Pharmaceutical Excipients as a Proxy for Measuring Pharmaceuticals in the Environment: A Case Study with Polyethylene Glycol

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ABSTRACT

BARKLEY, RACHEL AUSERMAN. Pharmaceutical Excipients as a Proxy for Measuring Pharmaceuticals in the Environment: A Case Study with Polyethylene Glycol (Under the direction of Dr. Tamara Pandolfo).

Pharmaceutical waste presents a problem where industry's production of pharmaceuticals and their waste are fast out-pacing the regulations around this waste. In the past, previous methods have framed the active pharmaceutical ingredients (APIs) as the most important ingredient which must be assessed to determine environmental risk. Here, a proxy is investigated as a means of quantifying the waste which communities may detect in wastewater. Pharmaceutical excipients are much more prevalent across manufacturing and therefore would provide the early datasets needed to assess how widespread these APIs could be. The framework uses polyethylene glycol (PEG) as a case study for how to implement this framework.

DEDICATION

Dedicated to Joanna Eden Carmel. Wish you were here.

BIOGRAPHY

Rachel Barkley has been working in the pharmaceutical and biotech industry since graduating from University of North Carolina at Chapel Hill in 2011 with a BS in Biology. Her first laboratory experience came in high school when she interned in Dr. Lola Reid's laboratory, working with Dr. Eliane Wauthier to culture fetal liver tissue for Dr. Wauthier's experiments. Her work experience has covered cellular biology, analytical chemistry, and regulatory compliance. Since 2019, she became involved in the Ecological Society of American (ESA) and the Society of Ecological Restoration (SER) to make connections with environmental scientists interested in pharmaceuticals in the environment. She has presented at meetings hosted by SER on the topic of ecopharmacovigilance, and she has organized sessions on pharmaceuticals in the environment for ESA's annual meeting over the past three years. She considers this project a culmination of her professional experience and career passions.

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Pharmaceuticals in the Environment

The current approach to tackling pharmaceuticals in the environment is based on the EPA's focus on the active ingredients (Kostich et al, 2014). However, addressing the concerns around pharmaceuticals in the environment requires a shift in focus to include not only the active ingredients but also the inactive ingredients also known as "excipients."

Within the industry, an excipient is defined as any intentionally included material with the dosage form which aids in the manufacturing of the drug product (Table 1; IPEC Federation, 2021). These ingredients do not interfere with the efficacy of the active pharmaceutical ingredient (API); in many formulations, they extend the shelf life of an API and enhance bioavailability (Wheatley, 2000). A critical part of understanding use of these compounds across the industry is that they are not always inert. Some excipients considered 'inactive' in one formulation are considered the API in another (Table 2). Some compounds have biological targets which can interfere with API efficacy in certain patients, manifesting as sensitivity or even causing an allergic reaction (Winek, 2000; Pottel et al, 2020).

21 CFR Part 211, enforced by the FDA, require rigorous documentation which demonstrates "safety, identity, strength, quality, and purity" of each ingredient within a drug product as well as assuring the safety of these ingredients when combined. Many excipients have been recognized for their utility throughout human history; the United States Pharmacopeia (USP), founded in 1820 as an independent and nonprofit organization for standardizing medicinal substances and their preparation, published standards for excipients still in use today such as lactic acid and natural gums (Winek, 2000).

Table 1: FDA and ICH Definitions of Industry Terms for Pharmaceutical Products

Term	Definition
Drug (US FDA, 2017)	• A substance recognized by an official pharmacopoeia or formulary. • A substance intended for use in the diagnosis, cure, mitigation, treatment, or prevention of disease. • A substance (other than food) intended to affect the structure or any function of the body. • A substance intended for use as a component of a medicine but not a device or a component, part, or accessory of a device. • Biological products are included within this definition and are generally covered by the same laws and regulations, but differences exist regarding their manufacturing processes (chemical process versus biological process.)
Drug Product (ICH, 2003)	The dosage form in the final immediate packaging intended for marketing
Dosage form (ICH, 2003)	A pharmaceutical product type (e.g., tablet, capsule, solution, cream) that contains a drug substance generally, but not necessarily, in association with excipients.
Active Pharmaceutical Ingredient (API), also called Drug Substance (ICH, 2016)	Any substance or mixture of substances intended to be used in the manufacture of a drug (medicinal) product and that, when used in the production of a drug, becomes an active ingredient of the drug product. Such substances are intended to furnish pharmacological activity or other direct effect in the diagnosis, cure, mitigation, treatment, or prevention of disease or to affect the structure and function of the body. See also USC 21 CFR 210.3(b)(7)
Excipient (ICH, 2003)	Anything other than the drug substance in the dosage form. See also USC 21 CFR 210.3(b)(8)

The FDA maintains the Inactive Ingredient Database which includes the inactive ingredient used in every FDA-approved product (FDA, 2022), effectively acting as a list of approved excipients. In addition, US Code of Federation Regulations 21 CFR Parts 182, 184, and 186 lists out substances the US government affirms are generally regarded as safe (GRAS) for food manufacturing; although meant for food, this list has also become a starting point in drug formulation because companies can ensure the safety of the ingredients. The FDA also maintains

several programs for approving new substances which can serve as ingredients in food, drugs, cosmetics, or medical devices.

