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„Microplastic contamination in soil: analytical methods and the role of organic fertilizer”

Martin Löder
Animal Ecology I, University of Bayreuth

NETmicroplastic - Stakeholder Workshop, 27. April 2023

MP research at Animal Ecology I

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Analysis of MP in environmental samples:

- Strong focus on method development
- Optimisation of sampling, sample extraction, purification
  - ➢ Large area filtration system, density separation, enzymatic purification
- Spectroscopic detection: µFTIR and Raman analysis
  - ➢ Automated analysis of large imaging data sets
- Matrices: Water, sediment, soil, biota, air

Biological effects:

- Experiments with different aquatic and terrestrial invertebrate species

MP research at Animal Ecology I

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More than 12 years of microplastic research in more than 30 projects on microplastics…

Bundesministerium für Bildung und Forschung

Landesanstalt für Umwelt, Messungen und Naturschutz Baden-Württemberg

DFG

Bundesministerium für Wirtschaft und Technologie

BWPLUS

Freistaat Thüringen

Landesamt für Umwelt, Bergbau und Naturschutz

Horizon2020
European Union Funding
for Research & Innovation

DLR

FONA
Forschung für Nachhaltige Entwicklung
BMBF

OBERFRANKEN STIFTUNG

Bayerisches Staatsministerium für Umwelt und Verbraucherschutz

DBU

HESSEN
Hessisches Landesamt für Umwelt und Geologie

Rheinland-Pfalz
LANDESAMT FÜR UMWELT

Landesamt für Natur, Umwelt und Verbraucherschutz Nordrhein-Westfalen

Studienstiftung des deutschen Volkes

MP research at Animal Ecology I – soil related projects

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international

papillons

Plastic in Agricultural Production:
Impacts, Lifecycles and LONg-term Sustainability

national

DFG SFB MIKROPLASTIK

MiKoBo

Microplastics in organic fertilizer and their impacts on agricultural soils

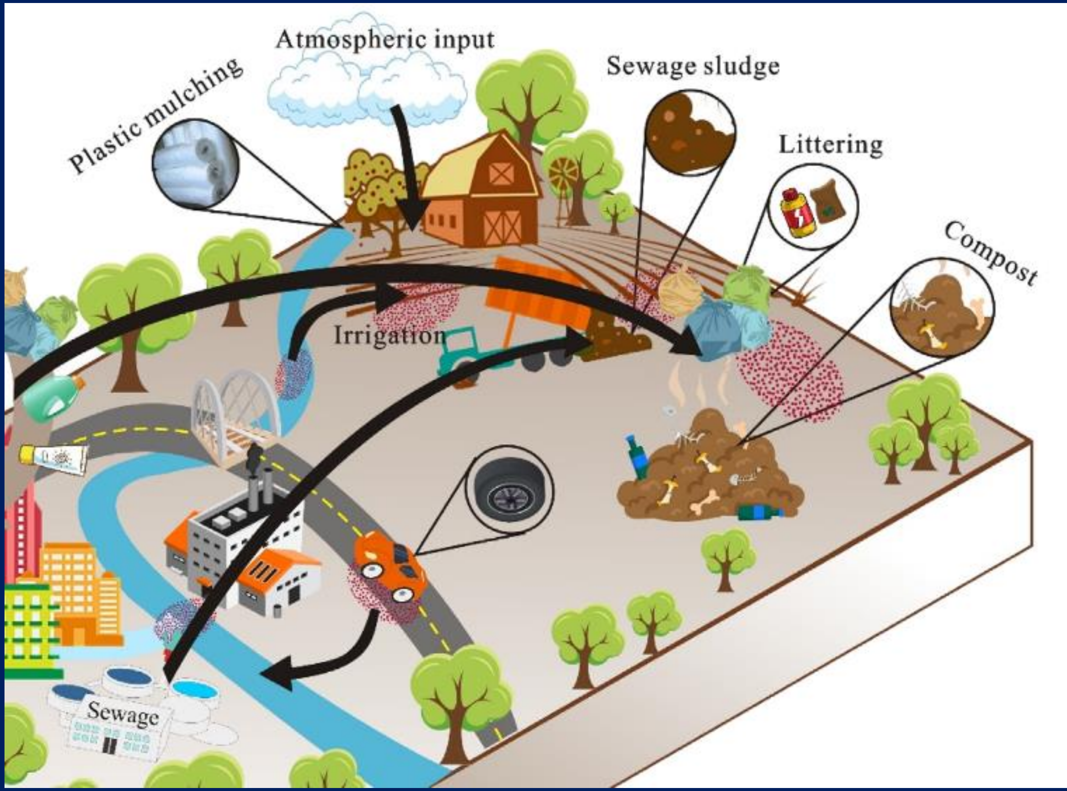
PLAWES

Microplastic contamination in the model system Weser - National Park Wadden Sea - a cross-ecosystem approach

BabbA

The fate of biodegradable plastic bags in industrial biowaste treatment

Microplastics in soils



Yang, L., Y. Zhang, S. Kang, Z. Wang and C. Wu (2021). "Microplastics in soil: A review on methods, occurrence, sources, and potential risk." Science of The Total Environment 780: 146546.

- Microplastics have potentially negative effects on: soil functions, plant growth, soil biota
- We need to know the level of contamination for risk assessment

Microplastics in soils

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ELSEVIER

Science of The Total Environment
Volume 880, 1 July 2023, 163294

Continents of plastics: An estimate of the stock of microplastics in agricultural soils

Mikaël Kedzierski, Delphine Cirederf-Boulant, Maialen Palazot, Marion Yvin, Stéphane Bruzaud

Highlights

- Worldwide, the stock of microplastics in agricultural soils could be 1.5 to 6.6 million tons.
- This is potentially more than the quantity of microplastics present on the surface of the oceans.
- The distribution of this stock between countries is probably not uniform.
- Improving data quality should be a priority to refine these estimates.

Comparability in reported units on microplastic is often lacking:

number: n	mass: mg

m^{-2} m^{-3} kg^{-1}

km^{-1}

$kg\ dw^{-1}$ l^{-1}

m^{-1}

km^{-2} $g\ ww^{-1}$

➢ **Standardisation/harmonisation of analytical techniques is urgently needed!**

Analysis of microplastics in soil samples

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- Visual identification in many early studies:

  … it looks like microplastic –
  so it must be microplastic!?…

- Reliable detection of microplastics is only possible with chemical analysis!

