Measurement of human-dispensed pharmaceuticals in complex matrices, like digestates and blackwater

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EWA 18th International Symposium
IFAT 2016, Munich, Germany 31.05.2016 – 01.06.2016
Contents

- Blackwater and Digestates in innovative sanitation systems
  - Background
  - characterization
  - pharmaceutical pollution
- Description of matrix, challenges for process controlling
  - Expectancy values
  - Normative substances for standardization
  - Distracting, complex molecules
- Developed procedure for process validation using GC-MS instead of LC-MS/MS
  - Procedure for sample preparation
  - Advantages of GC- MS comparing to LC-MS/MS
- Lessons learned and future applications of the new method
  - Cost reduced, pharmaceutical measurement in mostly ALL liquid matrices
  - Scientific usage and economic relevance
  - Continuous development for reliable results in sludge and solids
The background of the project:
- big networking project with different scientific and practical members (called KREIS)
- the main aim: "Linking sustainable energy generation to urban wastewater"
- closed loop for wastewater

Main focus and working task of the Chair in the whole project:
- separation of municipal wastewater in different technical parts for optimizing wastewater treatment (BW, GW, RW, PW)
- specified degradation of pharmaceuticals and pharmaceutical residues in wastewater
 Reasons for this experimental investigation

Separation of municipal wastewater in the different technical compounds...

blackwater  greywater  rainwater

The HAMBURG WATER Cycle ®

Greenery waste

biowaste

strategy: separation of the different technical compounds for a specified treatment application
Main aims of the research activities

- 4 targets of blackwater treatment including recovery of energy from organic residues
  - Pharmaceutical degradation
  - Approving treatment and biogas production stability of anaerobic treatment plants with a decentralized focus
  - Organic to liquid conversion
  - Are they combinable?
  - Are they conflicted?

- 2 challenges involved
  - Increasing of treatment efficiency by increasing digester gas production
  - Degradation of pharmaceuticals for decreasing of environmental pollution using anaerobic treatment conditions (CSTR + UASB)
  - Are these targets disputed?

Providing a comprehensive study using the CSTR and UASB technique with a variation of different reactor loadings by hydrolysis-substrate feeding
The problem with pharmaceuticals in wastewater and the environment

- human-related intruding of pharmaceuticals and pharmaceutical residues in different environmental compartments (water and soil)
- antidromic effect; increasing of analytical quality and increasing of pharmaceutical consumption → detection of pharmaceuticals and residues in every environmental area (groundwater, surface water, soil etc.)
- insufficient long-time- and big-scale- studies about human- and environmental- toxic effects and result of re-formation and interaction between different substances („World´s experiment“) Sattelberger, 1999
- human-depending loop of pharmaceuticals and residues → direct and indirect intruding into soil and water vs. direct and indirect recovery of freshwater, producing of animals and food plants
- Results: antibiotic resistance, infertility, hormonal balance
**Constructed pilot plant - input material and parameters/blackwater**

**Sampling and analytical procedure:**
- 1m³ of blackwater was sampled from Lübeck- Flintenbreite once a month
- stored at 4°C in climate chambers
- directly homogenized before feed to the different duplicates
- weekly analyzing of dry and organic matter
- physic- chemical analyzing twice a month
- pharmaceutical analyzing twice a month
- hygienic analyzing once a month

<table>
<thead>
<tr>
<th></th>
<th>Diclofenac</th>
<th>Ibuprofen</th>
<th>Metformin</th>
<th>Metoprolol</th>
<th>Amoxicillin</th>
<th>Carbamazepine</th>
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</thead>
<tbody>
<tr>
<td><strong>85%-Percentile</strong> [µg/l]</td>
<td>28,45</td>
<td></td>
<td></td>
<td>47,75</td>
<td></td>
<td>141,5</td>
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<tr>
<td><strong>Value Number [n]</strong></td>
<td>20</td>
<td>20</td>
<td>20</td>
<td>20</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td><strong>Sampling Duration [d]</strong></td>
<td>370</td>
<td>370</td>
<td>370</td>
<td>370</td>
<td>370</td>
<td>370</td>
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<thead>
<tr>
<th></th>
<th>acetic acid</th>
<th>propionic acid</th>
<th>TP</th>
<th>DOC</th>
<th>DIC</th>
<th>TOC</th>
<th>TN dissolved</th>
<th>COD</th>
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<tbody>
<tr>
<td><strong>85%-Percentile</strong></td>
<td>690</td>
<td>295,4</td>
<td>169,9</td>
<td>1</td>
<td></td>
<td>1,4</td>
<td></td>
<td></td>
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<tr>
<td><strong>Value Number [n]</strong></td>
<td>11</td>
<td>10</td>
<td>8</td>
<td>11</td>
<td>11</td>
<td>7</td>
<td>7</td>
<td>11</td>
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Main data of the pilot plant for the anaerobic treatment of separated blackwater

