

Regional Joint BCH and ABSCH Training of Trainers Workshop for Africa Region

Nairobi, 7-11 October 2024

Modern Biotechnology and BCH

Prof. Ossama Abdelkawy

Records

introduced or modified genetic element(s)
and those genetic elements may be present in fragments or truncated forms. Please see notes below, where applicable.

<input checked="" type="checkbox"/>	BCH-GENE-SCBD-14972-12	PHOSPHINOTHRICIN N-ACETYLTRANSFERASE GENE
Protein coding sequence Resistance to herbicides (Glyphosate)		
<input checked="" type="checkbox"/>	BCH-GENE-SCBD-14985-12	CRY1AB BACILLUS THURINGIENSIS - BT, BACILLUS, BACTU
Protein coding sequence Resistance to diseases and pests (Insects, Lepidoptera (butterflies and moths))		
<input checked="" type="checkbox"/>	BCH-GENE-SCBD-14975-5	BETA-LACTAMASE GENE (BACTERIA)
Protein coding sequence Resistance to antibiotics (Ampicillin)		
<input checked="" type="checkbox"/>	BCH-GENE-SCBD-100287-7	CAMV 35S PROMOTER
Promoter		
<input checked="" type="checkbox"/>	BCH-GENE-SCBD-100290-6	CAMV 35S TERMINATOR
Terminator		

Genetic element
Promoter
Terminator
Marker gene
Agrobacterium
Coding sequence
Truncated gene
Unique identifier
Transformation cassette
Gene gun
Risk Assessment
Detection and identification

Description

This LMO contains two copies of a truncated synthetic version of the full length *cry1Ab* gene from *Bacillus thuringiensis* subsp. *kurstaki*. The synthetic truncated *cry1Ab* gene encodes a protein that corresponds to the first 648 amino acids of the N-terminal of the 1155 amino acid full length native Cry1Ab protein and includes the portion of the native protein that is necessary for insect control.

Also note that the cassette has genetic elements belonging to corn to dupe the plant cell so that it does not recognize that

Additional information concerning the *bla* gene insert in this LMO:
 The *bla* gene from *Escherichia coli* is not expressed in plant cells, but was employed as selectable trait for screening bacterial colonies for the presence of the plasmid vector.

Additional information on the inserted genetic material:

| Basic concepts

Cartagena Protocol Art.3

Use of Terms

For the purposes of this Protocol:

- (g) "Living modified organism" means any living organism that possesses a novel combination of genetic material obtained through the use of modern biotechnology;
- (i) "Modern biotechnology" means the application of:
- In vitro nucleic acid techniques, including recombinant deoxyribonucleic acid (DNA) and direct injection of nucleic acid into cells or organelles, or
 - Fusion of cells beyond the taxonomic family;
- that overcome natural physiological reproductive or recombination barriers and that are not techniques used in traditional breeding and selection;

| Basic concepts

Biosafety vs Biosecurity

Biosafety (Prevention des Risques):

The efforts to avoid and minimize the environmental and human health risk resulting from modern biotechnology and its products.

Biosecurity (Biosécurité):

The efforts to prevent misuse of hazardous biological materials through loss, theft or diversion.

Basic concepts

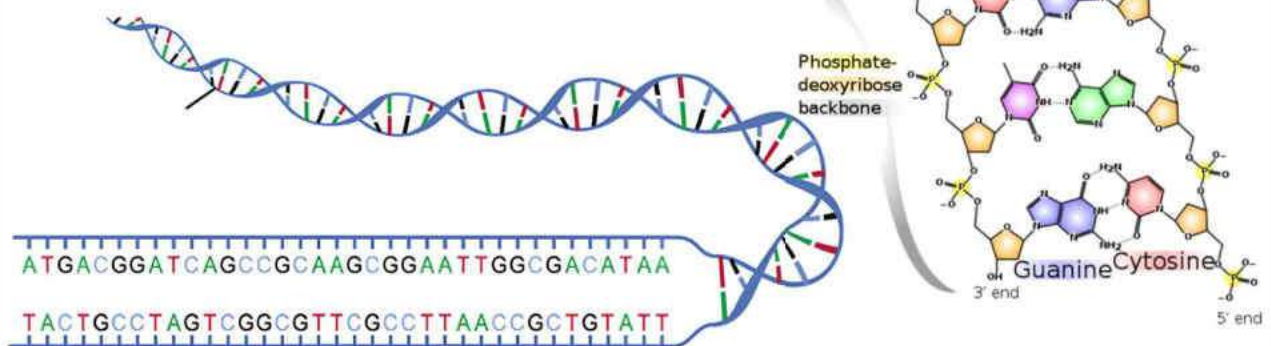
Genetic materials



The medium by which inherited characteristics of a living organism are transmitted from one generation to the next.

