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Stable isotopes of algae and macroinvertebrates in streams respond to watershed urbanization, inform management goals, and indicate food web relationships

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ABSTRACT

Watershed development and anthropogenic sources of nitrogen are among leading causes of negative impacts to aquatic ecosystems around the world. The $\delta^{15}N$ of aquatic biota can be used as indicators of anthropogenic sources of nitrogen enriched in ^{15}N, but this mostly has been done at small spatial extents or to document effects of point sources. In this study, we sampled 77 sites along a forest to urban land cover gradient to examine food webs and the use of $\delta^{15}N$ of periphyton and macroinvertebrate functional feeding groups (FFGs) as indicators of watershed development and nitrogen effects on streams. Functional feeding groups had low $\delta^{15}N$ variability among taxa within sites. Mean absolute differences between individual taxa and their respective site FFG means were < 0.55‰, whereas site means of $\delta^{15}N$ of FFGs had ranges of approximately 7–12‰ among sites. The $\delta^{15}N$ of periphyton and macroinvertebrate FFGs distinguished least disturbed streams from those with greater watershed urbanization, and they were strongly correlated with increasing nitrogen concentrations and watershed impervious cover. Nonmetric multidimensional scaling, using $\delta^{15}N$ of taxa, showed that changes in macroinvertebrate assemblages as a whole were associated with forest-to-urban and increasing nitrogen gradients. Assuming an average +3.4‰ per trophic level increase, $\delta^{15}N$ of biota indicated that detrital pathways likely were important to food web structure, even in streams with highly developed watersheds. We used periphyton and macroinvertebrate FFG $\delta^{15}N$ to identify possible management goals that can inform decisions affecting nutrients and watershed land use. Overall, the $\delta^{15}N$ of periphyton and macroinvertebrates were strong indicators of watershed urban development effects on stream ecosystems, and thus, also could make them useful for quantifying the effectiveness of nitrogen, stream, and watershed management efforts.

1. Introduction

Anthropogenic sources of nutrients continue to be among leading causes of negative impacts to stream ecosystems in the United States and in many parts of the world (Carpenter et al., 1998; Vörösmarty et al., 2010; Stoddard et al., 2016). In developed areas, human activities, growing populations, and impervious cover in watersheds increase nitrogen loads to downstream water bodies from wastewater, sewers, septic systems, fertilizers, and stormwater runoff (Kaushal et al., 2011; Kaushal et al., 2014; Pennino et al., 2016). Urbanization alters flow paths, hydrology, and geomorphology, and decreases vegetation, which further changes the transport and cycling of nutrients in watersheds and streams (Walsh et al., 2005; Wollheim et al., 2005; Kaushal and Belt, 2012). These changes in nutrient dynamics, habitat, and hydrology affect biogeochemistry, rates of primary production, and food webs in streams (Meyer et al., 2005; Miltner, 2010; Klose et al., 2012; Kautza and Sullivan, 2016), which can have negative downstream consequences for recreational opportunities, tourism, and property values, along with possible increased health risks (Dodds et al., 2009; Sobota et al., 2015). Nutrient pollution in watersheds also creates problems for estuarine and coastal ecosystems that are commonly nitrogen limited (Howarth et al., 2002; Seitzinger et al., 2005; Boyer et al., 2006). As a result, developing indicators based on ecological responses to nutrients and watershed land cover can assist with setting effects-based goals for stream ecosystem protection, management, and restoration, and with informing future decisions affecting

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land use and aquatic resources.

Given that human wastewater is enriched with the ^{15}N isotope and is an abundant source of nitrogen in urban and suburban watersheds (Heaton, 1986; Castro et al., 2003; Carey et al., 2013), measuring nitrogen stable isotope ratios ($\delta^{15}N$) in organisms has become a useful approach to identifying effects of different nitrogen sources on aquatic ecosystems (McClelland et al., 1997; Lake et al., 2001; Vander Zanden et al., 2005). In addition to point source discharges of wastewater, septic systems and leaking sewers are substantial sources of nitrogen entering streams (Groffman et al., 2004; Steffy and Kilham, 2004). Atmospheric deposition and inorganic lawn fertilizers could lower $\delta^{15}N$ in systems affected by wastewater because they typically range from 2–8‰ and −3 to 3‰, respectively, whereas human and animal wastes are typically > 10‰ (Heaton, 1986; Valiela et al., 2000). However, wastewater sources often dominate nitrogen loads in developed watersheds (Carey et al., 2013; Divers et al., 2014), and microbial denitrification, which benefits water quality by removing N, has preferential uptake of ^{14}N and increases $\delta^{15}N$ in downstream waters affected by inorganic fertilizers (Kellman and Hillaire-Marcel, 1998; Vander Zanden et al., 2005). Some evidence even suggests that nitrogen inputs to suburban streams from pet waste can exceed contributions from lawn fertilizers (Groffman et al., 2004; Carey et al., 2013). Despite differences in $\delta^{15}N$ from multiple sources, $\delta^{15}N$ in algae, macroinvertebrates, and fish generally increase in response to human and animal sources of nitrogen (Ulseth and Hershey, 2005; di Lascio et al., 2013; Hicks et al., 2017).

