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## Marine Pollution Bulletin

journal homepage: [www.elsevier.com/locate/marpolbul](http://www.elsevier.com/locate/marpolbul)

# Trends in nitrogen isotope ratios of juvenile winter flounder reflect changing nitrogen inputs to Rhode Island, USA estuarine systems

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## ARTICLE INFO

## Article history:

Received 30 November 2016

Received in revised form 27 January 2017

Accepted 5 February 2017

Available online xxxxx

## Keywords:

Stable nitrogen isotopes

Fish

Winter flounder

Estuaries

## ABSTRACT

Nitrogen isotope ratios ( $\delta^{15}\text{N}$ ) in juvenile winter flounder, *Pseudopleuronectes americanus*, were used to examine changes in nitrogen inputs to several Rhode Island, USA estuarine systems. Fish were collected over two three-year periods with a ten-year interval between sampling periods (2002–2004 and 2012–2014). During that interval numerous changes to nutrient management practices were initiated in the watersheds of these estuarine systems including the upgrade of several major wastewater treatment facilities that discharge to Narragansett Bay, which significantly reduced nitrogen inputs. Following these reductions, the  $\delta^{15}\text{N}$  values of flounder in several of the systems decreased as expected; however, isotope ratios in fish from upper Narragansett Bay significantly increased. We believe that low  $\delta^{15}\text{N}$  values measured in 2002–2004 were related to concentration-dependent fractionation at this location. Increased  $\delta^{15}\text{N}$  values measured between 2012 and 2014 may indicate reduced fractionation or that changes in wastewater treatment processes altered the nitrogen isotopic ratios of the effluents.

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## 1. Introduction

Efforts are underway worldwide to reduce nutrient inputs to coastal systems by changing land management practices or improving wastewater treatment systems (Pitt et al., 2009; Longphuir et al., 2015; Riemann et al., 2016). As an example, between 2002 and 2010 Rhode Island has upgraded all 11 of its wastewater treatment facilities (WWTFs) that directly discharge to upper Narragansett Bay or its tributaries to tertiary treatment. In addition, Massachusetts has upgraded 9 of 11 of its WWTFs that discharge into tributaries entering upper Narragansett Bay (RIDEM, 2005; Krumholz, 2012; NBEP, 2016). This has resulted in a reduction in the nitrogen load to the Bay from sewage of about 57% compared to 2000–2004 inputs (NBEP, 2016). In addition to sewage upgrades, a combined storm sewer overflow reservoir constructed under the city of Providence was completed in 2008. Together, these changes have resulted in large decreases in nitrogen inputs to the Bay. Also, in the watersheds of the Rhode Island coastal ponds, located on the south coast of the State adjacent to Narragansett Bay, improved nutrient management practices have been initiated including the increased use of denitrifying septic systems. In the watershed of Narrow River, a sub-estuary emptying into the southern part of Narragansett Bay, many households have been connected to municipal WWTFs.

Nitrogen isotope ratios ( $\delta^{15}\text{N}$ ) have been widely used to track anthropogenic nitrogen inputs to aquatic systems (Martinetto et al., 2006; Bannan and Roman, 2008). Nitrogen from sewage treatment facilities or in groundwater influenced by septic systems has elevated levels of  $^{15}\text{N}$  relative to  $^{14}\text{N}$  (Heaton, 1986; McClelland and Valiela, 1998). Therefore, measurements of  $\delta^{15}\text{N}$  in water, sediments and biota can be used to indicate the relative importance of sewage inputs to aquatic systems and to monitor changes over time at specific locations.

In a previous study (Pruell and Taplin, 2015) we measured nitrogen isotopes in juvenile winter flounder, *Pseudopleuronectes americanus*, collected between 2002 and 2004 from several Rhode Island estuarine systems, including Narragansett Bay, several coastal ponds and the Narrow River. The objective of the present study was to resample juvenile winter flounder from those same locations after ten years to assess whether changes in nutrient inputs could be detected in the biota of these systems. Therefore, flounder were collected over two three-year periods, ten years apart (2002–2004 and 2012–2014), before and after numerous steps had been undertaken in the watersheds to reduce nitrogen inputs to these estuarine systems.

## 2. Materials and methods

### 2.1. Fish collection

During two sampling periods (2002–2004 and 2012–2014) young-of-the-year winter flounder were collected from each of 20 stations

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(Fig. 1; Table 1) along the coast of Rhode Island, USA. These sampling locations included three shallow coastal lagoons, which are locally called salt ponds (Point Judith Pond, Ninigret Pond and Green Hill Pond), an estuarine river system (Narrow River) and the upper, middle and lower portions of Narragansett Bay (Fig. 1). Details on the

locations and physical characteristics of these stations are included in previous publications (Pruell et al., 2012; Pruell and Taplin, 2015). Fig. 1 provides information on the locations and discharge flows (averaged from 2002 to 2004) from the WWTFs in the Narragansett Bay watershed.

