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Diazepam stability in wastewater and removal by advanced membrane technology, activated carbon, and micelle–clay complex

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ABSTRACT

Stability and removal of the anti-anxiety drug diazepam (valium) from spiked wastewater samples were studied. An advanced wastewater treatment plant (WWTP), utilizing ultrafiltration (UF), activated charcoal (AC), and reverse osmosis (RO) after the secondary biological treatment showed that UF and RO were relatively sufficient in removing spiked diazepam to a safe level. Kinetic studies in both pure water (abiotic degradation) and in sludge (biotic degradation) at room temperature were investigated. Diazepam showed high chemical stability toward degradation in pure water, and underwent faster biodegradation in sludge providing two main degradation products. The degradation reactions in sludge and pure water showed first-order kinetics with rate constant values of 2.6×10^{-7} s^{-1} and 9.08×10^{-8} s^{-1}, respectively (half-life = 31 and 88 d, respectively). Adsorption of diazepam by activated carbon and composite micelle–clay (octadecyltrimethylammonium montmorillonite) complex was studied using both Langmuir and Freundlich isotherms. Based on the determination coefficient, Langmuir isotherm was found to better fit the data, indicating the retention of diazepam monolayer on both adsorbents. Filtration of 100 mg L^{-1} solutions of diazepam by micelle–clay filter yielded almost complete removal at flow rates of 2 mL min^{-1}.

Keywords: Diazepam; Wastewater treatment; Stability in sludge; HF membranes; Activated carbon; Micelle–clay complex

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1. Introduction

Persistent organic pollutants (such as polychlorinated biphenyls (PCBs) and polycyclic aromatic hydrocarbons (PAHs)) have for many years been investigated regarding their influence on the environment [1,2], and were found to have high acute toxicity and tendencies to undergo bioaccumulation and biomagnifications. They also have been reported to have a high resistance against degradation and long half-life in the environment.

Nowadays another concern is arising due to pharmaceuticals or their metabolites found in environmental samples. The presence of pharmaceutical residues in environmental bodies has increased the probability of toxicity risks for animals and humans in the last years [3–5].

It has been reported that the aquatic environment can become polluted with pharmaceutically active compounds (drugs) at low concentration because of the extensive consumption of pharmaceuticals in developed countries [6,7]. Persistence to biochemical degradation and polar structure are indicated as mainly responsible for the incomplete removal of pharmaceuticals during conventional wastewater treatment plant (WWTP) [8].

One important class of pharmaceuticals, which has received recent consideration, is known as benzodiazepines (structure **1**, Fig. 1). One of the most commonly used medicine among the members of the benzodiazepines is diazepam (structure **2**, Fig. 1) (7-chloro-l-methyl-5-phenyl-1,4-benzodiazepin-2-one). It is a long-acting benzodiazepine with anticonvulsant, anxiolytic, sedative, and muscle relaxant properties [9]. It is used for the treatment of acute management of all types of seizures in both adults and children [10]. Diazepam is administered orally, intravenously, or rectally [11].

Most pharmaceuticals undergo biotransformation in the human body, resulting in a release of significant amounts of a variety of metabolites into the aquatic environment, which can be further transformed during the sewage treatment processes (biotic degradation) [12]. Pharmaceuticals may also be chemically degraded by abiotic processes such as hydrolysis, oxidation, and photolysis. The degradation products can be also of concern because of their possible toxicity, which can sometimes be higher than that of the corresponding parent compound [13,14].

Diazepam (**2**) is metabolized in human liver via cytochrome P450 enzymes pathway. It has an elimination half-life of 20–100 h, and produces several pharmacologically active metabolites; the main active metabolites are obtained through demethylation mechanism (**3**, nordiazepam, $t_{½}$ = 36–100 h), temazepam (**4**, $t_{½}$ = 8–22 h), and oxazepam (**5,** $t_{½}$ = 4–15 h) as shown in Fig. 2 [15]. Diazepam and their metabolites are conjugated with glucuronide and are excreted primarily in the urine. From human metabolism studies, it has been shown that 30% of diazepam dose administered to patients is exerted without any change, 12% as nordiazepam (**3**), 15% as temazepam (**4**), 32% as

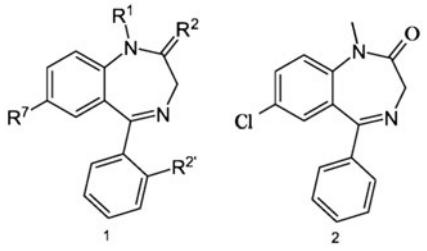

Fig. 1. Chemical structures of benzodiazepines basic structure (**1**), and diazepam (**2**).