Examining stable isotopes of multiple trophic levels is useful because urbanization affects basal resources and alters the biomass and abundance of macroinvertebrate functional feeding groups in streams, such as shredders, scrapers, collectors, and predators (Stepenuck et al., 2002; Sterling et al., 2016; García et al., 2017). Measuring $\delta^{15}N$ in aquatic organisms among different trophic levels (e.g., primary producers, consumers, and predators) provides information regarding how food webs change, because of trophic fractionation in biota (Fry, 1991; Vander Zanden and Fetzer, 2007; Layman et al., 2012). The $\delta^{15}N$ incorporated into the bodies of aquatic organisms reflects the isotope ratio of their nitrogen source or diet, with an average enrichment of +3.4‰ (typical range of 2–4‰) per higher trophic position (Post, 2002). The $\delta^{13}C$ of consumers and higher trophic levels, which has little to no fractionation (< 0.5‰), can provide additional insight into basal resources, energy transfer, and sources of carbon in food webs (France, 1995; Post, 2002; García et al., 2017). Stable isotopes of biota also integrate the exposure to and effects of urbanization and nitrogen from human sources reaching streams over time. Temporal variability in $\delta^{15}N$ of macroinvertebrates and primary producers still exists due to their internal turnover of nutrients, with long-lived organisms and higher trophic positions typically having less variability (Cabana and Rasmussen, 1996; Post, 2002; Jardine et al., 2014). However, within season variation is relatively low, whereas among season variation can be greater (Peipoch et al., 2012; Woodland et al., 2012; Pastor et al., 2014).

Studies of biota $\delta^{15}N$ in streams with developed watersheds mostly have focused on a small number of sites (typically < 20) with limited spatial extents (< 100 km^2 watersheds) or on direct upstream-to-downstream effects of wastewater treatment plants (e.g., Morrissey et al., 2013; Hicks et al., 2017). In this study, we used a spatially-balanced sampling design in a > 4400 km^2 coastal watershed, which targeted stream sites along a forest to urban land cover gradient, to determine if $\delta^{15}N$ of periphyton and macroinvertebrate consumers and predators were effective indicators of nitrogen and urban effects on stream ecosystems. We (1) documented within and among site variability of $\delta^{15}N$ for functional feeding groups, (2) identified responses of biota $\delta^{15}N$ to watershed urbanization and nitrogen concentrations, (3) used results to inform possible nitrogen management goals, and (4) examined if watershed urbanization affected food webs.

2. Materials and methods

2.1. Site selection and sampling

Wastewaters from point sources, sewers, and septic systems associated with watershed development and high population density are the leading contributors of nitrogen in the Narragansett Bay watershed (4421 km^2), which is located in northeastern USA, has 34.5% developed land cover, and 380 people km^{-2} (Castro et al., 2003; US EPA, 2007). We used ArcGIS 9.3 (Environmental Systems Research Institute, Redlands, California) and the 2006 National Land Cover Database (NLCD) to randomly select 105 possible 2nd to 4th order stream sites within categories of watershed impervious cover (< 1%, 1–5.5%, 5.5%–10%, 10–20%, 20–30%, and > 30%). For each of these categories, we used GIS to overlay a tessellated hexagon grid on the watershed and randomly selected one site within each hexagon. This survey design increased the likelihood of sampling sites along a continuous gradient of watershed development, while ensuring a representative spatial distribution of sites within each impervious cover category throughout the watershed (Smucker et al. 2016). We used NHDPlus Basin Delineator Software (www.horizon-systems.com) to delineate watersheds, which were checked for accuracy using US Geological Survey (USGS) 7.5-min quadrangles (1:24,000). We used NLCD 2006 for site selection because of available GIS tools that facilitate quick estimates of land cover, but once sites were selected we used photo-interpreted aerial imagery to quantify impervious cover and land cover in upstream watersheds for each site. Rhode Island land cover used 2003–2004 aerial imagery with 0.6 m resolution, and Massachusetts land cover used 2005 aerial imagery with 0.5 m resolution (RIGIS www.edc.uri.edu/rigis; MassGIS www.mass.gov/anf/research-and-tech/it-serv-and-support/application-serv/office-of-geographic-information-massgis).

We sampled 77 sites with watersheds < 200 km^2 between July and October 2012 (Fig. 1). Other sites were not sampled because they were

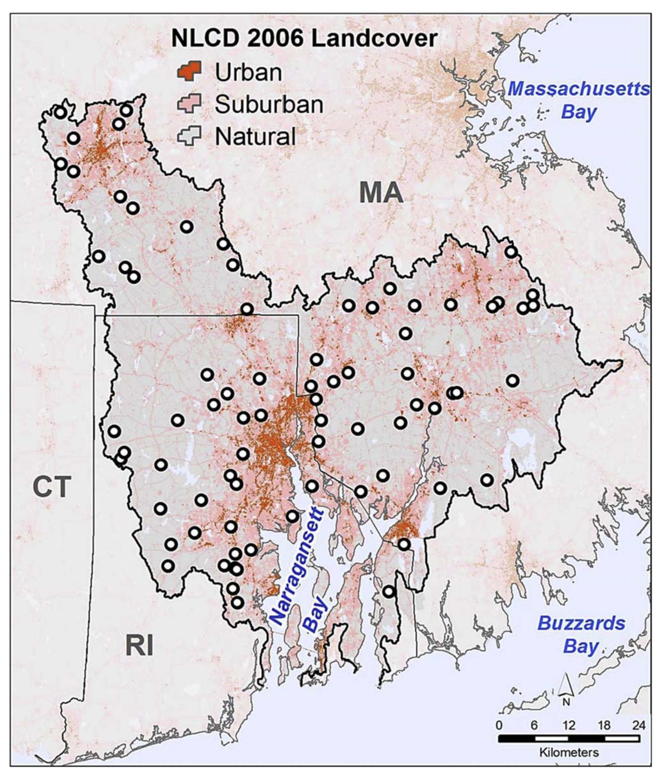

Fig. 1. Map showing the distribution of 77 sample sites in the Narragansett Bay watershed. Urban, suburban, and natural (forests and wetlands) land cover are shown. MA = Massachusetts, RI = Rhode Island, CT = Connecticut, NLCD = National Land Cover Database.