Environmental Remediation of Antineoplastic Drugs

Subjects: Oncology
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The global burden of cancer is on the rise, and as a result, the number of therapeutics administered for chemotherapy is increasing. The occupational exposure, recalcitrant nature and ecotoxicological toxicity of these therapeutics, referred to as antineoplastic (ANP) drugs, have raised concerns about their safe remediation. This review provides an overview of the environmental source of ANPs agents, with emphasis on the currently used remediation approaches. Outpatient excreta, hospital euents, and waste from pharmaceutical industries are the primary source of ANP waste. The current review describes various biotic and abiotic methods used in the remediation of ANP drugs in the environment. Abiotic methods often generate transformation products (TPs) of unknown toxicity. In this light, obtaining data on the environmental toxicity of ANPs and its TPs is crucial to determine their toxic e ect on the ecosystem. We also discuss the biodegradation of ANP drugs using monoculture of fungal and bacterial species, and microbial consortia in sewage treatment plants. The current review e ort further explores a safe and sustainable approach for ANP waste treatment to replace existing chemical and oxidation intensive treatment approaches. To conclude, we assess the possibility of integrating biotic and abiotic methods of ANP drug degradation.

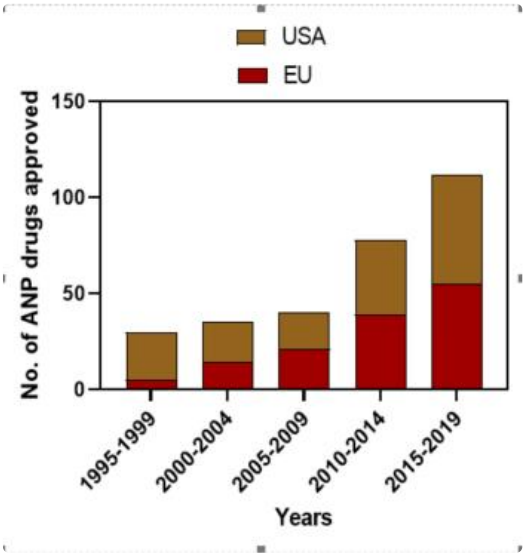
Keywords: antineoplastic drug ; environment ; toxicity ; remediation ; biodegradation

1. Introduction

Last few decades have experienced rising concerns over the release of pharmaceutical drugs into the environment. Though pharmaceutical compounds have targeted effects on the human body, the knowledge about the direct impact of their transformation products (TPs) and metabolites on other organisms and indirect effects on human health is scarce. Antineoplastic (ANP) drugs (also known as anticancer or cytostatic) are a specific group of pharmaceutical compounds which prevent, inhibit, or terminate the development of cancer. However, due to their non-specific mode of action, affecting both cancerous and healthy cells, ANP drugs exhibit cytotoxic, genotoxic, mutagenic, carcinogenic and teratogenic effects in all eukaryotic cells [1][2][3]. Nevertheless, due to their low environmental concentrations (10–100 ng/L or below), there is not enough evidence to accurately assess whether or not ANPs have an impact on the environment[4]. However, since they are designed to disrupt or prevent cellular proliferation, usually by interfering in DNA synthesis, their fate and transport in the environment should be explored.

The World Health Organization estimated the global burden of cancer at 18.1 million new cases and 9.6 million deaths in 2018 [5]. As per an evaluation by the American Cancer Society, 1,806,590 new cancer cases and 606,520 cancer deaths are projected to occur in the United States by 2020 [6]. In compliance with this trend of increasing cancer prevalence, new ANP drugs are also being designed, tested, and manufactured at an increasing rate [7]. Over the past few years, 70 new ANP drugs have been released to treat 20 variants of tumors (cancerous growths), the number of ANP drugs has expanded by more than 60% [8]. More than 500 companies are currently pursuing ANP drug development, with 300 companies having cancer drugs under clinical development stages [8]. Figure 1a,b shows the total number of ANP drugs approved in USA and EU, and total ANP molecules under different phases of development, respectively. In 10 years from 2010 to 2020, ANP drug production is expected to double [9]. The production of novel ANP drugs has varied greatly across countries over the years. For example, in 2004, Canada and Australia consistently produced a higher volume of ANP drugs (51 and 39 for Australia and Canada, respectively) whereas United States and Germany produced significantly lower volume (29 and 17 for Germany and United States, respectively). On the other hand, in 2014, United States and Germany produced higher volume of novel ANP drugs [10]. A comparative global heatmap of some prominent ANP drug producing countries in 2004 and 2014 is shown in Figure 1c. The raw data of Figure 1c is given. In addition to the production, it is also crucial to highlight the sites where ANPs are mostly released into the environment. However, there is not enough data to categorize sites in terms of ANP emission into the environment. The number of publications on occurrence of ANP compounds in environment is scarce, and most of studies to date are almost exclusively focused on

