UN environment programme

Project

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Detection and identification of GMOs

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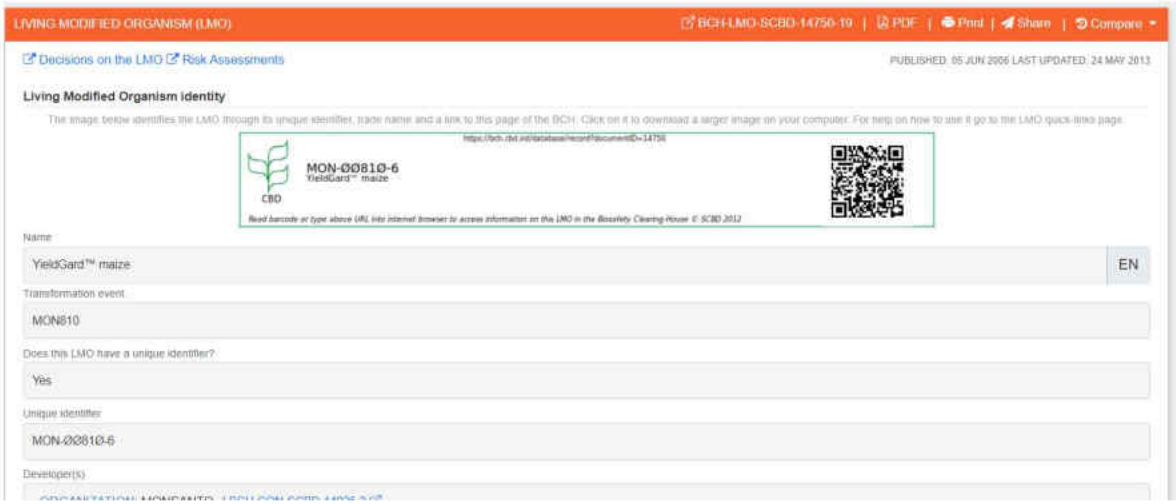
LMOs Records

https://bch.cbd.int/en/database/record?documentID=14750

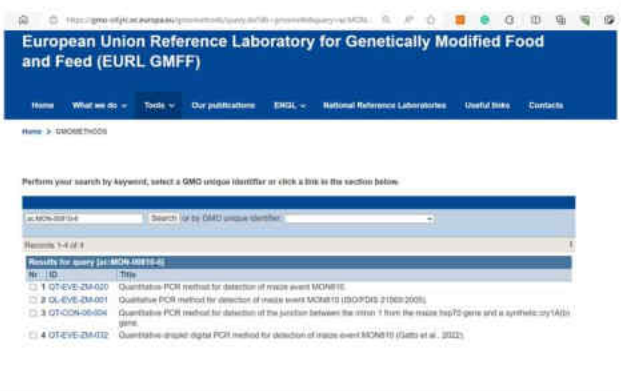


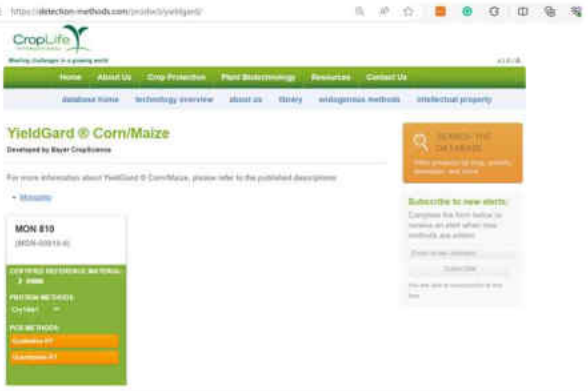
Detection and identification of GMOs

Detection method(s)

External link(s)

- MON-ØØ81Ø-6 - EU Reference Laboratory for GM Food and Feed (EURL-GMFF) *[English]*
- MON-ØØ81Ø-6 - CropLife International Detection Methods Database *[English]*
- MON-ØØ81Ø-6 - EU Reference Laboratory for GM Food and Feed (EURL-GMFF) *(JRC) [English]*
- MON-ØØ81Ø-6 - CropLife International Detection Methods Database *(CropLife) [English]*





Concepts and General Considerations

Why Detect GMOs?

