Investigation on the behaviour of selected pharmaceuticals in the groundwater after infiltration of treated wastewater

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Abstract In a rural arid area without suitable receiving water the treated wastewater of a low loaded municipal wastewater treatment plant with full nutrient removal and additional post treatment steps is infiltrated into the unsaturated soil for groundwater recharge. Groundwater probes at increasing distances from the infiltration point have been sampled every two months over a period of 14 months. Beside conventional parameters (nutrients, carbon, and bacterial counts) samples were analysed for pharmaceutically active substances. Depending on and corresponding to their chemical structure and their fate during wastewater treatment, the selected substances showed different behaviour in the saturated zone of the groundwater. The antiepileptic Carbamazepine behaves very conservative and only is removed negligible even after long flow times within the subsurface zone. For other substances like the tranquilizer Diazepam or the analgesics Diclofenac a partial elimination during the different steps of wastewater treatment can be observed. Further degradation could be observed during the subsequent subsurface passage. In correlation with flow time additional removal of these substances from the aqueous phase can be observed. The musk substances Galaxolide and Tonalide were removed to some extend but not as good as the previous mentioned compounds.

Keywords Groundwater protection; pharmaceuticals; wastewater infiltration; wastewater treatment

Introduction
As a large amount of the water utilized by humans as source for drinking water or irrigation originates from groundwater, sustainable care has to be taken in prevention of pollution. Until now, traditional parameters, including nitrate, oxygen consumption capacity, content of organic carbon, pesticides, hygienic parameters and others were taken into consideration when investigating and monitoring groundwater quality. Only recently public, scientific and political focus was put on substances that occur in the aquatic environment in very low concentrations like endocrine modulators or pharmaceutically active compounds (PhACs) and personal care products. After consumption or disposal after usage those substances are discharged into the sewerage and further into a wastewater treatment plant. Because most of the substances are not readily degradable they are removed from the sewerage with different efficiency depending on the layout of the wastewater treatment plant (e.g. sludge retention time) or their adsorption behaviour to activated sludge (Clara et al., 2003). The effluent of wastewater treatment plants therefore is a significant source for those substances entering the aquatic environment via receiving waters, like rivers or streams (Ternes, 1998; Ternes et al., 1998; Heberer, 2002) but groundwater as well. Especially in arid or semi arid regions the effluent of wastewater treatment plants may be used for artificial groundwater recharge after percolation through the soil. For intended use of groundwater and the prevention of pollution, the knowledge on the behaviour of micropollutants in the groundwater is of central importance for both, the evaluation of potential risks and to avoid restrictions in the intended use. In this study selected PhACs and personal care products (see Table 1) were investigated for their behaviour in the saturated zone of the soil after infiltration of treated wastewater.
Methods

Substances investigated
In this study 10 PhACs with different application profiles were investigated. Substance name, application and corresponding consumption in Austria 1997 are summarised in Table 1.

Sample collection
During a period between April 2000 and June 2001 samples were taken from the investigated area with an interval of approx. 2 month resulting in 7 samples for all sampling sites. Sampling implements 5 sites associated to different treatment steps of the waste water treatment plant and 7 groundwater sites (see Table 2) including two sites with uninfluenced groundwater. The inflow of the wastewater treatment plant and the effluent after secondary clarification were collected as 24 h composite samples. Groundwater was sampled from groundwater probes exchanging the volume of the probes three times before sampling.

Analytical methods for PhACs
Two different detection methods were used for the analysis of the above mentioned compounds. Ibuprofen, Diclofenac and Bezafibrate were separated and analysed by GC-MS detection after derivatisation with diazomethane and a clean up step by silica gel chromatography. LC-MS-MS was employed for the analysis of Roxithromycin, Sulfamethoxazol, Carbamazepine, Diazepam and Iopromide. Ionisation of the analytes

Table 1: Investigated PhACs and consumption in Austria 1997 according to Sattelberger (1999)

