# Temporal Variation of Chemical Persistence in a Swedish Lake Assessed by Benchmarking

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Ⓢ Supporting Information

ABSTRACT: Chemical benchmarking was used to investigate the temporal variation of the persistence of chemical contaminants in a Swedish lake. The chemicals studied included 12 pharmaceuticals, an artificial sweetener, and an X-ray contrast agent. Measurements were conducted in late spring, late autumn, and winter. The transformation half-life in the lake could be quantified for 7 of the chemicals. It ranged from several days to hundreds of days. For 5 of the chemicals (bezafibrate, climbazole, diclofenac, furosemide, and hydrochlorothiazide), the measured persistence was lower in late spring than in late autumn. This may have been caused by lower temperatures and/or less irradiation during late autumn. The seasonality in chemical persistence contributed to changes in chemical concentrations in the lake during the year. The impact of seasonality of persistence was compared with the impact of other important variables determining concentrations in the lake: chemical inputs and water flow/dilution. The strongest seasonal variability in chemical concentration in lake water was observed for hydrochlorothiazide (over a factor of 10), and this was attributable to the seasonality in its persistence.
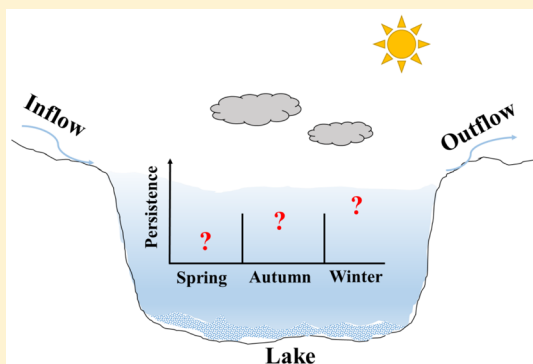
INTRODUCTION

Environmental persistence, often defined as the transformation half-life of a chemical, is an important factor controlling the exposure of organisms to the chemical, the extent of its transport to and impact in remote areas, and the reversibility of environmental contamination.[1,2] Various processes can contribute to the transformation of a chemical in the environment, including biotransformation, photolysis, redox reactions, and hydrolysis. These processes are influenced by environmental conditions such as temperature, pH, redox status, and solar irradiation. Thus, persistence is not an intrinsic property of a chemical but varies spatially and temporally. To date, there have been limited possibilities to measure persistence in the field, and hence, existing regulations rely on laboratory tests to evaluate chemical persistence. While laboratory tests may be less costly and applicable to a broader range of chemicals, without measurements of persistence in the field, one cannot know the ability of the laboratory tests to predict persistence in the environment. Furthermore, our understanding of the spatial and temporal variability of persistence remains limited. This is particularly true for chemicals for which biotransformation and/or photolysis are important transformation processes, as the transformation of these chemicals can be expected to be particularly variable in space and time and it is difficult to accurately simulate in the laboratory the environmental conditions that govern the transformation rate. Field measurements of persistence are thus a key component, along with laboratory experiments and modeling, of an integrated approach to assessing chemical persistence in the environment.

When persistence in aquatic systems has been measured in the field, it has often been with the mass balance approach.[3,4] In the mass balance approach, all of the significant chemical inflows and outflows to the aquatic system are measured over a certain period of time. In systems that are not at steady state, the change in the inventory of chemicals in the system over this time period is also measured. The equation for the conservation of mass is then used to derive the mass of chemical loss due to transformation, which is then used to calculate a transformation rate. While this method is scientifically rigorous, conducting a mass balance study can be difficult and costly, for instance, when an important chemical mass flow (e.g., volatilization, sedimentation) is variable in space or time.

Recently, we explored the potential of using chemical benchmarking to measure chemical persistence in lakes.[5] In this approach, the behavior of the test chemical is not studied in absolute terms, but rather relative to the behavior of another chemical with known persistence properties, which is referred to as the benchmark chemical. This is analogous to the use of an internal standard in chemical analysis, whereby in this case the ratio of the input to the lake of the test chemical and the benchmark chemical must be known or measurable. We created a framework for the application of the benchmark approach and showed that under certain conditions chemical persistence
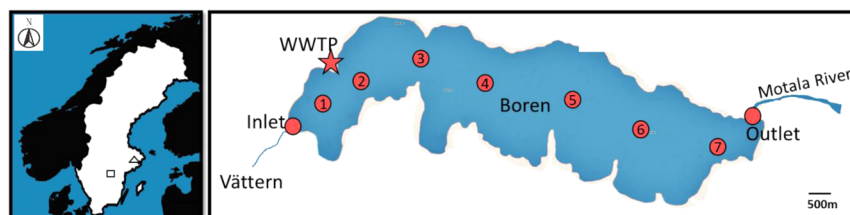
Figure 1. Left: location of Boren (square) in Sweden (triangle shows Stockholm). Right: map of the study site, Boren, showing the seven sampling points within the lake, the sampling point at the inlet where water enters Boren from Vättern, the sampling point at the outlet of the lake (all red dots), and the location of Karshult WWTP (red star).

could be quantified by simply measuring the ratio of the concentrations of the test and benchmark chemicals in the major source of chemical input to the lake and in the lake itself.[5] Depending on the time scale of the persistence of chemicals that one is interested in, different lake systems can be used. The framework was successfully applied to measure the persistence of several pharmaceuticals in Norra Bergundasjön, a small lake in Sweden, with transformation half-lives ranging from several days to several months.[6]

In this study, we apply the benchmarking approach to study the temporal variability of persistence in another Swedish lake, Boren. We were particularly interested in studying chemicals with a transformation half-life of weeks to months. We thus chose a lake with a hydraulic residence time of the order of a month, which gave us the possibility to study the temporal variability on a seasonal scale. We designed the sampling program to focus on three seasons in which we expect there could be distinct differences in persistence: late spring (stronger light, higher temperature), late autumn (little light, lower temperature), and winter (ice covered). A suite of chemicals emitted from a WWTP comprising 12 pharmaceuticals, an artificial sweetener, and an X-ray contrast agent were studied. We compare the seasonality in persistence with the seasonality in chemical inputs and the seasonality in water flow/dilution to identify the primary determinant of the seasonal variability in the ecosystem's exposure to the contaminants.

