

11-21-2016

First occurrence of the invasive hydrozoan *Gonionemus vertens* A. Agassiz, 1862 (Cnidaria: Hydrozoa) in New Jersey, USA

John J. Gaynor

Montclair State University, gaynorj@mail.montclair.edu

Paul A.X. Bologna

Montclair State University, bolognap@montclair.edu

Dena J. Restaino

Montclair State University, restainod1@mail.montclair.edu

Christie Barry

Montclair State University, castellano1@montclair.edu

Follow this and additional works at: <https://digitalcommons.montclair.edu/biology-facpubs>

Part of the [Biodiversity Commons](#), [Genomics Commons](#), [Marine Biology Commons](#), and the [Population Biology Commons](#)

MSU Digital Commons Citation

Gaynor, John J.; Bologna, Paul A.X.; Restaino, Dena J.; and Barry, Christie, "First occurrence of the invasive hydrozoan *Gonionemus vertens* A. Agassiz, 1862 (Cnidaria: Hydrozoa) in New Jersey, USA" (2016). *Department of Biology Faculty Scholarship and Creative Works*. 3.

<https://digitalcommons.montclair.edu/biology-facpubs/3>

Published Citation

BioInvasions Records (2016) Volume 5, Issue 4: 233–237

Rapid Communication

First occurrence of the invasive hydrozoan *Gonionemus vertens* A. Agassiz, 1862 (Cnidaria: Hydrozoa) in New Jersey, USA

John J. Gaynor*, Paul A.X. Bologna, Dena Restaino and Christie L. Barry

Montclair State University, Department of Biology, 1 Normal Avenue, Montclair, NJ 07043 USA

E-mail addresses: gaynorj@montclair.edu (JG), bolognap@montclair.edu (PB), restainod1@montclair.edu (DR), castellanoc1@montclair.edu (CB)

*Corresponding author

Received: 16 August 2016 / Accepted: 31 October 2016 / Published online: 21 November 2016

Handling editor: Stephan Bullard

Abstract

Gonionemus vertens A. Agassiz, 1862 is a small hydrozoan native to the Pacific Ocean. It has become established in the northern and southern Atlantic Ocean as well as the Mediterranean Sea. We report on the first occurrence of this species in estuaries in New Jersey, USA, and confirm species identification through molecular sequence analysis. Given the large number of individuals collected, we contend that this is a successful invasion into this region with established polyps. The remaining question is the vector and source of these newly established populations.

Key words: clinging jellyfish, Mid-Atlantic, 16S rDNA, COI

Introduction

Invasive species are well documented to have significant impacts on communities they have invaded (Thomsen et al. 2015). However, many diminutive species have been transported globally, yet seemingly have few impacts on the systems they have invaded (Hewitt et al. 2004; Gasith et al. 2011). Small hydrozoans in particular have invaded estuaries globally with seemingly no substantial impact. For example, the hydrozoan *Blackfordia virginica* Mayer, 1910, originally thought to be a native of the western mid-Atlantic, originated from the Black Sea and has successfully become established globally (Harrison et al. 2013).

The native range of *Gonionemus vertens* A. Agassiz, 1862, the clinging jellyfish, includes coastal regions of the North Pacific Ocean (Fofonoff et al. 2003). Its common name originates from the behavior of the medusa stage to “cling” to vegetation using modified adhesive tentacle pads. The medusae are predators of small zooplankton and epifaunal organisms that share the same vegetated habitats. Edwards (1976) suggests that two variants of this

species exist and exhibit different levels of sting potency, with the more venomous organisms present from the western North Pacific, while those from the eastern North Pacific have less potent stings. This species has now been documented to have invaded the coast of Norway, the Baltic Sea, the Mediterranean Sea, the northwest Atlantic, and the coast of Argentina (Fofonoff et al. 2003, and references within; Schuchert 2016). Govindarajan and Carman (2016) review and document the invasions within the Cape Cod region of Massachusetts and occurrences in other regional New England estuaries. Interestingly, the initial introduction in the late 1800s appears to be of the less venomous form and the potential re-introduction in the 1990s is of the more toxic variety. Here we report on successful invasion into New Jersey’s estuaries of the more venomous variety.

