Risk Assessment of Cytostatics in the Aquatic Environment

In recent decades, a number of studies have reported the presence of pharmaceutical and personal care products in the environment. These compounds are usually present at only low concentrations in the environment, but can cause adverse effects in exposed organisms because their mechanisms of action are designed to be effective in small doses.

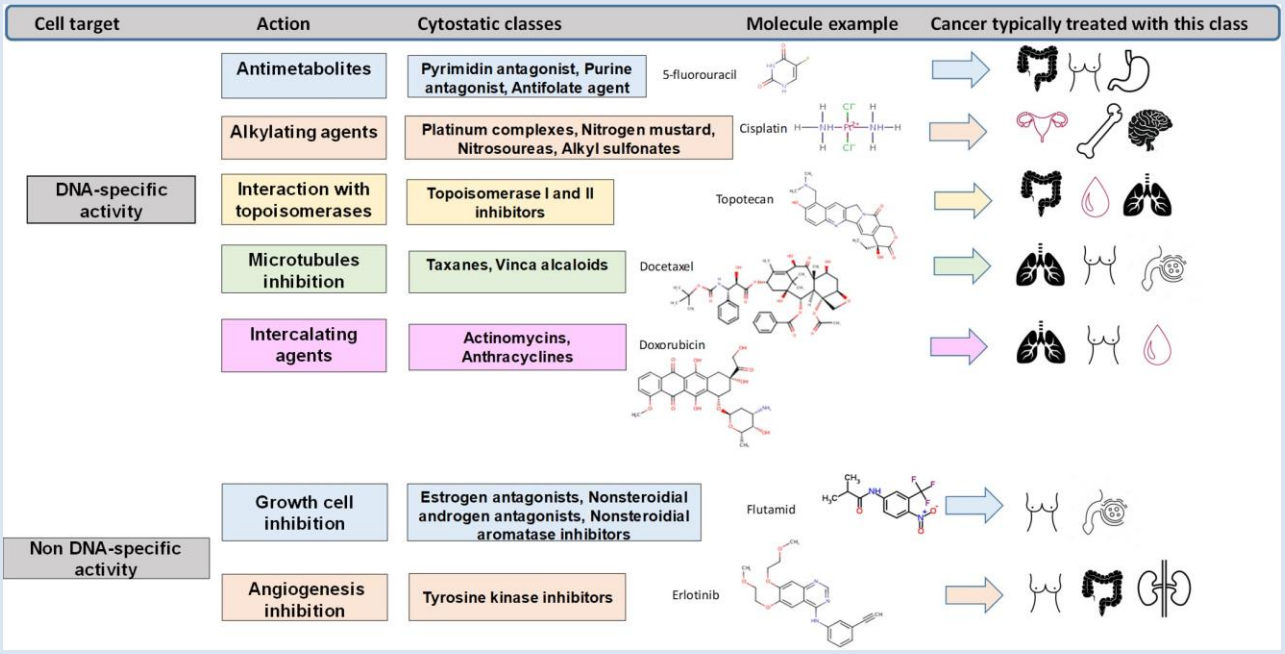
Cytostatics are pharmaceutical molecules, also known as antineoplastics or anticancer drugs, widely used in cancer treatment. Their use is increasing by around 10% per year in developed countries (Kümmerer et al., 2016). These substances are designed to kill rapidly growing cells such as those found in cancer tumours, although they are not specific to this type of cell. Consequently, all organisms may be susceptible to their toxicity. In addition, drugs used in breast and prostate cancers are considered as endocrine disruptors due to their specific hormonal or anti-hormonal properties. Drugs used for cancer treatment are thus suspected to represent a specific risk for aquatic non-target species.

This fact sheet provides an overview of the available information on occurrence, fate, bioaccumulation and toxicity of 48 cytostatic drugs approved by Health Canada for therapeutic use. Based on these data, the potential environmental risk associated with the presence of cytostatics in the aquatic environment is established, as well as the need for further studies to supplement the available data.

