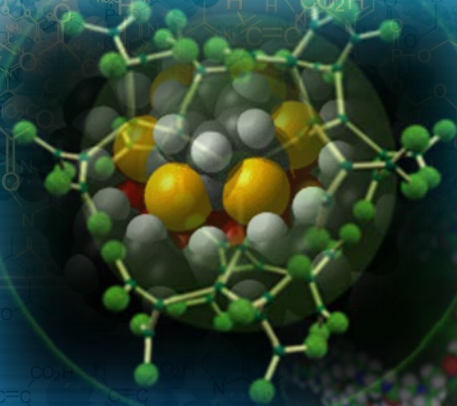




*Biotechnological approaches in  
Pest Management*





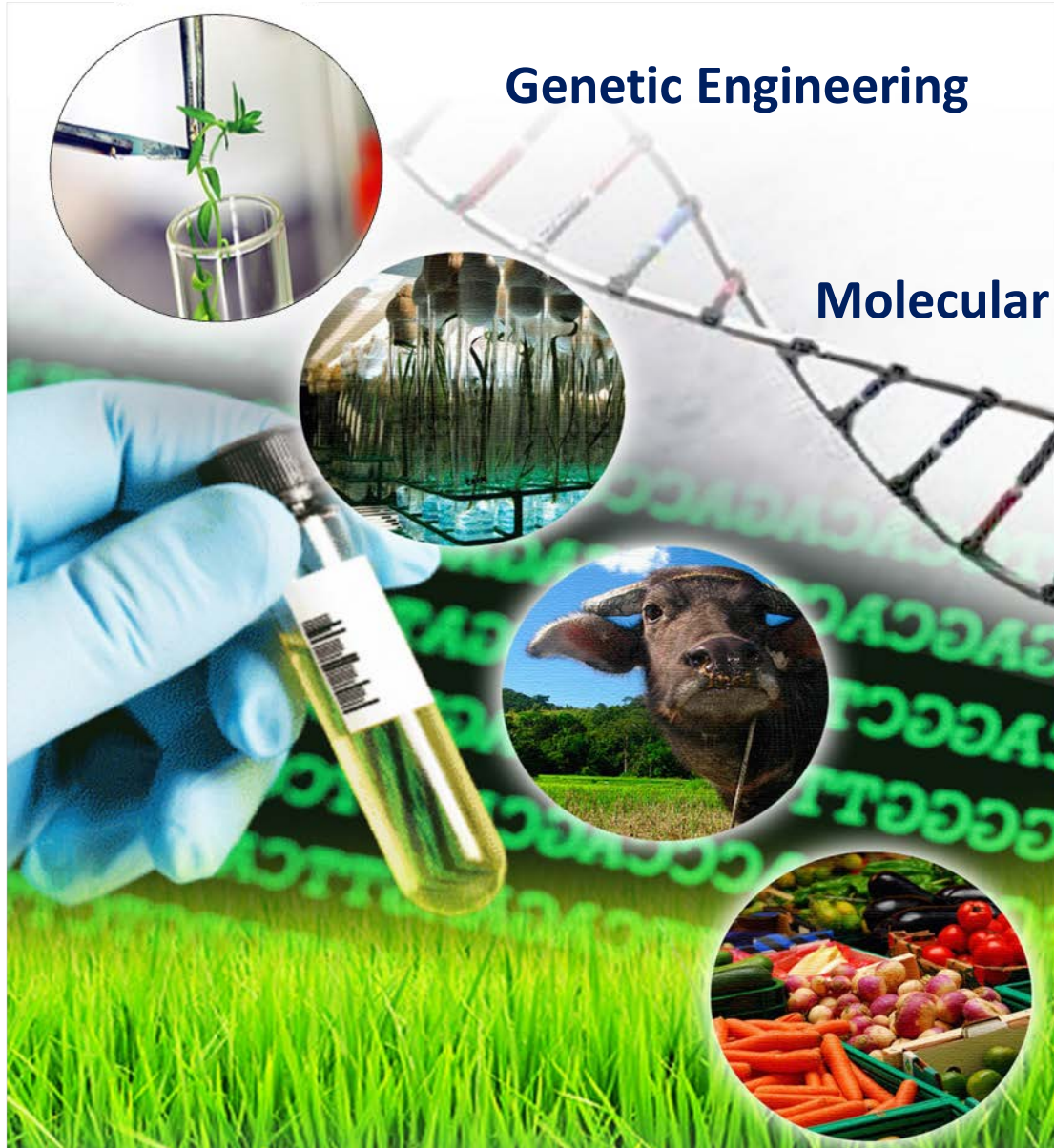
## **Biotechnology in Agriculture?**

**Any technique that uses living organisms or substances from these organisms, to make or modify a product, to improve plants or animals or to develop substance for specific uses.**





# How is Agricultural Biotechnology used?



**Genetic Engineering**

**Molecular markers**

**Molecular diagnostics**

**Vaccines**

**Tissue culture**

# Timeline of Biotechnology in Agriculture

Sporeine from France, first commercial product

First commercial Transgenic crop – Virus resistance tobacco by China

FlavrSavr® tomato- 1<sup>st</sup> genetically modified crop in USA and France

Field release of Bt cotton

1938

1962

1990

1994

1995

1996

2002

kurstaki, isolated as highly potent strain in France.

Chymosin -1<sup>st</sup> product of rDNA in food supply.

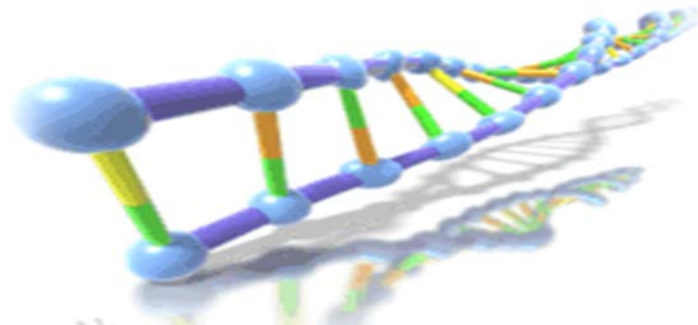
first experiment on transgenic plant in field

