The following appendix accompanies the article

Differentiating successful and failed molluscan invaders in estuarine ecosystems

A. Whitman Miller[1,]*, Gregory M. Ruiz[1], Mark S. Minton[1], Richard F. Ambrose[2]

[1]Smithsonian Environmental Research Center, PO Box 28, Edgewater, Maryland 21037, USA

[2]Environmental Science and Engineering Program, Box 951772, University of California, Los Angeles, California 90095, USA

* Email: millerw@si.edu

Mar Ecol Prog Ser 332:041–051

APPENDIX 1. METHODS USED FOR ANALYZING VARIABLES

The following are detailed descriptions of the data incorporated in a multiple logistic regression which was used to differentiate successful and failed molluscan invaders of San Francisco Bay, California, USA. Additionally, an outline of the mathematical combinations approach used is included. Literature Cited includes all references from Appendices 1 & 2.

Categorical and ranked variables

Salinity zone classes

Typically, organisms do not span the entire salinity spectrum from 0 to ≥35 ‰. Instead, some species are confined to estuaries (true estuarine species) while others are primarily marine or freshwater organisms that stray into parts of the estuary. Animals are often described as stenohaline (occurring in a narrow range of salinities) or euryhaline (occurring in a wide range of salinities). Species were assigned to 4 categories based on each one's range of salinity tolerance and whether the organism is confined to estuarine conditions. The 4 categories included: (1) stenohaline-marine (a salinity range spanning 15 ‰ or less and including marine conditions of 32 ‰ or more), (2) stenohaline-estuarine (a salinity range spanning 15 ‰ or less, but confined to estuarine conditions), (3) euryhaline-marine (a salinity range spanning 16 ‰ or more, including marine conditions), and (4) euryhaline-estuarine (a salinity range spanning 16 ‰ or more, but confined to estuarine conditions). Salinity zone was dummy coded with stenomarine as the reference category.

Developmental mode

Each species was categorized according to its developmental mode. Bivalves and gastropods were listed as either planktonic developers (i.e. with planktonic eggs and/or larvae) or direct developers that lack a planktonic stage. Direct developer = 1, planktonic = 0.

Feeding mode

Bivalves were divided into suspension feeders, deposit feeders, or suspension and deposit feeders (i.e. organisms that can feed both ways). Feeding modes varied more broadly among gastropods and ranged from carnivores and ectoparasites to herbivores, scavenger/detritivores, and suspension feeders. Due to the possibility of multiple feeding modes, species were scored according to the presence or absence of each mode.

Substrate preference

Molluscs were divided into 3 substrate utilization categories: hard, soft, or both hard and soft substrates. Due to the possibility of multiple substrate usage, species were scored according to the presence or absence of each substrate preference.

Substrate and depth diversity

In addition to the general categorical substrate designation (soft, hard, soft and hard substrates), the diversity of benthic substrates (e.g. sand, mud, oyster shells, rock, wood, etc.) where each species naturally occurs was compiled and the range of substrates approximated by numerical index (1–5), where the index equals the number of substrates occupied. Species with very narrow substrate requirements received low numbers and generalists were assigned higher index values. Similarly, an index for depth diversity was assigned to each species (1–4). Subtidal species with broader depth ranges were given higher rankings than subtidal species with narrower depth distributions. Diversity scores are based on presence in various depth intervals. Species were given 1 point for each of the following categories: intertidal, 0–25 m depth, >25–100 m depth, >100 m depth (e.g. a species that occurs intertidally and subtidally to a depth of 35 m received a depth diversity index of 3, where the index equals the number of categories occupied).

Tidal height distribution

The occurrence with respect to tidal height was categorized for each molluscan species as intertidal, subtidal, or both. Species were therefore scored according to the presence or absence in each category.

Benthic placement

Each bivalve species was designated as either infaunal or epifaunal (0 or 1). Being mobile organisms, gastropods were not categorized for benthic placement.

