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Simultaneous Determination and Occurrences of Pharmaceuticals by Solid-Phase Extraction and Liquid Chromatography-Tandem Mass Spectrometry (LC-MS/MS) in Environmental Aqueous Samples

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Pharmaceuticals and personal care products (PPCPs) are emerging contaminants in the aquatic environment. Many pharmaceuticals are not completely removed during wastewater treatment, leading to their presence in wastewater treatment effluents, rivers, lakes, and ground water. Here, we developed analytical methods for monitoring ten pharmaceuticals from surface water by LC/ESI-MS/MS. For sample clean-up and extraction, MCX (mixed cation exchange) and HLB (hydrophilic-lipophilic balance) solid-phase extraction (SPE) cartridges were used. The limits of detection (LOD) in distilled water and the blank surface water were in the range of 0.006 - 0.65 and 1.66 - 45.05 pg/mL, respectively. The limits of quantitation (LOQ) for the distilled water and the blank surface water were in the range of 0.02 - 2.17 and 5.52 - 150.15 pg/mL, respectively. The absolute recoveries for fortified water samples were between 62.1% and 125.4%. Intra-day precision and accuracy for the blank surface water were 2.9% - 24.1% (R.S.D.) and -16.3% - 16.3% (bias), respectively. In surface wastewater near rivers, chlortetracycline and acetylsalicylic acid were detected frequently in the range of 0.017 - 5.404 and 0.029 - 0.269 ng/mL, respectively. Surface water near rivers had higher levels than surface water of domestic treatment plants.

Key Words: Pharmaceuticals and personal care products, LC/MS/MS, Surface water

Introduction

The fate and effects of pharmaceuticals and personal care products (PPCPs) and natural estrogens entering the environment has gained increasing attention. PPCPs include non-prescription and prescription pharmaceuticals for human and veterinary use, and the active and inert ingredients in personal care products.[1] PPCPs and EDCs (Endocrine disrupting chemicals) and their metabolites are continuously introduced into aquatic environments via sewage treatment plant effluent and/or agricultural runoff.[2] Animal waste in pasturage or confined animal-feeding operations is another potential source of pharmaceuticals. Direct discharge to the ground from livestock excretion can contaminate surface water and can infiltrate ground water.[3] Approximately 3000 different active substances are used in human and veterinary medicine,[4] so their detection in environmental waters is of significant interest.

Thus, there has been growing interest in measuring emerging contaminants that are not currently covered by existing regulations on water quality, but that may be candidates for future regulation, depending on their toxicity and potential effects in the environment and on human health.[5] PPCPs can originate from human usage and excretions and veterinary applications of a variety of products such as over-the-counter and prescription medications and fungicides and disinfectants used for industrial, domestic, agricultural, and livestock practices.[1] The presence of pharmaceuticals in aquatic environments typically results from human excretion of metabolized or parent drug passing into sewage or septic systems and subsequent discharge of wastewater and percolation of septic-system leachate into surface water or ground water.[6]

The concentrations of pharmaceuticals found in surface water downstream from sewage treatment plant effluents are generally in the ng/L range and do not necessarily represent a serious threat to drinking water quality. Unfortunately, the consequences of continuous exposure to low concentrations of pharmaceuticals in the ecosystem are still not fully known. A discussion of various aspects of ecotoxicology of pharmaceuticals in the environment can be found in recent reviews.[7,8,9]

Environmental risk assessment must be based on reliable data about the actual concentrations of pharmaceuticals in aquatic systems. Therefore, efficient analytical methods are of major importance.[10]

The quantitative analysis of antibiotics in the aqueous environment is difficult because of the fact that antibiotics exist in low concentration levels and the matrices are complicated and what's more, physico-chemical properties of antibiotics are diverse. Consequently, a highly sensitive and selective analytical method is crucially needed to monitor antibiotics in the aqueous environment.