This fact sheet provides a summary of a literature review on risk assessment of cytostatic drugs in the aquatic environment (CEAEQ, 2019). The document can be consulted on the website of the Ministère de l'Environnement et de la Lutte contre les changements climatiques Quebec (MELCC), in French only: http://www.ceaeq.gouv.qc.ca/ecotoxicologie/revue-cytostatiques.pdf.

Classification of cytostatic drugs

In most cases, cytostatic drugs act on the deoxyribonucleic acid (DNA) or ribonucleic acid (RNA) in cancer cells, or in the synthesis of proteins involved in the formation of the cytoskeleton of these cells. Depending on their mode of action, molecular structure and source, these cytostatic compounds can be separated into various categories, shown in Figure 1**.**



- **Antimetabolite**: chemical that inhibits the use of a metabolite, which is another chemical that is part of normal metabolism.
- **Alkylating agent**: compounds that work by adding an alkyl group to the guanine base of the DNA molecule.
- **Intercalating agent**: substance that inserts itself into the DNA structure of a cell.
- **Topoisomerases**: enzymes that participate in the overwinding or underwinding of DNA.
- **Microtubule**: a tube-shaped organelle involved in maintaining the structure of the cell.
- **Angiogenesis**: physiological process through which new blood vessels form from pre-existing vessels.

Figure 1 – Cytostatic classification based on their main cell target

Environmental contamination pathways for cytostatic compounds

Following patient consumption, cytostatic agents are emitted through hospital and city wastewater. Although these drugs are predominantly administered in hospitals, approximately 75% of patients return home after their treatment (Ferrando-Climent et al., 2013). Patients under cytostatic treatment excreted them as a mixture of parent compounds and metabolites via urine or feces.

Therefore, excreted cytostatics can potentially pass through municipal wastewater treatment plants, eventually reaching surface waters and sediments (Figure 2).

Wastewater treatment plants around the world are generally designed to remove organic matter, suspended solids and nutrients such as phosphorous. Although these facilities can reduce the presence of numerous contaminants in wastewater, they are not designed to remove cytostatic compounds. This is also true in Quebec. Consequently, depending on the wastewater treatment method used, various amounts of cytostatic compounds may enter the aquatic environment through municipal effluents. Several studies have shown the poor elimination of cytostatics by conventional wastewater treatment plants (Zhang et al., 2017; Franquet-Griell et al., 2017) and the rate of removal varies from one cytostatic compound to another, depending on its physico-chemical properties. Some cytostatic compounds can be found in residual sludge, which is used as a fertilizer in agriculture, effectively introducing cytostatics to the soil , as shown in Figure 2. Lastly, during wastewater treatment plant overflow events, or in cases where a municipality does not have a wastewater treatment plant, effluent, potentially loaded with cytostatic compounds, is directly discharged into the receiving environments. Consequently, wastewater treatments plants are considered to be an significant point source of cytostatic contamination to the aquatic environment.