Table 2: Purpose of Excipient in a Drug Formulation and Use as an API ((Fox, 2014)

Use in formulation	Example	Is this product also an API?
Fillers or diluent	Cellulose, Lactose, Starch	No
Binders	Natural gums, gelatin	No
Disintegrants	Sodium starch glycolate	No
Lubricants	Polyethylene glycol Stearic acid	Yes
Glidants	Colloidal silicon dioxide	No
Wetting agent	Sodium lauryl sulfate	Yes
Colors/pigment	FD&C dyes	No
Flavor/sweetener	Sucrose, mannitol	Yes
Ointment base	Petrolatum	Yes
Vehicle	Cottonseed oil	No
Emulsion agents	Mineral oil	No
Propellant	Chlorfluorocarbons (CFCs)	No

To comply with all regulatory requirements from various boards of health (BOH), a pharmaceutical company expects to spend 12-15 years of time and resources on one new drug product (Figure 1; Honaker & Clements, 2017). Companies may cut this time through clever management or resource distribution, but they cannot take shortcuts when it comes to quality, purity, efficacy, and safety. Repositories such as the Inactive Ingredient Database exist to provide companies both with starting points for formulation but more importantly signal to them that they are less likely to encounter delays in the various approval steps if they choose ingredients used in previously approved products. The FDA's guidance on the database states clearly: "once an inactive ingredient has appeared in an approved drug product for a particular route of administration, the inactive ingredient is not considered new and may require a less extensive review the next time it is included in a new drug product." (FDA, 2022)

Figure 1: The major phases of drug discovery and the average number of years for each phase. (Honaker and Clements, 2017)

Regulations for drug manufacturing have made medicine among the safest products anyone can buy, but these regulations only focus on impact to human health. Although post-market surveillance examines possible adverse effects of drugs on the individual consumers, companies are not responsible for collecting data on any adverse effects of their products on the environment. The FDA requires an environmental risk assessment (ERA) with new drug applications (FDA, 1998). However, these are product-based and do not account for the ecotoxicity of the individual ingredients or their degradants. By comparison, Japan and Australia's ingredient-based approach has its own limitations because the substance may enter the environment through various sources in addition to drug products (Josie et al, 2020).

The research into pharmaceuticals in the environment is still complicated because understanding the fate and transport of the APIs requires understanding the anthropogenic systems from which they emerge (Rieger et al, 2002). While research interest continuously grows in understanding the environmental impacts of ingredients in personal care products (Juliano & Magrini, 2017), it is also logical that new research includes APIs and excipients in pharmaceuticals since in many countries, the same government body will regulate both types of products.

To be successful, monitoring of pharmaceutical waste needs active buy-in from municipal partners such as local water and sewer authorities. Partnering with cities and towns to create water monitoring programs for emerging contaminants will not only inform municipal authorities about the quality of their drinking water but also help them take steps to lower the amount of those contaminants going into drinking water. The Environmental Monitoring for Public Access and Community Tracking (EMPACT) program was created in 1997 by the EPA as a means of providing environmental data to communities so they can make policies (EPA, 2001). EMPACT's Lead-Safe Yard Project provided a success story for how researchers, government bodies, and communities can partner together to turn scientific data into actionable policy. EMPACT's projects demonstrate the need for treating communities as equal partners and designing programs to fit the reality of those communities. In the case of pharmaceutical waste, the challenge would be for cities and states to set aside resources for monitoring potentially hundreds of APIs in the local environment.

However, cities and towns cannot feasibly run assays for every API which could appear in their water. Rather than focusing on APIs, municipal authorities can focus on the presence of certain excipients as a proxy for the relative amount of drug substances and drug products entering the environment. This project will identify how to select a proxy, the steps a community can take to implement a monitoring program, and potential policy goals that communities can pursue with the data they collect.

