Ecological Modelling 319 (2016) 137–146

Contents lists available at ScienceDirect

Ecological Modelling

journal homepage: www.elsevier.com/locate/ecolmodel

Modelling antibiotics transport in a waste stabilization pond system in Tanzania

Cathrine Christmas Møller[a,*], Johan J. Weisser[a], Sijaona Msigala[b], Robinson Mdegela[b], Sven Erik Jørgensen[a], Bjarne Styrishave[a]

[a] Toxicology Laboratory, Section of Advanced Drug Analysis, Department of Pharmacy, Faculty of Health and Medical Sciences, University of Copenhagen, Universitetsparken 2, DK-2100 Copenhagen, Denmark
[b] Department of Veterinary Medicine and Public Health, Faculty of Veterinary Medicine, Sokoine University of Agriculture, PO Box 3021, Morogoro, Tanzania

ARTICLE INFO

Article history:
Available online 21 October 2015

Keywords:
STELLA
Sulfamethoxazole
Ciprofloxacin
Metronidazole
Trimethoprim
Degradation
Photolysis
Hydrolysis

ABSTRACT

Antibiotics in wastewater have become a growing problem in urban and peri-urban areas in developing countries as a result of increased use and misuse of antibiotics. A simple dynamic model, that describes the most important removal processes of antibiotic from the wastewater stabilization pond system (WSP) "Mafisa" in Morogoro, Tanzania, was developed using STELLA® software package. The model was based on liquid chromatography tandem mass spectrometry (LCMS/MS) analysis of trimethoprim, in water collected in the WSP. Concentrations of trimethoprim measured in the dry season and the rainy season were used in development of the model. To determine the model's applicability to simulate the removal of trimethoprim, a calibration was performed using concentrations from the dry season and a validation was performed using concentrations from the rainy season. To test the model's capacity to simulate the removal of other antibiotics than trimethoprim, a second validation was performed for three other antibiotics; metronidazole, sulfamethoxazole and ciprofloxacin. A two-tailed *t*-test with a confidence interval of 95% showed no significant difference ($P = 0.7819$) between the values given by the model (CSIM) and the values measured by LCMS/MS (COBS) of the first validation, and the standard deviation (SD) between the differences was 1%. The second validation gave a mean SD = 18% (found by a two-tailed *t*-test with a confidence interval of 95%) of the differences between CSIM and COBS. The model was developed under the assumption that settling, biodegradation, hydrolysis and photolysis were the only removal processes other than outlet. The major removal processes for trimethoprim and sulfamethoxazole were through settling and the outlet. Ciprofloxacin was removed by settling in the first pond. Metronidazole was mainly removed through the outlet, but settling and hydrolysis/photolysis also played a role. A sensitivity analysis (±10%) showed that the soil adsorption coefficient, the amount of suspended matter and the ratio of flow rate and volume were the most sensitive parameters. To strengthen the model, the exact removal processes should be further analysed. To apply the model on other WSP, a calibration of the settling rate constant and the amount of suspended matter should be performed.

1. Introduction

The discovery of antibiotics in the 1940s came as a breakthrough in treating bacterial infections worldwide (Mshana et al., 2013). However, the extended use has resulted in pollution of environmental waters such as rivers, groundwater and surface water by antibiotics and their residues (Kummerer, 2009; Mutiyar and Mittal, 2014). Antibiotics reach the environment in various ways and are considered pseudo-persistent contaminants due to their continual introduction and persistence (Li et al., 2009). Studies have shown a relationship between the sale of human pharmaceuticals, and their presence in sewage treatment plants (Zhou et al., 2012). Depending on the type, approximately 30% of orally administered antibiotics are metabolized in the body, and 70% are excreted unmetabolized through urine and faeces (Kummerer, 2009). Through urine and faeces these antibiotics may enter

* Corresponding author. Tel.: +45 21286860.
E-mail address: cathrine.ccm@gmail.com (C.C. Møller).

http://dx.doi.org/10.1016/j.ecolmodel.2015.09.017
0304-3800/

wastewater treatment plants, and if their removal is insufficient, they may end up in the groundwater and surface water. Antibiotics reaching the environment also originate from veterinary drugs used as growth promoters and for treatment of diseases in live stock (Nonga et al., 2010). This study also found that 65% of the farmers administered antibiotics without consulting a veterinarian and 100% of the eggs investigated contained antibiotic residues. Eggs containing antibiotic residues, sold at markets are therefore a way of involuntary exposure of antibiotics to the general population. Manure used as fertiliser, if originated from animals treated with antibiotics, presents another potential source of contamination of groundwater, surface water and crops (Khachatourians, 1998).

Since 2006 antibiotics have been detected in wastewater at concentrations ranging from 0.02 ng/L for sulfamethoxazole (Sul) up to 2292 ng/L for ciprofloxacin (Cip) (Chang et al., 2010; Gracia-Lor et al., 2011; Li et al., 2009; Mutiyar and Mittal, 2014). With the presence of antibiotics in the environment, there is a greater risk of development of antimicrobial resistance in bacteria. The spread of antibiotic resistance has become a worldwide problem (Negreanu et al., 2012) especially in Africa where resistance rates have been rising to nearly all known pathogens within the past 50 years (Vlieghe et al., 2009). Aquatic environments may serve as reservoirs for antibiotic resistant genes (Negreanu et al., 2012). Even antibiotic concentrations below Minimal Inhibitory Concentration (MIC) can promote development of resistance (Gullberg et al., 2011). This suggest that occurrence of trace amounts of antibiotics in the environment may accelerate development of antibiotic resistant bacteria (Negreanu et al., 2012).

