



# **Biennial Review of 40 CFR Part 503 As Required Under the Clean Water Act Section 405(d)(2)(C)**

**Biosolids Biennial Review  
Reporting Period 2016–2017**

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405(d)(2)(C)**

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U.S. Environmental Protection Agency  
Office of Water  
Office of Science and Technology  
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## Definition of Biosolids

For the purposes of this Biennial Report, “biosolids” is used to mean sewage sludge, as defined in 40 CFR 503:

**Sewage sludge** is solid, semi-solid, or liquid residue generated during the treatment of *domestic sewage* in a *treatment works*. Sewage sludge includes, but is not limited to, domestic septage; scum or solids removed in primary, secondary, or advanced wastewater treatment processes; and a material derived from sewage sludge. Sewage sludge does not include ash generated during the firing of sewage sludge in a sewage sludge incinerator or grit and screenings generated during preliminary treatment of domestic sewage in a treatment works. 40 CFR §503.9(w).

Terms in italics are defined as follows:

**Domestic sewage** is waste and wastewater from humans or household operations that is discharged to or otherwise enters a treatment works. 40 CFR §503.9(g).

**Treatment works** is either a federally owned, publicly owned, or privately owned device or system used to treat (including recycle and reclaim) either domestic sewage or a combination of domestic sewage and industrial waste of a liquid nature. 40 CFR §503.9(aa).

**Industrial wastewater** is wastewater generated in a commercial or industrial process. 40 CFR §503.9(n).

Taken together, these definitions mean that biosolids, or sewage sludge, for the purposes of 40 CFR Part 503, are the residues from treatment of domestic sewage, whether that domestic sewage is combined with industrial wastewater or not. It does not include sludge originating from treatment of industrial wastes in the absence of domestic sewage.

## Executive Summary

During the 2016-2017 biennial review process, the EPA collected and reviewed publicly available information on occurrence, fate and transport in the environment, and human health and ecological effects for pollutants that (1) have been identified in the Targeted National Sewage Sludge Survey (TNSSS; U.S. EPA, 2009), or in the open literature as having been found in biosolids; and (2) have not been previously regulated or evaluated (e.g., as potentially causing harm to humans or the environment) in biosolids.

The EPA identified 32 new articles as providing relevant data for chemical pollutants that may occur in U.S. biosolids. After initial review, information was gleaned from 15 of the new articles. Review of the 15 articles identified 28 new chemicals in biosolids: seven polybrominated diphenyl ethers (PBDEs); nine parabens and metabolites; five brominated flame retardants (BFRs); three other flame retardants; two perfluoroalkyl substances (PFASs); and two triclosan transformation products. These articles also identified new data for 31 chemicals previously identified in biosolids. Concentration data in biosolids were found for all 28 new chemicals and for two chemicals identified in a previous biennial review (diclofenac and tonalide). Human health toxicity values were found for three of the new chemicals (benzoic acid; 2,4-dichlorophenol; and hexabromobenzene) and two previously identified chemicals, perfluorooctanoate (PFOA) and perfluorooctanesulfonate (PFOS). ECOTOXicology knowledgebase (ECOTOX; U.S. EPA, 2018d) records were found for 17 newly identified chemicals and 26 previously identified chemicals. Physical-chemical properties were identified for 22 new chemicals and 20 chemicals previously identified in biosolids; and bioconcentration or bioaccumulation factors were identified for 23 new chemicals (11 in terrestrial systems and 13 in aquatic; there was one chemical with both) and 24 previously identified chemicals.

In addition, six articles were identified as providing relevant data for microbial pollutants that may occur in biosolids. Review of these articles identified no new microbial pollutants in biosolids. Data were found for seven previously identified microbial pollutants: *Cryptosporidium* spp., *Giardia* spp., antibiotic resistance genes (ARGs)/antibiotic resistant bacteria (AR bacteria), *Salmonella*, *Escherichia coli*, human norovirus, and human adenovirus.

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## **1. Introduction**

Section 405(d) of the CWA requires the EPA to review biosolids regulations every two years to identify additional pollutants that occur in biosolids and to regulate those pollutants if sufficient scientific evidence shows that they may harm (i.e., there is risk to) human health or the environment. The EPA considers risk to be the chance of harmful effects to human health or to ecological systems resulting from exposure to an environmental stressor (e.g., chemical). Risk assessment is a scientific process consistent with EPA policies and depends on three factors: 1) how much of a chemical is present in an environmental medium (e.g., biosolids); 2) how much contact a person or ecological receptor has with the contaminated environmental medium; and 3) the inherent toxicity of the chemical.

The biennial review process is intended to fulfill the CWA requirement to identify additional pollutants that occur in biosolids every two years. The data gleaned from the biennial review process will be analyzed to determine whether it is sufficient to be used for assessing potential risk. While not listed as a biennial review, an extensive literature search was conducted and published in 2003, and 10 pollutants were identified and prioritized for risk assessment in response to the 2002 National Research Council of the National Academies report (68 FR 75531). Subsequent biennial reviews are posted to EPA's website for 2005, 2007, 2009, 2011, 2013 and 2015: <https://www.epa.gov/biosolids/biennial-reviews-sewage-sludge-standards>.

Once additional pollutants that occur in biosolids are identified, the EPA must assess the pollutants to determine whether they pose a risk to human health or the environment. The EPA is in the process of developing a tool that will enable users to screen pollutants found in biosolids for potential risk. The screening results will be used to make informed decisions about the need to perform more refined risk assessments, or to address data gaps or uncertainties. The EPA is also in the process of developing a probabilistic risk assessment modeling framework to conduct refined risk assessments on those pollutants that fail the screening process.

Addressing the uncertainty around potential risk for pollutants identified in biosolids is the top priority for the EPA's Biosolids Program. The EPA continues to make significant progress in building capacity to assess pollutants by developing the necessary tools and data needed. The EPA expects to begin risk screening once public review of the screening tool has been completed.

## **2. Approach for Biennial Reviews**

Every two years the EPA develops biennial reviews by collecting and reviewing publicly available information on the occurrence, human health and ecological effects, and fate and transport in the environment of pollutants that have been found in U.S. biosolids. The three categories of information collected and presented here are needed to conduct risk assessments.

- **Concentration Data.** Both the ability to detect a given pollutant in biosolids and the determination of the concentration at which that pollutant is present are highly dependent on the existence of analytical methods for that pollutant in the biosolids matrix.
- **Toxicity to Human and Ecological Receptors.** For human toxicity, this type of data includes values such as a reference dose (RfD), reference concentration (RfC), cancer slope factor (CSF), or inhalation unit risk (IUR). For ecological toxicity, it includes values such as lethal dose, lethal concentration, or chronic endpoints related to survival, growth, and reproduction.
- **Environmental Fate and Transport Data.** These data are necessary for assessing exposure and include various physical-chemical properties, as well as bioconcentration or bioaccumulation factors, which describe the tendency of a chemical to move from one medium (e.g., soil) to another (e.g., plant matter).

The biennial review approach consists of two stages, as illustrated in **Figure 1**, for chemical and microbial pollutants:

- **Paper Review.** In this stage, the EPA conducts a systematic review including a literature search and evaluation to identify papers that provide evidence of the occurrence of chemical or microbial pollutants in biosolids that have not previously been identified, or new data that fills data gaps for pollutants previously identified in biosolids.
- **Chemical and Microbial Review.** Using the list of chemicals identified in the Paper Review stage, the EPA extracts data (typically concentration, and environmental fate and transport data) from the identified papers. For newly identified chemical pollutants, the EPA then collects additional data on human and ecological toxicity and environmental fate and transport data, not limited to the biennial review period. These data are collected from a set of established sources (see **Section 2.2**). The EPA is currently revising the approach for microbial pollutants. As a result, there is no analogous data collection step (collection of data on human and ecological toxicity, and environmental fate and transport) for microbial pollutants for the 2016-2017 Biennial Review. The revised approach and results for the microbial review are expected to be reported in the 2018-2019 Biennial Review.

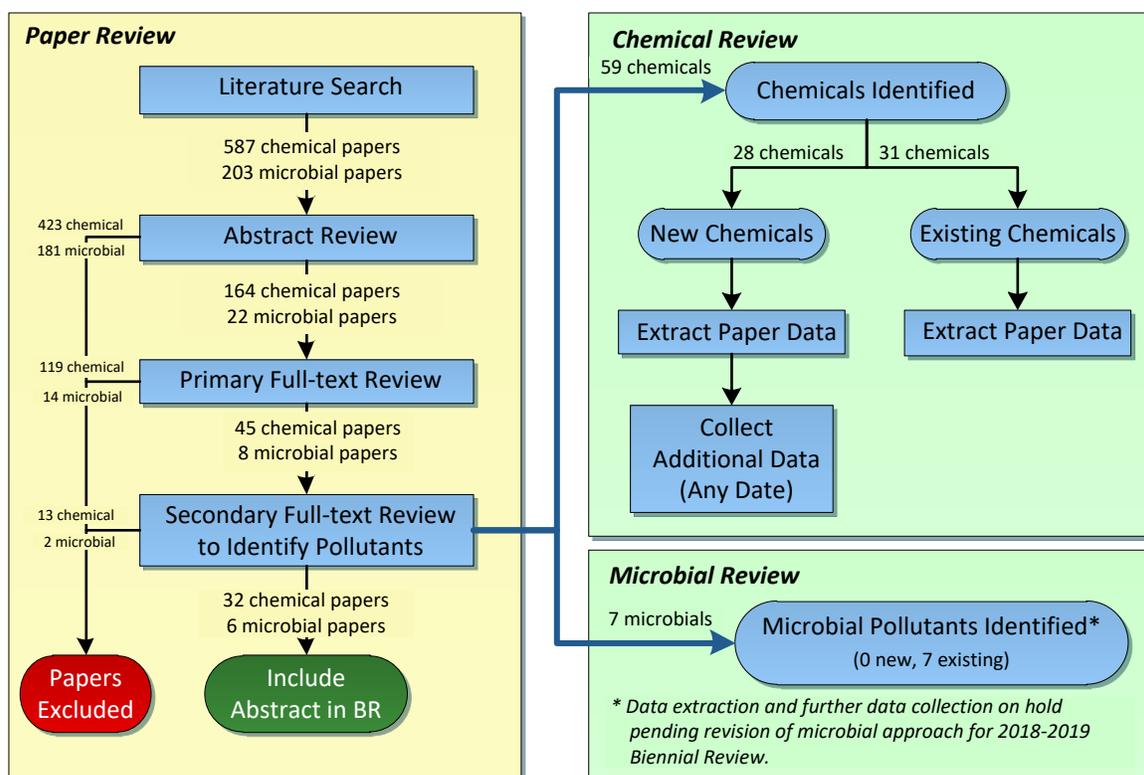


Figure 1. Overview of the Biennial Review Approach and 2016–2017 Summary for Chemical and Microbial Pollutants

## 2.1 Paper Review: Identify, Review and Select Papers

For reporting period 2016–2017, the EPA conducted a literature search from January 2016 through December 2017 for articles published in English in peer-reviewed journals to identify data published (e-pub or print) since the previous search was performed in support of the 2015 Biennial Review (EPA-822-S18-003). The bibliographic databases searched included PubMed, Web of Science, Toxicology Literature Online (TOXLINE), Fish, Fisheries, & Aquatic Biodiversity Worldwide, Environment Complete, CAB Abstracts, and Science Direct. Conference abstracts, reports, comments, letters, and editorials were excluded.

Literature searches for chemical and microbial pollutants were conducted separately. For both chemicals and microbial pollutants, the search strategy for previous reporting periods was used with modifications. For the chemical search, keywords applicable only to microbial pollutants were eliminated. Likewise, keywords applicable only to chemicals were eliminated for the microbial pollutant search. To avoid limiting potential findings, the geographical search terms restricting the searches to studies conducted in the U.S. and Canada (the approach taken for past biennial reviews) were not used for the 2016-2017 reporting period for either chemical or microbial pollutants. The data in the studies found from other countries were analyzed in the review process for appropriateness for future risk determinations.

For *chemical pollutants*, health-related keywords were combined with chemical-related keywords, and land application keywords were dropped to broaden the search. Specifically, to be identified as a candidate, a paper had to have at least one **biosolids-related** keyword and at least one **chemical- or health-related** keyword:

**Biosolids-related keywords:** (sewage sludge OR biosolids OR treated sewage OR sludge treatment OR sewage treatment)

AND

**Chemical- and health-related keywords:** (pollutant\* OR toxic\* [toxicant, toxicology, etc.] OR chemical OR constituent OR contaminant\* OR metal\* OR dioxin\* OR inorganic\* OR organic\* OR flame retardant\* OR pharmaceutical\* OR steroid\* OR hormone\* OR antibiotic\* OR personal care product\*) OR (effect OR effects OR occurrence OR concentration).

For *microbial pollutants*, health-related and microbial pollutant-related keywords were kept separate to ensure that the chemical search was not duplicated (by requiring a microbial-related keyword). Land-application keywords were retained, because 40 CFR Part 503 includes site restrictions specific to land application of Class B biosolids [40 CFR 503.32(b)(5)] to allow time for environmental conditions to further reduce pathogen levels (U.S. EPA, 1994). Specifically, to be identified as a candidate, a paper had to have at least one **biosolids-related** keyword and at least one **land application-related** keyword and at least one **microbial pollutant-related** keyword and at least one **health-related** keyword:

**Biosolids-related Keywords:** (sewage sludge OR biosolids OR treated sewage OR sludge treatment OR sewage treatment)

AND

**Land Application-related Keywords:** (land application OR farm OR agriculture OR soil)

AND

**Microbial Pollutant-related Keywords:** (pathogen\* OR Salmonella OR microb\*)

AND

**Health-related Keywords:** (occurrence OR concentration OR effect OR effects OR propert\* OR fate OR transport OR health OR health effects).

For the papers identified in both literature searches (chemical and microbial), the EPA first screened the abstracts and then the full text (for abstracts not screened out) and excluded papers that met the following criteria:

- No abstract AND insufficient information in the title to determine whether it might be relevant.
- Endpoints not pollutant-specific (i.e., overall effects of biosolids on plant growth, crop yield, soil microbe community, or soil nutrients).

- Media evaluated not primarily sewage sludge, as defined in 40 CFR Part 503, including:
  - *Influent and effluent wastewater*;
  - *Industrial sewage sludge* (e.g., paper mill biosolids) - sewage sludge is defined in 40 CFR Part 503 to exclude sludge originating solely from industrial wastes with no domestic sewage component;
  - *Activated carbon* - this is derived from sewage sludge and is not biosolids;
  - *Activated sludge* - this is the sludge from secondary treatment, not biosolids, which typically includes primary sludge as well; 40 CFR Part 503 specifies that sewage sludge does not include these intermediate sludges;
  - *Biochar* - these are residuals from burning biosolids, which are also excluded by 40 CFR Part 503; or
  - *Biosolids compost* - many of these are a mixture of human biosolids, industrial sludge, plant waste, and other ingredients.
- Describes only an analytical method or effectiveness of treatment methods.

The EPA then reviewed the remaining papers to identify new pollutants with evidence regarding biosolids and pollutants that had previously been identified in either the Targeted National Sewage Sludge Survey (TNSSS; U.S. EPA, 2009) or a previous biennial review for which there were new or additional data in the papers. See **Appendix A** for a list of new and previously identified chemical and microbial pollutants.

During full-text reviews, papers were excluded when:

- Countries had treatment technologies, regulatory requirements or soil types that were not comparable to the U.S. and Canada AND reported only concentration data. Note that studies for countries that were not comparable to the U.S. and Canada but had other types of data (e.g., toxicity, fate and transport) were not excluded.
- Only spiked concentration data were reported.
- No evidence of the occurrence of new pollutants in biosolids AND no new data for chemicals previously identified were reported.
- The only reported data were for metals previously regulated in biosolids.<sup>1</sup>
- New classes of chemicals (e.g., nanoparticles) were found in biosolids but no data were available.

Abstracts of the papers retained after full-text review for chemical pollutants are provided in **Appendix B** (15 papers) and **Appendix C** (14 papers excluded because they contained data only on metals already regulated in biosolids, and three papers that identified new classes of

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<sup>1</sup> For more information on pollutants previously regulated or evaluated in biosolids, see the [Statistics Support Documentation for the 40 CFR Part 503 - Volume 1](https://www.epa.gov/sites/production/files/2015-04/documents/statistics_1992_support_document_-_biosolids_vol_i.pdf) ([https://www.epa.gov/sites/production/files/2015-04/documents/statistics\\_1992\\_support\\_document\\_-\\_biosolids\\_vol\\_i.pdf](https://www.epa.gov/sites/production/files/2015-04/documents/statistics_1992_support_document_-_biosolids_vol_i.pdf)) and EPA's response to the National Research Council of the National Academies report on biosolids ([https://www.epa.gov/sites/production/files/2015-06/documents/technical\\_background\\_document.pdf](https://www.epa.gov/sites/production/files/2015-06/documents/technical_background_document.pdf)).

chemicals in biosolids with no data). Abstracts of the papers retained after full-text review for microbial pollutants are provided in **Appendix D** (six papers).

## **2.2 Chemical Review: Evaluate Data Availability for Chemicals to be Included in the Biennial Review**

For both the new and previously identified chemicals, the data presented in the papers (e.g., concentration, physical-chemical properties, transfer factors) were extracted.

For chemicals newly identified in biosolids, the EPA also collected additional data required for risk assessment determinations (e.g., human and ecological toxicity, and environmental fate and transport data) that were not limited to the biennial reporting period (2016–2017). The preferred sources for the additional data are described in the next sub-sections.

For previously identified chemicals, the EPA extracted only data that had not previously been available. However, data were extracted for previously identified chemicals of particular interest for which limited data are available (e.g., triclosan, triclocarban, and perfluoroalkyl substances).

### **2.2.1 Human Health Toxicity Values Data Sources and Selection**

To estimate the potential for adverse human health effects from land application of biosolids, the EPA uses RfDs and RfCs to evaluate non-cancer risk from oral and inhalation exposures, respectively. The EPA uses oral CSFs and IURs to evaluate risk for carcinogens from oral and inhalation exposures.<sup>2</sup>

The EPA's Integrated Risk Information System (IRIS; U.S. EPA, 2018a) Program develops human health risk assessments, including toxicity values, using Agency guidance and standardized methods for hazard identification and dose response.<sup>3</sup> IRIS human health assessments are thoroughly peer reviewed and publicly available. The EPA's primary source of human health toxicity values to evaluate the potential human health risk is IRIS. However, not all chemicals have a toxicity value in IRIS, and some chemicals with a toxicity value in IRIS do not necessarily have all four toxicity values (RfD, RfC, CSF, IUR). Thus, a variety of other sources are used. To make efficient use of resources, the EPA developed a hierarchy that gives higher priority to sources of information that:

- Are developed specifically for use in human health risk assessment using methodologies similar to those used by IRIS;
- Have been peer reviewed and have a transparent basis for the values; and
- Are more recent than published IRIS values.

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<sup>2</sup> For more information about these toxicity values, see <https://www.epa.gov/iris/basic-information-about-integrated-risk-information-system>.

<sup>3</sup> For more information about these methods, see <https://www.epa.gov/iris/basic-information-about-integrated-risk-information-system#guidance>.

Note, however, that during the 2016-2017 biennial review process, only Tier 1 and Tier 2 sources of the EPA hierarchy were searched for each chemical.

