Removal Pathways for Target Pharmaceuticals in Constructed Wetlands for Wastewater

Introduction

Over the last ten to twenty years, pharmaceuticals and their degradation byproducts have been identified as emerging contaminants of concern. For example, one study found of the 33 target pharmaceutical compounds, 27 were detected in streams in Southeast China (Hong et al., 2018). Pharmaceuticals are becoming ubiquitous in surface waters, and even at trace levels, mixtures of these compounds can damage the local ecosystem (Riva et al., 2019). These products are used frequently and have been identified in wastewater streams. Traditional wastewater filtration plants have shown low to moderate success at removing these compounds before discharging them into streams and other waterways (Zhang et al., 2013). Additional tertiary treatment methods have been found to effectively reduce pharmaceutical compound concentrations but at high costs (Matamoros et al. 2011).

It is widely agreed that constructed wetlands are an effective and low-cost alternative to traditional tertiary treatment methods for enhanced removal of numerous wastewater compounds, including emerging contaminants such as pharmaceuticals (Ilyas et al., 2019). Several processes occur in constructed wetlands that work to remove pharmaceuticals from wastewater. This paper explores the possible removal pathways within wetlands for selected pharmaceuticals and how constructed wetland design and chemical characteristics of the compounds can impact the removal efficiency of these pathways.

Methods

The effectiveness of constructed wetlands for removing pharmaceuticals from wastewater has been widely studied. One such review paper will be used as the bases of this paper. Ilyas et al. synthesized their findings on the design factors for constructed wetlands and their impacts on the removal of pharmaceuticals into a review paper (2019). Several removal mechanisms are active within a wetland that facilitates the removal of certain pharmaceutical compounds (Table 1) (Ilyas et al., 2019). This paper will examine these compounds and their removal mechanisms in more depth. Once target compounds were identified, the references within Illyas' study were reviewed to examine how efficiently the wetlands worked to remove the target compounds and what removal mechanisms might be in play within the wetland. In addition to the references from Illyas' work, a Google Scholar search provided additional studies to help better understand the prevalence of pharmaceuticals in wastewater and detailed explanations of the selected removal mechanisms. Using the selected reference studies, this paper examines how effectively selected pharmaceuticals are removed from wastewater using constructed wetlands and the chemical properties or wetland design factors that dictate the active removal pathways at work.

Table 1 – Study Compounds & Removal Mechanisms

REMOVAL MECHANISM	COMPOUNDS
ADSORPTION Through Substrate Media	▪ Clarithromycin ▪ Ofloxacin
Plant Uptake	▪ Sulfamethazine
Biodegradation	▪ Diclofenac ▪ Ofloxacin ▪ Sulfamethazine ▪ Gemfibrozil
Photodegradation	▪ Diclofenac ▪ Ketoprofen ▪ Clarithromycin

Adsorption through Substrate Media

Adsorption through substrate media occurs when compounds in the water column bind with the constructed wetland's soil or gravel. How effectively a compound can bind to the bed media depends on multiple factors for both the compound and the media. Influencing factors include chemical structure, water-solubility, pH, organic matter content of the media, ionic strength, and hydrophobic characteristics (Zhang et al. 2014). One driving factor for adsorption is electrostatic interactions between the positively and negatively charged surfaces. Zhang et al. determined this mechanism works best with bed material that is primarily organic matter, clays, or metal oxides (2014). Other factors for determining how effectively a compound will bind to substrate media is to look at either its sorption coefficient (K_d), organic carbon coefficient (K_{oc}), or its octanol-water partitioning coefficient (K_{ow}). The sorption coefficient is the ratio between the percent of the compound in the aqueous phase to the percent sorbed to soil or sediment. The organic carbon coefficient is the K_d normalized for organic carbon content. The partition coefficient plays a part in describing the hydrophobicity of the compound. The above coefficients positively correlate with a compound's ability to absorb the substrate media.

Of the compounds covered, clarithromycin and ofloxacin are identified as predominantly removed via substrate media adsorption or sorption to carbon-rich surfaces (Ilyas et al., 2019). Clarithromycin was studied by Hijosa-Valsero et al. (2011) and Berglund et al. (2014). Clarithromycin removal was studied as a tertiary treatment through mesocosm-scale constructed wetlands for wastewater effluent. Initial concentrations were 0.4 μgL^{-1} (Berglund et al. 2013) and 110 ngL^{-1}(Hijosa-Valera et al. 2011). In Hijosa-Valsero's studies, wetlands were designed to have an Hydrualic Rention Time (HRT) between 2.1 and 6.1 days (2014). Bergland designed all his study wetlands to have an HRT of 5.7 days (2011). Hijosa-Valsero's study had removal efficiencies between 50% (HRT 6.1 days) and 22% (HRT 2.1 days). Bergland found an average removal efficiency of 39%. Hijosa-Valsero also reported the K_{ow} of Clarithromycin as 3.16 mgL^{-1} and the K_{oc} as 2.17 mgL^{-1}.