METHODS

Study Site. The studied system was a Swedish lake, Boren, located 195 km southwest of Stockholm (Figure 1). It has a surface area of 27.7 km^2, an average depth of 5.4 m, and a pH of 8.0.[7] It is ice covered for 128 days per year on average. The major water inflow into Boren is from Vättern, Sweden's second largest lake with a hydraulic residence time of 60 years.[8] The Karshult wastewater treatment plant (WWTP; 58°34′13.95′′ N, 15°5′45.75′′ E), which serves 26 900 population equivalents, discharges its effluent into the lake close to the inlet. The dilution factor of the effluent by the inflowing lake water is 300−400. The only outflow is via the Motala River.

Sampling was conducted between April 15 and June 9, 2013 and between November 04, 2013 and February 16, 2014. The average water temperature in the lake (measured at 5 m depth at the lake outlet) was 10.6 °C during the late spring period (April 15−June 9), 5.1 °C during the late autumn period (Nov. 4−Dec. 1), and 1.3 °C during the winter period (Dec.2, 2013−Feb.16, 2014). The irradiation at the lake's surface in late spring is the highest and decreases from autumn to winter (30 year (1961−1990) average of 140−145 kWh m^{-2} in May, 21−24 kWh m^{-2} in November, and 6−8 kWh m^{-2} in December).[8] There is a similar seasonal trend for sunshine hours (205−230

h in May, 60−70 h in November, and 30−40 h in December).[8] The water outflow, which is regulated by a dam at the inlet from Vättern, ranged between 40 and 50 m^{3}s^{-1} with the exception of the 4 weeks of the autumn period, when it was ∼25 m^{3}s^{-1} (Figure S1 in the Supporting Information).

Chemicals. A group of 14 chemicals was selected for the study comprising 12 pharmaceuticals (acetaminophen, bezafibrate, carbamazepine, climbazole, diclofenac, fluconazole, furosemide, gemfibrozil, hydrochlorothiazide, ibuprofen, ketoprofen, and sulfamethoxazole), one X-ray contrast agent (diatrizoic acid), and one artificial sweetener (acesulfame K). Twelve of them had been included in the study that measured chemical persistence in Norra Bergundasjön.[6] The authentic standards, their isotope-substituted internal standards, and other chemicals used in this study are described in Text S1 in the Supporting Information.

Benchmarking Approach. The benchmarking approach to quantify persistence in lakes, as described in our previous work,[5] assumes that the lake is well-mixed and at steady state. We tested these assumptions, as described in the following section. If, furthermore, water advection is the dominant physical removal process for the chemicals in question (test chemicals), then the persistence of the test chemicals in the lake (defined as the water column and surficial (nonburied) sediment) can be estimated by measuring the ratio between the mass flows of a benchmark chemical and the test chemical into the lake and the ratio between their concentrations in the outflow of the lake (eq 1).

$$t_{0.5R} = \frac{\ln(2)}{k'_{R,\text{Test}}} = \ln(2)\frac{\tau_A}{\left(\frac{I_{\text{Test}}}{I_{\text{BM}}}\right)/\left(\frac{C_{W,\text{Test}}}{C_{W,\text{BM}}}\right) - 1} \tag{1}$$

where k'_R is the first-order rate constant for transformation of the test chemical in the lake, day^{-1}; τ_A is the hydraulic residence time of the lake, day; I is the mass flow of chemical entering the lake from all sources, g day^{-1}; C_W is the concentration of chemical in the lake water, g m^{-3}; the subscripts "Test" and "BM" refer to the test and the benchmark chemicals, respectively. The benchmark chemical is assumed to have the same dominant physical removal process as the test chemical (i.e., advection in this case) and to have a negligible degradation in the lake.

All 14 chemicals in this study have a low air−water distribution coefficient (log $D_{AW} < -6.95$) and octanol−water distribution coefficient with log $D_{OW} < 4.33$ at pH 8.0 (Table S1, Supporting Information). A multimedia model simulation of Boren indicated that chemicals with these partitioning properties mainly stay in the water phase and that water advection is the dominant process for their physical removal from the lake (Figure S2, Supporting Information).

Table 1. Concentrations of 14 Chemicals (Mean ± Standard Deviation of Triplicate Samples) at the Outlet of Boren during Different Periods, ng L^{-1a}. The Magnitude of the Seasonality in Chemical Concentrations and the Primary Causes Are Also Listed

	late spring	late autumn	early winter	midwinter	seasonality[b]	primary causes
acesulfame K	63.1 ± 4.4	71.8 ± 2.0	72.0 ± 2.1	56.4 ± 1.3	28%	dilution
acetaminophen	<0.29	<0.29	<0.29	<0.29		
bezafibrate	0.21 ± 0.10	0.14 ± 0.01	0.19 ± 0.04	0.13 ± 0.02	62%	chemical input
carbamazepine	1.97 ± 0.04	2.60 ± 0.03	2.62 ± 0.03	2.03 ± 0.03	33%	dilution
climbazole	0.05 ± 0.002	0.04 ± 0.01	0.03 ± 0.01	0.05 ± 0.01	67%	chemical input
diclofenac	3.38 ± 0.35	2.34 ± 0.07	2.49 ± 0.30	4.08 ± 0.18	74%	chemical input
diatrizoic acid	<2.23	<2.23	<2.23	<2.23		
fluconazole	1.07 ± 0.03	0.89 ± 0.08	1.04 ± 0.12	0.83 ± 0.07	29%	chemical input
furosemide	<0.33	1.67 ± 0.33	1.43 ± 0.38	1.10 ± 0.21	>a factor of 4	all 3 factors
gemfibrozil	0.56 ± 0.05	0.32 ± 0.03	0.31 ± 0.03	0.34 ± 0.01	81%	chemical input
hydrochlorothiazide	<0.21	2.24 ± 0.21	2.24 ± 0.25	1.70 ± 0.20	>a factor of 10	persistence
ibuprofen	<0.52	<0.52	<0.52	1.44 ± 0.18[c]		
ketoprofen	<0.14	<0.14	<0.14	<0.14		
sulfamethoxazole	0.34[c] ± 0.03	0.38 ± 0.06	0.44 ± 0.08	0.35 ± 0.05	>29%	dilution

[a]$n = 8$ for late spring; $n = 4$ for late autumn; $n = 5$ for early winter; and $n = 6$ for mid-winter. [b]The seasonality was calculated by (Max − Min)/Min. [c]Concentration was between MDL and MQL (method quantification limit).

