Kinetic and modeling of radiolytic decomposition of antibiotics

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Abstract

There has been recent growing interest in the presence of antibiotics in different environmental compartments. One considerable concern is the potential development of antibiotic-resistant bacteria in the environment, even at low concentrations. Kinetic experiments were conducted to compare two kinetic models for radiolytic decomposition of various antibiotics such as cephradine, amoxicillin, sulfamethazine, tetracycline, and lincomycin. Batch kinetic experiments with initial aqueous concentrations of 2, 6, 8, 20 mg/L showed the decomposition of antibiotics using gamma radiation followed a pseudo first-order reaction, and the dose constant increased with lower initial concentrations. For comparison of a kinetic model for radiolytic decomposition of various antibiotics, the Monod equation was used. The kinetic parameters of maximum reaction rates (k_{max}) and half-velocity coefficient (K_s) are obtained. The k_{max} values ranged from 0.29 to 0.99 μM/Gy. The K_s values for antibiotics decreased in the order of: cephradine > lincomycin > amoxicillin > sulfamethazine > tetracycline. Radiolytic decomposition of antibiotics by gamma radiation was well described by both a pseudo first-order reaction and the Monod equation.

Keywords: radiation, antibiotics, kinetics, modeling.

1 Introduction

Reports in the literature show that pharmaceuticals used in human and animal husbandry are present in soil, sediment, surface water, and groundwater. As important pharmaceuticals in today's medicine, antibiotics are used mainly to prevent and treat human and animal diseases, as well as for growth promoters in

WIT Transactions on Ecology and the Environment, Vol 109, © 2008 WIT Press
www.witpress.com, ISSN 1743-3541 (on-line)
doi:10.2495/WM080051

the intensive farming system. The antibiotics can be introduced to the various environments as parent compounds or metabolites by an excretion, disposal of unused or expired drugs, and accidental spills [1–3]. These antibiotics are incompletely degraded by the biological processes, and ultimately detected from μg/L to ng/L in aquatic environments [4–7]. The continual exposure to antibiotics, even at low concentration, may lead to developing antibiotic-resistance genes in bacterial strains and unknown adverse effects on humans and the ecosystem such as genotoxic potentials and disruption of the aquatic ecosystem [1,7–10].

Recently, many researches have been conducted on the treatment of antibiotics found in groundwater, surface water, and wastewater. Ingerslev et al. [11] showed that antibiotics under aerobic and anaerobic conditions were not readily biodegradable. Advanced oxidation processes (AOPs) have been suggested in recent years as a suitable alternative for the removal of refractory organic compounds found in a variety of environments. AOPs by using free radicals such as the hydroxyl radical (OH) include ozone, ozone/UV, TiO_2 photo catalysis, Fenton's reaction, H_2O_2/UV, H_2O_2/O_3 and ionizing radiation [4,12,13]. Ozone is a strong oxidant and disinfectant for drinking water and industrial wastewater, and can be effectively used as a pretreatment process to improve the biological degradation efficiency of wastewater containing antibiotics [4,14,15]. Abellan et al. [15] recently reported that 82% of sulfamethoxazole degradation efficiency was obtained with UV and TiO_2 as a catalyst.

Among these AOPs, ionizing radiation has promise as a removal process of the toxic organic pollutants. In water radiolysis, the primary species (OH, e_{aq}^-, and H) and molecular products (H_2, H_2O_2) are generated by the following equation [16]:

$$H_2O \sim\sim> \cdot OH + H\cdot + e_{aq}^- + H_2 + H_2O_2 + H_3O^+ \quad (1)$$

These reactive species/radicals react with target compounds for decomposition of solutes present in water. There have been a number of studies examining the radiolytic decomposition of the refractory organic compounds such as nitrobenzene, diazinon pesticide, methyl tert-butyl ether (MTBE), polychlorinated biphenyl (PCB), and trinitrotoluene (TNT) [17–22]. Yu et al. [23] also showed that 30 mg/L of cefaclor was completely degraded after 1 kGy by gamma radiation. However, it is very difficult to compare the radiolytic removal efficiencies for various toxic organic chemicals due to the use of different kinetic figures-of-merit. The development of the most reliable kinetic models and figure-of-merit will be useful to predict the irradiation dose requirement and obtain an optimization in an irradiation system.