Material and methods

The first reported *G. vertens* was collected in the Manasquan River estuary (Figure 1) with subsequent initial samples (n = 5) collected from the Shrewsbury

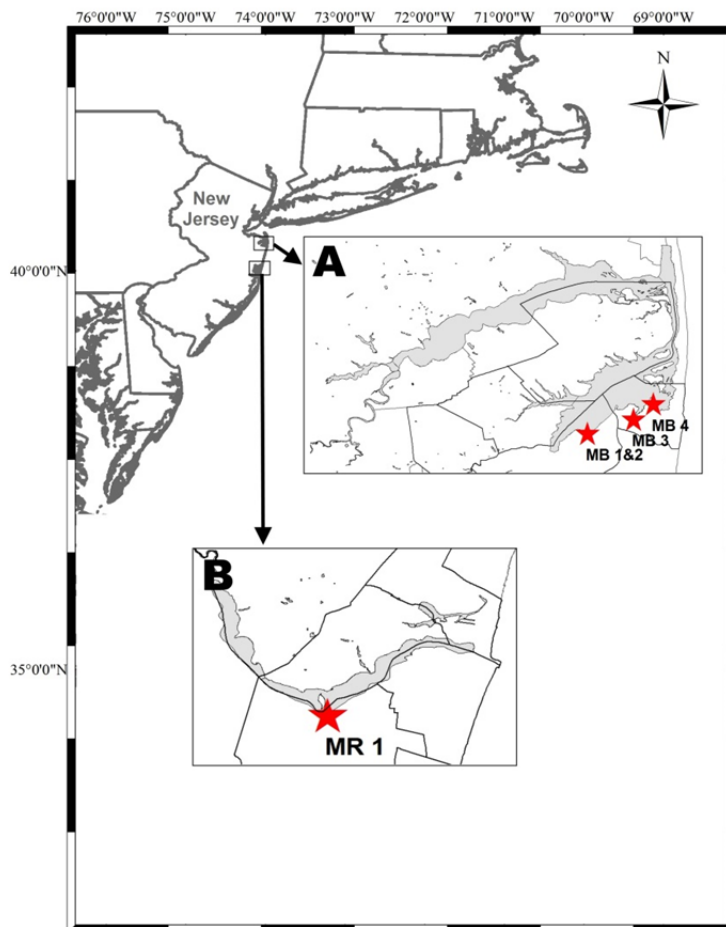


Figure 1. Map of the distribution of *Gonionemus vertens* in New Jersey estuaries, A: Shrewsbury River, B: Manasquan River (see Supplementary material Table S1 for geo-referenced locations).

River estuary). Individuals (e.g., Figure 2) were taken to Montclair State University (Montclair, New Jersey, USA) where tentacles were dissected from these five samples. Tentacles were excised from each individual (4 from each sample) and extracted using a CTAB/NaCl protocol (Winnepennickx et al. 1993) with the following modifications. All extractions were carried out in 500 μ L volumes in 1.7 mL microcentrifuge tubes and homogenized by grinding for 30 s with a micropestle (Eppendorf). Homogenized samples were incubated at 60 $^{\circ}$ C for 60 min, and RNA was digested by incubation with RNase A (Sigma-Aldrich; 10 μ g for 30 min at 37 $^{\circ}$ C) prior to precipitation. DNA was precipitated with 2/3 volume of isopropanol, pelleted in a microfuge (16,100 \times g for 10 min at 4 $^{\circ}$ C), washed twice with ice-cold 70% (v/v) ethanol, briefly dried in a Speed-Vac, and resuspended in 20 μ L of TE (10 mM Tris, 0.1 mM EDTA, pH 8). DNA concentrations and OD_{260/280} ratios were determined in a NanoDrop ND-1000.

PCR amplifications were performed using ChoiceTaq Master Mix (Denville Scientific, Denville, New Jersey, USA) according to manufacturer's directions with the exception that we used 20 μ L reaction volumes. Primers used for amplification of the 16S rDNA and COI loci are listed in Table 1. PCR was carried out in a ProFlex Thermal Cycler (Applied Biosystems, Inc.) according to the following parameters: 95 $^{\circ}$ C for 1 min (1 \times); 95 $^{\circ}$ C for 20 s, 55 $^{\circ}$ C for 20 s, and 72 $^{\circ}$ C for 30 s (30 \times); 72 $^{\circ}$ C for 7 min (1 \times); followed by a hold at 4 $^{\circ}$ C. Positive and negative template controls were always run and typically 10 μ L of each sample was run on a 1% (w/v) agarose gel to assess purity and yield of PCR product. PCR products were subject to direct Sanger dideoxy sequencing on an ABI3130 Genetic Analyzer. Sequencing reactions were carried out using BigDye Terminator Ready Reaction Mix V3.1 according to manufacturer's instructions except that we used 1/16 diluted reactions. Excess unincorporated fluorescent dideoxy

Table 1. List of PCR Primers.