Biogeographical faunal province of origin

Oyster associates were classified into 3 categories, according to their biogeographical faunal provinces of origin. These faunal provinces are based on the western Atlantic shallow water mollusc categorizations of Franz & Merrill (1980a,b). Franz & Merrill's Boreal and Arctic categories have been combined here as 'Northern.' The 3 faunal provinces used are as follows: (1) Northern = species whose northern and southern geographic boundaries lie to the north of Cape Cod, MA, and Cape Hatteras, NC, respectively. The Northern category encompasses species that are considered arctic or boreal. (2) Northern Transhatteran = species ranging to the north of Cape Cod and to the south of Cape Hatteras, NC. (3) Southern Transhatteran = species with northern limits do not extend north of Cape Cod and southern geographic boundaries that extend south of Cape Hatteras. The native faunal group was dummy coded with Northern-TransHatteran as the reference category.

Amphi-Atlantic distribution

Species whose natural distribution includes the eastern and western Atlantic are amphi-Atlantic species. Each species was categorized as either amphi-Atlantic or not (1 or 0). Species believed non-native to the western Atlantic (i.e. introduced by humans in historical times) are not treated as amphi-Atlantic species.

Continuous numerical variables

Latitudinal range

The spatial distribution along the western Atlantic coast was determined for each of the molluscan species analyzed; southern limit, northern limit are reported in terms of degrees latitude. In general, the geographic extents of bivalve species were based on distribution descriptions from Abbott (1974). Northern and southern geographic ranges were converted to latitude for analysis. The latitudinal ranges of gastropods were taken from the Academy of Natural Science of Philadelphia's online database Malacolog 4.1 (Rosenberg 2005). In a few instances, published accounts of a species' range that superseded the above references were used. In cases in which the geographic location of a range boundary was not specified precisely (e.g. Nova Scotia), the named location's northern and southern limits were averaged, and this was operationally considered as the species' range boundary.

Maximum depth

Maximum depth for living organisms was reported in meters below the water's surface.

Salinity distribution: lower, upper limits, range

The lower salinity limits for the adult stages of bivalves and gastropods were gleaned from the literature. Care was taken to report only lower salinity ranges experienced by organisms in their native habitats rather than in laboratory situations. The extreme variability of acclimatization regimes and tolerance endpoints (e.g. LD_{50}, loss of ciliary movement) made comparisons among laboratory based experiments untenable. Each species' upper and lower salinity tolerance limits were reported in parts-per-thousand (‰). Although field distributions do not necessarily indicate absolute tolerance limits, they probably reflect natural conditions that are typical for successful growth and reproduction.

Shell length

Maximum adult shell length was estimated in millimeters. Shell sizes were obtained primarily from Abbott (1974), Gosner (1971, 1978), Rehder (1997) and Malacolog 4.1 online database (Rosenberg 2005). Shell sizes were log transformed to approximate better a normal distribution before analysis.

Historical abundance (1862 to 1910 surveys)

Abundance estimates were based on historical surveys conducted in the waters surrounding New York City and vicinity (the source of oysters shipped to the west coast of the United States and northwestern Europe) between 1862 and 1899, corresponding to the time of peak oyster transplantation (Smith 1862, 1887, Hubbard & Smith 1865, Perkins 1869a,b, Smith & Prime 1870, Balch 1899). In these studies, species abundance was ranked qualitatively as ‘absent’, ‘rare’, ‘uncommon’, ‘common’, and ‘abundant.’ Modifiers such as ‘very’, ‘extremely’, ‘moderately’, and ‘locally’ were sometimes applied. While impossible to know how closely calibrated various authors’ estimates were, all employed ranked categories to estimate abundance. As the author of 4 of the 6 studies used here, Sanderson Smith likely brings consistency to the Staten Island and Long Island region estimates. To calculate a mean abundance score for each species, a numerical ranking system was developed to describe each of the 11 qualitative abundance categories identified in the historical surveys (Miller 2000). The numerical rankings range from 0 (absent) to 10 (extremely abundant). Use of a linear abundance scale is a conservative approach, underestimating the true magnitude of differences between low and high abundance, but allowing for a robust test of abundance as a predictor of invasion success. Site-specific abundance estimates were compiled for each species by historical source, including all locations surveyed, and then averaged across sources. Since abundance values are integrated across locations for the time-period of interest, they represent an estimate of the average relative abundance of oyster associates at the time. By extension, abundance represents the relative availability of these organisms (both juvenile and adult life stages) for uptake as bycatch in dredges and subsequent transport with oysters.