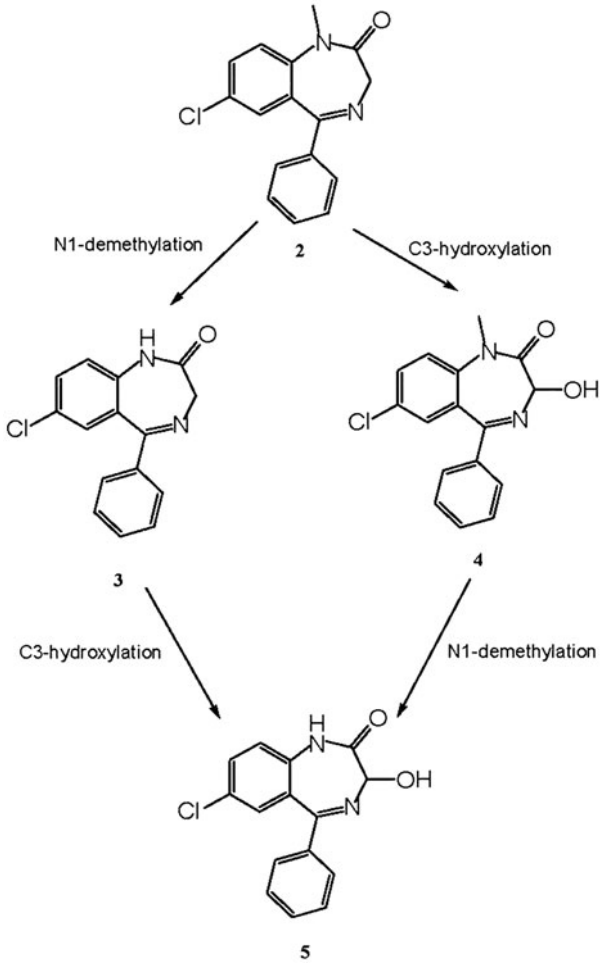

Fig. 2. Metabolism pathways for diazepam (**2**) in humans.

oxazepam (**5**), and 11% of the remaining dose is still unidentified [16]. The diazepam metabolism is mediated by a number of cyctochrome P450 enzymes and proceeds by both N-dealkylation and C3-hydroxylation reactions [15–17].

The two major aims of this study were: (i) to investigate diazepam stability in pure water (abiotic conditions) as well as in activated sludge (biotic conditions) and (ii) to determine the efficiency of advanced wastewater treatment technology, which consists of a combination of activated sludge process with ultrafiltration (UF) membranes, hollow fiber and spiral wound membranes, activated carbon (AC) adsorption, reverse osmosis (RO), and micelle–clay filters, in removing diazepam and its metabolites.

2. Materials and methods

2.1. Chemicals

All chemicals were of analytical grade. Diazepam was obtained from Birzeit Pharmaceutical Company (Palestine) with 99% purity, and was used as received. Wyoming Na-Namontmorillonite SWY-2 clay was obtained from the Source Clays Registry (Clay Mineral Society, Colombia, MO). Quartz sand (size 0.8–1.2 mm) was obtained from Negev industrial minerals (Israel). Octadecyltrimethylammonium (ODTMA) bromide was obtained from Sigma Aldrich (Munich, Germany). Powdered activated charcoal (PAC) with particle size ≤ 60 μm cat. No. 97876 and granular activated charcoal (GAC) with particle size ≤ 700 μm cat. No. 37771 were obtained from Sigma (Sigma Chemical Company, USA). The powder was used for batch adsorption experiments while the granules were used in column experiments. De-ionized water, methanol, and acetonitrile were of HPLC grade, and were purchased from Sigma Aldrich (Munich, Germany).

For sample enrichment and purification, SPE 1 g C-18 6 mL disposable cartridges (Waters, Milford, MA, USA) were used. Samples were shaken using Big Bill, (Banstaed/Themolyne, USA).

2.2. Advanced WWTP

The WWTP employed in this study (Fig. 3) is located at Al-Quds University-Palestine and was described in detail elsewhere [18,19]. Normally, the effluent from this plant is recycled for the irrigation of plants cropped in the field at the University Campus.

