Antibiotic Contamination in Wastewater of Phranangklao Hospital

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Abstract

The issue of residual antibiotics in wastewater from hospital treatment systems has become a major factor driving the emergence of antimicrobial resistance globally. This study investigates the sources and evaluates the extent of antibiotic contamination in hospital wastewater, with a case study of Phranangklao Hospital in Nonthaburi Province. Wastewater samples were collected from three representative buildings: Jindamanee Building (Outpatient Department Building; OPD Building), the 18-story Medical Center Building (Inpatient Department Building; IPD Building) and Ngeun Tung Daeng Building (Hemodialysis Building) in April 2024. Samples were also taken from the hospital's centralized wastewater treatment system, including influent, the flow measurements tank, effluent from the sedimentation tanks using both grab sampling and composite sampling methods, and the effluent discharge points in December 2023, during the dry season to minimize contamination from rainwater. Sampling was conducted between 10:00 a.m. and 12:00 p.m., during peak hospital water usage periods. The results showed that the IPD Building had the highest contamination levels, followed by the OPD Building and the Hemodialysis Building. Sulfamethoxazole (SMX) was the most frequently detected antibiotic across all operational units, indicating its environmental persistence. The hospital's centralized wastewater treatment system was able to reduce certain antibiotic residues, such as ciprofloxacin (CIP), by more than 90%, but SMX was still found in the treated effluent. The study highlights the need for improvements in hospital wastewater treatment systems to reduce antibiotic residues in effluent, as well as to minimize contamination at the source, such as from various medical activities. The findings can serve as a foundation for developing effective wastewater treatment measures and contamination control strategies to prevent long-term environmental and health impacts.

Introduction

Currently, the contamination of antibiotics in hospital wastewater has been increasingly detected in various areas of Thailand. This issue has garnered significant attention as the wastewater treatment systems in many hospitals are inefficient in removing antibiotics and their derivatives. This leads to residual antibiotics in the environment, particularly in natural water bodies that receive untreated or inadequately treated hospital wastewater [1-2]. This problem has become a critical concern that is being studied and addressed globally. Residual antibiotics in the environment can spread widely, with contamination levels in natural environments ranging from nanograms to micrograms per liter [3-4]. These antibiotics are persistent and resistant to natural degradation, resulting in environmental accumulation. This poses a risk of antimicrobial resistance (AMR) that can spread through food chains and impact humans, particularly in cases where bacterial infections fail to respond to treatment, potentially leading to fatalities [5-7]. Antibiotics found in hospital wastewater and natural environments are from various groups, such as beta-lactams, tetracyclines, and sulfonamides. Thus, reducing the contamination of antibiotics in hospital wastewater before it enters natural environments, especially water bodies, is the best preventive approach. [8] One key strategy is improving centralized wastewater treatment systems in hospitals to enhance treatment efficiency. Additionally, efforts should focus on reducing antibiotic contamination at the source, originating from various hospital activities. An assessment of relevant legal tools in Thailand reveals that hospital wastewater is regulated under the "Standards for Controlling Wastewater Discharge from Certain Types and Sizes of Buildings." [9] In this regulation, hospital wastewater must undergo additional bacterial control before discharge, limiting Total Coliform Bacteria to no more than 5,000 MPN/100 ml, Fecal Coliform Bacteria to no more than 1,000 MPN/100 ml, and Residual Chlorine to a range of 0.2-1.0 mg/l as Cl_2, in compliance with the Hospital Accreditation (HA) standards set by the Institute of Hospital Quality Improvement and Accreditation under the Ministry of Public Health [10]. However, there are currently no established thresholds for antibiotics discharged into the environment. Preventing and controlling antibiotic contamination requires process improvements related to the removal of antibiotics from hospital wastewater sources. This study aims to identify and quantify antibiotic residues in wastewater from Phranangklao Hospital. It evaluates the effectiveness of current treatment processes in removing these contaminants. Seasonal variations in antibiotic levels will also be analyzed. The research assesses potential environmental and health risks, particularly in relation to antimicrobial resistance. Finally, it proposes strategies to improve hospital wastewater management and reduce antibiotic pollution.