Sampling Campaign. The sampling campaign was designed to evaluate the persistence of the chemicals during three pronounced seasons typical for this site: late spring (characterized by high irradiation and high water temperatures), later autumn (low irradiation, low water temperatures), and winter (ice covered). To this end, samples of inflowing water from Vättern and effluent from the Karshult WWTP (the two major sources of chemical inputs to Boren) as well as outflowing water were collected. The effluent sampling is described below. For the late spring period, the inflowing water from Vättern and the outflowing water were sampled on June 12. In anticipation of the formation of ice, the sampling for the late autumn period was conducted on Dec. 9. After no ice was formed, further lake water samples were collected on Jan. 4 and Feb. 17. This resulted in the four study time periods that we call late spring (2013/04/15−06/09), late autumn (2014/11/04−12/01), early winter (2013/12/02−2014/01/05), and mid-winter (2014/01/06−02/16). Inflowing and outflowing water was collected from a depth of 20 cm below the surface.

Additional sampling was conducted to test the well-mixed and steady state assumptions. The well-mixed assumption was tested by collecting lake water samples at seven locations within the lake (Boren) and at the outlet of the lake on June 12, 2013 (Figure 1). Samples were collected in hand-held sampling bottles at a depth of 20 cm from a moving canoe. The steady state assumption was tested by collecting samples from WWTP effluent and Vättern water over extended periods of time. Seven day flow-proportional wastewater effluent samples were collected continuously from Apr. 15 to June 9, 2013 and from Nov. 4, 2013 to Feb. 16, 2014. Vättern water was sampled from the inlet to the Motala town waterworks (located 3 km from the entrance of the short river flowing from Vättern to Boren) once a week between Dec. 7, 2013 and Feb. 16, 2014.

A flow-proportional sampling system that is permanently installed at the WWTP was used to take effluent samples. The effluent in the sampler was kept at 2 °C. Every 24 h, the effluent was transferred to a container and frozen. Aliquots of these 24 h samples were combined to give a flow-proportional sample for each week. All wastewater and lake water samples were collected in HDPE bottles, transported to the laboratory, and stored in the freezer at −28 °C until analysis.

Analytical Methods. The methods employed in the Norra Bergundasjön study were used with some modifications to extract the lake water and wastewater samples.[6] In summary, after the addition of 3 mL of an isotope-substituted internal standard solution, 1.5 L of lake water was vacuum filtered, pH adjusted to 3, and extracted on SPE cartridges (see Table S2, Supporting Information). The 6 mL cartridges were packed with three sorbents (from top to bottom: Oasis MAX/Oasis HLB/Isolute ENV+, 120 mg/220 mg/140 mg, respectively). After extraction, the cartridges were eluted with 7 mL of methanol, 7 mL of 2% NH$_4$OH in methanol, and 7 mL of 2% formic acid in methanol. The combined eluates were evaporated to dryness under a nitrogen stream. The final extract was reconstituted in 150 μL of water/acetonitrile (80/20, v/v, 10 mM acetic acid). For wastewater samples, 50 mL was filtered after the addition of 100 μL of an isotope-substituted internal standard solution (see Table S2, Supporting Information). The pH was adjusted to 3, and Oasis HLB cartridges (60 mg/3 mL) were used for extraction. The cartridges were eluted with methanol, and after drying, the eluate was finally reconstituted in 150 μL of water/acetonitrile (80/20, v/v, 10 mM acetic acid).

The extracts were analyzed on UHPLC/MS/MS (ACQUITY UPLC System with Xevo TQ-S mass spectrometer from Waters) using an ACQUITY UPLC HSS T3 column (1.8 μM, 2.1 × 100 mm, Waters). The mobile phase, separation gradient program, MS/MS MRM transitions, and other mass spectrometer parameters were the same as in our previous work[6] except for acetaminophen and ibuprofen (see Table S3 in the Supporting Information).

Quality Control. All wastewater and lake water samples (except Vättern water samples taken from the inlet to the Motala town waterworks) were analyzed in triplicate. The recoveries of the chemicals in the lake water were measured by analyzing lake water sampled on Feb. 21, 2013 that had been spiked with the target chemicals (see Text S2, Supporting Information). For every 6 samples, spiked Milli-Q water was used as a control sample and nonspiked Milli-Q water was used as a method blank, following the same procedure as for the lake water samples. A field blank for lake water sampling was prepared by pouring Milli-Q water into a sampling bottle in the
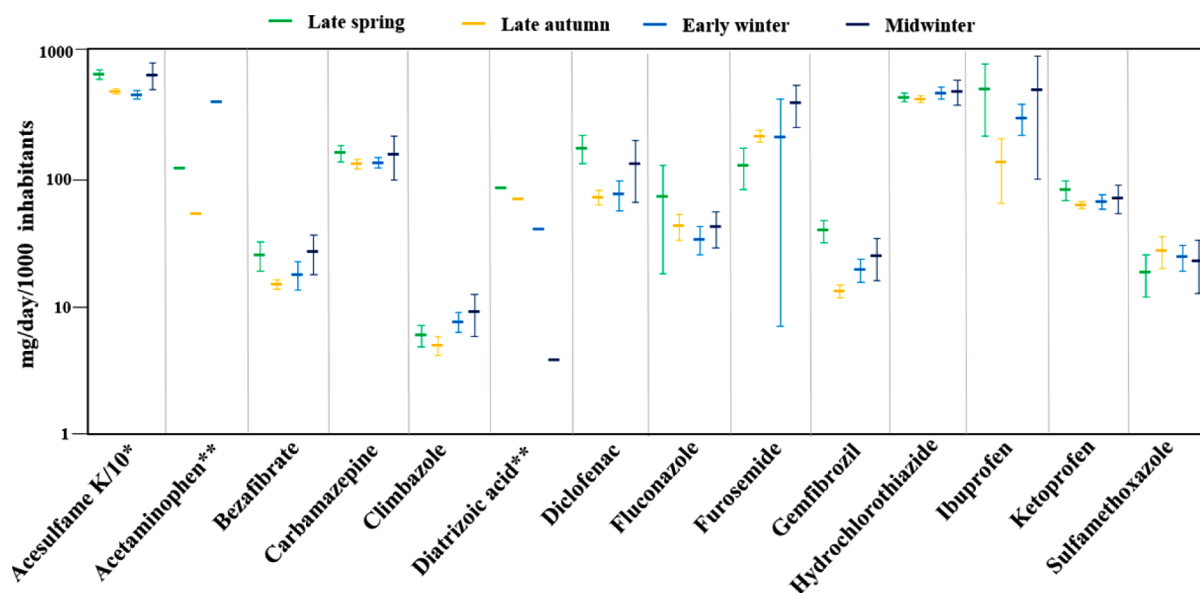
Figure 2. Chemical emissions from the Karshult WWTP during different periods (mg/day/1000 inhabitants); $n = 8$ for late spring samples (green); $n = 4$ for late autumn samples (yellow); $n = 5$ for early winter samples (light blue); $n = 6$ for midwinter samples (dark blue). The symbols and error bars refer to the mean and the standard deviation, respectively. *Note that the unit for acesulfame K is mg/day/100 inhabitants. **Due to the large standard deviation for acetaminophen and diatrizoic acid, their error bars are not shown in the plot; for acetaminophen, the emission in midwinter is off the scale (1380 mg/day/1000 inhabitants).