The simultaneous determination of different analytes requires chromatographic techniques.[5] Because of the recent awareness of the potentially dangerous consequences of the presence of pharmaceuticals in the environment, the analytical methodology for their determination in complex environmental matrices is still evolving, and the number of methods described in the literature has grown considerably.[11]

The development of analytical procedures for pharmaceutical residue analysis is facilitated by considerable experience in pesticide residue analysis. Common procedures include a pre-

Table 1. Classification of pharmaceuticals

Group	Pharmaceuticals	M.W.	pKa
Group-A	Acetaminophen	151.2	9.5
	Lincomycin	406.5	7.79
	Sulfamethazine	278.3	7.4
	Sulfamethoxazole	253.3	5.8
	Sulfathiazole	255.3	7.2
	Ttrimethoprim	290.3	6.6
Group-B	Chlortetracycline	478.9	3.33/7.55/9.33
	Oxytetracycline	460.4	3.22/7.46/8.94
	Ciprofloxacin	331.3	6.09/8.74
	Enrofloxacin	359.4	6.0/6.9

concentration and clean-up step by solid-phase extraction or related techniques, followed by chromatography in combination with mass spectrometry (MS) as a detector. For complex matrices, established sample preparation protocols may fail to allow sufficient sample clean-up to avoid ionization suppression in the atmospheric pressure ionization source of the MS detector.[10]

High-Performance liquid chromatography coupled to mass spectrometry (LC-MS), and particularly tandem mass spectrometry (LC-MS/MS), are the primary techniques used to determine ultra-trace concentrations of pharmaceuticals in wastewater, surface water, and ground water.[5,11,12,13,14,15]

Here, we have developed a sensitive and reliable analytical method for the simultaneous determination of ten pharmaceuticals in aquatic samples using a combined solid-phase extraction (SPE) isolation and clean-up, and LC/MS/MS with electrospray ionization (ESI) and multiple reaction monitoring (MRM). The limits of detection (LOD), limits of quantitation (LOQ), accuracy, and precision of the assay are described. The method was applied to target pharmaceuticals from surface water in South Korea. We selected the top ten pharmaceuticals prescribed in South Korea from 2000 to 2006,[16] both human and veterinary antibiotics (three sulfonamides (sulfamethazine, sulfamethoxazole, sulfathiazole), two tetracyclines (chlortetracycline, oxytetracycline), lincomycin, trimethoprim and two fluoroquinolones (enrofloxacin, ciprofloxacin)), as well as the analgesic/anti-inflammatory drugs, acetaminophen.

For chromatographic separation and mass spectrometric analysis, ten pharmaceuticals were classified into two groups (Table 1). We also describe the impact of matrix effects when measuring these compounds at environmentally relevant concentrations.

Experimental

Chemicals and standards. Sulfamethazine, sulfamethoxazole, sulfathiazole, lincomycin, acetaminophen, chlortetracycline, and oxytetracycline were purchased from Sigma-Aldrich Co. (St Louis, MO, USA) and trimethoprim, enrofloxacin and ciprofloxacin from Fluka Co. (Seelze, Germany), all pharmaceutical standards were of analytical grade and high purity (> 90%). The reference compounds, used as surrogate standards, sulfamethazine-6-^{13}C, ciprofloxacin-3-^{13}C and ibuprofen-6-^{13}C, and internal standard (terbutylazine) were purchased from Cambridge Isotope Laboratories Co. (Andover, MA, USA) and Fluka Co. (Seelze, Germany), respectively. Methanol, acetonitrile, and acetone were of HPLC grade (J.T. Baker Co, NJ, USA). All reagents were obtained from commercial sources and typically were at purities of 90% or greater : Na_2-EDTA and H_2SO_4 (Junsei Co., Tokyo, Japan), HCl (Waco Co., Osaka, Japan), ammonium formate (Sigma-Aldrich Co., St Louis, MO, USA), ammonium acetate (Merck Co., Darmstadt, Germany), formic acid (Fluka Co., Seelze, Germany), and ammonium hydroxide (Samchun Co., Gyeonggi-do, Korea).