Analysis of microplastics in soil samples

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Detection methods for microplastics:

➢ **Thermoanalytical methods:**
- Mass of microplastics per polymer type
- Mass balances possible

➢ **Spectroscopic methods:**
- Amount of microplastics per polymer type
- Info on number, size, shape of particles

➢ **Methods are to be seen as complementary techniques!**

➢ **For toxicological investigations, data on polymer type, shape and size are required.**

⇨ **Spectroscopic methods like <u>FTIR</u> or Raman spectroscopy** ⇦

- Detection limit FTIR: ~10 µm
- Detection limit Raman: ~0.5 µm

Analysis of microplastics in soil samples

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Soil - one of the most complex matrices in microplastic analysis:

- Mineral fraction:
  sand, silt, clay
- Organic fraction:
  plant & animal material
- Microplastics

Analysis of microplastics in soil samples

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Soil - one of the most complex matrices in microplastic analysis:

- **Natural matrix largely dominates**
- **Microplastics: „needle in the haystack“**

- **Sample purification prior to analysis of microplastics is essential!**

Analysis of microplastics in soil samples

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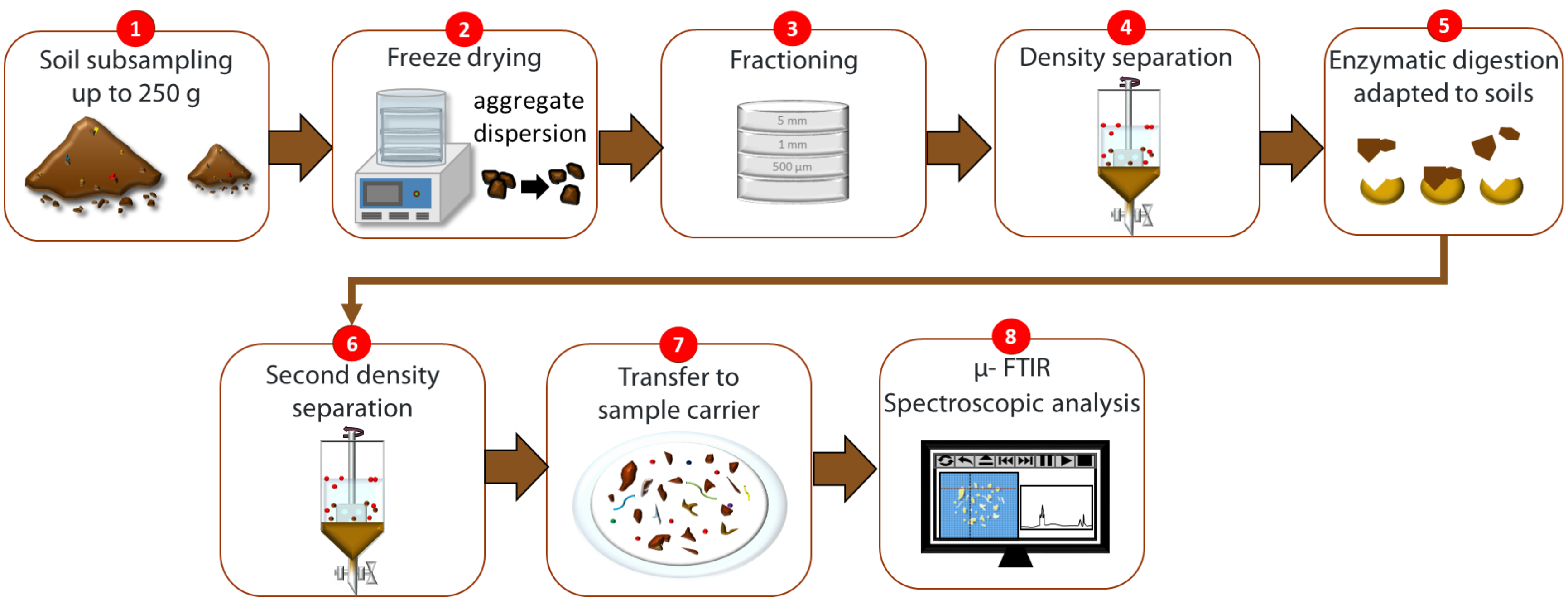
Protocol for MP purification and analysis of soil samples

Environmental Chemistry

Tackling the Challenge of Extracting Microplastics from Soils: A Protocol to Purify Soil Samples for Spectroscopic Analysis

Julia N. Möller,[a,*] Ingrid Heisel,[a] Anna Satzger,[a] Eva C. Vizsolyi,[a] S.D. Jakob Oster,[a] Seema Agarwal,[b] Christian Laforsch,[a,*] and Martin G.J. Löder[a,*]



Analysis of microplastics in soil samples

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Sample type	Soil sample	
Pre-treatment	Sample homogenisation, evtl. subsampling	
	Freeze drying	
	Size fractionation (500 µm sieve)	
	Particles 500 – 5000 µm	Particles 10 – 500 µm
Sample processing		
Visual sorting	x	
Density separation I		x
Enzymatic and oxidative digestion		x
Density separation II		x
Subsampling (if necessary)		x
Chemical identification		
ATR-FT-IR (particle count)	x	
FPA-µFT-IR (particle count)		x

Analysis of microplastics in soil samples

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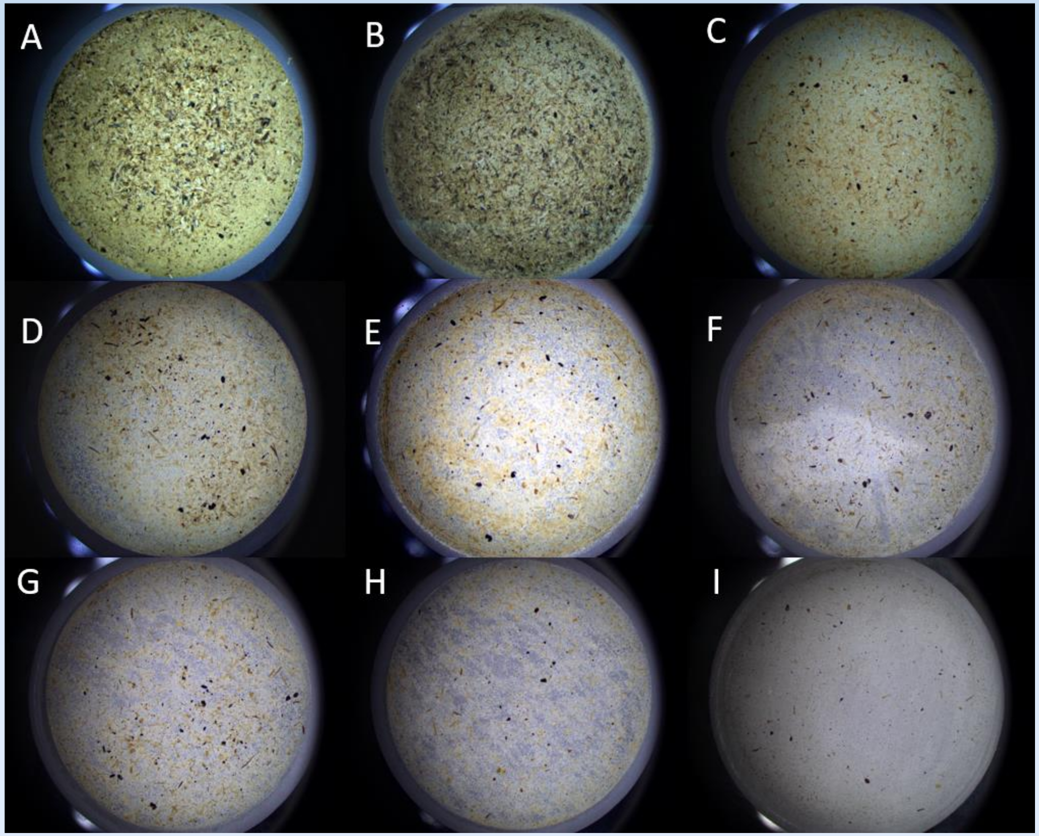
Protocol for purification of soil samples

