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A Foundation for Integrated Water and Species Policy: Wastewater Treatment Plant Effluent Overlaps With Wildlife in California Watersheds

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A Foundation for Integrated Water and Species Policy: Wastewater Treatment Plant Effluent Overlaps With Wildlife in California Watersheds

A Thesis submitted in partial satisfaction
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in

Evolution, Ecology, and Organismal Biology

by

Anna Peiming Cassady

March 2021

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This Thesis is dedicated to my family.

ABSTRACT OF THE THESIS

A Foundation for Integrated Water and Species Policy: Wastewater Treatment Plant Effluent Overlaps With Wildlife in California Watersheds

by

Anna Peiming Cassady

Master of Science, Graduate Program in Evolution, Ecology, and Organismal Biology
University of California, Riverside, March 2021
Dr. Helen Regan, Chairperson

The spread of human settlement has imperiled fresh waterbodies; however, it has also led to the generation of novel water conservation strategies, including the reuse of treated wastewater, or effluent. Effluent reuse is an increasingly common aspect of watershed management, and thus far, research has been concentrated on its effects to water quality and efforts to describe effects to wildlife species have been relatively piecemealed. In this study, we evaluate the overlap between wastewater treatment plants and federal and state-listed endangered and threatened wildlife species in order to present a holistic view of the intersection of effluent and species management and the potential need for effluent considerations in species conservation. We show that there is substantial overlap between the presence of species and the presence of treatment plants in California watersheds, and with this overlap, a large potential for unintended consequences. As such, species conservation goals should be considered when making decisions related to effluent reuse.

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Introduction

Freshwater streams and rivers are among the world's most imperiled ecosystems (Dudgeon et al. 2006; Reid et al. 2018a; Richter et al. 1997; Sala et al. 2000). These ecosystems and the species they support continue to be at the forefront of biodiversity decline across the planet (Almond, R.E.A., Grooten M. and Petersen 2020; Jenkins 2003; Reid et al. 2018a). The freshwater biodiversity index has experienced an 84% decline since 1970 (World Wildlife Fund 2020) and this loss is strongly correlated with various anthropogenic stressors (Dudgeon et al. 2006; Gleick 2000; Reid et al. 2018a) – of which the predominant consequence is altered hydrologic flow regime.

The hydrologic flow regime, hereafter referred to as "flow regime," is the variation in water discharge within a waterbody that results from seasonal and interannual changes within the water cycle and surrounding watershed (Poff et al. 1997; Poff and Zimmerman 2010). Flow regime can be characterized by water flow magnitude, frequency, duration, timing, and rate of change of the water supply (Poff and Ward 1989; Poff et al. 1997; Richter et al. 1996). Together, these characteristics drive various hydrologic phenomena that occur within freshwater systems, including differing size and intensity of flooding sequences, low-flow and drought events, and seasonal variability in stream flow. These phenomena are considered vital for the life-cycle regulation of riverine species (Bruckerhoff, Leasure, and Magoulick 2018; Leigh et al. 2016) and the creation of shifting, dynamic habitat types that bridge aquatic and terrestrial landscapes (Poff and Zimmerman 2010; Stanford, Lorang, and Hauer 2005). For example, some benthic macroinvertebrate communities rely on a fluctuating flow regime to mediate biological assembly processes such as the recolonization of a stream reach (Miller, Wooster, and Li 2007); fishes such as salmon return from the ocean to freshwater streams to spawn following surges in flow from fall and early winter rainfall (Brown, Moyle, and Yoshiyama 1994); amphibians begin their life cycles in relatively permanent waterbodies before undergoing a metamorphosis that allows them to disperse into adjacent wetland and terraced habitat (Holgerson et al. 2019); and riparian biota establishes along waterways as a function of the frequency and duration of flooding (Stromberg et al. 2007).

Flow regimes have been altered from their natural state (i.e., uninterrupted by human-mediated interventions) for millennia, beginning with irrigation by the earliest human civilizations (Harrower 2008). Industrialized human settlement has required extensive hydrological modification to meet various societal needs (Gleick 2000). Examples include groundwater pumping, waterway damming, flood attenuation, and installation of urban infrastructure (Gleick 2000). These modifications have a well-documented history of significantly altering the landscape such that native species are precluded from their habitats (Mount et al. 2019; Walsh et al. 2005). Flow modification alters aquatic habitat, but can also reduce or eliminate overbanking potential, leading to degraded riparian and terraced habitats (Poff et al. 1997; Walsh et al. 2005). Further implications have included the proliferation of “flashier” streams, eutrophication, and the restructuring of ecological communities (Walsh et al. 2005).

For example, flow regime modification in the form of flood control infrastructure (e.g., channelization, levees, water detention basins) can lead to higher-velocity flows that can increase scour and result in more destructive flooding events (Liao 2014; Williams 1990). Additional ecosystem impacts range from the invasion of non-native fishes that result from the human-mediated stabilization of flow (Comte, Grantham, and Ruhi 2021), to the proliferation of invasive, stress-adapted *Tamarix* vegetation in regulated and flood-suppressed waterways (Stromberg et al. 2007), to reduced downstream sediment transport which can have secondary effects on species such as the Santa Ana sucker that relies on a specific substrate composition for various stages of its life cycle (Wright and Minear 2019).

While the spread of human settlement has imperiled fresh waterbodies, it has also led to the generation of novel water conservation strategies, including the reuse of treated wastewater (Gleick 2000; Hamdhani, Eppehimer, and Bogan 2020; National Research Council 2002; Olivieri et al. 2014). During the first half of the 20th century in the United States, human waste and wastewater historically percolated into the ground via underground cesspools and septic tanks before concern over local groundwater supplies facilitated the disposal of untreated discharge directly into nearby waterbodies (National Research Council 2002). As this method of disposal was also a detriment to human health, rudimentary treatment processes were developed to filter the wastewater. The passing of the Clean Water Act in 1972 further codified the need for wastewater treatment processes and government subsidies aided the creation and use of wastewater treatment plants in urbanized locations throughout the United States (National Research Council 2002). Following continued advancement of purification technology and overall freshwater scarcity in many urbanized regions, treated wastewater, hereafter referred to as “effluent,” has been utilized as an inexpensive alternative to naturally occurring freshwater (e.g., water from reservoirs, rivers, groundwater). As such, effluent replaces freshwater in a growing number of utilities, including agricultural irrigation, landscape irrigation, and potable water generation (Hamdhani et al. 2020; Olivieri et al. 2014). Effluent is also used to aid in groundwater recharge and surplus is discharged into oceans and streams – effectively supplementing flow in regions that experience anthropogenic flow withdrawal (Hamdhani et al. 2020).

When evaluating water withdrawal and flow regime modification, effluent reuse is an increasingly common aspect of watershed management and therefore has been the subject of numerous research studies that evaluate its effects. A recent review of literature on effluent-fed streams found that 85% of the 147 evaluated studies identified water quality as a major focus (Hamdhani et al. 2020). Water quality investigations primarily focus on chemical, physical, and biological components such as the presence of pathogens (Sanders, Yuan, and Pitchford 2013), pharmaceutical compounds (Brozinski et al. 2013), endocrine disruptors (Vajda et al. 2011), temperature (Kinouchi, Yagi, and Miyamoto 2007), particulate matter (Sánchez-Morales, Sabater, and Muñoz 2018), eutrophication (Martí, Riera, and Sabater 2010), and salinity (Herbert et al. 2015). Effluent-fed streams containing endocrine-disrupting compounds have been shown to cause fish demasculinization (Vajda et al. 2011), which in turn has also been shown to limit reproductive success and lead to long-term population decline (Harris et al. 2011). Another study found that an increase in particulate material stemming from treatment plants (e.g., “clogging”) leads to an overall decrease in species biodiversity in impacted waterbodies (Sánchez-Morales et al. 2018).

Another major emphasis within effluent literature is the direct ecological impact to various taxonomic groups, with nearly all literature dedicated to aquatic-related taxa (Hamdhani et al. 2020). Excessive eutrophication has also been documented in effluent-fed streams, and treatment plants are estimated to contribute over 50% of nitrogen and phosphorous concentrations within the waterbody (Martí et al. 2010), a phenomenon that has been shown to restructure freshwater communities towards more tolerant or invasive taxa (Bellinger, Cocquyt, and O’Reilly 2006; Dunck et al. 2015; Ota et al. In revision) through the removal of nutrient constraints that naturally lead to species heterogeneity (Cook et al. 2018). Eutrophication within effluent-fed streams has also been observed leading to increased production of algal basal resources, including filamentous red algae (Murdock, Roelke, and Gelwick 2004). Aquatic invertebrate diversity has been observed decreasing after exposure to effluent (Grantham et al. 2012), again with emphasis placed on common, tolerant taxa that exploit the higher nutrient density at outfall structures (Arnon, Avni, and Gafny 2015). Fishes at treatment plant outfalls have been observed at higher abundance, likely due to increased production of basal resources, but species composition again tends to favor common and non-native species (McCallum et al. 2019).