A stock solution of 1000 μg/mL was prepared in methanol and stored at −20 °C. Working standard solutions, at different concentrations, were prepared by appropriate dilution of the stock solution with methanol and stored in a refrigerator at 4 °C. Surrogate mixtures were at a concentration of 100 μg/mL and stored at −20 °C.

Sampling and sample preparation. Samples were collected from surface water of four major rivers (the Han River, the Nakdong River, the Geum River and the Youngsan River) that receive the effluent of the waste water treatment plants. Amber glass bottles with Teflon-lined caps rinsed with ultra-pure water were used for sample collection. To characterize the nature of aquatic samples, conductivity, pH, and temperature were measured during the sampling period. Samples were stored at 4 °C until sample extraction (within two weeks).

Before sample extraction, samples were centrifuged at 11,300*g for 20 min and subsequently filtered through a 1.2 μm glass fiber filter (Whatman, Clifton, NJ, USA) and a 0.45 μm membrane filter (PALL Life Sciences, Mexico) to remove particulate matter prior to solid-phase extraction.

The sample extraction procedure was modified from reference (Karthikeyan and Meyer, 2006)[15] for the simultaneous analysis. Samples (500 mL) were added to 5 mL of 0.1 μg/mL Na_2-EDTA and 25 μL of 10 μg/mL surrogate standards (sulfamethazine-6-^{13}C, ciprofloxacin-3-^{13}C and ibuprofen-6-^{13}C) and adjusted to pH 3 with 3.5 M sulfuric acid. Oasis HLB, 200 mg, 6 cc (Waters, Milford, MA, USA), a copolymer made from a balanced ratio of hydrophilic N-vinylpyrrolidone and lipophilic divinylbenzene and Oasis MCX, 150 mg, 6 cc (Waters, Milford, MA, USA), were preconditioned sequentially with 2 mL of distilled water, 2 mL of methanol, 2 mL of 5% NH_4-hydroxide-methanol solution, 2 mL of distilled water, and 2 mL of distilled water (pH 3.0). The HLB and MCX cartridges were connected in tandem. Samples were loaded through the combined SPE cartridge (HLB-MCX) at 10 mL/min. After sample loading, combined cartridges were separated. The HLB cartridge was then washed with 1 mL of distilled water and eluted with 8 mL of methanol, and the MCX cartridge was washed with 1 mL of distilled water. The two cartridges were re-combined and loaded with 2 mL of methanol, followed by elution with 6 mL of methanol. After removing the HLB cartridge from the MCX cartridge, MCX was eluted separately with 4 mL of 5% NH_4-hydroxide-methanol solution. After eluent collection, 25 μL of 10 μg/mL terbutylazine (for Group-A and -B) as internal standard was added. The combined aliquots were evaporated to dryness with a Turbo Vap (Zymark, Hopkinton, MA, USA) evaporative concentrator, using nitrogen at 40 °C. Samples were re-

Table 2. HPLC chromatographic conditions and mass spectrometric parameters

Parameters	Group-A	Group-B
Column	Eclipse Plus C18 column, 2.1mm i.d., 100 mm length, 3.5 μm particle size	Luna C8 column, 3mm i.d., 150 mm length, 3 μm particle size
Mobile phase	A: 20mM ammonium acetate (pH 6.5) B: Acetonitrile	A: 20mM ammonium formate (0.3% formic acid) B: Acetonitrile
Gradient	Time(min) 0 3 10 12 15 15.1 25 Solvent B(%) 10 10 50 85 85 10 10	Time(min) 0 3 10 11 11.1 20 Solvent B(%) 20 20 80 80 20 20
Column flow rate	0.3 mL/min	0.3 mL/min
Injection volume	10 μL	10 μL
Column temp.	25 °C	25 °C
Ionization mode	Positive ion electrospray	Positive ion electrospray
Capillary voltage	3.50 kV	3.50 kV
Gas temperature	350 °C	350 °C
Gas flow	8 L/min (N_2)	8 L/min (N_2)
Nebulizer	35 psi	35 psi

constituted with 500 μL of ammonium acetate (pH 9.0). The reconstituted sample was filtered through a 0.45 μm syringe filter before LC/MS/MS analysis.