Choosing the Excipient

Modern processing techniques require the use of many inactive ingredients as additives in pharmaceuticals. If we want to use the presence of an excipient as a proxy for drug product presence in wastewater, our parameters should be as follows:

- *It cannot be an ingredient that the human body would treat as food.* An ingredient such as gluten, some sweeteners, lactose, or gelatin would be fully digested and may not be detectable once waste products are excreted from the body.
- *It must be synthetic.* An ingredient derived from an organism such as natural dyes, plant-derived oils, or amino acids could be present in food waste or occur due to natural environmental processes. These potentially also face the same issue as ingredients that the body would treat as food.
- *It must not be a food additive.* Any ingredient such as dyes, artificial flavor agents, plus many thickeners bulking agents, anticaking agents, antifoaming agents, emulsifiers, stabilizers, and preservatives are all present in highly processed food products. Their ubiquitous nature disqualifies them as a suitable proxy for pharmaceutical waste.
- *It must remain in the environment long enough to be measured.* Any ingredient, such as most volatile organics, that may degrade in route to the wastewater treatment plant would not be an appropriate proxy.
- *It must be well-documented in the literature.* While an ingredient which was made exclusively for use in pharmaceuticals is potentially a good proxy, the issue is that novel excipients approved by the FDA in the past twenty years are not well-documented in the literature and may not be ubiquitous in manufacturing.

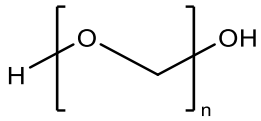
Based on these criteria, there are several groups of ingredients that could be used as proxies for pharmaceuticals, such as petroleum-derived chemicals. This project will focus on the petroleum-derived chemical polyethylene glycol (PEG), a polyether valued for its versatility

compared to its relatively safety to human and animal patients. This chemical was chosen for its conformance to these criteria:

- The human body does not treat PEG as food.
- PEG is synthetic.
- PEG is not a food additive.
- PEG takes, on average, six months to degrade in the environment.
- PEG is well-documented in the literature.

Polyethylene glycol (PEG) as Proxy

Polyethylene glycol (PEG) was first synthesized by Lourenco and Wurtz in 1859 (Leous, 1993). PEG is comprised of a repeating subunit is two carbons connected to an oxygen atom with each chain terminating in an alcohol unit. The general formula, where n equals the average numbers of subunits is



A scalable method for producing longer PEG chains was developed by Fordyce, Lovell and Hibbert (1939; Figure 2)

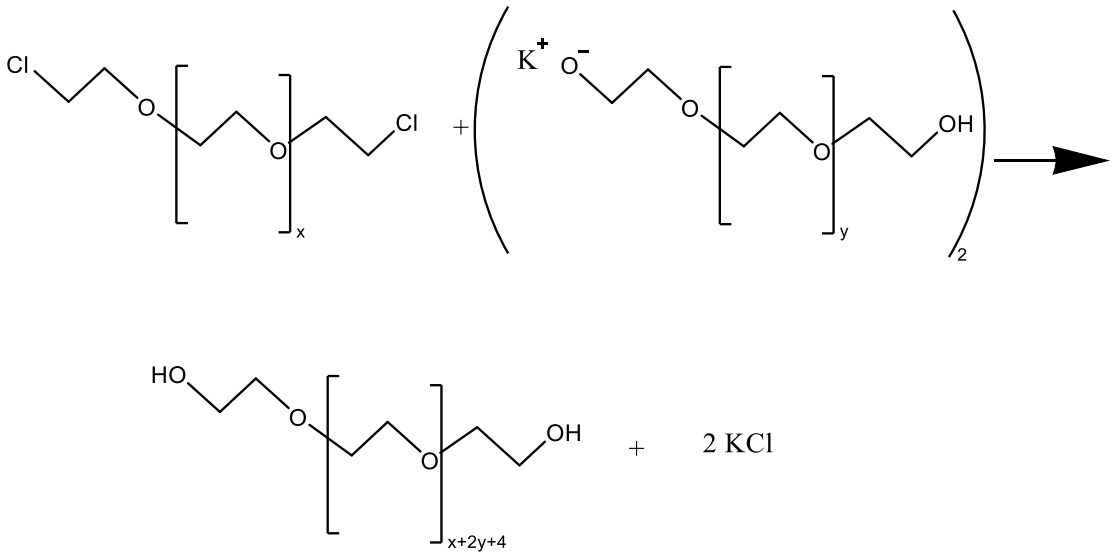


Figure 2: Synthesis of PEG as described by Fordyce, Lovell, and Hibbert (1939). Drawings created by author

By 1993, the synthesis method had become a stepwise reaction to create more uniform preparations (Leous, 1993). Starting with ethylene oxide and water, the two are reacted together to produce ethylene glycol. After creating ethylene glycol, ethylene oxide is added again to create two ethyl chains with terminating hydroxyl groups bonded to a single oxygen (Figure 3).

