Adsorption and removal at low atrazine concentration in an MBR pilot plant
G. Buttiglieri, L. Migliorisi and F. Malpei

ABSTRACT

Atrazine is a persistent organic pollutant and it has been widely used in agriculture and forestry in the world for more than fifty years. Atrazine shows ecotoxicity effects in aquatic ecosystems even at very low level concentrations with endocrine disruptor activity. Few studies were carried out on atrazine removal performances in drinking and waste-water by biological treatments, especially in membrane bio-reactors (MBRs). MBR technology might be more efficient than the conventional one in the removal of micro-pollutants. The fate of atrazine in wastewater treatment plants and its influence on the biomass activity was evaluated in this study. The experimental work was divided in three different phases: inhibition studies on different types of biomass (by means of microcalorimetry); adsorption studies on different sludges (conventional activated sludge (CAS) – and MBR) calculating adsorption isotherms and, finally, atrazine removal in an MBR pilot plant (simulating a treatment of atrazine and nitrate contaminated groundwater). The absence of significant inhibition was observed; higher atrazine adsorption on MBR sludge was detected for lower atrazine concentration (<50 µg L⁻¹); the removal efficiency in the MBR pilot plant was lower than 25% but higher than the theoretical one (based on adsorption isotherms).

Key words | adsorption, atrazine, calorimetry, inhibition, MBR

INTRODUCTION

Atrazine (2-chloro-4-ethylamino-6-isopropylamino-s-triazine, C₈H₁₄ClN₅) is a herbicide widely used for the protection of corn, sugarcane, grain sorghum, tea, and fruit crops. With 70,000 to 90,000 t applied per year atrazine was one of the most widespread triazine herbicides in the world (Steinberg et al. 1995). Actually it is banned in the European Community (2004/248/EC) but still in use all over the world and in US (around 40,000 t year⁻¹) mostly for maize cultivation. It is generally applied to soil pre-planting or pre-emergence, but is sometimes also applied to the foliage post-emergence (Dewey 1986; Graymore et al. 2001; US EPA 2008).

It may have detrimental effects on the aquatic ecosystem and human health and carcinogenic and endocrine disrupter activity in mammals (Friedman 2002; Hayes et al. 2002; Juliani et al. 2008). Chronic low-dose exposure to toxicants may cause silent damages in the reproductive system (Stevens et al. 1994); this might imply that low doses of atrazine could represent a stressful condition to the organism exposed to this herbicide for a long time.

Most of literature is on atrazine presence, adsorption and removal in soil systems and/or pure cultures, or on chemical and physical methods only, to treat contaminated sources. Very few scientific researches deal with atrazine removal in drinking and waste-water by biological treatment (Meakins et al. 1994; Nitschke et al. 1999), especially in membrane bio-reactors (MBRs). The MBR technology might be more efficient than conventional activated sludge in the removal of persistent compounds and micro-pollutants: the membrane retains the whole biomass inside the reactor, it is possible to work at high sludge retention time (SRT) and, hence, to promote the growth of slow bacteria community (Clara et al. 2005; Bernhard et al. 2006; Buttiglieri & Knepper 2007; Bouju et al. 2008). Bioaugmentation, using genetically engineered microorganism, might be a feasible way for removing recalcitrant chemicals like atrazine from wastewater, especially in MBRs and at high atrazine loading (Liu et al. 2008). Moreover alternating anoxic and aerobic phases (as applied in the present research) has been recommended to get high growth of
Pseudomonas ADP, a strain able to degrade atrazine (Katz et al. 2001).

It has to be noted, finally, that most of scientific reports on atrazine degradation deal with much higher concentrations than the ones measured in groundwater. In the present work concentrations close to the environmental ones were used (influent MBR concentration of 10 µg L⁻¹).

The MBR pilot plant here presented was previously tested to be highly efficient in denitrification of drinking water sources (Buttiglieri et al. 2005). Aim of this study was to evaluate the fate of atrazine in wastewater treatment plants (WWTPs), simulating a treatment of atrazine and nitrate contaminated groundwater, and its influence on the biomass. The experimental plan included:

- toxicity tests on MBR biomass;
- atrazine adsorption isotherms (MBR and CAS sludges) in laboratory tests;
- atrazine removal in an MBR pilot plant continuously fed for 98 days.