Chromatography and mass spectrometry. LC-MS/MS measurements were performed on an Agilent Technologies (Palo Alto, USA) model 1200 series HPLC equipped with a binary pump system and a coupled Agilent 6410 triple quadrupole mass spectrometer with an electrospray ionization (ESI) interface. After obtaining full scan spectra in scan mode, product ions were obtained from precursor ions in product ion mode, and then characteristic ions were selected for multiple reaction monitoring (MRM).

The chromatographic separation for Group-A was performed on a Eclipse Plus C_{18} column (100 × 2.1 mm I.D.) with a particle size of 3.5 μm (Agilent Technologies, Palo Alto, USA) and for Group-B on a Luna C_8 column (150 × 3.0 mm I.D.) with a particle size of 3.0 μm (Phenomonex, Torrance, CA, USA). Optimal separation was achieved with a binary mobile phase, and the mobile phase composition and gradients for two methods are shown in Table 2.

The optimal working parameters for the mass spectrometer were: capillary voltage, 3.5 kV; nebulizer temperature, 350 °C; drying gas flow, 8 L/min (N_2). The electrospray ionization modes for Group-A and -B were positive ion.

Results and Discussion

Chromatographic separation and mass spectrometry. Instrumental analysis of pharmaceuticals was performed according to the physico-chemical properties. To improve mass peak separation quality and maximize method sensitivity, we modified the mobile phase composition with different organic modifiers. The optimal gradients of HPLC are shown Table 2. In each group, pharmaceuticals were separated effectively without overlap, and total ion chromatogram (TIC) and extracted ion chromatograms (EIC) are shown in Fig. 1-2.

Because Group-A and -B pharmaceuticals are basic compounds, the base peak in full scan spectrum was typically $[M+H]^+$, and the precursor ion was $[M+H]^+$. For analysis of Group-A and -B, chromatographic retention time, precursor ion, product ions, and collision energies are shown in Table 3-4. The "quantitation ion" with the higher intensity was used as a quantifier, and "confirmation ion" with lower intensity was used for qualifier.

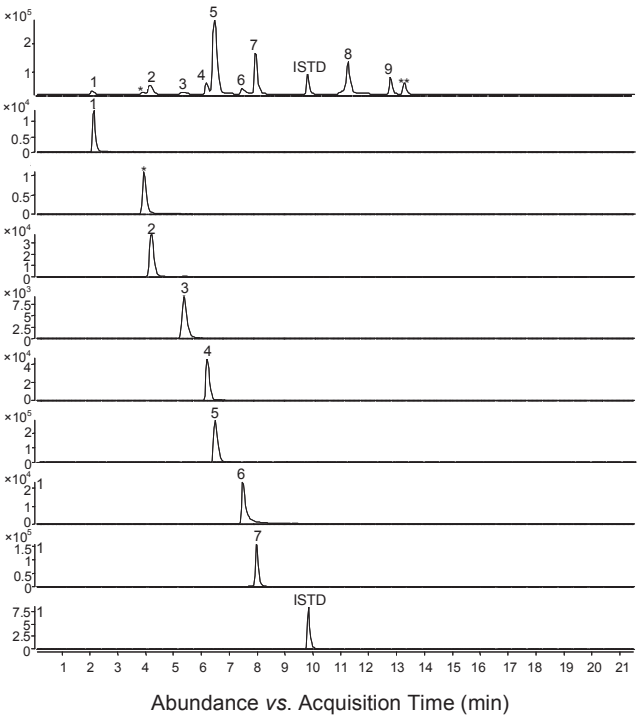

Figure 1. Total ion chromatogram and extracted ion chromatograms of Group-A pharmaceuticals: 1 = acetaminophen, 2 = sulfathiazole, 3 = sulfamethoxazole, 4 = carbadox, 5 = sulfamethazine, 6 = trimethoprim, 7 = lincomycin and ISTD = terbutylazine.