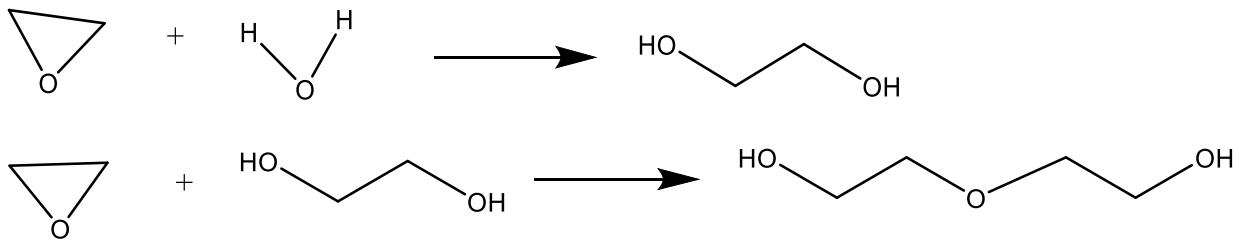


Figure 3: Synthesis of PEG described by Leous (1993). Drawing created by author

The hydroxyl group on the chain reacts with each added ethylene oxide, in a process called polymerization. One continues to add oxyethylene groups to the polymer until it reaches the desired molecular weight (Leous, 1993). To create various weights of PEG on a large scale,

industrial production starts with ethylene glycol as the raw material, then adds a caustic catalyst such as sodium hydroxide to react the raw material with ethylene oxide (Leous, 1993). Reaction rates can be speeded up through addition of pressure and heat. This reaction is so predictable and well-controlled that when creating large chains of PEG, the same reaction can be initiated with low-molecular weight PEG as the raw material. All PEGs remain stable for long-term storage, are non-volatile, and considered nontoxic (Pietrelli et al, 2021). Because of these characteristics, and because PEG's production is so well-controlled, the ingredient has become ubiquitous across many industries (Table 3).

Table 3: Common uses of PEG across industries

Industrial Categories	Specific Uses
Medical	Basis of laxatives for colonoscopy preparation
Pharmaceutical	Drug delivery system via PEGylation
Chemical	Ingredient in heat-resistant coats
	Ingredient in water-resistant paints
Consumer products	Ingredient in skin cremes, toothpastes, denture adhesives
Industrial	Binder in high-temperature ceramic molding
	Component of transistor manufacturing

Nomenclature for this group is based on molecular weight. For example: PEG 200 when synthesized has an approximate molecular weight of 200 g/mol (Table 4). According to the FDA's Inactive Ingredients database, it is rare for pharmaceutical manufacturers to use a PEG with a weight higher than 10,000 g/mol (Table 5). However, PEG 10,000 and above is used in other industrial processes. Therefore, analyzing detected PEG based on molecular weight can help separate pharmaceutical waste from other manufacturing waste. PEGs under 1000 g/mol appear as clear viscous liquids while PEGs 1500 g/mol and above appear as white powders or white flakes (ThermoFisher Scientific, 2021).

Table 4: Properties of some common PEGs. Sources: SDSs provided by ThermoFisher Scientific (2021) and Leous (1993).

PEG Product Number	**200**	**400**	**600**	**1500**	**4000**	**6000**
Average value of n	4	8-9	12-13	29-36	68-84	158-204
Molecular Weight Range (g/mol)	190-210	380-420	570-630	1300-1600	3000-3700	7000-9000
Viscosity (mPa-s at 20°C)	60-70	7.3	16-19	(solid)	(solid)	(solid)
Melting Range (°C)	-65	4-8	17-22	44-48	50-58	55-60
Flash point (°C)	171	176	252	>250	250	>250
pH	5.0-7.0	4.5-7.5	5.0-7.0	5.0-7.0	4.0-7.0	5.0-7.0
Density (g/cm^3)	1.125	1.128	1.127	1.21	1.21	1.21
Bioconcentration Factor	3.162 L/kg					
Partition Coefficient (log P_{ow})	-0.698					

A major property of PEG which has made it so desirable in pharmaceutical manufacturing is its solubility. Due to the terminating hydroxyl groups and ether groups, PEGs under 1000 g/mol dissolve readily into water, and PEGs over 1000 g/mol can dissolve readily when heated as part of dissolution (Leous, 1993). The same functional groups allow PEGs to also dissolve into polar organic solvents. However, PEG does not dissolve well into nonpolar organic solvents. Because of the reasons described above, PEG is also an effective solvent into which many polar compounds can dissolve including water and alcohols. In fact, PEGs over 1500 g/mol can dissolve water-insoluble compounds (Leous, 1993). PEG's solubility and solvency make it highly valuable as an excipient in drug product formulation. A survey of the FDA's Inactive Ingredients Database indicated that PEGs of different molecular weights are utilized in a variety of dosage forms and routes (Table 5).