Mathematical combinations

Mathematical combinations are same-sized groups with unique membership. The general formula for calculating a combination number is $C(n,k)=n!/k!(n-k)!$. The formula calculates the number of unique ways that groups of k elements can be taken from a set containing n elements (Anderson et al. 1994). The following example demonstrates their use when a pool consisting of 5 letters from the alphabet is used ($n = 5$).

Question: If 5 letters (A, B, C, D, E) are placed in a box, what is the probability that a random grab of 4 letters will contain the 2 letter combination AB?

The following is a comprehensive list of 4 letter combinations that can be pulled from the pool of 5 letters: ABCD, ABCE, ABDE, ACDE, and BCDE. By inspection, one can see that AB is included in 3 of 5 combinations. Thus, AB could be expected to occur in 4 letter combinations 60% of the time.

Solving the problem using combinatorial mathematics requires 3 steps. (1) Calculating the total number of 4 letter combinations that can be taken from a 5 letter set. (2) Calculating the subset of these 4 letter combinations that would contain 2 particular letters (AB). (3) Dividing Step 2 by Step 1.

Step 1:

The total number of 4 letter combinations (*C*) chosen from 5 letters is:

$$C(5,4) = \frac{[5!]}{[4!(5-4)!]} = 5 \quad (1)$$

Step 2:

Since the combinations of interest must include the 2 letter combination AB, these letters can be subtracted from the pool of 5 letters (A,B,C,D,E becomes C,D,E ($n = 3$)). Likewise, the set size chosen from the new pool will also decrease by 2 letters ($k = 4$ becomes $k = 2$). The goal is to calculate the number of 2 letter combinations that can be taken from a pool of 3 letters (C,D,E) such that when the 2 letter combination AB is added to each, the resulting list represents the total number of 4 letter combinations that include AB. This can be represented in the following equation.

$$C(5-2,4-2) = C(3,2) = \frac{[3!]}{[2!(3-2)!]} = 3 \qquad (2)$$

Therefore, there are three 2 letter combinations, that when linked to the AB combination, represent the total number of 4 letter combinations containing AB. These combinations are CD, CE, DE, and become ABCD, ABCE, and ABDE when AB is added. By inspection, these are the same combinations as found above.

Step 3:

The probability of choosing a combination of 4 letters that contains AB from a pool of 5 letters is:

$$\frac{C(5-2,4-2)}{C(5,4)} = \frac{\frac{3!}{2!(3-2)!}}{\frac{5!}{4!(5-4)!}} = 0.6 \qquad (3)$$

LITERATURE CITED

Abbott RT (1974) American seashells: the marine molluska of the Atlantic and Pacific Coasts of North America, 2nd edn. Van Nostrand Reinhold Company, New York, NY

Anderson DR, Sweeney DJ, Williams TA (1994) Introduction to statistics: concepts and applications, 3rd edn. West Publishing Company, New York, NY

Andrews J (1953) Fouling organisms of Chesapeake Bay Inst. Inshore Survey Program, Ref. 53-3, Interim Report 17

Balch FN (1899) List of marine mollusca of Coldspring Harbor, Long Island, with descriptions of two new species of nudibranchs. Boston Soc Nat Hist Proc 29:133–162

Barnes RSK (1994) The brackish-water fauna of Northwestern Europe. Cambridge University Press, Cambridge

Belding DL (1912) Report upon the scallop fishery of Massachusetts. Wright and Potter Printing Co., Boston, MA

Carriker ML (1955) Critical review of biology and control of oyster drills *Urosalpinx* and *Eurpleura.* US Fish Wildl Serv Spec Sci Rep 148:1–50

Castagna M, Chanley P (1973) Salinity tolerance of some marine bivalves from inshore and estuarine environments in Virginia waters on the western Mid-Atlantic coast. Malacologia 12(1):47–96

Chanley P, Andrews JD (1971) Aids for identification of bivalve larvae of Virginia. Malacologia 11(1):45–119

Chestnut AP (1980) Distribution, population dynamics, and reproductive biology of the burrowing clam, *Diplothyra smithii* Tryon. PhD dissertation, University of Southern Mississippi, Hattiesburg, MS

