Evidence for *Perkinsus marinus* Resistance In Natural *Crassostrea virginica* Populations from Virginia Waters

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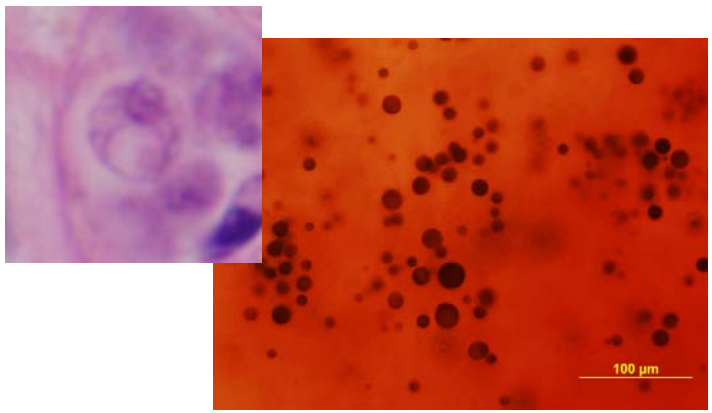
Definitions: “Resistance” vs. “Tolerance”

- **Roy and Kirchner 2000 (Evolution 54: 51-63):**
  - → **“Resistance”: host limits infection**
  - → **“Tolerance”: infection not limited, but fitness consequences reduced**
  - → **Parasite virulence is likely variable geographically (Bushek and Allen, Jr., 1996) and perhaps temporally, which complicates interpretations, but with *C. virginica*-*P. marinus* we may see both resistance and tolerance**

Perkinsus marinus

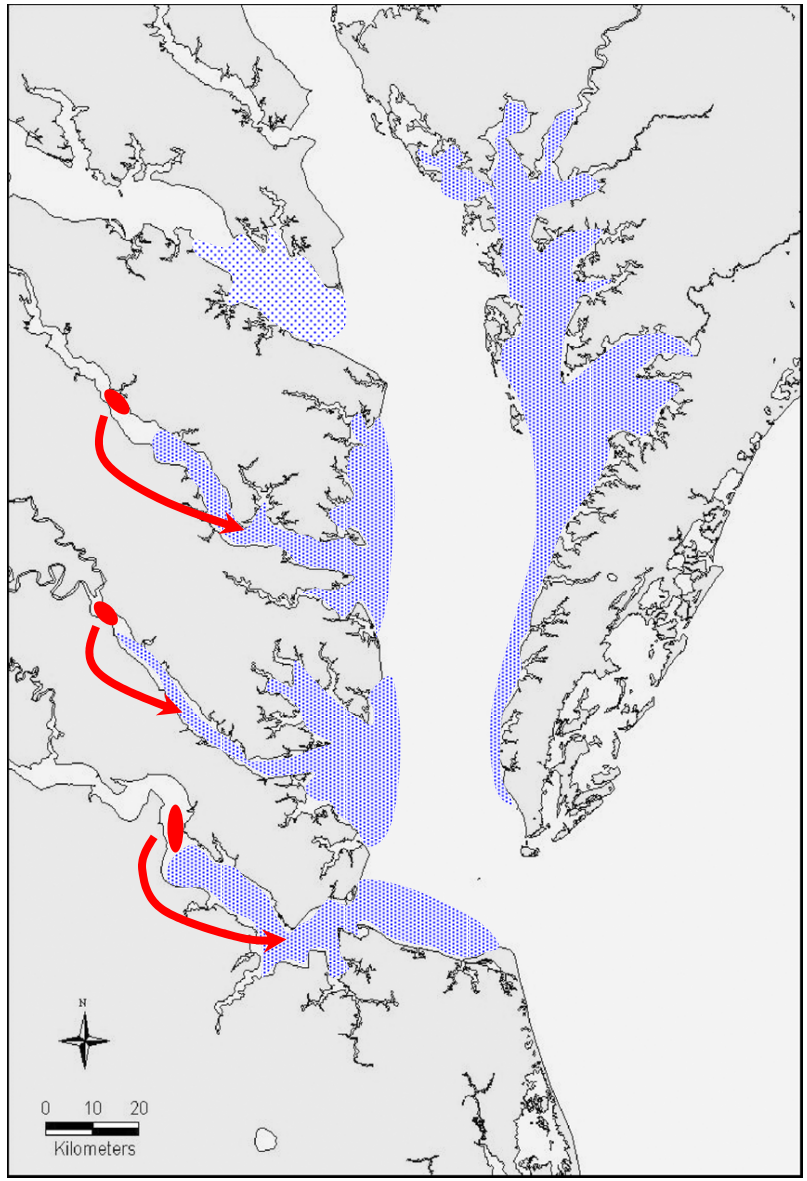
- Agent of perkinsosis or “dermo” disease, known since the 1940s in the Gulf of Mexico (Mackin et al. 1950) and the 1950s in Chesapeake Bay (Hewatt and Andrews 1954)
- *P. marinus* a protistan endoparasite, directly transmissible among oysters
  - → Acquired during feeding, released with feces and upon death & disintegration
- Dermo a wasting disease of the warmer months (temperatures > 20°C, and of waters > 12-15 psu (but *P. marinus* is tolerant of extended periods of lower salinity)
- Prevalence of *P. marinus* may be 100%, and dermo-caused mortality > 70%
- Considered a primary impediment to *C. virginica* aquaculture and restoration



