
Laboratory test strategy to investigate side-effects of genetically modified plants on the habitat function of soils



Fraunhofer

Institut
Molekularbiologie und
Angewandte Oekologie

April 20th, 2004

**Kerstin Hund-Rinke
Markus Simon**

Stepwise procedure for an ecotoxicological risk assessment of chemicals/pesticides

Chemical / pesticide

I. Single species tests (guidelines; lab):

Screening tests: Basic information on potential risk

Assessment: Comparison of effects with threshold values or environmentally relevant concentration → potential risk existent?



?

Risk assessment of GMPS

yes

II. Field test:

Specific information: Results - high environmental relevance

Long test duration; interpretation more difficult (manifold interactions, abiotic/biotic variability)



Essential information concerning the habitat function

Microflora

Function



Structure

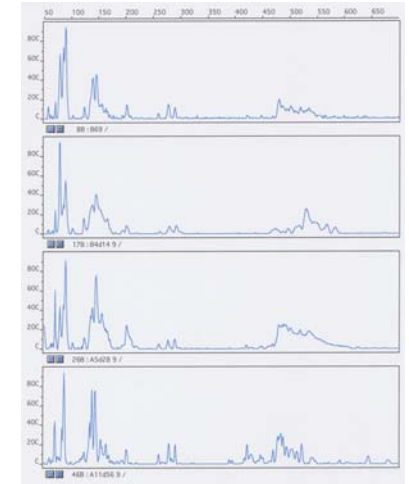
Soil organisms

Selected methods available from chemicals/pesticides testing

Micro-organisms:

Function:

- Respiration (e.g. ISO 17155)
- Nitrification (e.g. ISO 15685)



Structure of microbial biocoenosis:

- finger prints: e.g. PLFA, DGEE, T-RLFP

Soil fauna:

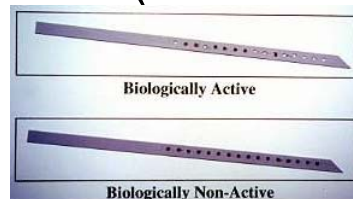
- Earthworms: reproduction test (ISO 11268-2)
- Collembola: reproduction test (ISO 11267)