Table 5: Survey of entries in the FDA's Inactive Ingredients Database for PEG

PEG form	Route	Dosage form(s)
PEG 200	Auricular	Drops
	Oral	Capsule; solution; extended-release tablet; tablet
	Topical	Ointment
PEG 300	Auricular	Drops
	Intramuscular	Injection; solution
	Intravenous	Injection; liquid; solution
	Ophthalmic	Ointment
	Topical	Cream; Lotion; Ointment; Solution
PEG 400	Intramuscular	Injection
	Intravenous	Injection
	Nasal	Metered spray
	Ophthalmic	Solution; Drops
	Oral	Capsule; soluble film; concentrate; suspension; syrup
	Rectal	Suppository
	Topical	Cream; emulsion; gel; liquid; lotion
PEG 600	Oral	Capsule; solution
	Topical	Ointment; solution
	Intravenous	Injection
PEG 1000	Oral	Solution; concentrate; film-coated tablet
	Rectal	Suppository
	Topical	Foam; cream; gel
	Transdermal	Gel
	Vaginal	Suppository
PEG 1450	Oral	Extended-release capsule; solution; suspension; tablet
	Topical	Ointment
	Urethral	Suppository
PEG 1600	Dental	Gel
	Oral	Tablet, coated tablet
	Rectal	Suppository
PEG 3350	Intra-articular	Injection
	Intramuscular	Injection
	Oral	Extended-release capsule; suspension; chewable tablet
	Soft tissue	Injection
	Rectal	Suppository
	Topical	Cream; ointment

Another important aspect of PEG's use is PEGylation, a process whereby PEG molecules are covalently attached to another molecule. By using covalent bonds instead of ionic bonds, PEG's addition to a given molecule change the molecule's behavior (Mastumura, 2003). In the case of certain vaccines such as the Pfizer-BioNTech COVID-19 vaccine, a lipid assisting in

liposome delivery was PEGylated as part of vaccine formulation to ensure efficient delivery of the messenger RNA (Fauci, 2021). PEGylation has been used in several pharmaceutical and biological products (Table 6). PEG does not normally elicit an immune response from a patient, and so PEGylation on liposomes within a biological product is used to "mask" the payload from the immune system until it reaches the target organ (Ogris, et al., 2003).

Table 6: PEGylation Examples, selected from "Polyethylene Glycol as an Embedment for Microscopy and Histochemistry" (ed. Kuixiong Gao, 1993)

Uses of PEGylation
Interferon α-2a PEGylated with a branching polymer to improve efficacy for hepatitis C treatment
Luteinizing hormone PEGylated for experimenting
Doxorubicin micelles conjugated with a hydrophobic polymer PEGylated to improve cancer treatment
PEG-coated liposome containing doxorubicin as drug delivery for cancer treatment
Uricase PEGylated as part of gout treatment

In the 1970s, PEG's ubiquity across industrial-scale manufacturing made it of special importance to researchers wanting to understand the persistence of xenobiotic chemicals in the environment (Bernhard et al, 2008). The partition coefficient for PEG shows that it moves through soil compartments in the environment to migrate into water compartments, and it is believed to accumulate in water reservoirs such as oceanwater, streams, and groundwater. Current research suggests that PEG does not accumulate in the food chain (Kawai, 2015). However, PEGylation can change the water solubility of many different molecules, including hydrophobic molecules, meaning that the properties regarding the fate and transport of PEG within the environment can be transferred to those molecules.

Implementing a Monitoring Plan

Due to the widespread use of PEG in industrial settings and its relatively low toxicity, PEG is not considered a chemical of concern for wastewater monitoring (Corti et al, 1998). However, the literature does contain a long history of experiments for detecting PEG presence in the environment to determine its fate and transport (Pirvono et al, 2012). The main challenge with a PEG monitoring program is that one must account for a variety of PEG weights and determine the optimal range for detection. Structure of the monitoring program will be limited to existing and available methods which can efficiently analyze samples in a timely manner.

Materials and Preparation

The field team should use glass containers for initial sampling start with grab samples as part of proof-of-concept testing. All laboratory containers and glassware that will directly touch the samples should be washed according to the general preparation method provided in the Handbook of Water and Wastewater Treatment Plant Operations (2008): first wash with a phosphate-free detergent, wash thrice with tap water, wash thrice with distilled or deionized water.