- **Tier 1: Highest Quality EPA Sources:** Sources in Tier 1 contain values developed by the EPA specifically for human health risk assessment according to standard methods and represent the highest quality human health toxicity values available. These toxicity values are frequently used to support EPA risk analyses.
  - ***Integrated Risk Information System (IRIS):*** IRIS is the EPA’s primary repository for human health toxicity values that have been developed specifically for human health risk assessment using standardized methods and have been thoroughly peer reviewed. IRIS is considered the most preferred source for human health toxicity values for most EPA risk assessment. For pesticides, toxicity values are developed by the EPA’s Office of Pesticide Programs (U.S. EPA, 2018b).
  - ***Human Health Benchmarks for Pesticides (HHBPs):*** The EPA develops chronic oral health benchmarks (RfDs and CSFs) for pesticides for surface and groundwater sources of drinking water using health effects data submitted during the pesticide registration process (U.S. EPA, 2018b).
  - ***Provisional Peer Reviewed Toxicity Values (PPRTVs):*** The Superfund Health Risk Technical Support Center (in the National Center for Environmental Assessment, Office of Research and Development) develops PPRTVs using the same methods as IRIS (U.S. EPA, 2018c).
  - ***Health Effects Support Documents (HESDs):*** The Office of Science and Technology (in the EPA Office of Water) develops toxicity values for chemicals in drinking water using the same methodology as IRIS.
- **Tier 2: Non-EPA Sources Using a Similar Methodology to Tier 1:** Sources in Tier 2 contain toxicity values developed specifically for human health risk assessment by another organization using methods similar to IRIS. They represent the highest quality human health toxicity values available and are frequently used to support EPA risk analyses.
  - ***Agency for Toxic Substances and Disease Registry (ATSDR) Minimum Risk Levels (MRLs):*** ATSDR develops MRLs, which are oral non-cancer toxicity values equivalent to RfDs (ATSDR, 2016).
  - ***California Environmental Protection Agency (CalEPA) Reference Exposure Levels (RELs) and Cancer Potency Factors (CPF)s:*** CalEPA develops RELs, which are non-cancer toxicity values equivalent to RfDs or RfCs (CalEPA, 2016) and CPFs, which are cancer toxicity values equivalent to CSFs or IURs (CalEPA, 2011).

### 2.2.2 Ecological Toxicity Value Data Sources and Selection

To assess the potential for ecological risks from biosolids, the EPA assesses direct contact and ingestion pathways for aquatic and terrestrial species. For the direct contact exposure pathway,

species assemblages (or communities) are assessed in soil, sediment, and surface water where they are assumed to be exposed through direct contact with the contaminated medium. For the ingestion pathway, species are assumed to ingest contaminated food and prey from biosolids-treated agricultural fields and farm pond that receives runoff from a biosolids-treated field. The ecological toxicity values are expressed in terms of media concentration (e.g., mg/L for surface water and mg/kg for soil) for the direct contact pathway and in terms of dose (mg/kg-d) for the ingestion pathway.

The EPA does not have a single repository for approved ecological toxicity values directly comparable to IRIS for human toxicity values; however, the *ECOTOXicology knowledgebase* (ECOTOX; U.S. EPA, 2018d) was searched for all newly identified and existing chemicals to identify the number of papers and species, if any, that were available. ECOTOX is a comprehensive, publicly available knowledgebase providing single chemical environmental toxicity data on aquatic life, terrestrial plants and wildlife. Studies in the database must meet the following minimum criteria based on the ECOTOX applicability criteria:

- (1) The toxic effects are related to single chemical exposure;
- (2) There is a biological effect on live, whole organisms;
- (3) Chemical test concentrations are reported;
- (4) There is an explicit duration of exposure;
- (5) Toxicology information is reported for the chemical of concern;
- (6) The paper is published in the English language;
- (7) The paper is available as a full article (not an abstract);
- (8) The paper is publicly available;
- (9) The paper is the primary source of the data;
- (10) A calculated endpoint is reported or can be calculated using reported or available information;
- (11) Treatment(s) are compared to an acceptable control;
- (12) The location of the study (e.g., laboratory vs. field) is reported; and
- (13) The tested species is reported (with recognized nomenclature).

Studies from the open literature papers that pass the ECOTOX screen of applicability are considered potentially relevant and further evaluated for inclusion in risk assessments.

### **2.2.3 Environmental Fate and Transport Data**

The EPA uses risk assessment models that require physical-chemical properties and transfer factors to estimate the potential for chemical transport and uptake from agricultural lands amended with biosolids to drinking water, produce, animal products, fish, and ecological receptor diet items.

The EPA's preferred source for physical-chemical properties is the PHYSPROP database, which is incorporated in the Estimation Programs Interface (EPISuite™; U.S. EPA, 2017). EPISuite is

a suite of physical/chemical property and environmental fate estimation programs developed by the EPA and Syracuse Research Corp. (SRC) and packaged with the PHYSPROP database. It uses available physical-chemical inputs (either from PHYSPROP, input by the user, or a combination of both) to estimate missing physical-chemical properties and bioconcentration or bioaccumulation factors. Physical-chemical properties include:

- Molecular weight
- Solubility
- Vapor pressure
- Henry's law constant
- Log octanol-water partition coefficients ( $\log K_{ow}$ )
- Soil-water partitioning coefficients ( $K_d$  and  $K_{oc}$ )
- Degradation rates in various media
- Diffusivities in air and water
- Bioavailability

For many organic chemicals, the plant and animal product uptake/transfer factors can be estimated using empirical relationships between the transfer factor and  $\log K_{ow}$ . Bioaccumulation factors are preferred if available. Fish bioconcentration factors (BCFs) can be estimated using EPISuite, if some physical-chemical property data are available. Transfer factors include the following:

- Air-to-plant transfer factors
- Root uptake factors for above-ground vegetation
- Root concentration factors (for root vegetables)
- Bioconcentration factors for fish
- Bioaccumulation factors for various plants and small fauna eaten by ecological receptors
- Bioconcentration factors for animal products (e.g., meat and milk)

### 3. Results from the Biosolids Biennial Review for the 2016–2017 Reporting Period

During the Paper Review stage of the literature review process completed for the 2016–2017 reporting period, the EPA identified 32 articles that met the eligibility criteria for chemicals. Review of these articles identified 28 new chemicals in biosolids (see **Section 3.1**): seven polybrominated diphenyl ethers (PBDEs); nine parabens and metabolites; five brominated flame retardants (BFRs); three other flame retardants; two perfluoroalkyl substances (PFASs); and two triclosan transformation products. These articles also provided new or additional data on 31 previously identified chemicals (see **Section 3.2**). Of the 32 papers, 15 identified new chemicals or provided data on previously identified chemicals (abstracts provided in **Appendix B**); 14 contained data on already regulated metals (abstracts provided in **Appendix C, Section C.1**); and three contained information on novel chemical groups (i.e., nanoparticles and microplastics)

(abstracts provided in **Appendix C, Section C.2**). During the Chemical Review stage, data identified include the following:

- Concentration data were identified for all 28 newly identified chemicals and two previously identified chemicals (diclofenac and tonalide).
- Human health toxicity data were identified for three newly identified chemicals (see **Section 3.1.1**) and two previously identified chemicals (see **Section 3.2.1**).
- ECOTOX records were found for 17 newly identified chemicals (see **Section 3.1.2**) and 26 previously identified chemicals (see **Section 3.2.2**).
- Physical-chemical property data were identified for 22 newly identified chemicals (see **Section 3.1.3**) and 20 previously identified chemicals (see **Section 3.2.3**).
- Bioconcentration or bioaccumulation factors were identified for 23 newly identified chemicals (see **Section 3.1.3**) and 24 previously identified chemicals (see **Section 3.2.3**).

The EPA also identified six articles that met the eligibility criteria for microbial pollutants (abstracts provided in **Appendix D**). Review of these articles identified no newly identified microbial pollutants in biosolids and provided potentially useful data on seven previously identified microbial pollutants: *Cryptosporidium* spp., *Giardia* spp., antibiotic resistance genes (ARGs)/antibiotic resistant bacteria (AR bacteria), *Salmonella*, *Escherichia coli*, human norovirus, and human adenovirus.

### **3.1 Chemicals Newly Identified in the 2016-2017 Biennial Review**

**Table 1** lists the 28 new chemicals identified in the 2016–2017 reporting period. As stated previously, for chemicals newly identified in biosolids, the EPA also collected data required to inform risk assessments from preferred sources not limited to the biennial reporting period (2016–2017) (see **Sections 2.2.1** and **2.2.2** in this review). Chemical concentrations in biosolids were reported in the U.S., Canada, Australia, Spain and India. The EPA will evaluate these data to determine whether they are representative of U.S. biosolids.

**Table 1. Chemicals Newly Identified in Biosolids in the 2016–2017 Reporting Period and Types of Data Available**

Chemical Name	CAS No.	Class	Concentration in Biosolids	Physical-chemical Properties	Bio-transfer Factors	Human Toxicity Values	Ecological Toxicity Values
BDE-17	147217-75-2	PBDEs	✓	✓	✓	x	x
BDE-184	117948-63-7	PBDEs	✓	✓	✓	x	x
BDE-191	446255-30-7	PBDEs	✓	✓	✓	x	x
BDE-196	446255-39-6	PBDEs	✓	✓	✓	x	x
BDE-197	117964-21-3	PBDEs	✓	✓	✓	x	x
BDE-206	63387-28-0	PBDEs	✓	✓	✓	x	x
BDE-207	437701-79-6	PBDEs	✓	✓	✓	x	x
Benzoic acid	65-85-0	Parabens	✓	✓	✓	✓	(✓)
Benzyl paraben	94-18-8	Parabens	✓	✓	✓	x	x
Bis(2,4,6-tribromophenoxy)ethane, 1,2-	37853-59-1	Brominated flame retardants	✓	✓	✓	x	(✓)
Butyl paraben	94-26-8	Parabens	✓	✓	✓	x	(✓)
Decabromodiphenyl ethane	84852-53-9	Brominated flame retardants	✓	x	✓	x	x
Dechlorane 602	31107-44-5	Other flame retardants	✓	x	✓	x	(✓)
Dechlorane 603	13560-92-4	Other flame retardants	✓	x	✓	x	(✓)
Dechlorane Plus	13560-89-9	Other flame retardants	✓	✓	✓	x	(✓)
Dichlorophenol, 2,4-	120-83-2	Triclosan transformation products	✓	✓	✓	✓	(✓)
Dihydroxybenzoic acid, 3,4-	99-50-3	Parabens	✓	✓	✓	x	(✓)
Ethyl paraben	120-47-8	Parabens	✓	✓	✓	x	(✓)
Hexabromobenzene	87-82-1	Brominated flame retardants	✓	✓	✓	✓	(✓)
Hexabromocyclododecane	3194-55-6	Brominated flame retardants	✓	✓	✓	x	(✓)
Hydroxybenzoate, 4-	99-96-7	Parabens	✓	✓	✓	x	(✓)
Methyl paraben	99-76-3	Parabens	✓	✓	✓	x	(✓)
Methyl protocatechuate	2150-43-8	Parabens	✓	x	x	x	x
Methyl triclosan	4640-01-1	Triclosan transformation products	✓	x	x	x	x
Pentabromoethylbenzene	85-22-3	Brominated flame retardants	✓	x	x	x	(✓)
Perfluorotetradecanoic acid	376-06-7	PFASs	✓	✓	x	x	(✓)
Perfluorotridecanoic acid	72629-94-8	PFASs	✓	✓	x	x	(✓)
Propyl paraben	94-13-3	Parabens	✓	✓	✓	x	(✓)

Key: ✓ = Some data available.

(✓) = ECOTOX papers or species found.

X = No data available from standard sources searched (see **Section 2.2**).

### 3.1.1 Human Health Toxicity Values for Newly Identified Chemicals

Human health toxicity values were found for three of the new chemicals identified in biosolids in the 2016–2017 reporting period: benzoic acid; 2,4-dichlorophenol; and hexabromobenzene (**Table 2**).

**Table 2. Human Health Toxicity Values Found for Chemicals Newly Identified in the 2016–2017 Reporting Period**

Chemical Name	RfD (mg/kg-d)	Last Revised	Source
Benzoic acid	4	1/31/1987	IRIS
Dichlorophenol, 2,4-	0.003	1/31/1987	IRIS
Hexabromobenzene	0.002	3/31/1987	IRIS

IRIS = Integrated Risk Information System (U.S. EPA, 2018a).

RfD = Reference Dose.

mg/kg-d = milligram/kilogram/day.

### 3.1.2 ECOTOX Results for Newly Identified Chemicals

ECOTOX papers (U.S. EPA, 2018d) were found for 17 newly identified chemicals (**Table 3**).<sup>4</sup> These papers will require further evaluation for relevance for inclusion in risk assessments.

**Table 3. Summary of Papers Found in ECOTOX for Chemicals Newly Identified in the 2016–2017 Reporting Period<sup>5</sup>**

Chemical Name	# Aquatic Papers	# Aquatic Species	# Terrestrial Papers	# Terrestrial Species
Benzoic acid	16	16	12	12
Bis(2,4,6-tribromophenoxy)ethane, 1,2-	2	3	1	3
Butylparaben	5	4	0	0
Dechlorane 602	1	1	0	0
Dechlorane 603	1	1	0	0
Dechlorane Plus	2	2	0	0
Dichlorophenol, 2,4-	66	56	17	17
Dihydroxybenzoic acid, 3,4-	2	4	3	3
Ethyl paraben	2	2	0	0
Hexabromobenzene	3	2	1	1
Hexabromocyclododecane	3	3	3	3
Hydroxybenzoate, 4-	2	2	5	5
Methylparaben	4	4	0	0
Pentabromoethylbenzene	2	2	0	0
Perfluorotetradecanoic acid	1	1	1	1
Perfluorotridecanoic acid	0	0	1	1
Propyl paraben	3	4	0	0

<sup>4</sup> All ECOTOX searches were completed using the Search function, rather than Explore function. Most searches were completed in June 2018 using the legacy version of ECOTOX; a few (Dechlorane 602, Dechlorane 603, Dechlorane Plus, and hexabromocyclododecane) were completed in September 2018 using ECOTOX version 5.

<sup>5</sup> ECOTOX results were not found for all newly identified chemicals.

### 3.1.3 Environmental Fate and Transport Properties for Newly Identified Chemicals

**Table 4** presents log  $K_{ow}$  values for nine of the newly identified chemicals from the papers reviewed; however, no further physical chemical properties data are available for these chemicals. Values for Henry's law constant (HLC),  $K_{oc}$ , log  $K_{ow}$ , vapor pressure (VP), and solubility (Sol) are available in PHYSPROP or can be estimated using EPISuite (U.S. EPA, 2017) for 13 more of the newly identified chemicals. Molecular weight (MW) for all chemicals in Table 4 is calculated from chemical structure.

**Table 4. Physical-Chemical Properties Found for Chemicals Newly Identified in the 2016–2017 Reporting Period**

Chemical	MW <sup>a</sup> (g/mol)	log $K_{ow}$ <sup>b</sup> (log L/kg)	$K_{oc}$ (L/kg)	HLC (atm·m <sup>3</sup> /mole)	VP (mmHg)	Sol (mg/L)	Source
BDE-17	436.92	5.88	NR	NR	NR	NR	Navarro et al., 2017 <sup>b</sup>
BDE-184	722.48	9.44	NR	NR	NR	NR	Navarro et al., 2017 <sup>b</sup>
BDE-191	722.48	9.44	NR	NR	NR	NR	Navarro et al., 2017 <sup>b</sup>
BDE-196	801.38	10.33	NR	NR	NR	NR	Navarro et al., 2017 <sup>b</sup>
BDE-197	801.38	10.33	NR	NR	NR	NR	Navarro et al., 2017 <sup>b</sup>
BDE-206	880.28	11.22	NR	NR	NR	NR	Navarro et al., 2017 <sup>b</sup>
BDE-207	880.28	11.22	NR	NR	NR	NR	Navarro et al., 2017 <sup>b</sup>
Benzoic acid	122.12	1.87	17	1.08E-07	7.0E-04	3,400	PHYSPROP, EPISuite <sup>c</sup>
Benzyl paraben	228.25	3.56	3,229	2.92E-10	3.8E-06	122	PHYSPROP, EPISuite <sup>c</sup>
Bis(2,4,6-tribromo-phenoxy)ethane, 1,2-	687.64	8.9	4.5E+04	7.32E-09	1.9E-10	0.2	PHYSPROP, EPISuite <sup>c</sup>
Butylparaben	194.23	3.57	522	8.45E-09	2.5E-04	207	PHYSPROP, EPISuite <sup>c</sup>
Dechlorane Plus	653.73	11.27	4.8E+07	7.44E-06	2.4E-11	4.4E-08	PHYSPROP, EPISuite <sup>c</sup>
Dichlorophenol, 2,4-	163	3.06	499	3.08E-07	1.2E-01	5,520	PHYSPROP, EPISuite <sup>c</sup>
Dihydroxybenzoic acid, 3,4-	154.12	0.86	28	1.17E-15	1.8E-07	18,200	PHYSPROP, EPISuite <sup>c</sup>
Ethyl paraben	166.18	2.47	157	4.79E-09	9.3E-05	885	PHYSPROP, EPISuite <sup>c</sup>
Hexabromobenzene	551.49	6.07	2,807	2.15E-05	1.7E-08	1.6E-04	PHYSPROP, EPISuite <sup>c</sup>
Hexabromocyclododecane	641.7	7.74	9.7E+04	1.72E-06	4.7E-07	8.6E-03	PHYSPROP, EPISuite <sup>c</sup>
Hydroxy benzoic acid, 4-	138.12	1.58	21	1.13E-11	1.9E-07	5,000	PHYSPROP, EPISuite <sup>c</sup>
Methylparaben	152.15	1.96	86	3.61E-09	2.4E-04	2,500	PHYSPROP, EPISuite <sup>c</sup>
Perfluorotetradecanoic acid	714.12	8.83	NR	NR	NR	NR	Navarro et al., 2017 <sup>b</sup>
Perfluorotridecanoic acid	664.11	8.16	NR	NR	NR	NR	Navarro et al., 2017 <sup>b</sup>
Propyl paraben	180.21	3.04	287	6.37E-09	3.1E-04	500	PHYSPROP, EPISuite <sup>c</sup>

<sup>a</sup> All molecular weights were calculated from the chemical formula.

<sup>b</sup> The log  $K_{ow}$  values from Navarro et al. (2017) were estimated using EPISuite v 4.1.

<sup>c</sup> Estimated using EPISuite v4.11.

NR = Not reported.

**Tables 5 and 6** present, respectively, terrestrial bioaccumulation factors (BAFs) for 11 newly identified chemicals, and aquatic bioconcentration factors (BCFs) for 13 newly identified chemicals.

**Table 5. Terrestrial Bioaccumulation Factors Found for Chemicals Newly Identified in the 2016–2017 Reporting Period (All Values Unitless)**

Chemical Name	Receptor	Minimum	Median	Maximum	Reference <sup>a</sup>
BDE-17	Earthworm	8.41	NR	10	Navarro et al., 2016
BDE-17	Spinach	2.28	NR	3.08	Navarro et al., 2017
BDE-184	Earthworm	0.69	1.34	1.51	Navarro et al., 2016
BDE-191	Earthworm	1.35	2.215	3.4	Navarro et al., 2016
BDE-196	Earthworm	0.48	0.55	0.65	Navarro et al., 2016
BDE-196	Tomato fruit	NR	0.11	NR	Navarro et al., 2017
BDE-196	Tomato leaf	0.12	NR	0.16	Navarro et al., 2017
BDE-196	Tomato stem	NR	0.03	NR	Navarro et al., 2017
BDE-197	Earthworm	0.26	0.605	0.79	Navarro et al., 2016
BDE-197	Tomato fruit	NR	0.11	NR	Navarro et al., 2017
BDE-197	Tomato leaf	0.1	NR	0.11	Navarro et al., 2017
BDE-206	Earthworm	0.42	1.115	4.27	Navarro et al., 2016
BDE-206	Tomato fruit	0.72	NR	0.8	Navarro et al., 2017
BDE-206	Tomato leaf	0.11	NR	0.55	Navarro et al., 2017
BDE-207	Earthworm	0.45	0.72	1.22	Navarro et al., 2016
BDE-207	Tomato fruit	0.5	NR	0.62	Navarro et al., 2017
BDE-207	Tomato leaf	0.11	NR	0.3	Navarro et al., 2017
Decabromodiphenyl ethane	Tomato leaf	0.05	NR	1.08	Navarro et al., 2017
Decabromodiphenyl ethane	Tomato root	0.03	NR	0.1	Navarro et al., 2017
Decabromodiphenyl ethane	Tomato stem	NR	0.05	NR	Navarro et al., 2017
Dechlorane 602	Earthworm	0.96	3.03	5.61	Navarro et al., 2016
Dechlorane 603	Earthworm	0.28	0.37	0.95	Navarro et al., 2016
Dechlorane Plus <sup>b</sup>	Earthworm	3.26	3.56	56.9	Navarro et al., 2016
Dechlorane Plus <sup>b</sup>	Spinach	19.06	NR	28.29	Navarro et al., 2017
Dechlorane Plus <sup>b</sup>	Tomato fruit	0.55	NR	1.1	Navarro et al., 2017
Dechlorane Plus <sup>b</sup>	Tomato leaf	0.48	NR	0.64	Navarro et al., 2017
Dechlorane Plus <sup>b</sup>	Tomato root	0.83	NR	1.6	Navarro et al., 2017
Dechlorane Plus <sup>b</sup>	Tomato stem	0.21	NR	0.3	Navarro et al., 2017

NR = Not reported.