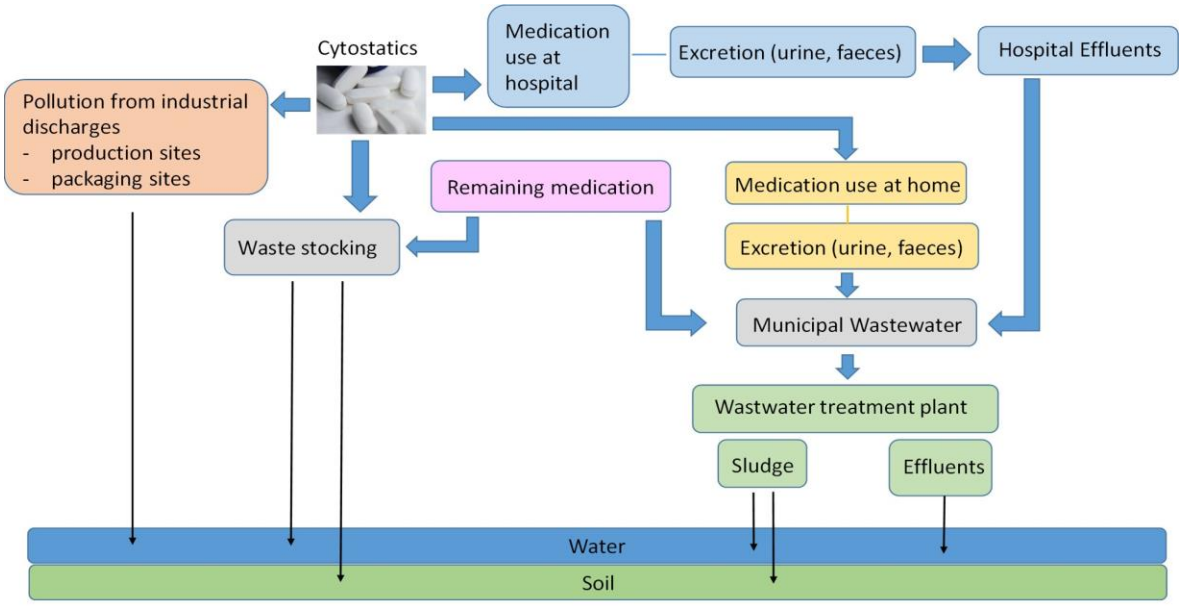


Figure 2 – Cytostatic pathways of environmental contamination (adapted from Besse, 2010)

Presence of cytostatics in the aquatic environment

Several studies have demonstrated the presence of cytostatic compounds in hospital effluents and municipal wastewater treatment plant influents and effluents, as well as in surface waters of their receiving environments. Concentrations ranging from 0 to 25,000 ng/L have been measured in various places around the world (Ffigure 3). Only a few measurements are available in Quebec, for 10 of the 48 cytostatic compounds studied (Table 1). With some exceptions,

concentrations in Quebec are relatively low (0 to 924 ng/L) compared to those reported elsewhere in the world.

Table 1 – Concentrations of cytostatic compounds measured in hospital effluents, municipal wastewater treatment plant influents and effluents, and their receiving environments in Quebec

Matrix	Location	Concentration (ng/L)	Reference
Class: Anthracyclines			
Epirubicin			
Municipal influents	Montreal	≤ 18	Rabii et al., 2014
Municipal effluents		≤ 18	
Class: Antifolates (Folic Acid Antagonists)			
Methotrexate			
Hospital effluents	11 Quebec hospitals	≤ 0.5 – 68.4	Vaudreuil et al., 2020
Municipal influents	6 Quebec municipalities	4.34 – 27.3	
Municipal effluents		≤ 0.5 – 25	
Municipal influents	Montreal	17 – 60	Rabii et al., 2014
Municipal effluents		≤ 12 – 53	
Municipal influents	Montreal	59	Garcia-Ac et al., 2009
Municipal effluents		≤ 16	
Surface water	St. Lawrence River, Montreal	≤ 6	
Class: Pyrimidine Antagonists			
Capecitabine			
Hospital effluents	11 Quebec hospitals	≤ 1 – 6.13	Vaudreuil et al., 2020
Municipal influents	6 Quebec municipalities	4.18 – 64.4	
Municipal effluents		8.62 – 52.2	
Cytarabine			
Hospital effluents	11 Quebec hospitals	≤ 5	Vaudreuil et al., 2020
Municipal influents	6 Quebec municipalities	74.4 – 924	
Municipal effluents		54.8 – 349	
5-Fluorouracil			
Hospital effluents	11 Quebec hospitals	≤ 2	Vaudreuil et al., 2020
Municipal influents	6 Quebec municipalities	≤ 2	
Municipal effluents		≤ 2	
Gemcitabine			
Hospital effluents	11 Quebec hospitals	≤ 5 – 31.4	Vaudreuil et al., 2020
Municipal influents	6 Quebec municipalities	≤ 5	
Municipal effluents		≤ 5	
Municipal influents	Montreal	≤ 20	Rabii et al., 2014
Municipal effluents		≤ 20	
Class: Topoisomerase I inhibitors			
Irinotecan			
Municipal influents	Montreal	≤ 19	Rabii et al., 2014
Municipal effluents		≤ 19	