2.3. Methods

2.3.1. Diazepam quantification

Diazepam analysis was performed by using HPLC Waters 2,695 equipped with a photodiode array. Data acquisition and control were carried out using Empower ™ software (Waters: Israel). Analytes were separated by using a C18 XBridge® column (4.6 × 150 mm, 5 μm particle size). HPLC conditions were: acetonitrile; water (1:1; v/v) as mobile phase; flow rate of 1.0 mL min^{-1}; UV detection at a wavelength of 230 nm. Acrodisc® syringe filters with GHP membrane

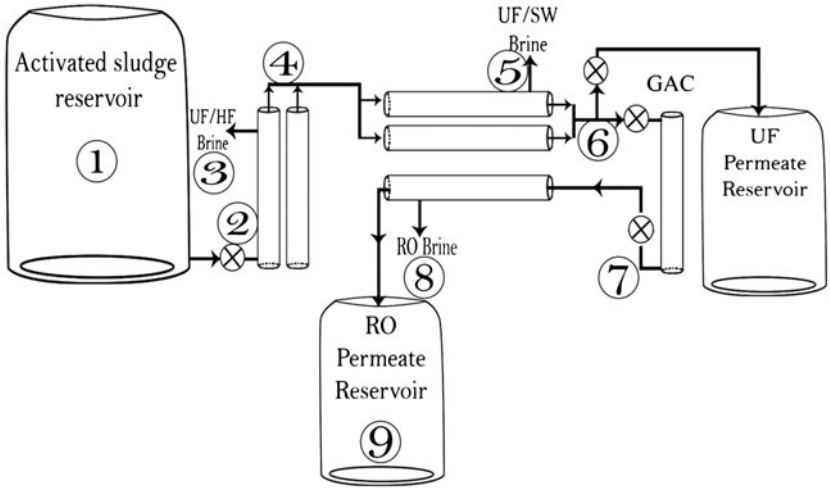

Fig. 3. A schematic representation of the WWTP at Al-Quds University; (1) activated sludge reservoir; (2) activated sludge reservoir effluent; (3) UF/HF, hollow fiber ultrafiltration membrane Brine; (4) UF/HF Permeate; (5) UF/SW, spiral wound ultrafiltration membrane Brine; (6) UF/SW Permeate; (7) GAC, GAC filter effluent; (8) RO, reverse osmosis Brine; and (9) RO, Permeate reservoir.

(hydrophilic polypropylene, 0.45 μm porosity) from Waters were always used for all analytical filtration requirements.

2.3.2. Characterization of wastewater

The wastewater was characterized for physical, chemical, and biological properties before the experiments according to standard methods [20].

2.3.3. Diazepam stability in pure water and WWTP removal efficiency

Prior to perform any other experiment, the stability of diazepam dissolved in pure water and in the activated sludge collected from the WWTP (Fig. 3) was determined to ascertain if hydrolysis or biodegradation reactions had taken place before the filtration stages. For this reason, samples were collected at specific times and analyzed by HPLC. Pure water and the activated sludge samples were spiked with the same quantity of the drug and continuously shacked during the experimental time; moreover, aeration was permitted for sludge samples to preserve the bacterial growth. The concentration of diazepam at each time interval was determined using a calibration curve and the percentage of degraded drug was calculated as the difference to the initial concentration.

The efficiency of the different filtration stages [hollow fiber (UF-HF), spiral wound (UF-SW), AC, and RO] for the removal of diazepam from wastewater was studied by spiking separately the secondary effluent in the activated sludge reservoir (1,000 L) with 1.0 mg L^{-1} of diazepam.

SPE-C18 disposable cartridges were used to preconcentrate 10 mL of each sample by adsorption of analytes. A part of the methanolic solution eluted from SPE cartridge (20 μL) was injected into the HPLC and analyzed as described in the subsection "Diazepam quantification".

2.3.4. Micelle–clay complex preparation

The complex was prepared as described elsewhere [18,21–28]. The micelle–clay complex was obtained by stirring 12 mM of ODTMA with 10 g L^{-1} clay for 72 h.

The suspension was centrifuged for 20 min at 15,000 g, the supernatant was discarded, and the complex was lyophilized. The obtained complex by virtue of its positive charge and hydrophobic region is capable of efficiently binding neutral and negatively charged organic molecules [18,21–24,28].