Europe [11]. Nevertheless, the number of cancer cases in different countries can be a governing factor that dictates the introduction of these compounds in the environment. The country with highest number of cases will consume the most ANP drugs and hence there will be a greater probability of introduction of these compounds into the environment. The global heatmap of the number of cancer cases in different countries per 100,000 people is given in Figure 1d. The raw data was obtained from the GLOBOCAN online database [12].



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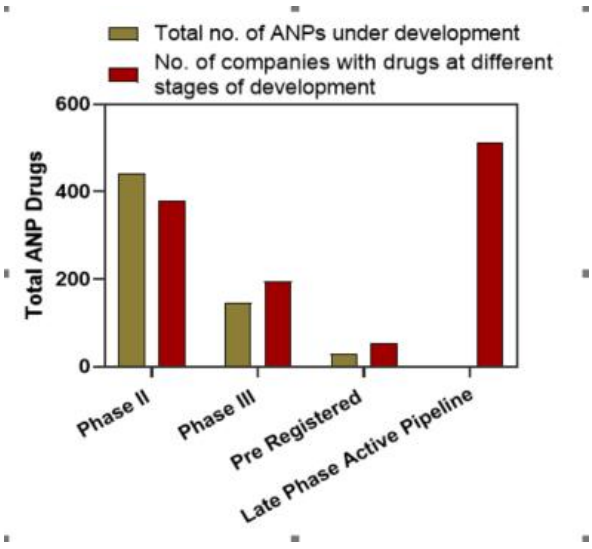

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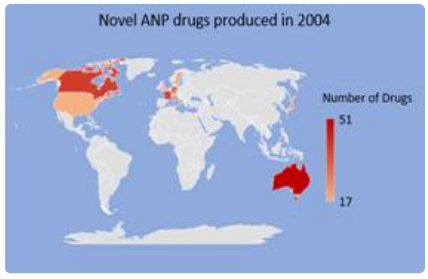

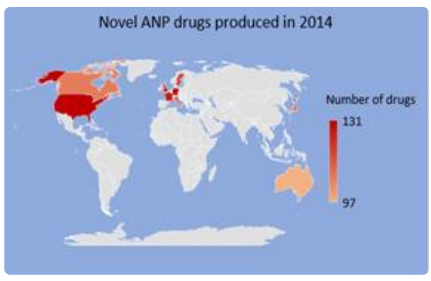

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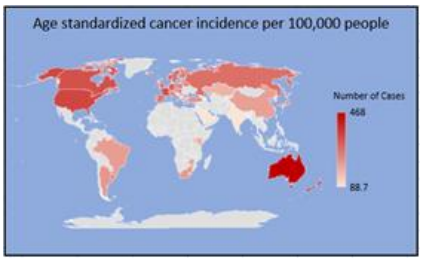

(d)

Figure 1. (**a**) Number of antineoplastic (ANP) drugs approved between 1995 and 2019 in USA and European Union (EU) [13][14][15]; (**b**) The Global Late Phase Oncology Pipeline in 2015; (**c**) comparative global heatmap of ANP drug produced in some select countries in 2004 and 2014 ; (**d**) age standardized cancer incidence in select countries in 2018 . (Phase II and Phase III refers to ANP drugs that are at second and third phase of clinical trials, respectively).

ANP drugs are categorized into different groups based on the organ or system on which they act and their therapeutic, pharmacological, and chemical properties [16]. These different categories of ANP drugs differ in their chemical structures and physicochemical properties. The physicochemical properties play an indispensable role in the potential fate of these drugs in the environment [17]. Building a physicochemical profile of ANPs will allow for their partitioning, and help us study the fate of ANPs within aquatic and terrestrial ecosystems [18]. In total, 102 active antineoplastic drugs have been identified which are environmentally relevant [19]. Section 2 of this review describes the parameters that determine the fate and distribution of ANPs in the environment.