Chronic Susceptibility

- **While resistance to *P. marinus* is evident in domesticated oyster strains (e.g., Andrews DEBY™; Ragone Calvo et al. 2003), natural populations are considered highly susceptible**
- **Sustained by either:**
  - → **(1) Recruitment derived from susceptible stocks in low salinity parasite refuges; or**
  - → **(2) Reproduction of small, susceptible and doomed but pre-dermo individuals (the industry perspective; *P. marinus* infections increasing with age, larger oysters are unable to contribute reproductively)**

Is this really how natural oyster populations function in dermo-enzootic waters?



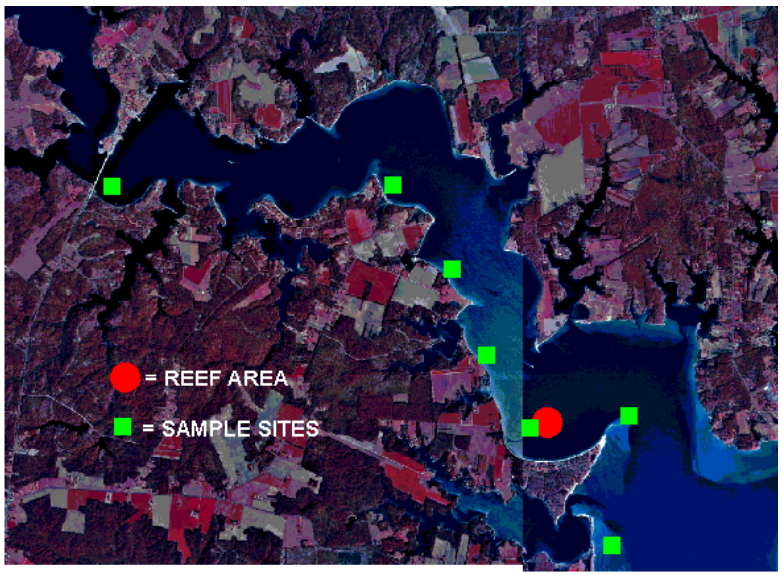
1) Are Natural Oyster Populations from Dermo Disease-Enzootic Waters Intolerant of *P. marinus*?

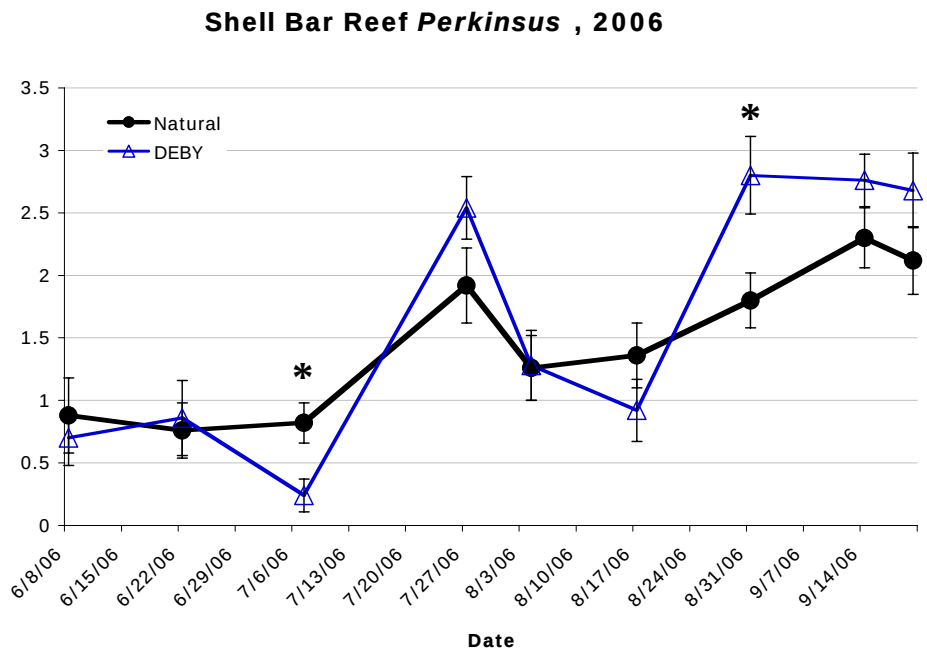
- **Field comparisons of a deployed domesticated, disease-resistant *C. virginica* stock (Andrews DEBY™) vs. naturally recruited oysters in a disease-enzootic location (GREAT WICOMICO RIVER FIELD ANALYSIS--2005/6)**

