Tiered approach to evaluating impact of GM crops on non-target organisms

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Assessment of risk of insect-resistant transgenic crops to nontarget arthropods

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Objective

To develop a **scientifically-sound, generic, and rigorous** approach to assessing the risks of **insecticidal transgenic crops** to non-target organisms, with emphasis on **terrestrial arthropods**, that meets the needs of environmental decision makers.
Involvement of scientists from diverse institutions (Europe & North America)

- Public research institutes
- Agricultural biotechnology industry
- Regulatory agencies
- Commercial testing laboratory

The group has experience in environmental risk assessment from a research and regulatory perspective
Subgroups - Topics

Problem formulation

Framework

Species selection

Study design
Problem formulation

I. Defines **scope of the risk assessment**
   
   - Identifies **assessment endpoints** reflecting **management goals** (protection goals, policy)
Problem formulation

I. Defines **scope of the risk assessment**
   - Identifies **assessment endpoints** reflecting **management goals** (protection goals, policy)

Example

*Management goal* – „protection of biodiversity“

*Assessment endpoint* – measurable attribute of the environment; e.g. abundance of certain NT arthropods
Problem formulation

I. Defines **scope of the risk assessment**

- Identifies **assessment endpoints** reflecting **management goals** (protection goals, policy)
- Generates relevant **risk hypotheses** (changes to assessment endpoints)
Problem formulation

I. Defines **scope of the risk assessment**

- Identifies **assessment endpoints** reflecting **management goals** (protection goals, policy)
- Generates relevant **risk hypotheses** (changes to assessment endpoints)
- Identifies **data requirements** (data to provide powerful tests of the risk hypotheses)
Problem formulation

II. Considers precursor information

Are there meaningful differences between the GM plant and its non-transformed comparators besides the introduced trait?
Crop/plant characterization

Agronomic/ morphological characterization

- Dormancy
- Growth
- Reproduction
- Seed dispersal
- Volunteer potential
- Insect-, disease-plant interactions
- …

Compositional analysis

- Macro- and micronutrients
- Toxicants
- Anti-nutrients
- …
Problem formulation

II. Considers **precursor information**

Are there meaningful differences between the GM plant and its non-transformed comparators besides the introduced trait?

- **if No**, the remaining ERA is focused on the expressed trait as stressor (insecticidal protein)
- **if Yes**, then the novel or different characters of the plant become additional stressors that also need to be evaluated
Typical risk hypothesis for regulatory risk assessment

„The expressed Bt protein is not toxic to non-target organisms at the concentration present in the field“
Stressor characterization

- Expression profile (time, tissue, level, etc.)
- Agronomic practice (location, timing, area, etc.)

⇒ Identifies NTOs likely to be exposed
Cry3Bb1 concentrations in maize arthropods

Predators

Phloem feeders

Mesophyll feeders

Pollen feeders

Others

Stressor characterization

• Expression profile (time, tissue, level, etc.)
• Agronomic practice (location, timing, area, etc.)
  ➡ Identifies NTOs likely to be exposed

• Mode of action
• Spectrum of activity against pests (or NTOs)
  ➡ Identifies NTOs likely to be sensitive

➢ Guides risk assessment and testing requirements
Properties of the framework (how to test)

- Conduct Field Studies
  - Sufficient Data?
- Conduct Semi-Field Studies
  - Sufficient Data?
- Conduct Laboratory Studies
  - Sufficient Data?
- Analyze Available Data

Move through the framework to acquire sufficient data to make a regulatory decision
Example framework

Field Studies

Semi-Field Studies

Laboratory Studies

Previous Data

Sufficient Data for Decision

Not all hypotheses require the same testing
Testing risk hypotheses

• “Testing” does not imply that a new study is required
• Existing data may corroborate risk hypotheses with sufficient certainty

Transgenic crops expressing *Bacillus thuringiensis* toxins and biological control
Jörg Romeis, Michael Meisels & Franz Bigler
Testing risk hypotheses

Example

Cry protein expressed in a new crop (e.g., Cry1Ab expressed in pigeonpea)

