

# Fitness and life history: incorporating sublethal effects on invertebrates into risk assessment of GMO toxins

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## Introduction

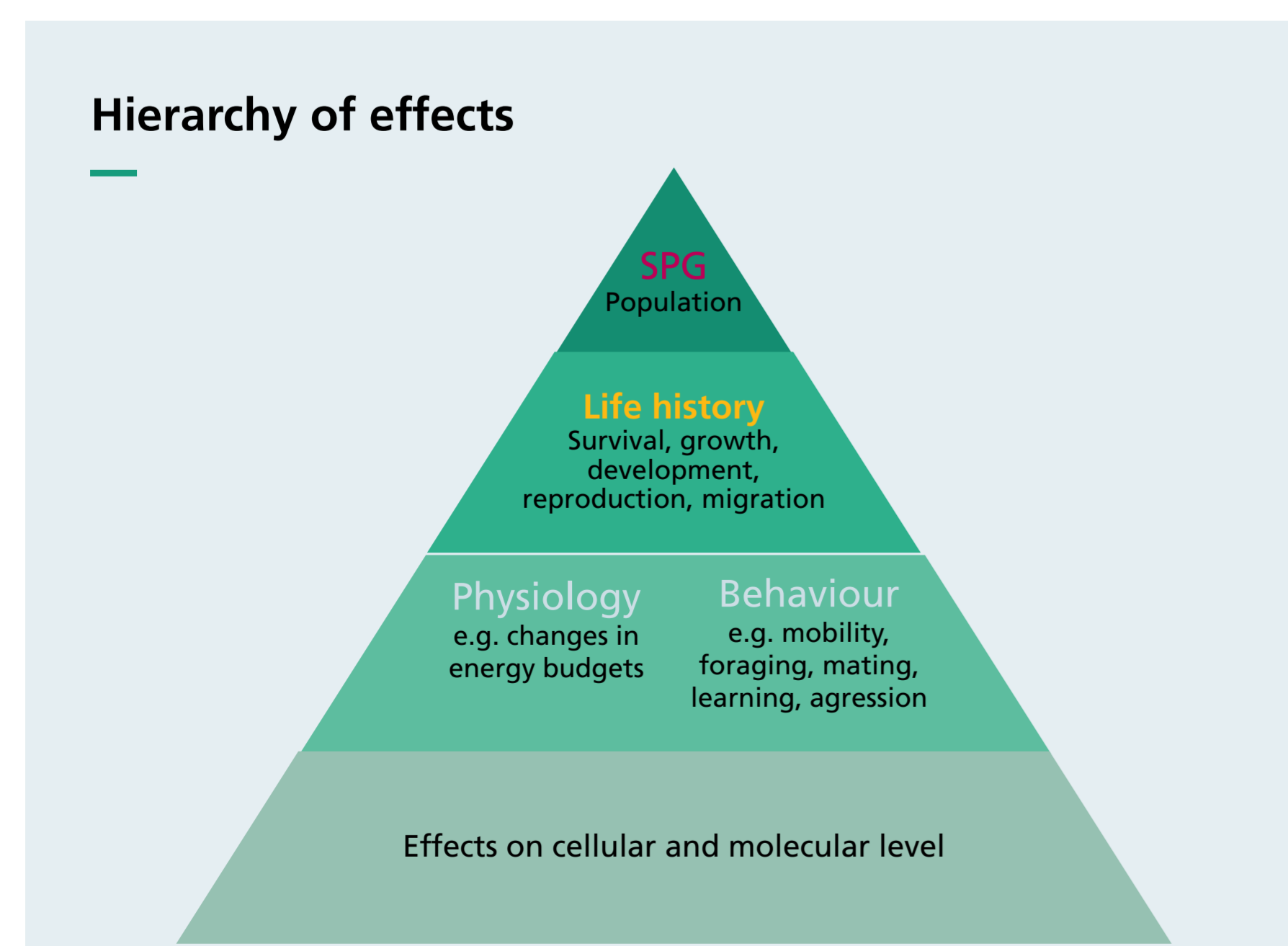
- Genetically Modified Plants (GMP's) can be designed to express toxins with biocidal properties. These can be e.g. proteins like Bt-proteins or RNA molecules to induce gene silencing.
- One aspect of the environmental risk assessment for GMPs is to assess the risk to non-target organisms (NTO's).
- The main route of exposure of NTO's to GMP toxins is uptake via the diet, either directly by feeding on living or dead plant material or indirectly via the food chain.
- The project aims to review available literature on the assessment of sublethal effects of GMP's on non-target invertebrates. The current practice for the risk assessment of plant protection products (PPP's) was used for comparison.