<table>
<thead>
<tr>
<th>Substance</th>
<th>CAS-Nr.</th>
<th>Use</th>
<th>Consumption Austria (kg/a)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bezafibrate</td>
<td>41859-67-0</td>
<td>Lipid regulator</td>
<td>4,474</td>
</tr>
<tr>
<td>Carbamazepine</td>
<td>298-46-4</td>
<td>Antiepileptic</td>
<td>6,334</td>
</tr>
<tr>
<td>Diazepam</td>
<td>439-14-5</td>
<td>Tranquilizer</td>
<td>125</td>
</tr>
<tr>
<td>Diclofenac</td>
<td>15307-86-5</td>
<td>Analgetic, antiphlogistic</td>
<td>6,143</td>
</tr>
<tr>
<td>Galaxolide (HHCD)</td>
<td>1222-05-5</td>
<td>Polycyclic musk</td>
<td>n.a.</td>
</tr>
<tr>
<td>Ibuprofen</td>
<td>15687-27-1</td>
<td>Analgetic, antiphlogistic</td>
<td>6,696</td>
</tr>
<tr>
<td>Iopromid</td>
<td>73334-07-3</td>
<td>Contrast media</td>
<td>5,386</td>
</tr>
<tr>
<td>Roxithromycin</td>
<td>80214-83-1</td>
<td>Antibiotic</td>
<td>n.a.</td>
</tr>
<tr>
<td>Sulfamethoxazol</td>
<td>723-46-6</td>
<td>Antibiotic</td>
<td>63</td>
</tr>
<tr>
<td>Tonalide (AHTN)</td>
<td>1506-02-1</td>
<td>Polycyclic musk</td>
<td>n.a.</td>
</tr>
</tbody>
</table>

n.a. = data not available

Table 2: Sites sampled in this study and their characteristics

<table>
<thead>
<tr>
<th>Sampling site</th>
<th>Abbreviation</th>
<th>Typ*</th>
<th>Flow time** (d)</th>
<th>Content of treated wastewater (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reference site with uninfluenced ground water</td>
<td>uninf gw</td>
<td>-50</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Inflow WWTP</td>
<td>Inflow wwp tp</td>
<td></td>
<td>(-)</td>
<td>100</td>
</tr>
<tr>
<td>Effluent WWTP</td>
<td>Eff wwp tp</td>
<td>(10)</td>
<td>100</td>
<td></td>
</tr>
<tr>
<td>Polishing lagoon</td>
<td>Pol.Lag. wwp tp</td>
<td>(10)</td>
<td>100</td>
<td></td>
</tr>
<tr>
<td>Garvel filter</td>
<td>Gr.Filter wwp tp</td>
<td>(2)</td>
<td>100</td>
<td></td>
</tr>
<tr>
<td>Sicker biotope</td>
<td>Sicker wwp tp</td>
<td>(5)</td>
<td>100</td>
<td></td>
</tr>
<tr>
<td>Groundwater probe (well)</td>
<td>hp1 gw</td>
<td>18</td>
<td>100</td>
<td></td>
</tr>
<tr>
<td>Groundwater probe (well)</td>
<td>hp3 gw</td>
<td>25</td>
<td>100</td>
<td></td>
</tr>
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<td>Groundwater probe (well)</td>
<td>hp2 gw</td>
<td>75</td>
<td>91</td>
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<tr>
<td>Groundwater probe (well)</td>
<td>hp4 gw</td>
<td>105</td>
<td>72</td>
<td></td>
</tr>
<tr>
<td>Groundwater probe (well)</td>
<td>hp4 gw</td>
<td>140</td>
<td>76</td>
<td></td>
</tr>
</tbody>
</table>

* gw = groundwater; wwp tp = wastewater treatment plant; ** mean values related to infiltration in sicker biotope as t = 0; mean hydraulic retention time in treatment steps within WWTP in brackets
was done by electro spray ionisation in positive mode. Prior to the sample extraction a surrogate standard (josamycine, tylosine and dihydrocarbamazepine or meclofenamic acid) was added to the samples. Two different solid phase extraction phases (cyclohexane (CH)– and ENV+ – phase) were employed for LC-MS-MS sample preparation. The samples were acidified in the case of the C-18 and CH– solid phase extraction cartridges, whereas the addition of a neutral EDTA buffer solution was necessary for the ENV+ phase. The water samples were extracted and enriched by C-18 solid phase cartridges prior to the analysis with GC-MS. To overcome problems due to ion suppression in the LC-MS method, recoveries of the surrogate standard and measurements of multiple dilutions of the extracts were performed. The limit of quantification (LOQ) was set with 20 ng l⁻¹ and the limit of detection (LOD) with 10 ng l⁻¹ for all regarded substances.