<sup>a</sup> Reference notes:

Navarro et al. (2016) presented BAF data for earthworms for a control and four treatments. Navarro et al. control values were not used. If only one treatment had a value, it is shown here as the median. If there were two treatment values, they are shown here as the minimum and maximum. If there were three or four treatment values, the minimum, maximum, and median (either the middle value of three or the mean of the two middle values of four) are shown.

Navarro et al. (2017) presented BAF data for spinach and tomato plant parts for a control and two treatments. Navarro et al. control values were not used. If only one treatment had a value, it is shown as the median. If there were two treatment values, they are shown as the minimum and maximum.

<sup>b</sup> All values from Navarro et al. (2017) were for anti-DP, while the values from Navarro et al. (2016) were for total DP (anti-DP + syn-DP). The values for anti- and syn-DP were very similar, so the anti-DP values are presented in the table as they were slightly higher.

**Table 6. Aquatic Bioconcentration Factors Found for Chemicals Newly Identified in the 2016–2017 Reporting Period (All Data from EPISuite)**

Receptor	TL3 Fish Value ([mg/kg]/[mg/L])	TL4 Fish Value ([mg/kg]/[mg/L])
Benzoic acid	5.2	6.5
Benzyl paraben	13	9.8
Bis(2,4,6-tribromophenoxy)ethane, 1,2-	1,200	780
Butylparaben	34	27
Dechlorane Plus	7.8	5.4
Dichlorophenol, 2,4-	36	34
Dihydroxybenzoic acid, 3,4-	1.1	1.0
Ethyl paraben	8.8	8.2
Hexabromobenzene	1,200	880
Hexabromocyclododecane	1,200	850
Hydroxy benzoic acid, 4-	3.0	3.4
Methylparaben	4.0	3.9
Propylparaben	19	16

### 3.2 Data Found on Chemicals Previously Identified in Biennial Reviews

In each biennial review, in addition to reporting newly identified chemicals in biosolids, the EPA also reviews the literature for concentration data, physical-chemical properties, ecological toxicity data, and environmental fate data for chemical pollutants previously identified in biosolids in the TNSSS, open literature, or previous biennial reviews. **Appendix A** provides a complete list of all previously identified chemicals. Previously identified chemicals from the EPA's 1989 National Sewage Sludge Survey and the EPA's 2001 National Sewage Sludge Survey are presented also in Appendix A. Previously identified chemicals for which new data were found are shown in **Table 7**, along with the types of data available for these chemicals, whether those data were found in this biennial review or an earlier one.

**Table 7. Previously Identified Chemicals with Data Found in the 2016–2017 Reporting Period and Types of Data Available**

Chemical Name	CAS	Class	Concentration in Biosolids	Physical-chemical Properties	Bio-transfer Factors	Human Toxicity Values	Ecological Toxicity Values
BDE-100	97038-97-6	PBDEs	•	✓	✓	X	X
BDE-153	68631-49-2	PBDEs	•	•	✓	•	(✓)
BDE-154	207122-15-4	PBDEs	•	✓	✓	X	X
BDE-183	207122-16-5	PBDEs	•	✓	✓	X	(✓)
BDE-209	1163-19-5	PBDEs	•	•	✓	•	(✓)
BDE-28	6430-90-6	PBDEs	•	✓	✓	X	X
BDE-47	5436-43-1	PBDEs	•	•	✓	•	(✓)
BDE-66	84303-45-7	PBDEs	•	✓	✓	X	X
BDE-99	60348-60-9	PBDEs	•	•	✓	•	(✓)
Diclofenac	15307-86-5	Pharmaceuticals	✓	•	•	•	(✓)
Ethinylestradiol, 17 $\alpha$ -	57-63-6	Hormones	X	•	✓	X	(✓)
Galaxolide	1222-05-5	Musk fragrances	•	•	✓	X	(✓)
Nonylphenol	25154-52-3	Surfactants	X	•	X	X	(✓)

Chemical Name	CAS	Class	Concentration in Biosolids	Physical-chemical Properties	Bio-transfer Factors	Human Toxicity Values	Ecological Toxicity Values
(continued)							
Nonylphenol diethoxylate	30-53-3	Surfactants	x	✓	x	x	(✓)
Nonylphenol monoethoxylate	27986-36-3	Surfactants	x	✓	x	x	(✓)
Perfluorobutanesulfonate	45187-15-3	PFASs	•	✓	x	x	(✓)
Perfluorobutanoate	375-22-4	PFASs	•	✓	✓	x	(✓)
Perfluorodecanoate	335-76-2	PFASs	•	✓	✓	x	(✓)
Perfluorododecanoate	307-55-1	PFASs	•	✓	✓	x	(✓)
Perfluoroheptanoate	375-85-9	PFASs	•	✓	✓	x	(✓)
Perfluorohexanesulfonate	108427-53-8	PFASs	•	✓	x	x	(✓)
Perfluorohexanoate	307-24-4	PFASs	•	✓	✓	x	(✓)
Perfluorononanoate	375-95-1	PFASs	•	✓	✓	x	(✓)
Perfluorooctane sulfonamide	754-91-6	PFASs	•	✓	x	x	(✓)
Perfluoropentanoate	2706-90-3	PFASs	•	✓	✓	x	(✓)
Perfluoroundecanoate	2058-94-8	PFASs	•	✓	✓	x	(✓)
PFOA	335-67-1	PFASs	•	✓	✓	✓	(✓)
PFOS	2795-39-3	PFASs	•	✓	✓	✓	(✓)
Tonalide (AHTN)	1506-02-1	Musk fragrances	✓	•	✓	x	x
Triclocarban	101-20-2	Antimicrobials	•	•	✓	x	(✓)
Triclosan	3380-34-5	Antimicrobials	•	•	✓	•	(✓)

Key: ✓ = Data found in this 2016-2017 biennial review reporting period.

• = Data from an earlier biennial review.

(✓) = ECOTOX papers or species found.

X = No data available from standard sources searched (see Section 2.2).

### 3.2.1 Human Health Toxicity Values for Previously Identified Chemicals

Table 8 presents data for two previously identified chemicals—perfluorooctanoate (PFOA) and perfluorooctanesulfonate (PFOS)—for which a human health toxicity value was found during the biennial review for the 2016–2017 reporting period.

**Table 8. Human Health Toxicity Values Found in the 2016–2017 Reporting Period for Chemicals Previously Identified**

Chemical Name	RfD (mg/kg-d)	CSForal (per mg/kg-d)	Source
PFOA	2E-5	7E-2	HESD for PFOA U.S. EPA (2016a)
PFOS	2E-5	na	HESD for PFOS U.S. EPA (2016b)

CSForal = Oral cancer slope factor.

RfD = Reference dose.

mg/kg/day = milligram/kilogram/day.

HESD = Health Effects Support Document.

### 3.2.2 ECOTOX Results for Previously Identified Chemicals

ECOTOX papers (U.S. EPA, 2018d) were found for 26 previously identified chemicals (Table 9).<sup>6</sup> These papers will require further evaluation for relevance for inclusion in risk assessments.

**Table 9. Summary of Papers Found in ECOTOX in the 2016–2017 Reporting Period for Chemicals Previously Identified<sup>7</sup>**

Chemical Name	# Aquatic Papers	# Aquatic Species	# Terrestrial Papers	# Terrestrial Species
BDE-153	1	1	0	0
BDE-183	1	1	0	0
BDE-209	2	2	9	6
BDE-47	9	15	0	0
BDE-99	6	6	0	0
Diclofenac	11	13	7	3
Ethinylestradiol, 17 $\alpha$ -	73	219	17	17
Galaxolide	12	11	4	5
Nonylphenol	42	54	6	5
Nonylphenol diethoxylate	3	4	0	0
Nonylphenol monoethoxylate	2	1	0	0
Perfluorobutanesulfonate	2	2	0	0
Perfluorobutanoate	3	3	1	2
Perfluorodecanoate	4	3	3	4
Perfluorododecanoate	1	1	1	1
Perfluoroheptanoate	2	1	2	2
Perfluorohexanesulfonate	0	0	2	2
Perfluorohexanoate	0	0	2	2
Perfluorononanoate	3	5	3	5
Perfluorooctane sulfonamide	0	0	1	1
Perfluoropentanoate	2	1	1	1
Perfluoroundecanoate	2	2	2	2
PFOA	21	23	7	9
PFOS	11	14	3	3
Triclocarban	13	29	0	0
Triclosan	25	31	7	9

<sup>6</sup> All ECOTOX searches were done using the Search function, rather than the Explore function. Most searches were completed in June 2018 using the legacy version of ECOTOX. Two searches (nonylphenol diethoxylate and nonylphenol monoethoxylate) were completed in September 2018 using ECOTOX version 5.

<sup>7</sup> ECOTOX results were not found for all newly identified chemicals.

### 3.2.3 Environmental Fate and Transport Properties for Previously Identified Chemicals

**Table 10** presents data for 20 previously identified chemicals for which physical-chemical properties data were found in the 2016–2017 reporting period.

**Table 10. Physical-Chemical Properties Found in the 2016–2017 Reporting Period for Chemicals Previously Identified**

Chemical	MW <sup>a</sup> (g/mol)	log K <sub>ow</sub> <sup>b</sup> (log L/kg)	K <sub>oc</sub> (L/kg)	HLC (atm·m <sup>3</sup> /mole)	Source
BDE-100	564.69	7.66	NR	NR	Navarro et al. (2017)
BDE-154	643.59	8.55	NR	NR	Navarro et al. (2017)
BDE-183	722.48	9.44	NR	NR	Navarro et al. (2017)
BDE-28	406.90	5.88	NR	NR	Navarro et al. (2017)
BDE-66	485.80	6.77	NR	NR	Navarro et al. (2017)
Nonylphenol diethoxylate, 4-	308.235	4.17	NR	2.2E-7–9.1E-7 <sup>c</sup>	Clarke et al. (2016)
Nonylphenol monoethoxylate, 4-	264.403	4.48	NR	1.4E-5–2.9E-5 <sup>c</sup>	Clarke et al. (2016)
Perfluorobutane sulfonate	300.1	1.82	NR	NR	Navarro et al. (2017)
Perfluorobutanoic acid	214.039	2.14	NR	NR	Navarro et al. (2017)
PFOS	499.118	4.49	372	NR	Navarro et al. (2017) (logKow) HESD (Koc)
Perfluorodecanoic acid	514.086	6.15	NR	NR	Navarro et al. (2017)
Perfluorododecanoic acid	614.101	7.49	NR	NR	Navarro et al. (2017)
Perfluoroheptanoic acid	354.062	4.15	NR	NR	Navarro et al. (2017)
Perfluorohexane sulfonate	400.11	3.16	NR	NR	Navarro et al. (2017)
Perfluorohexanoic acid	314.054	3.48	NR	NR	Navarro et al. (2017)
Perfluorononanoic acid	464.078	5.48	NR	NR	Navarro et al. (2017)
Perfluorooctane sulfonamide	499.142	5.8	NR	NR	Navarro et al. (2017)
PFOA	414.07	4.81	115	NR	Navarro et al. (2017) (logKow) HESD (Koc)
Perfluoropentanoic acid	264.047	2.81	NR	NR	Navarro et al. (2017)
Perfluoroundecanoic acid	564.093	6.82	NR	NR	Navarro et al. (2017)

<sup>a</sup> All molecular weights were calculated from the chemical formula.

<sup>b</sup> The log K<sub>ow</sub> values from Navarro et al. (2017) were estimated using EPISuite v 4.1.

<sup>c</sup> Clarke et al. (2016) provided minimum and maximum of a uniform distribution for unitless K<sub>H</sub> of 9.3E-6–3.8E-5 for nonylphenol diethoxylate and 5.8E-4–1.2E-3 for nonylphenol monoethoxylate. These values have been converted to atm·m<sup>3</sup>/mol (shown in this table) using a temperature of 20 C and EPA's On-line Tools for Site Assessment, available at <https://www3.epa.gov/ceampubl/learn2model/part-two/onsite/henryslaw.html>.

NR = Not reported.

**Table 11** presents terrestrial BAFs for 24 previously identified chemicals. For five of these (BDE-47, -99, -153, -209, and triclosan), the new data do not fill data gaps, but are potentially useful for validating transfer factors estimated from log K<sub>ow</sub> for these chemicals.

**Table 11. Terrestrial Bioaccumulation Factors Found in the 2016–2017 Reporting Period for Chemicals Previously Identified**

Chemical Name	Receptor	Minimum	Median	Maximum	Reference <sup>a</sup>
BDE-100	Earthworm	10.6	17.9	20.3	Navarro et al. (2016)
BDE-100	Spinach	NR	0.45	NR	Navarro et al. (2017)
BDE-100	Tomato fruit	0.47	NR	0.65	Navarro et al. (2017)
BDE-100	Tomato leaf	0.53	NR	0.59	Navarro et al. (2017)
BDE-100	Tomato root	0.97	NR	0.98	Navarro et al. (2017)
BDE-153	Earthworm	2.12	3.18	3.57	Navarro et al. (2016)
BDE-153	Spinach	0.06	NR	0.38	Navarro et al. (2017)
BDE-153	Tomato leaf	0.07	NR	0.1	Navarro et al. (2017)
BDE-153	Tomato root	NR	0.28	NR	Navarro et al. (2017)
BDE-154	Earthworm	4.14	5.055	5.64	Navarro et al. (2016)
BDE-154	Spinach	0.22	NR	0.24	Navarro et al. (2017)
BDE-154	Tomato fruit	NR	4.47	NR	Navarro et al. (2017)
BDE-154	Tomato leaf	0.06	NR	0.07	Navarro et al. (2017)
BDE-154	Tomato root	NR	0.22	NR	Navarro et al. (2017)
BDE-154	Tomato stem	NR	0.01	NR	Navarro et al. (2017)
BDE-183	Earthworm	0.39	1.105	1.3	Navarro et al. (2016)
BDE-183	Spinach	NR	0.37	NR	Navarro et al. (2017)
BDE-183	Tomato fruit	NR	0.21	NR	Navarro et al. (2017)
BDE-183	Tomato root	NR	1.16	NR	Navarro et al. (2017)
BDE-209	Earthworm	3.04	3.71	4.74	Navarro et al. (2016)
BDE-209	Tomato fruit	1.35	NR	1.41	Navarro et al. (2017)
BDE-209	Tomato leaf	0.56	NR	1.58	Navarro et al. (2017)
BDE-28	Earthworm	6.94	13.2	18.4	Navarro et al. (2016)
BDE-28	Tomato fruit	5.44	NR	7.54	Navarro et al. (2017)
BDE-28	Tomato leaf	1.94	NR	3.3	Navarro et al. (2017)
BDE-28	Tomato root	3.21	NR	9.43	Navarro et al. (2017)
BDE-47	Earthworm	16.2	21.1	26.9	Navarro et al. (2016)
BDE-47	Spinach	1.02	NR	2.12	Navarro et al. (2017)
BDE-47	Tomato fruit	2.15	NR	2.17	Navarro et al. (2017)
BDE-47	Tomato leaf	1.6	NR	1.64	Navarro et al. (2017)
BDE-47	Tomato root	1.77	NR	2.11	Navarro et al. (2017)
BDE-66	Tomato stem	0.03	NR	0.12	Navarro et al. (2017)
BDE-99	Earthworm	12.0	13.4	14.4	Navarro et al. (2016)
BDE-99	Spinach	0.45	NR	0.93	Navarro et al. (2017)
BDE-99	Tomato fruit	0.23	NR	0.35	Navarro et al. (2017)
BDE-99	Tomato leaf	0.33	NR	0.63	Navarro et al. (2017)
BDE-99	Tomato root	1.61	NR	2.93	Navarro et al. (2017)
Ethinylestradiol, 17 $\alpha$ -	Wheat root	0.229	0.294	0.404	Cantarero et al. (2017)
Ethinylestradiol, 17 $\alpha$ -	Wheat shoot	0.104	0.141	0.192	Cantarero et al. (2017)
Galaxolide	Earthworm	NR	1.91	NR	Havranek et al. (2017)
Perfluorobutanoate	Tomato fruit	30.87	NR	69.82	Navarro et al. (2017)
Perfluorobutanoate	Tomato leaf	NR	94.41	NR	Navarro et al. (2017)
Perfluorobutanoate	Tomato stem	15.35	NR	21.88	Navarro et al. (2017)
Perfluorodecanoate	Earthworm	4.76	9.08	23.3	Navarro et al. (2016)
Perfluorodecanoate	Tomato fruit	0.02	NR	0.02	Navarro et al. (2017)
Perfluorodecanoate	Tomato root	0.22	NR	2.7	Navarro et al. (2017)
Perfluorodecanoate	Tomato stem	0.1	NR	0.24	Navarro et al. (2017)

(continued)

Chemical Name	Receptor	Minimum	Median	Maximum	Reference <sup>a</sup>
Perfluorododecanoate	Earthworm	82.9	154.5	402	Navarro et al. (2016)
Perfluoroheptanoate	Tomato fruit	NR	0.91	NR	Navarro et al. (2017)
Perfluoroheptanoate	Tomato root	NR	5.33	NR	Navarro et al. (2017)
Perfluorohexanoate	Tomato fruit	3.64	NR	5.06	Navarro et al. (2017)
Perfluorohexanoate	Tomato leaf	NR	6.91	NR	Navarro et al. (2017)
Perfluorohexanoate	Tomato root	NR	1	NR	Navarro et al. (2017)
Perfluorohexanoate	Tomato stem	NR	1.77	NR	Navarro et al. (2017)
Perfluorononanoate	Tomato leaf	NR	0.28	NR	Navarro et al. (2017)
Perfluorononanoate	Tomato root	NR	4.53	NR	Navarro et al. (2017)
Perfluoropentanoate	Spinach	NR	1.08	NR	Navarro et al. (2017)
Perfluoropentanoate	Tomato fruit	NR	31.22	NR	Navarro et al. (2017)
Perfluoropentanoate	Tomato leaf	NR	27.84	NR	Navarro et al. (2017)
Perfluoropentanoate	Tomato root	NR	6.12	NR	Navarro et al. (2017)
Perfluoropentanoate	Tomato stem	NR	11.67	NR	Navarro et al. (2017)
Perfluoroundecanoate	Earthworm	50.5	NR	123	Navarro et al. (2016)
PFOA	Earthworm	1.17	NR	2.89	Navarro et al. (2016)
PFOA	Spinach	NR	1.63	NR	Navarro et al. (2017)
PFOA	Tomato fruit	NR	0.08	NR	Navarro et al. (2017)
PFOA	Tomato leaf	3.55	NR	4.14	Navarro et al. (2017)
PFOA	Tomato root	1.54	NR	4.37	Navarro et al. (2017)
PFOA	Tomato stem	0.35	NR	0.55	Navarro et al. (2017)
PFOS	Earthworm	10.7	26.5	30.3	Navarro et al. (2017)
PFOS	Spinach	3.82	NR	4.47	Navarro et al. (2017)
PFOS	Tomato fruit	NR	0.06	NR	Navarro et al. (2017)
PFOS	Tomato leaf	0.36	NR	1.24	Navarro et al. (2017)
PFOS	Tomato root	1.44	NR	2.25	Navarro et al. (2017)
PFOS	Tomato stem	0.05	NR	0.45	Navarro et al. (2017)
Tonalide (AHTN)	Earthworm	NR	0.34	NR	Havranek et al. (2017)
Triclocarban	Deer mouse liver	NR	0.2	NR	Sherburne et al. (2016)
Triclocarban	Earthworm	NR	0.79	NR	Sherburne et al. (2016)
Triclocarban	Kestrel eggs	NR	0.05	NR	Sherburne et al. (2016)
Triclocarban	Starling eggs	NR	0.25	NR	Sherburne et al. (2016)
Triclosan	Deer mouse liver	NR	0.5	NR	Sherburne et al. (2016)
Triclosan	Earthworm	NR	10.9	NR	Havranek et al. (2017)
Triclosan	Earthworm	NR	67	NR	Sherburne et al. (2016)
Triclosan	Kestrel eggs	NR	0.77	NR	Sherburne et al. (2016)
Triclosan	Starling eggs	NR	2	NR	Sherburne et al. (2016)
Triclosan	Wheat root	0.3	0.615	0.927	Cantarero et al. (2017)
Triclosan	Wheat shoot	0.025	0.0385	0.067	Cantarero et al. (2017)

<sup>a</sup> Reference notes:

Cantarero et al. (2017) presented BAF data for wheat roots and shoots for six soil/biosolids combinations. Data shown are minimum, maximum, and computed median.