- Biosafety Compliance: Adherence to regulations and standards for safe use of GMOs.
- Labeling and Traceability: Ensures accurate labeling and tracking of GMO content in products and meets consumer and market demands.
- Trade and Export: Supports countries in complying with international trade standards and maintaining the integrity of food and crop exports.
- Environmental Monitoring: Helps monitor the release of GMOs into the environment to safeguard ecosystems.

Concepts and General Considerations

General consideration for detection

Sampling:

- Determines how representative the results are of the lot from which the sample was taken.
- Heterogeneity, Sample Size and Particle Size, Lot Size and Impact on Limits of Detection (LOD) and Quantification (LOQ) need to be considered

Extraction:

- Extract proteins or DNA from the sample while minimizing interference.
- Matrix Effects (affect extraction efficiency) and Interferents.

Concepts and General Considerations

General consideration for detection

Reference materials:

- Serves as an external standard to validate methods and determine sensitivity and specificity.
- Ensure the reference material reflects the type and amount of GMO being tested.

Concepts and General Considerations

General consideration for detection

Method Validation:

- Specificity: Ability to accurately identify the target GMO without cross-reacting with non-target substances.
- Sensitivity: Capability to detect even small amounts of GMO.
- Accuracy: Correctness of the test results compared to known standards or reference materials.
- Reproducibility: Consistency of results across different tests, operators, and conditions.

Concepts and General Considerations

Brief overview of most commonly used methods

Protein-based Methods:

- Identify GMO-specific proteins expressed from the inserted genes.

DNA-based Methods:

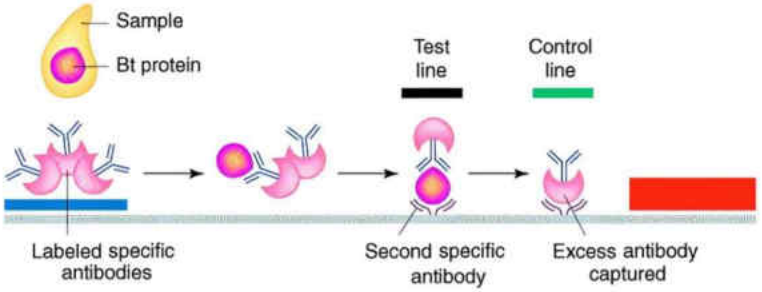
- Detect specific DNA sequences introduced in GMOs through genetic modification.

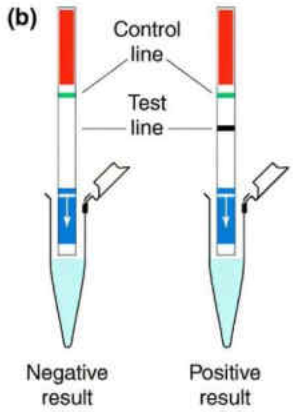
- E.g. Roundup Ready GM soy has been modified to resist the herbicide Roundup by inserting a gene that makes the soy plants produce a special enzyme (CP4 epsps) that can withstand glyphosate, the active ingredient in Roundup.

Key Considerations: The method selection depends on several factors such as available expertise, regulatory frameworks, sample types (raw, processed), and detection sensitivity requirements.

Protein based methods

Lateral flow strip



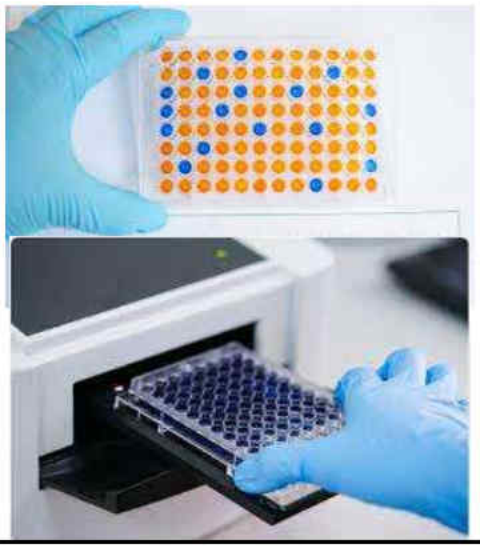


- Simple, portable test for on-site detection of GMO proteins. Strips use antibodies to bind GMO-specific proteins, with results visible as colored lines.
- Advantages: Fast, cost-effective for quick field testing.
- Limitations: Lower sensitivity and specificity compared to lab-based tests.