2.3.5. Batch adsorption experiments

Batch adsorption experiments were carried out on diazepam at different concentrations. Experiments were performed in 250 mL Erlenmeyer flasks containing 200 mg of either micelle–clay complex or PAC; 50 mL of diazepam solutions having known initial concentrations were introduced into each flask. The flasks were shaken in an oscillating shaker for three hours at room temperature then the content of each flask was centrifuged (10,000 g) for 5 min and filtered using 0.45 μm filters. The equilibrium concentration of diazepam was then obtained using HPLC analysis with the conditions reported above. The retention time of diazepam was 4.2 min.

2.3.6. Column filtration experiments

Column filtering experiments were performed using 50/1 (w/w) mixtures of quartz sand and either ODTMA–clay complex, or granular-activated charcoal (GAC), 20 cm layered in borosilicate columns of 25 cm length and 5 cm diameter. Each column contained 13 g of complex, or GAC. The bottom of the column was covered with 3 cm layer of quartz sand. Quartz sand was thoroughly washed by distilled water and dried at 105˚C for 24 h before use. Solutions in pure water (1 L each) containing different diazepam concentrations (0.01, 1, 10, and 100 mg L^{-1}) were passed through either micelle–clay or GAC columns (one column for each solution). In all cases, the flow rate was regulated to 2.0 mL min^{-1}. Eluted fractions were collected in all column experiments and analyzed.

All experiments were performed in three replicates and average values and standard deviations were calculated.

2.3.7. Calibration curves

Linearity of the proposed analytical method was verified by analyzing standard solutions in the range of 0.1–100 mg L^{-1} for diazepam. The calibration curve was obtained with a determination coefficient R^2 of 0.9996. The repeatability of triplicate subsequent injections was ranging from 98.5 to 99.5%, depending on the sample concentration.

3. Results and discussion

3.1. Wastewater characteristics

Table 1 summarizes the chemical, physical, and biological characteristics of wastewater collected from Al-Quds WWTP-activated sludge reservoir. Table 1

Table 1
Physical, chemical, and biological parameters of wastewater

Parameters	Results	Units	Parameters	Results	Units
pH	7.32 ± 0.01	–	TSS	3,700 ± 100	mg L^{-1}
Conductivity	2,000 ± 8	μSm cm^{-1}	BOD	900 ± 150	mg L^{-1}
Temperature	15.5 ± 0.2	℃	COD	1,900 ± 300	mg L^{-1}
Turbidity	5,000 ± 200	NTU	NH_4-N	59.5 ± 0.1	mg L^{-1}
DO	0.40 ± 0.01	mg L^{-1}	PO_4-P	14.3 ± 0.1	mg L^{-1}
TS	4,200 ± 200	mg L^{-1}	FC (*E. coli*)	$2.9 \times 10^5 \pm 1.5 \times 10^5$	cfu/100 mL
TDS	620 ± 50	mg L^{-1}	TC	$6.5 \times 10^6 \pm 3.0 \times 10^6$	cfu/100 mL
Settable solids	240 ± 5	mg L^{-1}	HPC	$2.6 \times 10^7 \pm 1.3 \times 10^7$	cfu/100 mL

Note: DO: Dissolved Oxygen; TS: Total Solids; TDS: Total Dissolved Solids; TSS: Total Suspended Solids; HPC: Heterotrophic Plate Count; TC: Total Coliform; and FC: Fecal Coliform.

shows that this wastewater contains high amounts of suspended solids and organic pollution load, the relatively high values of TSS and COD may be attributed to residues of chemicals in the wastewater from laboratories, which were not well removed by the sedimentation stage and secondary biological treatment. Moreover, high values of electrical conductivity and total dissolved solids are typical of municipal wastewaters and must be reduced if WWTP effluents are to be reused for crop irrigation purposes.

The large populations of bacteria detected are responsible for the fouling process affecting the UF and RO membranes. Al-Quds activated sludge was found to contain Enterobacter and Pseudomonas species: *Escherichia coli*, *Enterobacter sakazakii*, *Citrobacter freundii*, *Pseudomonas aeruginosa*, *Klebsiella pneumonia*, *Enterobacter cloacae*, *Enterobacter amnigenus*, *Enterobacter aerogenes*, *Salmonella spp.*, and *Serratia liquefaciens* [29].

3.2. Stability of diazepam in pure water and in sludge

The kinetic parameters of diazepam hydrolysis in pure water (100 mg L^{-1}) are illustrated in Fig. 4 as natural logarithm of diazepam concentration vs. time (days). The determination coefficient R^2 of the first-order hydrolysis reaction was 0.9981, and the rate constant was 9.08×10^{-8} s^{-1} at pH 7.3.