Havranek et al. (2017) presented BTF data for earthworms from sludge and estimates of BAFs for earthworms in sludge-amended soil, assuming a fixed volume of soil and uniform mixing. They note these latter values were more comparable to other literature values than the sludge transfer factors, thus they are presented here. While multiple values were provided in tabular form, the text (and table) summarized these to a single mean value, which is presented here.

Navarro et al. (2016) presented BAF data for earthworms for a control and four treatments. Navarro et al. control values were not used. If only one treatment had a value, it is shown as the median. If there were two treatment values, they are shown as the minimum and maximum. If there were three or four values, the minimum, maximum, and median (either the middle value of three or the mean of the two middle values of four) are shown.

Navarro et al. (2017) presented BAF data for spinach and tomato plant parts for a control and two treatments. Navarro et al. control values were not used. If only one treatment had a value, it is shown as the median. If there were two values, they are shown as the minimum and maximum.

## 4. Conclusions

Every two years the EPA develops biennial reviews by collecting and reviewing publicly available information on the occurrence, human health and ecological effects, and fate and transport in the environment of pollutants that have been found in U.S. biosolids. The kinds of information collected and presented in the 2016-2017 Biennial Review are needed to conduct risk assessments.

The EPA identified 32 new articles as providing relevant data for chemical pollutants that may occur in U.S. biosolids. After initial review, information was gleaned from 15 of the new articles. Review of the 15 articles identified 28 new chemicals in biosolids: seven polybrominated diphenyl ethers (PBDEs); nine parabens and metabolites; five brominated flame retardants (BFRs); three other flame retardants; two perfluoroalkyl substances (PFASs); and two triclosan transformation products. These articles also identified new data for 31 chemicals previously identified in biosolids. Concentration data in biosolids were found for all 28 new chemicals and for two chemicals identified in a previous biennial review (diclofenac and tonalide). Human health toxicity values were found for three of the new chemicals (benzoic acid; 2,4-dichlorophenol; and hexabromobenzene) and two previously identified chemicals, perfluorooctanoate (PFOA) and perfluorooctanesulfonate (PFOS). ECOTOXicology knowledgebase (ECOTOX; U.S. EPA, 2018d) records were found for 17 newly identified chemicals and 26 previously identified chemicals. Physical-chemical properties were identified for 22 new chemicals and 20 chemicals previously identified in biosolids; and bioconcentration or bioaccumulation factors were identified for 23 new chemicals (11 in terrestrial systems and 13 in aquatic; there was one chemical with both) and 24 previously identified chemicals.

In addition, although no new microbial pollutants in biosolids were identified, potentially useful data on seven previously identified microbial pollutants were found: *Cryptosporidium* spp., *Giardia* spp., antibiotic resistance genes (ARGs)/antibiotic resistant bacteria (AR bacteria), *Salmonella*, *Escherichia coli*, human norovirus, and human adenovirus.

Addressing the uncertainty around potential risk for pollutants identified in biosolids is the top priority for the EPA's Biosolids Program. The EPA continues to make significant progress in building capacity to assess pollutants by developing the necessary tools and data. The EPA expects to begin risk screening of pollutants found in biosolids once public review of its screening tool has been completed.

## 5. Additional Information

For additional information about the EPA's Biosolids Program, please visit the website at: <http://epa.gov/biosolids>.

## 6. References

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## Appendix A. Pollutants Identified in Biosolids

Table A-1. Chemical Pollutants Identified in Biosolids

Pollutant	CAS No.	Category	TNSSS analyte	When Identified	Last BR Mention <sup>a</sup>
Acetaminophen	103-90-2	Other drugs	X	2005BR	2005
Albuterol/Salbutamol	18559-94-9	Other drugs	X	2005BR	2013
Alprazolam	28981-97-7	Other drugs		2013BR	2013
Aluminum	7429-90-5	Metals	X	2005BR	2007
Amitriptyline	549-18-8	Other drugs		2013BR	2013
Amlodipine	88150-42-9	Other drugs		2013BR	2013
Amphetamine	300-62-9	Other drugs		2007BR	2007
Androstenedione	63-05-8	Hormones	X	2009 TNSSS	2009
Androsterone	53-41-8	Hormones	X	2009 TNSSS	2009
Anhydrochlortetracycline	13803-65-1	Antibiotics	X	2009 TNSSS	2009
Anhydrotetracycline	4496-85-9	Antibiotics	X	2009 TNSSS	2009
Antimony	7440-36-0	Metals	X	2005BR	2005
Aspirin	50-78-2	Other drugs		2005BR	2005
Atenolol	29122-68-7	Other drugs		2013BR	2013
Atorvastatin	134523-00-5	Other drugs		2013BR	2013
Azithromycin	83905-01-5	Antibiotics	X	2007BR	2011
Barium	7440-39-3	Metals	X	2009 TNSSS	2009
BDE-17 (2,2',4-TrBDE)	147217-75-2	PBDEs		2017BR	2017
BDE-28 (2,4,4'-TrBDE)	6430-90-6	PBDEs	X	2009 TNSSS	2017
BDE-47 (2,2',4,4'-TeBDE)	5436-43-1	PBDEs	X	2009 TNSSS	2017
BDE-66 (2,3',4,4'-TeBDE)	84303-45-7	PBDEs	X	2009 TNSSS	2017
BDE-85 (2,2',3,4,4'-PeBDE)	32534-81-9	PBDEs	X	2009BR	2009
BDE-99 (2,2',4,4',5-PeBDE)	60348-60-9	PBDEs	X	2009 TNSSS	2017
BDE-100 (2,2',4,4',6-PeBDE)	97038-97-6	PBDEs	X	2009 TNSSS	2017
BDE-138 (2,2',3,4,4',5'-HxBDE)	67888-98-6	PBDEs	X	2009 TNSSS	2009
BDE-153 (2,2',4,4',5,5'-HxBDE)	68631-49-2	PBDEs	X	2009 TNSSS	2017
BDE-154 (2,2',4,4',5,6'-HxBDE)	207122-15-4	PBDEs	X	2009 TNSSS	2017
BDE-183 (2,2',3,4,4',5',6-HpBDE)	207122-16-5	PBDEs	X	2009 TNSSS	2017
BDE-184 (HpBDE, 2,2',3,4,4',6,6'-)	117948-63-7	PBDEs		2017BR	2017
BDE-191 (HpBDE, 2,3,3',4,4',5',6-)	446255-30-7	PBDEs		2017BR	2017
BDE-196 (OcBDE, 2,2',3,3',4,4',5,6'-)	446255-39-6	PBDEs		2017BR	2017
BDE-197 (OcBDE, 2,2',3,3',4,4',6,6'-)	117964-21-3	PBDEs		2017BR	2017
BDE-206 (NoBDE, 2,2',3,3',4,4',5,5',6-)	63936-56-1	PBDEs		2017BR	2017
BDE-207 (NoBDE, 2,2',3,3',4,4',5,6,6'-)	437701-79-6	PBDEs		2017BR	2017
BDE-209 (2,2',3,3',4,4',5,5',6,6'-DeBDE)	1163-19-5	PBDEs	X	2009BR	2017
Benz(a)anthracene	56-55-3	PAHs		2005BR	2005

Pollutant	CAS No.	Category	TNSSS analyte	When Identified	Last BR Mention <sup>a</sup>
Benzenesulfonic acid, 2,2'-(1,2-ethenediyl)bis[5-amino]	42615-29-2	Other drugs		2005BR	2005
Benzo(a)pyrene	50-32-8	PAHs	X	2005BR	2005
Benzo(b)fluoranthene	205-99-2	PAHs		2005BR	2005
Benzo(k)fluoranthene	207-08-9	PAHs		2005BR	2005
Benzoic acid	65-85-0	Pharmaceuticals		2017BR	2017
Benzoylcegonine	519-09-5	Other drugs		2013BR	2013
Benzotropine	86-13-5	Other drugs		2013BR	2013
Benzyl-4-chlorophenol, 2-	120-32-1	Antimicrobial		2015BR	2015
Benzyl paraben	94-18-8	Preservatives		2017BR	2017
Beryllium	7440-41-7	Metals	X	2009 TNSSS	2009
Bezafibrate	41859-67-0	Other drugs		2005BR	2005
Bis(2,4,6-tribromophenoxy)ethane, 1,2-	37853-59-1	Flame retardant		2017BR	2017
Bis (2-ethylhexyl) phthalate	117-81-7	SVOCs	X	2005BR	2005
Bis (5-chloro-2hydroxyphenyl)methane	97-23-4	Antimicrobial		2015BR	2015
Bisphenol A	80-05-7	Plastics		2007BR	2015
Boron	7440-42-8	Metals	X	2005BR	2005
Butylated hydroxy toluene	128-37-0	Other drugs		2005BR	2005
Butylparaben	94-26-8	Preservatives		2017BR	2017
Caffeine	58-08-2	Other drugs	X	2005BR	2011
Calcium	7440-70-2	Inorganics	X	2007BR	2007
Campesterol	474-62-4	Steroids	X	2009 TNSSS	2009
Carbadox	6804-07-5	Antibiotics	X	2005BR	2005
Carbamazepine	298-46-4	Other drugs	X	2005BR	2013
Carbon tetrachloride	56-23-5	Organics		2005BR	2005
Cefotaxime	63527-52-6	Antibiotics	X	2009 TNSSS	2009
Cerium	7440-45-1	Metals		2005BR	2005
Chloro-4-phenylphenol, 2-	92-04-6	Antimicrobial		2015BR	2015
Chloroaniline, 4-	106-47-8	SVOCs	X	2009 TNSSS	2009
Chloroform	67-66-3	Organics		2005BR	2005
Chloronaphthalene, 2-	91-58-7	Organics		2005BR	2005
Chlortetracycline	57-62-5	Antibiotics	X	2009BR	2009
Cholestanol	80-97-7	Steroids	X	2009 TNSSS	2009
Cholesterol	57-88-5	Steroids	X	2005BR	2007
Chrysene	218-01-9	PAHs		2005BR	2005
Cimetidine	51481-61-9	Other drugs	X	2005BR	2005
Ciprofloxacin	85721-33-1	Antibiotics	X	2005BR	2011
Clarithromycin	81103-11-9	Antibiotics	X	2007BR	2009
Clinafloxacin	105956-97-6	Antibiotics	X	2009 TNSSS	2009
Clindamycin	18323-44-9	Antibiotics		2011BR	2011
Clofibric acid	882-09-7	Other drugs		2005BR	2005
Clotrimazole	23593-75-1	Antibiotics		2011BR	2011

Pollutant	CAS No.	Category	TNSSS analyte	When Identified	Last BR Mention <sup>a</sup>
Cloxacillin	61-72-3	Antibiotics	X	2009 TNSSS	2009
Cobalt	7440-48-4	Metals	X	2005BR	2007
Cocaine	50-36-2	Other drugs		2013BR	2013
Codeine	76-57-3	Other drugs	X	2005BR	2005
Coprostanol (3-beta)	360-68-9	Steroids	X	2007BR	2007
Cotinine	486-56-6	Other drugs	X	2005BR	2005
Cresol, p- (4-methylphenol)	106-44-5	Preservative		2005BR	2007
Cyanide	57-12-5	Organics		2005BR	2005
Cyclophosphamide	50-18-0	Other drugs		2005BR	2005
Decabromodiphenyl ethane	84852-53-9	PBDEs		2017BR	2017
Decamethylcyclopentasiloxane (D5)	541-02-6	Emollients		2015BR	2015
Dechlorane 602	31107-44-5	Flame retardant		2017BR	2017
Dechlorane 603	13560-92-4	Flame retardant		2017BR	2017
Dechlorane Plus	13560-89-9	Flame retardant		2017BR	2017
DEET (N,N-diethyltoluamide)	134-62-3	Pesticides		2005BR	2013
Dehydronifedipine	67035-22-7	Other drugs	X	2009 TNSSS	2009
Demeclocycline	127-33-3	Antibiotics	X	2009 TNSSS	2009
Desmethyldiltiazem	130606-60-9	Other drugs		2013BR	2013
Desmosterol	313-04-2	Steroids	X	2009 TNSSS	2009
Diazepam	439-14-5	Other drugs		2005BR	2005
Dichlorobenzene, 1,3-	541-73-1	Pesticides		2005BR	2005
Dichlorobenzene, 1,4-	106-46-7	Pesticides		2005BR	2005
Dichlorocarbanilide	1219-99-4	Antibiotics		2011BR	2011
Dichlorophenol, 2,4-	120-83-2	Other organics		2017BR	2017
Diclofenac	15307-86-5	Pharmaceuticals		2011BR	2017
Diclofenac sodium	15307-79-6	Other drugs		2005BR	2005
Digoxigenin	1672-46-4	Other drugs	X	2009 TNSSS	2009
Digoxin	20830-75-5	Other drugs	X	2005BR	2005
Dihydroequilin, 17 $\alpha$ -	651-55-8	Hormones	X	2009 TNSSS	2009
Dihydroxybenzoic acid, 3,4-	99-50-3	Metabolites		2017BR	2017
Diltiazem	42399-41-7	Other drugs	X	2005BR	2011
Dimethoate	60-51-5	Pesticides		2005BR	2005
Dimethyl phthalate	131-11-3	Organics		2005BR	2005
Dimethyl-3,5,-dinitro-4-tert-butylacetophenone, 2,6-	81-14-1	Odorants		2005BR	2005
Dimethylaminophenazone	58-15-1	Other drugs		2005BR	2005
Dimethylxanthine, 1,7-	611-59-6	Other drugs	X	2005BR	2005
Di-n-butyl phthalate (Butoxyphosphate ethanol, 2-)	84-74-2	Plasticizers		2005BR	2005
Di-n-octyl phthalate	117-84-0	Organics		2005BR	2005
Diphenhydramine	58-73-1	Other drugs	X	2007BR	2013

Pollutant	CAS No.	Category	TNSSS analyte	When Identified	Last BR Mention <sup>a</sup>
Di-tert-butylphenol, 2,6-	128-39-2	Other drugs		2005BR	2005
Doxycycline	564-25-0	Antibiotics	X	2005BR	2009
Endosulfan, $\alpha$	959-98-8	Pesticides		2005BR	2005
Endosulfan, $\beta$	33213-65-9	Pesticides		2005BR	2005
Enrofloxacin	93106-60-6	Antibiotics	X	2009 TNSSS	2009
Epianhydrochlortetracycline, 4-	158018-53-2	Antibiotics	X	2009 TNSSS	2009
Epianhydrotetracycline, 4-	4465-65-0	Antibiotics	X	2009 TNSSS	2009
Epichlortetracycline, 4-	14297-93-9	Antibiotics	X	2009 TNSSS	2009
Epicoprostanol	516-92-7	Steroids	X	2009 TNSSS	2009
Epioxytetracycline, 4-	14206-58-7	Antibiotics	X	2009 TNSSS	2009
Epitetracycline, 4-	23313-80-6	Antibiotics	X	2009 TNSSS	2009
Equilenin	517-09-9	Hormones	X	2009 TNSSS	2009
Equilin	474-86-2	Hormones	X	2005BR	2005
Ergosterol	57-87-4	Steroids	X	2009 TNSSS	2009
Erythromycin	114-07-8	Antibiotics	X	2005BR	2009
Estradiol, 17 $\alpha$ -	57-91-0	Hormones	X	2005BR	2005
Estradiol, 17 $\beta$ -	50-28-2	Hormones	X	2005BR	2009
Estradiol-3-benzoate, $\beta$ -	50-50-0	Hormones	X	2009 TNSSS	2009
Estriol (estradiol)	50-27-1	Hormones	X	2005BR	2005
Estrone	53-16-7	Hormones	X	2005BR	2011
Ethanol, 2-butoxy-phosphate	78-51-3	Organics		2005BR	2005
Ethylbenzene	100-41-4	Organics		2005BR	2005
Ethyl paraben	120-47-8	Preservatives		2017BR	2017
Ethynyl estradiol, 17 $\alpha$ -	57-63-6	Hormones	X	2005BR	2017
Fenofibric acid	26129-32-8	Other drugs		2005BR	2005
Fenthion	55-38-9	Pesticides		2005BR	2005
Fipronil	120068-37-3	Antibiotics		2011BR	2011
Floxacillin	5250-39-5	Antibiotics		2005BR	2005
Flumequine	42835-25-6	Antibiotics	X	2009 TNSSS	2009
Fluoranthene	206-44-0	PAHs	X	2009 TNSSS	2009
Fluoride	16984-48-8	Inorganics	X	2005BR	2005
Fluoxetine	54910-89-3	Other drugs	X	2005BR	2007
Furosemide	54-31-9	Other drugs		2013BR	2013
Galaxolide	1222-05-5	Fragrance		2005BR	2017
Gemfibrozil	25812-30-0	Other drugs	X	2005BR	2011
Glyburide	10238-21-8	Other drugs		2013BR	2013
Heptabromodibenzofuran, 1,2,3,4,6,7,8-	107555-95-3	PBDF		2015BR	2015
Heptabromodibenzofuran, 1,2,3,4,7,8,9-	161880-51-9	PBDF		2015BR	2015
Heptabromodibenzo-p-dioxin, 1,2,3,4,6,7,8-	103456-43-5	PBDD		2015BR	2015
Heptachlor epoxide	1024-57-3	Pesticides		2005BR	2005
Hexabromobenzene	87-82-1	Other organics		2017BR	2017
Hexabromobiphenyl, 2,2',4,4',5,5'-	59080-40-9	PBBs		2005BR	2005