Equipment and Methods

Survey on the usage and dispensing of target antibiotics in 8 groups or at least 5 groups of Phranangklao Hospital under the guidance of the Department of Health and surveying sources of pollution to find the origins of antibiotics. Collecting wastewater samples was done by grab sampling aligned with the sampling requirements for wastewater characteristics following the wastewater standard [10]. Before collecting samples, all containers were cleaned and sterilized to prevent contamination. Samples were immediately placed in sterile, amber glass bottles to minimize photodegradation of antibiotic compounds. Transportation to the laboratory was conducted in cooled containers (4°C), and analysis was carried out within 24 hours to ensure sample integrity. To study the origins of hospital antibiotics, wastewater samples were collected from representative buildings: Jindamanee Building (Outpatient Department Building; OPD Building), 18-story Medical Center Building (Inpatient Department Building; IPD Building), and Ngeun Tung Daeng Building (Hemodialysis Building), as shown in Figure 1. Collection of water samples from the Completely Mix Activated Sludge (CAS) was made as follows: grab sampling for influent (S1), flow measurements tank (S2), sedimentation tank (S3), composite sampling for sedimentation tank (S4), and effluent (S5), as shown in Figure 2. Sampling was conducted from 10:00 to 12:00 a.m., during the peak hospital water usage periods (it is thought that there was a high use of

antibiotics during that time period). The highest wastewater flow rates were recorded between 8:00 a.m. and 3:00 p.m. [11]. The confirmation of the type and amount of antibiotic residues in the wastewater sources and the Completely Mixed Activated Sludge (CAS) wastewater treatment system of Phranangklao Hospital was conducted. The type and amount of antibiotics detected in the wastewater were analyzed by preparing wastewater samples for antibiotic quantification following EPA Method 1694 (2007) [12] and analyzing them using LC/MS/MS and LC/MS/MS-QTOF techniques.

Experimental Results and Discussion

Antibiotic Contamination in Wastewater from Phranangklao Hospital

From the survey of wastewater from buildings on March 8, 2024, along with interviews with responsible officials, it was found that most antibiotic contamination in hospital wastewater comes from the excretion of waste of the patients, which is a major source of antibiotic contamination in the hospitals. The source of antibiotic contamination from service buildings is the OPD Building, IPD Building, and Hemodialysis Building. Collecting wastewater samples from the service buildings was performed during the dry season (April 2024) to reduce the chance of contamination from rainwater, as shown in Figure 1: For the OPD Building, wastewater samples were selected from the 3 toilets of outpatient users only to avoid collection of wastewater from the toilets of hospital staff. The wastewater sampling location in the building is an equalization tank (EQ). For the IPD Building, wastewater samples are taken from the overnight patient water usage activities only, such as toilets, showers, and sinks. The wastewater sampling point is outside the building, which is the collection point of the buildings before entering Completely Mix Activated Sludge (CAS). The hemodialysis building is used for dialysis only but has 3 main water usage activities including (1) dialysis water for patients during treatment (2) water for washing vessels and dialysis cones, and (3) RO wastewater. Due to the nature of water usage in the Hemodialysis Building, it is necessary to install specific water sampling equipment for each activity to enable detailed analysis of

residual antibiotics, as the combination of wastewater from different activities may affect the analysis of antibiotic contamination resulting from the treatment of hemodialysis patients.