Basic concepts

Genetic code



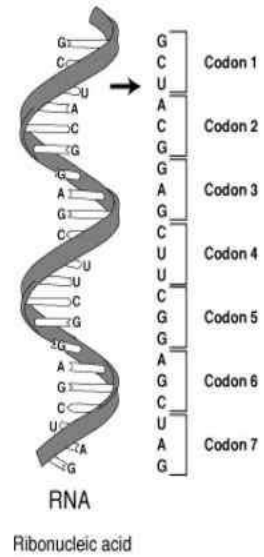
Sequences of nucleic acids that contain instructions for cell development and functions

Basic concepts

Genetic code

		1st base							
		U		C		A		G	
2nd base	U	UUU	Phenylalanine	UCU	Serine	UAU	Tyrosine	UGU	Cysteine
		UUC	Phenylalanine	UCC	Serine	UAC	Tyrosine	UGC	Cysteine
		UUA	Leucine	UCA	Serine	UAA	Stop	UGA	Stop
		UUG	Leucine	UCG	Serine	UAG	Stop	UGG	Tryptophan
C	CUU	Leucine	CCU	Proline	CAU	Histidine	CGU	Arginine	
	CUC	Leucine	CCC	Proline	CAC	Histidine	CGC	Arginine	
	CUA	Leucine	CCA	Proline	CAA	Glutamine	CGA	Arginine	
	CUG	Leucine	CCG	Proline	CAG	Glutamine	CGG	Arginine	
A	AUU	Isoleucine	ACU	Threonine	AAU	Asparagine	AGU	Serine	
	AUC	Isoleucine	ACC	Threonine	AAC	Asparagine	AGC	Serine	
	AUA	Isoleucine	ACA	Threonine	AAA	Lysine	AGA	Arginine	
	AUG	Methionine (Start)	ACG	Threonine	AAG	Lysine	AGG	Arginine	
G	GUU	Valine	GCU	Alanine	GAU	Aspartic Acid	GGU	Glycine	
	GUC	Valine	GCC	Alanine	GAC	Aspartic Acid	GGC	Glycine	
	GUA	Valine	GCA	Alanine	GAA	Glutamic Acid	GGA	Glycine	
	GUG	Valine	GCG	Alanine	GAG	Glutamic Acid	GGG	Glycine	

Nonpolar, aliphatic Polar, uncharged Aromatic Positively charged Negatively charged

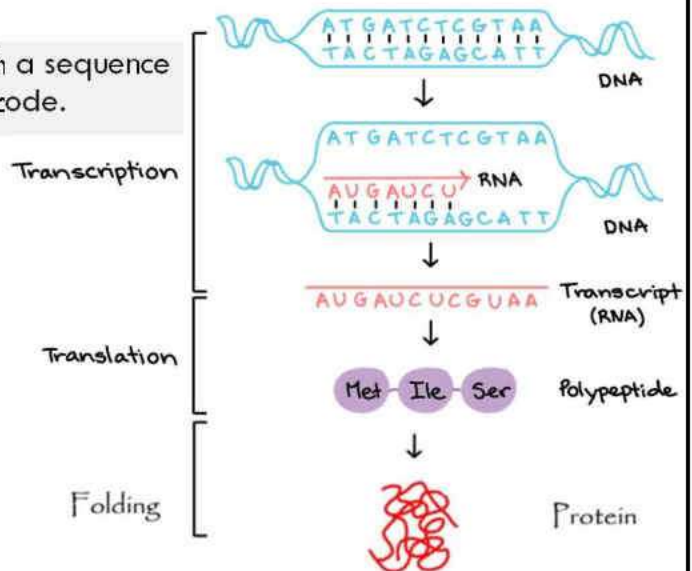
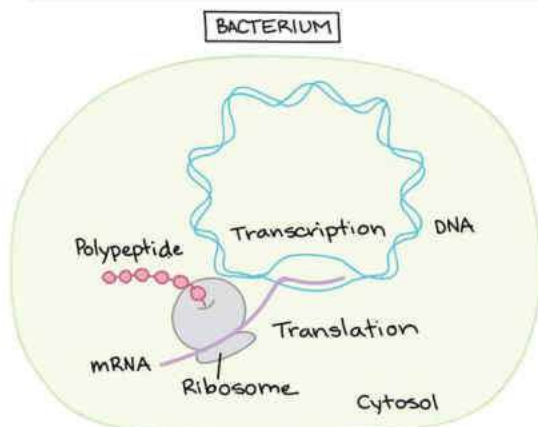