Similarly, kinetics of diazepam in Al-Quds activated sludge at room temperature was studied starting from the same concentration used in pure water. The determination coefficient R^2 in this case was 0.9987 (Fig. 4), and the rate constant was 2.60×10^{-7} s^{-1}. The degradation half-life was diminished from 88.3 d in pure water to 30.4 d in the activated sludge, where the concentration of diazepam was found at a concentration of 14.4 mg L^{-1} after 60 d of incubation. The degradation rate in the sludge medium was about threefold faster than in pure water.

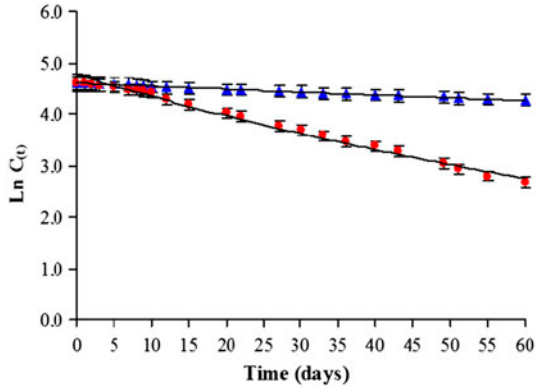

Fig. 4. Plot of Ln of diazepam vs. time in pure water (▲) (SD ± 0.12) and in sludge (●) (SD ± 0.17). T = 25℃, pH 7.3.

The accelerated degradation can be attributed to the bioactivity of bacteria populations present in the activated sludge.

3.3. Efficiency of WWTP for diazepam removal

The efficiency of WWTP at Al-Quds University for the removal of diazepam was studied. The activated sludge reservoir (point 1 in Fig. 3) was spiked with diazepam at concentration of 1.0 mg L^{-1}, which is an amount close to concentration levels of diazepam actually found in wastewater [30].

Samples were taken from different collecting sites of WWTP (Fig. 3). Analytical results of water effluent from the hollow fiber ultra-filtration membrane (UF-HF) indicated that the diazepam removal at this stage was about 82.1%, whereas about 90.4% of the drug was cumulatively removed after passing the spiral wound (UF-SW) membrane (Table 2).

Additionally, there was no complete removal of diazepam from the effluent exiting the GAC filter

Table 2
Removal of diazepam from wastewater by different treatment units in Al-Quds WWTP; average values of three replicates

Sample description		Sampling point as in Fig. 4	Concentration of DZ mg L^{-1}	
			Means ± S.D.	Removal %
The initial concentration of Diazepam in storage tank		1	0.98 ± 0.10	
UF-HF	influent	2	0.84 ± 0.05	
	brine produced	3	0.57 ± 0.03	
	effluent	4	0.15 ± 0.02	82.1
UF-SW	brine	5	0.15 ± 0.04	
	effluent	6	0.08 ± 0.01	90.4
GAC effluent		7	0.05 ± 0.02	93.7
RO	brine	8	0.07 ± 0.01	
	effluent	9	b.l.d.	≈100.0

Note: b.l.d. = below the limit of detection.

(93.7%); whereas, the RO unit showed 100% diazepam removal. It should be worth noting that the concentration of diazepam in the GAC effluent was 0.05 mg L^{-1}, whereas after the passage into the UF filters the concentration in the GAG influent water was just a little bit higher (0.08 mg L^{-1}).

For this reason, the removal of diazepam by RO was a necessary step for reaching a complete purification. Nevertheless, the advanced technology adopted in the Al-Quds WWTP is still affected by problems that are common to all plants: the production of brine in which a large portion of the contaminants ended being concentrated. For this reason, different methods of water filtration and purification should be tested.

3.4. Adsorption isotherms

The micelle–clay composite used in this study is positively charged, has large surface area, and includes large hydrophobic domains. It has been shown by X-ray diffraction, electron microscopy, and adsorption experiments that the material characteristics of the micelle–clay complex are different from those of an organo–clay complex, which is formed by adsorption of the same organic cations ODTMA as monomers [28]. Micelle–clay composites have already been proven to be efficient in the removal of about 20 neutral and anionic pollutants [18,21–27,31,32].