Antibiotic content in wastewater from the representative buildings

The study results revealed that the IPD Building had the highest amount of antibiotic contamination, followed by the OPD Building and Hemodialysis Building, respectively, as shown in Table 1, which is consistent with the data of Panadda, 2562 [13], Chiemchaisri W., *et al.* (2022) [14], Hamjinda *et al.* (2018) [15], Muhammad T.K *et al.* (2021) [16], Rozman U., *et al.* (2020) [17], which reported higher antibiotic loads in wastewater from inpatient activities due to greater water use. Sulfamethoxazole (SMX) was the most frequently detected antibiotic, present in all collected samples, reflecting its persistence in the environment. Previous studies corroborate SMX's widespread presence in river water, wastewater, and imported fish, highlighting its environmental resilience. According to Si LY.L. *et al.* (2022) [8], a study on antibiotics in rivers worldwide across 76 countries, encompassing over 600 research articles from 1999 to 2021 and collecting more than 90,000 data points, revealed the presence of 169 types of antibiotics in river water and sediments. Sulfamethoxazole (SMX) and trimethoprim (TMP) were found to be the most persistent, with average concentrations in river water at 4,320 ng/L and 1,200 ng/L, respectively. The data also indicated that the continents with the highest levels of antibiotic contamination in rivers were Africa, followed by Asia. These findings suggest that SMX and TMP can serve as effective indicators of environmental antibiotic contamination. The persistence of SMX and other antibiotics in hospital wastewater poses a significant risk for the development and spread of antimicrobial resistance (AMR). Their continued presence in effluents discharged into municipal wastewater systems or natural water bodies could contribute to the proliferation of resistant bacteria, which has global public health implications. Therefore, monitoring and managing antibiotic residues in hospital wastewater is essential for reducing the risk of AMR spread.
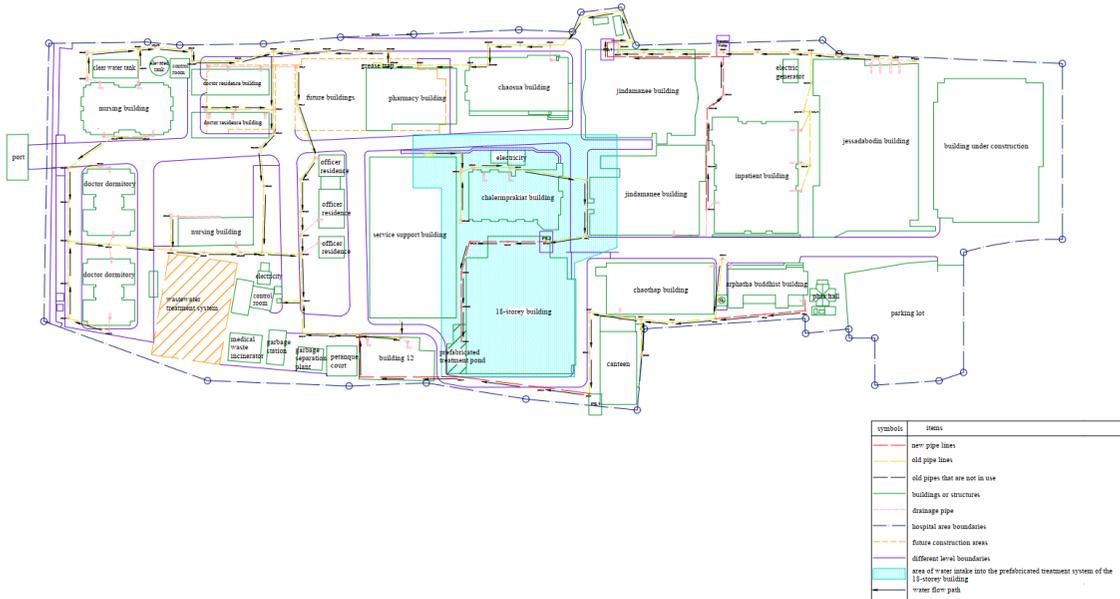
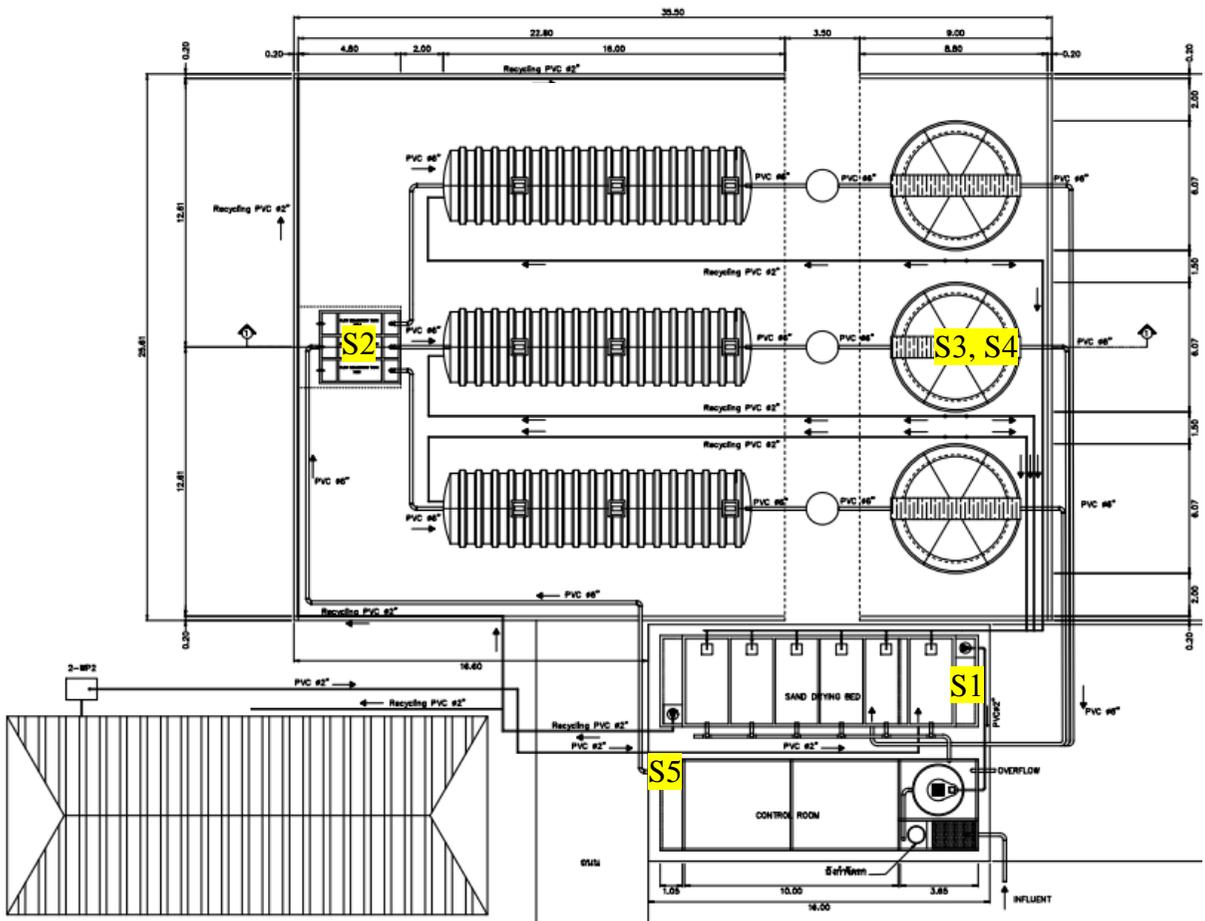**Figure 1** Diagram of wastewater collection system of Phranangklao Hospital