commercial cultivation of Bt cotton in India

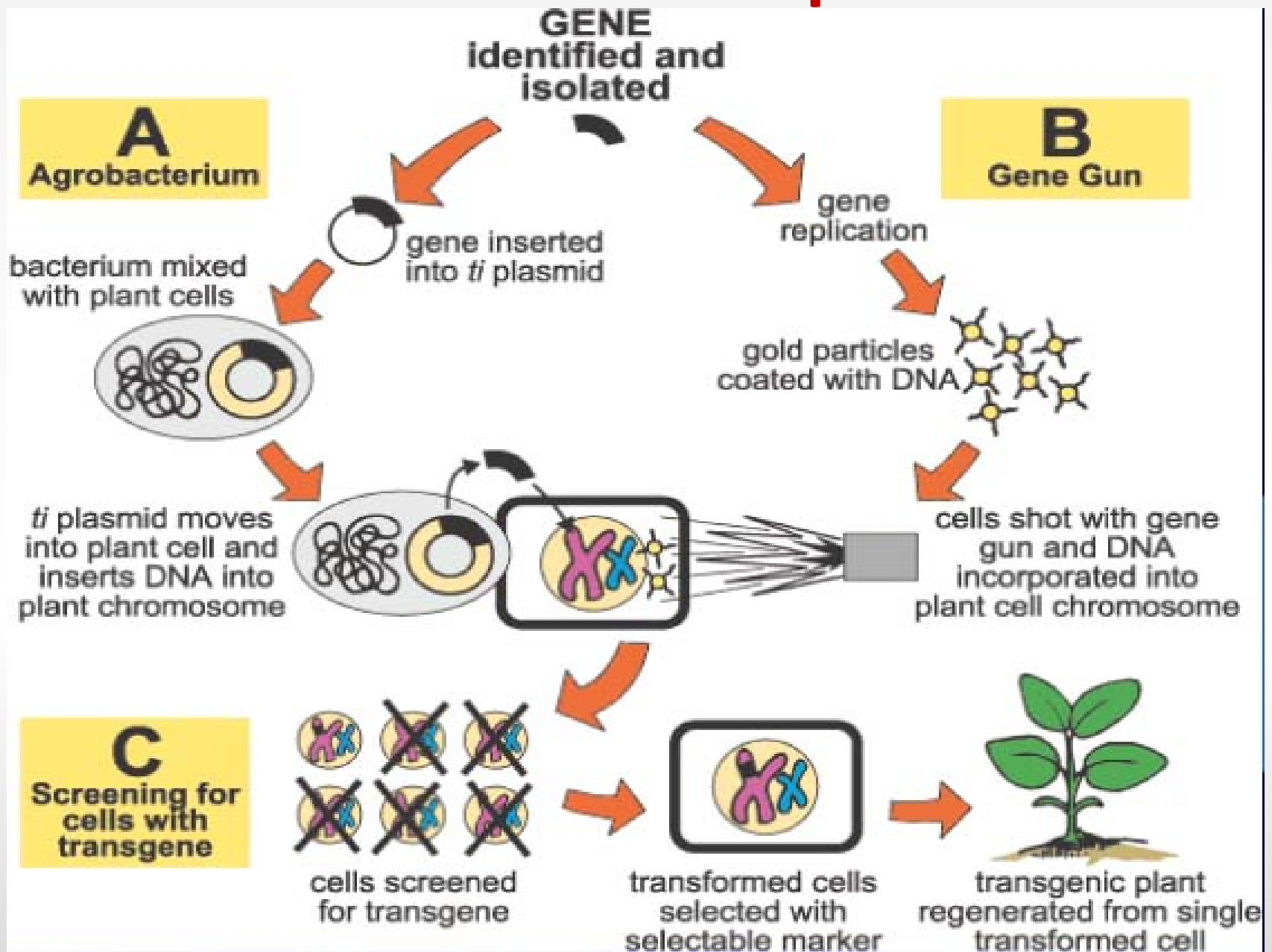


# Application of Biotechnology in Agriculture

1. **Crop improvement: Improved oil quality in Soybean and Canola**
2. **Herbicide resistance: Cotton, Corn, Soybean and Rice**
3. **Insect Resistance: Cotton, Corn, Rice, Tomato and Potato**
4. **Virus resistance: Papaya, Squash and Potato**
5. **Slow-ripening and softening: tomato and melon**
6. **Male sterility: Canola and Corn.**



# Gene transfer in plants



# Development of transgenic crops expressing insecticidal genes

- **Cry toxins *Bt***: Cry 1 Ab, Cry 1 Ac, Cry IIa, Cry 9c, Cry IIB, Vip I, Vip II etc.
  - **Plant metabolites** : Flavonoids, alkaloids, terpenoids
  - **Enzyme inhibitors** : SBTI, CpTi
  - **Enzymes** : Chitinase, Lipoxigenase
  - **Plant Lectins** : GNA, ACAL, WAA
  - **Toxins from predators** : Scorpion, spiders
  - **Insect harmones** : Neuropeptides and peptide hormones
- Insecticidal genes from sources other than *Bacillus thuringiensis*