- **Field comparisons of Andrews DEBY™ oysters, naïve Ross Rock (Rappahannock River) oysters, and representatives from two disease-enzootic locations (Wreck Shoal in the James, Aberdeen Rock in the York)(YORK RIVER BEACH TRIAL--2006)**

Great Wicomico River, 2006

- **Millions of Andrews DEBY™ oysters deployed to Shell Bar Reef to examine the impact a broodstock addition will have on subsequent recruitment at a restoration site**
- ***P. marinus*, *H. nelsoni* parasitism assessed biweekly through 2006 in samples of deployed DEBYs and naturally recruited oysters**
- **H_0: No difference in weighted prevalence between DEBYs & natives**



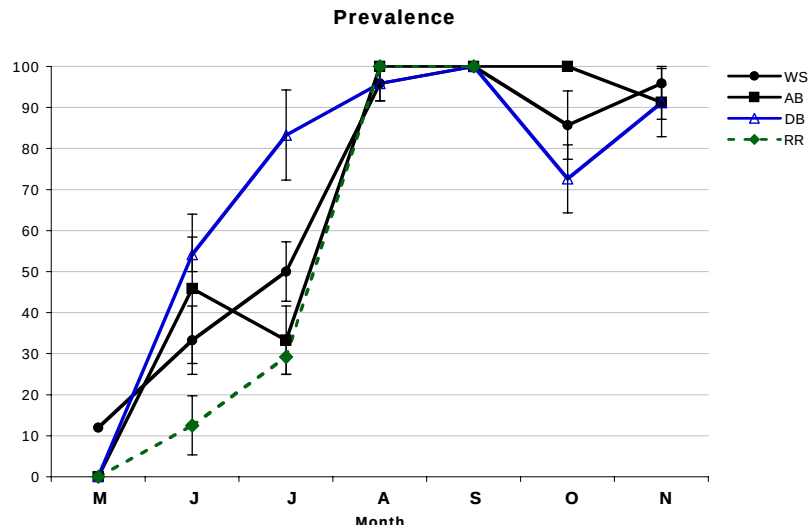


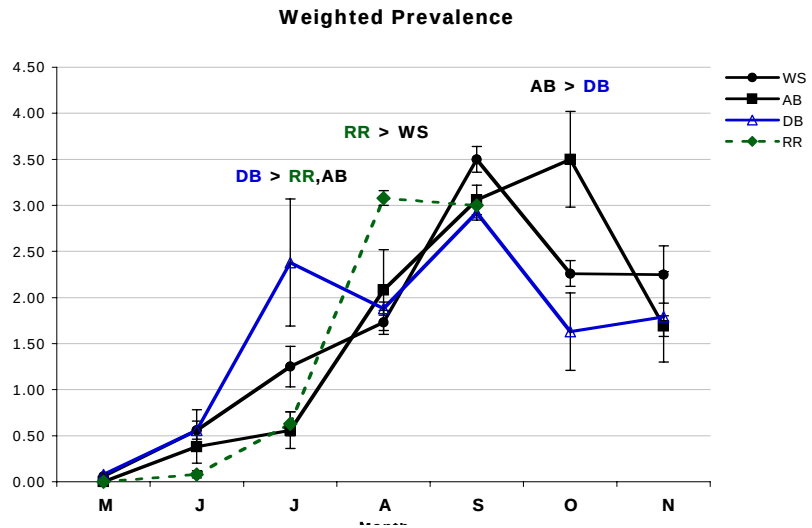
* **Kruskal-Wallis test significant at $\alpha = 0.05$**