Ofloxacin was studied in a mesocosm-scale constructed wetland to observe how different substrate media affect the removal efficiency of pharmaceuticals from wastewater (Chen et al., 2016). Influent concentrations of ofloxacin were around 7.26 ngL^{-1}(Chen et al., 2016). On average, the removal efficiency for ofloxacin was 100% (Chen et al., 2016). Samples collected from the substrate found ofloxacin concentration between 10.8ng/g and 45.8ng/g (dry weight)(Chen et al. 2016). The K_d of ofloxacin, between 1.03mgL^{-1} and 1.11mgL^{-1}, suggests that substrate media plays a large role in removing this compound (Chen et al., 2016). It was also observed that over time the aqueous concentration of ofloxacin decreases, and the substrate concentration increases (Chen et al., 2016). There was a consensus among the selected studies that the selected compounds were removed via adsorption within the constructed wetlands.

Plant Uptake

Plants can absorb pharmaceuticals from wastewater in a wetland through mass flow or active uptake. Mass flow works where the compounds are diffused from an area of higher concentration (in the wastewater) to lower concentrations (inside the plant). Active uptake of the pollutants happens when the compounds are directly absorbed into the plant through the roots. The plant can store the pollutant in three possible locations: cell vacuole, extracellular space, or integration into the cell membrane (Stottmeister et al., 2003). Pharmaceuticals that are likely to be taken up by plants have several characteristics in common. The compounds typically removed via this pathway have a K_{ow} range between 0.5 and 3 (Zhang et al., 2013). Pharmaceuticals with log values within this range can diffuse through the plant membrane. Lower log K_{ow} values mean the pollutant is more water-soluble and is less likely to be transported into the plant (Dan et al., 2013). The pollutants should also be lipophilic to pass through cell membranes. Another important physicochemical characteristic is the ionization constant (p*K*a). This constant examines how likely a compound will break down into its constituent ions in water. The pKa indicates how strong an acid is in solution; the higher the pKa value, the stronger the acid. When designing wetlands for the vegetative uptake of pollutants, it is necessary to consider the seasonal growth patterns of plants. Nivala et al. stated that plant development and activity follow annual cycles that could limit the effectiveness of a wetland in removing pollutants (2019). Plant uptake should not be relied upon to remove pharmaceuticals during the non-growing seasons.

The removal of sulfamethazine from raw wastewater was studied using mesocosm-scale constructed wetlands by Xian et al. (2010) and in pilot-scale wetlands by Dan et al. (2013). Sulfamethazine levels in the influent ranged from 100 μgL^{-1} (Xian et al., 2010) to 3.7 ngL^{-1} (Dan et al., 2013). When examining the removal of sulfamethazine over 35 days, the compound was mostly removed within the first eight days, and then levels plateaued (Xian et al. 2010). Removal efficiencies ranged greatly from 0.4% (Dan et al., 2013) to 99.5% (Xian et al., 2010). The lowest removal efficiencies might be skewed by the low levels of Sulfamethazine entering the wetlands that were near the analysis method's limit of quantitation (Dan et al., 2013). High removal efficiencies were found in a mesocosm scale wetland with floating macrophyte bed systems (Xian et al., 2010). When the sulfamethazine concentration was measured in the wetland plant biomass, it helped prove that this compound is directly absorbed into the plant via the roots (Xian et al. 2010).

Sulfamethazine's physicochemical properties are likely why plant uptake can effectively remove this compound. Its log K_{ow} is 0.76, and p*K*a is 7.49 (Dan et al., 2013). This log K_{ow} value is within the ideal range to be taken up by the wetland vegetation. Plant uptake via active uptake or diffusion of pharmaceuticals is an important removal mechanism for a constructed wetland. Based on the selected studies, plant uptake is likely the main removal mechanism for sulfamethazine.