Pollutant	CAS No.	Category	TNSSS analyte	When Identified	Last BR Mention <sup>a</sup>
Hexabromocyclododecane	3194-55-6	Brominated flame retardants		2017BR	2017
Hexabromodibenzofuran, 1,2,3,4,7,8-	70648-26-9	PBDF		2015BR	2015
Hexabromodibenzofuran, 1,2,3,6,7,8-	107555-94-2	PBDF		2015BR	2015
Hexabromodibenzofuran, 1,2,3,7,8,9-	161880-49-5	PBDF		2015BR	2015
Hexabromodibenzofuran, 2,3,4,6,7,8-	60851-34-5	PBDF		2015BR	2015
Hexabromodibenzo-p-dioxin, 1,2,3,4,7,8-	110999-44-5	PBDD		2015BR	2015
Hexabromodibenzo-p-dioxin, 1,2,3,6,7,8-	110999-45-6	PBDD		2015BR	2015
Hexabromodibenzo-p-dioxin, 1,2,3,7,8,9-	110999-46-7	PBDD		2015BR	2015
Hydrocodone	125-29-1	Other drugs		2013BR	2013
Hydroxyamitriptyline, 10-	1246833-15-7	Other drugs		2013BR	2013
Hydroxy benzoic acid, 4-	99-96-7	Preservatives		2017BR	2017
Ibuprofen	15687-27-1	Other drugs	X	2005BR	2005
Indole	120-72-9	Fragrance		2007BR	2007
Indometacine	53-86-1	Other drugs		2005BR	2005
Iron	7439-89-6	Metals	X	2005BR	2005
Isochlortetracycline	514-53-4	Antibiotics	X	2009 TNSSS	2009
Ketoprofen	22071-15-4	Other drugs		2005BR	2005
Limonene, d-	5989-27-5	Fragrance		2007BR	2007
Lincomycin	154-21-2	Antibiotics	X	2009BR	2009
Lomefloxacin	98079-51-7	Antibiotics	X	2009 TNSSS	2009
Magnesium	7439-95-4	Metals	X	2007BR	2007
Manganese	7439-96-5	Metals	X	2009 TNSSS	2009
Mefenamic acid	61-68-7	Other drugs		2005BR	2005
Mesalazine	89-57-6	Other drugs		2005BR	2005
Mestranol	72-33-3	Other drugs		2005BR	2005
Metformin	657-24-9	Other drugs	X	2009 TNSSS	2009
Methamphetamine	537-46-2	Other drugs		2007BR	2009
Methylenedioxymethamphetamine, 3,4-	42542-10-9	Other drugs		2009BR	2009
Methylnaphthalene, 2-	91-57-6	PAHs	X	2005BR	2005
Methylparaben	99-76-3	Preservatives		2017BR	2017
Methyl protocatechuate	2150-43-8	Metabolites		2017BR	2017
Methyl triclosan	4640-01-1	Metabolites		2017BR	2017
Metoprolol	37350-58-6	Other drugs		2005BR	2013
Miconazole	22916-47-8	Other drugs	X	2009 TNSSS	2009
Minocycline	10118-90-8	Antibiotics	X	2009 TNSSS	2009
Molybdenum	7439-98-7	Metals	X	2009 TNSSS	2009
Monuron	150-68-5	Pesticides		2005BR	2005

Pollutant	CAS No.	Category	TNSSS analyte	When Identified	Last BR Mention <sup>a</sup>
Nadolol	42200-33-9	Other drugs		2005BR	2005
Naproxen	22204-53-1	Other drugs	X	2005BR	2015
Napthalene	91-20-3	PAHs		2005BR	2005
Nitrate	14797-55-8	Inorganics	X	2009 TNSSS	2009
Nitrite	14797-65-0	Inorganics	X	2009 TNSSS	2009
Nitrofen	1836-75-5	Pesticides		2005BR	2005
Nitrogen	7727-37-9	Inorganics		2007BR	2007
Nitrogen, organic	14798-03-9	Organics		2007BR	2007
Nitrophenol, p-	100-02-7	Organics		2005BR	2005
N-nitrosodibutylamine (NDBA)	924-16-3	Nitrosamines		2015BR	2015
N-nitrosodiethylamine (NDEA)	55-18-5	Nitrosamines		2015BR	2015
N-nitrosodimethylamine (NDMA)	62-75-9	Nitrosamines		2015BR	2015
N-nitroso-di-n-propylamine (NDPA)	621-64-7	Nitrosamines		2015BR	2015
N-nitrosodiphenylamine (NDPhA)	86-30-6	Nitrosamines		2015BR	2015
N-nitrosopiperidine (NPIP)	100-75-4	Nitrosamines		2015BR	2015
N-nitrosopyrrolidine (NPYR)	930-55-2	Nitrosamines		2015BR	2015
Nonylphenol	25154-52-3	Surfactants		2005BR	2017
Nonylphenol (branched), 4-	84852-15-3	Surfactants		2005BR	2005
Nonylphenol monoethoxylate	27986-36-3	Surfactants		2007BR	2017
Nonylphenol, 4-	104-40-5	Surfactants		2005BR	2007
Nonylphenol diethoxylate	30-53-3	Surfactants		2007BR	2017
Norethindrone (norethisterone)	68-22-4	Hormones	X	2005BR	2005
Norfloxacin	70458-96-7	Antibiotics	X	2005BR	2011
Norfluoxetine	57226-68-3	Antibiotics		2011BR	2013
Norgestimate	35189-28-7	Other drugs	X	2009 TNSSS	2009
Norgestrel (levonorgestrel)	797-63-7	Hormones	X	2005BR	2005
Norverapamil	67812-42-4	Other drugs		2013BR	2013
Octabromodibenzofuran, 1,2,3,4,6,7,8,9-	103582-29-2	PBDF		2015BR	2015
Octabromodibenzo-p-dioxin, 1,2,3,4,6,7,8,9-	2170-45-8	PBDD		2015BR	2015
Octylphenol	67554-50-1	Organics		2005BR	2005
Octylphenol, 4-	1806-26-4	Organics		2007BR	2007
Ofloxacin	82419-36-1	Antibiotics	X	2009 TNSSS	2009
Ormetoprim	6981-18-6	Antibiotics	X	2009 TNSSS	2009
Oxacillin	66-79-5	Antibiotics	X	2009 TNSSS	2009
Oxolinic acid	14698-29-4	Antibiotics	X	2009 TNSSS	2009
Oxycodone	76-42-6	Other drugs		2013BR	2013
Oxytetracycline	79-57-2	Antibiotics	X	2005BR	2009
Paroxetine	61869-08-7	Other drugs		2013BR	2013
Penicillin G	61-33-6	Antibiotics	X	2009 TNSSS	2009
Penicillin V (phenoxymethylpenicillin)	87-08-1	Antibiotics	X	2005BR	2005
Pentabromodibenzofuran, 1,2,3,7,8-	107555-93-1	PBDF		2015BR	2015

Pollutant	CAS No.	Category	TNSSS analyte	When Identified	Last BR Mention <sup>a</sup>
Pentabromodibenzofuran, 2,3,4,7,8-	131166-92-2	PBDF		2015BR	2015
Pentabromodibenzo-p-dioxin, 1,2,3,7,8-	109333-34-8	PBDD		2015BR	2015
Pentabromoethylbenzene	85-22-3	Other organics		2017BR	2017
Pentachloronitrobenzene	82-68-8	Pesticides		2005BR	2005
Perfluorobutanesulfonate (PFBS)	45187-15-3	PFASs		2013BR	2017
Perfluorobutanoate (PFBA)	375-22-4	PFASs		2013BR	2017
Perfluorodecanoate (PFDA)	335-76-2	PFASs		2013BR	2017
Perfluorododecanoate (PFDoDA)	307-55-1	PFASs		2013BR	2017
Perfluoroheptanoate (PFHpA)	375-85-9	PFASs		2013BR	2017
Perfluorohexanesulfonate (PFHxS)	108427-53-8	PFASs		2013BR	2017
Perfluorohexanoate (PFHxA)	307-24-4	PFASs		2013BR	2017
Perfluorononanoate (PFNA)	375-95-1	PFASs		2013BR	2017
Perfluorooctane sulfonamide (PFOSA)	754-91-6	PFASs		2013BR	2017
Perfluorooctanesulfonate (PFOS)	2795-39-3	PFASs		2013BR	2017
Perfluorooctanoate (PFOA)	335-67-1	PFASs		2013BR	2017
Perfluoropentanoate (PFPeA)	2706-90-3	PFASs		2013BR	2017
Perfluorotetradecanoate (PFTeDA)	376-06-7	PFASs		2017BR	2017
Perfluorotridecanoate (PFTrDA)	72629-94-8	PFASs		2017BR	2017
Perfluoroundecanoate (PFUnDA)	2058-94-8	PFASs		2013BR	2017
Phenanthrene	85-01-8	PAHs		2007BR	2007
Phenazone	60-80-0	Other drugs		2005BR	2005
Phosphate (total)	14265-44-2	Inorganics		2005BR	2005
Phosphorus	7723-14-0	Inorganics	X	2007BR	2007
Polyethylene glycol	25322-68-3	Organics		2005BR	2005
Potassium	7440-09-7	Metals		2007BR	2007
Progesterone	57-83-0	Hormones	X	2005BR	2009
Promethazine	60-87-7	Other drugs		2013BR	2013
Propoxyphene	469-62-5	Other drugs		2013BR	2013
Propranolol	525-66-6	Other drugs		2005BR	2013
Propyl paraben	94-13-3	Preservatives		2017BR	2017
Pyrene	129-00-0	PAHs	X	2009 TNSSS	2009
Quinine sulfate	7778-93-0	Other drugs		2005BR	2005
Ranitidine	66357-35-5	Other drugs	X	2005BR	2005
Roxithromycin	80214-83-1	Antibiotics	X	2007BR	2007
Rubidium	7440-17-7	Metals		2005BR	2005
Salicylic acid	69-72-7	Other drugs		2005BR	2005
Sarafloxacin	98105-99-8	Antibiotics	X	2009 TNSSS	2009
Sertraline	79617-96-2	Other drugs		2013BR	2013
Silver	7440-22-4	Metals	X	2009 TNSSS	2009
Sitosterol, $\beta$ -	83-46-5	Steroids	X	2007BR	2007
Skatole	83-34-1	NA		2007BR	2007
Sodium	7440-23-5	Metals	X	2009 TNSSS	2009

Pollutant	CAS No.	Category	TNSSS analyte	When Identified	Last BR Mention <sup>a</sup>
Sodium valproate	1069-66-5	Other drugs		2005BR	2005
Stigmastanol, $\beta$ -	19466-47-8	Steroids	X	2007BR	2007
Stigmasterol	83-45-4	Steroids	X	2009 TNSSS	2009
Styrene	100-42-5	Organics		2005BR	2005
Sulfachloropyridazine	80-32-0	Antibiotics	X	2009 TNSSS	2009
Sulfadiazine	68-35-9	Antibiotics	X	2009 TNSSS	2009
Sulfadimethoxine	122-11-2	Antibiotics	X	2009BR	2009
Sulfamerazine	127-79-7	Antibiotics	X	2005BR	2005
Sulfamethazine	57-68-1	Antibiotics	X	2005BR	2009
Sulfamethizole	144-82-1	Antibiotics	X	2009 TNSSS	2009
Sulfamethoxazole	723-46-6	Antibiotics	X	2009 TNSSS	2009
Sulfanilamide	63-74-1	Antibiotics	X	2009 TNSSS	2009
Sulfasalazine	599-79-1	Other drugs		2005BR	2005
Sulfathiazole	72-14-0	Antibiotics	X	2009 TNSSS	2009
tert-Butyl-4-hydroxy anisole, 3-	25013-16-5	Other drugs		2005BR	2005
Testosterone	58-22-0	Hormones	X	2009BR	2009
Tetrabromobisphenol A	79-94-7	Organics		2005BR	2005
Tetrabromodibenzofuran, 2,3,7,8-	67733-57-7	PBDF		2015BR	2015
Tetrabromodibenzo-p-dioxin, 2,3,7,8-	50585-41-6	PBDD		2015BR	2015
Tetrachloroethylene	127-18-4	Solvents		2005BR	2005
Tetracycline	60-54-8	Antibiotics	X	2009BR	2009
Thallium	7440-28-0	Metals	X	2005BR	2005
Thiabendazole	148-79-8	Other drugs	X	2009 TNSSS	2009
Tin	7440-31-5	Metals	X	2005BR	2005
Titanium	7440-32-6	Metals	X	2009 TNSSS	2009
Toluene	108-88-3	Solvents		2005BR	2005
Tonalide (AHTN)	21145-77-7	Fragrance		2007BR	2017
Triamterene	396-01-0	Other drugs		2013BR	2013
Trichlorobenzene, 1,3,5-	108-70-3	Organics		2005BR	2005
Trichlorofon	52-68-6	Pesticides		2005BR	2005
Trichlorophenol, 2,4,5-	95-95-4	Antimicrobial		2015BR	2015
Triclocarban	101-20-2	Antibiotics	X	2007BR	2017
Triclosan	3380-34-5	Antibiotics	X	2005BR	2017
Trimethoprim	738-70-5	Antibiotics	X	2005BR	2009
Triphenyl phosphate	115-86-6	Pesticides		2005BR	2005
Tris(2-chloroethyl) phosphate	115-96-8	Organics		2005BR	2005
Tylosin	1401-69-0	Antibiotics	X	2005BR	2007
Valsartan	137862-53-4	Other drugs		2013BR	2013
Vanadium	7440-62-2	Metals	X	2005BR	2005
Verapamil	52-53-9	Other drugs		2013BR	2013
Virginiamycin	11006-76-1	Antibiotics	X	2005BR	2009
Warfarin	81-81-2	Other drugs	X	2009 TNSSS	2009
Xylene, m-	108-38-3	Solvents		2005BR	2005

Pollutant	CAS No.	Category	TNSSS analyte	When Identified	Last BR Mention <sup>a</sup>
Xylene, musk	81-15-2	Odorants		2005BR	2005
Xylene, o-	95-47-6	Solvents		2005BR	2005
Xylene, p	106-42-3	Solvents		2005BR	2005
Yttrium	7440-65-5	Metals	X	2005BR	2005

Table A-2. Microbial Pollutants Identified in Biosolids

Pollutant	Category	When Identified	Last BR Mention <sup>a</sup>
Aerobic endospores	Bacteria	2013BR	2013
<i>Aeromonas</i> spp.	Bacteria	2009BR	2009
Antibiotic-resistant bacteria (ARB) or Antibiotic-resistant genes (ARG)	Bacteria	2013BR	2017
<i>Clostridia</i> spp.	Bacteria	2007BR	2011
Coronavirus HKU1	Virus	2013BR	2013
Cosavirus	Virus	2013BR	2013
<i>Cryptosporidium parvum</i>	Protozoan parasite	2007BR	2017
Enterovirus	Virus	2009BR	2013
<i>Escherichia coli</i> ( <i>E. coli</i> )	Bacteria	2009BR	2017
Endotoxin	Microbial toxin	2007BR	2007
<i>Giardia</i> spp.	Protozoan parasite	2009BR	2017
Human adenoviruses	Virus	2009BR	2017
Human polyomaviruses	Virus	2011BR	2011
Klassevirus	Virus	2013BR	2013
<i>Listeria</i> spp.	Bacteria	2009BR	2011
Human norovirus	Virus	2013BR	2017
<i>Salmonella</i> spp.	Bacteria	2007BR	2017

<sup>a</sup> Most recent biennial review that mentions this pollutant. That does not necessarily indicate new data were found.

Table A-3. EPA National Sewage Sludge Surveys Pollutants Identified in Biosolids

Pollutant	CAS No.	Category	When Identified
3,3',4,4'-TeCB (PCB 77)	32598-13-3	PCB	2001
3,4,4',5'-TeCB (PCB 81)	70362-50-4	PCB	2001
2,3,3',4,4'-PeCB (PCB 105)	32598-14-4	PCB	2001
2,3,4,4',5'-PeCB (PCB 114)	74472-37-0	PCB	2001
2,3',4,4',5'-PeCB (PCB 118)	31508-00-6	PCB	2001
2',3,4,4',5'-PeCB (PCB 123)	65510-44-3	PCB	2001
3,3',4,4',5'-PeCB (PCB 126)	57465-28-8	PCB	2001
2,3,3',4,4',5'-HxCB (PCB 156)	38380-08-4	PCB	2001
2,3,3',4,4',5',5'-HxCB (PCB 157)	69782-90-7	PCB	2001
2,3',4,4',5,5'-HxCB (PCB 167)	52663-72-6	PCB	2001
3,3',4,4',5,5'-HxCB (PCB 169)	322774-16-6	PCB	2001

Pollutant	CAS No.	Category	When Identified
2,3,3',4,4',5,5'-HpCB (PCB 189)	39635-31-9	PCB	2001
Octachlorodibenzo-P-Dioxin	3268879	Dioxins/Furans	1989
Octachlorodibenzofuran	39001020	Dioxins/Furans	1989
Total Heptachlorodibenzo-P-Dioxin	37871004	Dioxins/Furans	1989
Total Heptachlorodibenzofurans	38998753	Dioxins/Furans	1989
Total Hexachlorodibenzo-P-Dioxins	344654608	Dioxins/Furans	1989
Total Hexachlorodibenzofurans	1_201	Dioxins/Furans	1989
Total Pentachlorodibenzo-P-Dioxins	36088229	Dioxins/Furans	1989
Total Pentachlorodibenzofurans	30402154	Dioxins/Furans	1989
Total Tetrachlorodibenzo-P-Dioxins	41903575	Dioxins/Furans	1989
Total Tetrachlorodibenzofurans	55722275	Dioxins/Furans	1989
1,2,3,4,6,7,8-Heptachlorodibenzo-P-Dioxin	35822469	Dioxins/Furans	1989
1,2,3,4,7,8-Hexachlorodibenzo-P-Dioxin	39227286	Dioxins/Furans	1989
1,2,3,4,7,8-Hexachlorodibenzofuran	70648269	Dioxins/Furans	1989
1,2,3,4,7,8,9-Heptachlorodibenzofuran	55673897	Dioxins/Furans	1989
1,2,3,6,7,8-Hexachlorodibenzo-P-Dioxin	57653857	Dioxins/Furans	1989
1,2,3,6,7,8-Hexachlorodibenzofuran	57117449	Dioxins/Furans	1989
1,2,3,7,8-Pentachlorodibenzofuran	5711746	Dioxins/Furans	1989
1,2,3,7,8,9-Hexachlorodibenzo-P-Dioxin	19408743	Dioxins/Furans	1989
2,3,4,6,7,8-Hexachlorodibenzofuran	60851345	Dioxins/Furans	1989
2,3,4,7,8-Pentachlorodibenzofuran	57117314	Dioxins/Furans	1989
2,3,7,8-Tetrachlorodibenzo-P-Dioxin	1746016	Dioxins/Furans	1989
2,3,7,8-Tetrachlorodibenzofuran	51207319	Dioxins/Furans	1989
Acetic Acid	94757	Pesticide	1989
Hexanoic Acid	142621	Semivolatile Organic	1989
Methylene Chloride	75092	Volatile Organic	1989
2-Propanone	67641	Volatile Organic	1989

## Appendix B. Reference Abstracts: Chemicals

Armstrong, D. L., et al. (2017). "Influence of thermal hydrolysis-anaerobic digestion treatment of wastewater solids on concentrations of triclosan, triclocarban, and their transformation products in biosolids." *Chemosphere* 171: 609-616.

The growing concern worldwide regarding the presence of emerging contaminants in biosolids calls for a better understanding of how different treatment technologies at water resource recovery facilities (WRRFs) can influence concentrations prior to biosolids land application. This study focuses on the influence of solids treatment via the Cambi Thermal Hydrolysis Process in conjunction with anaerobic digestion (TH-AD) on concentrations of triclosan (TCS), triclocarban (TCC), and their transformation products in biosolids and sludges. Concentrations of the target analytes in biosolids from the TH-AD process (Class A), sludges from the individual TH-AD treatment steps, and limed biosolids (Class B) from the same WRRF were compared. TCC concentrations were significantly lower in Class A biosolids than those in the Class B product - a removal that occurred during thermal hydrolysis. Concentrations of TCS, methyl triclosan, and 2,4-dichlorophenol, conversely, increased during anaerobic digestion, leading to significantly higher concentrations of these compounds in Class A biosolids when compared to Class B biosolids. Implementation of the TH-AD process had mixed effect on contaminant concentrations.