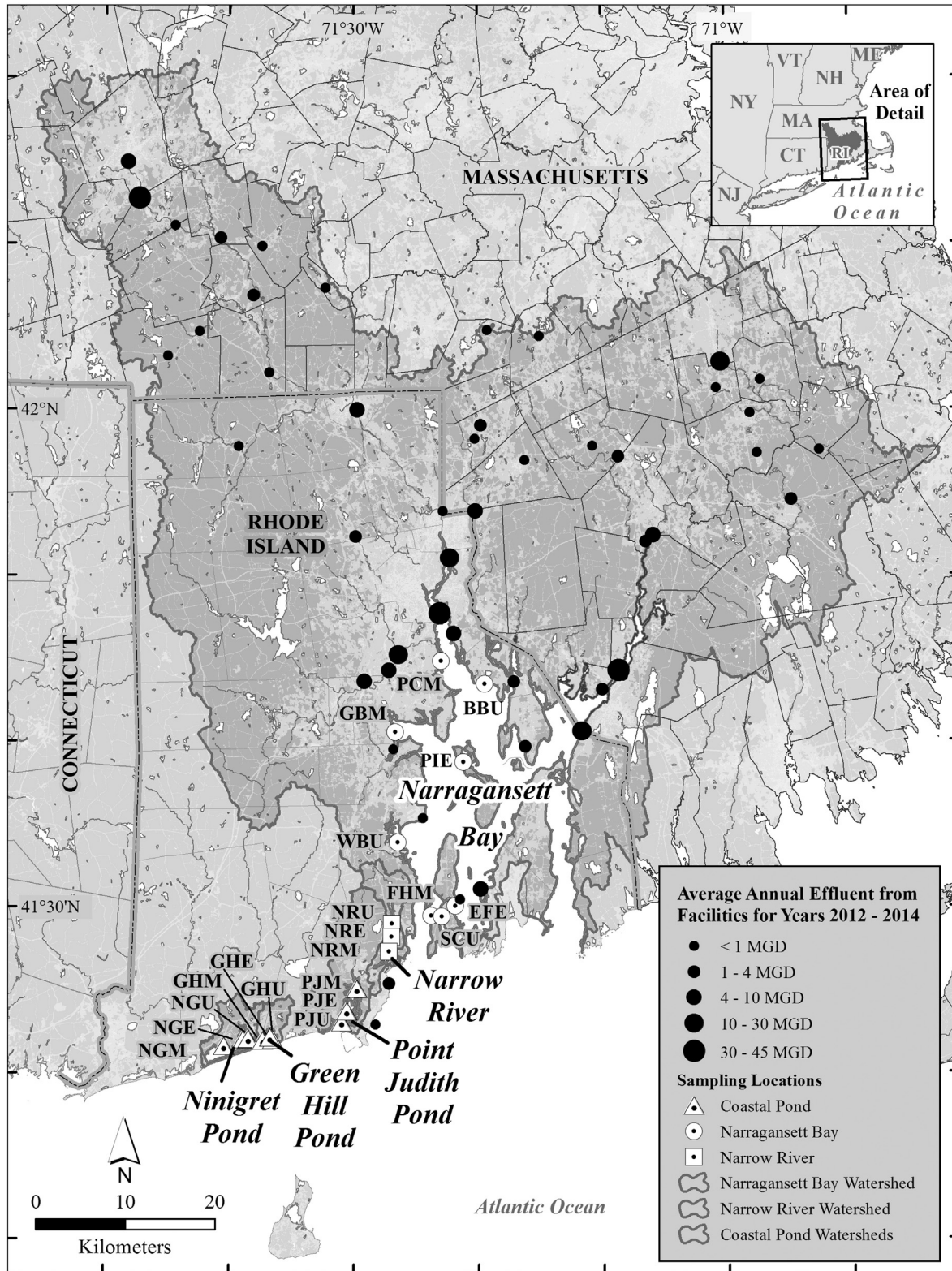


Fig. 1. Sampling locations for juvenile winter flounder along the coast of Rhode Island, USA. The sampling sites included three coastal ponds (Ninigret Pond, Green Hill Pond and Pt. Judith Pond), an estuarine river system (Narrow River) and Narragansett Bay. The station identifications are listed in Table 1. Locations and annual effluent flows for the wastewater treatment facilities in the area averaged over 2012–2014 are also shown.

Please cite this article as: Pruell, R.J., et al., Trends in nitrogen isotope ratios of juvenile winter flounder reflect changing nitrogen inputs to Rhode Island, USA estuarine syst..., Marine Pollution Bulletin (2017), <http://dx.doi.org/10.1016/j.marpolbul.2017.02.009>

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