Figure 2 Wastewater sampling point of the Completely Mix Activated Sludge (CAS)
wastewater treatment system

Table 1 Amount of antibiotics detected in wastewater of representative buildings

Group	Antibiotic group	Antibiotic name	Antibiotic Concentration (μg/L)[1]		
			OPD building	IPD building	Hemodialysis building
1	Beta-Lactam	Ampicillin	0.061-1.134	0.709-3.645	0.412-2.219
3	Cephalosporin	Ceftriaxone	NF-0.238	NF-0.184	NF-3.619
4	Macrolide	Erythromycin	NF-10.129	NF	NF
5	Tetracycline	Oxytetracycline	NF-0.504	NF-0.647	NF-0.176
		Doxycycline	NF	NF	NF
6	Quinolones	Ofloxacin	NF-1.131	0.275-2.511	NF
		Levofloxacin	NF-1.152	0.264-2.377	NF
		Norfloxacin	0.137-0.626	0.148-1.926	NF-0.059
7	Sulfonamide	Sulfadiazine	NF	NF	NF-0.674
		Sulfamethoxazole	NF-0.924	0.369-40.620	NF
		Trimethoprim	NF-0.051	NF-4.097	NF

Note: [1] Wastewater samples were collected from buildings before entering the hospital's centralized wastewater treatment system during the dry season (April 2024).