The diazepam adsorption at several initial concentrations on micelle–clay complex and activated charcoal was investigated. Equilibrium relationships between adsorbent and adsorbate can be described by the Langmuir adsorption isotherm [18], represented by Eq. (1):

$$\frac{C_e}{Q_e} = \frac{1}{kQ_{\max}} + \frac{C_e}{Q_{\max}} \quad (1)$$

where C_e (mg L^{-1}) is the equilibrium concentration of the drug in the solution, Q_e (mg g^{-1}) is the equilibrium mass of adsorbed drug per gram of complex or activated charcoal, k (L mg^{-1}) is the Langmuir binding constant, and $Q_{\max}$ (mg g^{-1}) is the maximum mass of drug removed per gram of complex.

The data fitted well the Langmuir equation giving $R^2 = 0.9945$ for activated charcoal, and 0.9970 for the micelle–clay (Table 3). The calculated Langmuir constants k and $Q_{\max}$ are presented in Table 3. The values of k and $Q_{\max}$ parameters for the adsorption isotherm obtained using micelle–clay complex were larger than activated charcoal, suggesting the former to be a more efficient adsorbent for diazepam removal. In

Table 3
Langmuir adsorption parameters (k and $Q_{\max}$) and determination coefficients (R^2) obtained from the adsorption of diazepam on the micelle–clay complex and activated charcoal

Adsorbent	k (L mg^{-1})	$Q_{\max}$ (mg g^{-1})	R^2
Micelle–clay complex	5.3 ± 0.2	31.2 ± 1.7	0.997
Activated charcoal	3.8 ± 0.3	28.9 ± 1.5	0.994
Ratio micelle–clay/charcoal	1.4	1.1	–

particular, the Langmuir binding constant $< k >$ for micelle–clay complex was about 1.4-fold greater than activated charcoal, and the value of Q_{max} was nearly 1.1-fold higher for the former.

3.5. Filtration results

Diazepam solutions were passed through filters which included the micelle–clay complex or activated charcoal mixed with excess sand at 1:50 ratio (w/w). The results shown in Table 4 indicate a significant advantage of the micelle–clay filter in removing diazepam compared to the amount removed by the activated charcoal. The efficiency of filter filled with activated charcoal and sand was adequate only for the lowest diazepam concentration, while the micelle clay system was able to remove the drug also at the higher concentration experimented. This result is not surprising, since the parameters obtained for adsorption isotherms of the two adsorbents clearly showed that the micelle–clay complex was more efficient than activated charcoal in adsorbing and removing diazepam from water.

Previously reported experiments demonstrated the poor capability of AC filters toward removing anionic and some neutral pollutants [18,21,22,25–27,31,32].

Karaman et al. [18] have shown that micelle–clay filter is more efficient than AC filter in removing diclofenac potassium from wastewater. Polubesova et al. [23], using column filters filled with a mixture of quartz sand and micelle–clay complex demonstrated high efficiency for this filter in removing tetracycline and sulfonamide pharmaceuticals from wastewater. In another study, Polubesova et al. [24] have succeeded to achieve an excellent removal of three anionic pollutants (imazaquin, sulfentrazone, and sulfosulfuron) and four neutral pollutants (alachlor, acetochlor, chlorotoluron, and bromacil) by micelle–clay complexes in aqueous dispersions. On the other hand, Zadaka et al. [32] have tested column filters with either a mixture of quartz sand and organic micelle—montmorillonite or zeolite and they found that both filters were capable to remove ethylene dibromide, anionic pollutants as sulfosulfuron, imazaquin and sulfentrazone, and neutral compounds such as bromacil and chlorotoluron from aqueous environments. In contrast to the striking success of the micelle–clay complexes, filters filled with the same weight of AC and sand have shown only partial removal of these pollutants.

Recently, it was suggested [21,22,25] that the integration of micelle–clay complex filters in existing WWTPs may be helpful for improving removal efficiency of recalcitrant residues of non-steroid anti-inflammatory drugs (NSAIDs). Moreover, Khamis et al. [14] concluded that the integration of micelle–clay filters in sewage treatment systems can be a promising technology.

It can be argued that in addition to diazepam residues, wastewater usually includes other recalcitrant organic pollutants. In such cases, GAC filters can be used as first-stage tertiary process to remove the majority of neutral pollutants, and additional micelle–clay filters can be adopted as second stage to eliminate anionic pollutants, and neutral compounds not retained by GAC filters.