**Pyramiding genes:** Engineering transgenic crops with more than one gene to get multimechanistic resistance.



# Bt cotton

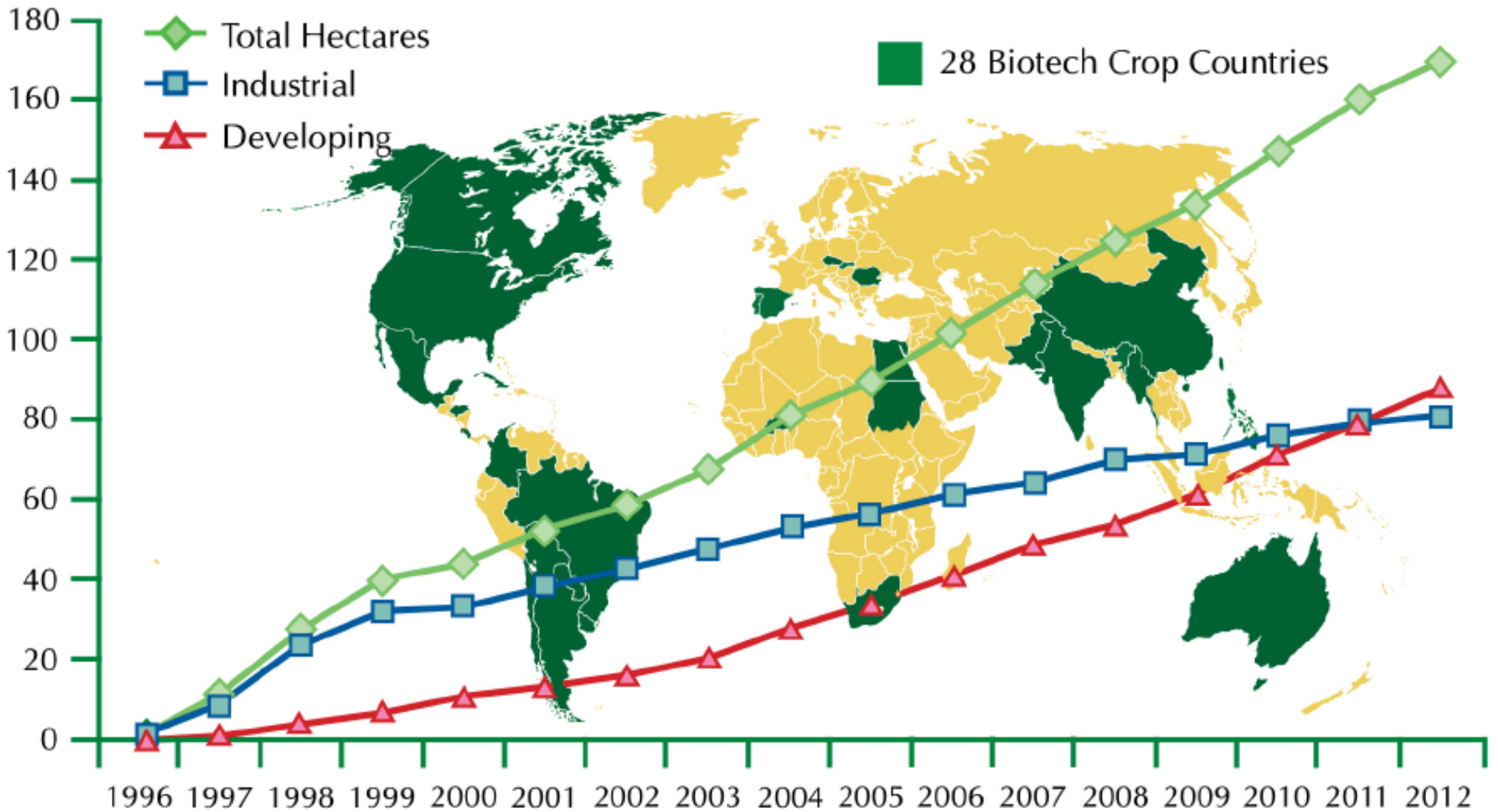
- 1961- Bt was registered as pesticide to the EPA
- 2002: Bt cotton was introduced in India
- India has the largest hectarage of cotton and one third of the total cotton are planted in the world
- Cotton yield increased from 308 Kg/ha in 2001-02 to 500 kg/in 2011-12.