**MATERIALS AND METHODS**

**SPE and HPLC analyses**

Analyses were done by HPLC (high performance liquid chromatography) with previous solid phase extraction (SPE). C₁₈ (ODS) Merck cartridges, volume 6 mL and 500 mg of filtering resin, were used for SPE. After activating the cartridges (with methanol, HPLC grade with purity 99.8%), a volume of 40–100 mL of sample was filtered and the adsorbed atrazine was then recovered in 5 mL of acetonitrile (HPLC grade, purity 99.9%, Advanced Biotech Italia). Later on evaporation with nitrogen flux (TurboVap II, Zymark evaporator) was assessed with constant flux (1.6 bar pressure, 35 °C water temperature) to dry the sample (80 minutes). Final re-eluting was with 0.5 mL of solution acetonitrile:water = 2:8.

Atrazine (analytical standard powder, HPLC grade) analyses were done with HP1050 HPLC. An Hypersil ODS column (length 20 cm, diameter 4.6 mm, particle size of 5 µm, Agilent) and an ODS precolumn (length 10 mm, diameter 5.2 mm) were used. The mobile phase was acetonitrile and a solution 1 mM of ammonium acetate in water. The flux was 0.8 mL min⁻¹, with a linear gradient from 20% to 70% of acetonitrile in 35 min; the column temperature was 25 °C. The injection volume was 100 µL. The detection system was an UV lamp with a DAD (diode-array detector) and the wavelength for atrazine quantification was 250 nm.

**Adsorption tests**

Lab-scale batch adsorption tests were performed in a 1 L beaker, continuously stirred, filled with 500 mL of activated sludge (previously inactivated with HgCl₂ – 100 mg L⁻¹), at 2 and 24 hours contact times. Atrazine was spiked at 0.5–1–5–10–20–50–75–100 µg L⁻¹ (additional sample for 24 hours contact time at 2.5 µg L⁻¹) from a standard solution (20 mg L⁻¹ atrazine). Membranes (Durapore PVDF, diameter 73 mm, porosity 0.45 µm, Millipore Stericup) were used for sample filtration. A preliminary test to evaluate atrazine retention on the filtering apparatus provided a negligible loss of atrazine (less than 1%, 0.5 L of standard solution filtered).

Experimental adsorption data were fitted according to the Freundlich isotherm:

\[ q = K_f \frac{C}{C^*}^{1/n} \]

where \( q \) is the adsorbed pollutant quantity per adsorbent mass unit (mg_adsorbed kg_adsorbent⁻¹); \( C \) is the equilibrium concentration in aqueous phase (mg L⁻¹); \( K_f \) is the Freundlich constant (L kg⁻¹); \( n \) is an adimensional coefficient.

Adsortion experiments were made on two sludges coming from:

- the MBR pilot plant described in the next section; 3 g_TSS L⁻¹ and a VSS/TSS ratio of 92%; contact times: 2 and 24 hours.
- a full scale municipal WWTP (CAS, 110,000 PE, 40,000 m³ d⁻¹); 2.8 g_TSS L⁻¹ and a VSS/TSS ratio of 64%; contact time 24 hours.

**Calorimetry – inhibition tests**

A high-resolution bench-scale calorimeter (Bio-RC1 – Mettler Toledo AG), especially suited for microbiological studies, has been used to investigate atrazine influence in anoxic and aerobic conditions. A standard 2-liter jacketed glass reactor was operated in the isothermal mode (more details in Daverio et al. 2003). Therefore, when a process dissipates or takes up heat, \( T_j \) respectively decreases or increases. The resulting temperature gradient across the reactor wall is directly proportional to the thermal power \( Q_t \) (W), produced or absorbed by the process, according to the equation:

\[ Q_t = UA(T_r - T_j) \]
where $U$ is the overall heat-transfer coefficient (W m$^{-2}$ K$^{-1}$), $A$ is the heat transfer surface (m$^2$). Dissolved oxygen concentration and pH are on-line acquired.

The globally generated or adsorbed heat amount during the bacterial activity, corresponding to the enthalpic variation associated to the whole metabolic process, is obtained integrating the thermal power throughout the peak period.

Calorimetric tests to evaluate the inhibition were based on the comparison between the heat, produced by the biomass dosing a specific substrate (ethanol for heterotrophic or ammonia for autotrophic bacteria), before and after atrazine spikes. In every inhibition test atrazine was in ethanol solution (10 g atrazine L$^{-1}$) due to its low water solubility.