Calibration curve. The internal standard for Group-A and -B, terbutylazine, eluted within the appropriate chromatographic time frame as the analytes, responded well in positive ion ESI mode, and did not exhibit noticeable matrix effects. Terbutylazine was not detected in any of the aquatic samples selected as the reference matrices for quantification.

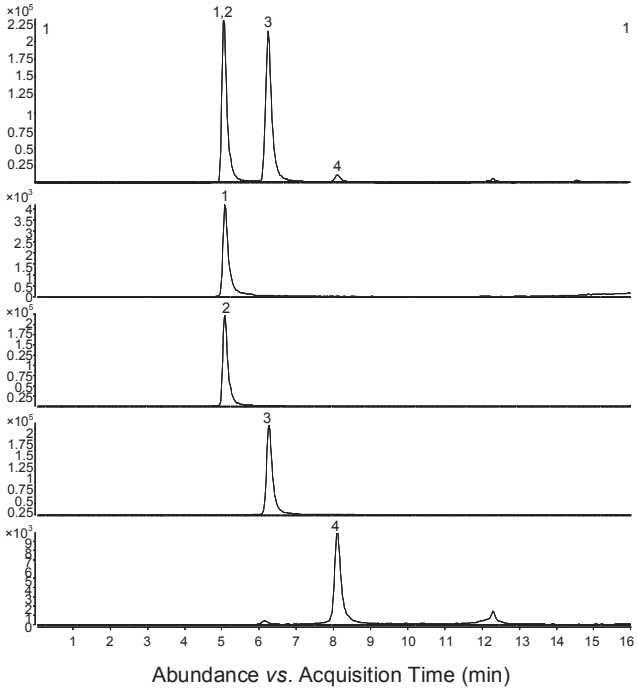

Figure 2. Total ion chromatogram and extracted ion chromatograms of Group-B pharmaceuticals: 1 = oxytetracycline, 2 = ciprofloxacin, 3 = enrofloxacin and 4=chlortetracycline.

The calibration curves were made from the peak area ratio of the pharmaceuticals to the internal standard (terbutylazine). The concentrations of the spiked analytes in the blank surface water were between 0.01 ~ 1 ng/mL. Calibration curves were linear with correlation coefficients ($R^2 > 0.99$) (except oxytetracycline) in the spiked concentration level (Table 5).

Limit of detection (LOD) and limit of quantitation (LOQ). The limit of detection (LODs) was defined as the lowest concentration at which the signal to noise ratio (S/N) were ≥ 3, and the limit of quantitation (LOQs) was defined as a concentration where S/N is ≥ 10, with accuracy and precision both within 20%. The LOD and LOQ for pharmaceuticals were determined by spiking with each concentrations of pharmaceuticals in distilled water and blank surface water. Blank surface water samples with no target antibiotics detected were used as blank surface water.

The limits of detection for the distilled water and the blank surface water were in the range of 0.006 ~ 0.65 and 1.66 ~ 45.05 pg/mL, respectively. The limits of quantitation for the distilled water and the blank surface water were in the range of 0.02 ~ 2.17 and 5.52 ~ 150.15 pg/mL, respectively (Table 6). These result from the matrix effect could negatively interact with the SPE cartridges (saturation of the SPE active sites with natural organic matter) leading to lower absolute recoveries and ionization suppression in MS.

In this study, the limit of quantitation for the established analytical method was found to be lower than 10 ~ 50 pg/mL reported in previous studies,[15] proving to be a sensitive method.

Recovery, precision, and accuracy. Intra-day precision and accuracy were evaluated by three replicates of spiked distilled water and blanked surface water at low (0.1 ng/mL) and high (1 ng/mL) concentrations on the same day. The precisions and accuracies for the distilled water were 3.2 ~ 14.6% (R.S.D.) and −12.4 ~ 20.6% (bias), respectively. The precisions and accuracies for the blank surface water were 2.9 ~ 24.1% (R.S.D.)