Figley B (2003) Marine life colonization of experimental reef habitat in temperate ocean waters of New Jersey. NJ Department of Environmental Protection, Division of Fish and Wildlife, Bureau of Marine Fisheries, Trenton, NJ

Flores-Andolais F, Garcia-Cubas A, Toledano-Granados A (1988) Sistematica y algunos aspectos ecologicos de los moluscos de la Laguna de la Mancha, Veracruz, Mexico. An Instituto Cienc Mar Limnologia, Univ Nac Auton Mex 15(2):235–258

Fofonoff PW, Ruiz GM, Steves B, Hines AH, Carlton JT (2005) National Exotic Marine and Estuarine Species Information System. http://invasions.si.edu/nemesis/. Access date: 27 June 2005

Fox R (2004) Invertebrate anatomy online *Diodora cayenensis* ©Keyhole Limpet web site, http://www.lander.edu/rsfox/310DiodoraLab.html. Access date: 01 July 2004

Franz DR (1973) The ecology and reproduction of a marine bivalve *Mysella planulata* (Erycinacea). Biol Bull 144:93–106

Franz DR, Merrill AS (1980a) Molluscan distribution patterns on the continental shelf of the middle Atlantic Bight (Northwest Atlantic). Malacologia 19:209–225

Franz DR, Merrill AS (1980b) The origins and determinants of distribution of molluscan faunal groups on the shallow continental shelf of the Northwestern Atlantic. Malacologia 19:227–248

Frey DG (1946) Oyster bars of the Potomac River. US Fish Wildl Serv Spec Sci Rep 32:1–93

Galtsoff PS (1964) The American oyster *Crassostrea virginica* Gmelin. US Fish Wildl Serv Fish Bull 64:1–480Gosner KL (1971) Guide to identification of Marine and Estuarine Invertebrates. Wiley-Interscience, New York, NY, pp 693

Gosner KL (1978) A field guide to the Atlantic Seashore from the Bay of Fundy to Cape Hatteras. Houghton Mifflin Company, Boston, MA

Hubbard JW, Smith S (1865) Catalogue of the mollusca of Staten Island, N.Y. Ann Lyceum Nat Hist (NY) 8:151–154

Hughes RN (1986) A functional biology of marine gastropods. The Johns Hopkins University Press, Baltimore, MD

Larsen PF (1985) The benthic macrofauna associated with the oyster reefs of the James River Estuary, Virginia, USA. Int Rev Ges Hydrobiol 70:797–814

Leathem W, Maurer D (1975) The distribution and ecology of common marine and estuarine gastropods in the Delaware Bay area. Nautilus 3:73–79

Lippson AJ, Lippson RL (1997) Life in the Chesapeake Bay, 2nd edn. The Johns Hopkins University Press, Baltimore, MD

Loosanoff VL, Davis HC, Chanley PE (1966) Dimensions and shapes of larvae of some marine bivalve mollusks. Malacologia 4(2):351–435

Marsh GA (1976) Ecology of gastropod epifauna of eelgrass in a Virginia estuary. Chesapeake Sci 17(3):182–187

Maurer D, Watling L (1973) The biology of the oyster community and its associated fauna in Delaware Bay. Delaware Bay Rep Ser 6:1-97

Menzel RW (1964) Checklist of the marine flora and fauna of the St. George's Sound area. Oceangr. Inst., Florida State University, Tallahassee, FL

Micheli F, Peterson CH (1999) Estuarine vegetated habitats as corridors for predator movements. Conserv Biol 13:869–881

Mikkelsen, PS, Mikkelsen PM (1984) Comparison of *Acteocina canaliculata* (Say, 1826), *A. candei* (d'Orbigny, 1841), and *A. atrata* spec. nov. (Gastropoda: Cephalaspidea). Veliger 27(2):164–192

Miller AW (2000) Assessing the importance of biological attributes for invasion success: eastern oyster *(Crassostrea virginica)* introductions and associated molluscan invasions of Pacific and Atlantic coastal systems. DEnv dissertation, University of California, Los Angeles, CA