Despite the focus on negative effects of effluent, recent research has emerged documenting positive, and perhaps unforeseen, impacts as well. In highly modified hydrologic settings, such as the arid western region of the United States, effluent has inadvertently served as substitute aquatic habitat in historically intermittent or perennial river systems that have had their baseflows diverted for anthropogenic purposes. The Santa Cruz River in southern Arizona is an example of a river that historically supported perennial reaches and in the present-day experiences staggering anthropogenic water withdrawals that have caused groundwater levels to drop and the river to flow ephemerally. Studies of this river have shown that wetted portions of the river subsisting on effluent discharge have returned to their perennial flow regime and are the only sections of the river to support dense riparian woodlands and a rich biodiversity of plants and animals (Boyle and Fraleigh 2003; Eppehimer et al. 2020; Stromberg et al. 1993). Effluent-fed rivers have supported rapid colonization and a high diversity of Odonata (dragonfly and damselfly) species, suggesting that effluent can serve as an important habitat source in urbanized landscapes (Bogan et al. 2020). In the Santa Ana River in southern California, a river that also experiences extensive human-mediated water extraction, treatment plant effluent has been evaluated for its role in creating variable species compositions in the urbanized reaches (Saffarinia et al. In review). In this same river system, non-native, generalist fish species showed differences in their trophic niches across a gradient of effluent input (Ota et al. In revision).

The use of effluent to create aquatic habitat by replacing anthropogenic water withdrawal can also result in unintended consequences during periods of water scarcity. For example, government-mediated water conservation efforts during California’s most recent drought from 2013 to 2017 led to reduced water intake to treatment plants (e.g., influent), leading to less available water for reuse (Tran, Jassby, and Schwabe 2017), higher concentrations of total dissolved solids in discharged water, and overall less discharge released into streams (Schwabe et al. 2020). Treatment plant effluent has also been documented as comprising greater than 50% of intermittent stream discharge during low-flow and drought conditions, which has been shown to magnify both nutrient and salinity concentrations (Hur et al. 2007).

Thus far, research into the unintended consequences of effluent use has been concentrated on its effects to water quality. However, given the growing body of research exemplifying effluent serving an additional role as habitat in hydrologically modified systems, we posit that there may be further unintended consequences in relation to biodiversity. The ecological effect of effluent is a growing field of study that has presented a patchwork of information at the waterbody and species or taxa level. The role of effluent within a hydrologic setting is inherently challenging to study at broader landscape scales because hydrologic modification is not uniformly applied across a landscape and effluent’s contribution to a waterbody is interdependent with other types of hydrologic modification, abiotic conditions of the landscape, and other management actions in the region. Water management priorities are often in conflict with one another (Mount et al. 2019), with agencies at the local, state, and federal levels that have differing objectives and responsibilities. However, despite these challenges, we feel it is important to attempt to identify patterns at a broader spatial scale in order to better understand the interdependence between effluent and species, to close existing knowledge gaps with a goal of lending support to effluent management decisions that could have far-reaching consequences for biodiversity.

Here, we aim to evaluate the overlap between wastewater treatment plants and wildlife species in order to present a holistic view of the intersection of effluent and species management and the potential need for effluent considerations in species conservation. We also intend to assess the permitted discharge of wastewater treatment plants (i.e., the amount of treated wastewater that treatment plants can release into waterbodies per the conditions of their legal permits) in relation to the recorded baseflow discharge of their receiving waterbodies. The intention of this analysis is to provide an overview of the contribution of effluent—an artificially created water source—to the baseflow quantities of the receiving waterbody. These analyses were conducted at the watershed level within the state of California, where freshwater biodiversity loss is particularly pervasive due to high levels of species endemism, widespread land conversion, increasing natural disturbances, and extensive hydrologic modification concomitant with rapid human population growth in the region (Grantham et al. 2017; Moyle, Katz, and Quiñones 2011; Moyle and Williams 1990). Analysis was confined to wildlife species listed under the federal or California Endangered Species Act. These species are already imperiled and exposed to myriad other threats; however, they are also the most visible from a species conservation perspective and would experience the greatest influence from water management decisions.

In this analysis, we approach the question of whether effluent is beneficial or detrimental from a position of neutrality; rather, we aim to demonstrate that the influence of effluent is a variable that should be considered by water managers and species conservation managers during decision-making processes. When not considered, we will show that there is a large potential for unintended consequences, particularly in urban settings where our analyses show effluent discharge overlaps with endangered and threatened species habitat.

Methods

Data Collection

a) Species Data

Wildlife species occurrence data was obtained from the California Natural Diversity Database (CNDDB), administered by the California Department of Fish and Wildlife (CDFW 2019). This dataset returned all species' spatial occurrences that had been inventoried in the database through January 5, 2019. Wildlife species returned from CNDDB were selected if, at the time of the dataset download, they were listed by the federal Endangered Species Act as "Endangered," "Threatened," "Candidate," "Proposed Threatened," or "Proposed Endangered." Analogous categories were used for species listed by the California Endangered Species Act. Hereafter, all federal- and state-listed endangered, threatened, candidate, proposed endangered, and proposed threatened wildlife species will be referred to as "species."

Each species was categorized by the floodplain habitat type they are most associated with, if applicable. For the purposes of this analysis, a floodplain is defined as all habitat types associated with a watercourse that rely on some frequency of flooding. Each species was associated with one of the following floodplain habitat types: aquatic habitat (meaning the species spends some portion of its life cycle submerged in water, e.g., fishes and amphibians); aquatic emergent vegetation (e.g., wetlands); riparian vegetation; terraces (e.g., alluvial fans, marine terraces); or uplands (meaning that the species is not associated with floodplains in any way). Within each habitat type, a frequency of use was determined: each species was classified as an obligate user (e.g., they are only found in this habitat) or a facultative user (e.g., they are equally found in this habitat, but may be found in other habitats as well). The habitat associations for each species were determined by a review of available literature, including resource agency management plans and species reviews, as available, as well as other reputable natural history websites for more obscure species. Further analysis was conducted only with floodplain-associated species that were either obligate or facultative users. Finally, while much of the associated discussion focuses on freshwater bodies, California supports marine species and wastewater treatment plants because it is a coastal state. These species and treatment plants were included within the following analyses in order to present a full picture of species and wastewater management within the state.

b) Watershed Data

All watershed analyses were conducted using the boundaries of the eight-digit Hydrologic Unit Codes (HUC 8), as defined by U.S. Geologic Survey (USGS). The HUC 8 is a component of the Watershed Boundary Dataset that captures the spatial extent of surface water drainage within the United States. The more digits of HUC (ranging from 2-12), the more refined the drainage area becomes. HUC 8 is referred to as a sub-basin and is generally the accepted watershed unit used by land management practitioners. The watershed data provided by the USGS is attributed to the California ecoregion (Region 18) and contained area that extends outside of the California political boundary. Because species and wastewater treatment plant data were confined to the political boundary, the watersheds were clipped to the political boundary for consistency purposes.

c) Wastewater Treatment Plant Data

Information on wastewater treatment plants was obtained from the California State Water Resources Control Board "Interactive Regulated Facilities Report" tool (SWRCB 2019). This tool queried each wastewater treatment facility with active permits within the state of California. From this output, we analyzed all facilities that possessed a National Pollutant Discharge Elimination System (NPDES) permit. The NPDES is a federal program that regulates the discharge of wastewater to waters of the United States (i.e., the legal definition for waterways that are federally regulated). Other wastewater treatment facilities regulated under other permitting structures were excluded because those treatment plants discharged to land-based resources and were not considered applicable to the research question. The list of wastewater treatment plants contained locations for most facilities; however, a portion were missing and were manually added using information in the public record. Two treatment plants were excluded from the analysis given the lack of locational data available. An additional 13 treatment plants were excluded because they were determined to be spatial duplicates (i.e., two data points for the same geographic location). A total of 270 treatment plants were used in the analysis. Finally, the list of treatment plants contained the associated permitted maximum discharge amount for each treatment plant based on their individual NPDES permits. For treatment plants where this data was missing, the permitted discharge amount was manually added through review of the individual NPDES permit. It is important to note that, in relation to the subsequent analysis, a treatment plant's permitted discharge and its true discharge (i.e., the real quantity released into waterways) are different; however, the latter values are difficult to obtain because they are not compiled by the state. With this being said, use of the permitted discharge still provides insight into the potential dominance of effluent.

d) Flow Data

In order to estimate the proportion of discharge potentially attributed to wastewater treatment plant outflow, water discharge data for the receiving streams of wastewater treatment plants was collected using the "StreamNetworkTools" R package (Kopp 2018). This R package streamlines the collection of covariates from the NHDPlus V2 dataset via the input of geographic coordinates. For this analysis, the geographic coordinates of all wastewater treatment plants were used to identify the nearest waterbody at a maximum distance of 1,500 meters. Ten wastewater treatment plants were excluded from this portion of the analysis because they were further than 1,500 meters from the receiving waterbody. Flow covariates were then collected for each waterbody. Covariates collected included cumulative mean annual runoff, minimum mean monthly discharge, maximum mean monthly discharge, coefficient of variation of mean monthly discharge, mean annual velocity, minimum mean monthly velocity, maximum mean monthly velocity, and coefficient of variation in mean monthly velocity. Flow covariates were available for a subset (181 of 270) of the receiving waterbodies attributed to the treatment plants and therefore the ensuing analysis was on this subset only. Out of 72 total watersheds that contain treatment plants, 38 had flow covariate data for every treatment plant. A total of

29 watersheds had partial representation of flow covariate data for their treatment plants, and five watersheds were not represented with flow covariate data. The remaining 68 watersheds do not contain treatment plants. All discharge data underwent a ($\log_{10}$+1) transformation for data visualization purposes.