Protein based methods

ELISA (Enzyme-Linked Immunosorbent Assay):

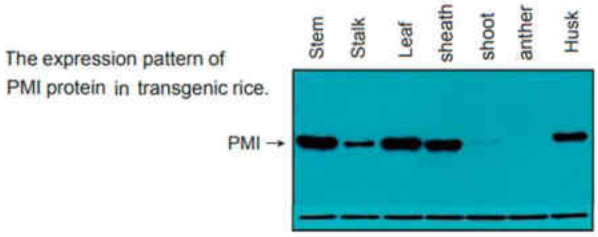
- Antibodies bind to GMO proteins in a micro-well plate, and a color change signals the presence of GMOs. ELISA can be quantitative or qualitative.
- Advantages: Widely used, high-throughput, suitable for screening.
- Limitations: Only detects specific proteins, making it unsuitable for processed products where proteins degrade.

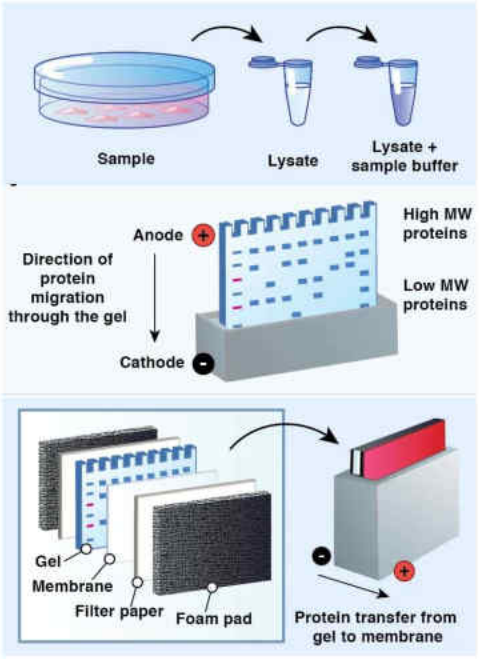


Protein based methods

Western Blotting

- Proteins are separated by size through gel electrophoresis and detected using antibodies.
- **Advantages:** High specificity for identifying individual proteins.
- **Limitations:** Time-consuming and complex compared to other methods.





Protein based methods

Limitations

- Cannot detect genetic elements that don't produce proteins (e.g., regulatory sequences or new gene silencing technologies like double-stranded RNA).
- Relies on antibodies recognizing a specific antigen in the transgenic protein, but changes in the protein structure during sample processing (due to heat or chemicals) can make this method ineffective.
- Detection capability is also influenced by the level of transgenic protein expression, which can vary in different tissues, life stages, and external factors like climate and soil.

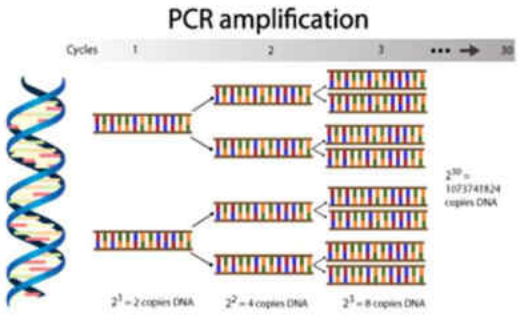
DNA based method

- PCR uses short synthetic DNA sequences ("primers") to replicate target DNA, allowing detection of GMO DNA in a sample.
- The type of PCR detection depends on the primers used, and it can be:
  - GMO screening: Detects common GMO elements, like the 35S promoter or NOS terminator, found in most GM crops.
  - Transgene-specific: Identifies specific genes, like Cry1Ab (insect resistance) or EPSPS (herbicide tolerance).
  - Construct-specific: Targets the region between two DNA elements, like the promoter and gene.
  - Event-specific: The most precise, detecting the unique junction between the host DNA and inserted gene.

DNA based method

Polymerase Chain Reaction (PCR):

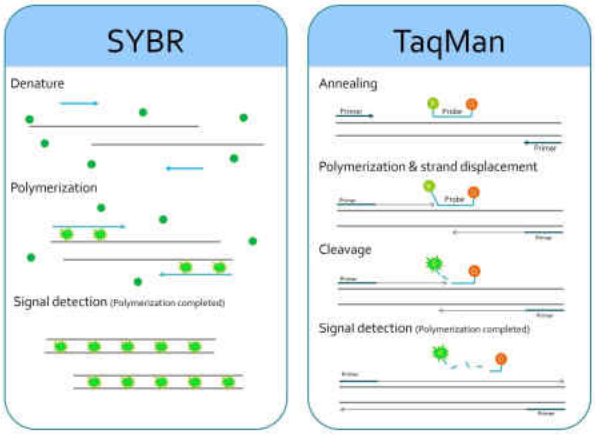
- Amplifies specific DNA sequences introduced through genetic modification (e.g., 35S promoter, nos terminator).
- **Advantages:** Highly sensitive, capable of detecting trace amounts of GMOs.
- **Applications:** Frequently used for compliance with labeling regulations and food traceability.