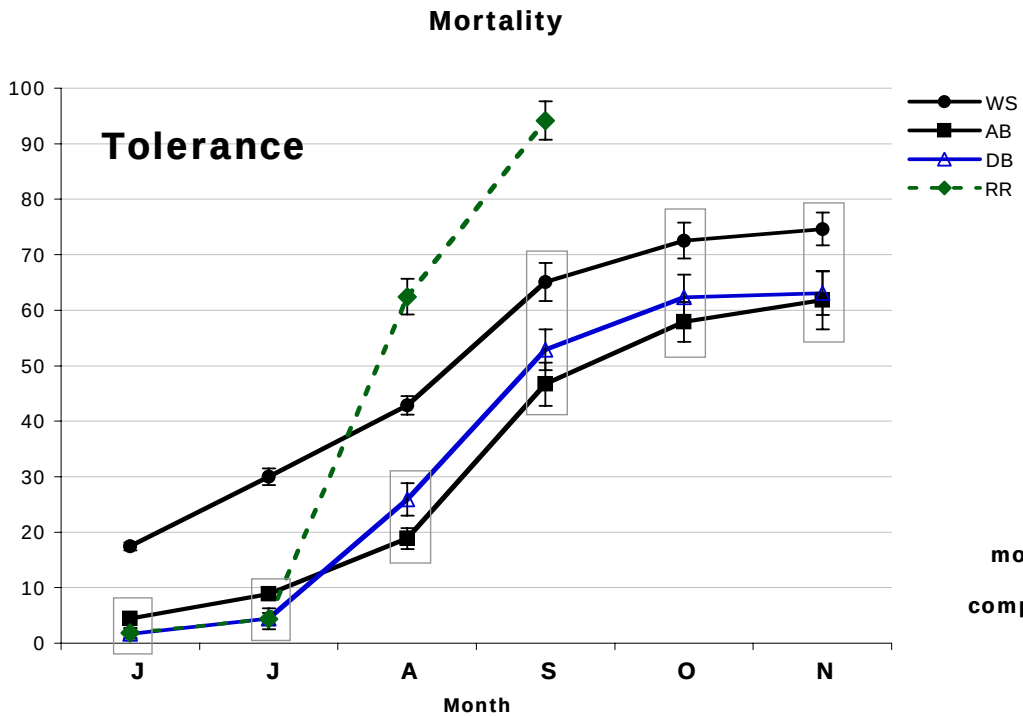
York River Beach Trial, 2006

- **Four wild-set oyster groups deployed in triplicate trays in the York River, April-May 2006:**
  - → **Wreck Shoal, James River (*P. marinus*-enzootic)**
  - → **Aberdeen Rock, York River (*P. marinus*-enzootic)**
  - → **Ross Rock, Rappahannock River (naïve oysters, *P. marinus* rarely observed)**
  - → **Andrews DEBY™ from Mobjack Bay (disease-resistant)**
- **Monthly evaluation of growth, mortality, disease**
- **H_0: No differences among stocks**

York River Beach Trial, 2006



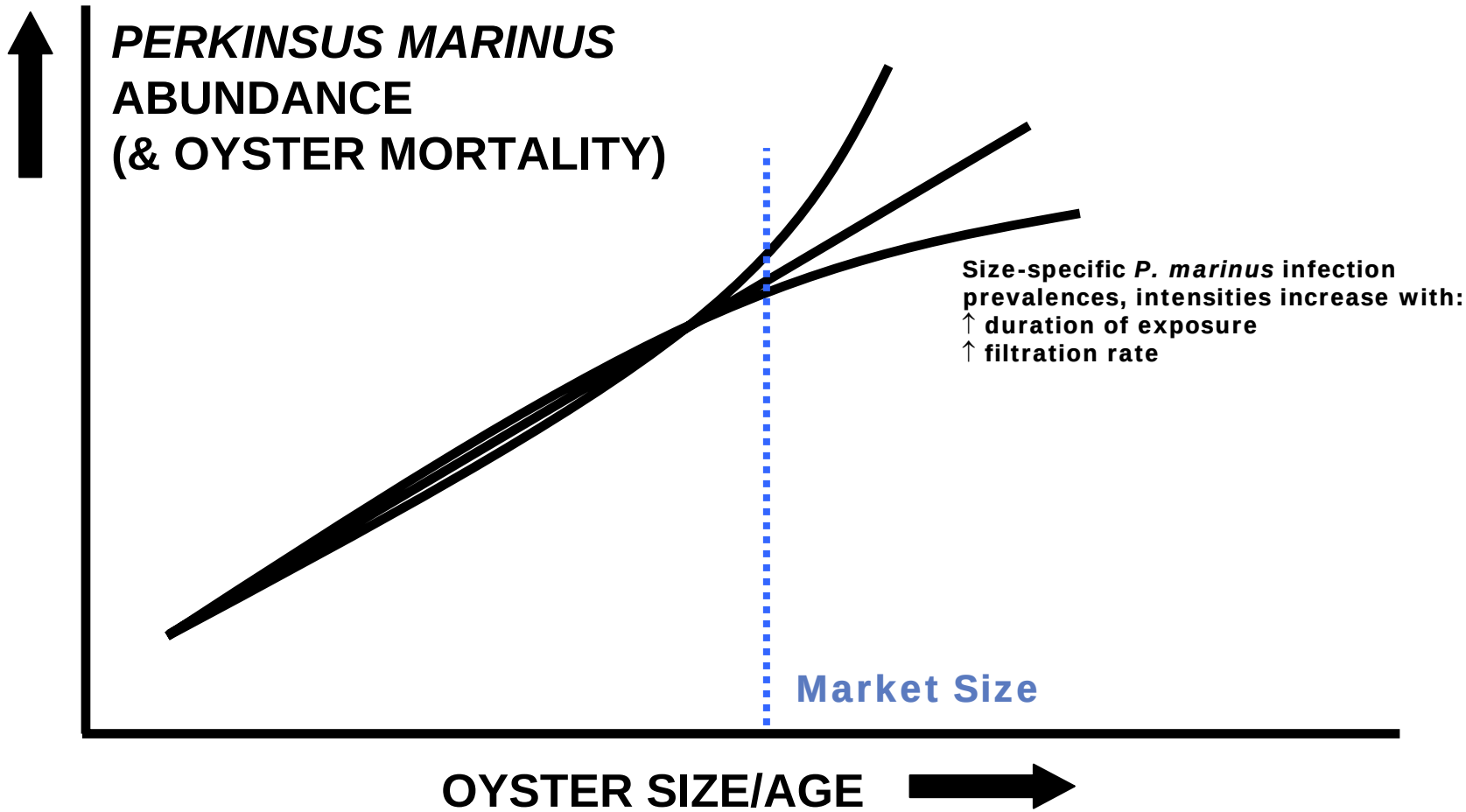




K-W analyses of intensity scores significant at $\alpha = 0.05$ in July, Aug, Oct. Multiple comparisons using Nemenyi Test

One-way ANOVAs of arcsine-transformed mortality data significant every month. Multiple comparisons using Tukey's Test

2) Does *P. marinus* Parasitism Increase with Oyster Size / Age, So Infected Oysters Make Decreasing Reproductive Contributions Over Time?