- Density separation I: > 99 % matrix reduction, removal of mineral content
- Enzymatic-oxidative purification: Further removal of 73-85 % of organics



A	Sample after density separation I
B	48 h SDS
C	1 h Fenton's reagent
D	12 h Protease
E	48 h Pectinase
F	48 h Viscozyme
G	24h Cellulase
H	1 h Fenton's reagent
I	24h density separation II

Analysis of microplastics in soil samples

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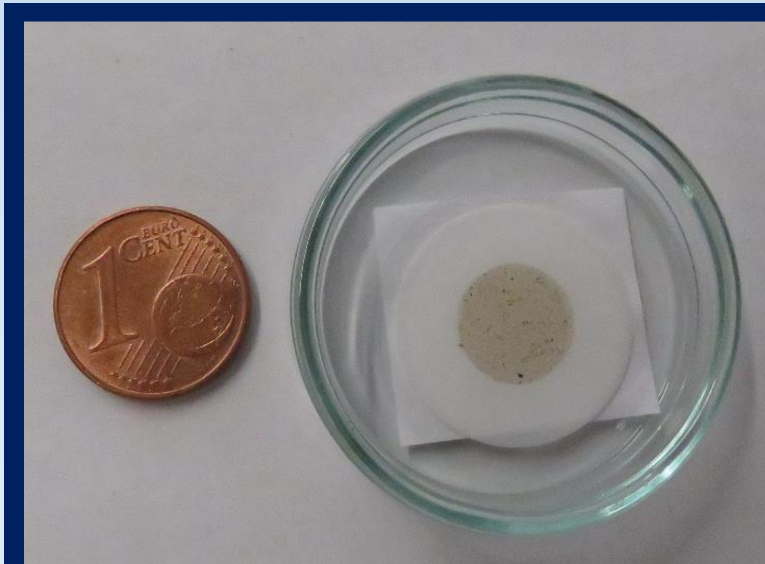
Protocol for purification of soil samples

- Highly efficient reduction by >> 99 % of initial sample weight

250 g Soil sample (TG)



Purified sample on filter for FTIR analysis

Analysis of microplastics in soil samples

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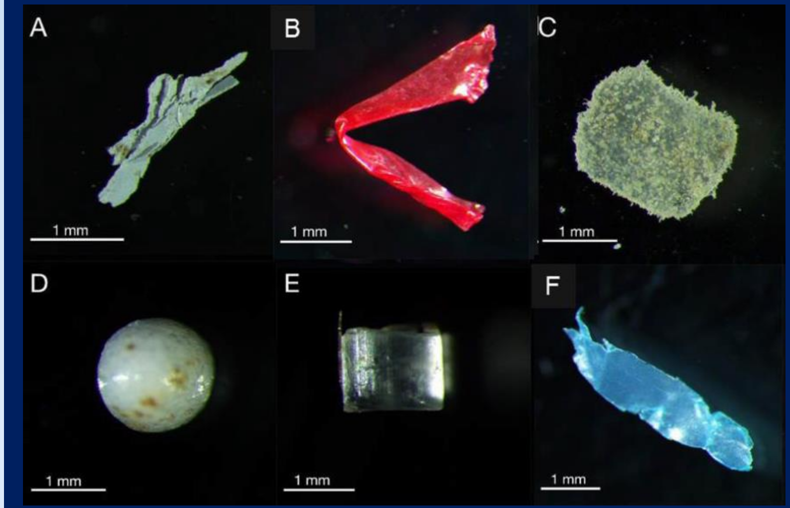
Two sample fractions:

> 500 µm particles

<500 µm particles



Sorted particles for single particle ATR-FTIR analysis

Purified sample on filter for FTIR analysis

Analysis of microplastics in soil samples

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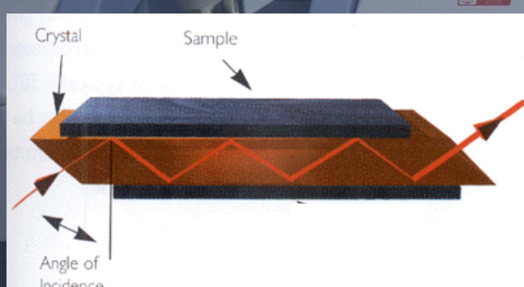
FTIR Methods

ATR unit:
(attenuated total reflection)

For larger particles

> 0,5 mm from manual sorting

Recording of single spectra



FTIR microscope:

For microscopic particles 10 – 500 µm on filters

(Pixel resolution ca. 5 µm)

Transmission
(Reflection)
(Micro-ATR)

Analysis of microplastics in soil samples

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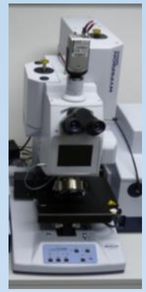
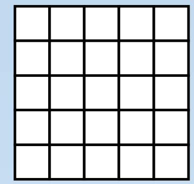
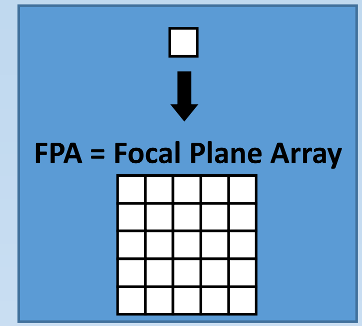
IR microscope



+

FPA detector





IR microscope: measurement of particles > 10 µm

FPA detector: simultaneous recording of thousands of IR spectra in a single measurement

Sucessive measurement of the whole sample filter in less than 2,5 hours

Chemical Imaging via µFTIR spectroscopy:
Combination of spatial and chemical information with high resolution on whole sample filters possible