Table 4
Removal of diazepam by filtration of 1L of water solutions diazepam (100, 10, 1.0, and 0.01 mg L^{-1}) through laboratory filters consisting of either micelle–clay (MC) or GAC mixed with excess sand at 1:50 (w/w) ratio; means of three replicates

Initial concentration (mg L^{-1})	Column type[a]	Average eluted concentration (mg L^{-1})	±SD
100	MC	0.7	0.1
100	GAC	1.95	0.5
10	MC	b.l.d.	–
10	GAC	1.4	0.9
1.0	MC	b.l.d.	–
1.0	GAC	0.1	0.02
0.01	MC	b.l.d.	–
0.01	GAC	b.l.d.	–

[a]Flow rate, 2 mL min^{-1}; temperature, 25°C; b.l.d., below the detection limit of the analytical method used.

4. Conclusions

Diazepam was found to undergo incomplete degradation both in water and sludge. Hence, further removal of this pharmaceutical from WWTP effluents is a required step. Advanced WWTP with RO as final stage was found to be highly efficient in removing diazepam from spiked wastewater samples. A micelle–clay complex and AC were identified as good adsorbents for this drug with high efficiency. The results indicate that integration of micelle–clay complex into WWTP is very promising in achieving complete elimination of this drug from wastewater effluents.

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References

[1] K. Breivik, A. Sweetman, J. Pacyna, K. Jones, Towards a global historical emission inventory for selected PCB congeners—A mass balance approach. Global production and consumption, Sci. Total Environ. 290(1–3) (2002) 181–198.
[2] Commission Regulation (EC) No 1881/2006, Maximum levels for certain contaminants in food stuffs, OJEU, 20 December 2006, L 364/5.
[3] B. Hailing-Sorensen, S.N. Nielsen, P.F. Lanzky, F. Ingerslev, H.C.H. Lutzhoft, S.E. Jorgensen, Occurrence, fate and effects of pharmaceutical substances in the environment—A review, Chemosphere 36 (1998) 357–393.
[4] C. Daughton, T. Jones-Lepp (Eds.), Pharmaceuticals and Personal Care Products in the Environment: Scientific and Regulatory Issues, American Chemical Society, Washington, DC, 2001.
[5] O.A. Jones, J.N. Lester, N. Voulvoulis, Pharmaceuticals: A threat to drinking water, Trends Biotechnol. 23 (2005) 163–167.
[6] T.A. Ternes, Occurrence of drugs in German sewage treatment plants and rivers, Water Res. 32 (1998) 3245–3260.
[7] C.G. Daughton, T.A. Ternes, Pharmaceuticals and personal care products in the environment: Agents of subtle change, Environ. Health Perspect. 107 (1999) 907–938.
[8] T. Heberer, Occurrence, fate, and removal of pharmaceutical residues in the aquatic environment: A review, Toxicol. Lett. 131 (2002) 5–17.
[9] J.E.F. Reynolds (Ed.), Martindale, The Extra Pharmacopoeia, Royal Pharmaceutical Society, London, 1996, pp. 700–706.
[10] J.C. Cloyd, R.L. Lalonde, T.E. Beniak, G.D. Novack, A single-blind crossover comparison of the pharmacokinetics effects of a new diazepam rectal gel with intravenous Diazepam, Epilepsia 39 (1998) 520–526.
[11] E. Rey, J.M. Treluyer, G. Pons, Pharmacokinetic optimization of benzodiazepine therapy for acute seizures. Focus on delivery routes, Clin. Pharmacokinet. 36 (1999) 409–424.
[12] M. Winkler, J.R. Lawrence, T.R. Neu, Selective degradation of ibuprofen and clofibric acid in two model river biofilm systems, Water Res. 35 (2001) 3197–3205.
[13] R. Henschel, A. Wenzel, M. Diedrich, A. Fliedner, Environmental hazard assessment of pharmaceuticals, Regul. Toxicol. Pharm. 25 (1997) 220–225.
[14] M. Khamis, R. Karaman, F. Ayyash, A. Qtait, O. Deeb, A. Manssra, Efficiency of advanced membrane wastewater treatment plant towards removal of aspirin, salicylic acid, paracetamol and p-aminophenol, J. Environ. Eng. Sci. 5 (2011) 121–137.
[15] O.A.H. Jones, N. Voulvoulis, J.N. Lester, Human pharmaceuticals in wastewater treatment processes, Crit. Rev. Env. Sci. Technol. 35 (2005) 401–427.