Data Analysis

The CNDDB functions as a data repository for rare species and has limitations to its utilization. Species absences are rarely noted in the database, so the occurrence data was used solely as presence-only data. Due to the propensity to survey at higher frequencies in urban and urban-adjacent settings, the data was used to document species presence and not species abundance. Finally, given the disproportionate precision of spatial accuracy between species occurrences in the database, each species polygon was reduced to its centroid for consistent analysis. These limitations meant the species data was obtained and presented at a coarse resolution; however, when analyzed at a broader spatial scale, such as at the HUC 8 watershed-level or state-level, patterns can be analyzed.

a) Wastewater Treatment Plant and Species Densities

The software ArcMap 10 was used to organize and evaluate the spatial data. Each watershed was attributed with the number of unique floodplain-associated species and wastewater treatment plants contained within its boundary. The relationship between the density of wastewater treatment plants and density of floodplain-associated species within each watershed was plotted and evaluated using Spearman's rank correlation coefficient (ρ), which was determined to be the appropriate statistic due to its ability to appraise nonparametric relationships. For aquatic species, density was calculated using the area of NHD watercourse polygons as a coarse proxy for waterway area within each watershed. For other species, density was calculated using the total watershed area. Species and wastewater treatment plant density was used for the analysis in order to account for watersheds having a large variation in size. Density data underwent a $\log_{10}$ transformation for data visualization purposes. Density was the best way to control for variation in area among watersheds, however, we also evaluated the unadulterated numbers, which showed the same pattern.

b) Species Range

Each species' range was quantified in relation to the presence of wastewater treatment plants within the state of California. For the purposes of this analysis, the species' range was defined as the watersheds in California that contained a positive occurrence of each individual species. For example, if least Bell's vireo (*Vireo bellii pusillus*) was documented as present in 44 of 140 total HUC 8 watersheds, least Bell's vireo's "range" was mapped as the extent of these watersheds. This extent was overlain with the extent of wastewater treatment plants. Similarly, if 33 of the 44 watersheds containing least Bell's vireo also contained a wastewater treatment plant, 75% of the species' range overlaps effluent-fed watersheds. This analysis was conducted for each of the species and organized visually.

c) Flow Analysis

The wastewater treatment plant data was downloaded using the "Interactive Regulated Facilities Report" included the maximum design flow discharge in the unit "million gallons per day." The maximum permitted discharge was then compared with the baseflow discharge quantities of the receiving waterbodies, as determined through the "StreamNetworkTools" R package. Bar plots were used to visually represent the difference between the regulated discharge quantities and the receiving waterbody baseflow discharge quantities. Wastewater treatment plants were then collapsed to their respective watershed and the median value of the flow covariates was attributed to each watershed. The same bar plots were created to evaluate differences at the watershed level.

Results

The state of California contains 140 HUC-8 watersheds and 285 wastewater treatment plant facilities that discharge into waterways, of which 270 were included in the final analysis. These facilities are contained within 72 (51%) of California's watersheds. As indicated in Figure 1, the majority of wastewater treatment plants are clustered in areas disturbed either by urbanization or agricultural production, such as the Los Angeles Basin and Inland Valleys of southern California, the San Francisco Bay Area in northern California, and the Central Valley region. A total of 157 threatened and endangered wildlife species have been documented within all of California's watersheds. The spatial distributions of the watersheds, treatment plants, and species are depicted on Figure 1. While Figure 1 depicts the full spatial distribution of the species occurrences per the CNDDB, each polygon was reduced to its centroid for the ensuing analysis.

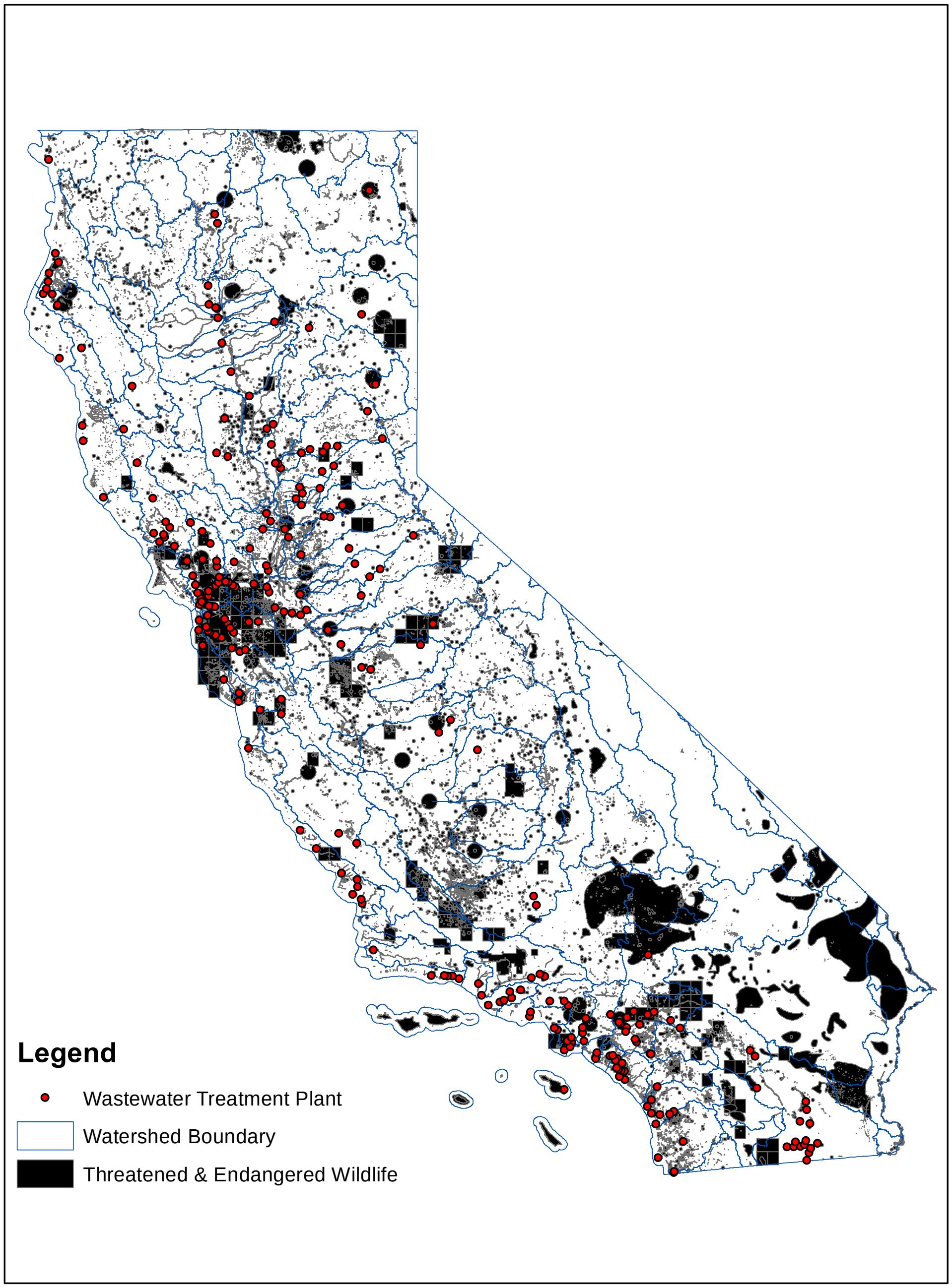

Figure 1: The spatial distribution of wastewater treatment plants, threatened and endangered wildlife species polygons, and HUC-8 watersheds within California.

Fifty-six percent of all threatened and endangered wildlife species in California were identified as obligates to floodplain habitats (e.g., aquatic habitat, aquatic emergent vegetation, riparian vegetation; terraces) – meaning that these species will almost always be found in their respective floodplain habitats. An additional 11% of these species are associated with floodplain habitats in a facultative capacity, meaning that they can be found in floodplain habitats, but also in upland habitats. In total, we found that 67% of species are associated with floodplains in some capacity. Obligate freshwater species make up the largest proportion of threatened and endangered species, comprising 31% of the total. A further breakdown of the different floodplain habitats associated with species is provided in Table 1. Endangered and threatened floodplain-associated species were found in 138 of 140 (99%) total watersheds and in all watersheds that also contain wastewater treatment plants.