- projection period: 6 months
- construction period:
  - CST- reactors: 3 months
  - UASB- reactors: 6 months
- treatment capacity for blackwater:
  - low- load conditions ca. 20l/d
  - high- load conditions ca. 76l/d
- used co- substrates for variation of reactor loading:
  - biowaste, hydrolyzed organic materials (greenery, kitchen waste, biowaste), fat separator deposits
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</tr>
</thead>
<tbody>
<tr>
<td>Diclofenac</td>
<td>anti- rheumatic agent</td>
<td>4 (301)</td>
<td>&lt;10 [3]</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ibuprofen</td>
<td>anti- rheumatic agent</td>
<td>2 (002)</td>
<td>90 [4]</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Metoprolol</td>
<td>selective- β receptor blocker</td>
<td>4 (301)</td>
<td>60 [4][5]</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Amoxicillin</td>
<td>antibiotic agent</td>
<td>1.187.403</td>
<td>30 [7]</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Carbamazepine</td>
<td>anti- convulsing agent</td>
<td>1.166.333</td>
<td>4 (301)</td>
<td>&lt;10 [8]</td>
<td></td>
</tr>
<tr>
<td>Sulfamethoxazol</td>
<td>antibiotic agent</td>
<td>198.734</td>
<td>6 (303)</td>
<td>28 [10]</td>
<td></td>
</tr>
</tbody>
</table>

Metabolism, daily doses, pbt- index & degradation performances of selected pharmaceuticals

(in extracts) chemical structure of:

Diclofenac

Metformin

Amoxicillin
No! - Diclofenac as one example!
Balancing of pharmaceuticals during the anaerobic treatment

http://www.vfa.de/static/generated/12817-ffdl2011-s25.jpg
Balancing of pharmaceuticals during the anaerobic treatment

- Human produced metabolites
- Raw substance
- Unknown products

- State of knowledge: good, moderate, poor

- Mineralisation into CH4, CO2

Diagram:
- CSTR (Continuous Stirred Tank Reactor)
- Analysis: CH4, O2, CO2, H2, H2S
- Abluft mit vorgeschaltetem Reinigung
- Magnetventil
- Kondensatfalle
- Vorlagebehälter
- Entnahme
- PUR 12mm
- Tiefwalzlösung

Flow paths:
- Raw substance
- Human produced metabolites
- Unknown products
Balancing of pharmaceuticals during the anaerobic treatment - how to solve that problems?

Identified problems and future works in our field:
- Balancing of treatment efficiency not proofed using raw substances
- Unknown products of both sites of the system; efficiency rates
- Ratio of the pharmaceutical inactivation during the process?
- „look at that what you want to see!“ → problems with the target analytics
- Possible options for solving some of that problems → Non-targets, new measurement methods

State of knowledge
- good
- moderate
- poor

Raw substance
Human produced metabolites
Anaerobic metabolites
Unknown products
Balancing of pharmaceuticals during the anaerobic treatment - heavy matrices
### Advantages/Disadvantaged for pharmaceutical measurement

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<th>GC- MS</th>
<th>LC-MS/MS</th>
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<tbody>
<tr>
<td>- Reduction of matrix → clean up of impurities</td>
<td>- Clean up vs. transformation of origin sample</td>
</tr>
<tr>
<td>- Increased stability of measurement by ionisation using EI- electron impact/ Diclorprop as standard substance</td>
<td>- Decreased stability of measurement by using ESI- electron spray injection → Ionsuppression depending on salt, fats, humic acid etc.</td>
</tr>
<tr>
<td>- Diclorprop as internal standard for all measurements</td>
<td>- Internal, deuterated C\textsuperscript{13}- standard substances are needed for each molecule</td>
</tr>
<tr>
<td>- Possibilities for non-target screening using stable molecule mass library</td>
<td></td>
</tr>
<tr>
<td>- At this juncture: method only useable for acetous pharmaceuticals which have to be derivate</td>
<td>- Valid method can be used for all “well-known” raw pharmaceutical substances</td>
</tr>
</tbody>
</table>
Balancing of pharmaceuticals during the anaerobic treatment - significant GC-MS measurements
Lessons learned and future applications of the new method

- Cost reduced, pharmaceutical measurement in mostly ALL liquid matrices
  - Equipment/Invest of GC- MS new: 60000€
  - Equipment/Invest of LC- MS/MS new: 150000€ – 200000€
  - Reducing the analytical prize/sample around 2/3

- Scientific usage and economic relevance
  - Fast and approved measurements for validation of experimental results in lab- as well as pilot scale
  - Potential and useful solution for process controlling and validation in municipal and innovative wastewater treatment systems

- Continuous development for reliable results in sludge and solids

- Increasing the number of pharmaceutical substances for GC- MS detection

- Development of non- abrasive and non- distracting methods for sample clean up and preparation

- Development of non- abrasive and non- distracting methods for solid analyzing (activated sludge, granular sludge, soil and concrete)
PALMQUIST, H.; JÖNSSON, H. (2003): Urine, faeces, greywater and biodegradable solid waste as potential fertilisers, ecosan- closing the loop; proceedings of the 2nd international symposium on ecological sanitation; 7th – 11th April 2003; Lübeck, Germany
ROENNEFAHRT, I. (2005): Verbrauchsmengen in der Bewertung des Umweltrisikos von Humanarzneimitteln; In: Umweltbundesamt (Hrsg); Arzneimittel in der Umwelt – Zu Risiken und Nebenwirkungen fragen Sie das Umweltbundesamt; Dessau, Germany; UBA-text publishing 29/05
UBA (2002): Arzneimittelwirkstoffe im Zu- und Ablauf von Kläranlagen; Berichte des österreichischen Umweltbundesamt
UBA (2011): Zusammenstellung von Monitoringdaten zu Umweltkonzentrationen von Arzneimitteln
Universität Dortmund (2003): Untersuchungen zum Eintrag und zur Elimination von gefährlichen Stoffen in kommunalen Kläranlagen Teil 1; Fachbereich Chemietechnik, Lehrstuhl Umwelttechnik

References
Thank you for your attention!

Special thanks goes to the BMBF for funding and also to the involved colleges from the b.is and from the other participated partners in the project.

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