It is universal in all living organisms with negligible exceptions. Three consecutive bases (codon) code for one amino acid

Basic concepts

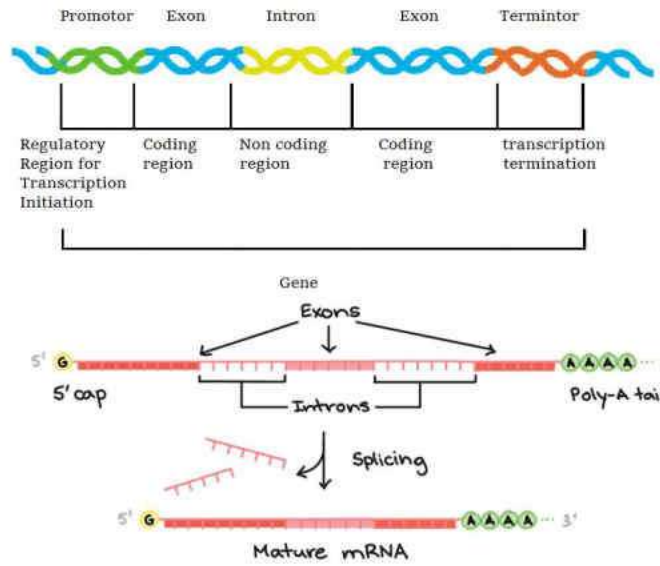
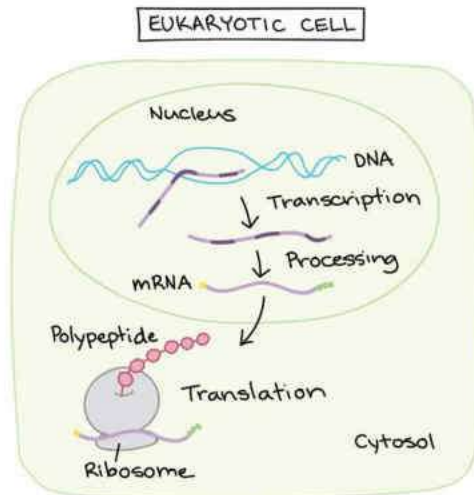
Gene expression

Is the synthesis of a specific protein with a sequence of amino acids encoded in the genetic code.



Basic concepts

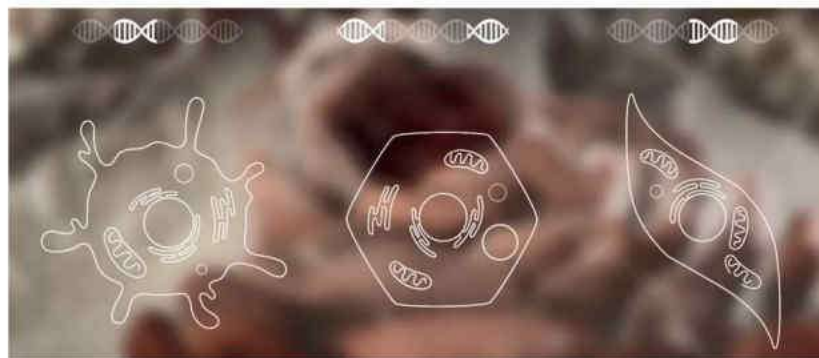
Gene expression



Basic concepts

Gene silencing

Our body consists of different types of cells (skin, muscles, or bone cells) with identical genetic materials.

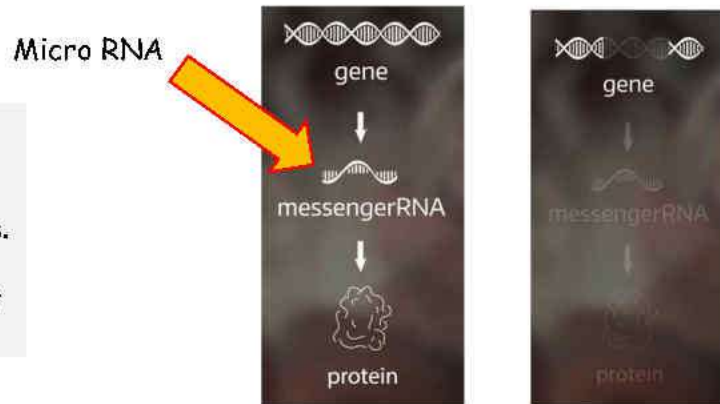


Through gene silencing genetic information is switched off so during development a cell only reads instructions that are necessary for gaining the characteristics structures and functions.