Biodegradation

Biodegradation is the transformation of compounds via aerobic and anaerobic microbial communities. Microorganisms can transform compounds via mineralization, transformation into hydrophobic compounds, which patrician onto the solid phase, or transformation into hydrophilic compounds that remain aqueous (Zhang et al., 2014). The selected studies for this research did not identify the physiochemical properties of target compounds that facilitate this process, but one potential factor is chemical structure. For example, several compounds chemically similar to diclofenac have varying biodegradability, and the only difference between these compounds is the presence or absence of an aryl group (Zhang et al., 2014).

Although more research is still needed to determine the chemical characteristics that determine biodegradability, scientists can determine what microbial communities are present in constructed wetlands. Scientists can tell whether microbial communities within a wetland are aerobic or anaerobic by looking at the oxidation-reduction potential (ORP) and dissolved oxygen (DO) levels in the effluent of the wetland. If the effluent has a positive ORP and high levels of DO, this is a good indicator that aerobic microbial communities are present within the wetland (Hijosa-Valsero et al., 2010). Microbial activity is also dependent on temperature. Hijosa-Valsero et al. stated that the ideal temperature for most microbial activity, particularly nitrifying and proteolytic bacteria, is between 15° and 25°C (2010). During colder months, these microbial pathways are shut down and take several months to rebound to ideal operating temperatures

(Nivala et al., 2019). Hijosa-Valsero et al. also stated the type of vegetation present could affect the biodegradation of pharmaceutical compounds (2010). The rhizosphere surrounding a plant's root system provides nutrients and a large surface area to promote a diverse microbial community (Zhang et al., 2014), which can facilitate the biodegradation of some pollutants. Liu found a positive relationship between soil pH and microbial diversity which could also help to stimulate the biodegradation of pharmaceuticals in wetlands (2014). When designing a wetland, the water level can be a key determining factor as to whether the wetland will be aerobic or anaerobic (Liu et al. 2014). Subsurface flow in a wetland likely promotes anaerobic microbial communities. These compounds can be removed via metabolic or cometabolic pathways, but cometabolic pathways require specific enzymes that are generated during the metabolic process to initiate this removal pathway (Kahl et al. 2017). Carbonaceous Biological Oxygen Demand (CBOD) measures oxygen utilized by aerobic compounds during the biodegradation process. Lower CBOD levels could indicate that cometabolic pathways are being used (Khal et al., 2017). While cometabolic pathways might be used initially, it is still possible that metabolic pathways would be used later in the biodegradation process (Kahl et al. 2017).

Some researchers have stated that biodegradation alone is not a sufficient removal mechanism for pharmaceuticals from wastewater (Zhang et al., 2014). Pharmaceuticals can have negative impacts on the microbial communities in wetlands, such as temporary negative effects on biofilms and compounds that are bioactive may inhibit the function of bacteria (Zhang et al. 2014). Lower concentrations of pharmaceuticals in influent may also be insufficient to stimulate necessary enzyme production in the bacteria (Zhang et al., 2014).

Ilyas et al. stated that the compounds primarily removed through biodegradation are diclofenac, ofloxacin, sulfamethazine, and gemfibrozil (2019). Diclofenac was studied by Aviala et al. (2010 and 2014), Chen et al. (2016), He et al. (2018), Kahl et al, (2017), Hijosa-Valsero et al. (2010 and 2012), and Nivala et al. (2017). From these studies, diclofenac in the wetland effluents ranged from .37 µg/L (Hijosa-Valsero et al., 2010) to 6 µg/L (Nivala et al., 2019). Removal efficiencies were comparatively low, ranging from 17% to 52% in Hijosa-Valsero et al's 2010 study to 25% to 75% in Nivala et al's 2019 study. Both studies were designed around multi-cell subsurface wetlands, Hijosa-Valsero et al (2010) at a mesocosm-scale and Nivala et al (2019) at the pilot scale. In the mesocosm-scale wetland, most of the removal of diclofenac occurred within the first treatment cell where anaerobic pathways were present (Hijosa-Valsero et al., 2019). The pilot-scale study concluded that vegetated cells with high DO and aerobic microbial communities successfully removed diclofenac. The removal of diclofenac is highly variable and poorly understood (Kahl et al. 2017). This compound can be removed via aerobic or anaerobic biodegradation. Hijsoa-Valsero et al's 2010 study speculated that due to the low ORP in the wetlands where diclofenac was effectively removed, this compound was likely removed via anaerobic pathways. These anaerobic pathways are driven by reductive dehalogenation by anaerobic degraders (Chen et al., 2016). The lower removal rates of diclofenac could be due to the chemical composition of this compound. In contrast, Nival et al. reported high ORP and DO levels in the effluent, indicating that in this study, aerobic biodegradation likely played a role in removing diclofenac (2019).