field. The method detection limit (MDL) and method quantification limit (MQL) were defined as signal-to-noise ratios of 3 and 10, respectively.

RESULTS AND DISCUSSION

Quality Control. The recoveries of the 14 chemicals in lake water samples were between 50% and 105% except for acesulfame K (10%) and diatrizoic acid (5%) (Figure S3, Supporting Information). An isotope-substituted internal standard was used for every analyte, correcting for recovery. The triplicate analysis showed a relative standard deviation (RSD) of <20% for 319 of 322 wastewater data points and 94 of 96 lake water data points (including the inflowing water from Vättern, the lake outlet, and transect water samples) above the MQL. Despite the low recovery of acesulfame K, the use of an isotope substituted internal standard resulted in excellent method precision, with an average RSD of the triplicate analyses of the lake water samples of 4% (see Table 1). In the control samples, the RSD was <10% for all analytes. Method blanks and field blanks were all below the MDL.

Seasonality in Emissions from the WWTP. All 14 analytes were quantified in the effluent from the Karshult WWTP, with mean concentrations ranging from 57.6 ng L^{-1} for bezafibrate to 14 900 ng L^{-1} for acesulfame K (Tables S4–S7 in the Supporting Information). The concentrations were generally consistent with concentrations measured in other European WWTPs (see Text S3 and Table S4, Supporting Information).

The emissions during four time periods are illustrated in Figure 2. In a given time period, the weekly emissions of most chemicals had a RSD <36% (see error bars in Figure 2 and Tables S8–S10, Supporting Information), indicating relatively constant emissions within a period. The exceptions were fluconazole (75% in late spring), sulfamethoxazole (44% in midwinter), diclofenac (50% in midwinter), furosemide (97% in early winter), and ibuprofen (>57% in late spring, late

autumn, and midwinter). Furthermore, both acetaminophen and diatrizoic acid had a RSD >100% during all periods. Diatrizoic acid is an X-ray contrast agent for which a single dose can be up to 200 g,[9] so the weekly emissions from the Karshult WWTP could have originated from the treatment of a single individual. Emissions of this chemical can thus be expected to be highly variable for small- and medium-sized WWTPs. The variability in acetaminophen emissions may reflect a high week-to-week variability in demand for its pain killing and fever reducing effects. The peak emission of acetaminophen in mid-February corresponded with the peak in the incidence of influenza in this area of Sweden during 2013–2014.[10] This high variability indicates that long-term integrated sampling is needed to obtain accurate estimates of the acetaminophen loading to WWTP recipients.

Seasonal differences in emissions were most prevalent between late spring and late autumn. A significant difference was observed for 8 chemicals ($p < 0.05$ in Table S11, Supporting Information). Bezafibrate, gemfibrozil, carbamazepine, ketoprofen, diclofenac, acesulfame K, and ibuprofen had lower emissions in late autumn than in late spring, while furosemide had higher emissions in late autumn. The biggest differences in the mean emissions between late spring and late autumn were found for gemfibrozil, diclofenac, and furosemide (73%, 61%, and 62%, respectively), while for the others it was <50%. The differences in the emissions between other pairs of periods were not significant for most chemicals (Table S11, Supporting Information). A comparison of the seasonality in emissions from Karshult with literature reports is provided in Text S3, Supporting Information.

The mean emissions for the time periods ranged over less than a factor of 2 for 8 of the chemicals. This indicates that for these 8 chemicals the seasonality in WWTP emissions only has a modest impact on the seasonality in contaminant levels in water. For acetaminophen, ibuprofen, and diatrizoic acid, the range in mean period emission was considerably larger, but the
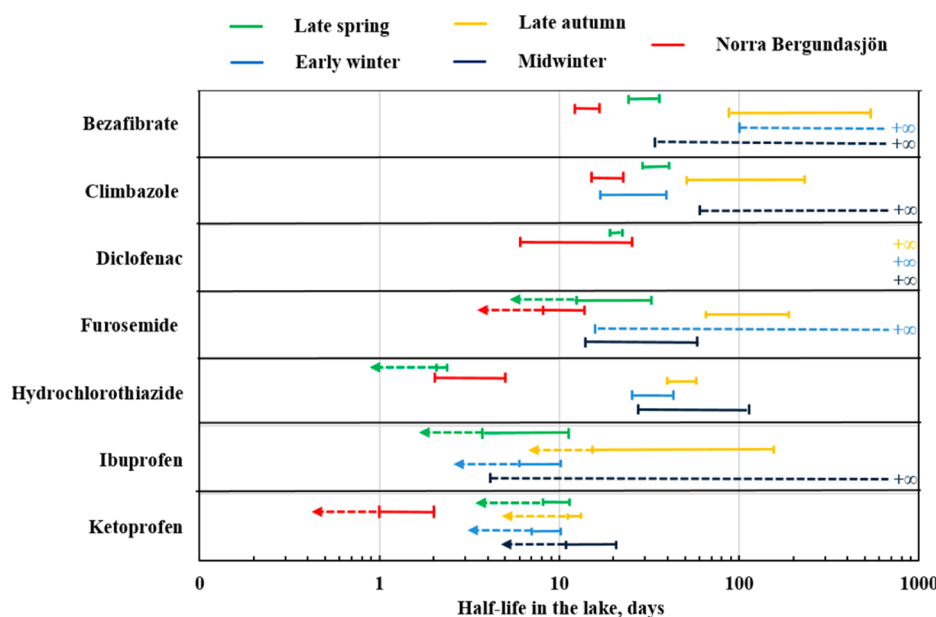
Figure 3. Estimated half-lives $t_{0.5R}$ of seven pharmaceuticals during the 4 study time periods in Boren and during spring in another Swedish lake (Norra Bergundasjön; Zou et al. 2015).[6] The segment ⊢—⊣ refers to the 95% confidence interval for $t_{0.5R}$ (green for late spring, yellow for late autumn, light blue for early winter, and dark blue for midwinter; red for Norra Bergundasjön in late spring). For the chemicals for which the concentration in the outlet of the lake was < MDL, the 95% confidence interval of an upper boundary of $t_{0.5R}$ (calculated using the MDL as the concentration in the outlet) is shown as ⊢—⊣, together with a dotted arrow indicating that the lower boundary of the half-life is unknown. For chemicals for which $t_{0.5R}$ calculated with eq 1 was negative, it is shown as "+∞". See the text for more details.