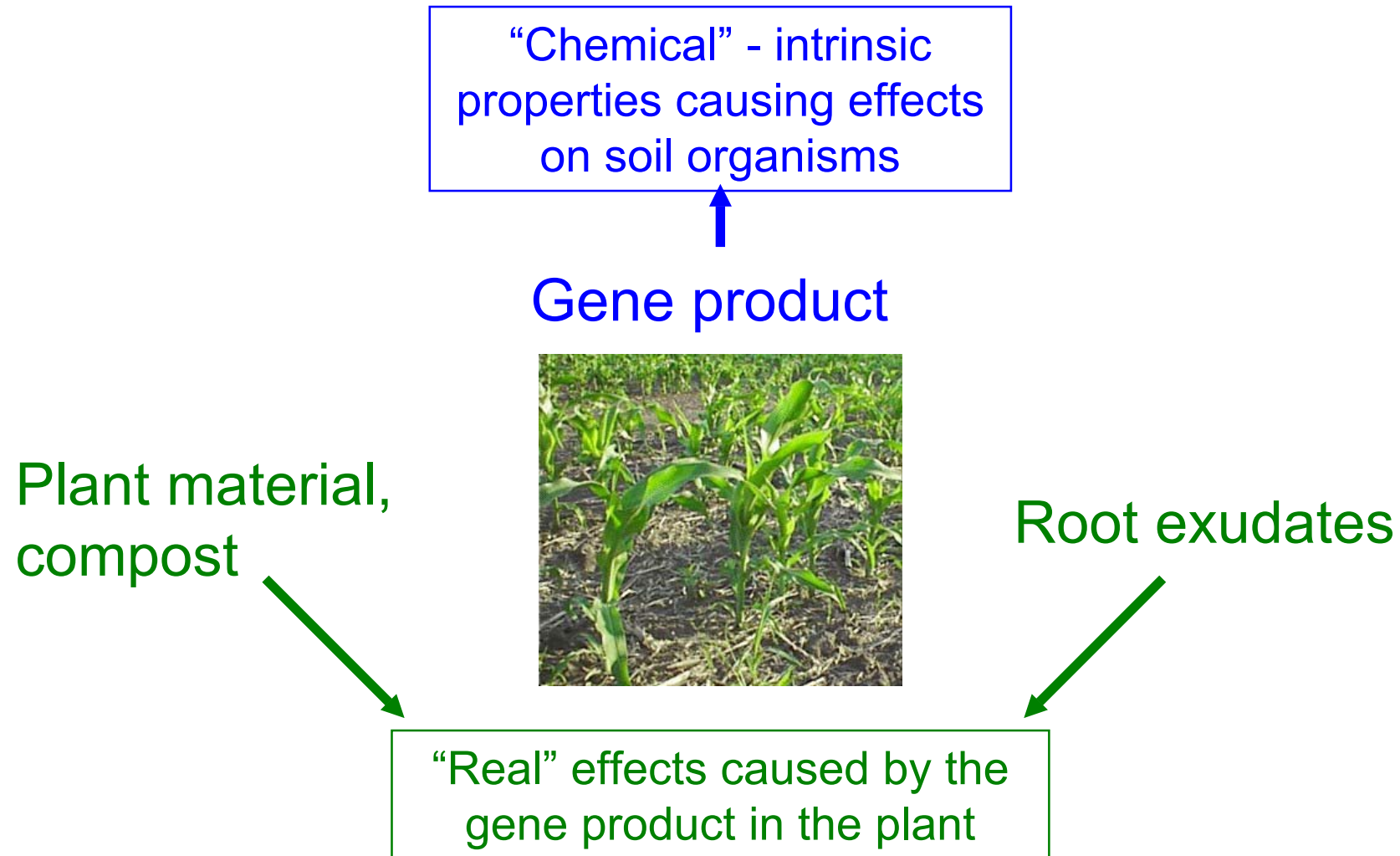
- Different exposure routes
- Sensitive endpoint

Functional test:

- bait lamina



Information for risk assessment regarding the habitat function



Test design

Prerequisite: sufficient amount of the substance

I. Application to soil

II. Measurement of effect: microflora, representative soil organisms

**Plant material,
compost**

Gene product

Root exudates



- I. Growing of GMPs, drying of plant material, grinding and application to test soil**
- II. Measurement of effect: microflora; representative test organisms, feeding with plant material instead of food described in guidelines**

- I. Use of soil in which GMPs were grown**
- II. Measurement of effect: microflora, representative soil organisms**

Test soil

Soil according to test philosophy of standardized ecotox tests:

Sandy soil (worst case: minimized sorption of test product)

Recommendation:

Worst case, taking special requirements of GMP into account

Example

Lysozyme

Pure substance (analytical quality)

Bt-Toxin:

Pesticide

Synthesized Bt-Toxin

Bt-maize – plant material

Bt-maize – root exudates

2 Soils:

Sandy soil – low sorption capacity

Loamy soil

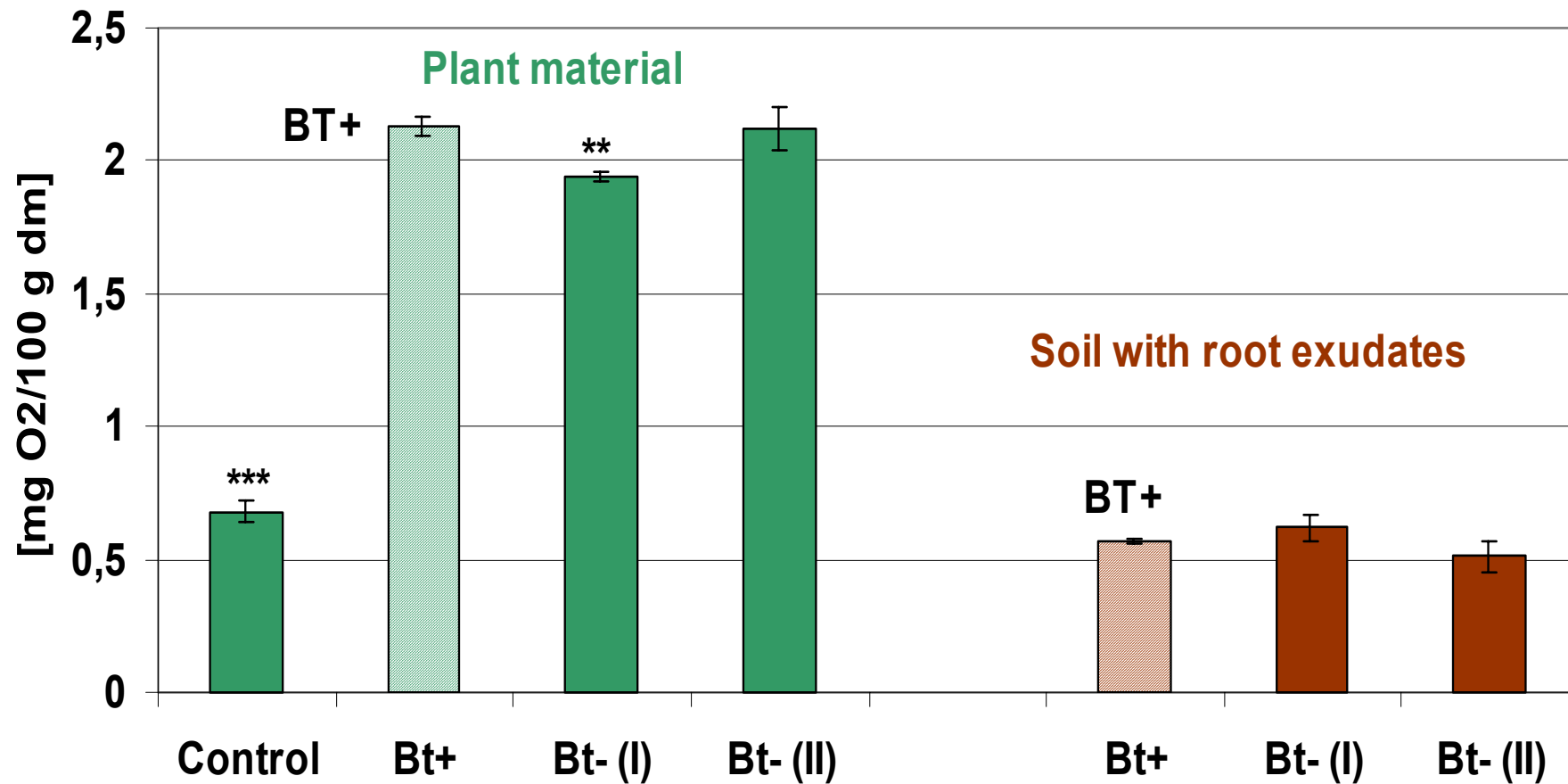
Testing of “pure substance” in a sandy soil (I)

Test parameter	Bt-pesticide (Dipel) (environmentally relevant concentration)	Cry1Ab (synthesized Bt-toxin; comparable concentration)
Micro-organisms: basal respiration	+	-
Micro-organisms: SIR	-	+
Nitrification	-	+
PLFA: gram+/gram -	+	-
Earthworms: reproduction	+	-
Collembola: reproduction	-	-
Bait lamina	-	-