Belhaj, D., et al. (2016). "Estrogenic compounds in Tunisian urban sewage treatment plant: occurrence, removal and ecotoxicological impact of sewage discharge and sludge disposal." *Ecotoxicology* 25(10): 1849-1857.

The occurrence, fate and ecotoxicological assessment of selected estrogenic compounds were investigated at Tunisian urban sewage treatment plant. The influents, effluents, as well as primary, secondary and dehydrated sludge, were sampled and analyzed for the target estrogens to evaluate their fate. All target compounds were detected in both sewage and sludge with mean concentrations from 0.062 to 0.993  $\mu\text{g L}^{-1}$  and from 11.8 to 792.9  $\mu\text{g kg}^{-1}$  dry weight, respectively. A wide range of removal efficiencies during the treatment processes were observed, from 6.3 % for estrone to 76.8 % for estriol. Ecotoxicological risk assessment revealed that the highest ecotoxicological risk in sewage effluent and dehydrated sludge was due to 17 $\beta$ -estradiol with a risk quotient (RQ) of 4.6 and 181.9, respectively, and 17 $\alpha$ -ethinylestradiol with RQ of 9.8 and 14.85, respectively. Ecotoxicological risk after sewage discharge and sludge disposal was limited to the presence of 17 $\beta$ -estradiol in dehydrated-sludge amended soil with RQ of 1.38. Further control of estrogenic hormones in sewage effluent and sludge is essential before their discharge and application in order to prevent their introduction into the natural environment.

Cantarero, R., et al. (2017). "Effects of applying biosolids to soils on the adsorption and bioavailability of 17 $\alpha$ -ethinylestradiol and triclosan in wheat plants." *Environmental Science & Pollution Research* 24(14): 12847-12859.

Biosolids contain inorganic and organic contaminants, including pharmaceutical and personal care products (PPCPs) that have accounted for a series of emerging contaminants, such as triclosan (TCS) and the hormone 17 $\alpha$ -ethinylestradiol (EE2). The general aim of this study was to evaluate the effects of biosolid application on EE2 and TCS adsorption and bioavailability in soils through testing with wheat plants. For the bioavailability study, sand and two soils, Lampa and Lo Prado, were used. The sand and soils were treated using two biosolid application rates (0 and 90 mg ha), and the EE2 and TCS concentrations in the biosolids were determined as  $0.54 \pm 0.06$  and  $8.31 \pm 0.19$  mg kg, respectively. The concentration observed in wheat plants indicated that EE2 and TCS are mainly concentrated in the roots rather than in the shoots. Furthermore, the bioavailability of the compounds in plants depends on the properties of the contaminants and the soil. Adsorption studies showed that increasing the soil organic matter content increases the adsorption of TCS and EE2 on these substrates and that both compounds follow the Freundlich adsorption model. The desorption procedure indicated that availability for both TCS and EE2 depended on the soil type because TCS and EE2 were small in the Lampa soil with and without biosolid application and TCS increased by nearly 50% in the Lo Prado soil. The Lo Prado soil had an acidic pH (5.9) and the Lampa soil had a neutral pH of 7.3, and the organic carbon content was smaller.

Chen, J., et al. (2017). "Occurrence, temporal variation, and estrogenic burden of five parabens in sewage sludge collected across the United States." *Sci Total Environ* 593-594: 368-374.

Five parabens used as preservatives in pharmaceuticals and personal care products (PPCPs) were measured in sewage sludges collected at 14 U.S. wastewater treatment plants (WWTPs) located in nine states. Detected concentration ranges (ng/g, dry weight) and frequencies were as follows: methyl paraben (15.9 to 203.0; 100%), propyl paraben (0.5 to 7.7; 100%), ethyl paraben (<0.6 to 2.6; 63%), butyl paraben (<0.4 to 4.3; 42%) and benzyl paraben (<0.4 to 3.3; 26%). The estrogenicity inherent to the sum of parabens detected in sewage sludge (ranging from 10.1 to 500.1 pg/kg 17 $\beta$ -estradiol equivalents) was insignificant when compared to the 10(6)-times higher value calculated for natural estrogens reported in the literature to occur in sewage sludge. Temporal monitoring at one WWTP provided insights into temporal and seasonal variations in paraben concentrations. This is the first report on the occurrence of five parabens in sewage sludges from across the U.S., and internationally, the first on temporal variations of paraben levels in sewage sludge. Study results will help to inform the risk assessment of sewage sludge destined for land application (biosolids).

Clarke, R., et al. (2016). "A quantitative risk ranking model to evaluate emerging organic contaminants in biosolid amended land and potential transport to drinking water." *Human and Ecological Risk Assessment* 22(4): 958-990.

A quantitative risk ranking model was developed for human exposure to emerging contaminants (EC) following treated municipal sewage sludge (biosolids) application to Irish agricultural land. The model encompasses the predicted environmental concentration (PEC) in soil, surface runoff, groundwater, and subsequent drinking water ingestion by humans. Human exposure and subsequent risk was estimated for 16 organic contaminants using a Monte Carlo simulation approach. Nonylphenols ranked the highest across three environmental compartments: concentration in soil (PEC<sub>soil</sub>), runoff (PEC<sub>runoff</sub>), and groundwater (PEC<sub>groundwater</sub>), which had mean values of 5.69mg/kg,  $1.15 \times 10^{-2}$   $\mu\text{g/l}$ , and  $2.22 \times 10^{-1}$   $\mu\text{g/l}$ , respectively. Human health risk was estimated using the LC50 (chemical intake toxicity ratio, (RR)) as a toxicity endpoint combined with PEC<sub>runoff</sub> and PEC<sub>groundwater</sub>. NP ranked highest for LC50 combined with PEC<sub>runoff</sub> and PEC<sub>groundwater</sub> (mean RR values  $1.10 \times 10^{-4}$  and  $2.40 \times 10^{-3}$ , respectively). The model highlighted triclocarban and triclosan as ECs requiring further investigation. A sensitivity analysis revealed that soil sorption coefficient and soil organic carbon were the most important parameters that affected model variance (correlation coefficient -0.89 and -0.30, respectively), highlighting the significance of contaminant and soil properties in influencing risk assessments. This model can help to prioritize emerging contaminants of concern requiring vigilance in environmental compartments.

Fu, Q., et al. (2016). "Meta-analysis of biosolid effects on persistence of triclosan and triclocarban in soil." *Environ Pollut* 210: 137-144.

Biosolids are extensively used in agriculture as fertilizers while offering a practical solution for waste disposal. Many pharmaceutical and personal care products (PPCPs), such as triclosan and triclocarban, are enriched in biosolids. Biosolid amendment changes soil physicochemical properties, which may in turn alter the persistence of PPCPs and hence the risk for secondary contamination such as plant uptake. To delineate the effect of biosolids on PPCPs persistence, triclosan and triclocarban were used as model compounds in this study and their sorption ( $K_d$ ) and persistence ( $t_{1/2}$ ) were determined in different soils before and after biosolid amendment. Biosolids consistently increased sorption of triclosan and triclocarban in soil. The  $K_d$  of triclosan increased by 3.9-21 times following amendment of a sandy loam soil with biosolids at 2-10%. The persistence of both compounds was prolonged, with  $t_{1/2}$  of triclosan increasing from 10 d in the unamended soil to 63 d after biosolid amendment at 10%. The relationship between  $t_{1/2}$  and  $K_d$  was further examined through a meta-analysis using data from this study and all relevant published studies. A significant linear relationship between  $t_{1/2}$  and  $K_d$  was observed for triclosan ( $r^2 = 0.69$ ,  $p < 0.01$ ) and triclocarban ( $r^2 = 0.38$ ,  $p < 0.05$ ) in biosolid-amended soils. On the average, when biosolid amendment increased by 1%,  $t_{1/2}$  of triclosan was prolonged by 7.5 d, while  $t_{1/2}$  of triclocarban was extended by 4.7 d. Therefore, biosolid amendment greatly enhances persistence of triclosan and triclocarban, likely due to enhanced sorption or decreased chemical bioavailability. This finding highlights the importance to consider the effect of biosolids when evaluating the environmental risks of these and other biosolid-borne PPCPs.

Gallen, C., et al. (2016). "Occurrence and distribution of brominated flame retardants and perfluoroalkyl substances in Australian landfill leachate and biosolids." *J Hazard Mater* 312: 55-64.

The levels of perfluoroalkyl substances (PFASs), polybrominated diphenyl ethers (PBDEs) and hexabromocyclododecane (HBCDDs) were studied in Australian landfill leachate and biosolids. Leachate was collected from 13 landfill sites and biosolids were collected from 16 wastewater treatment plants (WWTPs), across Australia. Perfluorohexanoate (PFHxA) (12-5700ng/L) was the most abundant investigated persistent, bioaccumulative and toxic (PBT) chemical in leachate. With one exception, mean concentrations of PFASs were higher in leachate of operating landfills compared to closed landfills. Polybrominated diphenyl ethers (PBDEs) and hexabromocyclododecane isomers (HBCDDs) were detected typically at operating landfills in comparatively lower concentrations than the PFASs. Decabromodiphenyl ether (BDE-209) (<0.4-2300ng/g) and perfluorooctanesulfonate (PFOS) (<LOD-380ng/g) were the predominant PBTs detected in biosolids. Using data provided by sites, the volume of leachate discharged to WWTPs for treatment was small (<1% total inflow), and masses of PBTs transferred reached a maximum of 16g/yr (PFHxA). A national estimate of masses of PBTs accumulated in Australian biosolids reached 167kg/yr (BDE-209), a per capita contribution of 7.2+/-7.2mg/yr. Nationally, approximately 59% of biosolids are repurposed and applied to agricultural land. To our knowledge this study presents the first published data of PFASs and HBCDDs in Australian leachate and biosolids.

Gottschall, N., et al. (2017). "Brominated flame retardants and perfluoroalkyl acids in groundwater, tile drainage, soil, and crop grain following a high application of municipal biosolids to a field." *Sci Total Environ* 574: 1345-1359.

Dewatered municipal biosolids (DMB) were applied at a rate of 22Mgdwha(-1) to an agricultural field in fall 2008. Concentrations of polybrominated diphenyl ethers (PBDEs; BDE-47, -99, -100, -153, -154, -183, -197, -207, -209), other brominated flame retardants (BFRs; HBB, PBEB, DBDPE, BTBPE) and perfluoroalkyl acids (PFAAs; PFHxS, PFOS, PFDS, PFOSA, PFHpA, PFOA, PFNA, PFDA, PUnA, PFDaA, PFTA) were monitored in tile drainage, groundwater (2m, 4m and 6m depth), soil cores (0-0.3m) pre- and post-application, DMB aggregates incorporated into the soil post-application, and in wheat (*Triticum* spp.) planted post-application. Several compounds were detected in soil and water pre-application and on a reference field plot. PBDEs, other BFRs and PFAAs were detected in tile drainage and 2m groundwater throughout the post-application study period; a few PBDEs were also detected sporadically at lower depths in groundwater. Some of these compounds had not been detected pre-application, while some exceeded reference field plot/pre-application levels (some significantly ( $p < 0.05$ ) in tile drainage); both cases indicating biosolid-based water contamination. In DMB aggregates, several PBDE congeners were found to have dissipated exponentially, with reductions >90% in many of them within 1 year post-application. Exponential dissipation of other BFRs and PFAAs in DMB aggregates were not significant. No PBDEs, other BFRs, or PFAAs were detected in wheat grain.

Havranek, I., et al. (2017). "Uptake and elimination kinetics of the biocide triclosan and the synthetic musks galaxolide and tonalide in the earthworm *Dendrobaena veneta* when exposed to sewage sludge." *Environ Toxicol Chem* 36(8): 2068-2073.

Sewage sludge is an important amendment that enriches soils with organic matter and provides plants with nutrients such as nitrogen and phosphorus. However, knowledge on the fate and effects of organic pollutants present in the sludge on soil organisms is limited. In the present study, the uptake of triclosan, galaxolide, and tonalide in the earthworm *Dendrobaena veneta* was measured 1 wk after amendment of agricultural soil with sewage sludge, while elimination kinetics were assessed over a 21-d period after transferring worms to clean soil. After 1-wk exposure, earthworms had accumulated 2.6 +/- 0.6 mug g(-1) galaxolide, 0.04 +/- 0.02 mug g(-1) tonalide, and 0.6 +/- 0.2 mug g(-1) triclosan. Both synthetic musks were efficiently excreted and below the limit of quantification after 3 and 14 d of depuration for tonalide and galaxolide, respectively. Triclosan concentrations, on the other hand, did not decrease significantly over the depuration period, which may lead to the transfer of triclosan in the food web.

Karthikraj, R., et al. (2017). "Occurrence and fate of parabens and their metabolites in five sewage treatment plants in India." *Sci Total Environ* 593-594: 592-598.

Parabens (p-hydroxy benzoic acid esters) are antimicrobial agents, used widely as preservatives in personal care products (PCPs), pharmaceuticals, foods, and beverages. PCPs that contain parabens are a major source of these chemicals in sewage treatment plants (STPs). Very few studies have demonstrated the occurrence of parabens in wastewater. In this study, the occurrence and fate of six parabens, methyl-(MeP), ethyl-(EtP), propyl-(PrP), butyl-(BuP), heptyl-(HpP) and benzyl-(BzP) parabens, and five of their metabolites (4-HB, 3,4-DHB, OH-MeP, OH-EtP and BA) were studied in five STPs in India. The concentrations of parent parabens ( summation operator6parent parabens) in five STPs ranged between 131 and 920ng/L in influent; 16 and 67ng/L in effluent; and 104 and 1090ng/g, dry weight, in sludge samples. The sum concentrations of five paraben metabolites ( summation operator5paraben metabolites) in five STPs ranged between 4110 and 34,600ng/L in influent; 2560 and 3800ng/L in effluent; and 1220 and 35,900ng/g, dry weight, in sludge samples. These values were higher than those reported for many industrialized countries. We calculated the mass loadings, removal efficiencies, and environmental emissions of parabens based on the measured concentrations in influents, effluents, and sludge. The mean removal efficiencies of parent parabens and their metabolites ranged from 80% to 100% and 28% to 76%, respectively.

Milinic, J., et al. (2016). "Sorption of perfluoroalkyl substances in sewage sludge." *Environ Sci Pollut Res Int* 23(9): 8339-8348.

The sorption behaviour of three perfluoroalkyl substances (PFASs) (perfluorooctanesulfonic acid (PFOS), perfluorooctanoic acid (PFOA) and perfluorobutanesulfonic acid (PFBS)) was studied in sewage sludge samples. Sorption isotherms were obtained by varying initial concentrations of PFOS, PFOA and PFBS. The maximum values of the sorption solid-liquid distribution coefficients ( $K_{d,max}$ ) varied by almost two orders of magnitude among the target PFASs: 140-281 mL g(-1) for PFOS, 30-54 mL g(-1) for PFOA and 9-18 mL g(-1) for PFBS. Freundlich and linear fittings were appropriate for describing the sorption behaviour of PFASs in the sludge samples, and the derived  $K_F$  and  $K_d$ , linear parameters correlated well. The hydrophobicity of the PFASs was the key parameter that influenced their sorption in sewage sludge. Sorption parameters and  $\log(K_{OW})$  were correlated, and for PFOS (the most hydrophobic compound), pH and Ca + Mg status of the sludge controlled the variation in the sorption parameter values. Sorption reversibility was also tested from desorption isotherms, which were also linear. Desorption parameters were systematically higher than the corresponding sorption parameters (up to sixfold higher), thus indicating a significant degree of irreversible sorption, which decreased in the sequence PFOS > PFOA > PFBS.

Navarro, I., et al. (2016). "Bioaccumulation of emerging organic compounds (perfluoroalkyl substances and halogenated flame retardants) by earthworm in biosolid amended soils." *Environ Res* 149: 32-39.

In the present work, the bioaccumulation behavior of 49 target emerging organic compounds (20 perfluoroalkyl substances, PFASs, and 29 halogenated flame retardants, HFRs) was studied in soil invertebrates (*Eisenia andrei*). Multi species soil systems (MS.3) were used to assess the fate and the effects associated with the application of four biosolids in agricultural soil on terrestrial soil organisms. Biosolid amendment increased concentrations 1.5-14-fold for PFASs, 1.1-2.4-fold for polybrominated diphenyl ethers, PBDEs, and 1.1-3.6-fold for chlorinated flame retardants, CFRs. Perfluorooctanesulfonate, PFOS, (25%) and BDE-209 (60%) were the predominant PFAS and HFR compounds, respectively, in biosolids-amended soils. Total concentrations (ng/g dry weight) in earthworms from biosolid-amended soils ranged from 9.9 to 101 for PFASs, from 45 to 76 for PBDEs and 0.3-32 for CFRs. Bioaccumulation factors (BAFs) were calculated to evaluate the degree of exposure of pollutants in earthworms. The mean BAF ranged from 2.2 to 198 for PFASs, 0.6-17 for PBDEs and 0.5-20 for CFRs. The relationship of PFAS and PBDE BAFs in earthworms and their log Kow were compared: PFAS BAFs increased while PBDE BAFs declined with increasing log Kow values. The effect of the aging (21 days) on the bioavailability of the pollutants in amended soils was also assessed: the residence time affected differently to the compounds studied.

Navarro, I., et al. (2017). "Uptake of perfluoroalkyl substances and halogenated flame retardants by crop plants grown in biosolids-amended soils." *Environ Res* 152: 199-206.

The bioaccumulation behavior of perfluoroalkyl substances (PFASs) and halogenated flame retardants (HFRs) was examined in three horticultural crops and earthworms. Two species, spinach (*Spinacia oleracea*) and tomato (*Solanum lycopersicum* L.), were grown in field soil amended with a single application of biosolids (at agronomic rate for nitrogen), to represent the scenario using commercial biosolids as fertilizer, and the third crop, corn (*Zea mays*) was grown in spiked soil (~50mg PFOS/kg soil, ~5mg Deca-BDE/kg soil and a mixture of both, ~50mg PFOS and ~5mg Deca-BDE/kg soil) to represent a worst-case scenario. To examine the bioaccumulation in soil invertebrates, earthworms (*Eisenia andrei*) were exposed to the spiked soil where corn had been grown. PFASs and HFRs were detected in the three crops and earthworms. To evaluate the distribution of the compounds in the different plant tissues, transfer factors (TFs) were calculated, with TF values higher for PFASs than PBDEs in all crop plants: from 2 to 9-fold in spinach, 2 to 34-fold in tomato and 11 to 309-fold in corn. Bioaccumulation factor (BAF) values in earthworms were also higher for PFASs (4.06+/-2.23) than PBDEs (0.02+/-0.02).

Rastetter, N. and A. Gerhardt (2017). "Toxic potential of different types of sewage sludge as fertiliser in agriculture: ecotoxicological effects on aquatic, sediment and soil indicator species." *Journal of Soils and Sediments* 17(1): 106-121.