Table 1: Floodplain Use Attributes

Floodplain Habitat & Frequency of Use	Number of Species	Percent of Total
Aquatic		
Obligate - Marine	6	3.8
Obligate - Freshwater	48	30.6
Facultative	3	1.9
Aquatic Vegetation		
Obligate	12	7.6
Facultative	1	0.6
Riparian Vegetation		
Obligate	15	9.6
Facultative	3	1.9
T***erraces (Marine)***		
Obligate	3	1.9
Facultative	3	1.9
Terraces (Freshwater)		
Obligate	4	2.5
Facultative	7	4.5
Floodplain Subtotal	***105***	***66.9***
Upland		
Obligate	44	28.0
Seasonal pools	8	5.1
Upland Subtotal	***52***	***33.1***
Grand Total*	**157**	**100**

* Note: totals may not add due to rounding.

Species Range

There are many species that have high proportions of their ranges overlapping watersheds that contain treatment plant effluent. As shown in Table 1, a total of 105 species are associated with floodplain habitats. A total of 80% of these species have some proportion of their range overlapping watersheds containing wastewater treatment plants, further broken down to 25% (26 species) at 100% of their range; 38% (66 species) at over

50%, but less than 100%; and 17% (18 species) at greater than 0%, but less than 50%. A plot exemplifying the range information is provide as Figure 2. Breaking this down by habitat type, 72% of aquatic-associated species, 92% of aquatic emergent vegetation species, 78% of riparian species, and 100% of terrace species have a proportion of their range overlapping watersheds with treatment plant effluent, as visualized in Figure 3.

In contrast, only 20% (21 species) do not have any part of their range overlapping watersheds that contain wastewater treatment plants. In general, these species are extremely range-restricted and tend to occur in more remote locations such as the Mojave Desert or Sierra Nevada mountain range, where generally there is little urbanization and therefore no municipal need for wastewater treatment processes.

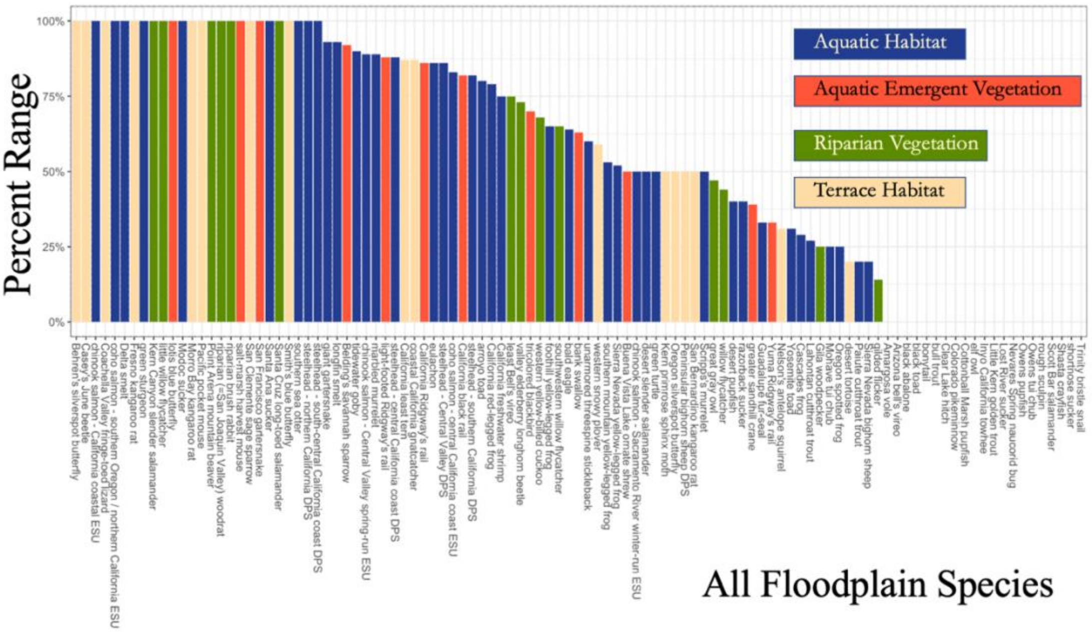

Figure 2: The proportion of each floodplain-associated species' watershed range that also contains wastewater treatment plants. Each species is color-coded based on its floodplain habitat use. Each habitat type is equally represented within the graph, indicating that there is high overlap of treatment plant effluent with each floodplain habitat type.

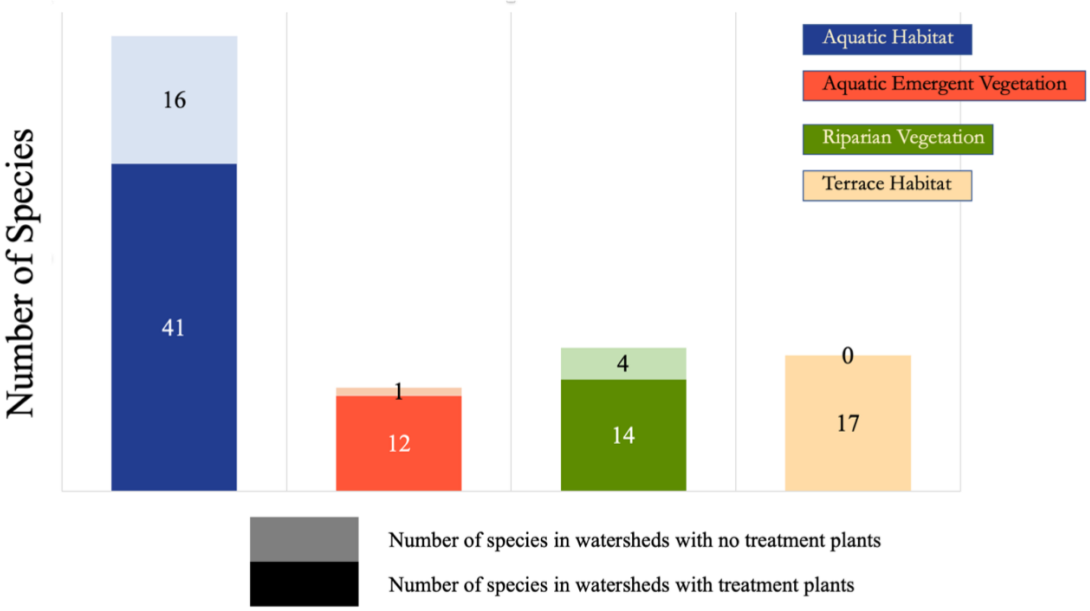

Figure 3: The proportion of species for each floodplain habitat type that have ranges overlapping watersheds with treatment plants. With every habitat type, the majority of species overlap watersheds with treatment plants.

Wastewater Treatment Plants and Species Densities

There is high overlap between species and treatment plants in California watersheds. The density of wastewater treatment plants in watersheds is positively correlated with the density of species that use floodplain habitats ($\rho = 0.45$, $p = 2.5\text{x}10^{-8}$; Figure 4). This correlation is observed most strongly with species associated with terraces ($\rho = 0.57$, $p = 2.5\text{x}10^{-5}$) and riparian-associated habitat ($\rho = 0.51$, $p = 7.9\text{x}10^{-8}$), followed by aquatic habitat ($\rho = 0.41$, $p = 1.2\text{x}10^{-6}$) and aquatic emergent vegetation ($\rho = 0.41$, $p = 8.9\text{x}10^{-6}$; Figure 5). While the relationships presented somewhat linearly, they each contained outliers typically in the form of watersheds that contain no treatment plants or watersheds with smaller land area which in turn inflated the density calculations. Plots exemplifying these relationships are provided as Figures 4 and Figure 5. Finally, a plot of the unadulterated number of treatment plants and species for each watershed as Figure 6.