differences were not significant ($p > 0.05$) due to high variability within the periods.

Seasonality in the Flow/Dilution. After emission from the WWTP, the chemicals were diluted in the lake. The extent of the dilution is inversely proportional to the residence time of the water in the lake. As described in the Methods section, the flow rate of water into and out of the lake was 40−50 m³ s⁻¹ in late spring and winter but lower in late autumn (∼25 m³ s⁻¹). In other words, the dilution was nearly twice as high in late spring and winter as in late autumn (Figure S1, Supporting Information). This means that for persistent chemicals with the same emission the concentration in the lake would be 60−90% higher in late autumn than during other time periods.

Seasonality in Persistence. *Well-Mixed and Steady-State Assumptions.* When calculating chemical persistence in a lake with eq 1, it is assumed that the water body is well-mixed and that the chemicals are near steady state. The well-mixed assumption can be assessed using the spatial distribution of the chemicals in the lake. On June 12, 2013, seven lake water samples were taken on a transect along the lake in addition to the sample at the lake outlet. The concentrations of the persistent chemicals (acesulfame K, carbamazepine, flucona-zole, gemfibrozil, and sulfamethoxazole; see below) were relatively homogeneous throughout the lake (RSD <23%, Table S12, Supporting Information). This indicates that the WWTP effluent was mixed throughout the whole lake, i.e., that there was no strong preferential flow pathway for the effluent out of the lake. However, homogeneous concentrations of persistent chemicals are not sufficient evidence of a well-mixed situation; this is also consistent with a plug flow model of the lake's hydrology. On the other hand, homogeneous concen-trations of more labile substances for which the WWTP was a strong source demonstrate a well-mixed state. Bezafibrate showed a mass loss of ∼40% in the lake (Figure S4 in the Supporting Information) and had concentrations well above

the MDL (Table S12, Supporting Information). The RSD of the bezafibrate concentration after the WWTP (i.e., after sampling point 2) was 8%, which is comparable to the persistent chemicals. There was no trend of decreasing concentrations after the WWTP, indicating that a well-mixed model is more appropriate than a plug flow model of water flow through the lake. The transformation half-life of bezafibrate in the lake ($t_{0.5R}$) was estimated to be 29 days. Thus, the 8% RSD corresponds to a difference in residence time of 3.5 days, which is just 10% of the hydraulic residence time (36 days). This assessment of spatial distribution indicates that the well-mixed assumption is a reasonable approximation.

To assess the steady-state assumption, we studied the temporal variation of the two major chemical inputs into Boren: via the effluent from Karshult WWTP (weekly flow proportional samples taken in late spring, late autumn, and early and midwinter) and via the inflowing water from Vättern (weekly grab samples taken from the inlet to the Motala town waterworks during winter). The temporal variability of the chemical discharge from the WWTP was discussed above and illustrated in Figure 2. With the exception of acetaminophen and diatrizoic acid, the temporal variability on a weekly time scale, expressed as RSD, was <36% in most cases. Since the variability was high for diatrizoic acid and acetaminophen, they were not included in the persistence assessment. Variations in WWTP input on the time scale of days will have a small influence on the persistence determination given the water residence time of 1−2 months. The RSD of the concentrations of four of the five chemicals detected in Vättern water collected in winter (weekly samples for 10 weeks) was <28%, while for the fifth chemical (diclofenac) it was 81% (Table S13, Supporting Information). This indicates that the mass flows of chemicals from Vättern were relatively constant on a seasonal time scale, which was anticipated given the long hydraulic residence time of Vättern and the absence of

population centers or other known chemical sources to Vättern in the vicinity of its outlet. The appropriateness of the steady state assumption was verified by comparing the ratio of the concentrations of the test chemical and the benchmark chemical in Boren predicted using eq 1 and a nonsteady state model. The difference was ≤10%, confirming that the steady state assumption was a reasonable approximation (Text S4, Supporting Information).

Chemical Persistence Determined by the Benchmarking Approach. In previous work, acesulfame K was shown to be conservative in a Swedish lake with a hydraulic residence time of 121 days, and it was used as the benchmark chemical to quantify persistence.[6] In the current study, acesulfame K also had a closed mass balance during all 4 periods (Figure S4 in the Supporting Information), so it was again used as the benchmark chemical. Four other chemicals (carbamazepine, fluconazole, gemfibrozil, and sulfamethoxazole) also had a closed mass balance in the lake (Figure S4, Tables S8–S10 and S14–S15, Supporting Information), whereby the mass balance was uncertain for sulfamethoxazole because its concentration was <MQL in the Vättern samples. The persistence of the remaining seven pharmaceuticals ($t_{0.5R}$) was estimated for each period using the benchmarking approach according to eq 1 (see Text S5 in the Supporting Information for details of the calculations).