Matrix	Location	Concentration (ng/L)	Reference
Class: Nitrogen Mustards			
Cyclophosphamide			
Hospital effluents	11 Quebec hospitals	≤ 0.5 – 2.2	Vaudreuil et al., 2020
Municipal influents	6 Quebec municipalities	≤ 0.5 – 118	
Municipal effluents		≤ 0.5 – 18.2	
Municipal influents	Montreal	≤ 4 – 22	Rabii et al., 2014
Municipal effluents		≤ 4 – 21	
Municipal influents	Montreal	9	Garcia-Ac et al., 2009
Municipal effluents		≤ 9	
Surface water	St. Lawrence River, Montreal	≤ 9	
Ifosfamide			
Hospital effluents	11 Quebec hospitals	≤ 1 – 144	Vaudreuil et al., 2020
Municipal influents	6 Quebec municipalities	≤ 1	
Municipal effluents		≤ 1	
Municipal influents	Montreal	≤ 4	Rabii et al., 2014
Municipal effluents		≤ 4	
Class: DDT Derivatives			
Mitotane			
Surface water	St. Lawrence River and its tributaries (Richelieu, Yamaska, Saint-François and Nicolet rivers)	≤ 0.01 to 0.232	Pham et al., 1996

Bioaccumulation of cytostatics in aquatic organisms

Most cytostatic compounds have a low bioconcentration factor (BCF) in aquatic organisms; BCF is the ratio between the compound's concentration in the organism and in the environment (ChemSpider, 2019). In theory, this implies that these substances do not tend to accumulate in organisms. Indeed, based on the *Persistence and Bioaccumulation Regulations*, who set the criteria used to determine if a substance is persistent or bioaccumulative under certain sections of the *Canadian Environmental Protection Act, 1999*, cytostatics are not considered persistent nor bioaccumulative (EC, 2015). Nevertheless, depending on their physico-chemical properties, a few cytostatic compounds can bioconcentrate in organisms, including antiestrogens (e.g., tamoxifen) and tyrosine kinase inhibitors (e.g., lapatinib), which have been demonstrated to have high BCF values (Jean et al., 2012; Orias et al., 2015; Booker et al., 2014).

Toxic potential of cytostatics in aquatic ecosystems

Experimental data on the toxicity of various cytostatic compounds to plants and animals are limited. In the small number of studies available on each of these compounds, the threshold effect levels are associated with a high level of uncertainty.

Nevertheless, a few studies have shown that some cytostatics could have adverse effects on aquatic organisms in the receiving environment at the low concentrations found in effluents and surface water.

According to the toxicity data obtained, the concentrations found in the environment are too low to cause mortality in aquatic organisms (Figure 3). However, at concentrations near those measured in the effluents from some wastewater treatment plants and in some aquatic environments, studies report that cytostatic compounds can interfere with or inhibit growth, damage DNA (genotoxicity) and potentially act as endocrine disruptors in exposed organisms. This is notably the case for pyrimidine antagonists, particularly 5-fluorouracil, which, at concentrations found in receiving environments, may affect the growth of certain algae and bacteria (Zaleska-Radziwill et al., 2014; Zounkova et al., 2007). Anthracyclines, mainly doxorubicin, can damage the DNA of aquatic invertebrates at the concentrations measured in the influents and effluents of municipal wastewater treatment plants (Parrella et al., 2015). This type of damage has also been observed following exposure to etoposide, a topoisomerase II inhibitor, at concentrations measured in wastewater treatment plant effluents (Parrella et al., 2015). Lastly, several studies have shown that non-steroidal antiandrogens (flutamide) and antiestrogens (tamoxifen) can affect the reproduction of aquatic invertebrates, fish and amphibians, as well as cause significant endocrine disruption, at concentrations measured in the environment (Rajakumar et al., 2012; Van der Ven et al., 2007; Williams et al., 2007).