Cong et al. (2015) notes that UV light has become a tool for PEGylating proteins to render them inert, and then using light again to cleave off the PEG and reactivate the protein (Georgianna et al, 2010). Thus, exposing samples to light could cause PEGylation of PEG with nearby proteins which would be abundant in wastewater. To minimize this, samples should be transferred to amber glass vials with rubber stoppers; PEG may remain stable, but its degradation may result in a high count of low-weight PEG pieces which could skew results.

Sample Collection

A wastewater treatment plant's current monitoring program does not require modification to its sampling protocols to capture samples for testing PEG presence. Literature surveyed for this project did not provide any specific recommendations on sample collection or preservation. As part of initial method development, test samples within 24 hours of collection. Analysts should determine expiry through additional testing after 24 hours to determine when PEG in the sample will undergo significant degradation; do not add preservatives. Although it is established that raw material PEG remains stable at room temperature (20-25°C) (ThermoFisher Scientific, 2021), related work with polyethers in wastewater suggest that cold preservation (4°C) is best practice (Thurman, et al., 2017). Amber glass vials are designed to block light, but it is still advisable to keep the samples in storage away from fluorescent light rather than on the benchtop. Prior to analysis, samples should be filtered through PTFE filters 0.2 micron (Thurman, et al., 2017). Once the method has been fully developed, composite samples can then provide an average and allow a wastewater treatment plant to decide on an appropriate monitoring range for PEG. If a treatment plant wants to establish where PEG fits into its suite of test programs for different sources, the plant should start with testing for PEG where the sources of possible pollutants come from septic systems and municipal sewage systems; testing should only focus on the point sources which would capture pharmaceutical waste.

Instrumentation and Analysis

While chromatographic methods exist, these instruments require trained operators and are expensive to acquire and maintain. To fit this proxy into the existing suite of tests performed as part of ongoing wastewater treatment and management, facilities can use existing spectrophotometers (Hoffman, 1983; Fejfer et al, 2021) which are often used for testing levels of

nitrates and chlorides. For any municipal plant with few resources there is a newly developed, low-cost, open-source miniature spectrophotometer which can provide quantitative PEG analysis with absorbance in the 450nm to 750nm range (Laganovska, et al., 2020).

Calibration and Standards

PEG itself does not absorb electromagnetic radiation at the UV-Vis wavelengths, but one can add reagents to samples to react with the PEG and create byproducts that can be detected (Jia & Tian, 2009). Developed by pharmacy professor Johann Dragendorff in 1866, Dragendorff's reagent (DR) is a solution of potassium bismuth iodide which allowed for the detection of alkaloids using early chromatographic methods (Raal, et al., 2020). The reagent can be obtained as a commercially available solution or prepared in a laboratory by reacting bismuth oxynitrate and potassium iodide in a diluted acid (Raal, et al., 2020). The color complex resulting from the reagent and PEG is detectable by UV-Vis spectrophotometry; the ion pair created in the reaction has a color ranging brown to red to orange to yellow. Notably, some alkaloids such as caffeine do not react with DR (Raal, et al., 2020). In the case of detecting pharmaceutical waste, DR not reacting with certain alkaloids helps support PEG as a proxy because this detection method is less likely to incorrectly detect other ingredients and APIs.

A modified DR method, using a sodium acetate buffer, has been utilized in the development of a calibration curve to measure various weights of PEG via spectrophotometer (Jia and Tien, 2009; Fejfer et al., 2021). The calibration curve encompassed PEG weights of 100, 400, 800, 1200, 4000, 6000, 10K, and 20K with standards created via a consecutive solution method. (Fejfer et al., 2021).

This calibration curve is ideal because according to the FDA's Inactive Ingredients database, the highest molecular weight for PEG found in pharmaceuticals is 8000 MW (FDA,

2022). This creates a relatively narrow group of PEG molecules to look for and is more likely to exclude the forms of PEG used outside pharmaceuticals and personal care products.

When developing a reliable method for quantifying PEG in environmental samples, the UV-Vis spectrophotometer method should be validated by comparison against other methods. Solid phase extraction (Guermant et al, 1995; Szymanski et al, 2001) size-exclusion chromatography (Mortensen et al, 2007), or liquid chromatography with mass spectroscopy (Thurman, et al., 2017) are all excellent ways to verify accuracy and precision of the PEG-Dragendorff method.