Basic concepts

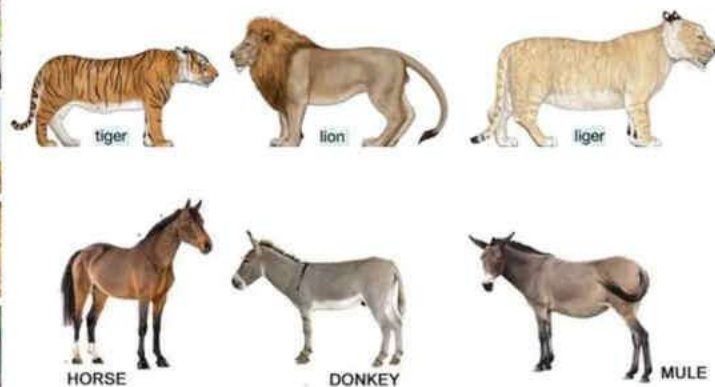
Gene silencing

Micro RNA molecules (miRNA) turn off genes by inactivating mRNA necessary for translating genetic information into proteins. They participate in regulating the cells from their development to their death.



Basic concepts

A species

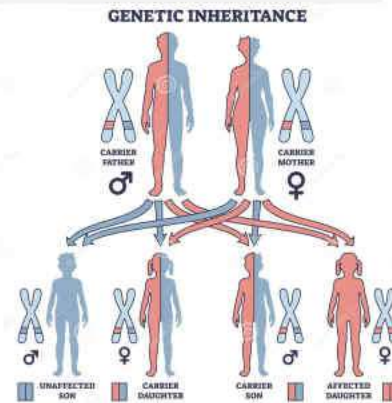
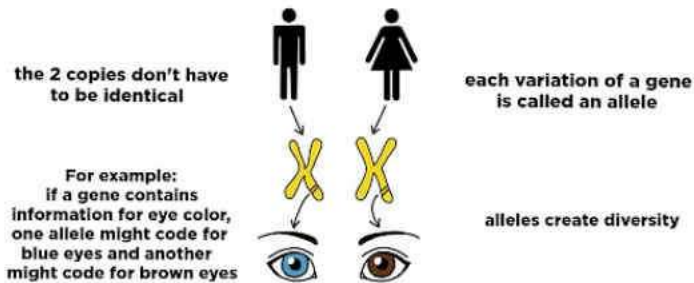


A group of living organisms consisting of similar individuals capable of exchanging genes or interbreeding to produce fertile offspring.

Basic concepts

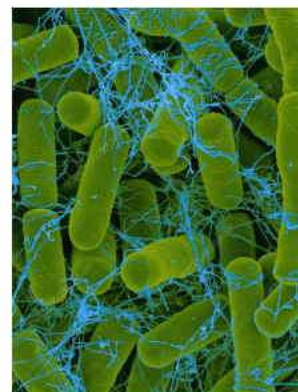
ALLELES

Individuals resulting from interbreeding inherit 2 copies of every gene, one copy from each parent.



Basic concepts

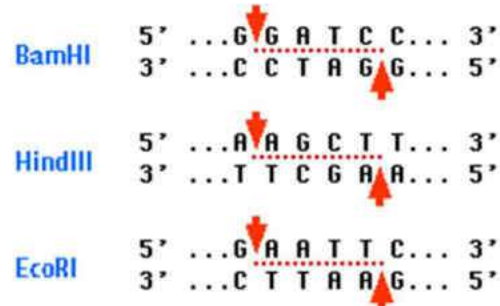
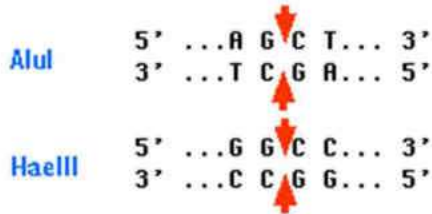
Genetic barriers between species



Each living cell can identify foreign genetic materials belonging to other species and will make it unfunctional by destroying it or by repairing its own DNA, creating barriers between species.