The $t_{0.5R}$ of the seven pharmaceuticals ranged from several days to several hundred days (Figure 3 and Table S16, Supporting Information). For chemicals that were detected in the outlet water from Boren but not in Vättern water, 0 and MDL for Vättern water were used to calculate the upper and the lower boundaries, respectively, of the central estimate of $t_{0.5R}$ (see Text S6, Supporting Information). For chemicals that were detected neither in the outlet water from Boren nor in Vättern water, the upper boundary of the central estimate of $t_{0.5R}$ was calculated using the MDL for the outlet water from Boren together with 0 for Vättern water; no lower boundary could be obtained in these cases. The 95% confidence interval of the central estimates of $t_{0.5R}$ was calculated on the basis of the variability in the weekly mass flows of the test and the benchmark chemicals in the WWTP effluent. First, the 95% confidence interval of k'_R was calculated, and then, the 95% confidence interval of $t_{0.5R}$ was obtained by dividing ln(2) by the 95% confidence interval boundaries of k'_R (see Text S6 in the Supporting Information for details). When a negative value was obtained from the calculations for the upper boundary of $t_{0.5R}$, positive infinity ($+\infty$) was used. In those cases where the central estimate of $t_{0.5R}$ was a range (delineated by the upper and lower boundaries, see above), the 95% confidence interval of the end points of the range was calculated. For most of the chemicals, the 95% confidence interval of $t_{0.5R}$ was less than 1 order of magnitude (Figure 3).

We measured the persistence of ten of the chemicals studied here in another Swedish lake (Norra Bergundasjön) during the late spring of 2013.[6] Somewhat longer half-lives were observed in Boren for bezafibrate (30–66% longer) and climbazole (21–63% longer). For diclofenac, furosemide, hydrochlorothiaxide, and ketoprofen, the 95% confidence intervals for $t_{0.5R}$ overlapped (Figure 3).

Norra Bergundasjön is a hypertrophic lake with very strong algal blooms during spring and summer, whereas Boren is classified as noneutrophied with good nutrient status.[11] The higher biological activity in Norra Bergundasjön could contribute to enhanced biodegradation of xenobiotics com-

pared to Boren. On the other hand, the intense algal blooms in Norra Bergundasjön will limit light penetration, so that photodegradation is expected to be lower in Norra Bergundasjön despite its somewhat shallower depth (3.3 m vs 5.4 m). Bezafibrate is subject to biodegradation in the environment.[12−14] Although laboratory studies have shown that it can be degraded photolytically,[15] we found no work showing that photodegradation is significant under environmental conditions. The shorter $t_{0.5R}$ of bezafibrate in Norra Bergundasjön could be due to more rapid biotransformation in that system. As for climbazole, no published data on the environmental transformation of this compound was found. The removal efficiency of climbazole in a WWTP was 30–51%,[16] which might have involved microbial degradation or sorption to the activated sludge. Photodegradation of climbazole at different pH values under laboratory conditions was reported,[17] so photodegradation cannot be ruled out either.

Evaluation of Seasonality. For bezafibrate, climbazole, diclofenac, furosemide, and hydrochlorothiazide, $t_{0.5R}$ was longer in late autumn than in late spring (Figure 3). The lower water temperature in late autumn than in late spring (mean of 5.1 °C in late autumn versus 10.6 °C in late spring) could have resulted in lower microbial activity and slower biotransformation in late autumn. This could explain the longer $t_{0.5R}$ for bezafibrate and climbazole, for which biotransformation is suspected to be a major loss process (see above). The same explanation could apply to furosemide, which is biodegraded in sediment-water biodegradation tests.[18]

The largest seasonal difference was observed for hydrochlorothiazide, for which the upper boundary of the 95% confidence interval of $t_{0.5R}$ in late spring was 20 times lower than the lower boundary in late autumn. This very strong seasonality in persistence points to phototransformation as the dominant removal process, since there is a strong seasonality in irradiance at this site (see above), while seasonality of this magnitude is unlikely for biodegradation or hydrolysis. Furthermore, low removal of hydrochlorothiazide in WWTPs[19,20] suggests that biotransformation of this compound is slow. In water-sediment biodegradation tests, the concentration of hydrochlorothiazide decreased in all assays including the sterile and water-only controls,[18] suggesting that nonphotolytic abiotic transformation (e.g., hydrolysis) was more important than biodegradation. However, phototransformation of hydrochlorothiazide has been reported, and in a laboratory study, phototransformation was found to be more rapid than transformation in dark controls.[21,22] In summary, these laboratory studies are consistent with the conclusion that hydrochlorothiazide was removed from Lake Boren primarily via photodegradation.

Diclofenac has also been shown to be mainly degraded by photolysis.[3,23−25] Our results were consistent with this; no degradation was observed during the late autumn, early winter, or midwinter periods, while $t_{0.5R}$ was 19−23 days during spring. For comparison purposes, the photolysis half-life of diclofenac at 60° N during late spring was calculated using a reprogrammed version of the GCSOLAR model.[26] Assuming a quantum yield of 0.15 and clear skies, a half-life of 11 days was obtained. While the uncertainty in this estimate is considerable (e.g., due to the clear sky assumption and uncertainty in the quantum yield, which ranges from 0.04 to 0.22 in the literature),[27−29] it does indicate that the laboratory based understanding of the photolysis of diclofenac is broadly consistent with the field observations from this study.

Phototransformation could also have contributed to the shorter $t_{0.5R}$ in late spring for the other 4 chemicals, as laboratory studies have shown that they are also subject to phototransformation (see above for bezafibrate and climbazole and refs 30 and 31 for furosemide). However, the much lower seasonality in $t_{0.5R}$ indicates that phototransformation was not the dominant degradation process. For the remaining two chemicals in Figure 3 (ketoprofen and ibuprofen), only the maximum $t_{0.5R}$ could be estimated, so it was not possible to assess differences in $t_{0.5R}$ between periods.

