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IN REPLY REFER TO:

June 29, 2011

To: Brannon Ketcham, Hydrologist, Point Reyes National Seashore
Project Manager

From: Raymond M. Sauvajot, Natural Resource Program Chief, Pacific West Region
Peer Review Coordinator

Subject: Completion of Peer Review for Grosholz, "Estimating the Relative Abundance of
Naturalized Manila Clams and Invasive Fouling Species in Drake's Estero"

As peer review coordinator for the subject report, I am writing to confirm that the peer review process for this report has been completed consistent with National Park Service Interim Guidance for Peer Review (January 31, 2008) and Director's Order #11B. In particular, I coordinated the peer review for the report and submitted, through you, resulting peer review comments to the report author. Three anonymous peer reviews were obtained for the report. In general, suggested edits and revisions from the peer reviewers to the author were relatively straightforward, and were satisfactorily addressed by the author in a revised final report submitted to you. Consequently, I believe that the report should now be considered "final" with National Park Service peer review completed.

If you have any questions about the peer review process, please let me know. I hope the review process was helpful in assessing the utility and relevance of this report.

Raymond M. Sauvajot, Ph.D.

Cc: Cicely Muldoon, Superintendent, PORE
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Report to National Park Service

OP Fund # 42498

**Estimating the Relative Abundances of Naturalized Manila Clams
and Invasive Fouling Species in Drakes Estero**

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Introduction and Rationale for Project

I report on the results of two studies conducted over the past several months from August – December 2010 in Drakes Estero, CA. The first study has as its aim to determine if the invasive Manila clam (*Venerupis philippinarum*) has developed naturalized populations in Drakes Estero. Manila clams have been raised in bag culture in new areas within Drakes Estero in the past few years. There is currently no record of Manila clams being established in Drakes Estero, although definitive studies have never been conducted. This report documents the first evidence consistent with Manila clams being established in this system.

The second study was to examine the community of fouling species inhabiting hard substrate provided by oyster racks and comparing this with surrounding habitats including rock substrate and eelgrass. The primary question concerns the degree to which oyster racks provide additional or even novel habitat to non-indigenous fouling species. Several studies in other systems have shown that artificial hard substrate can provide novel habitat for fouling species like this (Glasby et al. 2007, Marzinelli et al. 2009). This report documents the relative abundance of some common fouling species, particularly the invasive tunicate *Didemnum vexillum* on oyster racks and in adjacent seagrass beds.

Manila Clams

We conducted two days of surveys of mudflats in the vicinity of Bull Point (Table 1). These efforts consisted of examining mudflats by kayak to identify likely habitat for Manila clams. We chose three distinct mudflats in the vicinity of Bull Point (Figure 1). Sites were chosen based on rapid visual assessment of substrate grain size, aspect, tidal elevation, presence or absence of key benthic species and other factors known to represent appropriate habitat for Manila clams (Whiteley and Bendell-Young 2007, Tsai et al. 2010). They were also chosen based on the presence of commercial clam bags with Manila clams (Mudflat 1) and without clam bags (Mudflat 2 and 3).

At each mudflat, we chose a tidal elevation believed to be consistent with the observed tidal distribution of Manila clams (Grosholz et al. 2001). Parallel to the tide line, we laid out a 30 m transect line and selected six random points along this. At each of the six locations, we placed a plastic coring frame (approximately 0.55 x 0.35 x 0.20 m LxWxD) and excavated each with a shovel. Two persons were involved in sieved sediments through a 2 mm bucket sieve and all bivalves were removed, identified to species and measured to the nearest mm. These are long established sampling methods that have been shown to adequately quantify the distribution and abundance of Manila clams (Whiteley and Bendell-Young 2007).

Our samples included individuals other bivalves such as *Mya arenaria* and *Macoma* spp. which are typical of this type of site and tidal elevation. We also found clear evidence of the presence of a naturalized population of Manila clams in Drakes Estero. Our data collection from Mudflat 3 revealed three live juvenile Manila clams (9-11 mm maximum shell length) and one adult (45 mm) (Table 1). This size range and density for the juvenile clams strongly suggests that these clams recruited in this area within the past year based on growth rates of Manila clams in similar habitats in the region (Grosholz et al. 2001, Tsai et al. 2010). Therefore, it seems highly unlikely that the juvenile Manila clams in our transects could possibly be 'escapees' from clam bags present in previous years at this site. There were no clam bags present at the time of sampling at this mudflat and from past records no clam bags were ever present at Mudflat 3 (B. Becker, pers. comm.). The very low density of Manila clams in our samples at densities (approximately 5 /m²) also suggests that this population, if naturalized, has only become so recently. In other west coast sites where Manila clams have become established, densities often reach 100-200 or more per m² (Whiteley and Bendell-Young 2007, Tsai et al. 2010). These data and the resulting conclusions represent only a preliminary look at the degree to which these clams have become naturalized in the Drakes Estero