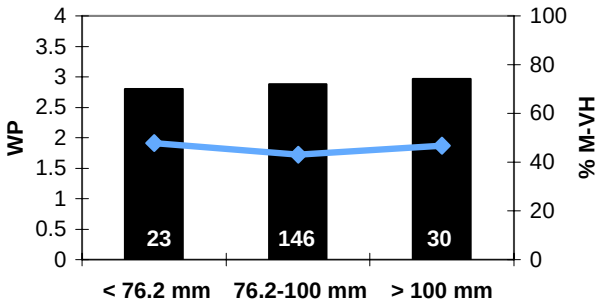
VIMS Oyster Disease Monitoring, 2002-2006

- 30+ sites routinely monitored
- 72 samples with *P. marinus* weighted prevalence > 2.00 since 2002
- 1827 *P. marinus* diagnoses

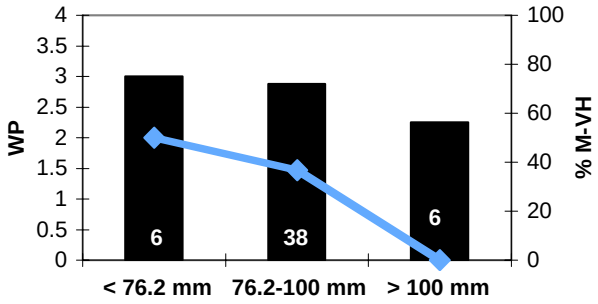
Size Category	n	WP	% N-LM	% M-VH	%MH-VH	%H-VH
< 76.2 mm	287	2.53	63.1%	36.9%	15.0%	6.3%
76.2-99.9 mm	1110	2.62	63.2%	36.8%	18.0%	7.7%
≥ 100 mm	430	2.57	65.1%	34.9%	17.2%	6.5%

P. marinus at Specific VIMS Monitoring Sites

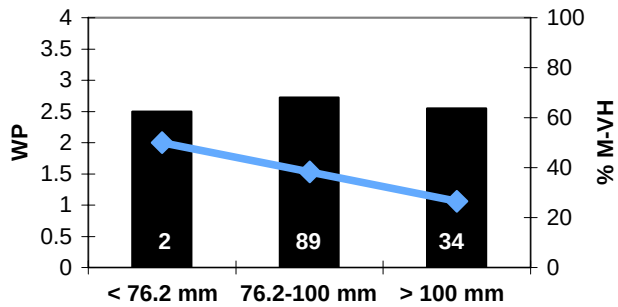
Rappahannock 2002



York 2005



James 2006



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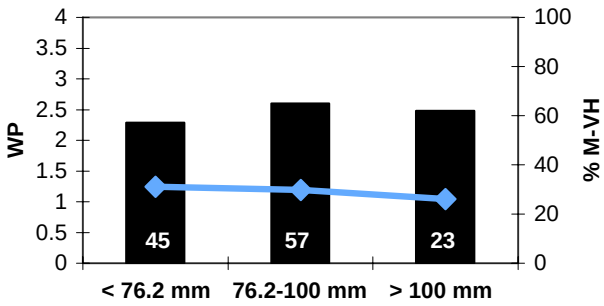

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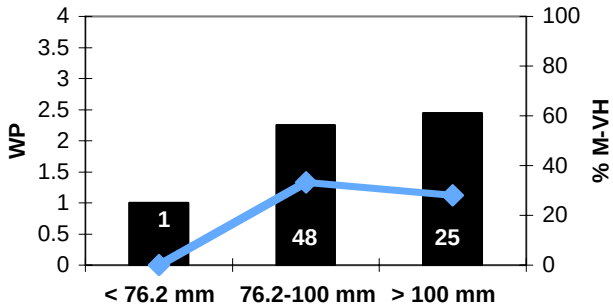
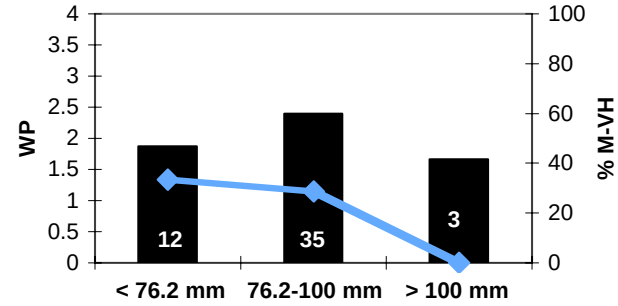
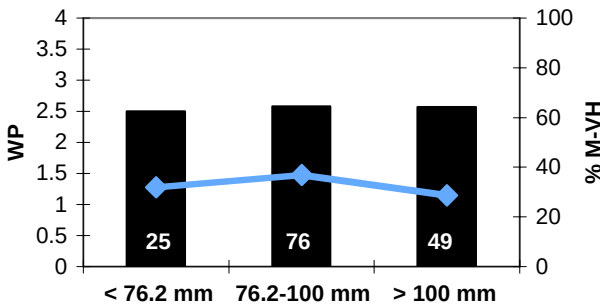