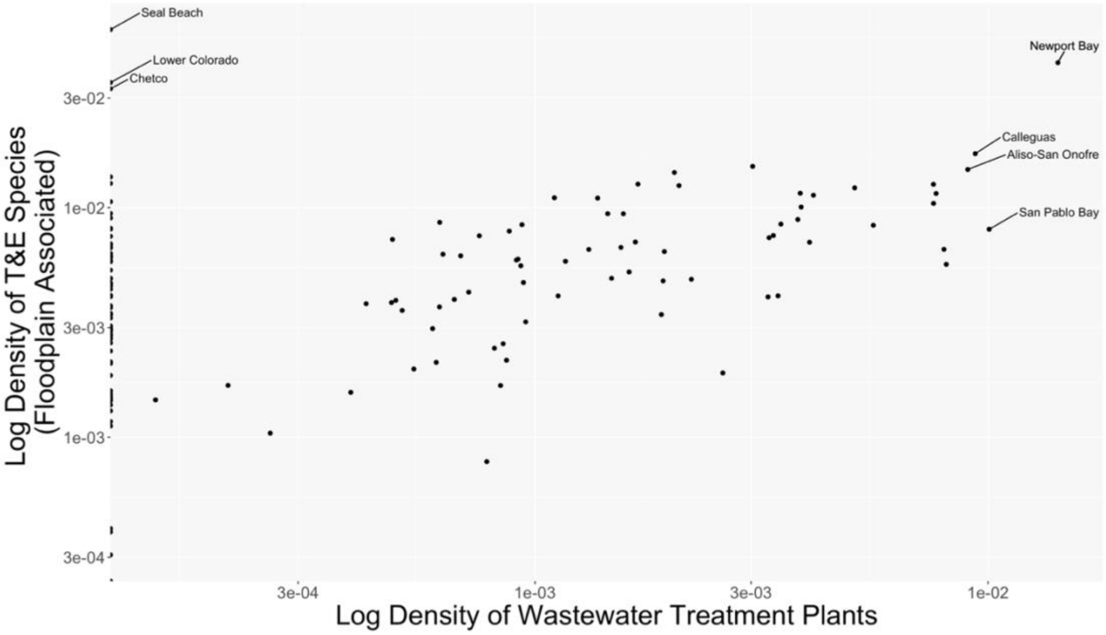

Figure 4: The relationship between the density of wastewater treatment plants and density of floodplain-associated species per watershed is positively correlated ($\rho = 0.45$, $p = 2.5x10^{-8}$).

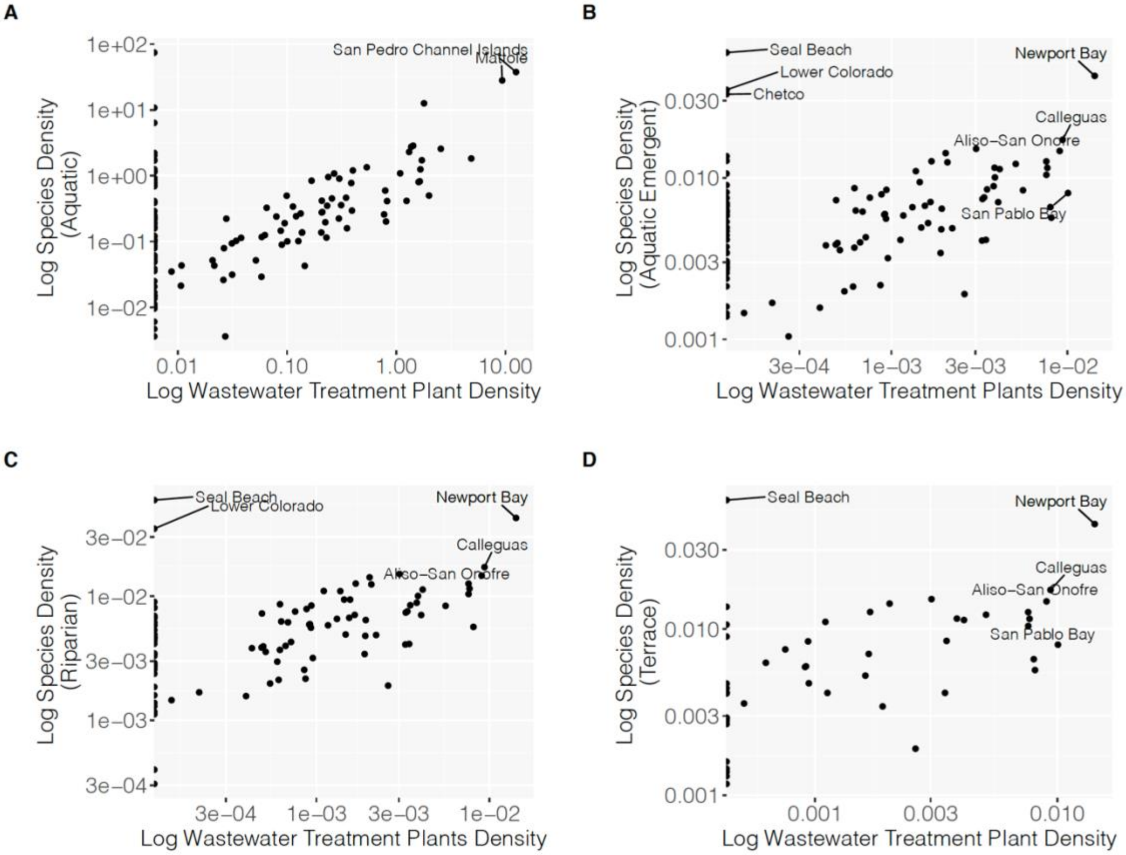

Figure 5: The relationship between the density of wastewater treatment plants and density of habitat-specific floodplain-associated species per watershed is positively correlated. A) $\rho= 0.41$, $p = 1.2 \times 10^{-6}$; B) $\rho= 0.41$, $p = 8.9 \times 10^{-6}$; C) $\rho= 0.51$, $p = 7.9 \times 10^{-8}$; D) $\rho= 0.57$, $p = 2.5 \times 10^{-5}$

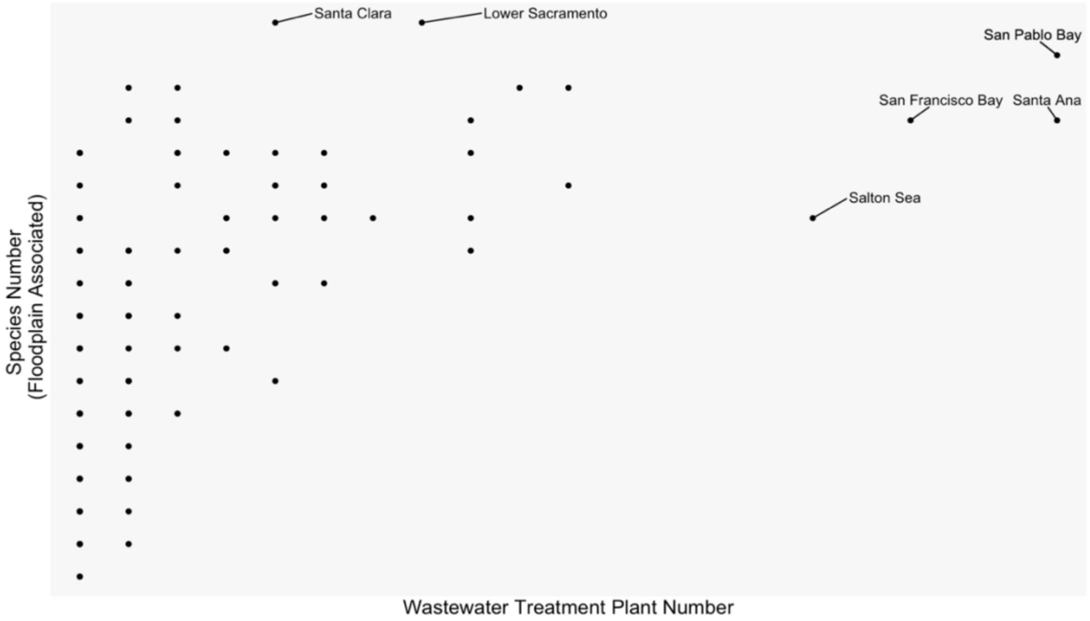

Figure 6: The relationship between the number of wastewater treatment plants and the number of floodplain-associated species is positively correlated (ρ= 0.64, p = 2.2×10^{-16}). The watersheds with the highest numbers of each resource are located in areas that have experienced urbanization or agricultural driven land conversion.

Flow Analysis

When evaluating the permitted treatment plant outflow in relation to the receiving waterbody baseflow, we found that there are many treatment plants and watersheds that have the potential to receive the majority of their cumulative watershed baseflow from effluent. Figure 7 depicts watersheds in the state that are receiving higher permitted treatment plant outflow compared to baseflow discharge. When comparing the maximum regulated discharge to the mean annual discharge of the receiving water, 83 out of 181 (46%) treatment plants and 23 out of 67 (34%) watersheds documented higher potential outflow discharge than the receiving waterbody baseflow. When comparing to minimum monthly mean discharge, 140 (77%) treatment plants and 46 (69%) watersheds documented higher permitted outflow discharge than the baseflow of its receiving waterbody. Finally, when comparing maximum monthly mean discharge, 60 (33%) treatment plants and 12 (18%) watersheds documented higher permitted outflow discharge than the receiving waterbody baseflow. It is important to note that the documented differences at the watershed level are considered an underestimate due to the lack of available data for all treatment plants within 29 watersheds. Bar plots exemplifying the relationships between maximum regulated discharge and receiving waterbody baseflow are provided as Figures 8 and 9.