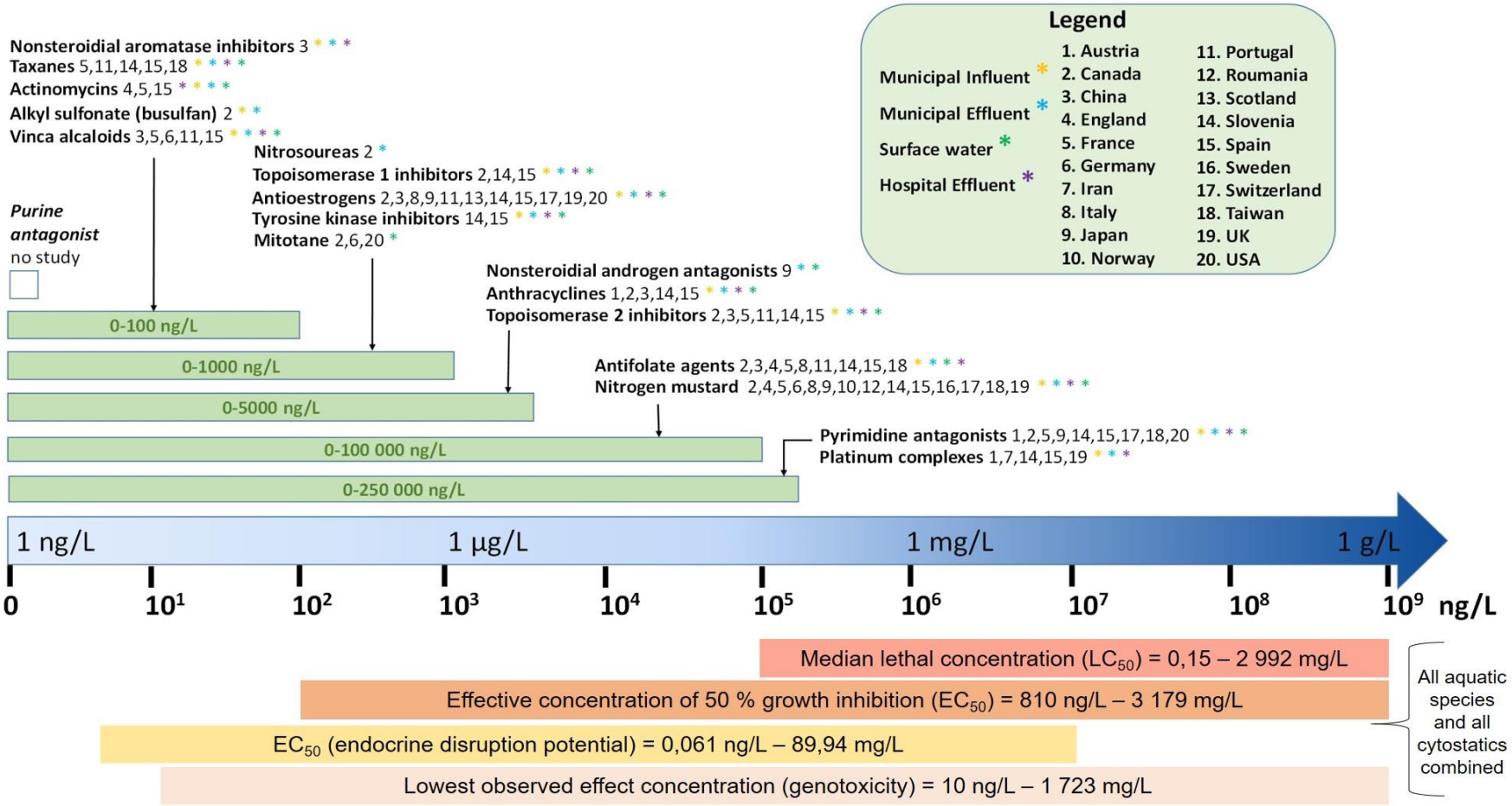


Figure 3 – Measured environmental concentrations of cytostatics classes in different aqueous sources around the world, as compared to toxic concentrations for aquatic organisms exposed to cytostatic in experimental studies. Toxicity is shown according to four different scales: lethal potential, growth toxicity potential, genotoxic potential and endocrine disrupting potential. Genotoxic potential includes all types of DNA damage. Endocrine disrupting potential includes all hormone imbalances in the exposed organism, as well as reproductive effects since hormones play a key role in reproductive function.