Locus	Primer Sequence (5' to 3')	Reference
16S	TCGACTGTTTACAAAAACATAGC ACGGAATGAACTCAAATCATGTAAG	Bridge et al. 1995
COI	GGTCAACAAATCATAAAGATATTGG TAAACTTCAGGGTGACCAAAAAATCA	Folmer et al. 1994



Figure 2. Image of *Gonionemus vertens* collected in New Jersey. Photograph by Christie Barry.

nucleotides were removed by Performa Gel DTR Gel Filtration Cartridges (EdgeBio, Gaithersburg, Maryland, USA). Both strands of each product were sequenced using the same primers used to generate the fragment. Raw sequences were edited and aligned using 4 Peaks (<http://nucleobytes.com/4peaks/index.html>) and CLUSTAL Omega (Sievers et al. 2011; <http://www.ebi.ac.uk/Tools/msa/clustalo/>), and searched for homology against all known genetic sequences using the BLAST algorithm (Altschul et al. 1990).

Results

Sequencing and bioinformatic analysis of both the 16S ribosomal and the cytochrome oxidase subunit I (COI) loci confirm the identity of our samples as the clinging jellyfish, *Gonionemus vertens*. Figure 3 shows the CLUSTAL Omega alignment of our sample (Genbank KX656923) with the two closest matches in Genbank for the 16S locus: our best match demonstrates a 99.4% homology (521/524 identities) with only 3 SNP's and one gap to a sample collected from the China Sea (Genbank KF962471). Alignment of our COI locus shows a similar strong homology to *Gonionemus vertens* COI as well (97.2% homology;

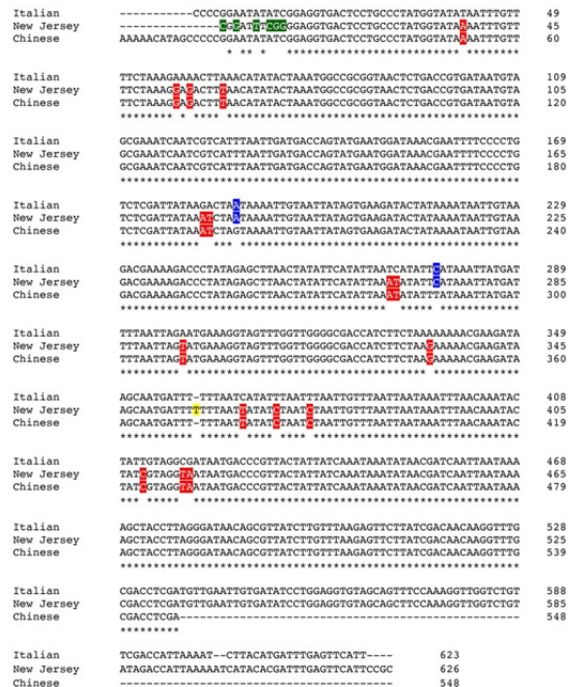


Figure 3. CLUSTAL Omega Alignment of *Gonionemus vertens* 16S rDNA Sequences. Asterisks show identity of all three aligned sequences. Red identifies unique homology between New Jersey and Chinese samples; blue identifies unique homology between New Jersey and Italian samples; green identifies bases unique only to New Jersey samples; and yellow demonstrates the point of insertion of one additional T in a run of 6 T's found in the other two samples. (Genbank Accession numbers for 16S loci are KF962471 (Chinese), EU293976 (Italian), and KX656923 (New Jersey).

633/651 identities) at the nucleotide level (data not shown). The fact that both of these loci, which are commonly employed for DNA barcoding of Hydrozoa (Harrison et al. 2013), point to the identification of our samples as *Gonionemus vertens*, strengthens our conclusions. Analysis of both the 16S and COI loci from all five New Jersey samples sequenced to date were identical. The lack of genetic variation in all of these samples, which were isolated from two populations nearly 20 miles distant from each other, suggests that they were the result of the same biological invasion event.

Discussion

The spread of non-native species throughout the world's oceans has accelerated due to anthropogenic means, and they may pose significant risks to biodiversity and ecosystem functions (Bax et al. 2003; Thomsen et al. 2015). In particular, several activities have promoted these invasions including aquaculture, bulk transport of cargo, travel, and exploration over the last 500 years (Katsanevakis et al. 2013). Processes which facilitate these invasions include hull fouling, ballast water discharge, and the construction of transport canals (see Hulme 2009). The development of the Panama and Suez Canals has opened pathways between distant ecoregions and their proposed expansions have highlighted the potential for greater invasion (Carman et al. 2011; Muirhead et al. 2015; Galil et al. 2015).