It would be expected that Manila clam density would increase over the next several years as would the mean size of clams as the size structure included adults. Manila clams are known to reach maturity (15-

20 mm) within the first year (Holland and Chew 1974), so significantly more adults would be expected within 1-2 years. It cannot be determined with certainty that these new colonists are the result of reproduction by Manila clams present in Drakes Estero. There is some possibility that clams in other estuaries such as San Francisco Bay or Tomales Bay given that the larval development period can be 3-4 weeks at local temperatures (Bourne 1982). However, Manila clams are not recorded from all bays and estuaries in this region (NEMESIS 2011), so connectivity is certainly limited. It is far more likely that the colonists were spawned locally, but this would require considerably more research to determine their origin.

Future work is strongly recommended to understanding the extent to which Manila clams and other commercial species are becoming, or have become, established in Drakes Estero. There is very little information regarding the impacts of introduced Manila clams on benthic habitats since their introduction into estuaries in western North America (Carlton 1999). However, recent studies in British Columbia have documented increased siltation, accumulation of organic matter and reduction of species richness in Manila clam production sites using predator control netting (Bendell-Young 2006). More recent studies looking specifically at clam effects found higher levels of ammonium, organic matter and silt and other changes in sediment biogeochemistry at sites where Manila clams were farmed in comparison with control sites (Bendell et al. 2010). Whether similar changes to the habitat would apply to the specific farming practices for Manila clams in Drakes Bay remains to be determined.

Other potential impacts of bivalve aquaculture in Drakes Estero involve the establishment of the Pacific oyster *Crassostrea gigas*, which has become more widely distributed in this region. In San Francisco Bay, *C. gigas* has become naturalized within the past ten years (Goodwin et al. 2010). Also within the past decade, isolated adult Pacific oysters have been found in Tomales Bay (E. Grosholz and D. Kimbro, pers. obs.) and recently (2010-2011) at Bull Point in Drakes Estero (E. Grosholz and B. Cheng, pers. obs.). Despite this species being cultured throughout California, Pacific oysters have not become naturalized in many regions. Its establishment in these new areas may have negative impacts on several native species including the native Olympia oyster in Drakes Estero. The status of Olympia oysters and their historic presence in Drakes Estero is currently being investigated (Konzak and Praetzelis 2011).

Fouling Community on Oyster Racks

We surveyed oyster racks in Drakes Estero on three dates in September-October 2010 in the vicinity of Bull Point and in the vicinity of Drakes Bay Oyster Company (Table 2). We selected racks that represented 'working' oyster racks that were currently in production as well as 'non-working' racks (=not currently in use') that had intact rack

structures, but that were not currently in production. We also sampled the surrounding substrate including some rocky outcrops and eelgrass beds adjacent to the racks. We selected racks haphazardly based on ease of kayak deployment and accessibility for sampling.

At each rack we haphazardly selected sampling points to remove samples of fouling organisms. The culture method in this case consisted of stakes mounted onto wooden racks so samples included both oysters growing on stakes and rack structures including wooden supports and plastic spacers used to separate oyster groups. Two persons collected samples from small areas typically on the scale of 0.10 x 0.10 m. These samples were selected visually to capture the diversity of species in this habitat and no way reflect a quantitative sampling regime to estimate relative abundance or cover of species. However, these samples should provide information regarding the total number of species (species richness) in these habitats as a function of sampling effort. We attempted to sample relatively completely the well-developed fouling communities at each site. At each sampling point, we removed organisms by hand or using a paint scraper and placed the organisms immediately into gallon Ziploc bags. Organisms were kept in coolers and transported to labs at either UC Davis or the Romberg Tiburon Center where subsamples of organisms were fixed in either 70% ethanol (for subsequent genetic identification) or 10% formalin (for subsequent morphological identification) within a few hours of collection. All identification and quantification were conducted in the lab using standard taxonomic keys. Verification of initial ID by experts for particular taxa is pending.