## What to protect?

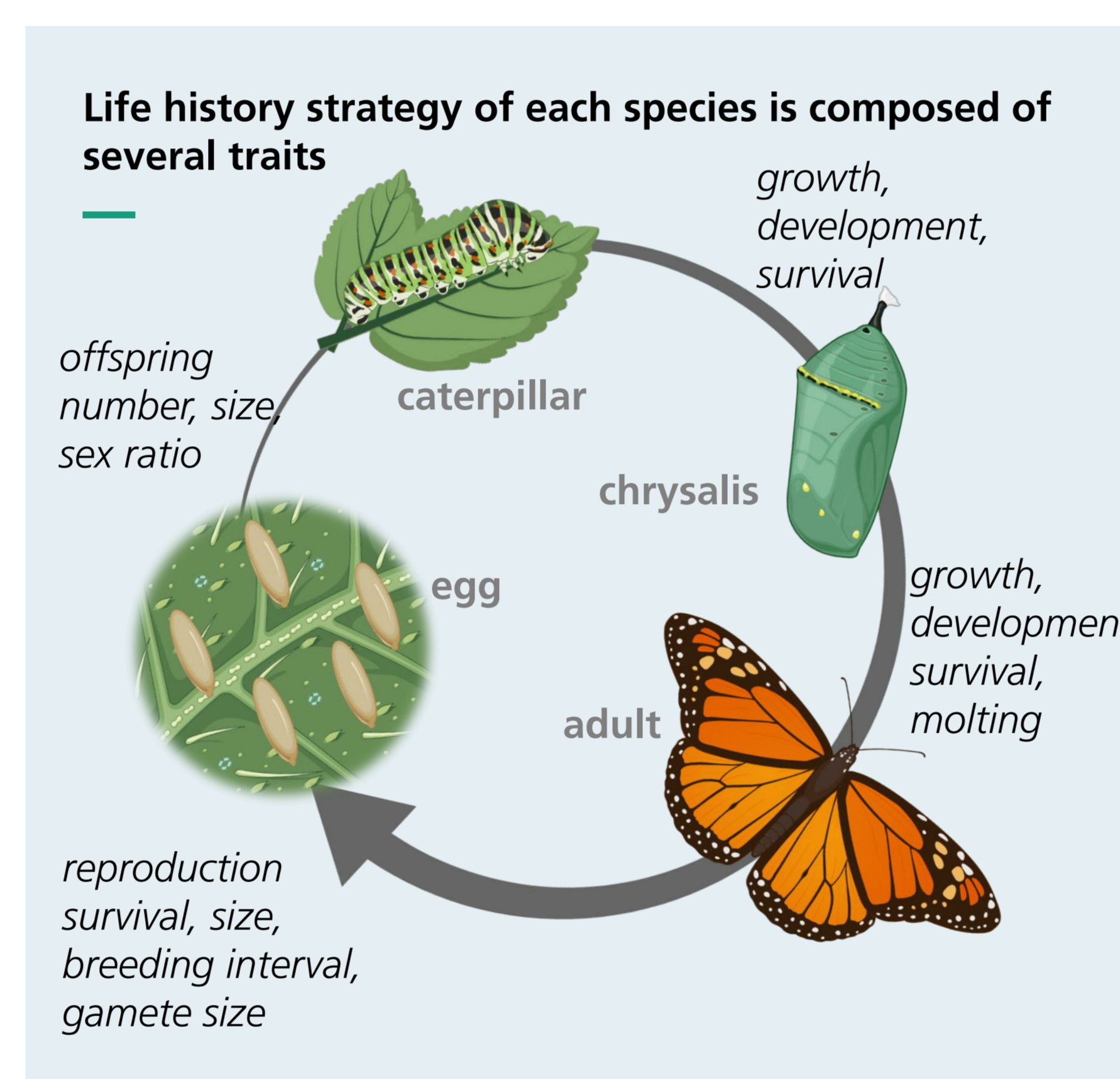
- Since the protection goals laid down in the EU Directive (EC) 2018/350 are relatively vague, the questions what should be protected where and when must be answered in the problem formulation for each risk assessment.
- In the EU, specific protection goals (SPGs) are actually derived using an **ecosystem services approach** (1).
- In most cases SPGs are defined on the level of the **population** and aim to protect the **abundance or biomass** of representative species providing ecosystem services.