This study focused on the evaluation of the kinetic parameters of different degradation models for the radiolytic decomposition of different groups of antibiotics. Among many kinds of antibiotics, the target antibiotics studied were: β–lactams (penicillins: **amoxicillin** and cephalosporins: **cephradine**); non β–lactams (tetracyclines: **tetracycline**, sulfonamide: **sulfamethazine** and lincosamides: **lincomycin**). The selected chemicals represent the most important antibiotics used in human and animal. The objectives of this study were: 1) to

investigate the removal of the five antibiotics in aqueous solution by gamma irradiation; and 2) to compare the decomposition kinetics of the antibiotics by the three figures-of-merit (dose constant and the Monod equation parameters (k_{max} and K_s)).

2 Materials and method

2.1 Chemicals

Amoxicillin, cephradine, tetracycline, lincomycin and sulfamethazine were obtained from Sigma-Aldrich, Co. (St Louis, MO, USA) with a purity higher than 99%. The chemical structures of the antibiotics used in this study are shown in Figure 1. All chemicals used for HPLC analysis were of HPLC grade and purchased from Sigma-Aldrich, Co. (St Louis, MO, USA), J.T. Baker (USA) and Kanto Chemical Co., Inc. (Japan). All solutions were prepared with Milli-Q purified water (Millipore).

2.2 Irradiation sources

Gamma irradiations were performed with a high-level ^{60}CO source (Nordion Inc., Canada) at the Korea Atomic Energy Research Institute (Jeongup, Korea). The radioactivity of the source was around 1.47 X 10^{17} Bq (= 397949 Ci). Aqueous antibiotics solutions were irradiated in 50 mL screw cap bottles and 2 mL septa-capped vial without any headspace. The solutions were prepared 24 hrs prior to irradiation and stored at 4°C. All the samples were prepared in equilibrium with an atmospheric pressure and room temperature (22°C±2) before irradiated, and were sealed with screw caps to avoid the contact with air.

Table 1: Column information of high performance liquid chromatography used in the analysis of antibiotics.

	Column stationary phase	Injection volume (μl)	Flow rate (mL/min)	UV wave length (nm)	Eluent
Amoxicillin	C8[a]	50	1.0	230	25 mM Potassium phosphate pH4.6 : Methanol (95 : 5)
Cephradine	Polar RP[b]	50	1.0	254	20 mM Ammonium formate pH3.5 : Methanol (65 : 35)
Sulfamethazine	C18[c]	50	0.6	254	0.1% Formic acid in water : 0.1% Formic acid in Acetonitrile (70 : 30)
Tetracycline	C8[a]	50	1.0	355	0.01 M Oxalic acid : Methanol : Acetonitrile (72: 8 : 20)
Lincomycin	C18[d]	75	0.7	210	1 mM Ammonium formate : Acetonitrile (65 : 35)

[a]Luna 5μ C8(2) 100A 150 × 4.6 mm (Phenomenex, Torrance, CA, USA).
[b]Synergi 4μ Polar-RP column 150 × 4.6 mm (Phenomenex, Torrance, CA, USA)).
[c]Onyx Monolithic C18 100 × 3.0 mm (Phenomenex, Torrance, CA, USA)).
[d]Zorbax sb-C18 250 × 3.0 mm (Agilent Technologies, Santa Clara, CA, USA).

2.3 Analytical methods

Antibiotics concentrations in the aqueous samples were measured using high performance liquid chromatography (HPLC) using an Agilent 1200 Series HPLC (Agilent Technologies, Santa Clara, CA, USA), equipped with an UV absorbance detector. The operation conditions of HPLC measurement are listed in Table 1. Triplicate samples for all experiments were analyzed for the reproducibility.