Ofloxacin was studied by Chen et al. (2016) and Yan et al. (2016) in mesocosm-scale constructed wetlands. Influent in these two studies ranged from 7.2 µg/L (Chen et al. 2016) to 500 µg/L (Yan et al. 2016). Both studies reported near complete removal of ofloxacin from the wastewater (Chen et al, 2016 & Yan et al, 2016). Yan et al described ofloxacin as mostly cations, which are easily removed via electrostatic interactions with negatively charged biomass and has a high metabolic rate (2019).

Sulfamethazine was studied by Chen et al (2016), Choi et al (2016), Liu et al (2014), and Xian et al (2010). Influent concentrations of sulfamethazine ranged from 8.07 ng/L (Chen et al. 2016) to 104 µg/L (Xian et al. 2016). Removal rates ranged from 40% (Liu et al., 2014) to 99.5% (Xian et al., 2016). Liu et al speculated that wetlands with higher oxidizing abilities could stimulate microbial activity (2014). It was also observed that microbial communities could feed and grow from sulfonamides (Liu et al., 2014).

Lastly, gemfibrozil was studied by Conkle et al (2008), Yi et al (2017), and Zhang et al (2018). Gemfibrozil concentrations in the influent of Conkle et al's wetlands were 1.65 µg/L (2008). Conkle et al found an average removal efficiency of 95% of gemfibrozil in his wetlands. Yi et al reported that the increase in BOD and CBOD could indicate an increase in biodegradable compounds broken down via cometabolic pathways, such as gemfibrozil (2017). Cometabolic pathways transform compounds using enzymes created during the metabolic process. Zhang et al stated dehydrogenase is a common oxidase with a strong relationship with gemfibrozil (2018). Yi et al found that autotrophic ammonia-oxidizing bacteria can stimulate cometabolic pathways (2017). Both Yi et al. and Zhang et al. agree that nitrification plays a large part in the breakdown of this compound, as evidenced by the strong positive relationship between gemfibrozil and ammonia (2017) (2018).

Photodegradation

Photodegradation is the physical breakdown of compounds via light. This process is also known as photolysis. Not much research has been done on what characteristics of the compounds facilitate this process. Several studies have described the wetland conditions that could facilitate this degradation pathway. Photolysis depends on solar irradiation, so it is most effective within the first 20cm of the water column (Rühmland et al., 2015). Studies have also found that this removal process is most effective in unplanted wetlands (Avila et al., 2014). Even plant litter accumulation on the water's surface could reduce the wetland's removal efficiency (Reyes-Contreras et al., 2012). Since this process is driven by solar irradiation, this process is affected by seasonal changes in daylight. Drops in removal efficiencies were seen during the winter months when daylight hours were shorter, and thus, there was a drop in solar irradiation (Matamoras et al. 2008).

Diclofenac, ketoprofen, and clarithromycin are believed to be primarily removed via photodegradation (Ilyas et al. 2019). Diclofenac was studied by Matamoros et al. (2008, 2012), Avila et al (2014), Rühmland et al (2015), and Francini et al (2018). Matamoros, Avila, and Fancini designed their studies to use mesocosm-scale constructed wetlands. Diclofenac removal rates were between 73% (Matamoros et al., 2008) and 96% (Matamoros et al., 2008). Lower removal rates can be attributed to the samples being collected in the winter (Matamoros et al., 2008). In Rühmland et al's study, samples were placed in sealed vials within the wetland at varying depths in the water column, and they found a 90% removal of diclofenac from the samples after six days (2015). Most other authors agreed that photodegradation was one of the main degradation pathways for diclofenac, but Rühmland et al's work helped to confirm this theory.

Ketoprofen is believed to be mainly removed via photodegradation (Ilyas et al., 2019). Matamoros et al studied this compound in 2008 and 2012, Reyes-Contreras et al in 2012, and Francini et al in 2018. All used mesocosm-scale constructed wetlands for their research.

Removal efficiencies for ketoprofen were between 5.4% (Reyes-Contreras et al., 2012) and 99% (Matamoros et al., 2008). Reyes-Contreras et al' study had better removal efficiencies for unplanted wetlands during summer months, as high as 96.8% (2012). It was assumed that the main removal mechanism for this compound is photodegradation because they saw drops in removal efficiencies over time as plant foliage and litter took over the wetlands and decreased the solar irradiation within the water column (Reyes-Contreras et al, 2012).