Local situation

The investigation area is located in the most eastern part of Austria. The region’s settlements are characterised by rural structures concentrating on agriculture and in some regional parts on viniculture as major economic income sources. Regarding the geo-morphological situation, the region is characterised by glacial sediment deposits resulting from an intensively braided tributary system of the former River Danube basin. The irregular but frequent rearrangements of the tributaries caused a very heterogeneous sediment structure with gravel as its predominant deposit. The high amount of sand components is characteristic. The predominant soil type according to OECD TG 106 is soil type 6 (OECD, 2000). The extent of the aquifer within the investigation region varies between 4 m to 14.5 m. The mean depths to groundwater within the region can be classified between 0.5 m and 6 m (Zessner et al., 2003). Mean annual precipitation is about 550 mm. Evaporation often exceeds the precipitation rate during the summer months which causes a very low regeneration rate of groundwater.

The wastewater treatment plant sampled

The investigated waste water treatment plant designed for 7,000 population equivalent (p.e.) and provides activated sludge treatment including intermittent nitrification/denitrification and phosphorus precipitation with FeCl₃. The WWTP consists of screen and grit chamber, two aeration tanks (V = 2 × 1,546 m³) and two secondary clarifiers (V = 2 × 949 m³, A = 2 × 304 m²) for final sedimentation. Post-treatment steps consist of a polishing lagoon (V = 3,000 m³), a gravel filter (A = 500 m², h = 1.05 m) and three infiltration ponds (total A = 1,500 m², h = 1.5 m) where the treated wastewater is infiltrated into the soil–aquifer system. The actual mean load of the facility is about 2,450 p.e. (1 p.e. = 120 g COD d⁻¹), the maximum loading as an average value of two weeks about 4,350 p.e. The excess sludge is removed very infrequently what effects a very high sludge retention time (SRT₂₀°C) > 25 d, depending on the seasonal charging.

Results and discussion

A ground water flow model was calibrated for use in the frame of the project (Zessner et al., 2003) allowing estimating hydraulic flow times in the underground for the sampled groundwater probes. Obtained flow times are summarized in Table 2. In addition the dilution of the infiltrated treated waste water by uninfluenced ground water was calculated using boron as a conservative tracer substance. Measured boron concentrations in the course of the wastewater treatment plant show no change as expected (Figure 1a) but measured concentrations in the groundwater showed decrease due to dilution by uninfluenced ground water. Postulating a persistent behaviour of boron in the underground, dilution factors for the ground water sampling sites were calculated (Table 2). Obtained dilution
factors were used to recalculate all measured data (conventional parameters as well as PhACs) in order to consider only reduction of concentrations due to adsorption or degradation processes in the saturated zone but to exclude decrease due to dilution. Both considerations (regarding and neglecting dilution) are necessary for different concerns. Measured concentrations are summarized in Table 3, plots in Figure 2a–h are based on recalculated data.

**Diclofenac (Figure 2a)**

The acidic Diclofenac is a substance assumed to be eliminated from waste water mainly by adsorption processes to activated sludge (Clara et al., 2003). As in the investigated plant the daily excess sludge production is low due to high sludge retention time, only low amount of fresh adsorbent is produced and low removal rates of about 14% are observed. Within the

**Figure 1 a)** Boron concentrations in the compartments of the WWTP (black dots) and the ground water. Measured data in the ground water as white dots; recalculated concentrations regarding dilution by uninfluenced groundwater (background) as white triangles. Linear regression indicating conservative behaviour of boron in the groundwater. **b)** DOC concentrations in the sampling sites and first-order degradation calculated for DOC removal in the groundwater.
next treatment step, the polishing lagoon a tremendous decrease occurs. As adsorption and possible biodegradation is not likely to be the reason for this observation because no highly active sludge flock photo degradation may be a possible explanation. A similar, but not that high decrease in concentration can be observed in the infiltration pond, which has an open water surface too. During the first 25 days of subsurface flow no additional removal is observed. Nevertheless concentrations are below LOD after additional 50 days of flow indicating a complete removal within the saturated zone.

**Bezafibrate (Figure 2b)**
Bezafibrate shows a very good removal during waste water treatment with 99% of the inflow concentration being removed in the aeration tank. During post-treatment steps as the polishing lagoon additional removal down to LOQ is observed, so finally a total decrease in concentration of 99.5% was observed in our study. During groundwater flow no significant additional removal could be observed and concentrations stay around the LOQ. Only after 140 days of flow time all measured concentrations were below LOD.