Analysis of microplastics in soil samples

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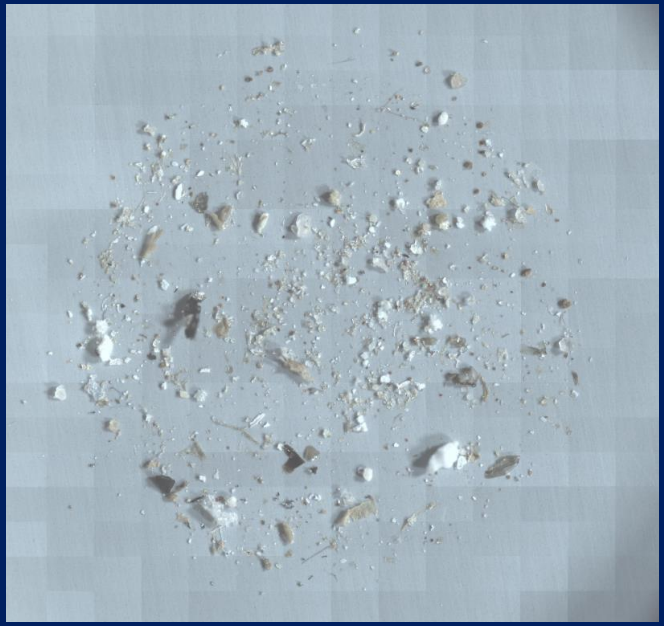
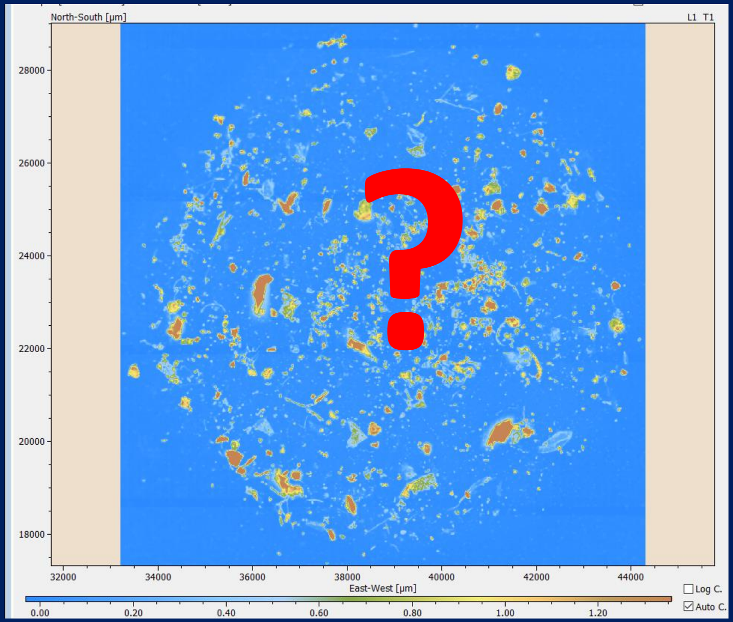
Sample filter



Purified sample on filter for µFTIR measurement

FTIR imaging measurement

Chemical Image



Spectral data of sample:

> 4,5 Million IR-spectra

manual analysis impossible

Analysis of microplastics in soil samples

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EPINA IMAGELAB

Automated analysis - Bayreuth Particle Finder

- Cooperation with TU Vienna, Prof. Lohninger
- Development of an automated analysis tool for microplastics in environmental samples
- Basic software ImageLab
- Plastic polymer identification using Random Forest classifiers

Target:

- 22 most common plastic types
- Fast classification of large measurement files
- Automatic size measurement of particles