# Major transgenic crops expressing Bt genes for Insect Resistance

<b>Transgenic Crop Plants</b>	<b>Foreign Gene</b>	<b>Target insect pests</b>
Cotton	Cry1A(b), Cry1A(c)	<i>H. armigera, H. zea, Heliothis virescens, Pectinophora gossypiella. S. exigua</i>
Maize	Cry1A(b), Cry1A(c), Cry9C	<i>Chilo partellus, H. zea</i>
Tomato	Cry1A(c)	<i>Manduca sexta</i>
Tomato	Bt(k)	<i>M. sexta, H. zea</i>
Rice	Cry1A(b), Cry1A(c), CryII(a)	<i>Scirpophaga incertulas, Cnaphalocrosis medinalis</i>
Potato	Cry 1A(b), Cry1A(b)6, CryIII A, CryIII B	<i>Phthorimaea operculella, Leptinotarsa sp.</i>
Tobacco	Cry1A(c )	<i>H. virescens, M. sexta,</i>
Brinjal	CryI AC	<i>Leucinodes orbonalis</i>

# GLOBAL AREA OF BIOTECH CROPS Million Hectares (1996-2012)

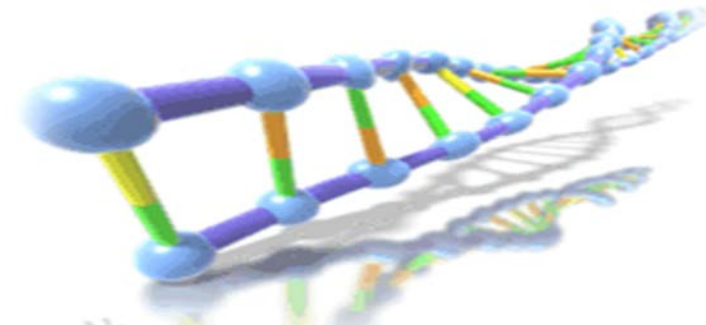


*A record 17.3 million farmers, in 28 countries, planted 170.3 million hectares (420 million acres) in 2012, a sustained increase of 6% or 10.3 million hectares (25 million acres) over 2011.*

A 3D ball-and-stick model of a DNA double helix, showing the sugar-phosphate backbone and nitrogenous base pairs. The model is rendered in shades of blue, purple, green, and orange, with a soft shadow cast below it.

# Requirements identified while producing transgenic plants

- Resistance should be controlled by single gene.
- Expression of transferred gene should occur in the desired tissue at the appropriate time.
- Safe for consumption
- Inheritance of the gene in the successive generations should be very stable.



# Plant derived genes



# Protease inhibitors

- Antimetabolic proteins which interferes with the process of digestion in insects- strategy by plants.
- Dietary protease inhibitors – detrimental to the growth and development of insects
- Ex: *Helicoverpa*, *Spodoptera*

# $\alpha$ – Amylase inhibitors

- Inhibit the digest enzymes of mammals and insects.
- Seeds of several varieties of common bean, *Phaseolus vulgaris* (BAAI) – exhibit resistance to bruchid beetles, *Callasobruchus* spp.
- Transgenic tobacco plants expressing amylase inhibitors from from wheat (wheat  $\alpha$ -amylase inhibitor, WAAI) increase the mortality of lepidopteran larvae by 30-40 per cent.

# Lectins

- ✦ Plant derived proteins that bind to oligo and polysaccharides
- ✦ Causes agglutination and cell aggregation.
- ✦ Carbohydrate binding lectin protein (including chitin) called phytohemagglutinin (PHA) found in seeds of common bean.
- ✦ It binds the chitin in peritrophic membrane of midgut thus interfere with nutrient uptake.
  - Wheat (wheat germ agglutinin, WGA) and snowdrop (*Galanthus nivalis* agglutinin, GNA) } Inhibitory to homopteran pests-aphids, plant hoppers and leaf hoppers
- ✦ Alternative to Bt delta endotoxins.

# Biotechnological methods employed for crop improvement

<b>Sl. No.</b>	<b>Technique</b>	<b>Application</b>	<b>Examples</b>
<b>1</b>	<b><i>Agrobacterium-based</i> plant transformation</b>	<b>Ti- plasmid –to carry novel DNA into plants</b>	<b>Bt insect resistant crop plants</b>
<b>2</b>	<b>Particle acceleration</b>	<b>DNA coated gold particles fired into growing tissue</b>	<b>Transgenic soybean</b>
<b>3</b>	<b>Electroporation</b>	<b>Electric current used to alter protoplast membranes permitting DNA uptake</b>	<b>Transgenic rice</b>
<b>4</b>	<b>Microinjection</b>	<b>DNA injected into the nucleus or cytoplasm of a protoplast</b>	<b>Transgenic tomato</b>
<b>5</b>	<b>RNA interference</b>	<b>Blockage of gene function by inserting short sequences of RNA</b>	<b>Potential for protecting cotton, rice and maize against insect pests</b>