**Ibuprofen (Figure 2c)**
Both being acidic substances, ibuprofen shows a similar behaviour as bezafibrate. Over 99% of the inflow concentration is removed during waste water treatment. Post-treatment steps show additional removal beneath LOQ. As the concentration in the infiltrated water already is below the LOD in most cases, no statement on the subsurface behaviour of ibuprofen can be given. Nevertheless behaviour similar to bezafibrate can be expected.
For diazepam (data not shown) the inflow concentrations of the investigated treatment plant already are in the range of LOQ, the effluent concentrations between LOQ and LOD. Even in the post treatment steps no significant change in concentrations was found. Despite of this uncertainty results from lab scale plants (Clara et al., 2003) showing no removal can be confirmed. In the ground water no change during the first 25 days of subsurface flow can be observed, but all data were below LOD after additional 50 days.

**Figure 2** a–h) Concentrations of respective PhACs for the particulate treatment steps of the investigated wastewater treatment plant (black dots) and in the groundwater (white dots). Flowtime for sampling sites according to Table 2. For the groundwater sites dilution by uninfluenced groundwater was taken into consideration for recalculating data from Table 3.

**Diazepam**

For diazepam (data not shown) the inflow concentrations of the investigated treatment plant already are in the range of LOQ, the effluent concentrations between LOQ and LOD. Even in the post treatment steps no significant change in concentrations was found. Despite of this uncertainty results from lab scale plants (Clara et al., 2003) showing no removal can be confirmed. In the ground water no change during the first 25 days of subsurface flow can be observed, but all data were below LOD after additional 50 days.
**Carbamazepine (Figure 2d)**

Showing only 10% removal between inflow and effluent, carbamazepine passes the wastewater treatment plant without any significant change in concentration. Post-treatment steps eliminate an additional 5%. During subsurface flow a very slow removal can be found resulting in about 30% removal compared to the inflow after 100 days. It is suspected, that this removal is rather due to half live time of the substance then to degradation processes.

**Tonalide (Figure 2e) and galaxolide (Figure 2f)**

Both polycyclic musks show a very similar behaviour during waste water treatment as well as during ground water flow; whereas galaxolide is measured in concentrations double the results of tonalide in all sampling sites. Both show removal rates of approximately 80% during wastewater treatment mainly due to adsorption because of their high adsorption coefficient \((\log K_D > 3 \, \text{l kg}^{-1})\). Compared to the inflow concentrations additional 10% are removed in the post-treatment steps, whereas no additional removal in the saturated zone is observed even after 140 day of flow time.

**Iopromide (Figure 2g)**

Only low influent concentrations were observed for iopromide, because there is no hospital providing X-ray in this rural catchment area. Nevertheless concentrations up to 120 ng l\(^{-1}\) iopromide can be found. In all samples of the effluent investigated concentrations were below LOD as in all post treatment steps. In the groundwater concentrations in the range of LOQ were observed for all sites and flow times. This may indicate a removal of Iopromide during wastewater treatment but hardly any additional removal in the groundwater.

**Roxithromycin (Figure 2h)**

For the antibiotic roxithromycin a removal rate of 46% was calculated within the wastewater treatment plant. Post treatment steps show no consistent picture, but regarding further concentrations in the groundwater after a flow time up to 25 days, additional removal for the post treatment steps can be stated with additional 25%. Similar to diazepam concentrations stayed unchanged until 25 days of subsurface flow but then decreased below LOQ.

**Conclusion**

During subsurface flow in the saturated zone monitored PhACs showed similar behaviour in regard to removal from the aqueous phase. Acidic drugs like diclofenac, bezafibrate and ibuprofen that are removed easily during wastewater treatment are subject to additional removal during post treatment steps like polishing lagoon, gravel filter or infiltration pond. Up to several µg l\(^{-1}\) of these substances can be removed efficiently. On the other hand, neutral substances like diazepam and carbamazepine that hardly show any removal during wastewater treatment remain stable during post treatment steps too as well as in the groundwater. The polycyclic musks tonalide and galaxolide show significant removal during waste water treatment and post treatment steps, but no significant further reduction during groundwater flow. As those substances are highly adsorbed to organic matter, a lack of absorbents in the ground water may be the reason for this behaviour.

Regarding the general behaviour of investigated substances it can be observed, that after a flow time of 75 days in the groundwater no additional removal for another 70 days could be observed. Concentrations reached after 75 days remain stable for the rest of the flow time observed. Actually it cannot be said if this stable state already is reached earlier, because there was no sampling site between a flow time of 25 and 75 days respectively.
Acknowledgements

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References


