Analysis of environmental DNA holds promise for monitoring biodiversity in changing ecosystems and may have broad applications for resource assessments. We here evaluate the availability of reference sequences of 47 plearonectiforms species (8 families), documented from North Carolina to the Gulf of Maine, matching to amplicons generated by a common eDNA mitochondrial marker, the Riaz 12S primers. GenBank ([https://www.ncbi.nlm.nih.gov](https://www.ncbi.nlm.nih.gov/)) provided full sequences of the Riaz amplicon region for 21 flatfishes (plus 2 partial sequences), indicating that 51% of 47 species lack reference sequences as of September, 2022. Using the Riaz 12S marker we compared flatfish taxa actually observed using eDNA metabarcoding on filtered water samples to a bottom trawl survey that identified fish using traditional morphological traits across this region during fall 2019. Twelve flatfish taxa were taken in X bottom trawl tows (2 identified as sp.). The eDNA survey ‘caught’ 11 flatfish taxa in 172 water samples (2 identified only to family); X taxa were observed in both surveys. Additional work is needed to shore up reference materials in GenBank for improving estimates of flatfish diversity. Regarding fishery applications, 5 flatfishes with formal assessments are represented by sequences in GenBank, as well as an additional 2 sequences that can only be identified to family level (Pleuronectidae 1 and Pleuronectidae 2). An initiative by NOAA Fisheries National Systematics Laboratory, Smithsonian, is designed to improve reference materials, as well as to evaluate the application of multi-barcodes in improving taxonomic identifications.

Environmental DNA holds promise for monitoring biodiversity in dynamic ecosystems and may have broad applications for resource assessments. We hereOur study of flatfishes evaluates the sufficiency of a common eDNA mitochondrial marker (i.e., the Riaz 12S primer set) to match all 47 pleuronectiform species (8 families) documented from North Carolina to the Gulf of Maine. For this marker, 51% of the 47 species lack reference sequences; full sequences for 21 flatfishes (plus 2 partial sequences) were available in GenBank (<https://www.ncbi.nlm.nih.gov>) as of September, 2022. UsingA real test of the Riaz 12S primers, marker we compared flatfish taxa observed by two surveys conducted across this region during fall 2019: 1) a bottom trawl survey identified 12 flatfish taxa (3 identified as sp.) using traditional morphological traits, and 2) an eDNA metabarcodingoceanographic survey using filtered water samples identified 11 taxa (2 identified to family) from amplicon sequence variants (ASVs) detected from filtered water samples; 5 taxa were identified to the same species in both surveys. Much less water was strained by the eDNA method to reveal the same biodiversity:. In terms of effort, we examined 301 trawl tows versus 346 liter seawater used for eDNA metabarcoding [YL: a measure of water volume]. Additional work is needed to shore up the reference materials in GenBank for improving estimates of flatfish diversity. Regarding fishery applications, 5 flatfishes with formal stock assessments are represented by sequences in GenBank, as well as an additional 2 sequences that can only be identified to family (Pleuronectidae). An initiative by NOAA Fisheries’ National Systematics Laboratory, Smithsonian, is designed to improve reference materials. Use of multiple markers will also improve taxonomic identifications.

Environmental DNA holds promise for monitoring biodiversity in dynamic ecosystems and may have broad applications for resource assessments. We here evaluates the sufficiency of a common eDNA mitochondrial marker (i.e., the Riaz 12S primers) to match all 47 pleuronectiform species (8 families) documented from North Carolina to the Gulf of Maine. For this marker, 51% of the 47 species lack reference sequences; 21 full and 2 partial sequences were available in GenBank (<https://www.ncbi.nlm.nih.gov>) as of September, 2022. Using the Riaz 12S primers, we compared flatfish taxa observed by two surveys conducted across this region during fall 2019: 1) a bottom trawl survey identified 12 flatfish taxa (3 identified as sp.) using morphological traits, and 2) an eDNA metabarcoding survey using filtered water samples identified 11 taxa (2 identified to family); 5 common species were identified in both surveys. Much less water was strained by the eDNA method to reveal the same biodiversity: 301 trawl tows vs. 346 liter seawater for eDNA metabarcoding. Additional work is needed to shore up the reference materials in GenBank for improving estimates of flatfish diversity. Regarding fishery applications, 5 flatfishes with formal stock assessments are represented by sequences in GenBank, as well as an additional 2 sequences that can only be identified to family (Pleuronectidae). An initiative by NOAA Fisheries’ National Systematics Laboratory, Smithsonian, is designed to improve reference materials. Use of multiple markers will also improve taxonomic identifications.