Outlook

The information presented in this fact sheet has led to the following findings:

- Several studies around the world have demonstrated the presence of cytostatic compounds in hospital effluents and municipal wastewater treatment plant influents and effluents, as well as in surface waters in their receiving environments. In general, wastewater treatment plants are not designed, or are inadequately designed, to remove these substances.

- Few studies indicate that some cytostatics have high bioconcentration factors, suggesting that they could accumulate along food chains.

- Several cytostatic compounds can produce acute, chronic or genotoxic effects or have endocrine disrupting potential at the concentrations measured in the environment around the world.

These findings have highlighted need for additional research to evaluate:

- The fate of cytostatic compounds in municipal wastewater treatment plants and in their receiving environments (water and sediments);

- The bioaccumulation of cytostatic compounds in aquatic organisms, given the paucity of available studies;

- The toxic potential to aquatic organisms of each class of cytostatic compounds, particularly their genotoxicity and its potential for endocrine disruption, which have remained relatively unexplored. Both laboratory and field studies are needed, to assess the status of aquatic communities exposed to these contaminants.

In response to these findings, several actions have been undertaken:

- Chemists at the Ministère de l'Environnement et de la Lutte contre les changements climatiques (MELCC) and Environment and Climate Change Canada (ECCC) are currently developing methods for analyzing cytostatic compounds (both pure and metabolized forms) commonly used in Quebec to treat cancer, in various environmental matrices.

- A project to determine the presence and effects of cytostatic compounds possibly present in municipal effluents discharged into the St. Lawrence, as well as the risks they pose to potentially exposed aquatic organisms, was included in the 2016–2021 programming for the St. Lawrence Action Plan (SLAP) under the research theme on urban wastewater. The project is a collaborative effort between MELCC and ECCC and has been renewed under the 2021–2026 SLAP programming. As part of this project, environmental concentrations of several cytostatic compounds will be measured, and studies will be carried out on the bioaccumulation and resulting effects on fish and aquatic invertebrates. Two studies are currently underway to determine the effects of several cytostatic compounds on Fathead Minnow (*Pimephales promelas)* embryos and on the Eastern Elliptio (*Elliptio complanata),* a freshwater mussel.

- MELCC is characterizing cytostatic compounds in the effluents of small wastewater treatment plants, with either aerated or non-aerated lagoons, that exclusively treat domestic wastewater.

All these projects will increase the amount of data available for evaluating the presence of cytostatic drugs in Quebec's aquatic ecosystems and the potential environmental risk associated with their presence for aquatic organisms.

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Prepared by

Nathalie Paquet and Gaëlle Triffault-Bouchet, Centre d'expertise en analyse environnementale du Québec, Ministère de l'Environnement et de la Lutte contre les changements climatiques

Editing

Anouka Bolduc, Ministère de l'Environnement et de la Lutte contre les changements climatiques

Mélanie Desrosiers, Centre d'expertise en analyse environnementale du Québec, Ministère de l'Environnement et de la Lutte contre les changements climatiques

François Gagné, Environment and Climate Change Canada

Christian Gagnon, Environment and Climate Change Canada

Cat. no. En154-139/2021E-PDF

ISBN: 978-0-660-41243-6

Published by authority of

Published by authority of the Ministère de l'Environnement et de la Lutte contre les changements climatiques

Également disponible en français sous le titre : *Évaluation du danger relié à la présence de cytostatiques dans le milieu aquatique*