Purpose Treated and processed sewage sludges (biosolids) generated during the treatment of wastewater usually contain substantial concentrations of nutrients, especially phosphorus, which is essential for plant growth. Sewage sludge therefore can be used as an alternative fertiliser in agriculture. But since sewage sludge could also contain pollutants, analysis and ecotoxicological tests on affected soil and stream water organisms are necessary in order to guarantee its harmless use. Materials and methods Three test species were chosen to cover the environmental compartments, water, sediment and soil. The following test species and parameters were applied to evaluate the acute effects of three sewage sludge samples: *Lemna minor* (growth inhibition, discolouration and colony breakup), *Gammarus fossarum* (mortality, behaviour) and *Eisenia fetida* (avoidance behaviour). Chemical assessment included nutrients, organic pollutants and heavy metals. Results and discussion The assessment of a non-dewatered sludge (S1) sample resulted in an inhibition of growth of *L. minor* starting from 0.6 g total solid (TS) l(-1) after 7 days (EC50 1.2 g TS l(-1)). *G. fossarum* displayed significantly decreased movement activity at 0.5 and 1.2 g TS l(-1) sludge concentration during an exposure time of 2 days, leading to decreased survival after 4 days of exposure in 0.5 g TS l(-1) (LC50 0.5 g TS l(-1)). After 2 days, *E. fetida* exhibited an increased avoidance behaviour of contaminated soil from 0.2 g TS kg(-1) sewage sludge (EC50 0.4 g TS kg(-1)). The dewatered sludge samples (S2 and S3) had a lower toxic effect on the test organisms. *G. fossarum* was the most sensitive test species in the applied test setups. The realistic application amounts of the tested sewage sludge samples of approximately 6.0 g TS kg(-1) (maximum allowed application amount of sewage sludge) and approximately 3 g TS kg(-1) (maximum agronomical relevant application amount) in worst case studies are higher than the analysed EC50/LC50 values of S1 and of the LC50 (*G. fossarum*) of S2 and S3. Conclusions All three tested sewage sludge samples have to be classified as toxic at high concentration levels under laboratory conditions. Realistic output quantities of S1 will negatively influence soil invertebrates and freshwater organisms (plants and crustacean), whereas the dewatered sludge samples will most likely not have any acute toxic effect on the test organisms in the field. Test with environmental samples should be conducted in order to support this hypothesis.

Sherburne, J. J., et al. (2016). "Occurrence of Triclocarban and Triclosan in an Agro-ecosystem Following Application of Biosolids." *Environ Sci Technol* 50(24): 13206-13214.

Triclocarban (TCC) and triclosan (TCS), two of the most commonly used antimicrobial compounds, can be introduced into ecosystems by applying wastewater treatment plant biosolids to agricultural fields. Concentrations of TCC and TCS were measured in different trophic levels within a terrestrial food web encompassing land-applied biosolids, soil, earthworms (*Lumbricus*), deer mice (*Peromyscus maniculatus*), and eggs of European starlings (*Sturnus vulgaris*) and American kestrels (*Falco sparverius*) at an experimental site amended with biosolids for the previous 7 years. The samples from this site were compared to the same types of samples from a reference (biosolids-free) agricultural site. Inter-site comparisons showed that concentrations of both antimicrobials were higher on the experimental site in the soil, earthworms, mice (livers), and European starling eggs, but not American kestrel eggs, compared to the control site. Inter-species comparisons on the experimental site indicated significantly higher TCC concentrations in mice (TCC: 12.6-33.3 ng/g) and in starling eggs (TCC: 15.4-31.4 ng/g) than in kestrel eggs (TCC: 3.6 ng/g). Nesting success of kestrels only was significantly lower on the experimental site compared to the reference site due to nest abandonment. This study demonstrates that biosolids-derived TCC and TCS are present throughout the terrestrial food web, including secondary (e.g., starlings) and tertiary (i.e., kestrels) consumers, after repeated, long-term biosolids application.

## Appendix C. Reference Abstracts: Other

### C.1 Abstracts of Papers Containing Only Regulated Metals

Alvarenga, P., et al. (2016). "Beneficial Use of Dewatered and Composted Sewage Sludge as Soil Amendments: Behaviour of Metals in Soils and Their Uptake by Plants." *Waste and Biomass Valorization* 7(5): 1189-1201.

In Portugal, where soils have a very low topsoil organic matter content, the use of sewage sludge (SS) as organic soil improvers seems an attractive option, because it would enable organic matter, N, P, K and other nutrients to be recycled. However, the risk of this practice must be properly assessed. The aim of this study was to evaluate the potential risk of the use of dewatered and composted SS as soil amendments. For this purpose, SS from two different wastewater treatment plants (SS1 and SS2), and a compost produced from SS and agricultural wastes (AWSSC), were characterized for their total metal concentrations, organic contaminants and indicator pathogens, and used in a pot experiment with three application rates, 6, 12 and 24 ton dry matter/ha, cultivated with a hybrid variety of sorghum and Sudan grass (*Sorghum bicolor* x *Sorghum sudanense* var. Rocket). SS1 and AWSSC met the legal requirements to be applied to soils, but SS2 had a high content of pathogens, which compromised its use. Both dewatered SS had a marked beneficial effect on plant production and on soil nutritional characteristics, more pronounced than in the case of AWSSC application, without a significant increase in total and in mobile metals concentration in soils. Bioaccumulation factors for metals in plants were low, and their concentrations in the aboveground plant material were lower than the maximum tolerable level for cattle, used as a risk indicator of metal entry into the human food chain. However, it will be necessary, in future studies, to evaluate the potential risk of the observed increase in the mobilisable content of Cu and Zn in soil, as a consequence of the application of these organic materials.

Bai, Y., et al. (2017). "Sewage Sludge Amendment Combined with Green Manuring to a Coastal Mudflat Salt-Soil in Eastern China: Effects on Soil Physicochemical Properties and Maize Yield." *International Journal of Agronomy*: 1-10.

Sewage sludge and green manure have become widely used organic amendments to croplands in many regions of the world. However, the amending effect of the combination of sewage sludge with green manuring in reclaimed coastal mudflat salt-soil has been unclear yet. This paper was one of earlier studies to investigate and evaluate the effects of sewage sludge amendment combined with green manuring on selected soil physicochemical properties of the mudflat soil in a rain-fed agroecosystem. The mudflat salt-soil was amended by one-time input of sewage sludge at the rates of 0, 30, 75, 150, and 300 t ha<sup>-1</sup>. After green manuring for three consecutive seasons, maize (*Zea mays* L.) was planted in 2013 and 2014. The results showed that SSA combined with green manuring decreased bulk density, pH, salinity, and exchangeable sodium percentage of the topsoil (0–20 cm soil layer) and increased aggregate stability, cation exchange capacity, and N and P concentration of the topsoil. As a result, the maize yield increased with the increase of SSA rates. Sewage sludge combined with green manuring can be applied in coastal mudflat salt-soil amendment, which provides an innovative way to create arable land resources and safe disposal of sewage sludge.

Barbarick, K., et al. (2017). "Meta-Analyses of Biosolids Effect in Dryland Wheat Agroecosystems." *J Environ Qual* 46(2): 452-460.

Land application to cropping systems is USEPA's preferred method of recycling biosolids. Determination of biosolids effect size through meta-analyses from two decades of field-location research at three sites should answer the question: Does 20 yr of biosolids application affect dryland wheat (L.) grain production, grain nutrient concentrations, and soil elemental extractability compared with equivalent rates of commercial N fertilizer? At two sites, biennial biosolids application rates to a wheat-fallow (WF) rotation were up to 11.2 dry Mg ha and up to 112 kg commercial N fertilizer ha, whereas rates at the third location varied to match soil-test information. Crop rotations included WF and wheat-corn (L.)-fallow. We completed meta-analyses of biosolids effects compared with N fertilizer on wheat yield, grain protein, grain total, and soil ammonium bicarbonate-diethylenetriaminepentaacetic acid (ABDTPA)-extractable P, Zn, Cu, Fe, and Ni concentrations at the aforementioned sites from 1993 through 2013. Results showed that biosolids produced greater grain P and Zn at one site. Biosolids rates at two sites resulted in greater grain Zn and ABDTPA P, Zn, Cu, and Fe. Meta-analyses tests for heterogeneity indicated that the variance for all sites and rates could be explained as consistent across treatments, whereas the test for the 20 yr showed that heterogeneity was large and other factors affected the variance (e.g., climatic variability between years). Meta-analysis showed the practical effect of biosolids over a 20-yr study and demonstrated that the primary biosolids effect was an improvement in Zn availability to wheat.

Belhaj, D., et al. (2016). "Effects of sewage sludge fertilizer on heavy metal accumulation and consequent responses of sunflower (*Helianthus annuus*)." *Environ Sci Pollut Res Int* 23(20): 20168-20177.

Use of sewage sludge, a biological residue produced from sewage treatment processes in agriculture, is an alternative disposal technique of waste. To study the usefulness of sewage sludge amendment for *Helianthus annuus*, a pot experiment was conducted by mixing sewage sludge at 2.5, 5, and 7.5 % (w/w) amendment ratios to the agricultural soil. Soil pH decreased whereas electrical conductivity, organic matter, total N, available P, and exchangeable Na, K, and Ca increased in soil amended with sewage sludge in comparison to unamended soil. Sewage sludge amendment led to significant increase in Pb, Ni, Cu, Cr, and Zn concentrations of soil. The increased concentration of heavy metals in soil due to sewage sludge amendment led to increases in shoot and root concentrations of Cr, Cu, Ni, and Zn in plant as compared to those grown on unamended soil. Accumulation was more in roots than shoots for most of the heavy metals. Moreover, high metal removal for the harvestable parts of the crops was recorded. Sewage sludge amendment increased root and shoot length, leaves number, biomass, and antioxidant activities of sunflower. Significant increases in the activities of antioxidant enzymes and in the glutathione, proline, and soluble sugar content in response to amendment with sewage sludge may be defense mechanisms induced in response to heavy metal stress. Graphical abstract Origin, fate and behavior of sewage sludge fertilizer.

Bogusz, A., et al. (2017). "Adsorption and desorption of heavy metals by the sewage sludge and biochar-amended soil." *Environ Geochem Health*.

The goal of the study was to evaluate the application of biochar (BC) to the sewage sludge (SL) on the adsorption and desorption capacity of Cd(II), Cu(II), Ni(II) and Zn(II). The effect of biochar contribution in the sewage sludge (2.5, 5 and 10%) was investigated. The isotherms data were fitted to the Langmuir (LM), Freundlich (FM) and Temkin (TM) models. The best fitting for kinetic study was obtained for the pseudo-second-order equation. The best fitting of the experimental data was observed for the LM in the case of SL and BC, and for the FM in the case of SL- and SL/BC-amended soil. SL was characterized by even four-order higher sorption capacity than BC. The addition of the BC to the SL and next to the soil increased the adsorption capacity of the soil and the SL-amended soil. In the case of all investigated potentially toxic elements (PTEs), the highest adsorption capacity was achieved for SL-amended soil in comparison with the control soil. In the case of other experimental variants, the adsorption capacity of metal ions was as follows: 2.5% BC > 5.0% BC > 10% BC. The negative correlation between hydrated radius of metal ions and the kinetics of sorption was observed. However, the desorption of PTEs from BC/SL-amended soil was significantly lower than for SL-amended soil (except of Cd) and non-amended soil. It can be concluded that the addition of the biochar enhanced the immobilization of PTEs and reduced their bioavailability and mobility in the soil amended by the sewage sludge.

Bonomo, M. M., et al. (2016). "Sewage sludge hazardous assessment: chemical evaluation and cytological effects in CHO-k1 cells." *Environ Sci Pollut Res Int* 23(11): 11069-11075.

Application of sewage sludge in agricultural lands is a growing practice in several countries due to its numerous benefits to soil and crops, where chemical and pathogen levels are determined by corresponding legislation. However, the presence of contaminants in residues must always be controlled before application due to their dangerous effects over the ecosystem and potential risks to human health. The main objective of this study was to integrate biological and chemical analysis in order to help elucidating the residue potential toxic, cytotoxic, and mutagenic effects. We evaluate samples of sewage sludge before and after the sanitizing treatment with lime in cytokinesis-block assay using CHO-k1 culture cells. The sanitizing treatment promoted a decrease in pathogen levels, which is the main purpose of this process. Even with chemical levels below the established by environmental agencies, results showed sewage sludge ability to enhance genotoxic and mutagenic effects, proving that residue should be handled with caution in order to minimize its environmental and human risk.

Cele, E. N. and M. Maboeta (2016). "Amelioration of iron mine soils with biosolids: Effects on plant tissue metal content and earthworms." *Environ Sci Pollut Res Int* 23(22): 23005-23016.

The achievement of environmentally sound and economically feasible disposal strategies for biosolids is a major issue in the wastewater treatment industry around the world, including Swaziland. Currently, an iron ore mine site, which is located within a wildlife sanctuary, is being considered as a suitable place where controlled disposal of biosolids may be practiced. Therefore, this study was conducted to investigate the effects of urban biosolids on iron mine soils with regard to plant metal content and ecotoxicological effects on earthworms. This was done through chemical analysis of plants grown in biosolid-amended mine soil. Earthworm behaviour, reproduction and bioaccumulation tests were also conducted on biosolid-amended mine soil. According to the results obtained, the use of biosolids led to creation of soil conditions that were generally favourable to earthworms. However, plants were found to have accumulated Zn up to 346 mg kg<sup>-1</sup> (in shoots) and 462 mg kg<sup>-1</sup> (in roots). This was more than double the normal Zn content of plants. It was concluded that while biosolids can be beneficial to mine soils and earthworms, they can also lead to elevated metal content in plant tissues, which might be a concern to plant-dependant wildlife species. Nonetheless, it was not possible to satisfactorily estimate risks to forage quality since animal feeding tests with hyperaccumulator plants have not been reported. Quite possibly, there may be no cause for alarm since the uptake of metals from soil is greater in plants grown in pots in the greenhouse than from the same soil in the field since pot studies fail to mimic field conditions where the soil is heterogeneous and where the root system possesses a complex topology. It was thought that further field trials might assist in arriving at more satisfactory conclusions.

Eid, E. M., et al. (2017). "The effects of different sewage sludge amendment rates on the heavy metal bioaccumulation, growth and biomass of cucumbers (*Cucumis sativus* L.)." *Environ Sci Pollut Res Int* 24(19): 16371-16382.

When sewage sludge is incorrectly applied, it may adversely impact agro-system productivity. Thus, this study addresses the reaction of *Cucumis sativus* L. (cucumber) to different amendment rates (0, 10, 20, 30, 40 and 50 g kg<sup>-1</sup>) of sewage sludge in a greenhouse pot experiment, in which the plant growth, heavy metal uptake and biomass were evaluated. A randomized complete block design with six treatments and six replications was used as the experimental design. The soil electrical conductivity, organic matter and Cr, Fe, Zn and Ni concentrations increased, but the soil pH decreased in response to the sewage sludge applications. As approved by the Council of European Communities, all of the heavy metal concentrations in the sewage sludge were less than the permitted limit for applying sewage sludge to land. Generally, applications of sewage sludge of up to 40 g kg<sup>-1</sup> resulted in a considerable increase in all of the morphometric parameters and biomass of cucumbers in contrast to plants grown on the control soil. Nevertheless, the cucumber shoot height; root length; number of leaves, internodes and fruits; leaf area; absolute growth rate and biomass decreased in response to 50 g kg<sup>-1</sup> of sewage sludge. All of the heavy metal concentrations (except the Cu, Zn and Ni in the roots, Mn in the fruits and Pb in the stems) in different cucumber tissues increased with increasing sewage sludge application rates. However, all of the heavy metal concentrations (except the Cr and Fe in the roots, Fe in the leaves and Cu in the fruits) were within the normal range and did not reach phytotoxic levels. A characteristic of these cucumbers was that all of the heavy metals had a bioaccumulation factor <1.0. All of the heavy metals (except Cd, Cu and Zn) had translocation factors that were <1.0. As a result, the sewage sludge used in this study could be considered for use as a fertilizer in cucumber production systems in Saudi Arabia and can also serve as a substitute method of sewage sludge disposal. Graphical Abstract The effects of different sewage sludge amendment rates on the heavy metal bioaccumulation, growth and biomass of cucumbers.

Eid, E. M., et al. (2017). "Effects of different sewage sludge applications on heavy metal accumulation, growth and yield of spinach (*Spinacia oleracea* L.)." *Int J Phytoremediation* 19(4): 340-347.

In this study, we present the response of spinach to different amendment rates of sewage sludge (0, 10, 20, 30, 40 and 50 g kg<sup>-1</sup>) in a greenhouse pot experiment, where plant growth, biomass and heavy metal uptake were measured. The results showed that sewage sludge application increased soil electric conductivity (EC), organic matter, chromium and zinc concentrations and decreased soil pH. All heavy metal concentrations of the sewage sludge were below the permissible limits for land application of sewage sludge recommended by the Council of the European Communities. Biomass and all growth parameters (except the shoot/root ratio) of spinach showed a positive response to sewage sludge applications up to 40 g kg<sup>-1</sup> compared to the control soil. Increasing the sewage sludge amendment rate caused an increase in all heavy metal concentrations (except lead) in spinach root and shoot. However, all heavy metal concentrations (except chromium and iron) were in the normal range and did not reach the phytotoxic levels. The spinach was characterized by a bioaccumulation factor <1.0 for all heavy metals. The translocation factor (TF) varied among the heavy metals as well as among the sewage sludge amendment rates. Spinach translocation mechanisms clearly restricted heavy metal transport to the edible parts (shoot) because the TFs for all heavy metals (except zinc) were <1.0. In conclusion, sewage sludge used in the present study can be considered for use as a fertilizer in spinach production systems in Saudi Arabia, and the results can serve as a management method for sewage sludge.

Eid, E. M. and K. H. Shaltout (2016). "Bioaccumulation and translocation of heavy metals by nine native plant species grown at a sewage sludge dump site." *Int J Phytoremediation* 18(11): 1075-1085.

In the present study, nine native plant species were collected to determine their potential to clean up nine heavy metals from soil of a sewage sludge dump site. Almost all nine plant species grown at sewage sludge dump site showed multifold higher concentrations of heavy metals as compared to plants grown at the reference site. All the investigated species were characterized by a bioaccumulation factor (BF) > 1.0 for some heavy metals. BF was generally higher for Cd, followed by Pb, Co, Cr, Cu, Ni, Mn, Zn, and Fe. The translocation factor (TF) varied among plant species, and among heavy metals. For most studied heavy metals, TFs were <1.0. The present study proved that the concentrations of all heavy metals (except Cd, Co, and Pb) in most studied species were positively correlated with those in soil. Such correlations indicate that these species reflect the cumulative effects of environmental pollution from soil, and thereby suggesting their potential use in the biomonitoring of most heavy metals examined. In conclusion, all tissues of nine plant species could act as bioindicators, biomonitors, and remediates of most examined heavy metals. Moreover, *Bassia indica*, *Solanum nigrum*, and *Pluchea dioscoridis* are considered hyperaccumulators of Fe; *Amaranthus viridis* and *Bassia indica* are considered hyperaccumulators of Pb; and *Portulaca oleracea* is considered hyperaccumulator of Mn.

Healy, M. G., et al. (2016). "Bioaccumulation of metals in ryegrass (*Lolium perenne* L.) following the application of lime stabilised, thermally dried and anaerobically digested sewage sludge." *Ecotoxicol Environ Saf* 130: 303-309.

The uptake and accumulation of metals in plants is a potential pathway for the transfer of environmental contaminants in the food chain, and poses potential health and environmental risks. In light of increased population growth and urbanisation, the safe disposal of sewage sludge, which can contain significant levels of toxic contaminants, remains an environmental challenge globally. The aims of this experiment were to apply municipal sludge, having undergone treatment by thermal drying, anaerobic digestion, and lime stabilisation, to permanent grassland in order to assess the bioaccumulation of metals (B, Al, Ti, V, Cr, Mn, Co, Ni, Cu, Zn, As, Nb, Mo, Sb, Ba, W, Pb, Fe, Cd) by perennial ryegrass over a period of up to 18 weeks after application. The legislation currently prohibits use of grassland for fodder or grazing for at least three weeks after application of treated sewage sludge (biosolids). Five treatments were used: thermally dried (TD), anaerobically digested (AD) and lime stabilised (LS) sludge all from one wastewater treatment plant (WWTP), AD sludge from another WWTP, and a study control (grassland only, without application of biosolids). In general, there was no significant difference in metal content of the ryegrass between micro-plots that received treated municipal sludge and the control over the study duration. The metal content of the ryegrass was below the levels at which phytotoxicity occurs and below the maximum levels specified for animal feeds.