➢ Bayreuth Particle Finder - successful

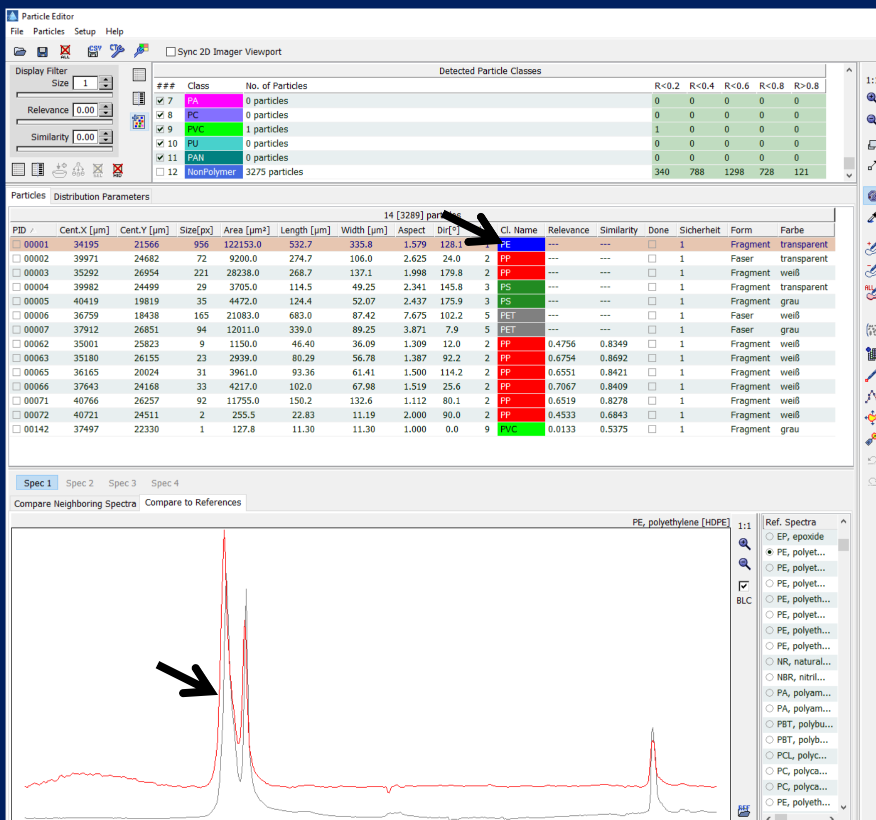
Purency

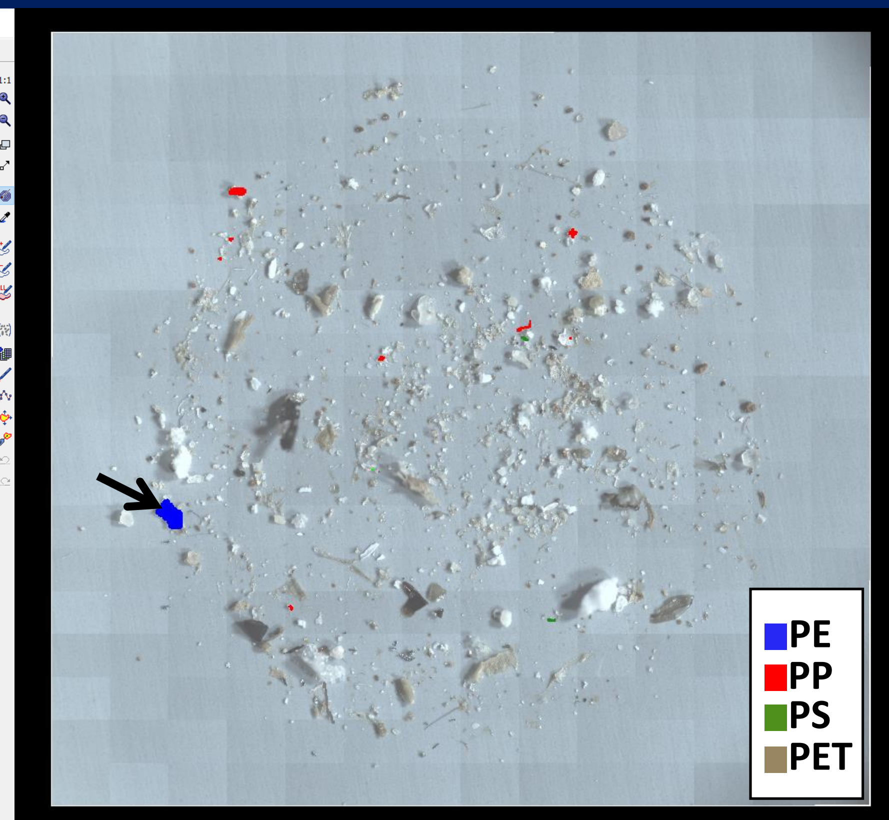
➢ Currently improvement with PURENCY GmbH

➢ Purency Microplastics Finder

Analysis of microplastics in soil samples

Example BPF – result:





Analysis of microplastics in soil samples

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Focal plane array detector-based micro-Fourier-transform infrared imaging for the analysis of microplastics in environmental samples

Martin Günter Joachim Löder [A B C], Mirco Kuczera [A], Svenja Mintenig [A], Claudia Lorenz [A] and Gunnar Gerdts [A]

Environmental Science & Technology — Article

Cite This: *Environ. Sci. Technol.* XXXX, XXX, XXX−XXX — pubs.acs.org/est

Enzymatic Purification of Microplastics in Environmental Samples

Martin G. J. Löder,[*,†,§] Hannes K. Imhof,[‡] Maike Ladehoff,[†,∥] Lena A. Löschel,[‡] Claudia Lorenz,[†] Svenja Mintenig,[†,⊥] Sarah Piehl,[‡] Sebastian Primpke,[†] Isabella Schrank,[‡] Christian Laforsch,[*,‡] and Gunnar Gerdts[*,†]

Analytical Methods — Royal Society of Chemistry

PAPER — View Article Online — View Journal | View Issue

Check for updates

Cite this: *Anal. Methods*, 2019, 11, 2277

A methodology for the fast identification and monitoring of microplastics in environmental samples using random decision forest classifiers†‡

Benedikt Hufnagl,[*a] Dieter Steiner,[a] Elisabeth Renner,[a] Martin G. J. Löder,[b] Christian Laforsch[b] and Hans Lohninger[a]

Environmental Chemistry

Tackling the Challenge of Extracting Microplastics from Soils: A Protocol to Purify Soil Samples for Spectroscopic Analysis

Julia N. Möller,[a,*] Ingrid Heisel,[a] Anna Satzger,[a] Eva C. Vizsolyi,[a] S.D. Jakob Oster,[a] Seema Agarwal,[b] Christian Laforsch,[a,*] and Martin G.J. Löder[a,*]

FTIR measurement:

µFTIR Imaging of whole sample filters

Sample purification:

Plastic-friendly enzymatic process suitable for simple and difficult matrices

Data analysis:

Fast automated analysis of imaging data using RDF classifiers

Purification/analysis of MP in soil samples:

Combination of the previous publications

The role of organic fertilizer: farmyard manure?

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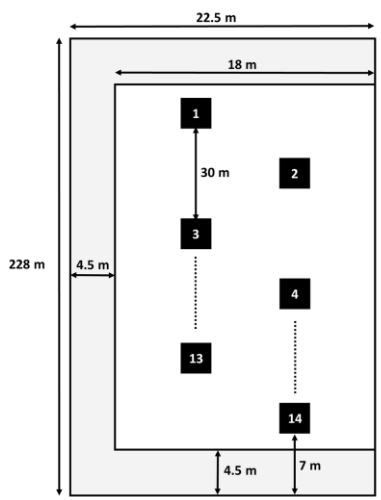
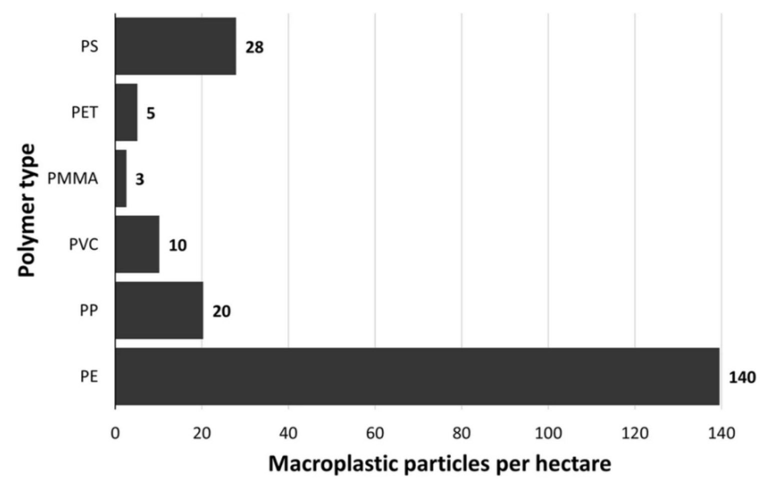
Identification and quantification of macro- and microplastics on an agricultural farmland

Sarah Piehl, Anna Leibner, Martin G. J. Löder, Rachid Dris, Christina Bogner & Christian Laforsch

Scientific Reports **8**, Article number: 17950 (2018) | Cite this article





"…plastic materials are omnipresent at farms, and unintentional breakdown of materials and spreading may introduce plastic debris into farmyard manures…"

The role of organic fertilizer: farmyard manure?