During the late autumn, early winter, and midwinter periods, the 95% confidence intervals for $t_{0.5R}$ overlapped for almost all compounds (Figure 3), i.e., no seasonal differences in persistence could be identified. The exceptions were climbazole, which had a shorter $t_{0.5R}$ in early winter (December) than in late autumn (November) and in midwinter (January and February), and furosemide, which had a shorter $t_{0.5R}$ in midwinter than in late autumn. We have no explanations for these observations. We note that ice typically forms on Boren during early winter. We designed the study to assess whether the presence of ice would affect the persistence of the chemicals. However, the winter of 2013/2014 was unusually warm and no ice formed. Consequently, the environmental conditions during these periods were similar.

Seasonality in Exposure in the Lake Ecosystem. The seasonality of the chemical concentrations (and hence the exposure of organisms) in the lake ecosystem can now be examined in light of the seasonality in chemical inputs, dilution, and persistence. The seasonal variability of chemical exposure in Boren varied widely from almost no variability for acesulfame K to more than a factor of 10 for hydrochlorothiazide (Table 1).

For those chemicals for which no degradation in Boren was observed (acesulfame K, carbamazepine, fluconazole, and gemfibrozil), the seasonal variability of chemical exposure in the lake system ranged from 28% for acesulfame K to 81% for gemfibrozil. For acesulfame K and carbamazepine, the highest concentrations were measured in late autumn and early winter. As discussed above, the emissions of acesulfame K and carbamazepine from Karshult WWTP decreased from late spring to late autumn. The input from Vättern also decreased (Tables S14 and S17, Supporting Information), so the total load into the lake was ~40% lower in late autumn than in late spring. However, the concentration in late autumn was slightly higher (Table 1) due to less dilution in late autumn. For fluconazole and gemfibrozil, the highest concentrations were measured in late spring. This was a result of higher inputs from both the Karshult WWTP and Vättern in late spring, which were so much higher that they were not neutralized by the higher dilution in late spring. For acesulfame K and carbamazepine, the seasonality in chemical exposure was dominated by the seasonality in dilution (flow), while for fluconazole and gemfibrozil, the seasonality in chemical exposure was dominated by the seasonality in emissions and chemical inflow from Vättern.

Hydrochlorothiazide is the chemical with the greatest seasonality in chemical exposure, with concentrations in late spring that are more than an order of magnitude lower than in the other periods. In this case, the contributions of seasonality in chemical inputs and dilution were negligible. The strong seasonality in chemical persistence, with $t_{0.5R}$ being an order of magnitude shorter in late spring than in the other periods

(Figure 3), was almost completely responsible for the strong seasonality in exposure.

A similar seasonality in chemical persistence was observed for diclofenac, for which $t_{0.5R}$ was also markedly shorter in late spring than in the other periods. However, the seasonality in chemical exposure in Boren showed the opposite trend. A strong seasonality in inputs from Vättern resulted in a 6 times higher total input of diclofenac to Boren in late spring than in late autumn. Although the chemical persistence was markedly shorter in the late spring, this could not fully compensate for the higher chemical input. This was because the persistence was still long compared with the residence time of the water in the lake (36 days). Even though the elimination of the chemical was faster in late spring, the amount removed was still comparatively small compared with the chemical input. This illustrates that the seasonality in persistence does not by itself give an indication of expected seasonality in chemical levels in the environment. The relative magnitudes of $t_{0.5R}$ and the hydraulic residence time of the system must also be considered.

The seasonality in chemical exposure for bezafibrate and climbazole was analogous to that for diclofenac. Despite having a markedly shorter chemical persistence in late spring than in late autumn, the concentrations of bezafibrate and climbazole in Boren were higher in late spring. The seasonality in exposure was dominated by the seasonality in chemical inputs instead of seasonality in chemical persistence because $t_{0.5R}$ in late spring (24–35 days for bezafibrate; 29–41 days for climbazole) was similar to the water residence time.

For furosemide, the chemical exposure was higher in the late autumn than in the late spring (Table 1). This was due to mutually supportive seasonal trends in all 3 factors: higher emissions, lower dilution, and longer persistence in late autumn.

The exposure of aquatic life to xenobiotics is directly related to the xenobiotic concentration in water to which the organisms are exposed. In Boren, the exposure to the selected xenobiotics varied seasonally, whereby the strongest variability was caused by variability in persistence (i.e., for hydrochlorothiazide). Clearly, persistence varies in time. The comparison of persistence in Boren and Norra Bergundasjön during the same season showed that it also varies in space. More field measurements are required if we are to further develop our understanding of the factors governing the spatial and temporal variability of chemical persistence and better link our theoretical concepts and laboratory observations to the reality in the environment.

■ ASSOCIATED CONTENT

Ⓢ Supporting Information

Chemicals, recoveries of the 14 chemicals from spiked lake water, comparison of the chemical concentrations in WWTP effluent with literature data, influence of the variability in the chemical loading from the WWTP and in water flow rate on the persistence estimation, persistence calculation by benchmarking approach, uncertainty analysis of the persistence determination, and additional figures and tables. The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.est.5b01720.

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■ **REFERENCES**

(1) Boethling, R.; Fenner, K.; Howard, P.; Klečka, G.; Madsen, T.; Snape, J. R.; Whelan, M. J. Environmental persistence of organic pollutants: guidance for development and review of POP risk profiles. *Integr. Environ. Assess. Manage.* **2009**, *5* (4), 539−556.

(2) Persson, L. M.; Breitholtz, M.; Cousins, I. T.; de Wit, C. A.; MacLeod, M.; McLachlan, M. S. Confronting unknown planetary boundary threats from chemical pollution. *Environ. Sci. Technol.* **2013**, *47* (22), 12619−12622.

(3) Tixier, C.; Singer, H. P.; Oellers, S.; Müller, S. R. Occurrence and fate of carbamazepine, clofibric acid, diclofenac, ibuprofen, ketoprofen, and naproxen in surface waters. *Environ. Sci. Technol.* **2003**, *37* (6), 1061−1068.

(4) Stoll, J. A.; Giger, W. Mass balance for detergent-derived fluorescent whitening agents in surface waters of Switzerland. *Water Res.* **1998**, *32* (7), 2041−2050.

(5) Zou, H.; MacLeod, M.; McLachlan, M. S. Evaluation of the potential of benchmarking to facilitate the measurement of chemical persistence in lakes. *Chemosphere* **2014**, *95*, 301−309.