Mendez, A., et al. (2017). "The effect of sewage sludge biochar on peat-based growing media." *Biological Agriculture & Horticulture* 33(1): 40-51.

Peat is the main component of growing media in horticulture. Increasing demand, environmental concerns and rising costs for peat make the search for alternative materials imperative. Much research has been performed aiming to find high quality and low cost substrates from different organic wastes such as compost and thus decrease peat consumption. Biochar is a carbon-rich material that has attracted important research as a soil amendment. However, its potential utilization as a peat substitute for growing media formulation remains less well explored. The aim of this study was to evaluate the effects of sewage sludge and sewage sludge biochar on peat properties as growing media and on lettuce (*Lactuca sativa*) growth. Sewage sludge transformation into biochar proved to be a sustainable waste management approach in order to promote their future use as growing media components. Addition of biochar from sewage sludge increased the N, P and K content of growing media. The biochar addition to peat at a 10% vol rate increased lettuce biomass production by 184-270% and the shoot length by 137-147% despite hydrophysical properties not being improved. Also, biochar addition had a positive effect on growing media microbial biomass which increased more than 966%. In spite of the higher metal concentration in biochar than in sewage sludge, their transfer to plants seems to be reduced when compared with direct sewage sludge use.

Milik, J., et al. (2017). "The concentration of trace elements in sewage sludge from wastewater treatment plant in Gniewino." *Journal of Ecological Engineering* 18(5): 118-124.

The sewage sludge originating from wastewater treatment plants (WWTP) serving rural areas is suggested for agricultural or natural usage. However, sewage sludge is beforehand subjected to the several pre-treatments, which involve stabilization, hygienisation and pre-composting. These methods mainly decrease the amount of organic substances and the presence of microorganisms, but hardly affect the concentrations of heavy metals. The advantages of using sludges as fertilizer for improving and sustaining soil fertility and crop production are numerous. The addition of sewage sludge to soils could affect the potential availability of heavy metals. Trace elements are distributed in the soil in various forms: solid phases, free ions in soil solution, soluble organic-mineral complexes, or adsorbed on colloidal particles. The most undesirable heavy metals in sewage sludge that are toxic for the living organisms include: cadmium, chromium, nickel, lead and mercury. In the study, the concentrations of trace elements (Pb, Cd, Cr, Hg, Ni, Zn, Al, As, Se, B, Ba, Br, Ca, Cu, Fe, Mn, Na, Ga, Li, Mo, Sr, Mg, K, Ru, Tl, V, U) were tested in the sewage sludge obtained from a WWTP serving rural areas (PE < 9 000). In each case, the tested sewage sludge was meeting the criteria of stabilization and was used for agriculture and land reclamation purpose. All the samples were collected in 2016 and subjected to microwave mineralization in a closed system in aqua regia. The total amounts of macro and microelements were determined with a spectrophotometer Coupled Plasma emission ICP-OES. It was found that the total concentrations of trace metals in all of sewage sludges are the same as the Polish regulation limit of pollutants for sludge to be used in agriculture. European legislation is less restrictive and permits higher contents of heavy metals in sludge used for agriculture than Asia. The trace elements (cadmium: 1.16 mg·kg<sup>-1</sup>/d.m. in the Polish sewage sludge, are much higher than those in the other countries. Copper and zinc were the most prevalent elements observed (111.28 mg·kg<sup>-1</sup>/d.m. and 282.94 mg·kg<sup>-1</sup>/d.m., respectively). The concentrations of copper in the Polish sewage sludge are much lower (49-130 mg·kg<sup>-1</sup>/d.m.) than European sewage sludge (522-562 mg·kg<sup>-1</sup>/d.m.). The two of the tested heavy metals (beryllium, bismuth) were under the detection limit, while gallium, molybdenum, thallium, vanadium and silver were detected in the concentrations lower than 0.005 mg·kg<sup>-1</sup>/d.m. According to the obtained results, in all the tested samples, the total amount of trace elements, did not exceed the limit values in sewage sludge for their use in agriculture and land reclamation.

Rorat, A., et al. (2017). "Vermiremediation of polycyclic aromatic hydrocarbons and heavy metals in sewage sludge composting process." *J Environ Manage* 187: 347-353.

The main objective of this work was to study the dynamics of the degradation of polycyclic aromatic hydrocarbons (PAHs) during sewage sludge vermicomposting. This eco-biotechnology employing earthworms as natural bioreactors for decomposing of organic matter may be used for vermicoremediation of particular pollutants present in various organic matter sources. In this experiment, sewage sludge was mixed with bulking agents and precomposted. Afterward, adult *Eisenia andrei* specimens were introduced into the process. Total heavy metal and PAHs concentration were measured in composts and earthworms before and after the process. While heavy metal concentrations fluctuated mildly in the substratum, several metals clearly accumulated in the earthworms' bodies. Body Accumulation Factors could be ranked as follows (Cd > Cu > Zn > Ni > Cr > Pb). Interestingly, addition of earthworms into the process has led to the high percentage of PAHs removal and some of the 16 priority PAHs analyzed in this study have been accumulated in earthworms' bodies. Applied conditions did not affect worms' viability but they almost completely inhibited their reproduction.

## C.2 Abstracts of Papers Containing Novel Chemicals without Data

Jesmer, A. H., et al. (2017). "The toxicity of silver to soil organisms exposed to silver nanoparticles and silver nitrate in biosolids-amended field soil." *Environ Toxicol Chem* 36(10): 2756-2765.

The use of engineered silver nanoparticles (AgNPs) is widespread, with expected release to the terrestrial environment through the application of biosolids onto agricultural lands. The toxicity of AgNPs and silver nitrate ( $\text{AgNO}_3$ ; as ionic  $\text{Ag}^+$ ) to plant (*Elymus lanceolatus* and *Trifolium pratense*) and soil invertebrate (*Eisenia andrei* and *Folsomia candida*) species was assessed using Ag-amended biosolids applied to a natural sandy loam soil. Bioavailable  $\text{Ag}^+$  in soil samples was estimated using an ion-exchange technique applied to  $\text{KNO}_3$  soil extracts, whereas exposure to dispersible AgNPs was verified by single-particle inductively coupled plasma-mass spectrometry and transmission electron microscopy-energy dispersive X-ray spectroscopy analysis. Greater toxicity to plant growth and earthworm reproduction was observed in AgNP exposures relative to those of  $\text{AgNO}_3$ , whereas no difference in toxicity was observed for *F. candida* reproduction. Transformation products in the AgNP-biosolids exposures resulted in larger pools of extractable  $\text{Ag}^+$  than those from  $\text{AgNO}_3$ -biosolids exposures, at similar total Ag soil concentrations. The results of the present study reveal intrinsic differences in the behavior and bioavailability of the 2 different forms of Ag within the biosolids-soils pathway. The present study demonstrates how analytical methods that target biologically relevant fractions can be used to advance the understanding of AgNP behavior and toxicity in terrestrial environments.

Kraas, M., et al. (2017). "Long-term effects of sulfidized silver nanoparticles in sewage sludge on soil microflora." *Environ Toxicol Chem* 36(12): 3305-3313.

The use of silver nanoparticles (AgNPs) in consumer products such as textiles leads to their discharge into wastewater and consequently to a transfer of the AgNPs to soil ecosystems via biosolids used as fertilizer. In urban wastewater systems (e.g., sewer, wastewater treatment plant [WWTP], anaerobic digesters) AgNPs are efficiently converted into sparingly soluble silver sulfides ( $\text{Ag}_2\text{S}$ ), mitigating the toxicity of the AgNPs. However, long-term studies on the bioavailability and effects of sulfidized AgNPs on soil microorganisms are lacking. Thus we investigated the bioavailability and long-term effects of AgNPs (spiked in a laboratory WWTP) on soil microorganisms. Before mixing the biosolids into soil, the sludges were either anaerobically digested or directly dewatered. The effects on the ammonium oxidation process were investigated over 140 d. Transmission electron microscopy (TEM) suggested an almost complete sulfidation of the AgNPs analyzed in all biosolid samples and in soil, with  $\text{Ag}_2\text{S}$  predominantly detected in long-term incubation experiments. However, despite the sulfidation of the AgNPs, soil ammonium oxidation was significantly inhibited, and the degree of inhibition was independent of the sludge treatment. The results revealed that AgNPs sulfidized under environmentally relevant conditions were still bioavailable to soil microorganisms. Consequently,  $\text{Ag}_2\text{S}$  may exhibit toxic effects over the long term rather than the short term.

Mahon, A. M., et al. (2017). "Microplastics in Sewage Sludge: Effects of Treatment." *Environ Sci Technol* 51(2): 810-818.

Waste water treatment plants (WWTPs) are receptors for the cumulative loading of microplastics (MPs) derived from industry, landfill, domestic wastewater and stormwater. The partitioning of MPs through the settlement processes of wastewater treatment results in the majority becoming entrained in the sewage sludge. This study characterized MPs in sludge samples from seven WWTPs in Ireland which use anaerobic digestion (AD), thermal drying (TD), or lime stabilization (LS) treatment processes. Abundances ranged from 4196 to 15385 particles kg<sup>-1</sup> (dry weight). Results of a general linear mixed model (GLMM) showed significantly higher abundances of MPs in smaller size classes in the LS samples, suggesting that the treatment process of LS shears MP particles. In contrast, lower abundances of MPs found in the AD samples suggests that this process may reduce MP abundances. Surface morphologies examined using scanning electron microscopy (SEM) showed characteristics of melting and blistering of TD MPs and shredding and flaking of LS MPs. This study highlights the potential for sewage sludge treatment processes to affect the risk of MP pollution prior to land spreading and may have implications for legislation governing the application of biosolids to agricultural land.

## Appendix D. Reference Abstracts: Microbials

Amoros, I., Moreno, Y., Reyes, M., Moreno-Mesonero, L. and Alonso, J. L. 2016. Prevalence of *Cryptosporidium* oocysts and *Giardia* cysts in raw and treated sewage sludges. *Environ Technol* 37(22):2898-904.

Treated sludge from wastewater treatment plants (WWTPs) is commonly used in agriculture as fertilizers and to amend soils. The most significant health hazard for sewage sludge relates to the wide range of pathogenic microorganisms such as protozoa parasites. The objective of this study was to collect quantitative data on *Cryptosporidium* oocysts and *Giardia* cysts in the treated sludge in wastewater treatment facilities in Spain. Sludge from five WWTPs with different stabilization processes has been analysed for the presence of *Cryptosporidium* and *Giardia* in the raw sludge and after the sludge treatment. A composting plant (CP) has also been assessed. After a sedimentation step, sludge samples were processed and (oo)cysts were isolated by immunomagnetic separation (IMS) and detected by immunofluorescence assay (IFA). Results obtained in this study showed that *Cryptosporidium* oocysts and *Giardia* cysts were present in 26 of the 30 samples (86.6%) of raw sludge samples. In treated sludge samples, (oo)cysts have been observed in all WWTP's analysed (25 samples) with different stabilization treatment (83.3%). Only in samples from the CP no (oo)cysts were detected. This study provides evidence that (oo)cysts are present in sewage sludge-end products from wastewater treatment processes with the negative consequences for public health.

Goberna, M., Simon, P., Hernandez, M. T. and Garcia, C. 2018. Prokaryotic communities and potential pathogens in sewage sludge: Response to wastewater origin, loading rate and treatment technology. *Sci Total Environ* 615:360-368. E-pub 10/11/2017.

Sewage sludge features high nitrogen and phosphorous contents encouraging its use as a biosolid in agriculture, but it bears potential chemical and microbiological risks. To tease apart the relative contribution of main factors determining the sludge chemical and microbial features, we analysed 28 treatment plants differing in the wastewater origin (municipal residues, agro-food or chemical industries), organic loading rate and treatment technology (extended aeration, activated sludge or activated sludge followed by anaerobic digestion). We found that the treatment technology and the organic loading rate are main determinants of the sludge chemical properties, including its organic load, nutrient and metal contents, and override the effect of the wastewater origin. Sludge bacterial and archaeal community structure and diversity, characterized through massive sequencing of the 16S rRNA gene, were also mostly determined by the treatment technology partly through shifts in the sludge nutrient load. The same factor conditioned the relative abundance of sequenced bacteria most closely related to potential pathogens, but not that of cultivable *Escherichia coli* or *Salmonella* spp. We did not find an effect of the geographic location of the plant on any of the variables at the regional scale of our study. Operational parameters appear as major determinants of the sludge chemical and microbial properties, irrespective of the source of wastewaters, thus leaving a broad management window for improving the agronomic value of sewage sludge.

Gondim-Porto, C., Platero, L., Nadal, I. and Navarro-Garcia, F. 2016. Fate of classical faecal bacterial markers and ampicillin-resistant bacteria in agricultural soils under Mediterranean climate after urban sludge amendment. *Sci Total Environ* 565:200-210.

The use of sewage sludge or biosolids as agricultural amendments may pose environmental and human health risks related to pathogen or antibiotic-resistant microorganism transmission from soils to vegetables or to water through runoff. Since the survival of those microorganisms in amended soils has been poorly studied under Mediterranean climatic conditions, we followed the variation of soil fecal bacterial markers and ampicillin-resistant bacteria for two years with samplings every four months in a split block design with three replica in a crop soil where two different types of biosolids (aerobically or anaerobically digested) at three doses (low, 40; intermediate, 80; and high, 160Mg.ha(-1)) were applied. Low amounts of biosolids produced similar decay rates of coliform populations than in control soil (-0.19 and -0.27log<sub>10</sub>CFUs g(-1) drysoilmonth(-1) versus -0.22) while in the case of intermediate and high doses were close to zero and their populations remained 24 months later in the range of 4-5 log<sub>10</sub> CFUs g(-1) ds. *Enterococci* populations decayed at different rates when using aerobic than anaerobic biosolids although high doses had higher rates than control (-0.09 and -0.13 log<sub>10</sub> CFUs g(-1) dsmonth(-1) for aerobic and anaerobic, respectively, vs -0.07). At the end of the experiment, counts in high aerobic and low and intermediate anaerobic plots were 1 log<sub>10</sub> higher than in control (4.21, 4.03, 4.2 and 3.11 log<sub>10</sub> CFUs g(-1) ds, respectively). Biosolid application increased the number of *Clostridium* spores in all plots at least 1 log<sub>10</sub> with respect to control with a different dynamic of decay for low and intermediate doses of aerobic and anaerobic sludge. Ampicillin-resistant bacteria increased in amended soils 4 months after amendment and remained at least 1 log<sub>10</sub> higher 24 months later, especially in aerobic and low and intermediate anaerobic plots due to small rates of decay (in the range of -0.001 to -0.008 log<sub>10</sub> CFUs g(-1) dsmonth(-1) vs -0.016 for control). Aerobic plots had relative populations of ampicillin-resistant bacteria higher than anaerobic plots with different positive trends. Dose (22%) and time (13%) explained most of the variation of the bacterial populations. Dynamics of fecal markers did not correlate with ampicillin-resistant bacteria thus making necessary to evaluate specifically this trait to avoid possible risks for human and environmental health.

Lau, C. H., Li, B., Zhang, T., Tien, Y. C., Scott, A., Murray, R., Sabourin, L., Lapen, D. R., Duenk, P. and Topp, E. 2017. Impact of pre-application treatment on municipal sludge composition, soil dynamics of antibiotic resistance genes, and abundance of antibiotic-resistance genes on vegetables at harvest. *Sci Total Environ* 587-588:214-222.

In many jurisdictions sludge recovered from the sewage treatment process is a valued fertilizer for crop production. Pre-treatment of sewage sludge prior to land application offers the potential to abate enteric microorganisms that carry genes conferring resistance to antibiotics. Pre-treatment practices that accomplish this should have the desirable effect of reducing the risk of contamination of crops or adjacent water with antibiotic resistance genes carried in these materials. In the present study, we obtained municipal sludge that had been subjected to one of five treatments. There were, anaerobic-digestion or aerobic-digestion, in both instances with and without dewatering; and heat-treatment and pelletization. Each of the five types of biosolids was applied to an agricultural field at commercial rates, following which lettuce, carrots and radishes were planted. Based on qPCR, the estimated antibiotic gene loading rates were comparable with each of the five biosolids. However, the gene abundance in soil following application of the pelletized biosolids was anomalously lower than expected. Following application, the abundance of antibiotic resistance genes decreased in a generally coherent fashion, except *sul1* which increased in abundance during the growing season in the soil fertilized with pelletized biosolids. Based on qPCR and high throughput sequencing evidence for transfer of antibiotic resistance genes from the biosolids to the vegetables at harvest was weak. *Clostridia* were more abundant in soils receiving any of the biosolids except the pelletized. Overall, the behavior of antibiotic resistance genes in soils receiving aerobically or anaerobically-digested biosolids was consistent and coherent with previous studies. However, dynamics of antibiotic resistance genes in soils receiving the heat treated pelletized biosolids were very different, and the underlying mechanisms merit investigation.

Lopes, Thiara Reis, Periotto, Fernando and Pletsch, Adelmo Lowe. 2017. Bacterial resistance in sanitary sewage sludge in different treatment systems. *Management of Environmental Quality: An International Journal* 28(1): 32-42.

**Purpose:** The purpose of this paper is to assess the occurrence and risk of dispersion in the environment of antibiotic-resistant microorganisms from sanitary sewage sludge produced in two conventional wastewater treatment systems adopted in the West of the State of Paraná, Brazil.

**Design/methodology/approach:** The sludge samples were collected for three months from two wastewater treatment plants, totaling six sample points, and sent to the laboratory where the physical-chemical and biological determinations were performed.

**Findings:** This work made possible to find that the sludge produced in the sewage treatment plants presents potential risks related to the spread of microorganisms due to the occurrence of resistant isolates of *Escherichia coli* and *Salmonella* sp. It was also possible to detect that the largest concentrations of metal ions in the sludge favored the occurrence of bacterial resistance to antibiotics. The occurrence of pathogens, heavy metals and other emerging pollutants in sewage indicates that the sludge requires proper treatment, to provide safe agricultural reuse or disposal.

**Practical implications:** The techniques applied for monitoring sludge were effective to check the risk of resistant microorganisms input into the environment. Studies concerning sewage treatment plants' final effluents can bring additional data about the incorporation of such microorganisms into aquatic environments.

**Originality/value:** The results made possible to observe the need to provide post-treatment for the sludge, especially of the sludge obtained from the anaerobic fluidized bed reactor, since the removal of pathogens, as well as the nutrients, is not satisfactory.

Tozzoli, R., Bartolo, I. di, Gigliucci, F., Brambilla, G., Monini, M., Vignolo, E., Caprioli, A. and Morabito, S. 2017. Pathogenic *Escherichia coli* and enteric viruses in biosolids and related top soil improvers in Italy. *Journal of Applied Microbiology* 122(1):239-247.

**Aims:** To investigate the presence of genomic traits associated with a set of enteric viruses as well as pathogenic *Escherichia coli* in top soil improvers (TSI) from Italy.

**Methods and Results:** Twenty-four TSI samples originating from municipal sewage sludges, pig manure, green and household wastes were analysed by real time PCR for the presence of hepatitis E virus (HEV), porcine and human adenovirus (HuAdV), norovirus, rotavirus and diarrhoeagenic *E. coli*. None of the samples was found positive for HEV or rotavirus. Four samples were positive for the presence of nucleic acids from human norovirus, two of them being also positive for HuAdV. Real time PCR screening gave positive results for many of the virulence genes characteristic of diarrhoeagenic *E. coli* in 21 samples. These included the verocytotoxin-coding genes, in some cases associated with intimin-coding gene, and markers of enteroaggregative, enterotoxigenic and enteroinvasive *E. coli*.

**Conclusions:** These results provide evidence that enteric viruses and pathogenic *E. coli* may be released into the environment through the use of sludge-derived TSI.

**Significance and Impact of the Study:** The results highlight that the TSI-related environmental risk for the food chain should be more deeply assessed.