UNIVERSITÄT BAYREUTH

Identification and quantification of macro- and microplastics on an agricultural farmland

Sarah Piehl, Anna Leibner, Martin G. J. Löder, Rachid Dris, Christina Bogner & Christian Laforsch

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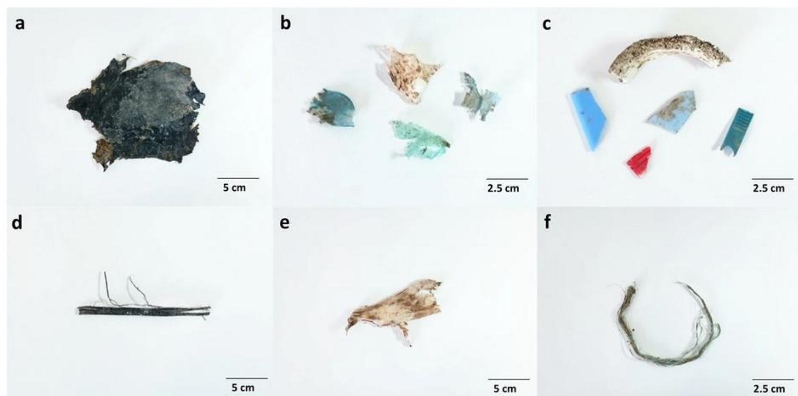

Exemplary pictures of macroplastic debris detected on the investigated agricultural farmland. Particles were grouped into three different shape categories: films (**a**,**b**), fragments (**c**), and others (**d**–**f**).

- ➢ **Estimations of macroplastics (> 5 mm):**
  276 to 510 pieces per hectare in the upper 5 cm
- ➢ **Estimations of microplastics (1 to 5 mm):**
  158,100 to 292,400 particles per hectare in the upper 5 cm

The role of organic fertilizer: compost/digestate?

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SCIENCE ADVANCES | RESEARCH ARTICLE

ENVIRONMENTAL STUDIES

Organic fertilizer as a vehicle for the entry of microplastic into the environment

Nicolas Weithmann,[1] Julia N. Möller,[2] Martin G. J. Löder,[2] Sarah Piehl,[2] Christian Laforsch,[2]* Ruth Freitag[1]

Table 1. Overview of plants and compartments. The total number of particles is shown as particles >1 mm per kilogram of dry weight.

	Plant A		Plant B				Plant C	Plant D	Plants E to N
Type	Biowaste composting		Biowaste digestion				Energycrop digestion	Biowaste digestion	Agricultural digestion
Sampled	CP 8 mm	CP 15 mm	Digest A	Digest B	Digest C	Digest D	End-of-process	Commercial binding	End-of-process
Particles per kilogram	20	24	70	122	146	14	0	895	0 to 11

- Number of microplastics in the final fertilizer strongly depends on the original input
- Highest contamination when biowaste from commerce/markets is processed

The role of organic fertilizer: compost/digestate?

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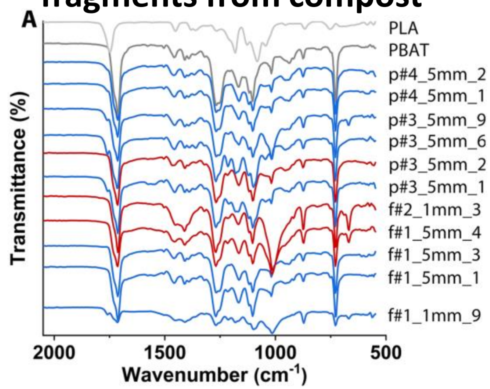
Municipal biowaste treatment plants contribute to the contamination of the environment with residues of biodegradable plastics with putative higher persistence potential

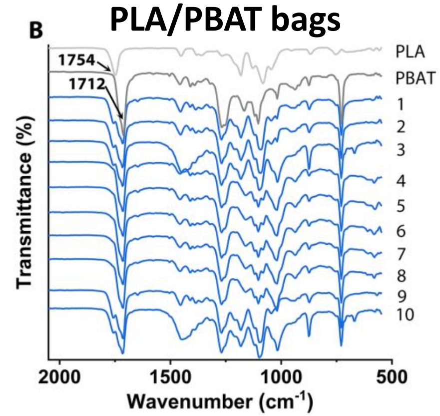
Thomas Steiner, Yuanhu Zhang, Julia N. Möller, Seema Agarwal, Martin G. J. Löder, Andreas Greiner, Christian Laforsch & Ruth Freitag

Scientific Reports **12**, Article number: 9021 (2022) | Cite this article

IR Spectra of PLA/PBAT fragments from compost



IR Spectra of commercial PLA/PBAT bags



- ➢ Conditions in the investigated biowaste treatment plants do not always ensure complete biodegradation of PLA/PBAT
- ➢ More research needed in cooperation with plastic manufacturers & plant operators

The role of organic fertilizer: sewage sludge?