DNA based method

Real-Time PCR (qPCR):

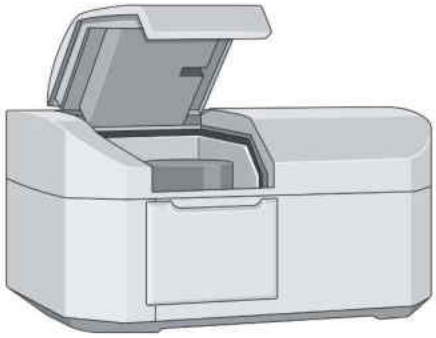
- A quantitative version of PCR that tracks DNA amplification in real-time, allowing for both detection and quantification of GMOs.
- **Advantages:** Provides precise quantification of GMO content, critical for compliance with thresholds (e.g., <1% GMO content in food).
- **Applications:** Determines GMO concentration in food products and crop samples.



DNA based method

DNA Microarrays:

- DNA samples are hybridized to multiple known sequences on a chip. Fluorescence signals indicate the presence of GMO-specific DNA sequences.
- **Advantages:** Allows for simultaneous detection of multiple GMOs in a single test, ideal for large-scale or high-throughput screening.
- **Applications:** Suitable for testing diverse food products or detecting GMOs in complex mixtures



DNA based methods

Advantages

- DNA has greater chemical stability than proteins, which allows it to withstand chemical and heat treatments.
- DNA is also present in all cells, and therefore any part of the organism can be used for testing.
- PCR methods are more versatile than protein methods, and PCR can be easily used to screen a sample for the presence of several potential LMOs simultaneously.

Detection and identification of GMOs

Challenges in GMOs Detection

- Emerging GM Technologies: Gene-editing techniques like CRISPR present challenges for traditional GMO detection, as these changes may not be as easily identifiable using current methods.
- Global Regulatory Differences: Varying thresholds for GMO content across countries create challenges for uniform detection and compliance, especially in international trade.
- Mixtures: It is not always possible to clearly distinguish between a sample containing material from different single transformation events and another sample with one or more stacked genetically modified (GM) events. Both may present similar detection profiles, making it difficult to differentiate between them using current testing methods.

Detection and identification of GMOs

Detection Method	Ease of Use	Need for Special Equipment	Duration	Cost per Sample (USD)	Employed Mainly In
Lateral Flow Strip	Very easy	Minimal (handheld reader)	~10-15 minutes	$2 - $10	On-site/field testing, rapid screening
ELISA	Moderate	Standard lab equipment (ELISA reader)	1-2 hours	$5 - $30	Laboratory-based, protein detection
Western Blot	Complex	Blotting apparatus and lab equipment	Several hours	$50 - $100	Research labs, protein analysis
PCR	Moderate	PCR machine required	2-4 hours	$10 - $50	Laboratory-based, DNA detection
Real-time PCR (qPCR)	Moderate	Real-time PCR machine required	2-4 hours	$50 - $100	Laboratory-based, precise DNA quantification
DNA Microarray	Complex	Specialized microarray equipment	1-2 days	$100 - $300	High-throughput GMO screening in research labs

Exercises

Searching for information

CASE STUDY (CSFI16):

You are a phytosanitary officer in Kenya. You received documentation for a cottonseed shipment to be imported from the United States (USA) for food processing. Use the BCH to answer the following questions:

Q1. What GM cotton might be in your shipment?

Q2. Are all of them approved to be imported or domestically used in Kenya?

Q3. How will you proceed if the shipment is labeled as 'GMO-free?'

Q4. How will you proceed if the shipment is labeled as it might contain GMO?

Searching for information

CASE STUDY (CSFI18)

A company based in Malawi is willing to import corn for cultivation from Malawi trade partners in the SADC Region. Use the BCH to answer the following questions:

Q1. Which corn GM varieties can be in the shipments?

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 !

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