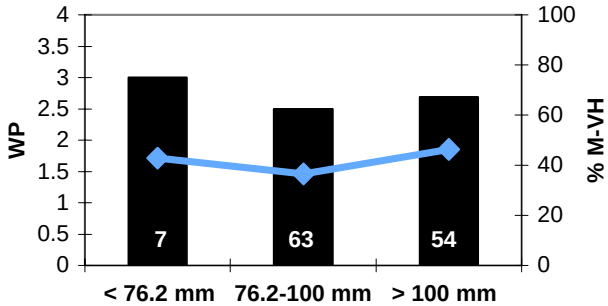
Great Wicomico 2006



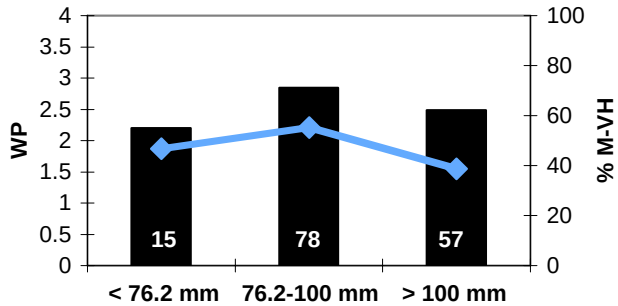
Lynnhaven 2005



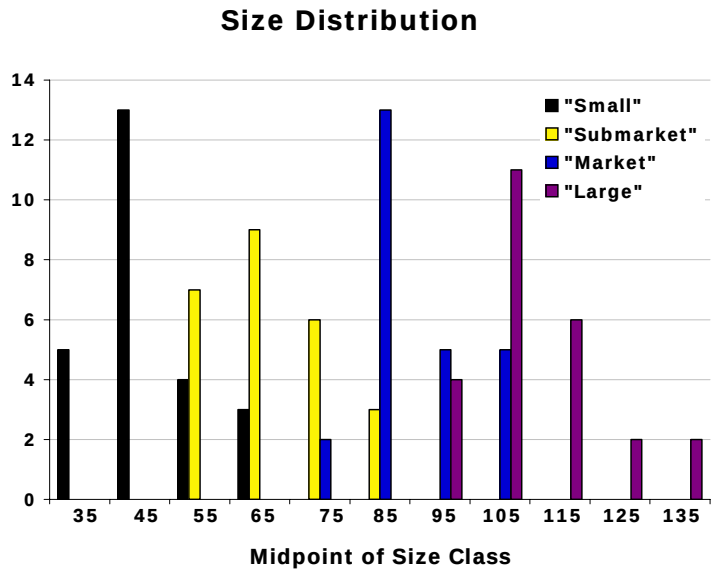
Rappahannock 2006

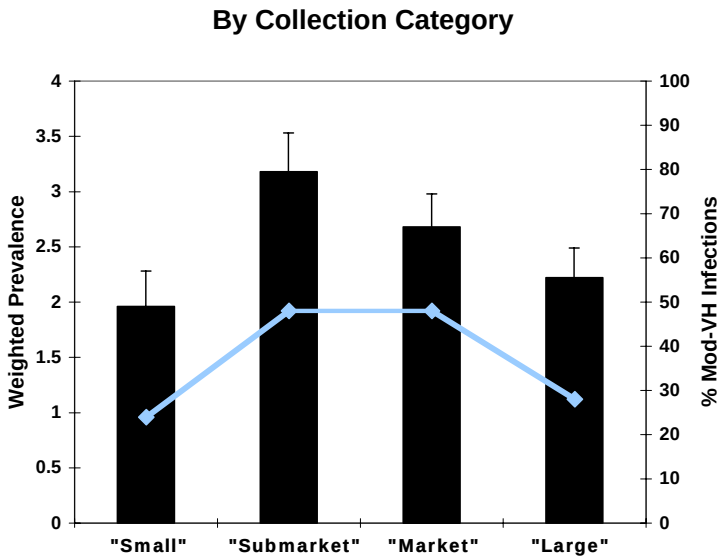


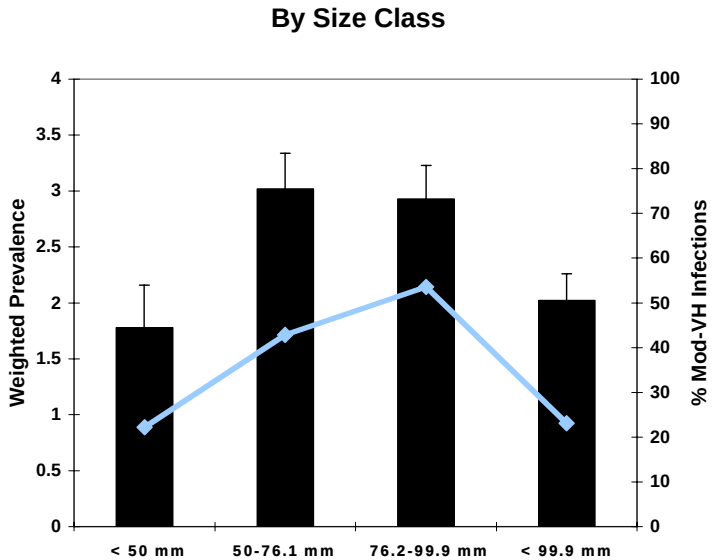
Lynnhaven 2006 (ex. PHC)



Pleasure House Creek, October 2006