Table 3. Retention time, precursor ion, product ions, and collision energies for Group-A analysis

Pharmaceuticals	R.T. (min)	Precursor ion (*m*/*z*)	Confirmation ion (*m*/*z*)		Quantitation ion (*m*/*z*)	Collision Energy (eV)
Acetaminophen	2.16	152	152	93	110	15
Sulfathiazole	4.22	256	108	92	156	15
Lincomycin	7.95	407	407	359	126	20
Sulfamethoxazole	5.36	254	108	99	156	15
Trimethoprim	7.47	291	123	261	230	25
Sulfamethazine	6.49	279	279	124	186	15
Sulfamethazine-6-^{13}C (surrogate standard)	6.49	285	124	285	186	20
Terbutylazine (ISTD)	9.84	212	86	114	156	15

Table 4. Retention time, precursor ion, product ions, and collision energies for Group-B analysis

Pharmaceuticals	R.T. (min)	Precursor ion (*m*/*z*)	Confirmation ion (*m*/*z*)		Quantitation ion (*m*/*z*)	Collision Energy (eV)
Chlortetracycline	8.09	479	444	154	462	20
Oxytetracycline	5.09	461	443	461	426	20
Ciprofloxacin	5.07	332	288	332	314	15
Enrofloxacin	6.26	360	316	360	342	15
Ciprofloxacin-3-^{13}C (surrogate standard)	5.07	335	290	335	317	15
Terbutylazine (ISTD)	10.8	212	86	114	156	15

Table 5. Correlation coefficients (R^2) and linear equation for the spiked surface water (concentration range : 0.01 - 1 ng/mL)

Group	Pharmaceuticals	Correlation Coefficient (R^2)	Linear equation
Group-A	Acetaminophen	0.9994	y = 1.5717x - 0.0132
	Trimethoprim	0.9991	y = 0.3847x - 0.0064
	Lincomycin	0.9993	y = 1.8639x - 0.0202
	Sulfamethazine	0.9988	y = 0.8051x - 0.0126
	Sulfamethoxazole	0.9991	y = 0.3714x - 0.0029
	Sulfathiazole	0.9977	y = 1.2245x - 0.0202
Group-B	Chlortetracycline	0.9900	y = 0.9453x - 0.0458
	Oxytetracycline	0.9802	y = 0.2666x - 0.0171
	Ciprofloxacin	0.9948	y = 1.3144x - 0.0845
	Enrofloxacin	0.9964	y = 3.997x - 0.2596

Table 6. Limit of detection (LODs) and limit of quantitation (LOQs) in distilled water and blank surface water (pg/mL) (n = 4)

Group	Pharmaceuticals	Distilled water		Blank surface water	
		LOD	LOQ	LOD	LOQ
Group-A	Acetaminophen	0.44	1.47	3.13	10.43
	Sulfathiazole	0.05	0.17	1.66	5.52
	Lincomycin	0.02	0.09	8.29	27.62
	Sulfamethoxazole	0.65	2.17	7.31	24.35
	Trimethoprim	0.18	0.60	9.48	31.59
	Sulfamethazine	0.006	0.02	11.65	38.82
Group-B	Chlortetracycline	0.05	0.17	12.61	42.02
	Oxytetracycline	0.12	0.40	45.05	150.15
	Enrofloxacin	0.04	0.15	24.88	82.92
	Ciprofloxacin	0.04	0.13	8.19	27.31

Table 7. Absolute recoveries, precision, and accuracy for pharmaceuticals in distilled water and blank surface water

