Enantioselective Degradation of Fungicides in Soils: Chiral Preference Changes with Soil pH

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Enantio-/Stereoselective Aspects in Plant Protection Chemistry

- > 50% of plant protection product actives are chiral

- Enantiomers/stereoisomers may differ in:
  - efficacy
  - biodegradation
  - toxicity/ecotoxicity
"Chiral Switch": Substitution of Racemic by Single-Isomer/Enantio-Enriched Actives

- lower application rates at similar efficacy
- lower residues in crops and in the environment
- less side effects on non-target organisms
"Chiral Switch": Substitution of Racemic by Single-Isomer/Enantio-Enriched Actives

- lower application rates at similar efficacy
- lower residues in crops and in the environment
- less side effects on non-target organisms
- higher costs for synthesis
- additional studies for registration
- lower costs with respect to formulants, storage, transport
- (new patent)
Residues of Chiral Compounds in the Environment

Residues often show different enantiomer-/stereoisomer composition e.g., mecoprop residues in water compartments

![Graph showing enantiomer fraction (EF) vs. number]

R > S

R < S

data from Hegeman
Factors Determining Enantiomer-/Stereoisomer Composition of Residues

- Composition of sources
  - *mecoprop* as herbicide in agriculture: racemic or enriched R-enantiomer
  - *mecoprop* as additive to bituminous membranes in roof sealing: racemic ester
Factors Determining Enantiomer-/Stereoisomer Composition of Residues

- Composition of sources
  - mecoprop as herbicide in agriculture: racemic or enriched R-enantiomer
  - mecoprop as additive to bituminous membranes in roof sealing: racemic ester

- Enantio-/stereoselective biodegradation in soils and natural waters
  - mecoprop: preferential degradation of both enantiomers observed

- Isomerization/enantiomerization
  - mecoprop: slow enantiomerization
Enantio-/Stereoselective Degradation in Soils: Data Gaps

- Limited understanding of the factors governing enantio-/stereoselectivity
- Usually studied with very few soils → extrapolation to other soils is questionable
- Configurational stability (isomerization, enantiomerization) → may be important with respect to efficacy and side effects
Fungicides Selected for Incubation Studies

Metalaxyl

Epoxiconazole

Cyproconazole
Definitions and Kinetic Aspects

\[ \ln[c] = \ln[c]_{t=0} - k \cdot t \]

Enantiomer Ratio \( ER = \frac{[R]}{[S]} \)

\[ \ln[ER] = \ln[ER]_{t=0} - (k_R - k_S) \cdot t \]

Enantioselectivity \( ES = \frac{k_R - k_S}{k_R + k_S} \)
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Stereochemistry of Metalaxyl

![R-metalaxyl](image1)

R-metalaxyl
(fungicidally active)

![S-metalaxyl](image2)

S-metalaxyl
(fungicidally inactive)
Primary Degradation of Metalaxyl
Incubation of (rac)-Metalaxyl in a Calcic Cambisol

pH (CaCl₂) 7.1
C_{org} 1.6 %
loam

Buerge et al ES&T, 2003
Incubation of (rac)-Metalaxyl in a Calcic Cambisol

pH (CaCl$_2$) 7.1
C$_{org}$ 1.6 %

TBDM-γ-CD

Buerge et al ES&T, 2003
Ferric Podzol

"reversed" enantioselectivity

pH (CaCl₂) 3.4
C₉₉₉ 6.3 %
sandy loam
Ferric Podzol

"reversed" enantioselectivity

Calcic Cambisol

pH (CaCl₂) 3.4
Cₗ₀ 6.3 %
sandy loam

pH (CaCl₂) 7.1
Cₗ₀ 1.6 %
loam

Buerge et al ES&T, 2003
Incubation of Metalaxyl in an Orthic Podzol

\[ ER = \frac{[R]}{[S]} \]

- \( A_h \)-horizon, \( ES = -0.62 \)
- \( E \)-horizon, \( ES = 0.07 \)
- \( B_s \)-horizon, \( ES = 0.25 \)

Buerge et al ES&T, 2003
Incubation of Metalaxyl in an Orthic Podzol

\[ \text{pH (CaCl}_2\text{)} \]

3.0
3.5
5.2

Buerge et al ES&T, 2003
Addition of Acid or Base to a Luvisol

\[ ER = \frac{[R]}{[S]} \]

Native Soil, ES = 0.03

Incubation Time [d]

Buerge et al ES&T, 2003
Addition of Acid or Base to a Luvisol

$ER = \frac{[R]}{[S]}$

- **H$_2$SO$_4$-treated, ES = -0.09**
- **Native Soil, ES = 0.03**
- **CaCO$_3$-treated, ES = 0.18**

Incubation Time [d] vs. Ret. Time [min]
Metalaxyl: Correlation between ES and pH

Buege et al ES&T, 2003
No Correlation between $k_R$ or $k_S$ and pH
Stereochemistry of Epoxiconazole
Stereochemistry of Epoxiconazole

Enantiomers

Epimers

Epimers

Enantiomers
cis-Epoxiconazole: Correlation between ES and pH

\[ y = -0.10 \times + 0.37 \]

\[ R^2 = 0.95 \]

\[ k(+) > k(-) \]

\[ k(+) < k(-) \]

Buerge et al ES&T, in prep
Stereochemistry of Cyproconazole
Stereochemistry of Cyproconazole
Cyproconazole: No/Weak
Correlations between ES and pH

$y = -0.02x + 0.14$
$R^2 = 0.03$

$y = -0.03x + 0.12$
$R^2 = 0.15$

Buerge et al. ES&T, in prep
Stereochemistry of Cyproconazole
Cyproconazole: Correlations between Epimer-Selectivity and pH

$y = -0.18x + 1.02$
$R^2 = 0.76$

$y = -0.13x + 0.80$
$R^2 = 0.75$

$y = -0.15x + 0.88$
$R^2 = 0.61$

$y = 0.10x - 0.68$
$R^2 = 0.60$

Buerge et al ES&T, in prep
Dichlorprop and Mecoprop: Correlation between ES and pH

\[ ES = \frac{k_R - k_S}{k_R + k_S} \]

Müller and Buser, 1995
Romero et al, 2001
Schneiderheinze et al, 1999

\[ y = 0.32 x - 2.28 \]
\[ R^2 = 0.95 \]

Müller and Buser, 1995
Romero et al, 2001
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\[ y = 0.36 x - 2.61 \]
\[ R^2 = 0.98 \]
### Observed Correlations between ES and Soil pH

<table>
<thead>
<tr>
<th>Compound</th>
<th>pH (ES = 0)</th>
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<td>cis-Epoxiconazole</td>
<td>3.7 ± 2.1</td>
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<td>Metalaxyl</td>
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Assumption: linear correlation (not strictly correct)
Errors indicate 95 % confidence intervals
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<tr>
<td>Metalaxyl-Acid</td>
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<td>Ethofumesate</td>
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Hypotheses for Observed pH Dependence

- Different microorganisms or microbial communities
- One microorganism/two distinct enzymes
- One microorganism/one enantioselective enzyme
- Enantioselectivity on level of
  - degradation
  - uptake
- ...

Conclusions

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• degradation of metalaxyl, cis-epoxiconazole, cyproconazole is stereo-/enantioselective
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- correlations between enantioselectivity and soil pH:
  - metalaxyl, cis-epoxiconazole, dichlorprop, mecoprop: yes
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  - cyproconazole, metalaxyl-acid, ethofumesate: no
- soil pH is important in understanding enantioselective degradation
- next steps: to (sub) cellular scale (microorganisms, enzymes)
Acknowledgments

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