1.5 L sludge samples from the MBR pilot were tested with the following protocols:

- **heterotrophic bacteria in anoxic conditions**: ethanol (165 mgCOD L$^{-1}$); nitrate (20 mgN-NO3 L$^{-1}$); atrazine (tests at 10–20–30 mg L$^{-1}$); nitrate (20 mgN-NO3 L$^{-1}$).
- **heterotrophic bacteria in aerobic conditions**: ethanol (110 mgCOD L$^{-1}$); atrazine (15 mg L$^{-1}$); ethanol (110 mgCOD L$^{-1}$).
- **autotrophic bacteria**: ammonia (3.3 mgN-NH4 L$^{-1}$); atrazine (15 mg L$^{-1}$); ammonia (3.3 mgN-NH4 L$^{-1}$).

### Pilot plant configuration and operation

The MBR pilot plant was designed to simulate the treatment of atrazine and nitrate contaminated groundwater (semi-synthetic feed). The water was taken from Maggiore Lake (Italy; mgN-NO3 L$^{-1}$ < 1; mgN-NO2 L$^{-1}$ < 0.015; mgN-NH4 L$^{-1}$ < 0.020; mgPO4 L$^{-1}$ < 0.030). Ethanol was used as the carbon source (Peng et al. 2007; Puig et al. 2007); with a load of 0.125 gCOD gVSS$^{-1}$ d$^{-1}$; nitrate load in the range 0.017–0.022 gN-NO3 gTSS$^{-1}$ d$^{-1}$ with a ratio 1.4–1.8 gN gTSS$^{-1}$.

It was made up of a first anoxic tank (90 L volume, 2.7–3.6 gTSS L$^{-1}$) followed by an aerobic tank (190 L, 5.6–6.9 gTSS L$^{-1}$). A peristaltic pump recycled the sludge from the aerobic to the anoxic tank. Inflow and recyle flow were constant and set at 252.9 L d$^{-1}$. Influent nitrate concentration was 96.8–124.5 mgN-NO3 L$^{-1}$ and influent COD was 697 mg L$^{-1}$; respective mean removal efficiencies were 85 and 98.8%. The SRT was maintained at a high value (79 days): high SRT (more feasible in MBRs) should be more efficient on the biodegradation/biotransformation since it should allow the development of substrate specific slowly growing biomass and lead to a more diverse biomass (Clara et al. 2005).

A microfiltration membrane module ZW-10 (zee-weed 10, Zenon-GE InfraWater, nominal surface of 0.93 m$^2$, nominal cut off 0.2 µm, back-flushing once a day, air flow 2.7 m$^3$ h$^{-1}$) was placed inside the aerobic tank. Scheme is presented elsewhere (Buttiglieri et al. 2005).

The MBR was continuously fed at a constant atrazine concentration of 10 µg L$^{-1}$ for 98 days. The herbicide concentration in the permeate was measured twice a day at the beginning of the test, once a day after the first week and, in the last 20 days, 3 times per week.

### RESULTS AND DISCUSSION

#### Inhibition tests

Biomass samples from the MBR pilot plant were investigated to check acute atrazine inhibition effects, at high concentrations (range 10–50 mg L$^{-1}$), in the hypothesis that if no inhibition is detected at high concentration there is no significant acute effect at lower concentrations too.

The MBR pilot plant was designed also for biological denitrification; tests on denitrificating bacteria in anoxic conditions were hence very important. Heights and areas under the two heat peaks following nitrate spiking (before and after atrazine, see Material and Methods), were compared to evaluate differences in degradation activity. Spiked solutions, heights of the peaks (height$\text{max}$) and differences between them (Δheight$\text{max}$) are presented in Table 1 and an example test in Figure 1.

The very intense but narrow peak (Figure 1, time 2.1 h) following atrazine spike was due to the physical mixing enthalpy of water with ethanol, as no degradation was possible with no nitrate available in this step.

