# ECOTOXICOLOGY OF MULCH FILMS

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**Environmental** 



RNTHAA

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## BACKROUND

- Mulch films are applied worldwide due to their **positive effects** (e.g. humidity, temperature, weed growth limitation) on agricultural farming
- Commonly applied polymers are polyethylene (PE) and the bio-based poly lactate acid (PLA) with addition of multiple, often unknown additives
- Despite the environentally friendly seeming labelling of bio-based and biodegradable, PLA can be even more toxic than petroleum based plastics<sup>2</sup> and only degraded under very harsh conditions<sup>3</sup>
- Degradation and later accumulation of mulch films in soil is fascilitated by their low thickness due to rising removal difficulty and generally high disposal costs<sup>4</sup>
- Plastic mulching is a relevant source of microplastics (MP) in the terrestrial environment with up to 324.5 kg/ha<sup>5</sup> which lies in the same magnitude of earthworm mortality<sup>6</sup> Impact of accumulation in soil is yet unsufficiently determined and requires further investigation<sup>7</sup>

The main applications, greenhouse and mulch flm, account for almost 75% of the total market, with more than one million tons being produced for each.<sup>1</sup>

> Plasticulture market growth is **5% annually** depending on region and application.<sup>1</sup>



We aim to investigate the effects of mulch film microplastics and their constituents on the living environment with focus on soil organisms.

Furthermore, we want to investigate the influence of sorbed pesticides on the toxicity of mulch film fragments.



This work is part of the iMulch project funded by the European Fond for regional development (EFRE).

iMulch – An investigation of the influence of polymers on a terrestrial ecosystem using the example of mulch films used in agriculture

### OBJECTIVE

- Assessment of toxicity mechanisms of mulch film constituents and potential synergistic effects
- Determination of MP toxicity on representative soil organisms
- Determination of the interaction of mulch film fragments and selected pesticides
  - Influence of mulch films on pesticide toxicity

INTERNAL	METHODS	
Extraction	Extracts & pesticide screening	Microplastic in soil
500mg polymer cut into 1x2cm	<ul> <li>Extract testing</li> <li>In vitro screening for dioxin-like</li> </ul>	Reference soil 01-A to approximate to natural conditions
Worst case scenario	activity, genotoxicity and endocrine/androgene activity	Scenarios



- Availability of pesticides in plastic spiked soil
- Determination of the baseline toxicity of pesticide active substances and products

# Thiacloprid Krazine

Tebuconazole Quinoxyfen Comparison of petroleum- and biobased mulch films conderning biological effects

# WHERE DO WE GO FROM HERE?

# In vitro

- Establishment of beta-Naphtoflavone as micro EROD induction standard
- Investigation of sorption behaviour in microtiter plates for extracts and pesticides

## In vivo

- Comparison of effects of new and aged mulch film
- Deduction for further testing with *Eisenia fetida* in chronic exposure scenarios (OECD 222, in soil) and decision on biomarkers
- Comparison between sensitivity of *Eisenia fetida* and

Per solvent 2x30min UAE

Solvents PE: acetonitrile/dichlormethane Solvents PLA: acetonitrile / isopropanol

### **Realistic scenario**

Shaking extraction: 24h, Millipore Water, 20±2°C, 100rpm

- Solid phase extraction
- Solvent: ethyl acetate, methanol
- Analysis with GC-MS

- Application of glass-coated microplates to prohibit sorption
- Determination of acute toxicity to earthworm Eisenia fetida (OECD 207, filter paper test)

# Pesticide testing

- In vitro assays to narrow down substances
- Active substances and products in comparison



- Exposure monitoring (soil, earthworm)
- Acute toxicity on *E.fetida* (OECD 207)
- OECD 232: realistic and worst case MP concentrations on *F. candida* reproduction



## **PRELIMINARY RESULTS**



\*results should be interpreted as trends



**Fig. 1** EROD activity for 2,3,7,8-TCDD in plastic plates (blue, n=5) and glass plates, n=4)

In vitro pesticide screening: positive, negative, unclear 

	Endocrine activity	Genotoxicity	Dioxin-like activity
Atrazine	$\checkmark$	$\checkmark$	?
Tebuconazole	$\checkmark$	×	$\checkmark$
Quinoxyfen	$\checkmark$	?	X

2,3,7,8-TCDD not usable in assay with glass coated plates (Fig. 1)

#### Lumbricus terrestris



Other project partners are also presenting at the SETAC 2020: Check out poster 2.08P.11 for information on biodegradability and poster 4.14P.1 for more information on the iMulch project and first results.

# OECD 207: Filterpaper test with EC50 [mg/mL]

- Tebuconazole (49±2), solvent extracts (no toxicity), water extracts (no toxicity)
- OECD 207: Acute toxicity test with active substance and EC50 [mg/mL]
  - Folicur (Tebuconazole, 116±27), Biscaya (Thiacloprid, <50)</li>



#### REFERENCES

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