The last compound assumed to be primarily removed via photodegradation is clarithromycin. Hijosa-Valsero et al (2011) and Berglund et al (2014) studied this compound. Both designed their studies around mesocosm-scale constructed wetlands. The maximum removal efficiency for this compound was 59% (Berglund et al., 2014). Clarithromycin is described as a recalcitrant compound that can be difficult to remove from wastewater (Hijosa-Valsero et al., 2011). Both studies agree that this compound is likely removed via photodegradation. Hijosa-Valsero et al stated that the long hydraulic retention time required to see partial removal of this compound indicates that this compound is removed via this removal pathway (2011).

Summary

Table 2 shows the removal efficiencies and the likely removal mechanisms for the target compounds from the selected studies.

Table 2 - Removal Efficiencies

Compound	Removal Mechanisms[1]	Maximum Removal Efficiency	Minimum Removal Efficiency
Clarithromycin	Adsorption & Photodegradation	59%[2]	22%[1]
Diclofenac	Biodegradation & Photodegradation	100%[5]	17%[6]
Gemfibrozil	Biodegradation	95%[8]	15.4%[8]
Ketoprofen	Photodegradation	99%[10]	5.4%[9]
Ofloxacin	Adsorption & Biodegradation	90%[11]	89%[11]
Sulfamethazine	Plant Uptake & Biodegradation	99.5% [13]	40% [12]

1. Ilyas et al. (2019) 2. Berglund et al. (2014) 3. Hijosa-Valsero et al. (2011) 4. Hijosa-Valsero et al. (2010) 5. Chen et al. (2016) 6. Hijosa-Valsero et al. (2010) 7. Conkle et al. (2008) 8. Zhang et al. (2018) 9. Reyes-Contreras et al. (2012) 10. Matamoros et al. (2008) 11. Yan et al. (2016) 12. Liu et al. (2014) 13. Xian et al. (2010)

Rühmland et al's study removed 90% of diclofenac via photodegradation after six days (2015). Other authors (Hijosa-Valsero et al., 2011) claimed that diclofenac was poorly understood and, thus, difficult to identify what characteristics could contribute to primary removal mechanisms. Ketoprofen had removal efficiencies as high as 96.8% within 2.1 days in a system where photodegradation was the main removal mechanism (Reyes-Contreras et al., 2012). Reyes-Contreras et al. stated that removal efficiency in these systems heavily depended on the season and amount of light in the system (2012). Hijosa-Valsero et al identified that clarithromycin was recalcitrant and difficult to remove (2011). Hijosa-Valsero et al found removal efficiencies of around 50% and identified the primary removal mechanism as adsorption. Yan et al identified that most ofloxacin was biodegraded within the first two hours of their study (2016). Ofloxacin can also be removed through adsorption, so additional HRT would be beneficial to ensure maximum removal from wastewater. Sulfamethazine was mostly removed within the first eight days of Xian et al's study via plant uptake (2010). Conkle et al found 95% removal of gemfibrozil in his study, which had an HRT of one to nine days (2008).

There are several seasonal considerations when designing wetlands to promote the removal of target compounds. Temperature and seasonal changes greatly affect plant uptake, biodegradation, and photodegradation. Reyes-Contreras et al reported that ketoprofen had 96.8% removal efficiency during the summer, but it dropped to 5.4% during the winter months (2012). During winter, wetlands experience colder temperatures that might suppress plant and microbial activity slowing the breakdown of these compounds. The winter months also have less daylight and solar irradiation, limiting the amount of photodegradation in the wetlands.

The wetland's design can influence removal efficiencies and should be carefully considered. Avila et al's 2014 study included a multi-cell design wetland that allowed for multiple removal mechanisms. In Reyes-Contreras et al's 2012 study, different wetland cells were designed to facilitate specific removal mechanisms; for example, unplanted cells allowed the photodegradation processes to excel. Vertical flow wetlands tend to have shorter hydraulic retention time than horizontal subsurface systems (Stottmeister et al., 2003).

This paper aimed to explore removal efficiencies and pathways of pharmaceuticals in constructed wetlands and the chemical characteristics and design features that play a role in this treatment process. Constructed wetlands are successful at removing pharmaceuticals from wastewater, but careful consideration needs to go into the design of wetlands to ensure that maximum removal efficiencies can be obtained. The optimal design would include multiple cells, each allowing pharmaceutical treatment through different removal pathways. The best option for Wastewater Treatment Plants that would like to explore this option would be to run pilot-scale studies at their treatment facility to better understand how their wastewater interacts with the designed wetland.

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