Small Δheight$\text{max}$ values were observed with a maximum reduction of around 14% at 30 g atrazine L$^{-1}$-1. The overall enthalpic variations, represented by the area under the

<table>
<thead>
<tr>
<th>Atrazine Input (mg L$^{-1}$)</th>
<th>EthOH Input (mgCOD L$^{-1}$)</th>
<th>VSS (g L$^{-1}$)</th>
<th>height$\text{max}$ I peak (W gVSS$^{-1}$)</th>
<th>height$\text{max}$ II peak (W gVSS$^{-1}$)</th>
<th>Δheight$\text{max}$ (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>1,649</td>
<td>2.2</td>
<td>0.236</td>
<td>0.225</td>
<td>4.66</td>
</tr>
<tr>
<td>10</td>
<td>4,946</td>
<td>2.7</td>
<td>0.224</td>
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</tr>
<tr>
<td>20</td>
<td>3,297</td>
<td>2.4</td>
<td>0.189</td>
<td>0.181</td>
<td>4.23</td>
</tr>
<tr>
<td>30</td>
<td>4,946</td>
<td>2.9</td>
<td>0.143</td>
<td>0.123</td>
<td>13.99</td>
</tr>
</tbody>
</table>
peaks, were always very similar (differences lower than 2%) confirming that no significant inhibition occurred.

Control tests to check inhibition induced by ethanol (tests at 1,649 and 4,946 mg\textsubscript{COD} L\textsuperscript{-1}, same protocol but without atrazine, Table 1) were made observing similar $\Delta$height\textsubscript{max} values.

It can be concluded that most of the inhibition effect to heterotrophic biomass in anoxic conditions was due to ethanol: the ethanol concentration was very high (up to almost 5,000 mg\textsubscript{COD} L\textsuperscript{-1}) and plausibly there was a substrate concentration inhibition. Nonetheless higher inhibition was observed when ethanol and atrazine spiked together (4,946 mg\textsubscript{COD} L\textsuperscript{-1}, 30 mg L\textsuperscript{-1} atrazine) with a difference of around 3%.

Similar inhibition tests were made on heterotrophic bacteria in aerobic conditions (atrazine 15 mg L\textsuperscript{-1}). Heights were similar: the first one around 1.9 W, the second one 1.8 W; the areas almost equals (0.2 Wh). It can be concluded that the two peaks are substantially equals and that atrazine had no significant effect on biomass metabolism (in agreement to Zagorc-Koncan \textsc{et al.} 1999, atrazine spiked at 30 mg L\textsuperscript{-1}).

Finally, similar tests were made on autotrophic bacteria (atrazine 15 mg L\textsuperscript{-1}) but it was not possible to evaluate any inhibition effect. Only very limited signals, both in the heat and in the dissolved oxygen concentration, were detected. This effect was probably due to the small ammonia load in the pilot plant (influent concentration less than 0.020 mg\textsubscript{N-NH\textsubscript{4}} L\textsuperscript{-1}) and presumably to the small autotrophic biomass population.

Still it must be considered that atrazine concentration in the environment is generally much lower than in the aforementioned tests. Consequently, atrazine inhibition on heterotrophic biomass in environmental conditions (and in the MBR pilot plant presented in the following) may be ruled out with a good safety factor.

### Adsorption tests

MBRs, compared to CAS, should be mainly more effective on adsorption of pollutants. The membrane retains big particles such as colloids inside the bioreactor enhancing the adsorption surface and therefore, the removal onto suspended solids (Bouju \textsc{et al.} 2008).

Adsorption tests were assessed on MBR biomass at a contact time of 2 and 24 hours. The latter is usually approximated to equilibrium conditions (Celis \textsc{et al.} 1998; Spongberg & Gangliang 2000); moreover this time was close to the MBR hydraulic retention time (around 26 hours).

Quantities adsorbed at 24 hours contact time were as an average 60–70\% higher than in 2 hours trials, with a minimum of 43\% and a maximum of 94\%. These differences confirmed the hypothesis that two hours time was not sufficient to reach the equilibrium.

In Figure 2 the experimental data correlation is shown in a bi-logarithmic graph, providing the following Freundlich isotherm (power interpolation, $R^2 = 0.98$):

$$q = 0.0275 C^{1.5328}$$

Sludge drawn from the oxidation tank recycle of the full scale WWTP was used for adsorption tests on CAS sludge (contact time 24 hours). $K_f$ was around $10^{-8}$ L g\textsubscript{VSS}\textsuperscript{-1}, $1/n$ equal to 5.334 ($n = 0.187$) higher than in MBR sludge (power interpolation $R^2 = 0.85$).
Table 2 compares $C_{eq}$ and $q$ obtained for the two sludges. At low atrazine dosages (<50 µg L$^{-1}$), MBR sludge provides a higher adsorption capability than CAS one (no adsorption at all under 10 µg L$^{-1}$).