Though used in minimal quantity, ANP drugs persist in the environment [19] and can be harmful even if present under low concentrations [20][21]. All ANP compounds are potent immunosuppressive agents and have a high pharmacological potency that is fatal to aquatic and terrestrial organisms [22]. Significant disadvantages and environmental concerns associated with the usage of ANP drugs include the following: (i) inevitable contamination of natural ecosystems (terrestrial and aquatic) by the drugs themselves and their potentially toxic transformation products (TPs); (ii) scarcity of complete and coherent knowledge on the environmental fate of these drugs, its human metabolites, or TPs; (iii) their biomagnification at various trophic levels which can have adverse effects on the flora and fauna of the contaminated ecosystems.

The aforesaid environmental concerns have made it imperative to develop a safe, economical, and environmentally friendly process to remediate residual ANP drugs in the ecosystem. To date, limited studies have been successful in using a single microbial system or microbial consortia for the complete or partial elimination of ANP wastes [23][24][25][26]. The integration of microbial bioremediation systems with abiotic remediation techniques can be beneficial as it will decrease the requirements of harsh chemicals and may also reduce the generation of toxic degradation products [27]. This review presents a case for integrating abiotic and biotic modes of ANP degradation. With our decade-long continuous exploration in the field of extremophilic bioprocessing and bioremediation [28][29][30][31][32][33][34], we have identified the scope of thermophilic microbes and thermophilic bioprocessing towards developing sustainable and environment friendly methods of ANP waste degradation.

The primary objectives of this review effort are to (i) outline the parameters that affect the circulation of ANP drugs in an environment; (ii) provide a brief overview of the source, types, and concentration of ANPs in the environment; (iv) compare and contrast the Environmental Risk Assessment (ERA) strategies being implemented in different regions; (iii) compile the information on existing biotic and abiotic methods of remediation of ANP drugs; (iv) discuss the possibility of designing a more energy efficient remediation method through integration of biotic and abiotic methods of treatment.

2. Sources of ANPs in the Environment

Since the very first detection of ANP drugs in the aquatic environment in 1985 [35], various studies have discovered different classes of ANP drugs in the aquatic environment, with negligible or almost unchanged structural conformation [36]. ANP agents are mostly introduced into the environment through the urine and feces of outpatients who consume the drug at home (oral chemotherapy), or patients who undergo chemotherapy at hospitals. Particularly effluents from cancer hospitals or hospital wards specialized in oncology are the key emission source of ANP drugs in the aquatic environment [37][38]. In fact, excretions of patients undergoing chemotherapy at hospitals is the main source of introduction of ANP

drugs in the environment . The excretion route (feces or urine) is dependent on the type of drug administered. For example, 70% of bleomycin (in less than 2 h) and 40% of doxorubicin (in 5 days) is excreted in urine, and 50% of irinotecan (ITN) (48 h) in feces . A few more ANP drugs such as MTX and pemetrexed (PEM) have shown high urinary excretion rates (≈ 90%). MTX and PEM are mainly excreted as human metabolites of the parent drug [39]. For example, the urinary excretion of 5-FU is only about 15% as parent compound (5-FU), and 80% as its metabolite R-fluoro-alanine [40]. The excretion rate of ANP drugs varies with medication (duration of medication, formulation), mode of application (intradermal, intravenous, oral), and metabolic rates among treated patients. Figure 2 shows the possible routes for the introduction of pharmaceutical drugs into the environment. The occurrence and concentration of ANPs in effluents is also dependent on many factors, such as the number of patients, the physico-chemical nature of the drug used, dosing, excretion rates, methods of sampling, storage, and transport, as well as daily water consumption, which can significantly dilute the effluent and affect the detectability of ANP and its TPs . ANPs through effluents can enter the aquatic environment unaltered or as TPs depending on the physicochemical characteristics discussed in Section 2. These drugs or their TPs can have detrimental effects on the environment, aquatic life, and human health . Due to danger that these drugs pose, it is important to gather data on the usage of ANPs in a decentralized way because every patient and hospital do not use the same type ANP drugs. Such decentralized categorization of data would also help in designing efficient environmental risk assessment and remediation strategies.