For *G. vertens*, its introduction to the Mediterranean has been speculated, but the prevalence of *G. vertens* invasions in northern Europe (Tambs-Lyche 1964; Bakker 1980), the western North Atlantic (Govindarajan and Carman 2016), and western South Atlantic (Rodriguez et al. 2014) suggest that this species has a great potential for transport and invasion (Schuchert 2016). Based upon our sequence analysis, the *G. vertens* discovered in New Jersey estuaries matches most closely a sample from the China Sea (Figure 3), indicating the more venomous group. This is in alignment with Govindarajan and Carman (2016) assertions. Consequently, the potential avenue for invasion may be a unique introduction to the area, but more likely comes from the Cape Cod, Massachusetts, USA, population; possibly transferred by local currents or vessels (Lavoie et al. 1999). Ultimately, determination of the precise invasion pathway may require a regional population genetics approach that compares genetic distance with physical distance.

Acknowledgements

This study was supported by a grant from the New Jersey Department of Environmental Protection (NJDEP) to PB and JG. We would also like to thank Gary Buchanan and Joe Bilinski of the NJDEP for assistance in field collections and for the use of research vessels from the NJDEP Bureau of Marine Water Monitoring. We also thank J. Hart, M. Carlo, the Parker Family, and C. Claus for assistance to getting access to specimens. We want to thank Annette Govindarajan and Mary Carman for their review of this manuscript and active discussions on the invasion and life history of *Gonionemus vertens*, as well as an anonymous reviewer who provided constructive comments to improve the manuscript.

References

- Altschul SF, Gish W, Miller W, Myers EW, Lipman DJ (1990) Basic local alignment search tool. *Journal of Molecular Biology* 215: 403–410, [http://dx.doi.org/10.1016/S0022-2836\(05\)80360-2](http://dx.doi.org/10.1016/S0022-2836(05)80360-2)
- Bakker C (1980) On the distribution of *Gonionemus vertens* A. Agassiz (Hydrozoa, Limnomedusae), a new species in the eelgrass beds of Lake Grevelingen (SW Netherlands). *Hydrobiological Bulletin* 14: 186–195, <http://dx.doi.org/10.1007/BF02260120>
- Bax N, Williamson A, Aguero M, Gonzalez E, Geeves W (2003) Marine invasive alien species: a threat to global biodiversity. *Marine Policy* 27: 313–323, [http://dx.doi.org/10.1016/S0308-597X\(03\)00041-1](http://dx.doi.org/10.1016/S0308-597X(03)00041-1)
- Bridge D, Cunningham CW, DeSalle R, Buss LW (1995) Class-level relationships in the Phylum Cnidaria: molecular and morphological evidence. *Molecular Biology and Evolution* 12(4): 679–689
- Carman MR, Bullard SG, Rocha RM, Lambert G, Dijkstra JA, Roper JJ, Goodwin A, Baker EM, Carman MM (2011) Ascidians at the Pacific and Atlantic entrances to the Panama Canal. *Aquatic Invasions* 6: 371–380, <http://dx.doi.org/10.3391/ai.2011.6.4.02>
- Edwards C (1976) A study in erratic distribution: the occurrence of the medusa *Gonionemus* in relation to the distribution of oysters. *Advances in Marine Biology* 14: 251–284, [http://dx.doi.org/10.1016/S0065-2881\(08\)60448-4](http://dx.doi.org/10.1016/S0065-2881(08)60448-4)
- Fofonoff PW, Ruiz GM, Steves B, Carlton JT (2003) California Non-native Estuarine and Marine Organisms (Cal-NEMO) System. <http://invasions.si.edu/nemesis/> (accessed 25-Oct-2016)
- Folmer O, Black M, Hoeh W, Lutz R, Vrijenhoek R (1994) DNA primers for amplification of mitochondrial cytochrome c oxidase I from diverse metazoan invertebrates. *Molecular Marine Biology and Biotechnology* 3(5): 294–299
- Galil BS, Boero F, Campbell ML, Carlton JT, Cook E, Fraschetti S, Gollasch S, Hewitt CL, Jelmert A, Macpherson E, Marchini A (2015) 'Double trouble': the expansion of the Suez Canal and marine bioinvasions in the Mediterranean Sea. *Biological Invasions* 17: 973–976, <http://dx.doi.org/10.1007/s10530-014-0778-y>
- Gasith A, Gafny S, Hershkovitz Y, Goldstein H, Galil BS (2011) The invasive freshwater medusa *Craspedacusta sowerbii* Lankester, 1880 (Hydrozoa: Olindiidae) in Israel. *Aquatic Invasions* 6 (Suppl 1): S147–S152, <http://dx.doi.org/10.3391/ai.2011.6.s1.033>
- Govindarajan AF, Carman MR (2016) Possible cryptic invasion of the Western Pacific toxic population of the hydromedusa *Gonionemus vertens* (Cnidaria: Hydrozoa) in the Northwestern Atlantic Ocean. *Biological Invasions* 18: 463–469, <http://dx.doi.org/10.1007/s10530-015-1019-8>
- Harrison GF, Kim K, Collins AG (2013) Low genetic diversity of the putatively introduced, brackish water hydrozoan, *Blackfordia virginica* (Leptothecata: Blackfordiidae), throughout the United States, with a new record for Lake Pontchartrain, Louisiana. *Proceedings of the Biological Society of Washington* 126: 91–102, <http://dx.doi.org/10.2988/0006-324X-126.2.91>
- Hewitt CL, Campbell ML, Thresher RE, Martin RB, Boyd S, Cohen BF, Currie DR, Gomon MF, Keough MJ, Lewis JA, Lockett MM (2004) Introduced and cryptogenic species in Port Phillip Bay, Victoria, Australia. *Marine Biology* 144: 183–202, <http://dx.doi.org/10.1007/s00227-003-1173-x>
- Hulme P (2009) Trade, transport and trouble: managing invasive species pathways in an era of globalization. *Journal of Applied Ecology* 46: 10–18, <http://dx.doi.org/10.1111/j.1365-2664.2008.01600.x>
- Katsanevakis S, Zenetos A, Belchior C, Cardoso AC (2013) Invading European Seas: assessing pathways of introduction of marine aliens. *Ocean and Coastal Management* 76: 64–74, <http://dx.doi.org/10.1016/j.ocecoaman.2013.02.024>
- Lavoie D, Smith L, Ruiz G (1999) The potential for intra-coastal transfer of non-indigenous species in the ballast water of ships. *Estuarine and Coastal Shelf Science* 48: 551–564, <http://dx.doi.org/10.1006/ecss.1999.0467>