Group	Pharmaceuticals	Conc. (ng/mL)	Distilled water			Blank surface water	
			Recovery (%)	RSD (%)	Bias (%)*	RSD (%)	Bias (%)*
Group-A	Acetaminophen	0.1	81.9	3.5	4.6	7.1	4.2
		1	86.9	8.1	−0.2	7.0	1.0
	Sulfathiazole	0.1	86.6	9.2	1.5	10.3	3.4
		1	79.7	7.8	2.0	7.8	−1.5
	Lincomycin	0.1	65.1	8.0	2.7	7.9	2.0
		1	62.1	6.2	1.9	3.0	0.7
	Sulfamethoxazole	0.1	94.1	5.3	8.2	4.1	1.6
		1	81.5	5.4	−1.7	9.9	−0.9
	Trimethoprim	0.1	95.8	8.4	−4.6	10.7	0.6
		1	92.2	7.9	4.3	5.8	−0.1
	Sulfamethazine	0.1	91.1	6.9	−6.9	9.6	−1.3
		1	83.9	7.3	4.7	16.2	−1.0
Group-B	Chlortetracycline	0.1	77.1	11.7	2.8	17.9	16.3
		1	75.5	11.2	−1.3	24.1	−16.3
	Oxytetracycline	0.1	125.4	14.2	20.6	4.0	11.1
		1	123.7	14.6	−6.1	2.9	9.5
	Enrofloxacin	0.1	72.8	5.5	−0.8	10.9	12.5
		1	82.3	7.6	−4.3	5.3	3.7
	Ciprofloxacin	0.1	84.4	3.2	−12.4	3.2	7.0
		1	96.9	9.0	3.7	4.3	3.5

$$*\text{Bias}(\%) = \frac{\text{Calculated Value} - \text{Measured Value}}{\text{Calculated Value}} \times 100$$

Table 8. Occurrence and fate of the pharmaceuticals from the surface water of the river that receives the effluent of the domestic WWTP (ng/mL)

Group	Pharmaceuticals	Surface water		
		Average Concentration of detected values	Concentration Range	Number of Detection
Group-A	Acetaminophen	0.006	N.D. - 0.137	1/24
	Sulfathiazole	0.082	N.D. - 1.882	4/24
	Lincomycin	0.020	N.D. - 0.246	8/24
	Sulfamethoxazole	0.018	N.D. - 0.168	5/24
	Trimethoprim	0.001	N.D. - 0.012	2/24
	Sulfamethazine	0.012	N.D. - 0.296	1/24
Group-B	Chlortetracycline	0.308	N.D. - 5.404	9/24
	Oxytetracycline	N.D	N.D	0/24
	Enrofloxacin	0.006	N.D. - 0.076	3/24
	Ciprofloxacin	0.0001	N.D. - 0.001	1/24

Table 9. Occurrence and fate of pharmaceuticals from the surface water of the river that receives the effluent of the livestock WWTP (ng/mL)

Group	Pharmaceuticals	Surface water		
		Average Concentration	Concentration Range	Number of Detection
Group-A	Acetaminophen	0.008	N.D. - 0.126	1/16
	Sulfathiazole	0.115	N.D. - 1.377	7/16
	Lincomycin	0.064	N.D. - 0.343	7/16
	Sulfamethoxazole	0.038	N.D. - 0.435	2/16
	Trimethoprim	0.002	N.D. - 0.021	2/16
	Sulfamethazine	0.025	N.D. - 0.304	2/16
Group-B	Chlortetracycline	0.168	N.D. - 2.237	9/16
	Oxytetracycline	N.D	N.D	0/16
	Enrofloxacin	0.006	N.D. - 0.087	2/16
	Ciprofloxacin	0.001	N.D. - 0.011	2/16

and −16.3 ~ 16.3% (bias), respectively. The precisions and accuracies were poorer in surface water than in distilled water. The tetracyclines (chlortetracycline and oxytetracycline) showed poor precisions and accuracies. The bias (%) was defined as $\frac{\text{Calculated Value} - \text{Measured Value}}{\text{Calculated Value}} \times 100$. These results are within acceptable values of −30% to +20% at each concentration level.[17] Recoveries, precision, and accuracy are shown in Table 7.

Occurrence and fate of pharmaceuticals. The modified and validated method was applied to the measurement of pharmaceuticals from the surface water of the river that receives the effluent of the domestic and livestock WWTP. Samples were collected twice at the same site on different dates: The first sampling date was the end of June, 2007, during the dry season (shortage of water), and the second sampling date was in the middle of August, 2007.