Basic concepts

Restriction enzymes



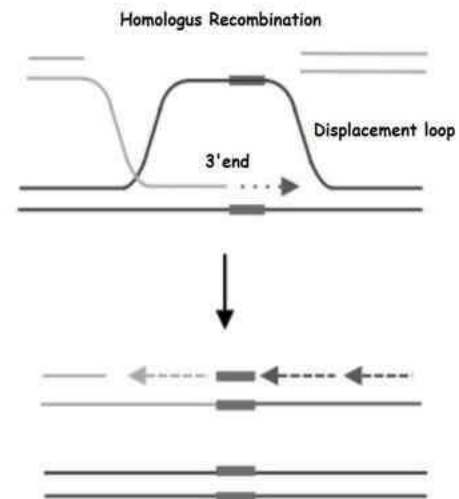
- Restriction enzymes are naturally occurring defense mechanisms to digest foreign DNA molecules.
- They recognize specific DNA sequences, mostly 4-6 bp, and cut DNA into fragments by breaking the phosphodiester linkage between two successive nucleotides of DNA.
- Now, if these restriction sites may be present in the organism's DNA, the DNA methylase enzymes carry out methylation of their DNA to protect it from digestion.

Basic concepts

Mechanisms for DNA Repair DSBs

Homologous Recombination (HR):

- Utilizes a homologous template (often a sister chromatid) to repair DSBs.
- Involves strand invasion and exchange of genetic material.
- High fidelity; preserves genetic information.
- Essential for meiosis and genetic diversity.
- Used in gene editing techniques like CRISPR.

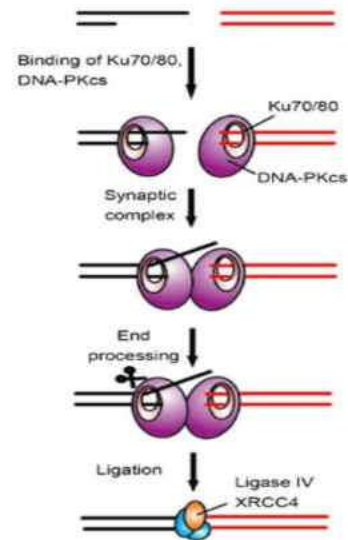


Basic concepts

Mechanisms for DNA Repair DSDBs

Non-Homologous End Joining (NHEJ):

- Directly joins the broken DNA ends without the need for a homologous template.
- Involves recognition of DNA ends and ligation.
- Prone to errors; can lead to insertions or deletions (indels).
- Important in response to radiation or other DNA-damaging agents.



Basic concepts

Genetic engineering (GE)– Modern Biotechnology



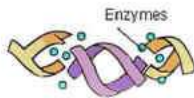
- Genetic material is altered or artificially introduced in vitro to induce a desirable new trait that does not occur naturally in the species.
- Inserted genes usually come from a different species.

Overview on the process of GE

1. Identify and isolate genetic sequence of interest from donor organisms and manipulate it in the laboratory to enhance their expression in the intended recipient organism.

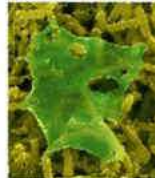


Bacillus thuringiensis

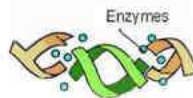


Cry1Ab gene

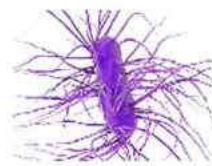
Resistance to Insects - Lepidoptera (butterflies and moths)



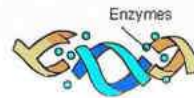
Streptomyces hygroscopicus



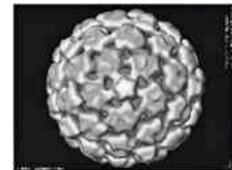
Phosphinothricin N-acetyltransferase gene
Resistance to herbicides - Glufosinate



Escherichia coli



Beta-lactamase gene
Resistance to antibiotics - Ampicillin



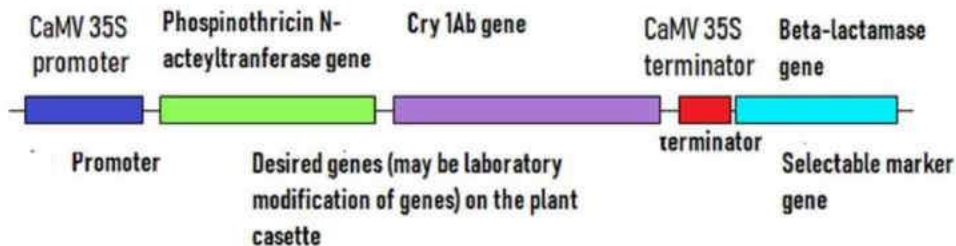
Cauliflower mosaic virus



CaMV 35S promoter
CaMV 35S terminator

Overview on the process of GE

2. Build the manipulated genes of interest and other nucleotide sequences needed for their proper functioning in an orderly sequence 'Transformation Cassette'