Discussion and Implications

PEG in Pharmaceuticals

While not a component of every possible drug product, PEG has been employed across such a wide variety of drug dosage forms and products that it can serve as a sufficient proxy when attempting to quantify pharmaceutical waste in municipal water systems. A review of entries in the DailyMed database maintained by the National Library of Medicine (2022) shows PEG as one of the ingredients in many extended-release tablet forms of drugs that were submitted to the FDA for approval in the past three years. Beyond tablet manufacturing, PEG has become part of the toolbox for researchers trying to perfect biologics, including vaccines, biomaterials used for aiding tissue regeneration (Lauto, Mawad, & Foster, 2008), and delivery for gene therapy (Ogris, et al., 2003).

Novartis's meningococcal vaccine (European Union Patent No. EP2462949A2, 2008) used PEG as a temperature protective agent for the vaccine formulation; the Program for Appropriate Technology in Health (PATH) filed patent in 2006 (United States Patent No.

US20060228369A1) for a generalized method using PEG to help stabilize and preserve vaccines sensitive to temperature changes. More recently, the COVID-19 vaccine was developed in a very short timeframe by leveraging similar technology (Fauci, 2021); components had already been developed and patented years before as part of work on a potential SARS vaccine (Aranda, et al., 2020). Thus, monitoring PEG in wastewater can serve as a proxy for the fate of unused vaccines which have been disposed.

PEG has also played an important role in cancer treatments. PEG-coated liposomes were developed to deliver the cancer drug doxorubicin to tumors in a more targeted manner compared to previous methods of flooding the body with drug (Mastumura, 2003). The liposomes stay circulating in the body for longer, meaning that patients need a lower dose than with other drugs which will lower impact of side effects and improve the patient's overall quality of life. PEGylated liposomes are part of the common suite of drug delivery systems available for new cancer drugs, and thus this technology will keep playing a role in new treatments. As with vaccines, monitoring PEG can serve to monitor if and how cancer drugs are entering the environment. Although the proxy cannot make a distinction between the different drugs, using PEG as a proxy will capture nearly every class of drug, including cancer drugs.

An oft-overlooked part of contaminants of concerns is nanoparticles which are already being developed and implemented in pharmaceuticals without much information on their environmental impact. Work by Garreta and Yoncheva (United States Patent No. US20080248125A1, 2005) shows the use of PEG-linked nanoparticles can assist in drug administration. Together, all these technologies demonstrate that PEG monitoring is robust proxy with staying power because it will capture waste generated by future drugs even as processes improve.

The use of PEG in the formulation of different antibiotics presents the strongest reasoning for why towns and cities should find a way to monitor the pharmaceuticals entering the bodies of water to which they are stewards. Antibiotics present an increasing danger to both human and environmental health. An important paradigm emerging across fields is One Health, an approach developed around the idea that public health must look beyond the human population to the stressors and the relationships they have with the other organisms on Earth (WHO, 2022). A concern among One Health researchers has been flow of antibiotics into the environment.

Industrial scaling of animal husbandry has led prophylactic use of antibiotics on livestock to minimize potential loss of production through animal illness and death. Wepking et al (2017) demonstrates that the manure from dairy cows contains the antibiotics they are given for managing disease and more importantly, the antibiotics present in the manure lead to great antibiotic resistance in soil microbial communities. The overabundance is also present in aquaculture (Cabello, 2006); like industrial meat and dairy productions, industrial aquaculture is relying on numerous antibiotics to keep stocked fish healthy enough to harvest because aquaculture puts higher stressors on the fish than their natural environment, making them all the more susceptible to infection and death. Cabello notes the growing connection between aquaculture and antibiotic resistance in human pathogens due to plasmid-sharing between the microbes from the aquaculture operation and microbes in the surrounding ecosystems. Through monitoring PEG levels, towns and cities can use that information as preliminary diagnostic indicators for determining when they should put resources toward testing for specific drugs such as antibiotics.

PEG Fate and Transport

The main drawback to PEG as a proxy is that it can be degraded by bacteria (Obradors & Aguilar, 1991; Kawai, 2015). It is not feasible to insist that there should be no PEG-degrading bacteria present in activated sludge; research suggests that the ability to degrade PEG may evolve spontaneously under the right selection pressures (Kawai, 2015). Instead, towns and cities would need the treatment plants to monitor the sludge to ensure that PEG degradation isn't interfering with monitoring. They would also have to exclude PEG degradation during treatment from how they measure PEG and instead only measure it when wastewater enters the plant.