NF = Not Found

Amount of antibiotics detected in the centralized wastewater treatment system of Phranangklao Hospital

Confirmation of the type and amount of antibiotic residue in the Completely Mix Activated Sludge (CAS) wastewater treatment system of Phranangklao Hospital was made. Therefore, the research team chose to collect wastewater samples in December 2023 (dry season) at the water sampling points, including grab sampling for influent (S1), flow measurements tank (S2), sedimentation tank (S3), composite sampling for sedimentation tank (S4), and effluent (S5), as shown in Figure 2. Table 2 shows the study results of the amount of antibiotics detected in wastewater of the Completely Mix Activated Sludge (CAS) wastewater treatment system in December 2023. It was found that the wastewater entering the system had the highest amount of contamination of ciprofloxacin (2.091-6.456 μg/l), followed by ampicillin (0.474-4.747 μg/l) and ceftriaxone

(NF-2.726 μg/l), respectively. The difference in the amount of antibiotics detected by the method of collecting water samples from the sedimentation tank using grab sampling (S3) or composite sampling (S4) showed no difference. However, composite sampling is recommended due to the operational nature of the hospital's wastewater treatment system, which functions intermittently, especially during peak hours (10:00–12:00 a.m.). When considering the half-life of antibiotics in water, it was found to be less significant compared to the solubility of each antibiotic. For instance, amoxicillin (AMX), with a half-life of 20 days [18], was not detected in any of the sampled water at positions S1–S5. Conversely, sulfamethoxazole (SMX), which has a shorter half-life of only 4.9 days [19], was detected in all samples. The degradation ratio of SMX varied across different treatment stages, from influent wastewater (S1) through primary treatment (S2) and secondary treatment (S3, S4). While SMX showed partial degradation during

Table 2 Amount of antibiotics detected in the Conventional Activated Sludge (CAS) system of Phranangklao Hospital

Group	Antibiotic group	Antibiotic name	Antibiotic Concentration (µg/L) [1]					Half-life in water [2]
			S1	S2	S3	S4	S5	
1	Beta-Lactam	Amoxicillin	NF	NF	NF	NF	NF	20 days
		Ampicillin	0.474-4.747	0.792-2.481	0.094-0.420	0.174-0.468	0.111-0.577	27 days
3	Cephalosporines	Ceftriaxone	NF-2.726	NF-2.530	NF	NF	NF	4.1 days
4	Macrolide	Clarithromycin	0.011-0.292	0.016-0.372	0.003-0.046	0.005-0.050	0.005-0.045	2 hr.
5	Tetracyclines	Oxytetracycline	0.377-0.400	0.295-0.450	0.340-0.498	0.320-0.403	0.365-0.400	66 hr.
		Doxycycline	0.450-0.665	0.402-0.618	0.251-0.301	0.278-0.307	0.241-0.281	107 hr.
6	Quinolone	Ofloxacin	0.435-1.301	0.773-2.542	0.388-0.662	0.274-0.692	0.391-0.488	10.6 days
		Levofloxacin	0.388-1.310	0.817-2.493	0.398-0.626	0.254-0.695	0.406-0.477	6.3 days
		Norfloxacin	1.062-1.663	0.648-1.178	0.083-0.155	0.091-0.129	0.122-0.133	77 days
		Ciprofloxacin	2.091-6.456	1.304-3.189	0.086-0.224	0.098-0.203	0.165-0.197	< 46 hr.
7	Sulfonamide	Sulfadiazine	1.469-2.161	1.237-1.409	0.603-0.803	0.635-0.814	0.615-0.843	13.2 hr.
		Sulfathiazole	0.063-0.090	0.049-0.061	0.037-0.041	0.034-0.044	0.033-0.044	13 days
		Sulfamethoxazole	1.753-1.961	1.291-2.995	0.089-0.189	0.124-0.289	0.332-0.610	4.9 days
-	Diaminopyrimidines	Trimethoprim	0.266-0.447	0.190-0.412	0.009-0.014	0.010-0.014	0.010-0.014	< 11.8 days
-	Lincosamide	Lincomycin	0.021-0.038	0.030-0.049	0.007-0.009	0.007-0.009	0.006-0.009	12 days