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Eine Initiative des Bundesministeriums für Bildung und Forschung
Plastik in der Umwelt
Quellen • Senken • Lösungsansätze
GEFÖRDERT VOM
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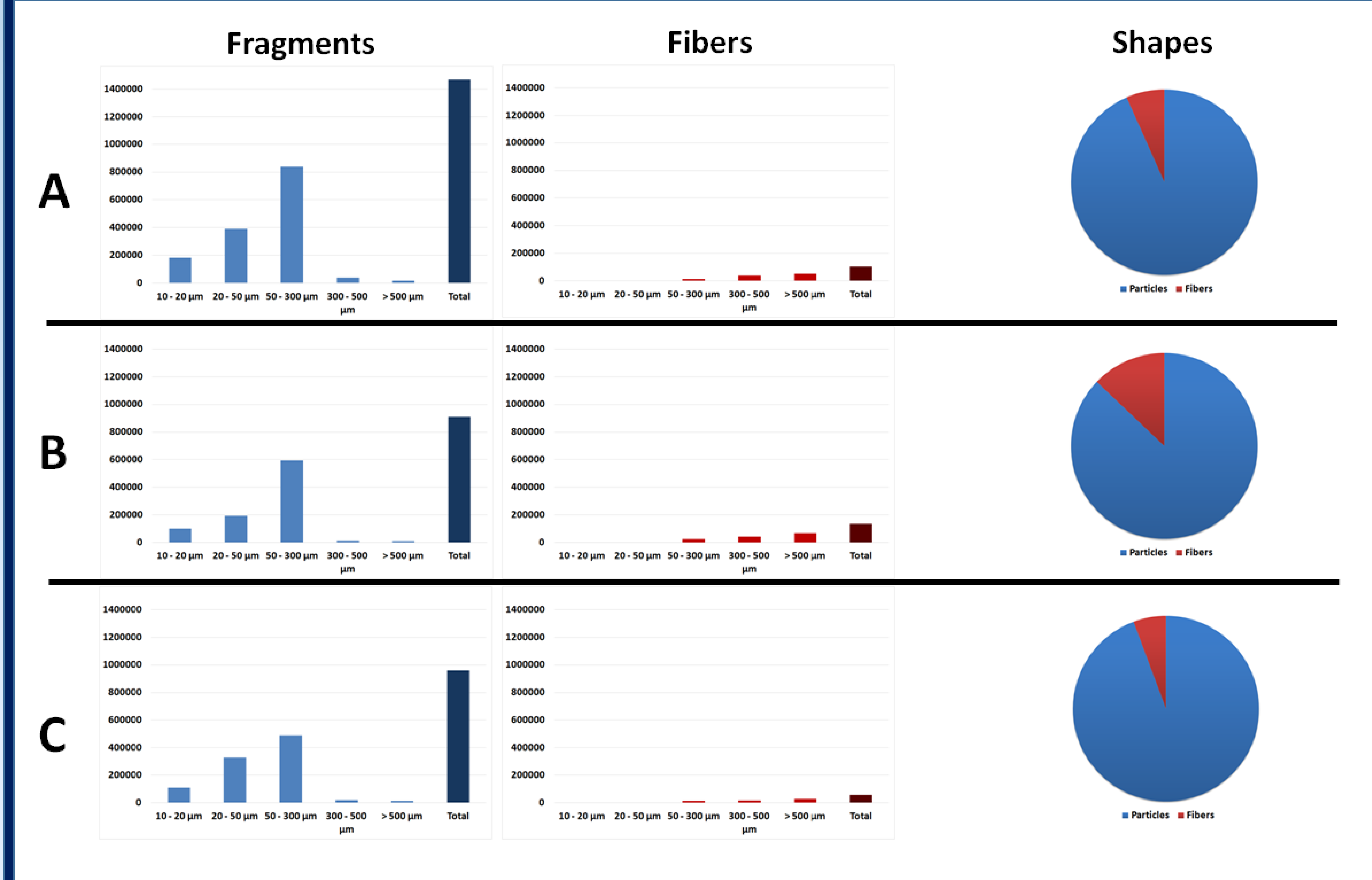
PLAWES

Analysis of sewage sludge samples as microplastic source:

- **Sewage sludge from 2018**
- **Used at LUFA Speyer (Landwirtschaftliche Untersuchungs- und Forschungsanstalt) in fertilization experiments**
- **Analysis of 3 subsamples with 10 g each**
- **Methodology as in Möller et al. 2021**

The role of organic fertilizer: sewage sludge?



The role of organic fertilizer: sewage sludge?

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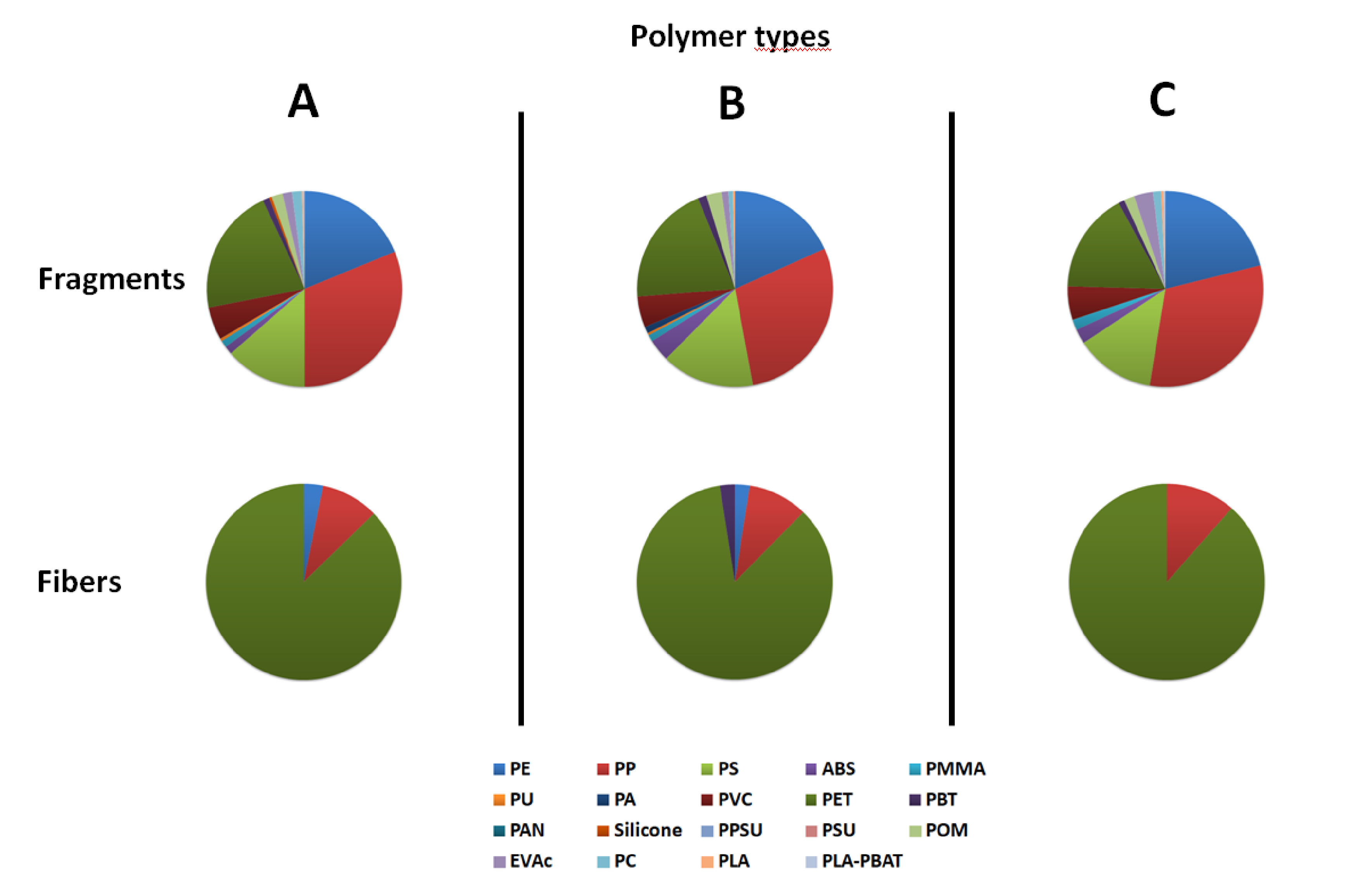

The role of organic fertilizer: sewage sludge?