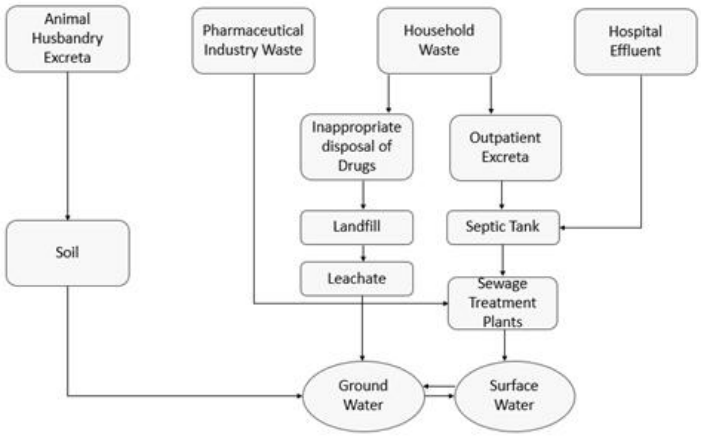

Figure 2. Sources for the introduction of ANP waste into the environment (modified from [41][42]).

3. Environmental Risk Assessment (ERA) of ANPs

Recent years have seen a sharp spike in the number of ANP drugs in the market which has promoted anticancer home treatments . As a result, more ANPs are being increasingly reported in wastewater and natural water bodies [43]. Due to their increased accumulation and highly hazardous nature, it has become imperative to quantify the concentration of ANPs in the environment and carry out an environmental risk assessment (ERA). The ERA methodology varies in different regions of the world. The European medical agency (EMA) calculates the predicted environmental concentration (PEC) in addition to screening the persistence, bioaccumulation, and toxicity of the drug during phase I assessment [44][45]. The PEC value is calculated using the following Equation (1) [45].

$$PEC_{SW} = (DOSE_{AS} \times F_{PEN})/(WASTEW_{INHAB} \times DILUTION) \quad (1)$$

where, PEC_{SW} predicted environmental concentration for surface water (mg/L); $DOSE_{AS}$ is the maximum daily dose of the active substance consumed per inhabitant (mg/inh·d); F_{PEN} is the fraction of a population receiving the active substance; $WASTEW_{INHAB}$ amount of wastewater per inhabitant per day (L/inh·d); DILUTION is the dilution factor.

As per the EMA standard guidelines, if the PEC_{SW} value is < 10 ng/L, the pharmaceutical drug is unlikely to represent a risk to the environment [45]. Persistence, bioaccumulation, and other toxicity tests are not required for ANPs that fall under this category. Lipophilic ANPs (log K_{ow} > 4.5) are directly moved to phase II because of their bioaccumulative nature. In contrast, the USA uses different ERA methodology where the drugs are first sorted based on its potency to enter the environment. Various tests such as the water solubility test, dissociation constant test, octanol/water partition coefficient (K_{ow}) test, and vapor pressure is conducted on the sorted drugs. The environment introduction concentration (EIC) is then estimated using the following Equation (2) [45].

$$EIC_{AQ} = A \times B \times C \times D \quad (2)$$

where, EICAQ: expected introduction concentration of an active moiety into the aquatic environment (ppb); A: amount produced for direct use as active moiety (kg/year); B: inverse of liters per day entering the publicly owned treatment works, (POTWs) (day/L); C: conversion factor (year/365 day); D: conversion factor (109 µg/kg). If the EIC_{AQ} < 1000 ng/L, the drug is excluded from further testing. PEC_{SW} is generally lower than EIC_{AQ} due to dilution factor.

When compared to EU and USA, Canada's ERA assesses not only the harmful effects of the substance on human health and environment but also their exposure potential [45]. Canada's ERA considers the lethality, mutagenicity, reproductive effects, and organ toxicity of the drug. This detailed testing weighs the impact of ANPs and is not carried out in the EU or USA. In Canada, PEC is calculated using the following Equation (3) [45].

$$PEC_{SW} = (\text{kg of API/year}) \times (1 \text{ year}/365 \text{ days}) / (\text{Wastewater L/day·capita}) \times (\text{population}) \quad (3)$$

where, PEC_{SW}: predicted environmental concentration in surface water (µg/L); kg of API/year: amount produced per year (kg/year); Wastewater L/day·capita: volume of wastewater generated per day per capita (L/day). The substance tested is designated to be toxic when PECsw divided by predicted no-effect concentration (PNEC) is ≥1.

One major obstacle in assessing the risk of ANPs in the environment is that the existing ERA procedures differ significantly in the European Union (EU), USA, and Canada. The most notable distinction is that the EU and USA regulate products (drug), whereas Canada regulates substances (active pharmaceutical ingredients (APIs))[45]. In EU and USA, existing drugs or new drug applications before the introduction of the ERA guidelines are not required for assessment whereas in Canada all substances entering or which may enter the environment are premised to be toxic until evaluation [45]. The EU and USA have implemented a tiered approach in which only the drugs that are suspected to be toxic are transferred onto the next stage of assessment. Canada, on the contrary, has adopted a classification-based approach in which the timing of notification is determined by the type of substances, i.e., polymer, chemical, or biological living systems [45]. However, in Canada, toxic and even non-toxic substances are checked for their accumulation impact to the land and water. The criteria of toxicity testing even after ERA clearance is vital as accumulation or circumstances of exposure to the substance may render the substance toxic. Table 1 gives a comparison of ERA procedures implemented in the USA, EU, and Canada.

