a constitutive promoter, then the fruits will never ripen and in such cases farmers are advised to apply ethylene gas exogenously. But by using the fruit specific promoter, there is no need for any exogenous application of ethylene. Here the fruit will ripen automatically, but slowly, and will have extended shelf life."

In the process, the scientists have been able to delay the shelf life by two or three weeks, which is a good time if the produce needs to be transported from one part of the country to the other and will not rot in the absence of refrigeration facility in rural households. Significantly, this is the first effort in the country to have successfully come up to this stage.

Dr Bansal and his team have been working on this project since the last four to five years with the support from the ICAR and DBT. They initially cloned the necessary genes and promoters and made the required gene constructs to develop the transgenics based on the antisense RNA approach. Since the promoter is fruit specific, until the plant has gone to the fruiting stage, the gene construct behaves as a silent gene and is not expressed. Only when the fruit starts ripening, the gene construct becomes functional, acts in the fruit specific manner and there is no ethylene production. These findings were also studied by the scientists at the molecular level in the lab. Now they have successfully developed the transgenic in the elite cultivar of tomato of Pusa institute, known as "Pusa Ruby", which is ready for field trials after the necessary approvals are taken from the government. The trials at the Phytotron level have already been done which clearly show that there is a delay in ripening.

The amount of loss that occurs annually due to the post harvest losses in fruits and vegetables runs into thousands of crores and with this technology we will be able to save losses worth Rs 1,000 crore annually in tomato alone. Besides, it will also help in diverting the surplus produce timely into the food processing industry" commented Dr Bansal.

Sharing his larger vision for tomato project, Dr Bansal stated that his group may like to transfer this trait to some other tomato lines, which can be used in hybrid seed production. This will be undertaken with the help of tomato breeders within the Institute and the ICAR system and later they may also like to share this technology with the private sector to enter into a public private partnership. "We would like to spread the transgenic tomato throughout the country, with the help of breeders and the private sector."

According to him, the product should be ready to hit the market in the next three to five years, if the regulatory approvals are obtained in time and the biosafety guidelines are not too stringent. Since there is no foreign gene, bacterial or otherwise, in this tomato, this timeline should not be difficult to achieve," he said.

Another critical project on which Dr Bansal and his team are working is the nutritional quality improvement in tomato, which is also being supported by the DBT. The red color of tomato is imparted by "Lycopene", which is one of the best dietary natural antioxidants available. The lycopene content in Indian cultivars is much less as compared to its European or American counterparts and there is a lot of scope for improvement in the Indian cultivars.

This project started about a year ago and is the only effort in the country to increase the lycopene content in tomatoes. Currently the scientists are studying all the 25 genes in the carotenoid biosynthetic pathway; lycopene is one of the products of this biosynthetic pathway. The aim is to understand the regulation of the gene expression in this pathway as the researchers want that there should be minimal or no disturbance to the whole pathway, except in the precise gene responsible for improved lycopene. Post this study, they will go for genetic engineering using the identified genes and promoters. "We have already identified a putative gene which is associated with increased lycopene content and have cloned that gene as well. Currently we are studying the regulation and interplay of all the genes involved," shared Dr Bansal.

"Our final goal is to combine the increased lycopene content with extended shelf life, so that it is nutritionally enriched with better keeping quality. I hope that in a year or two, at the most, we would like to combine both the traits and in possibly three years time we will come up with another product that is having both the traits together," he added.

This effort will go a long way in providing nutritional security along with affordability to the common man in India.

Rolly Dureha