[16] D.J. Greenblatt, I.S. Harmatz, H. Friedman, A. Locniskar, R.I.A. Shader, large-sample study of diazepam pharmacokinetics, Ther. Drug Monit. 11 (1989) 652–657.
[17] E. Charles, The photodegradation of diazepam in human and its metabolites, PhD diss., University of Plymouth, Plymouth, 2007.
[18] R. Karaman, M. Khamis, M. Quried, R. Halabieh, I. Makharzeh, A. Manassra, J. Abbadi, A. Qtait, S.A. Bufo, A. Nasser, S. Nir, Removal of diclofenac potassium from wastewater using clay-micelle complex, Environ. Technol. 33(11) (2012) 1279–1287.
[19] M. Dakiky, M. Khamis, A. Manasra, M. Mereb, Selective adsorption of chromium (VI) in industrial waste water using low cost abundantly available adsorbents, Adv. Environ. Res. 6 (2002) 533–540.
[20] American Public Health Association, Standard Methods for the Examination of Water and Wastewater, twenty-first ed., APHA, Washington, DC, 2005.
[21] S. Khalaf, F. Al-Rimawi, M. Khamis, S. Nir, S.A. Bufo, L. Scrano, G. Mecca, R. Karaman, Efficiency of membrane technology, activated charcoal, and a micelle-clay complex for removal of the acidic pharmaceutical mefenamic acid, J. Environ. Sci. Health. Part A 48(13) (2013) 1655–1662.
[22] S. Khalaf, F. Al-Rimawi, M. Khamis, D. Zimmerman, U. Shuali, S. Nir, L. Scrano, S.A. Bufo, R. Karaman, Efficiency of advanced wastewater treatment plant system and laboratory-scale micelle-clay filtration for the removal of ibuprofen residues, J. Environ. Sci. Health. Part B 48(9) (2013) 814–821.
[23] T. Polubesova, S. Nir, D. Zadaka, D.O. Rabinovitz, C. Serban, L. Groisman, B. Rubin, Water purification of organic pollutants by optimized micelle-clay systems, Environ. Sci. Technol. 39 (2005) 2369–2384.
[24] T. Polubesova, D. Zadaka, L. Groisman, S. Nir, Water remediation by micelle-clay system: Case study for tetracycline and sulfonamide antibiotics, Water Res. 40 (2006) 2369–2374.
[25] M. Qurie, M. Khamis, F. Malek, S. Nir, L. Scrano, S.A. Bufo, J. Abbadi, R. Karaman, Stability and removal of naproxen and its metabolite by advanced membrane wastewater treatment plant and micelle-clay complex, CLEAN – Soil Air Water 42(5) (2014) 594–600.
[26] M. Qurie, M. Khamis, A. Manassra, I. Ayyad, S. Nir, L. Scrano, S.A. Bufo, R. Karaman, Removal of Cr(VI) from aqueous environments using micelle-clay adsorption, Sci. World J. 2013 (2013) 7 pp., Article ID 942703, doi: http://dx.doi.org/10.1155/2013/942703.
[27] S. Sulaiman, M. Khamis, S. Nir, F. Lelario, L. Scrano, S.A. Bufo, R. Karaman, Stability and removal of dexamethasone sodium phosphate from wastewater using modified-clays, Environ. Technol. 35(15) (2014) 1945–1955.
[28] Y.G. Mishael, T. Undabeyita, G. Rytwo, B. Papahadjopoulos-Sternberg, B. Rubin, S. Nir, Sulfometuron adsorption via alkyl ammonium cations adsorption as monomers and micelles on montmorillonite, J. Agric. Food Chem. 50 (2002) 2856–2863.

[29] J. Abbadi, R. Saleh, S. Nusseibeh, M. Qurie, M. Khamis, R. Karaman, L. Scrano, S.A. Bufo, Microbial removal from secondary treated wastewater using a hybrid system of ultrafiltration and reverse osmosis, J. Environ. Sci. Eng. A1 (2012) 853–869.
[30] D. Bendz, N.A. Paxéus, T.R. Ginn, F.J. Loge, Occurrence and fate of pharmaceutically active compounds in the environment, a case study, J. Hazard. Mater. 122 (2005) 195–204.
[31] M. Khamis, R. Karaman, M. Qurie, J. Abbadi, S. Nusseibeh, A. Manassra, S. Nir, Performance of micelle-clay filters for removing pollutants and bacteria from tertiary treated wastewater, J. Environ. Sci. Eng. A1 (2012) 160–168.
[32] D. Zadaka, Y.G. Mishael, T. Polubesova, C. Serban, S. Nir, Modified silicates and porous glass as adsorbents for removal of organic pollutants from water and comparison activated carbons, Appl. Clay Sci. 36 (2007) 174–181.