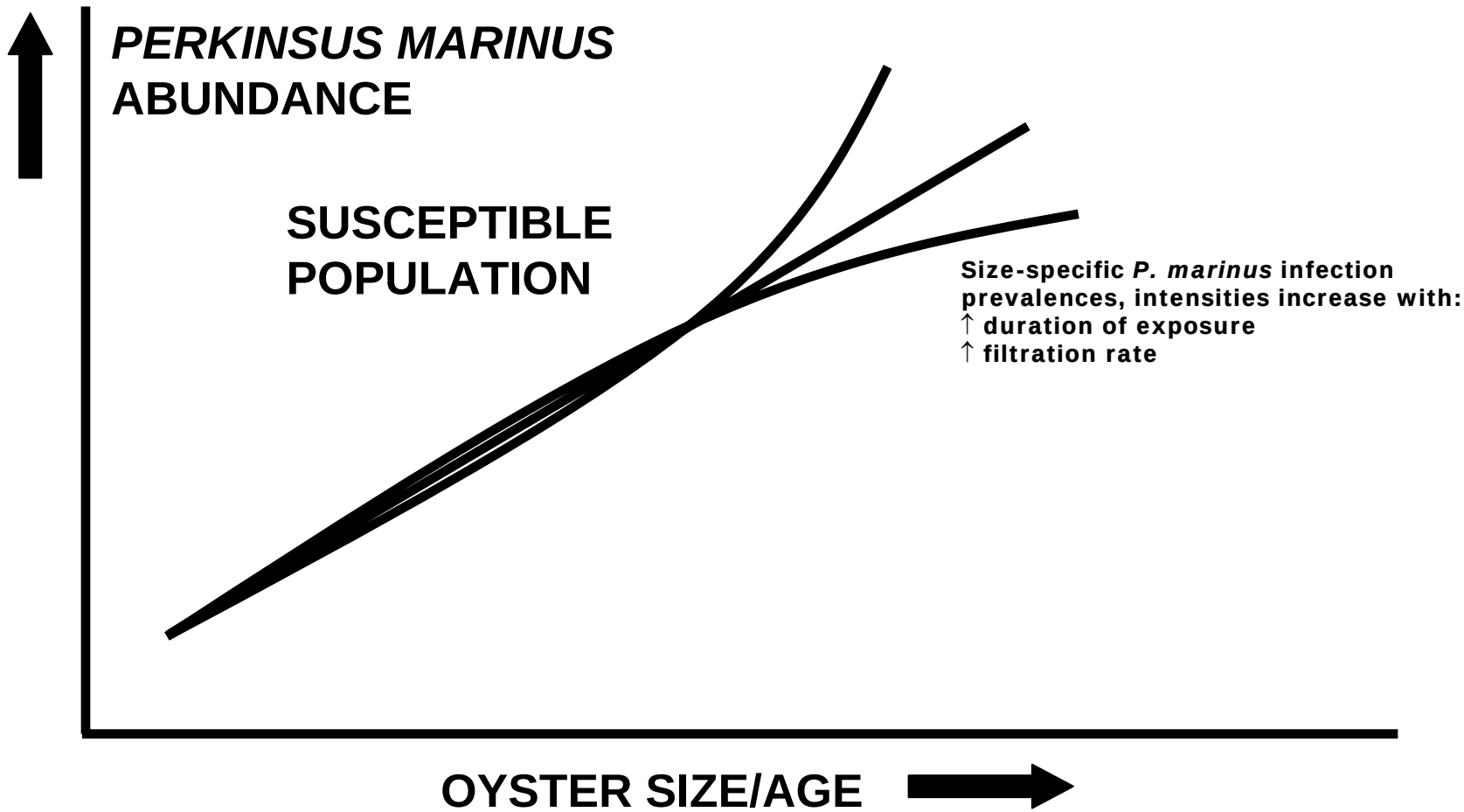




Susceptible vs. Resistant (?) Oyster Populations

- **Also observed by Calvo et al. (1996) in MD:**

"A plot of size versus infection stage revealed that stages 4 and above (i.e., > moderate) were less frequent in small (20-50 mm) oysters and very large (100-200 mm) oysters than in medium (50-100 mm) oysters."