## What drives population level effects?

- Dynamics of a population in a given area is driven by the number of births, deaths as well as migration. Birth and death rates result from the **life histories** of the individuals.
- Due to limiting resources an organism cannot optimize all its life history traits. Thus, there are **trade-offs**, e.g. between number and size of the offspring. The relevance of such trade-offs in assessing effects on toxins is not clear yet.



- Survival is directly linked to population abundance and assessed in many tests, while the assessment of sublethal effects in test protocols is much less established.
- The most relevant **sublethal effects** for assessing the risk on populations are those affecting growth, development and reproduction. Traits are e.g. size and age at first reproduction, interval between reproductive events, as well as number, size, fitness and sex ratio of offspring.
- These life history traits driving fecundity, result from effects on physiology or behaviour which can also affect survival and are consequences of effects at the cellular and molecular level.



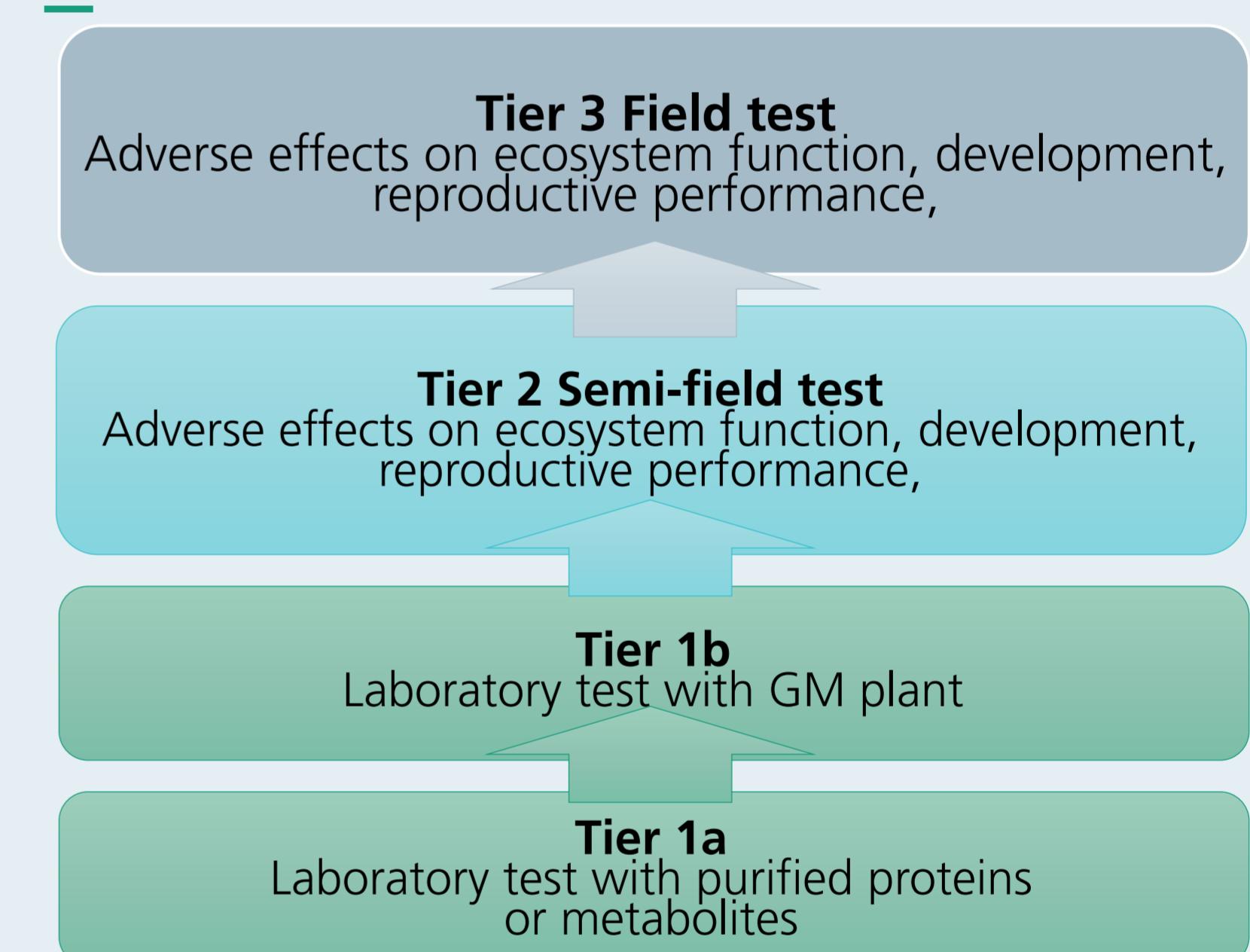
## How to measure sublethal effects?

