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Short Communication

IMPROVEMENT OF PAPAYA PRODUCTION USING GENETIC ENGINEERING: A REVOLUTION

Lincoln Mandal[1]*, Pooja Sahu[2], Ajit Kumar Mannade[1] & Jawahar Lal Katara[3]
[1]Department of Plant Molecular Biology and Biotechnology, IGKV, Raipur – 492012, India
[2]Department of Genetics and Plant Breeding, IGKV, Raipur – 492012, India
[3]Crop Improvement Division, CRRI, Cuttack, Odisha, India

ABSTRACT
Papaya (*Carica papaya*) is widely grown in the tropics and rich source of antioxidant nutrients such as carotenes, vitamin C and flavonoids. Papaya ringspot virus (PRSV) is often a limiting factor in the production of papaya worldwide. The "Transgenic virus resistance" and "RNA-interference" are excellent approaches for controlling viral diseases in papaya. The trait in papaya resistance to PRSV has been developed by use of transgenic technology. The transgenic papayas are virus-free plants. The first GM fruit crop papaya developed by a public institution has commercialized. In 1991, Gonsalves observed the first transformed plant that appeared to have PRV resistance. The papaya line "55-1"was first reported resistant under field conditions in 1992. The commercial cultivars 'Rainbow" and "SunUp" developed from line 55-1. The 'Sun-Up' is homozygous for the coat protein gene while 'Rainbow' is an Fl hybrid of 'SunUp' and the nontransgenic 'Kapoho'. The "Line 63-1"a new virus-resistance transgenic papaya line expressing the coat protein (CP) gene for papaya ring spot virus (PRSV) has developed. The "Line 63-1" could be used as a source for resistance to PRSV isolates. The transgenic papaya shows higher yield potential compare to non-transgenic. The transgenic dosage leads to increase disease resistance. So that the likely reason 'SunUp' showed broader resistance than 'Rainbow'.

KEYWORDS: *Carica papaya*, tropics, viral diseases, ring spot virus, Line 63-1, SunUp, Rainbow.

INTRODUCTION
Papaya, also called papaw or pawpaw, is an edible melon-like fruit of a tropical softwood tree (Carica papaya) of the family Caricaceae. Papaya is considered one of the most economically important and nutritious fruits, being a rich source of antioxidant nutrients such as carotenes, vitamin C and flavonoids; the B vitamins folate and pantothenic acid; the minerals potassium, magnesium and fiber. In addition papaya is the source of the digestive enzyme papain, which is an industrial ingredient. Gaining in popularity worldwide, papaya is now ranked third with 11.22 Mt, or 15.36 percent of the total tropical fruit production, behind mango with 38.6 Mt (52.86%) and pineapple with 19.41 Mt (26.58%). Global papaya production in 2010 was estimated at 11.22 Mt, growing at an annual rate of 4.35 percent between 2002 and 2010 .Global papaya production has grown significantly over the last few years, mainly as a result of increased production in India (FAOSTAT, 2012a).

Major production constraints in papaya production
A major production constraint for papaya worldwide is "papaya ringspot virus (PRSV)" a potyvirus that is rapidly transmitted by a number of aphid species in a nonpersistent manner (Gonsalves & Ishii, 1980). This virus affects production and productivity in every region of the world by decreasing photosynthetic capacity of the plants, which subsequently display stunted growth, deformed, inedible fruit and eventually plant mortality (Gonsalves, 1998). Because of limitation, scientists have turned to genetic engineering and the use of the PRSV "coat protein resistance" strategy to develop transgenic PRSV-resistant papaya.

Transgenic PRSV-resistant papaya
Transgenic PRSV-resistant papaya is a papaya that has been genetically engineered to contain a virus gene that encodes for the production of the coat protein of the virus. As a major component of viruses, the coat protein's primary function is to protect viral genetic information. The expression of this gene in the resulting papaya line renders the plants resistant to the virus. In other words, a gene from the pathogen is used to fight against the pathogen itself.

Historical aspect
In 1992, PRSV was discovered in the district of Puna on Hawaii Island, where 95% of Hawaii's papaya was grown. Within two years, PRSV was widespread and causing severe damage to the papaya in that area. Coincidentally, a field trial to test a PRSV-resistant transgenic papaya had started in 1992 and by 1995 the 'Rainbow' and 'SunUp' transgenic cultivars had been developed. These cultivars were commercialized in 1998. Licenses that were needed to commercialize the transgenic papaya were obtained by April, 1998.