NOAA NMSP (2005) Benthic fauna of Gray’s Reef National Marine Sanctuary web site http://www.seamonsters.noaa.gov/creatures.aspx. Access date: 27 June 2005

Pechenik JA (1986) Field evidence for delayed metamorphosis of larval gastropods: *Crepidula plana Say*, *C. fornicata* (L.), and *Bittium alternatum Say*. J Exp Mar Biol Ecol 97:313–319

Perkins GH (1869a, b) Molluscan fauna of New Haven. A critical review of all marine, freshwater and land molluscs of the region, with descriptions of many of the living animals and of two new species. Part I. Cephalapoda and Gasteropoda and Part II. Acephala and Bryozoa. Boston Soc Nat Hist Proc 13:109–135 and 139–163

Rehder HA (1997) National Audubon Society field guide to North American seashells. Alfred A. Knopf, New York, NY

Robertson R (1978) Spermatophores of six eastern North American pyramidellid gastropods and their systematic significance (with the new genus *Boonea*). Biol Bull 155:360–382

Robertson R (1994) Wentletrap egg capsules and veligers: what they are and how to see and study them. Am Conch 22(4):5–6

Robertson R (1996) A little-known Atlantic and Gulf coast American odostomian (Pyramidellidae) and its generic relationships. Am Malacol Bull 13(1-2):11–21

Robertson R, Mau-Lastovicka T (1979) The ectoparasitism of *Boonea* and *Fargoa* (Gastropoda: Pyramidellidae). Biol Bull 157:320–333

Rosenberg G (2005) Malacolog 4.1: a database of western Atlantic marine mollusca. Available at: www.malacolog.org

Ruppert E, Fox R (1988) Seashore animals of the Southeast. University of South Carolina Press, Columbia, SC

Scheltema RS (1984) Development and planktonic larvae of common benthic invertebrates of the Woods Hole Massachusetts region: summary of existing data and bibliographic sources. Woods Hole Oceanog Inst Tech Rept WHOI-84-13

Smith S (1862) On the Mollusca of Peconic and Gardiner’s Bays, Long Island, New York. Ann Lyceum Nat Hist (NY) 7:147–167

Smith S (1887) Catalogue of the mollusca of Staten Island. Nat Sci Assoc Staten Island Part 1 (Extra No. 5):53

Smith S, Prime T (1870) Report on the mollusca of Long Island, N.Y., and of its dependencies. Ann Lyceum Nat Hist (NY) 9:377–407

Sokolova IM (2000) Effects of salinity on the growth rate and survival of juvenile *Littorina saxatilis* gastropods from habitats with different salinity regimes. Russ J Mar Biol 26(6):427–431

StickleWB, Moore MN, Bayne BL (1985) Effects of temperature, salinity and aerial exposure on predation and lysosomal stability in the dog whelk *Thais* (*Nucella*) *lapillus* (L.). J Exp Mar Ecol 93:235–258

Sullivan CM (1948) Bivalve larvae of Malpeque Bay, PEI. Bull Fish Res Board Can 77:1–36

Thiriot-Quievreux C (1983) Summer meroplantonic Prosobranch larvae occurring off Beaufort, North Carolina. Estuaries 6(4):387–398

Verrill AE, Smith SI (1874) The invertebrate animals of Vineyard Sound and adjacent waters with an account of the physical features of the region. Rep US Comm Fish and Fisheries for 1871–1872. Government Printing Office, Washington, DC

Wass ML, Burrell WG, Zwerner D, Calder DG, Van Engel WG, Sandifer PA (1972) A checklist of the biota of Lower Chesapeake Bay with inclusions from the Upper Bay and the Virginian Sea. Va Inst Marine Sci Spec Rep 65:1–290

Wells HW (1961) The fauna of oyster beds, with special reference to the salinity factor. Ecol Monogr 31:239–266

Williams AB, Porter HJ (1971) A ten-year study of meroplankton in North Carolina estuaries: occurrence of postmetamorphal bivalves. Chesapeake Science 12:26–32

Winkley HW (1888) Mollusca found in the oyster beds of Cocagne, N. B., Bedeque and Summerside, P.E.I. Bull Nat Hist Soc New Brunswick 7:69–71