3. Finally, the cassette is integrated into the recipient organism's genome through a process known as 'transformation'

Overview on the process of GE

Techniques used for the modification

Search the list (min 3 chars to begin search) *0 keywords selected.*

- Agrobacterium-mediated DNA transfer
- Biolistic / Particle gun
- Cell fusion
- Cross breeding
- 'de novo' synthesis
- Direct DNA transfer
 - Electroporation
 - Heat shock
 - Microinjection
 - Osmotic shock
- Embryonic stem cell-mediated gene transfer
- Gene editing (e.g. CRISPR-Cas, etc.)
- Virus-mediated gene transfer
- Other

Commonly used techniques

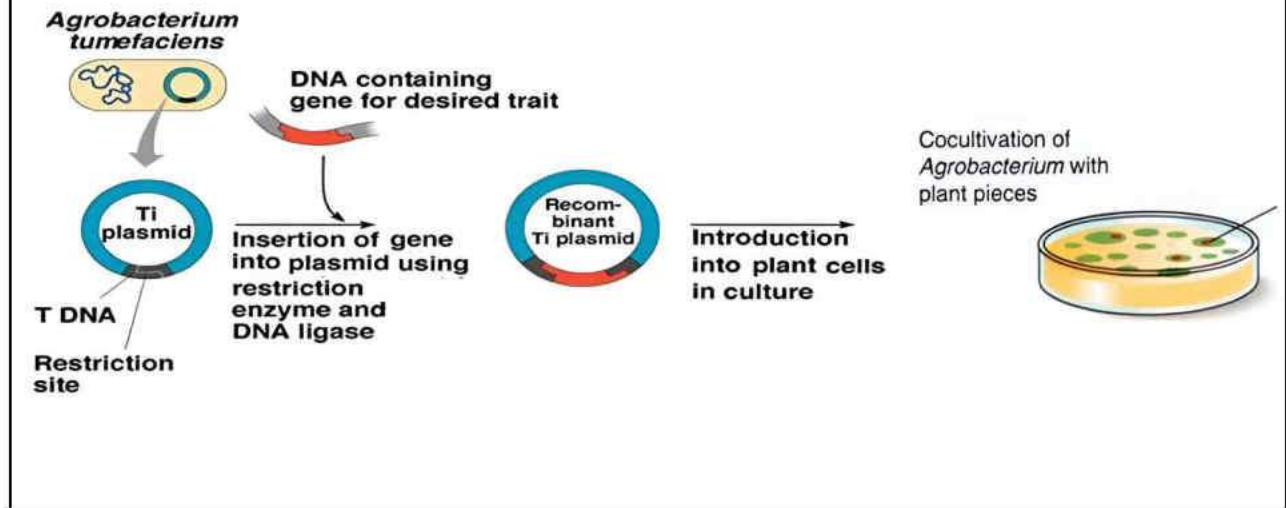
Transformation using *Agrobacterium tumefaciens*

- *Agrobacterium tumefaciens* is rod-shaped, Gram-negative soil bacterium.
- It is the causal agent of crown gall disease (the formation of tumors) in over 140 species of eudicots.