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Plastik in der Umwelt

Quellen • Senken • Lösungsansätze

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Bundesministerium für Bildung und Forschung

PLAWES

Analysis of sewage sludge samples as microplastic source:

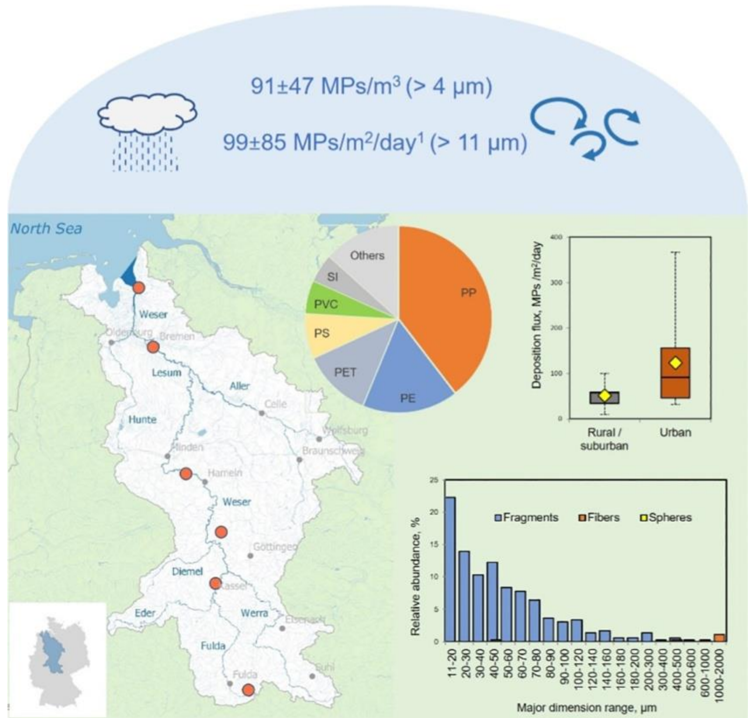
- Results of replicates similar: methodology suitable
- High contamination with microplastics:
  - Fragments: 1.112.426 ± 308.944 per kg dry weight
  - Fibers: 98.892 ± 38.650 per kg dry weight
- Diverse polymer composition represents our daily life with plastics

The role of organic fertilizer vs. atmospheric deposition

Science of The Total Environment
Available online 20 November 2021, 151812
In Press, Corrected Proof

Airborne microplastic concentrations and deposition across the Weser River catchment

Sarmite Kernchen [a], Martin G.J. Löder [b], Franziska Fischer [c], Dieter Fischer [c], Sonya R. Moses [b], Christoph Georgi [d], Anke C. Nölscher [a], Andreas Held [d], Christian Laforsch [b]



The role of organic fertilizer vs. atmospheric deposition

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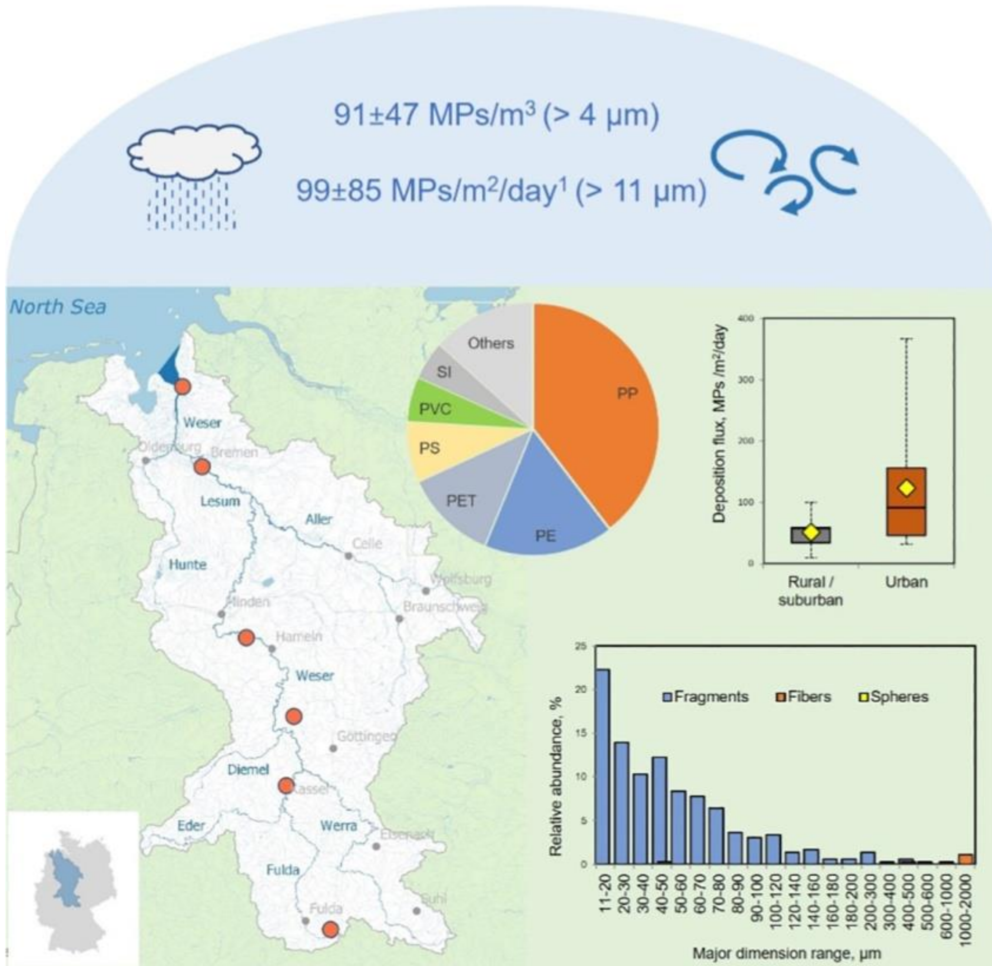

"…an estimated 232 metric tons of plastic being deposited in the Weser River catchment annually…"

Summary

MP analysis in soils:

- Chemical detection methods enable reliable analyses
- Thermoanalytical and spectroscopic detection methods are complementary
- Data on polymer type, shape and size in environmental samples are required for toxicological investigations
- Spectroscopic methods allow the detection of microplastics down to a size of approx. 10 µm with FTIR and below 1 µm with Raman
- Detection of nanoplastics in environmental samples is still a long way off

Summary

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MP contamination of organic fertilizers:

- ➢ MPs are ubiquitously present, thus also manure is a potential source
- ➢ The MP contamination of composts/digestates depends on the input material
  - citizens should collect biowaste plastic-free
  - commercial biowaste should be unpacked prior to treatment
- ➢ Sewage sludge is a massive source for MPs, usage should be restricted

Generally: as a precautionary principle MP input into the environment should be avoided where possible to prevent potential negative impacts!

Many thanks to…

- Funding agencies
- Team Animal Ecology I
- you for your attention!