3 Results and discussion

3.1 Radiolytic decomposition of antibiotics

A typical decomposition curve of antibiotics by gamma radiation is shown in Figure 1. Experiments were conducted in quintuplicate to ensure that the kinetic results were reproducible for each dose amount tested. The aqueous concentrations of antibiotics irradiated were all 30 mg/L in batch bottles without a head space (total volume = 150 mL). The irradiation doses were up to 4 kGy. As shown in Figure 1, the decomposition extent of antibiotics increased as the absorbed dose increased. Most of the antibiotics of 30 mg/L were completely degraded after 1 kGy, but sulfamthazine needed 4 kGy for the complete decomposition.

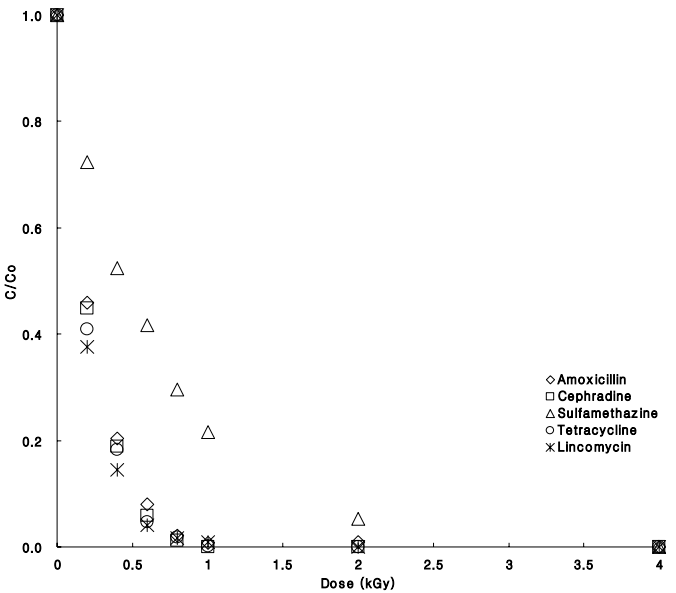

Figure 1: Radiolytic decomposition of antibiotics of 30 mg/L by gamma radiation.

3.2 Determination of chemical reaction constants for radiolytic decomposition of antibiotics

Most radiolytic degradations of organic compounds are represented by equation (2), which has been used to describe dose-dependant degradation rates [22, 24]

$$C = C_0 e^{-dD} \tag{2}$$

where C is the aqueous concentration of antibiotics, d the dose constant in reciprocal dose units, and D the absorbed dose.

Batch kinetic experiments were performed to study the initial aqueous concentration dependency on antibiotics decomposition by gamma radiation. Initial aqueous concentrations of antibiotics ranged from 2 to 50 mg/L and the absorbed doses from 0 to 120 Gy. All of the kinetic experiments were performed at the ambient temperature and atmospheric conditions.

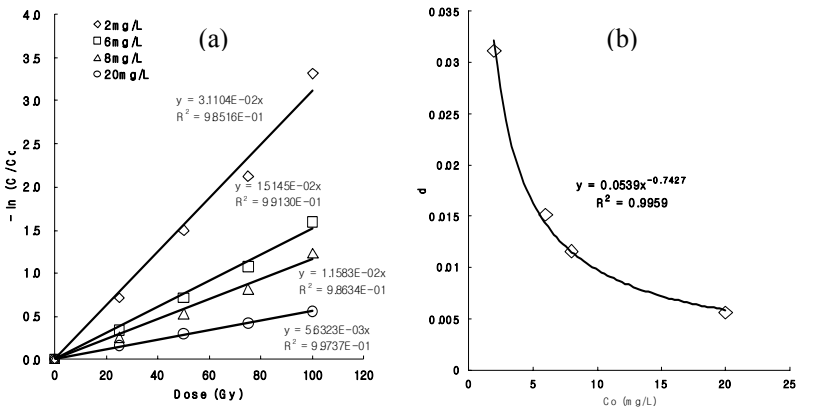

Figure 2: Kinetic results of radiolytic decomposition of cephradine at different initial aqueous concentrations: initial concentration dependency on dose constants.