(6) Zou, H.; Radke, M.; Kierkegaard, A.; MacLeod, M.; McLachlan, M. S. Using chemical benchmarking to determine the persistence of chemicals in a Swedish lake. *Environ. Sci. Technol.* **2015**, *49* (3), 1646−1653.

(7) Motalastream Water Management; http://www.motalastrom.org/ (accessed October 31, 2014).

(8) SMHI (Swedish Meteological and Hydrological Institue); http://www.smhi.se/ (accessed October 31, 2014).

(9) Pérez, S.; Barceló, D. Fate and occurrence of X-ray contrast media in the environment. *Anal. Bioanal. Chem.* **2007**, *387* (4), 1235−1246.

(10) Public Health Agency of Sweden. *Influenza in Sweden: Season 2013−2014;* http://www.folkhalsomyndigheten.se/pagefiles/18481/Influenza-in-Sweden-2013-2014.pdf.

(11) VISS (VattenInformationSystem Sverige); http://www.viss.lansstyrelsen.se/ (accessed October 31, 2014).

(12) Quintana, J. B.; Weiss, S.; Reemtsma, T. Pathways and metabolites of microbial degradation of selected acidic pharmaceutical and their occurrence in municipal wastewater treated by a membrane bioreactor. *Water Res.* **2005**, *39* (12), 2654−2664.

(13) Lewandowski, J.; Putschew, A.; Schwesig, D.; Neumann, C.; Radke, M. Fate of organic micropollutants in the hyporheic zone of a eutrophic lowland stream: results of a preliminary field study. *Sci. Total Environ.* **2011**, *409* (10), 1824−1835.

(14) Kunkel, U.; Radke, M. Biodegradation of acidic pharmaceuticals in bed sediments: insight from a laboratory experiment. *Environ. Sci. Technol.* **2008**, *42* (19), 7273−7279.

(15) Fatta-Kassinos, D.; Vasquez, M. I.; Kümmerer, K. Transformation products of pharmaceuticals in surface waters and wastewater formed during photolysis and advanced oxidation processes - degradation, elucidation of byproducts and assessment of their biological potency. *Chemosphere* **2011**, *85* (5), 693−709.

(16) Qi, W.; Singer, H.; Berg, M.; Müller, B.; Pernet-Coudrier, B.; Liu, H.; Qu, J. Elimination of polar micropollutants and anthropogenic markers by wastewater treatment in Beijing, China. *Chemosphere* **2015**, *119*, 1054−1061.

(17) Couteau, C.; Jadaud, M.; Peigne, F.; Coiffard, L. J. M. Influence of pH on the photodegradation kinetics under UV light of climbazole solutions. *Analusis* **2000**, *28*, 557−560.

(18) Radke, M.; Maier, M. P. Lessons learned from water/sediment-testing of pharmaceuticals. *Water Res.* **2014**, *55*, 63−73.

(19) Radjenović, J.; Petrović, M.; Barceló, D. Fate and distribution of pharmaceuticals in wastewater and sewage sludge of the conventional activated sludge (CAS) and advanced membrane bioreactor (MBR) treatment. *Water Res.* **2009**, *43* (3), 831−841.

(20) Bueno, M. J.; Gomez, M. J.; Herrera, S.; Hernando, M. D.; Agüera, A.; Fernández-Alba, A. R. Occurrence and persistence of organic emerging contaminants and priority pollutants in five sewage treatment plants of Spain: two years pilot survey monitoring. *Environ. Pollut.* **2012**, *164*, 267−273.

(21) Brigante, M.; DellaGreca, M.; Previtera, L.; Rubino, M.; Temussi, F. Degradation of hydrochlorothiazide in water. *Environ. Chem. Lett.* **2005**, *2* (4), 195−198.

(22) Boreen, A. L.; Arnold, W. A.; McNeill, K. Photodegradation of pharmaceuticals in the aquatic environment: a review. *Aquat. Sci.* **2003**, *65* (4), 320−341.

(23) Poiger, T.; Buser, H. R.; Müller, M. D. Photodegradation of the pharmaceutical drug diclofenac in a lake: pathway, field measurements, and mathematical modeling. *Environ. Toxicol. Chem.* **2001**, *20* (2), 256−263.

(24) Radke, M.; Ulrich, H.; Wurm, C.; Kunkel, U. Dynamics and attenuation of acidic pharmaceuticals along a river stretch. *Environ. Sci. Technol.* **2010**, *44* (8), 2968−2974.

(25) Buser, H. R.; Poiger, T.; Müller, M. D. Occurrence and fate of the pharmaceutical drug diclofenac in surface waters: rapid photodegradation in a lake. *Environ. Sci. Technol.* **1998**, *32* (22), 3449−3456.

(26) Zepp, R. G.; Cline, D. M. Rates of Direct Photolysis in Aquatic Environment. *Environ. Sci. Technol.* **1977**, *11* (4), 359−366.

(27) Poiger, T.; Buser, H. R.; Müller, M. D. Photodegradation of the pharmaceutical drug diclofenac in a lake: Pathway, field measurements, and mathematical modeling. *Environ. Toxicol. Chem.* **2001**, *20* (2), 256−263.

(28) Packer, J. L.; Werner, J. J.; Latch, D. E.; McNeill, K.; Arnold, W. A. Photochemical fate of pharmaceuticals in the environment:Naproxen, diclofenac, clofibric acid, and ibuprofen. *Aquat. Sci.* **2003**, *65* (4), 342−351.

(29) Andreozzi, R.; Raffaele, M.; Nicklas, P. Pharmaceuticals in STP effluents and their solar photodegradation in aquatic environment. *Chemosphere* **2003**, *50* (10), 1319−1330.

(30) Jakimska, A.; Śliwka-Kaszyńska, M.; Reszczyńska, J.; Namieśnik, J.; Kot-Wasik, A. Elucidation of transformation pathway of ketoprofen, ibuprofen, and furosemide in surface water and their occurrence in the aqueous environment using UHPLC-QTOF-MS. *Anal. Bioanal. Chem.* **2014**, *406* (15), 3667−3680.

(31) Isidori, M.; Nardelli, A.; Parrella, A.; Pascarella, L.; Previtera, L. A multispecies study to assess the toxic and genotoxic effect of pharmaceuticals: furosemide and its photoproduct. *Chemosphere* **2006**, *63* (5), 785−793.