- For PPP's, the legal data requirements include tests on earthworms, collembles, mites, wasps and bees. Guidance on additional species to be tested is available.
- There are no legal minimum data requirements regarding tests with NTO's for GMP's.
- Several existing laboratory test guidelines for PPPs include the assessment of sublethal effects. Sublethal effects are not yet routinely addressed in GMP risk assessment on NTO's.
- In most cases, the exposure situation in test guidelines is not representative of GMP's producing toxins. For examples, test guidelines for Non-Target Arthropods (2) focus on exposure via contact, while diet is the most relevant exposure route in the case of GMP toxins.
- PPP risk assessment for terrestrial invertebrates focusses on beneficial arthropods (predators and parasitoids as antagonists to pests as well as pollinators). For herbivorous species providing other ecosystem services such as cultural services and which may be exposed to GMP's (e.g. butterflies, grasshoppers) test guidelines are missing.

## How to assess sublethal effects?

- For PPP's, criteria on acceptability of effects or triggering higher tier tests (e.g. semi-field and field studies) are defined in Uniform Principles or guidance documents.
- In GMO risk assessment, only one general Guidance Document (3) and one Scientific Opinion on Non Target Organisms (4) is available. Criteria to decide on acceptability of effects or need for higher tier assessments can be defined by the applicant on a case by case basis.
- Population models are increasingly used in PPP risk assessment. The risk of Bt-Maize for butterflies has also been assessed based on modelling (5). However, this model considers only lethal effects on larvae but no sublethal effects.

## Tiered approach on the assessment of potential impacts of genetically modified plants on non-target organisms (modified from (4))



## Preliminary conclusions

- Specific protection goals** should be defined in a consistent and transparent way, independent of the type of the stressor (e.g. PPP or GMP).
- To facilitate comparable risk assessment of effects of GMP's on NTO's **more specific guidance** on what to test and how to test should be developed.
- Some established test guidelines need **modifications** to address the potential long-term exposure via the diet (GMP material or animals fed on GMP's).
- For some additional taxa, test guidelines would increase the overall quality and reliability of the results.
- Decision criteria** and triggers should be defined in guidance documents.
- The relevance of **sublethal effects** for the population should be addressed in higher tier testing or population modelling.
- However, there is potential to use tests on sublethal effects in order to **analyse life-history trade-offs**.

## References

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- Butterfly/caterpillar/egg/chrysalis image created with Biorender.com

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