Antibiotics are a very diverse group of chemicals with very different physico-chemical properties. Consequently, analysing a broad range of antibiotics in wastewater is a challenging task, demanding the availability of sophisticated technology and highly trained personnel. Alternative methods for evaluating the effectiveness of wastewater treatment systems such as Waste Stabilisation Pond (WSP) systems are therefore in high demand, in particular in low and middle-income countries (LMICs) where resources are scarce. Modelling the performance of such systems may be a useful alternative. Such models may predict the concentration of antibiotic in each sedimentation pond and the removal efficiency.

To the author's knowledge, no attempt had been made so far to model antibiotics from WSP. Therefore, the work presented in this paper is pioneer work on the topic. The aim of the study was to present a simple dynamic model using STELLA® (isee Systems) software package to describe the most important removal processes of antibiotics through the WSP system Mafisa in Morogoro, Tanzania. The model is based on measured concentrations in loco of four antibiotics belonging to four different classes. The antibiotics analysed were trimethoprim (Trim), metronidazole (Met), Sul and Cip. Trim is a dihydrofolate reductase inhibitor, Met a nitroimidazole, Sul a sulphonamide and Cip a quinolone. All four antibiotics are on the World Health Organisation's Model List of Essential Medicines (2013).

We attempted to answer two questions with the developed model: (1) what is the applicability of the model to determine the removal efficiency of Trim from the WSP system? (2) Can the model be applied to other antibiotics? These questions are answered by calibration of the model based on a set of observations for Trim, followed by a validation of the model against another set of observations for Trim and by a validation of the model against observations for all four antibiotics. The developed model was afterwards used to assess the relative importance of the four removal processes (settling, outlet, hydrolysis + photolysis and biodegradation).

2. Materials and methods

2.1. Location

The Mafisa WSP is located in Morogoro, Tanzania. Morogoro is a town with approximately 300.000 inhabitants located 200 km inland from Dar es Salaam. Mafisa is located next to the Morogoro River in the Northern part of the town, in an area with housing and farming activities (Fig. 1) and receives wastewater from Morogoro town. The WSP system consists of two receiving ponds and six sedimentation ponds. The ponds have different functions as well as different dimensions. Pond 1 is an anaerobic sedimentation pond, pond 2 is a facultative pond, while ponds 3–6 are aerobic stabilization ponds. The dimension, flow rate and pH of the individual ponds are summarized in Table 1. After the sewage water is guided through Mafisa, it joins the Morogoro River. During dry season, the water in the river is low; hence, water from Mafisa is used for irrigation of the fields, mainly rice fields, surrounding Mafisa and the river. In the rainy season, the water joins the river immediately after outlet. Evaluation of the water level was based on a visual inspection.

2.2. Sampling and analysis

2.2.1. Sampling

Six sampling points were implemented and sampling was conducted in triplets. The sampling points and a schematic overview of Mafisa are shown in Fig. 1. At each of the sampling points, 2.5 L of water was collected in glass amber bottles. To prevent any degradation during sample preparation and transport, pH was adjusted on site to around 3 using hydrochloric acid (HCl) (Carlo-Erba) and measured using universal pH indicator strips. The samples were transported to the laboratory where they were filtrated twice. The first filtration was through a grade 5 filter paper with 20 μm particle retention from Munktell. The second filtration was through a grade 120H filter paper with 1–2 μm particle retention, also from Munktell. A standard addition method was applied when analysing the samples, by adding an internal standard (IS) to the samples (Runnqvist et al., 2010). After filtration the samples were divided to 3×800 mL and spiked with 100 μL 2.5 ppm internal standard mix (IS mix). The IS mix contained ciprofloxacin-d_8 (d-Cip), sulfamethoxazole-d_4 (d-Sul) and trimethoprim-d_3 (d-Trim).

2.2.2. Sample preparation

Approximately 800 mL of water sample, pH adjusted to 3 and spiked with 100 μL IS, was loaded onto Oasis®HLB 6 cm^3 200 mg (30 μm) cartridges from Waters (Milford, MA, USA) using a vacuum manifold and pump. The vacuum manifold was a VacMaster from IST (Sweden) and the pump was from ScanVac (Denmark). The drop-rate was adjusted to 1.5 mL/min. Prior to loading; cartridges were pre-conditioned with 2 mL methanol (MeOH) followed by 2 mL distilled water. After loading the water samples, the cartridges were air-dried using vacuum and stored at −18 °C before shipping to Denmark. During transport the cartridges were stored in a cooler with a coolant. Upon arrival in Denmark they were stored at −18 °C until use.

Prior to analysis, antibiotics were eluted from the cartridges with 8 mL mobile phase B (0.01% formic acid in MeOH) after washing with 2 mL 5% MeOH in water. The eluent was evaporated to dryness under a gentle stream of nitrogen at 33 °C. Nitrogen (99.8%) was supplied by Air Liquid (Ballerup, Denmark) and the evaporator was a Dionex SE 500 (CA, USA). Elution and evaporation was done in 12 mL amber tubes. Afterwards, the samples were reconstituted in 100 μL mobile phase B and 900 μL water. Samples were then transferred to Eppendorf tubes and centrifuged at 0.4472 RCF for 5 min