Atrazine concentration in environmental conditions is anyway usually lower than 5 µg L$^{-1}$; hence, despite the differences between the sludges, adsorption removal should be small. For higher concentrations, on the other hand, significant removal might be obtained (e.g. for 100 µg atrazine L$^{-1}$ dosed the equilibrium concentration was around 56–59 µg L$^{-1}$).

**Atrazine removal in a MBR pilot plant**

MBRs are expected to be more effective than CAS for adsorption (as discussed before) and biodegradation/transformation of micropollutants. The biological degradation, in fact, may be improved by complete biomass retention and increased SRT (more feasible in MBRs). Comprehensively, MBRs should be able to lead to a greater removal of poorly biodegradable compounds (Clara et al. 2005; Bernhard et al. 2006), like atrazine.

The purpose of this section, and of the whole experimental work on the herbicide atrazine, was, hence, to evaluate whether real possibilities exist to remove atrazine by means of the MBR technology, simulating a nitrate and atrazine contaminated groundwater treatment system.

The trend in the permeate concentration along the first 7 days followed the expected one after a step increase in continuous stirred tank reactor without reaction or degradation.

After that, MBR atrazine permeate concentration was quite variable but, for most of the sample, in the range 7.6–9.8 µg L$^{-1}$ (Figure 3). Very few points above 10 µg L$^{-1}$ were due to technical problems in influent, permeate extraction and atrazine feeding. Only in few occasions concentrations lower than 6 µg L$^{-1}$ were measured with a single sample of 4.9 µg L$^{-1}$.

Taking into consideration the values after day 7 (excluding the very first days after the step increase) average permeate concentration was 8.5 µg L$^{-1}$ corresponding to 15% average removal.

<table>
<thead>
<tr>
<th>$C_{start}$ (µg L$^{-1}$)</th>
<th>$C_{eq}$ (µg L$^{-1}$) MBR</th>
<th>$C_{eq}$ (µg L$^{-1}$) CAS</th>
<th>$q$ (µg gSSV$^{-1}$) MBR</th>
<th>$q$ (µg gSSV$^{-1}$) CAS</th>
</tr>
</thead>
<tbody>
<tr>
<td>2.5</td>
<td>2.29</td>
<td>2.50</td>
<td>0.08</td>
<td>0</td>
</tr>
<tr>
<td>5</td>
<td>4.45</td>
<td>5.00</td>
<td>0.24</td>
<td>0</td>
</tr>
<tr>
<td>10</td>
<td>7.76</td>
<td>10.00</td>
<td>0.91</td>
<td>0</td>
</tr>
<tr>
<td>20</td>
<td>14.71</td>
<td>17.32</td>
<td>2.19</td>
<td>1.49</td>
</tr>
<tr>
<td>50</td>
<td>38.14</td>
<td>39.45</td>
<td>4.79</td>
<td>5.60</td>
</tr>
<tr>
<td>75</td>
<td>52.17</td>
<td>50.14</td>
<td>9.54</td>
<td>14.69</td>
</tr>
<tr>
<td>100</td>
<td>55.93</td>
<td>58.77</td>
<td>17.97</td>
<td>23.34</td>
</tr>
</tbody>
</table>

**Figure 2** | Linearized Freundlich isotherm for 24 hours contact time – MBR sludge.

**Figure 3** | Atrazine permeate concentration compared with the influent one (10 µg L$^{-1}$).
At 8.5 µg\textsubscript{atrazine} L\textsuperscript{-1}, taking into account Freundlich isotherms calculated in the previous section, the amount of adsorbed atrazine was around 0.73 µg\textsubscript{atrazine} g\textsubscript{VSS}\textsuperscript{-1}. The total pilot plant biomass (1,297 g\textsubscript{VSS}) should adsorb around 948 µg\textsubscript{atrazine} (lower than the daily atrazine load of 2.53 mg d\textsuperscript{-1}) and be already saturated after a single day. The adsorption system was consequently hypothesised to be at equilibrium after short time.