Note: [1] Wastewater samples were collected from the Conventional Activated Sludge (CAS) system during the dry season (December 2023)
[2] limit of detection (LOD) = 0.003 µg/L or 3 ng/L
NF = Not Found

secondary treatment, it still persisted in the treated effluent (S5). This finding suggests that the solubility of antibiotics plays a more critical role in their detectability in wastewater than their half-life alone. Some antibiotics, despite having a relatively long half-life, such as amoxicillin (AMX), with 20 days, may not be detected in the water samples if they are poorly soluble in water, readily degraded under environmental conditions (e.g., light, temperature), or strongly adsorbed onto sludge particles during the treatment process. In contrast, antibiotics with high water solubility and low affinity for adsorption, such as sulfamethoxazole (SMX), may remain present throughout the treatment stages and persist in the final effluent, even though their half-life in water is shorter. SMX is one of the most commonly found and environmentally persistent antibiotics, particularly in water bodies, as confirmed by Si LY.L., *et al.* (2022) [8]. Although ciprofloxacin (CIP) showed higher concentrations at S1 compared to other antibiotics, it was reduced by over 90% during the wastewater treatment process. Consistent with the report by Chiemchaisri *et al.* (2022) [14], it was found that certain groups of antibiotics were removed by more than 80% in the aeration tank and were adsorbed onto sludge. Therefore, many antibiotic resistant infections can be effectively treated using the CAS system. Consequently, it is highly likely that certain types or groups of antibiotics may not be detected in the treated effluent from hospital wastewater treatment systems.

These findings affirm that the CAS system used at Phranangklao Hospital is moderately effective at removing certain antibiotics, particularly those that adsorb onto sludge or degrade readily. However, the persistence of SMX and similar compounds in the effluent highlights the limitations of conventional treatment technologies. To address this, hospitals could consider Implementing advanced treatment technologies such as membrane filtration, ozonation, or advanced oxidation processes (AOPs) to improve removal efficiency. Segregating pharmaceutical wastewater streams at the source (e.g., from pharmacy or ICU departments) for specialized treatment. Conducting routine surveillance of antibiotic residues in effluent and sludge to track long-term trends and ensure compliance with environmental safety standards.

Conclusions

The study of residual antibiotic concentrations in wastewater from buildings and the Conventional Activated Sludge (CAS) system of Phranangklao Hospital in Nonthaburi Province has provided insights into the levels of antibiotic contamination in hospital wastewater. These findings can inform the development and improvement of effective strategies for addressing antibiotic contamination. The survey revealed that Phranangklao Hospital utilizes and dispenses antibiotics across 8 target groups. Most antibiotics are treated with more than 50% efficiency at each stage of the Conventional Activated Sludge (CAS) system. However, an analysis of the types and concentrations of residual antibiotics found that ampicillin, clarithromycin, ciprofloxacin, sulfathiazole, and lincomycin were not detected in wastewater from the representative buildings. Some residual antibiotics were observed to increase in concentration within the centralized treatment system, possibly due to wastewater inflow from other service buildings. On the other hand, the concentrations of certain residual antibiotics in wastewater from representative buildings decreased upon entering the hospital's centralized wastewater treatment system, as they were biodegraded during the process. For example, amoxicillin was completely degraded within the

treatment system. These findings highlight both the effectiveness and limitations of the current CAS system in treating antibiotic residues. The presence of certain antibiotics at increased concentrations after centralized treatment suggests the need for more comprehensive sampling and monitoring across all sources within the hospital. Additionally, the results emphasize the importance of optimizing existing wastewater treatment protocols and considering advanced treatment technologies to ensure the effective removal of residual antibiotics. This is crucial not only for environmental protection but also for mitigating the risk of promoting antimicrobial resistance in aquatic ecosystems.

Acknowledgments

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