Approaches
The "Transgenic virus resistance" is an excellent approach for controlling viral diseases in horti-cultural crops. The "Coat protein resistance Strategy" is very useful for development transgenic Papaya. The "RNA-interference" approach - many laboratories have shown that the pathogen-derived resistance to plant viruses is due to the

mechanism of post-transcriptional gene silencing or RNA-interference.

Transforming the papaya for virus resistance:

In 1987, researchers at the University of Hawaii (UH) and Cornell University in New York had begun trying to create a "transgenic" papaya with virus resistance. They used "genetic engineering" or "genetic transformation" techniques to insert part of the virus into the nucleus of a Papaya cell. At Cornell, Dr. Dennis Gonsalves isolated the virus' coat protein and he and Dr. Jerry Slightom of The Upjohn Company modified it for use in plants. In 1989, quantities of the gene were "shot" into cultured papaya tissue using a "gene gun" developed by Dr. John Sanford at Cornell. In 1991, Gonsalves observed the first transformed plant that appeared to have PRV resistance.

Transformation systems for Papaya

The promoter used in the transformation of papaya for various traits is the cauliflower mosaic virus (CaMV) 35S promoter. The most common selectable marker gene used in the production of transgenic papaya is the *neomycin phosphotransferase* (*nptII*) gene that confers kanamycin resistance. The first transformation of papaya using Agrobacterium was reported by Fitch *et al.,* in 1993. Gonsalves *et al.*,1998 used the gene gun in transferring an untranslatable cp gene derived from PRSV HA 5-1.

Varieties

The Papaya line "55-1"was first reported resistant under field conditions in 1992. Two varieties of papaya resistant to papaya ringspot virus have been developed using Biotechnology: "SunUp" and "Rainbow". The 'SunUp' is a transgenic red-fleshed Sunset that is homozygous for the coat protein geneand the 'Rainbow' is a yellow-fleshed F1 hybrid developed by crossing 'SunUp' and nontransgenic yellow-fleshed 'Kapoho' (Manshardt, 1998). The "Line 63-1"a new virus-resistance transgenic papaya line expressing the coat protein (CP) gene for papaya ring spot virus (PRSV) has developed. The "Line 63-1" originated from same transformation experiment that result in line 55-1 from which the transgenic commercial cultivars, "Rainbow" and "SunUp", were developed. The "Line 63-1" presents Hawaii with PRSV-resistance transgenic germplasm that could be used as a source for resistance to PRSV isolate within and outside of Hawaii (Tennant *et al.*, 2005).

Yield Potentials

The field trial of Steve Ferreira of the University of Hawaii showed that the transgenic 'SunUp' and'Rainbow' were resistant to virus and that these cultivars were of commercial quality. The data showed that 'Rainbow' yielded about 125,000 pounds of marketable fruit per acre per year, whereas the nontransgenic 'Sunrise' (all of which became infected by PRSV) yielded 5,000 pounds of fruit per acre per year (Ferreira *et al*., 2002). The transgenic papayas are virus-free. So, save yield potential of the varities. The 'Rainbow' is now widely planted and has helped to save the papaya industry from devastation by PRSV.

Impact of Transgenic Papaya

The "Rainbow" to be resistant to the several PRSV isolates from Hawaii but susceptible to a range of isolates from outside of Hawaii. Unlike Rainbow, however, "SunUp" is resistant to many PRSV strains from regions outside of Hawaii. A practical consequence is that increasing the transgene dosage can lead to increased resistance. Thus, transgene dosage is the likely reason that SunUp shows broader resistance than Rainbow, in that SunUp is homozygous for the inserted coat protein gene, while Rainbow is hemizygous (or has half the gene dosage of SunUp) because it is an F1 hybrid between SunUp and the nontransgenic Kapoho. Achieving "durable" resistance- Studies have shown that 'SunUp' papaya has broader resistance than 'Rainbow'.

Deregulation and safety aspect of PRSV-resistant papaya

Transgenic line 55-1, the parent of "SunUp" and "Rainbow" were deregulated within two years after documents were submitted to the Animal Plant Health Inspection Service (APHIS), the Environmental Protection Agency (EPA), and the Food and Drug Administration (FDA). Finally, Licenses to commercialize the transgenic papaya were obtained by the Papaya Administrative Committee in Hawaii by April, 1998. Development of genetically engineered crops and foods is monitored by three federal regulatory agencies: the U.S. Department of Agriculture, the Environmental Protection Agency, and the Food and Drug Administration. The agencies have examined the development of the 'SunUp' and 'Rainbow' papaya cultivars and approved them as safe.

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