We found an abundance of various fouling species of which *Didemnum vexillum* was prominent among them. This species was common on both 'working' and 'non-working' racks as well as on eelgrass (see below). The number of species of fouling organisms was slightly greater on the racks and was somewhat different in comparison with the eelgrass, although the area sampled was not always equivalent. We also found that the number of species on the older abandoned racks had a slightly different set of species than the working racks, for instance the presence of the introduced bryozoan *Watersipora subtorquata*. However, this pattern is confounded by the position along the estuary, since the 'non-working' racks were further towards the mouth of the estuary with more stable salinities.

Of real importance is our observation of large colonies of *Didemnum vexillum* growing on the distal portions of the leaf shoots of the native eelgrass *Zostera marina*. Our observations also documented that *D. vexillum* was overgrowing the hydrozoan *Obelia* sp. (status uncertain) growing within beds of benthic macroalgae. Although some eelgrass samples were also overgrown with a white sponge (species TBD), at least a small number of samples had *Didemnum* overgrowing several inches of the distal portion of the leaf shoots. This contrasts significantly with initial descriptions of its distribution of *Didemnum* along the Atlantic coast, which were limited to hard substrate (Bullard et al. 2007, Valentine et al. 2007, Carman et al. 2007), although more recent studies have also documented its distribution on eelgrass (Carman and Grunden 2010, Carman et al. 2010). This study is consistent

with the results of Fisk et al. (2005), which also found *D. vexillum* on oyster racks, however, they did not report this species growing on eelgrass at that point.

Considerable additional work is strongly recommended on this topic. The potential range of habitats that *Didemnum* and other invasive fouling species can use is very poorly known. A much more comprehensive survey is needed to provide convincing evidence of the role of oyster racks to support disproportionately large abundances of invasive species. Our data clearly show that most of the biomass of fouling species in these samples consists of invasive nonindigenous species. Our data also show significant fouling of eelgrass by *Didemnum*, which has also been demonstrated in the northeastern U.S. (Carman and Grunden 2010), although not noted elsewhere. There is a significant possibility of *Didemnum* having a substantial negative impact on eelgrass given the extent of fouling witnessed in Drakes Estero in just this limited sampling. This pattern should be investigated in other west coast estuaries to determine how widely eelgrass may be affected by this spreading invasion.

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Table 1. Dates and locations of mudflat samples taken to document Manila clam presence in Drakes Estero.

Date	Starting Site	Sample Location	Relative Elev.	Samples	Latitude	Longitude
8/11/2010	Bull Point	Mudflat 1	L	6	38.05825°	122.92967°
					38.05835°	122.93008°
8/12/2010	Bull Point	Mudflat 2	L	6	38.04878°	122.94363°
					38.04880°	122.94328°
8/12/2010	Bull Point	Mudflat 3	H	6	38.04792°	122.94333°
					38.04795°	122.94305°

Table 2. Dates, locations and substrates samples taken to describe fouling species on and adjacent to active and unused oyster racks in Drakes Estero.

Date	Starting Site	Sample Location	Latitude	Longitude	Substrate	# Samples
9/10/2010	Bull Point	Bull Point Rocky Bench	38.06117°	122.94423°	Rocky Bench	5
9/10/2010	Bull Point	Unused Rack 1	38.05962°	122.94620°	Rack	1
9/10/2010	Bull Point	Unused Rack 2	38.05892°	122.94633°	Rack	5
9/10/2010	Bull Point	Unused Rack 3	38.05930°	122.94693°	Rack	1
9/10/2010	Bull Point	Unused Rack 4	38.05988°	122.94292°	Rack	3
10/5/2010	DBOC	Active production rack 1	38.07605°	122.93813°	Oyster shell	18
10/5/2010	DBOC	Active production rack 1	38.07605°	122.93813°	Plastic spacer	4
10/5/2010	DBOC	Active production rack 1	38.07605°	122.93813°	Eelgrass	5
10/5/2010	DBOC	Active production rack 1	38.07605°	122.93813°	Rack	3
10/19/2010	DBOC	Active production rack 2	38.07030°	122.93556°	Oyster shell	4
10/19/2010	DBOC	Active production rack 3	38.06670°	122.93830°	Plastic spacer	1

1 **Figure 1. Map of sampling locations in Drakes Estero showing sites used for fouling species including 'Unused Racks' (yellow pins) and 'Active Racks' (red pins) as well as sites used for Manila clams (green pins).**

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