Table 1. Characteristics of Environmental Risk Assessment (ERA) regulatory approach implemented in USA, EU, and Canada.

	USA	**EU**	**Canada**
Implementing organization	Food and Drug Administration (FDA)	European Medical Agency (EMA)	Health Canada and Environment and Climate Change Canada
Regulated product	All drugs manufactured for sale in EU member states	New Drugs	New Substances
Timing of ERA	When applying for marketing approval	New Drug applications [56]	Before notification
ERA Methodology	Phase-tiered based approach (Phase I; Phase II-Tier A and Tier B)	Tiered based approach (Tier 1, Tier 2, Tier 3)	Classification based approach (polymers, living organisms, chemicals)
Drug exclusion criteria	PEC [a] < 10 ng/L	EIC [b] < 1000 ng/L	PEC < 100 ng/L
Risk Assessment Criteria	PEC/PNEC [c] ≥ 1	EC [d]$_{50}$/MEEC [e] < 10	PEC/PNEC ≥ 1

Strengths	Responsibility lies with government Tiered approach	Responsibility lies with government Tiered approach	Responsibility lies with government Analyzes substances that are presumed to be non-toxic.
Weaknesses	No ERA for existing drugs. Non-consistent ERA procedure between member states	No ERA for existing drugs	Non-tiered approach

Notes: [a]—Predicted environmental concentration (for details see Section 4 of the review); [b]—Environment Introduction Concentration (for details see Section 4 of the review); [c]—Predicted no effect concentration (for details see [45]); [d]—Concentration of a drug that gives half maximal response; [e]—Maximum expected environmental concentration.

Some ERA studies elucidated that ANP drugs that directly interact with DNA do not have any safe threshold concentrations [45], and as such the stipulated PEC and EIC values may give false positives for certain ANP drugs. For instance, Kidd et al. [46][47][48] showed that concentration of breast cancer drug 17α-ethynylestradiol (in the concentration range 5–6 ng/L) impacted reproductive health in the fish (*Pimephales promelas*), that lead to a decline in its population at a concentration lower than the EMA suggested toxic level (10 ng/L).

One major issue with accurate ERA is the persistence and accumulation of some ANP drugs in the environment over time. The recalcitrant nature of some ANP drugs in activated sludge environment indicates the possibility of these compounds being persistent in river water [49]. The analysis of data provided by the German Environmental Agency confirmed that nearly 30% of ANP compounds are persistent in the water phase . This persistence may gradually increase the actual environmental concentration. Rowney et al. [47] showed that alkylating ANP drugs (0–145 ng/L), antimetabolite ANPs (0–27.4 ng/L), and anthracycline ANPs (0–0.7 ng/L) were detected in the Thames catchment in the United Kingdom. Similarly, the data available for the measured environmental concentration of cyclophosphamide in surface water varies from a negligible 0.05 to 64.8 ng/L .

Since there are a large number of ANP drugs currently in the market, and several others in the pipeline (Figure 1), it is necessary to categorize ANPs that are widely prescribed and are likely to persist in the environment. In addition to persistence, the current ERA methodologies do not have any guidelines on dealing with the transformation products (TPs) or human metabolites (HMs) of these drugs. It is not possible to design strategies to study the toxicity of TPs beforehand as the generation of TPs depends on the type of remediation methods employed and environmental parameters such as pH, temperature, type of remediation technique, etc. . Some studies have suggested that TPs or HMs, in some cases, may be more toxic than the parent drug [49]. Besse et al. reasoned that the metabolites of ANP drug methotrexate could be more toxic than their parent drug. Similarly, the TPs of certain medications can be more active, even more polar, and therefore of higher mobility in aquatic environments than the parent drug [50]. Consequently, it seems plausible to redesign the current ERA methodologies keeping in mind the toxicity of HMs and TPs. Genotoxicity assessments of ANPs and their relevant TPs should be conducted to allow for a better development of biodegradation or a combination of both biotic and abiotic remediation techniques.

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