- Collect new data on protein expression
- Review existing data on the fauna associated with the crop in the proposed area of cultivation (e.g., India)
- Existing data on Cry protein toxicity (collected for cotton and maize risk assessments) may be sufficient to demonstrate low risk to non-pest species of pigeonpea expressing these proteins

Species selection

Select appropriate species to serve as surrogates for ecologically and economically important non-target organisms that can be tested to provide relevant data at proportionate costs in the laboratory.
Species selection - criteria

- Representation of different ecological functions
- Representation of the receiving environment

Key species or guilds that are representatives of different functional groups are known in most systems. Appropriate surrogates can therefore be selected that are relevant for the agro-ecosystem.
Species selection - criteria

- Representation of different ecological functions
- Representation of the receiving environment
- Information about the stressor (specificity, exposure profile)

Example 1
Cry3 proteins to provide protection against beetle pests (Corn rootworms, Boll weevil) are more likely to affect other Coleoptera than species belonging to other taxonomic groups.
Species selection - criteria

- Representation of different ecological functions
- Representation of the receiving environment
- Information about the stressor (specificity, exposure profile)

Example 2
Honeybees are only exposed to insecticidal proteins expressed by GM maize varieties when these are present in the pollen.
Species selection - criteria

- Representation of different ecological functions
- Representation of the receiving environment
- Information about the stressor (specificity, exposure profile)

- **Amenability** for testing
- **Availability** of test methods
- **Taxonomic recognition**
- **Anthropocentric values**

Adoption of the *surrogate species* concept
Study design – general requirements

- Purpose and **objectives** of the study clearly defined (directed by problem formulation)
- Study must provide data that are **interpretable** and can be related to an assessment endpoint
- Study results should assist decision-making by **reducing uncertainty** in the risk assessment
Study design – tiered assessment

**Laboratory studies**
- Effects tests
  - Transgenic plant material
  - Purified toxins
    - Elevated ("worst-case") dose
    - Dose response

**Semi-field studies**
- Tri-trophic studies
- Life-cycle tests

**Field studies**
- Study faunistic or population dynamics
- Complexity leads to increase in costs and labor

Complexity
Advantages of laboratory studies

- **Controlled conditions**
  - abiotic conditions
  - quality, age, etc. of test organisms
  - negative/positive control (thresholds)

  => **standardized**, replicable

- **Isolation** of biological impact of concern

- **Worst-case exposure** conditions

- High number of **replications** – high statistical power
Risk assessment continuum

Laboratory → Extended lab/semi-field → Field

Power in evaluation of hazard
Risk assessment continuum

- **Laboratory**
- **Extended lab/semi-field**
- **Field**

- Increase in realism of assessment
- Increase in ecological complexity
- Reduction in generality

- Power in evaluation of hazard
- Evaluation of consequences of hazard
Study design – considerations

(i) Specific measurement endpoints
• Depends on purpose of study
• Should be related to assessment endpoints

(ii) Life-stage to be tested

Selection criteria
• Level of likely exposure (adult vs. larva)
• Sensitivity to the insecticidal compound
• Amenability to testing („validated“ test system)
Study design – considerations

(iii) Availability of test protocols

• modified to account for
  - oral exposure pathways
  - mode of action of insecticidal proteins

(iv) Test validation (quality control standards)

• Assures repeatability, interpretability and quality of the study

• GLP standards recommended / mandatory

• Need for complete study/ data reconstructability
Higher tier testing

Conduct only when they

- **Reduce uncertainty** in the risk assessment
- **Are justified** by detection of unacceptable risks at lower tiers of testing
- When early tier studies are not possible
- Can be performed under conditions and rigour necessary to produce interpretable results
Conclusions (I)

- The approach ensures testing of clearly stated relevant hypotheses
- Tiered evaluation of potential hazards with representative surrogate species and conservative exposure estimates provides a rigorous and effective basis for estimating risk
- It thus minimizes the likelihood of false negatives which could result in the release of GM plants with undesirable effects
Conclusions (II)

- The approach minimizes collection of data that are irrelevant to the risk assessment.
- It makes maximum use of information that is already available.
- Decisions about acceptable risk can be made in a reasonable period of time.