Reported averages are derived using all quantifiable values. In surface water of the river that receives the effluent of the domestic WWTP, lincomycin and chlortetracycline were detected frequently, at 0.002 ~ 0.246 and 0.017 ~ 5.404 ng/mL, respectively. Oxytetracycline was not found (Table 8).

For the surface water of the river that receives the effluent of the livestock WWTP, the most frequently detected pharmaceuticals were sulfathiazole, chlortetracycline, and lincomycin, which are only for veterinary or both human and veterinary prescription (Table 9). Overall, surface water contamination near livestock WWTPs was higher than near domestic WWTPs (from median values).

Conclusions

We have developed a effective and sensitive method for quantitation of ten pharmaceuticals using simultaneous solid-phase extraction and LC-MS/MS in aquatic samples. This method allows simultaneous extraction and clean-up of ten pharmaceuticals compounds with different physical-chemical properties, measuring below concentrations of 0.02 ~ 2.17 pg/mL for distilled water and 5.52 ~ 150.15 pg/mL for blank surface water. The method could be successfully applied to monitoring surface water in the major rivers in South Korea.

References

1. Daughton, C. G.; Ternes, T. A. *Environ. Health Perspect.* **1999**, *107*, 907.
2. Kolpin, D.W. ; Furlong, E. T.; Meyer, M. T.; Thurman, E. M.; Zaugg, S. D.; Barber, L. B.; Buxton, H. T. *Environ. Sci. Technol.* **2002**, *36*, 1201.
3. Cahill, J. D.; Furlong, E. T.; Burkhardt, M. R.; Kolpin, D.; Anderson, L. G. *J. Chromatogr. A* **2004**, *1041*, 171.
4. Richardson, S. D. *Anal. Chem.* **2006**, *78*, 4021.
5. Hernandez, F.; Sancho, J. V.; Ibanez, M.; Guerrero, C. *Trends Anal. Chem.* **2007**, *26*, 466.
6. Ternes, T. A.; Hirsch, R.; Mueller, J.; Haberer, K.; Feresenius, J. *Anal. Chem.* **1998**, *363*, 329.
7. Cunningham, V. L.; Buzby, M.; Hutchinson, T.; Mastrocco, F.; Parke, N.; Roden, N. *Env. Sci. Technol.* **2006**, *40*, 3457.
8. Fent, K.; Weston, A. A.; Caminada, D. *Aquatic. Toxicol.* **2006**, *76*, 122.
9. Crane, M.; Watts, C.; Boucard, T. *Sci. Tot. Environ.* **2006**, *367*, 23.
10. Buchberger, W. W. *Anal. Chim. Acta* **2007**, *593*, 129.
11. Gomez, M. J.; Petrovic, M.; Fernandez-Alba, A. R.; Barcelo, D. *J. Chromatogr. A* **2006**, *1114*, 224
12. Abuin, S.; Codony, R.; Compano, R.; Granados, M.; Prat, M. D. *J. Chromatogr. A* **2006**, *1114*, 73.
13. Miao, X. S.; Bishay, F.; Chen, M.; Metcalfe, C. D. *Environ. Sci. Technol*. **2004**, *38*, 3533.
14. Renew, J. E.; Huang, C. H. *J. Chromatogr. A* **2004**, *1042*, 113.
15. Karthikeyan, K. G.; Meyer, M. T. *Sci. Total Environ.* **2006**, *361*, 196.
16. *Development of Analytical Method and Study of Exposure of Pharmaceuticals and Personal Care Products in Enviroment*; National Institute of Environmental Research, 2006.
17. Cherlet, M.; De Baere, S.; Croubels, S.; De Backer, P. *Anal. Chim. Acta* **2002**, *473*, 167.
18. Cha, J. M.; Yang, S.; Carlson, K. H. *J. Chromatogr. A* **2006**, *1115*, 46.