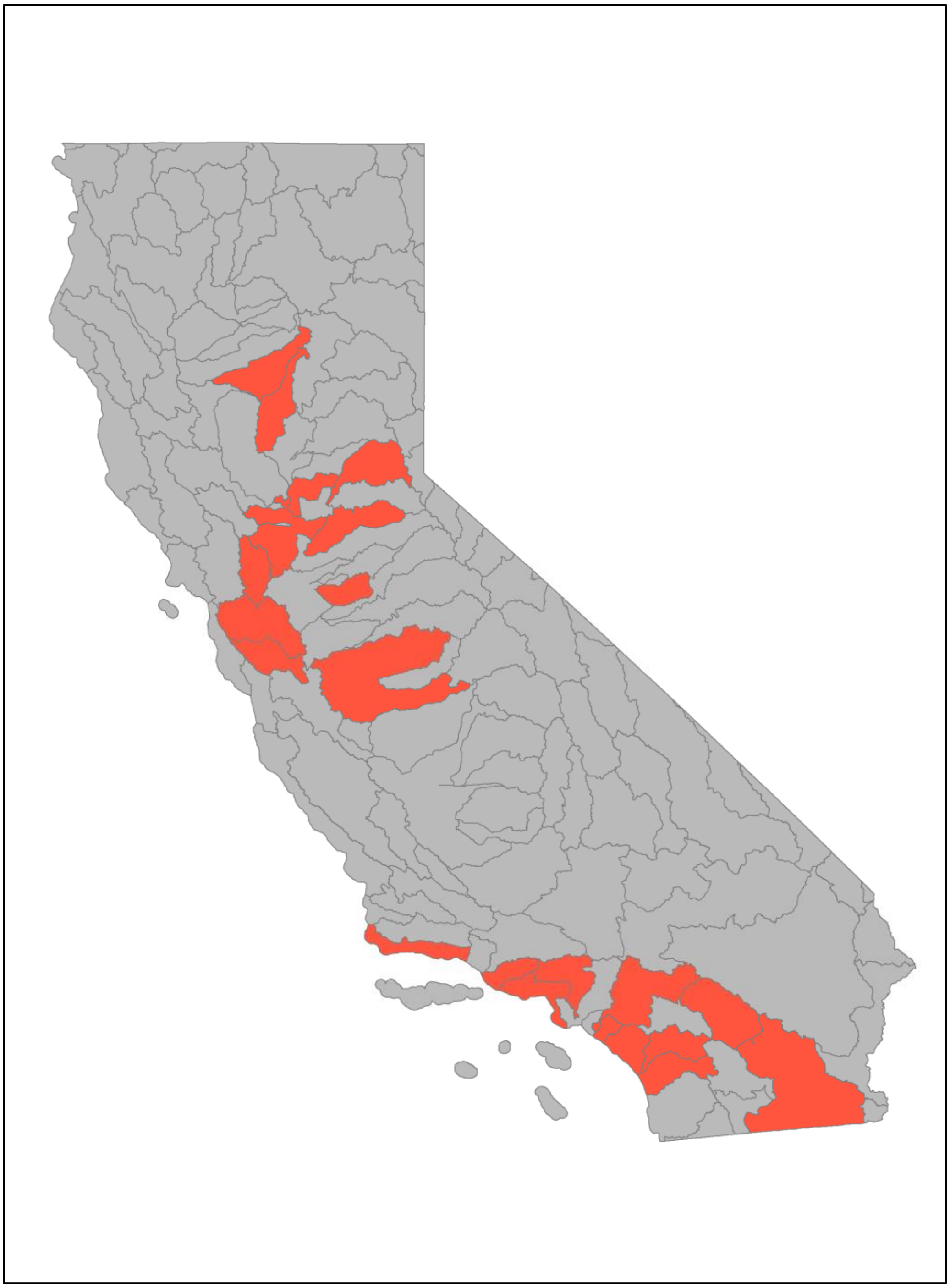

Figure 7: California watersheds that cumulatively receiving higher discharge from effluent than the underlying baseflow.

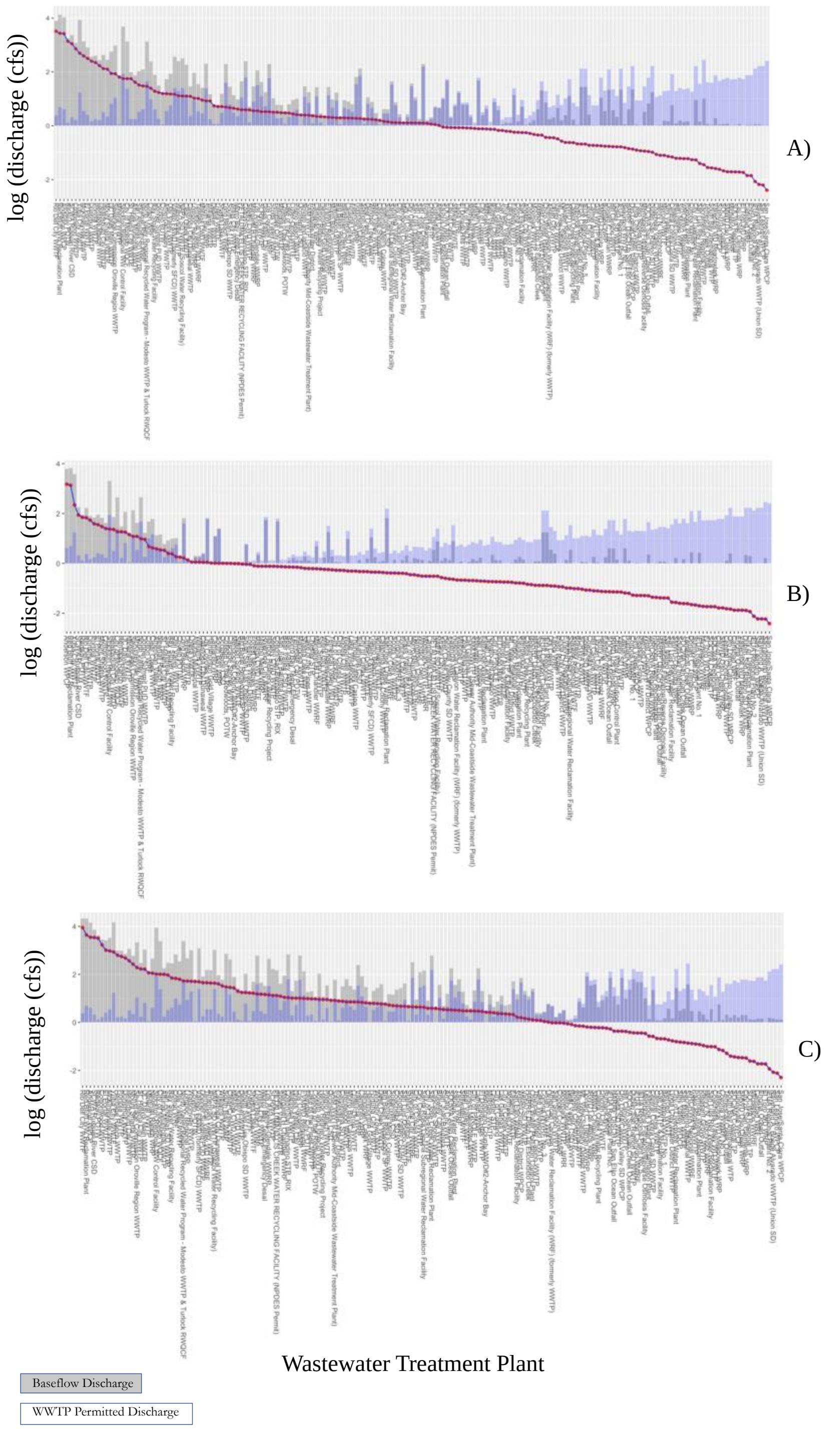

Figure 8: A) wastewater treatment plant permitted discharge compared with mean annual baseflow discharge; B) wastewater treatment plant permitted discharge compared with minimum monthly mean baseflow discharge; C) wastewater treatment plant permitted discharge compared with maximum monthly mean baseflow discharge.