Consequently, in the hypotheses that the system was at the equilibrium and the sole removal mechanism in the MBR pilot plant was adsorption on extracted surplus sludge (with no degradation occurring) the atrazine mass balance equations are:

\begin{align*}
Q(C_0 + C_2) - 2QC_1 &= 0 \quad \text{anoxic tank} \\
2QC_1 - 2QC_2 - QSC_2 - qX_S &= 0 \quad \text{aerobic tank}
\end{align*}

where \(C_0\) is the influent atrazine concentration (10 µg L\textsuperscript{-1}), \(C_1\) and \(C_2\) (the latter being theoretically equal to the permeate one) the equilibrium concentrations in the two tanks (unknown), \(Q\) the influent flow (252.9 L d\textsuperscript{-1}, equal to the recycle flow), \(Q_S\) the average daily surplus sludge flow (around 2.8 L d\textsuperscript{-1}), \(X_S\) the daily total suspended solid extraction (equal to around 16.56 g\textsubscript{VSS} d\textsuperscript{-1}) and \(q\) the adsorbed atrazine amount on surplus sludge extracted from the plant (µg \textsubscript{VSS} d\textsuperscript{-1} from Freundlich equation obtained in laboratory tests, MBR sludge, 24 hours contact time).

The resulting equilibrium concentrations in the tanks are 9.83 µg L\textsuperscript{-1} for the aerobic tank and 9.92 µg L\textsuperscript{-1} for the anoxic one, higher than the measured mean outflow concentration (8.5 µg L\textsuperscript{-1}).

This discrepancy might indicate different adsorption capabilities in pilot plant conditions compared to laboratory tests.

Still, partial chemical and/or biological degradation phenomena might be assumed in a few days with lower atrazine permeate concentrations. This hypothesis might be partially confirmed by the obtained chromatograms: in a few samples (mostly lower than 7 µg L\textsuperscript{-1}) different peaks other than atrazine were observed (not present in the other ones). More accurate investigations are, anyway, necessary to confirm them as atrazine metabolites.

Partial degradation phenomena and the growth of atrazine metabolising biomass may be hypothesised even if for very short periods. This biomass, if present, was not developed in a constant and reliable way as no stable degradation was gained. If this partial degradation is confirmed, a further comprehension of best operative conditions would become of great importance in order to obtain and maintain a biomass population able to degrade and remove atrazine in a satisfactory way.

Nonetheless it is apparent that, during the experimental plan, obtained removal efficiencies were very limited and they do not suggest a real application of the MBR system for this pollutant at these low concentrations (2.5 µg\textsubscript{atrazine} d\textsuperscript{-1} compared to 176 g\textsubscript{COD} d\textsuperscript{-1} of easily biodegradable ethanol) unless a proper biomass is not correctly acclimatised or its capabilities increased (at higher SRT or by bio-augmentation).

**CONCLUSIONS**

The removal of atrazine with semi-synthetic feed was researched by means of a MBR pilot plant.

Adsorption isotherms on activated sludge (sampled from the MBR plant and a small CAS) were calculated in a concentration range between 0.5 and 100 µg L\textsuperscript{-1}. For MBR sludge the Freundlich coefficient \(K_f\) was 0.0275 L\textsubscript{VSS} \textsuperscript{-1} and the exponent \(1/n\) equal to 1.53. At low atrazine dosages (<5 µg L\textsuperscript{-1}), MBR sludge provides a higher adsorption capability than CAS one. Moreover, by means of micro-calorimetry, the absence of atrazine inhibition effects on heterotrophic and autotrophic bacteria at low atrazine concentration was confirmed (slight inhibition effects were detected at much higher concentrations, mg L\textsuperscript{-1} order of magnitude).

For 98 days permeate and tanks atrazine concentrations were evaluated in the pilot plant. More than 80% of the samples had a limited removal efficiency, lower than 25%. The permeate concentration was calculated on the basis of lab-scale isotherms, at equilibrium conditions, in the hypothesis that no biodegradation was present but only atrazine removal by adsorption on surplus sludge extracted; it resulted equal to 9.83 µg L\textsuperscript{-1}. Real permeate concentrations, instead, were lower for most of the experimentation with an average value of around 8.5 µg L\textsuperscript{-1} (corresponding to 15% mean removal). In a real case study the adsorption process might be different from the hypothesised one and obtained in laboratory conditions. Nevertheless it is not possible to exclude a partial and inconstant chemical and/or biological removal. More investigations on metabolites, adsorption and at higher SRT or by means of bio-augmentation are needed.

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