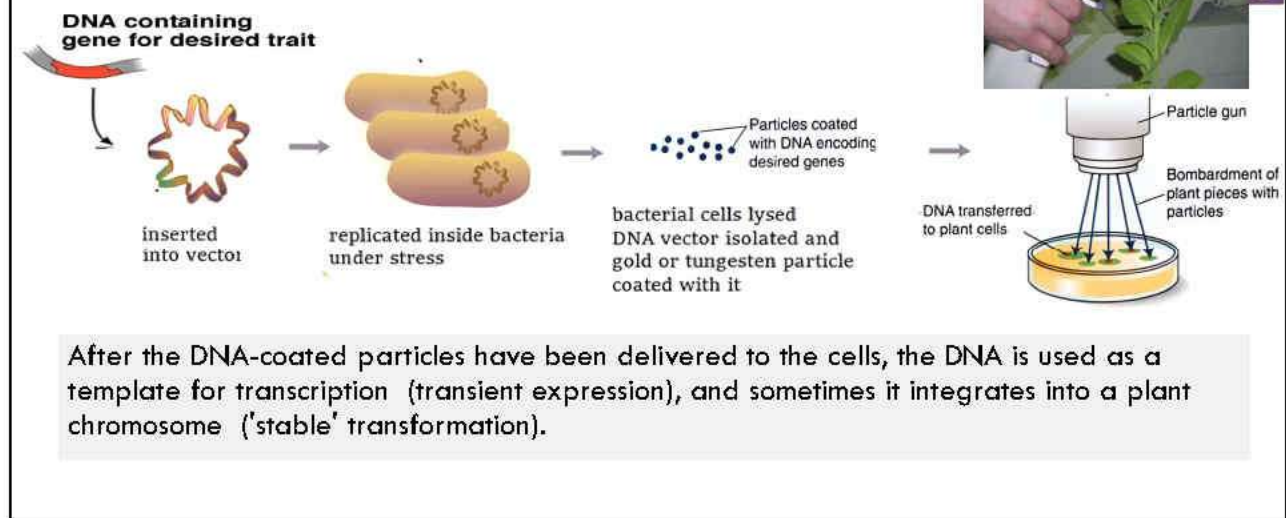
Commonly used techniques

Transformation using *Agrobacterium tumefaciens*

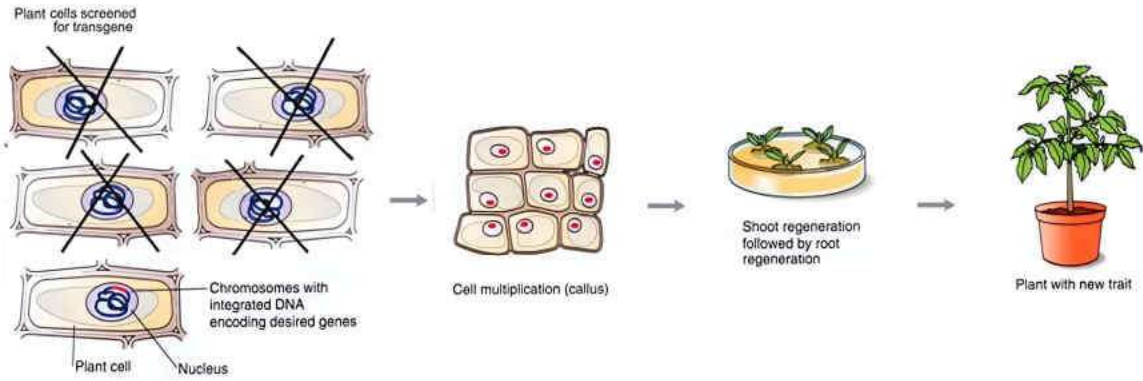


Commonly used techniques

Transformation using gene gun



LM plant generation



- Transformed cells are then selected, e.g., with the help of a marker gene
- Then are treated with a series of plant hormones, such as auxins and gibberellins, to divide and differentiate into an entire plant.
- The new plant that originated from a successfully transformed cell has new traits that are heritable (LMO).

Unique identifier

Genetic Element Registry

Total records: 928

Record ID	Unique identification	Identity & transformation event	Organism	Description
BCH-LMO-SCBD-11444-1	AAT-709AA-4	Pod Borer-resistant cowpea AAT709A	Vigna unguiculata Cowpea, Black eyed pea	Resistance to diseases and pests - Insects - Lepidoptera (butterflies and moths), Resistance to antibiotics - Kanamycin
BCH-LMO-SCBD-14752-6	ACS-BN0311-5	Navigator™ canola Oxy-235	Brassica napus Turnip, Rapeseed, Canola Plant, Oilseed Rape, Rape, BRANA	Resistance to herbicides - Bromoxynil
BCH-LMO-SCBD-15101-6	ACS-BN0318-4	Falcon™ rapeseed GS4030p4toe6/Ac	Brassica napus Turnip, Rapeseed, Canola Plant, Oilseed Rape, Rape, BRANA	Resistance to herbicides - Glufosinate
BCH-LMO-SCBD-14753-8	ACS-BN0301-4	inVigor™ canola RF1 (B93-101)	Brassica napus Turnip, Rapeseed, Canola Plant, Oilseed Rape, Rape, BRANA	Resistance to herbicides - Glufosinate, Resistance to antibiotics - Kanamycin, Changes in physiology and/or production - Fertility restoration
BCH-LMO-SCBD-14754-5	ACS-BN0302-5	inVigor™ canola RF2 (B94-2)	Brassica napus Turnip, Rapeseed, Canola Plant, Oilseed Rape, Rape, BRANA	Resistance to herbicides - Glufosinate, Resistance to antibiotics - Kanamycin, Changes in physiology and/or production - Fertility restoration
BCH-LMO-SCBD-14755-7	ACS-BN0303-6	inVigor™ canola RF3	Brassica napus Turnip, Rapeseed, Canola Plant, Oilseed Rape, Rape, BRANA	Resistance to herbicides - Glufosinate, Changes in physiology and/or production - Fertility restoration
BCH-LMO-SCBD-118285-1	ACS-BN0303-6 + MON-00073-7	Herbicide tolerant, male fertility restoring canola RF3 + RT73	Brassica napus Turnip, Rapeseed, Canola Plant, Oilseed	Resistance to herbicides - Glufosinate, Glyphosate, Changes in physiology and/or production - Reproduction, Fertility restoration