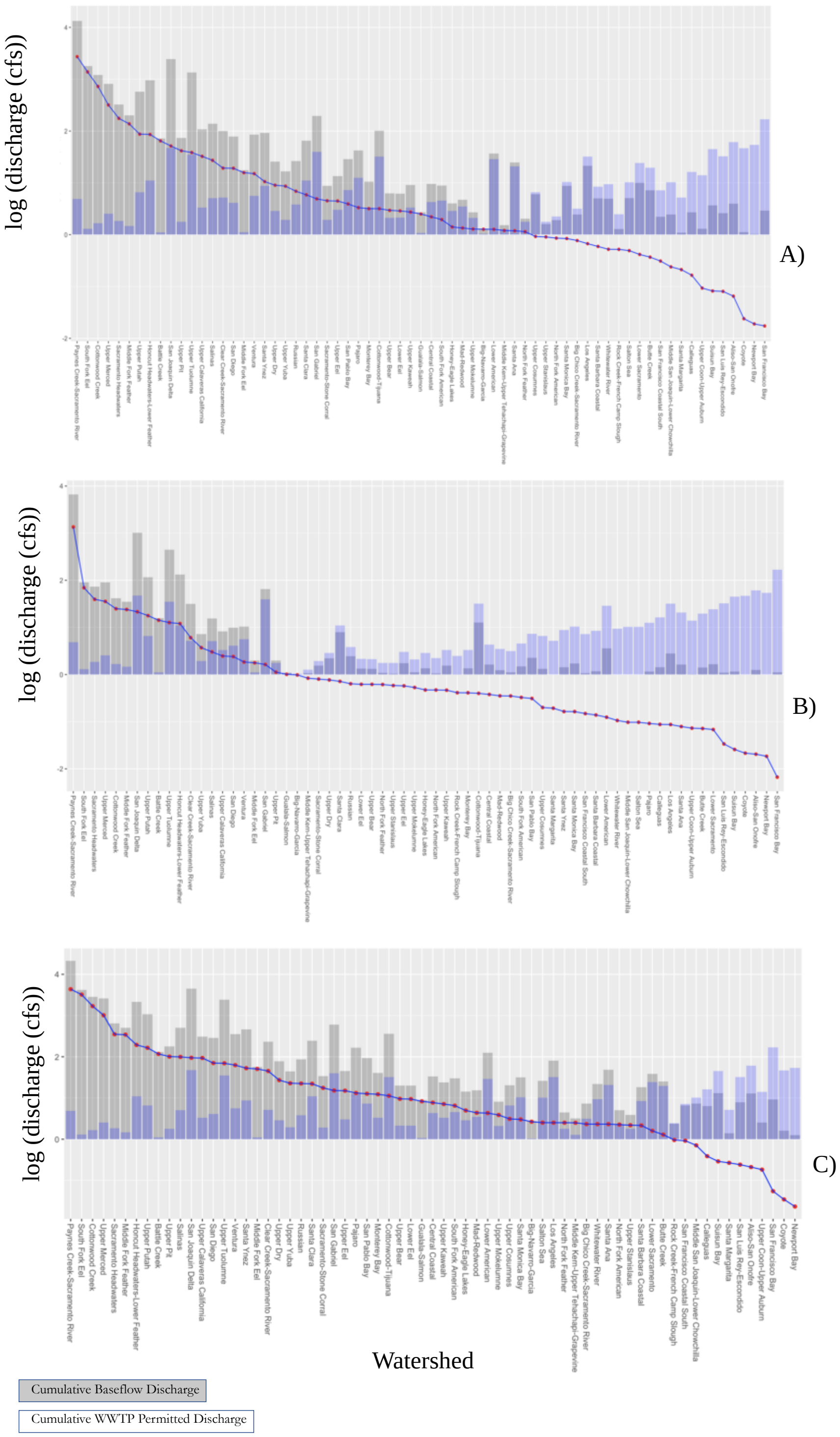

Figure 9: A) cumulative permitted watershed discharge compared with mean annual baseflow discharge; B) cumulative permitted watershed discharge compared with minimum monthly mean baseflow discharge; C) cumulative permitted watershed discharge compared with maximum monthly mean baseflow discharge.

Discussion

The overview presented in this paper is intended to illuminate and acknowledge the interdependence of wastewater treatment plant effluent and endangered and threatened wildlife species. Wastewater treatment plants were born out of the need to protect water quality and prevent sewage from contaminating waterways. However, this resource has been utilized to service numerous municipal needs (i.e., irrigation, drinking water, groundwater recharge) and biological needs (i.e., stream and river recharge) in areas that experience water scarcity. These needs often come into direct conflict with one another (Poff et al. 1997), and therefore it is necessary to evaluate potential unforeseen consequences. Documenting the degree of overlap between effluent dominance and species of concern is an important first step in mitigating potential conflicts.

Freshwater biodiversity is imperiled globally (Reid et al. 2018b) and we confirmed that this generalization also extends throughout the state of California. We found that the majority (67%) of species protected under either the United States Endangered Species Act (16 U.S.C. §1531 et seq.) or the California Endangered Species Act (F.G.C § 2050 - 2089.25) are associated with floodplain habitats, which we defined as comprising aquatic habitat, aquatic emergent vegetation, riparian vegetation, or terraces. We also found that there is a positive correlation between the presence of treatment plants and presence of species in California watersheds.

The purpose of this study is not to demonstrate a mechanism driving the correlation between the presence of treatment plants and species. In all reality, each variable is likely jointly responding to multiple drivers — land conversion, urbanization, flood control, to name some examples. As shown in Figure 1, the majority of wastewater treatment plants are assembled in areas that experience disproportionate levels of hydrologic modification either as a function of urbanization or agricultural production, including the Los Angeles Basin and Inland Valleys of southern California, the San Francisco Bay Area in northern California, and the Central Valley region. These areas are also likely to see the most complex interplay between water and species management objectives due to overlapping and conflicting water resource uses. This is further exemplified through our finding that there are many species that have high proportions of their range overlapping watersheds fed by treatment plant effluent. Through our analysis, we have shown that the fates of these two resources, treatment plant effluent and species, overlap greatly in their distributions and are therefore now inextricably linked.

In one example of management actions in California leading to unforeseen consequences from treatment plant discharge (Schwabe et al. 2020), water conservation efforts were enacted by the state legislature to curtail deleterious effects caused by drought. While these efforts did offset substantial water shortages exacerbated by drought, they also resulted in a lower volume of water going through the treatment process and discharging into rivers and streams. This caused extreme low flow events and high salinity concentrations within receiving waterbodies (Schwabe et al. 2020). Increases in salinity concentration not only reduces water quality, but can also have documented harmful effects on wildlife (Patnode et al. 2015). Based on our analysis in this paper, we can further suggest a reduction in effluent discharge in urbanized locations could lead to the degradation of habitat for species that have become accustomed to the presence of effluent as habitat.

In our study, we found that there are many species that have high proportions of their range overlapping watersheds fed by treatment plant effluent and each habitat type generally showed the same high overlap, indicating that all floodplain components have the potential for being influenced by treatment plant effluent. However, these habitat types and their associated species are likely to respond to effluent differently. Aquatic, in-stream species are expected to experience the most direct effects from effluent, particularly in regions with anthropogenic water withdrawal where effluent is comprising some of the only available aquatic habitat. For these species, variation in effluent discharge would also be expected to occur on the shortest time interval, as the variation can instantaneously translate to availability of aquatic habitat. Riparian and aquatic emergent vegetation species would also experience effects from effluent presence or absence, albeit at longer time intervals due to a lag effect from the time it would take for the habitat to respond to inundation or desiccation. Effluent has been documented leading to increased recruitment of riparian species (Stromberg et al. 1993) and anecdotally treatment plants can be marked by the expanse of riparian woodlands that extend downstream of the outfall structure. Terraces are formed on the longest time interval, as this habitat type is sustained by periodic flood events that occur on longer climactic cycle. While some effect to terrace species is expected from the presence or absence of effluent, this effect is expected to be limited in relation to larger impacts from flood control actions that attenuate the large flooding events that this habitat type relies on (Chock et al. 2020; 63 FR 51005).

While range overlaps did not generally differentiate among species based on habitat type, we did observe some differences in the degree that the respective densities of species were positively correlated with treatment plant densities in California watersheds. Highest correlations were recorded for terrace and riparian associated species, followed by aquatic and then aquatic emergent vegetation species. The association with aquatic species such as fishes perhaps makes the most intuitive sense. The dispersal pathway of aquatic species is strictly confined to aquatic habitats, as opposed to a bird or terrestrial mammal that can disperse via air or land through a patchy habitat mosaic in pursuit of suitable habitat for important life history functions. A positive correlation between aquatic-associated species with treatment plant abundance suggests that treatment plants could be providing aquatic habitat in areas that experience extensive hydrologic modification and have limited natural baseflow. We have seen this phenomenon occur at the local level in the Santa Ana River in southern California where, unbeknownst to conservation managers and treatment plant operators alike, periodic treatment plant shutoffs for routine maintenance were draining the river and killing the federally threatened Santa Ana sucker (*Catostomus santaanae*; Sahagun 2016; Center for Biological Diversity 2018). The clustering of treatment plants in urbanized and agricultural landscapes suggests that the produced effluent outflow may be substituting as habitat for aquatic-associated species in these locations. Further study to determine the correlation of this phenomenon with the extent of urbanization would be worthwhile to understand the role that urbanization plays as a driver for species and treatment plant presence.