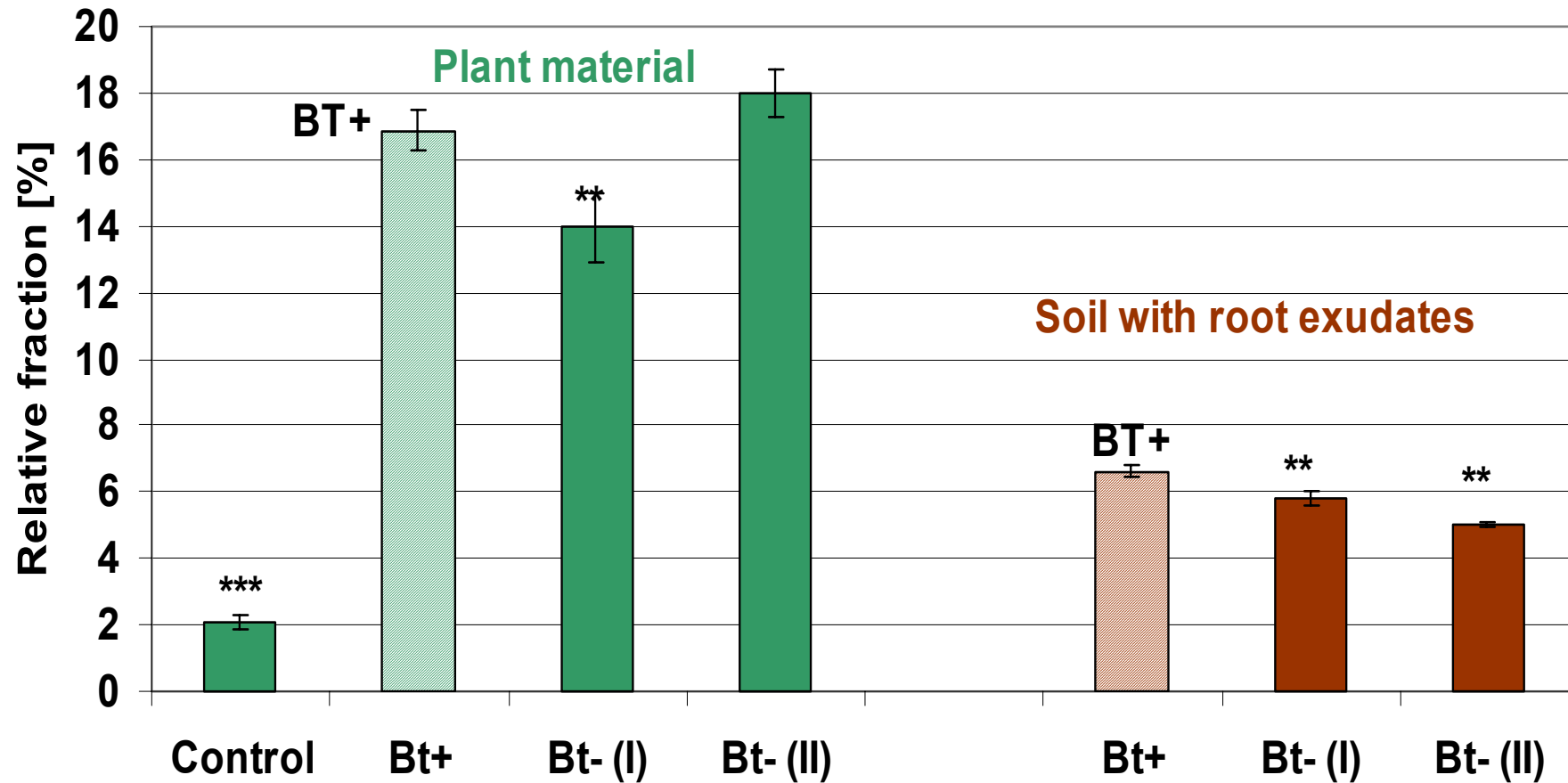
Testing of “pure substance” in a sandy soil (II)

- Due to the different effect patterns
 - Effects only in the sandy soil, no effect in the soil with high sorption capacity
- Ecotoxicological test systems are also suitable to detect effects caused by gene products