# Genetic engineering of Predator and Parasitoids

- Transgenic strain of *Metaseilus occidentalis*  
Predator of spider mite
- Maternal microinjection
- Transgenic strain can be used routinely in applied pest management programme



(Hoy, 2000)

# Genetic improvement of predators & Parasitoids

- **Resistance to pathogens**
- **Resistance to pesticides**
- **Adaptation to different environmental conditions**
- **High fecundity**
- **Improved host seeking ability**



# Potentials of biotechnology in IPM

- Low toxicity of protease inhibitors and Bt  $\delta$ - endotoxin as compared to conventional insecticide.
- Expression of toxins in all plant parts - No need of continuous monitoring of pest.
- Provide protection to those plant parts which are difficult to be treated with insecticides.
- There is no drift problem and ground water contamination.
- Safe to non target species and human beings.
- Eliminate the problem of shelf life and field stability faced by pesticide formulation.
- Inbuilt resistance to various insects.

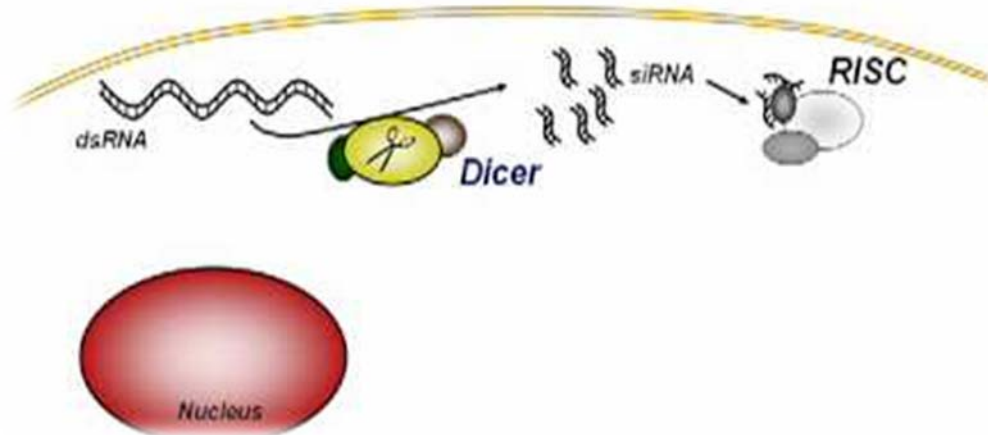
# **Risk associated with Biotechnological approaches**