- Muirhead JR, Minton MS, Miller WA, Ruiz GM (2015) Projected effects of the Panama Canal expansion on shipping traffic and biological invasions. *Diversity and Distributions* 21: 75–87, <http://dx.doi.org/10.1111/ddi.12260>
- Rodriguez CS, Pujol MG, Mianzan HW, Genzano GN (2014) First record of the invasive stinging medusa *Gonionemus vertens* in the southern hemisphere (Mar del Plata, Argentina) / Primer registro de la medusa urticante invasora *Gonionemus vertens* en el hemisferio sur (Mar del Plata, Argentina). *Latin American Journal of Aquatic Research* 42: 653, <http://dx.doi.org/10.3856/vol42-issue3-fulltext-23>
- Schuchert P (2016) *Gonionemus vertens* A. Agassiz, 1862. In: Schuchert P. World Hydrozoa database. Accessed through: World Register of Marine Species at <http://www.marinespecies.org/aphia.php?p=taxdetails&id=117768> on 2016-10-25
- Sievers F, Wilm A, Dineen DG, Gibson TJ, Karplus K, Li W, Lopez R, McWilliam H, Remmert M, Söding J, Thompson JD, Higgins D (2011) Fast, scalable generation of high-quality protein multiple sequence alignments using Clustal Omega. *Molecular Systems Biology* 7: 539, <http://dx.doi.org/10.1038/msb.2011.75>
- Tambs-Lyche H (1964) *Gonionemus vertens* L. Agassiz (Limnomedusae)—A zoogeographical puzzle. *Sarsia* 15: 1–8, <http://dx.doi.org/10.1080/00364827.1964.10409521>
- Thomsen M, Wernberg T, Schiel D (2015) Invasions by non-indigenous species. In: Crowe TP, Frid CL (eds), *Marine Ecosystems: Human Impacts on Biodiversity, Functioning and Services*. Cambridge University Press, pp 274–331, <http://dx.doi.org/10.1017/cbo9781139794763.010>
- Winnepenninckx B, Backeljau T, Dewachter R (1993) Extraction of high molecular weight DNA from mollusks. *Trends in Genetics* 9: 407, [http://dx.doi.org/10.1016/0168-9525\(93\)90102-N](http://dx.doi.org/10.1016/0168-9525(93)90102-N)

Supplementary material

The following supplementary material is available for this article:

Table S1. Records of the hydrozoan *Gonionemus vertens* from New Jersey, USA.

This material is available as part of online article from:

http://www.reabic.net/journals/bir/2016/Supplements/BIR_2016_Gaynor_etal_Supplement.xls