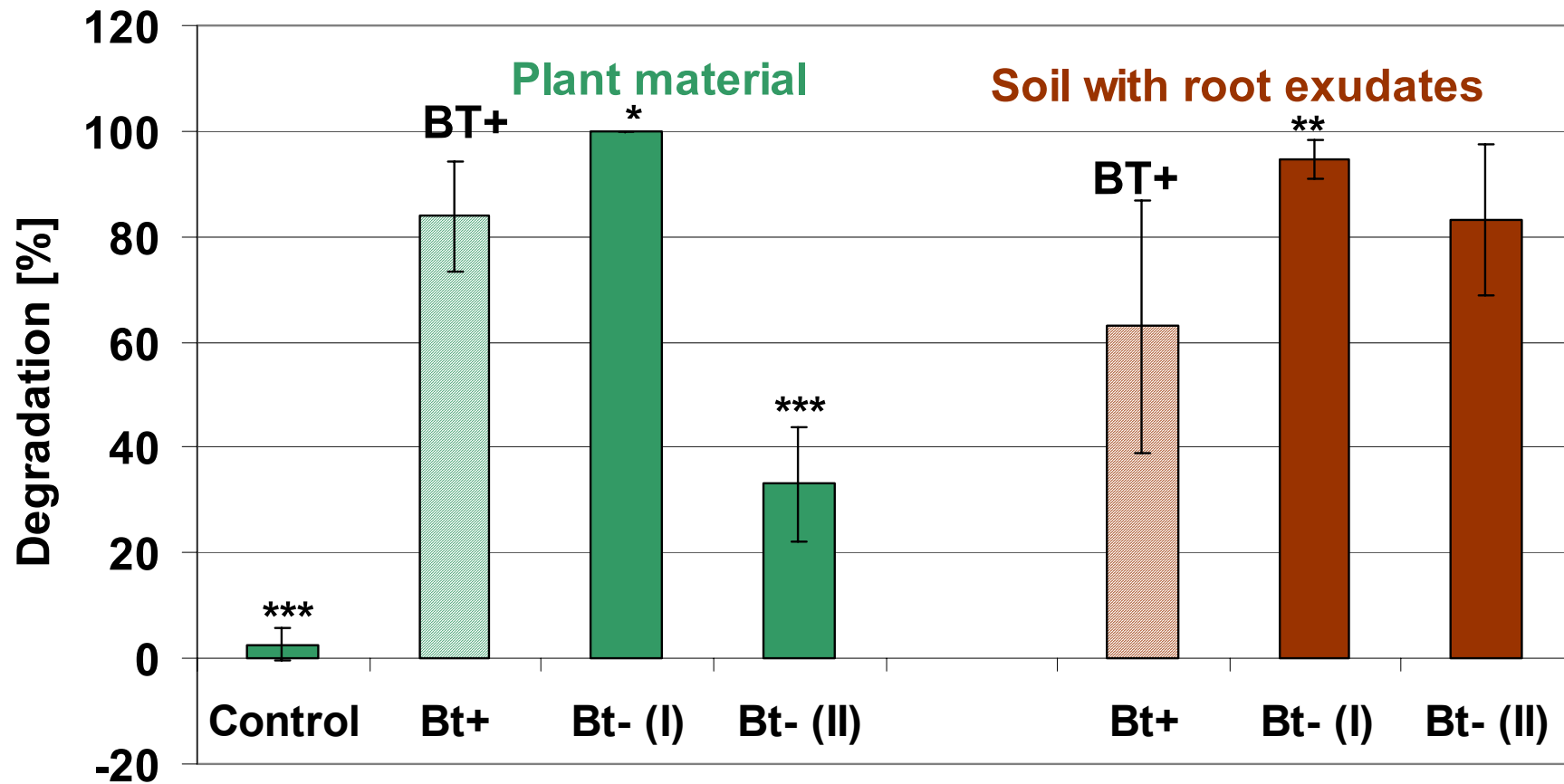
Testing of plant material / root exudates: SIR



Testing of plant material / root exudates: PLFA (Fungi)



Testing of plant material / root exudates: bait lamina



Summary: Screening of GMPs using ecotoxicity tests

- Applied test methods (developed for the testing of chemicals) are also suitable for the testing of GMPs
- Test design (testing of “gene product”, plant material, root exudates) is practicable
- Feeding of soil organisms with GMP is principally possible; adaptation necessary for each plant (nutrient status of the plant has to be taken into account)
- Assessment of plant material and root exudates: several isogenic plants have to be tested
- Test duration is shorter compared to field experiments

Conclusion: Screening of GMPs using ecotoxicity tests

- Differences can be pointed out → a potential risk may be existent
- Stepwise approach successfully applied for the risk assessment of chemicals → principally accepted approach → it should also be accepted for the assessment of GMPs concerning effects of the habitat function of soils