Table 2: Dose constants of antibiotics.

Antibiotics	Dose constant (d) (Gy^{-1})	R^2
Cephradine	$0.0539Co^{-0.7427}$	0.9959
Amoxicillin	$0.0633Co^{-0.8602}$	0.9996
Sulfamethazine	$0.0541Co^{-0.8983}$	0.987
Tetracycline	$0.1772Co^{-0.853}$	0.9783
Lincomycin	$0.0515Co^{-0.7244}$	0.9908

Figure 2 shows the results of a radiolytic decomposition of cephradine at different initial aqueous concentrations. The kinetic experiments were performed in duplicate, which revealed near identical results. All of the experimental data (Figure2) was fit to the pseudo first-order reaction model, which showed statistically reliable results (R^2 values higher than 0.985). The dose constants of five antibiotics at different initial concentrations are listed in Table 2.

3.3 Determination of kinetic parameters of the Monod equation

The radiation dose-dependent kinetics of removal reaction of the antibiotics were investigated by the measurement of the radiolytic degradation rates at different initial concentrations of the antibiotics. The radiolytic decomposition of a given antibiotic can be described using the Monod equation (equation 3), frequently used in enzyme kinetic studies [25, 26, 27, 28, 29, 30].

$$r = \frac{k_{\max} \times \mathrm{S}}{\mathrm{K_s} + \mathrm{S}} \tag{3}$$

Where r is the removal rate (μM/Gy) of the antibiotics concentration, k_{max} maximum specific reaction rate (μM/Gy), K_s half-velocity coefficient (μM) and S the aqueous concentration of antibiotics (μM).

Figure 3 shows the Monod equation curve of radiolytic decomposition of cephradine. Table 3 presents the k_{max} and K_s values obtained by fitting the experimental data. The highest k_{max} value was 0.99 μM/Gy of cephradine and the lowest K_s 1.49 μM of tetracycline, indicating that tetracycline is rapidly decomposed at lower concentrations while cephradine at higher concentrations.

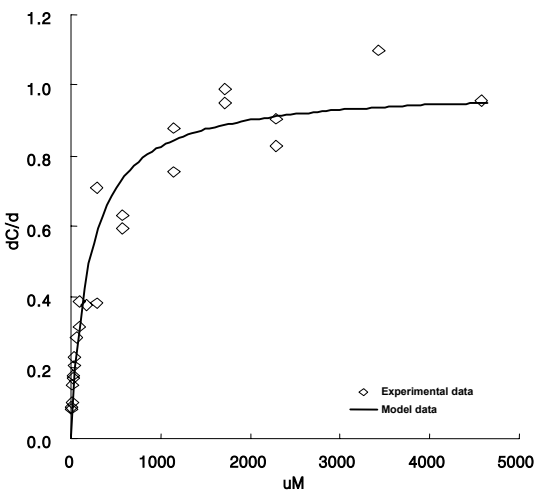

Figure 3: Initial degradation rates of cephradine by gamma radiation.

Table 3: Kinetic parameters of the Monod equation for radiolytic decomposition of five antibiotics in aqueous solution.

Antibiotics	k_{max} (μM/Gy)	K_s (μM)
Cephradine	0.99	199.64
Amoxicillin	0.56	56.42
Tetracycline	0.43	1.49
Sulfamethazine	0.29	6.36
Lincomycin	0.58	56.46

4 Conclusion

Radiation process was very effective for the degradation of antibiotics in aqueous solution. This study was designed to compare the figures-of-merit: chemical reaction dose constant, *d*, and kinetic parameters of the Monod equation, k_{max} and K_s. Mincher and Curry [24] suggested that the dose constant, *d*, is the most appropriate figure-of-merit for the majority of systems, due to its precision. However, the Monod equation model was not compared to the other figures-of-merit. The kinetic experimental results showed that the kinetic parameters of the Monod equation also represent the radiolytic decomposition of antibiotics at different concentration and are very useful to predict radiation dose requirements. The future study will include the comparison of the two different models for radiolytic decomposition of antibiotics at different concentrations.

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