PEG's prevalence in non-oral routes of drug dosing becomes important when looking at the larger picture of forecasting trends in pharmaceutical manufacturing. One market research firm, TechSci Research, estimates the global topicals market at $101.1 billion USD in 2021, $107.7 billion USD in 2022, and projects steady growth over the next five years at 6.8% (Research and Markets, 2022). Research and Markets, a publisher of market research reports, estimates the global transdermal drug delivery systems market at $6.0 billion USD in 2021 and project growth over the next six years at 6.4% (Research and Markets, 2022). The driver behind trends toward non-oral dosage routes comes down to demographics. The rise in global population is not being driven by any significant increase in live births but by lower mortality rates. The World Health Organization (WHO) predicts the number of people living past 60 years will grow 38% over the next ten years; the United Nations (UN) has declared 2021-2030 the Decade of Healthy Ageing to promote member nations to provide for an aging population (United Nations General Assembly, 2020). To maintain the same quality of life, infirmed and elderly patients will need medicines they can take which are easier for use, either by their caretakers or themselves. Oral dosages such as pills, lozenges, and drinkable solutions are

unusable if the patient cannot sit up or swallow safely (Logrippo, et al., 2017). Thus, excipients with properties like PEG are attractive to manufacturers when formulating the non-oral dosage of a drug product. The fallout of the COVID-19 pandemic and effects of environmental pollution have also left millions of people around the world with chronic health conditions (Schlatter, 1994; Sykes et al, 2021). Demand for these drugs will increase, and thus PEG will become more prevalent in manufacturing.

PEG monitoring and analysis be treated similarly to the use of total dissolved solids (TDS) in wastewater or particulate matter in air quality studies; these are proxies for water and air quality that ensure standards are met. The current schedule of parameters that determine quality of effluent from wastewater treatment plants (Table 7; Arundel, 2020) ensure water is safe for human consumption but cannot capture pharmaceutical waste because they were never designated for that purpose. The EPA has published various resources about pharmaceuticals in the environment, but the EPA has not yet moved to implement any regulations around acceptable levels of pharmaceuticals. In 2019, the EPA prohibited healthcare facilities from "sewering" pharmaceuticals waste—i.e., disposing of pharmaceuticals by flushing them or pouring them down sinks (EPA, 2022); while helpful, the burden of this prohibition falls strictly on healthcare facilities and doesn't require any infrastructure around them.

There are still gaps that must be addressed to implement PEG as a standard proxy in water quality monitoring for pharmaceutical waste. While the EPA is studying the issue of acceptable levels of any pharmaceutical, the EPA's work is still focusing on APIs and risk estimation of studied APIs rather than monitoring pharmaceutical proxies (Kostich et al, 2014). The goal of this work is for EPA to provide guidance to communities on the proxies they can use for monitoring pharmaceuticals in their local water systems; as demonstrated here, PEG is a

robust choice for monitoring by proxy. Consistent pre-treatment testing can help communities make decisions on waste management services and the infrastructure for these services. Compliance with future regulations will improve dramatically when cities and towns create their own monitoring programs and make policy decisions to provide the necessary services to organizations governed by such regulations.

Table 7: Schedule of Monitoring Programs Recommended for Wastewater Treatment Plants

Test Parameter	Purpose	Proxy?
Biological Oxygen Demand (BOD)	Measures the oxygen demand of organic matter and the live organisms on the sample.	Yes, for determining microbial activity
Chemical Oxygen Demand (COD)	Measures the oxygen demand of chemical reactions.	Yes, for quantifying organic molecules.
Total Suspended Solids (TSS)	Measures the dry weight of suspended solids which are not dissolved into water	No, means of determining if settlement tanks are working to get sediment out of wastewater
Total Dissolved Solids (TDS)	Measures the total of all inorganic and organic matter in the water.	Yes, for measuring metals in the water
Ammonia	Measures ammonia	No, direct measurements as ammonium is toxic to fish
Nitrate	Measures nitrates	No, direct measurement as high nitrate can cause eutrophication and toxic algal blooms.
pH	Measures hydronium ion	No, direct measurement as an indicator of overall water quality.

Implementing this framework within a community's existing monitoring program for wastewater is the first step toward tackling the problem of pharmaceutical waste. Starting with PEG as the proxy, local wastewater treatment plants can use their existing resources to develop reliable methods to capture current PEG levels. As the monitoring program continues, these facilities can partner with research institutions in the community such as universities and colleges to evaluate the data. With a reliable method developed, communities can start citizen science initiatives to quickly build a large dataset. With a sufficient dataset to analyze, communities can make decisions about how they are providing waste management services and

what they should change, such as providing more support for the businesses impacted by the EPA's regulations. These data can also drive interest for investing more infrastructure as communities decide what their priorities are. Finally, many communities collecting data at the same time can work together to advocate for support from state and federal agencies so that regulations can come into place to ensure industries can be held responsible for potential pollution they discharge with pharmaceutical waste.

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