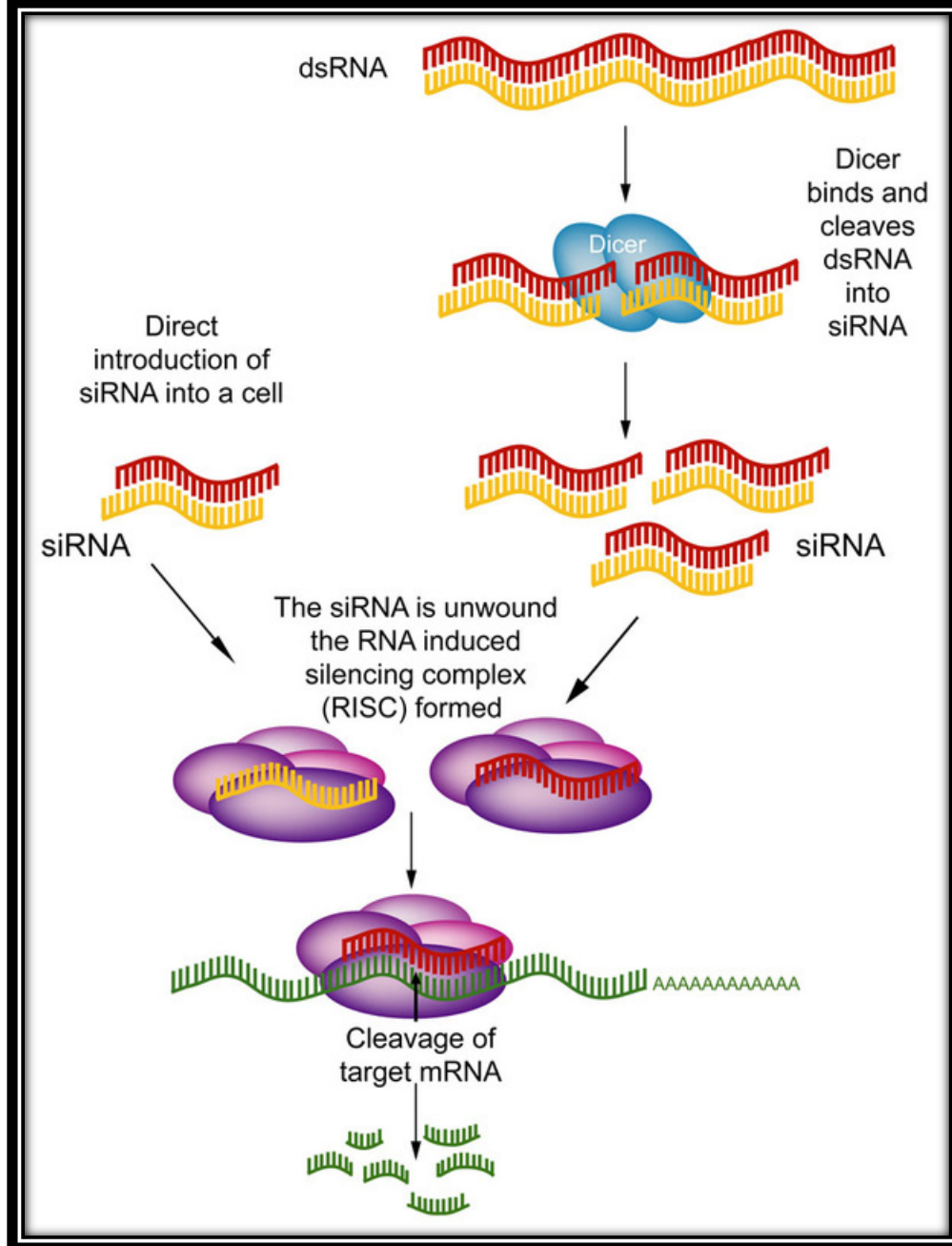
- **Human and animal health: Toxicity, food quality, allergenicity**
- **Risk for agriculture: loss of biodiversity, alternation in nutritional level, development of resistance.**
- **Risk for environment: persistence of gene, unpredictable gene expression, impact on non target organisms.**
- **Risk for horizontal transfer: interaction among different genetically modified organisms, genetic pollution through pollen or seed dispersal, transfer of gene to microorganism.**



# RNA interference

- Method of blocking gene function by inserting short sequences of double stranded ribonucleic acid (dsRNA) that match part of the target mRNA sequence, thus no proteins are produced.
- Knock down the expression of genes.





**Mechanism of RNA interference (RNAi) in cell**

# **Major Indian centres in transgenic research & application**

- **Seven such centres were set up initially at various Universities/Institutions namely,**
- **Jawaharlal Nehru University (New Delhi),**
- **Madurai Kamaraj University (Madurai),**
- **Tamil Nadu Agricultural University (Coimbatore),**
- **Osmania University (Hyderabad),**
- **National Botanical Research Institute (Lucknow) and**
- **Bose Institute (Kolkata).**
- **University of Delhi South Campus in 1997.**

# Conclusion

- **Biotechnological approaches play important role in insect-pest management.**
- **The efficacy of bio-control agents can be increased through rDNA technology**
- **DNA barcoding can help in quick and accurate identification.**
- **DNA fingerprinting helps for identification of biotypes and genetic changes in insect-pest.**