OECD Unique identifier

What is a unique identifier?

- It is a digital alphanumeric code for each living-modified plant approved for commercial use, including food or feed.
- Currently, there are discussions to extend to transgenic animals.
- Unique Identifiers are generated by the developers of a new transgenic plant and included in the dossiers that they forward to national authorities during the safety assessment process.
- Once approved, national authorities can forward the unique identifier to the OECD Secretariat for inclusion in the OECD's product database, from which the information is automatically shared with the Biosafety Clearing House.

OECD Unique identifier

Understanding the code

2 or 3 alphanumeric digits to designate the applicant

5 or 6 alphanumeric digits to designate the transformation event

One numerical digit for verification (to reduce errors by ensuring the integrity of the alphanumeric code)

MON = Monsanto
SYN = Syngenta
DAS = Dow Agro-Science
BCS = Bayer Crop-Science

MON-15985-7

SYN-EV176-9

DAS-Ø15Ø7-1

Genetic engineering – Modern biotechnology

What is a Stacked LMO?

- It is an LMO possessing new traits resulting from more than one transformation cassette. It can be produced by several approaches, including conventional cross-breeding involving two LMOs that are either single transformation events or already stacked events, the transformation of an LMO, or simultaneous transformation with different transformation cassettes or vectors.
- Accordingly, the cassettes containing the transgenes and other genetic elements inserted in the original transformation events may be physically unlinked (i.e., located separately in the genome) and can segregate independently.
- Stacked LMOs may occur in the field in cross-pollinating plants like maize (corn) if more than one LMO are planted near each other.

Genetic engineering – Modern biotechnology

What is a Stacked LMO?

- For stacked LMOs, the unique identifiers show the multiple combined GM events.

BCS-BNØ12-7 X **ACS-BNØØ3-6** X **MON-883Ø2-9** LMO with 3 stacked events

BCS-GHØØ2-5 X **BCS-GHØØ4-7** LMO with 2 stacked events



Searching for information

CASE STUDY (CSFI08)

You have recently been given a food product that indicates that it contains a genetically modified organism identified as 'SYN-EV176-9'. Use the BCH to answer the following questions:

- Q1. What type of organism is 'SYN-EV176-9'?
- Q2. How has 'SYN-EV176-9' been modified from its parent organism (i.e., what new characteristics does it display)?
- Q3. Is 'SYN-EV176-9' known by any other names?
- Q4. What gene has been inserted into 'SYN-EV176-9'? Where did the gene come from?
- Q5. Have any countries approved 'SYN-EV176-9' for human food, animal feed, or processing? Which ones?

Searching for information

CASE STUDY (CSFI08)

- Q6. Have any countries decided that 'SYN-EV176-9' cannot be used for any reason? If so, why?
- Q7. Where could you go for further information about this organism?
- Q8. What product does the inserted gene produce?
- Q9. What other organisms in the BCH have the same inserted traits as 'SYN-EV176-9'?
- Q10. What genes have been inserted into the other organisms to give these same traits?

Searching for information

CASE STUDY (CSFI15)

You are a phytosanitary officer in the Czech Republic. You are inspecting a field planted with MON-ØØ81Ø-6 - YieldGard™ maize.

- Q1. What stacked events can be present with this event?
- Q2. Are all of them approved in Malawi?
- Q3. Are any of those varieties banned in any European country, and why?
- Q4. If you are a phytosanitary officer in Malawi, how will you proceed if the corn shipment is labeled GMO-free?

Thank you !

For more information, please email

elkawyo@gmail.com

+201111561456