An additional clear association was demonstrated among both riparian-associated and aquatic emergent vegetation (e.g., wetland) species richness density and treatment plant density. Ecologically, these associations may seem less intuitive, but as described previously, we believe the correlation stems from riparian and wetland habitat types emerging as the direct result of an increased flow created by treatment plant effluent. This phenomenon has been documented in dry riverbeds that have seen a return to perennial flow from treatment plant effluent (White and Greer 2006; Stromberg et al. 1993). Anecdotally, oftentimes the locations of treatment plants are marked by clearly observable riparian shrubland or woodland communities immediately downstream of the outflow location. This can be seen within the Santa Ana River downstream of the Rapid Infiltration and Extraction (RIX) Facility in Colton, CA, as well as within the Mojave River downstream of the Victor Valley Wastewater Reclamation Authority WTP in Victorville, CA (Google Earth 2021). Similar to the analysis for aquatic-associated species, given that treatment plants are most prevalent within urbanized and agriculturally driven landscapes, an increase in aquatic habitat from effluent would also drive an increase in riparian and wetland habitat types and support the species that rely on these habitat types. Based on the findings in this paper, we can further expect that the persistence of these habitat types and the species they support are directly linked with future effluent management decisions.

Finally, the high correlation between terrace-associated species and treatment plants was unexpected because terraces are the only floodplain habitat component that would be expected to shrink with an increase in flow. Terraces are a unique habitat type formed by larger scale disturbances (e.g., flooding) that restructure nutrients and sediment (Chock et al. 2020). This type of habitat can also overlap with low-flow channels that experience the absence of flow (i.e., rivers in arid environments that run dry in the summer and fall). Treatment plants, however, produce outflow at near constant rates, creating perennial waterbodies in systems that may have historically existed with an ephemeral or intermittent flow regime, a phenomenon that has been documented in arid environments (Brooks, Riley, and Taylor 2006; Luthy et al. 2015). In this case, the presence of relatively continuous treatment plant outflow would be expected to decrease the quality of habitat for terrace-associated species. Additionally, as described previously, terrace habitats are largely threatened by anthropogenic water withdrawal and flood control measures that limit the extent of flooding that these habitat types require (63 FR 51005). In this case, it is possible that because treatment plants and flood control both exist as functions of urbanization, these variables have been confounded and the high correlation with terrace species is presenting as a link to treatment plant effluent. Again, further study to understand the role urbanization plays as a driver for species and treatment plant presence would be worthwhile to tease apart these differences. In addition, we believe this association could be artificially inflated in the statistical analysis due to the significantly fewer watersheds (n=48) that contain species associated with this habitat type. In addition to occurring in fewer watersheds, the terrace datasets also contained fewer outlier values which increased the correlation coefficient. For comparison, the other three habitat types (aquatic, aquatic vegetation, and riparian) each had greater than 100 watersheds that contained associated species, which also inherently led to more outlier values through an increase in presence of watersheds with no treatment plants, lowering the correlation coefficient.

We acknowledge that this grouping of species solely represents those that have made it through the multitude of legal hurdles required for inclusion on state and federal protection lists. This is important because there may be many other native species that are imperiled, but remain understudied and have not yet received the critical mass necessary to begin the listing process. Our analysis focused on legally protected species due to their visibility within species conservation management and the malleability of their future trajectories; however, it is likely that the findings in this study can be extended to benefit other native floodplain species in the state of California and beyond.

When evaluating the permitted treatment plant outflow in relation to the receiving waterbody baseflow, we found that there are many watersheds (34%) that could potentially receive the majority of their cumulative watershed baseflow from effluent. As shown in Figure 8, these watersheds are located in areas with dense urbanization or agricultural production such as the Los Angeles basin, San Francisco Bay Area, and Central Valley, suggesting that floodplain habitats (and therefore the species they support) in these regions could be changing in response to increases in hydrology that perhaps were not present historically. This further supports the positive correlation found between floodplain species and treatment plants documented earlier.

We also documented differences when comparing minimum monthly mean discharge and maximum monthly mean discharge to the regulated outflow from treatment plants. Generally, we found that 69% of studied watersheds are legally allowed to cumulatively produce more effluent than the baseflow discharge during minimum flow conditions (e.g., summer and fall), suggesting that in these cases, waterbodies may not reach their natural low-flow points. This further supports work that has been done signifying that ephemeral and intermittent streams are experiencing perennial flows as a result of treatment plant effluent (Brooks et al. 2006; Luthy et al. 2015), as well as work that has documented effluent increasing discharge during low-flow conditions (White and Greer 2006; Zimmerman et al. 2018). This raises the question as to whether native species adapted to seasonal fluctuation and natural flow withdrawals are further imperiled by alterations to this resource. Significantly fewer (18%) watersheds had cumulative permitted outflow that exceeded the maximum monthly mean discharges, indicating that at times of high-flow conditions (e.g., winter and spring), treatment plant effluent would likely not contribute as strongly to baseflow discharge.

The analysis used to compare treatment plants outflow to baseflow is relatively coarse because stream gage data was not available for the associated receiving waterbodies for all of the treatment plants (we analyzed data for 67% of the total treatment plants) and therefore we aggregated the data to the watershed level in order to better evaluate emerging patterns. However, this meant that some watersheds did not include effluent data for all of the treatment plants within their boundary. In instances where this was the case, the results may be an underrepresentation of the true contribution of effluent to baseflow. In addition, we were unable to account for whether the stream gage that measured the receiving waterbody baseflow was upstream or downstream of the treatment plant outflow location. In events where the stream gage is located downstream of the treatment plant, the discharges from the treatment plant and receiving waterbody would have looked similar because the receiving waterbody gage would be capturing the treatment plant discharge. This indicates that there is perhaps an underrepresentation of how much treatment plant effluent contributes to baseflow. Finally, the baseflow discharge does not take into account the multitude of other anthropogenic water withdrawals that could be occurring upstream of the stream gage location, particularly in urbanized landscapes. Irrespective of these challenges, the conclusion that treatment plant effluent plays a role in shaping species habitat within hydrologically modified landscapes does not change. An area of future study should include building upon the natural flow modeling framework presented in Zimmerman et al. 2018 and quantifying to what extent discharge from treatment plant effluent is replacing historical natural baseflows that have been removed from alternative forms of anthropogenic hydrologic modification. For instance, it would benefit water conservation policy for managers to understand the quantity of water that has been removed for municipal purposes compared to a natural flow regime, and then in what ways effluent is replacing that removal.

It is also important to note that the flow analysis compared stream gage data (which was presented at the mean annual, minimum monthly mean, and maximum monthly mean time intervals) to the wastewater treatment plant discharge permitted by the state. The permitted values represent the maximum discharge permissible; however, it does not capture temporal variability of discharge at the day-to-day level within each treatment plant. Treatment plants discharge effluent at a relatively constant rate when compared to a natural flow regime that includes seasonal and interannual fluctuation; however, treatment plant discharge still includes some variation. Generally, treatment plants produce outflow as a function of the inflow of wastewater they receive from their service region (i.e., how much municipal water is being sent down the drains). What is then not recycled for other municipal needs is discharged into rivers and streams at a maximum discharge rate outlined in the treatment plant's permit from the state. The factors that determine this rate are based on the design capacity of the treatment plant, other municipal needs for the effluent, and the size and complexity of the service region (MDE 2006). Human behavior also plays a role in discharge rates, with peak outflow occurring during daylight hours in response to the diurnal nature of most human activities (Butler and Graham 1995; Enfinger and Stevens 2014). While there is inherent variability in the discharge rate of a treatment plant relative to the maximum value listed on the permitting document, the variation is not comparable to what is seen in a flow regime intended to mimic natural conditions which would require seasonal and interannual differences. An area of future study should include evaluating the daily discharge rates of a treatment plant in order to understand the true variability in discharge and compare it with both the receiving waterbody daily discharge and the historical natural flow regime. This would help fill the knowledge gap of how treatment plant discharge truly compares to a natural flow regime and it could lead to actionable suggestions that treatment plant operators could implement in order to mimic natural conditions and assist in species conservation.

In this study, we have shown that there is substantial overlap between the presence of species and the presence of treatment plants in California watersheds, and with this overlap, a large potential for unintended consequences. We further posit that discharge from treatment plants may be providing and shaping habitat for floodplain-associated species, especially in urbanized and agriculturally driven landscapes. As such, species conservation goals should be considered when making decisions related to effluent reuse. There is no denying that water conservation management (in which effluent recycling is becoming a major component) is necessary and intentional within the state of California, a state that has undergone widespread modification to its water infrastructure in order to support its burgeoning human population and a state that is experiencing the adverse effects of a changing climate. Nevertheless, effluent recycling and species conservation cannot each be evaluated in a vacuum – it is important to acknowledge their interdependency. While the purpose of this study is not to assume any causality between the presence of treatment plants and species, our analysis demonstrates that the fates of these two resources are ultimately interconnected and we want to bring awareness to a potential future causality that could occur due to the unintended consequence of water policy. This study was presented at the landscape level in order to evaluate patterns that may have only emerged when reviewing the data holistically; however, decision-making related to species conservation and effluent reuse happens at the regional level and judgements need to be adapted to each situation on a case-by-case basis. We intend for this study to aid conservation practitioners with decision-making that can be beneficial for both water and species conservation efforts.

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