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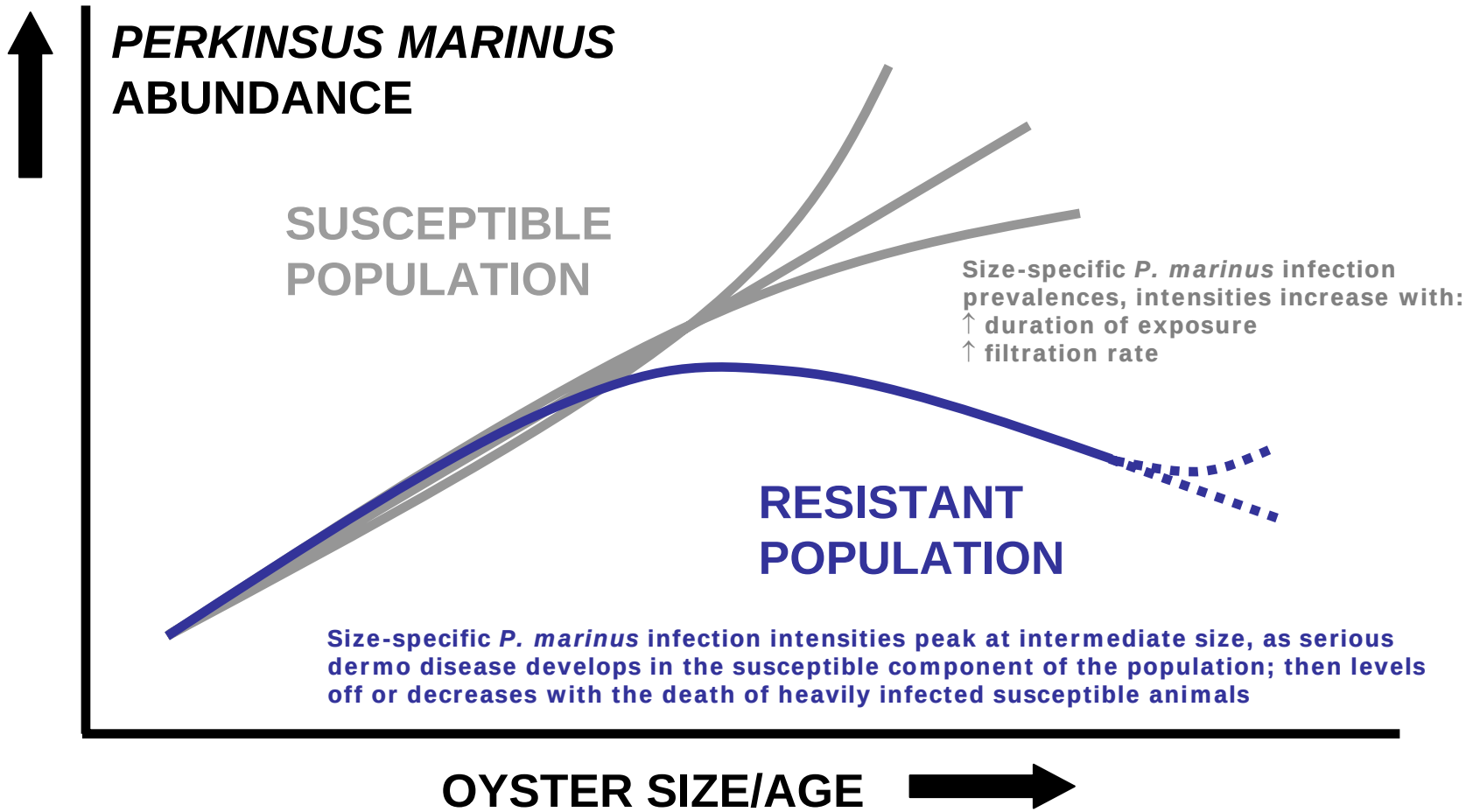

Discussion and Implications

- **Implications:**

  1) If this size-specific *P. marinus* parasitism trend does reflect the purging of dermo-susceptible individuals from the population, with a resulting enrichment for resistant oysters in larger size classes -- and the resistance of these oysters is heritable -- then a case can be made for the preservation of these larger, presumably fecund, resistant individuals

  → **A disproportionate reproductive contribution by these large, fecund, resistant oysters -- balancing or offsetting the contribution by susceptible but pre-dermo 1) local individuals and 2) recruits from low salinity sanctuaries -- may be the key to the evolution of resistance or tolerance in natural populations**

Discussion and Implications

2) Most oysters in VA waters have peak-season infections below moderate intensity

- → **Statistically significant impacts on oyster condition, growth are generally apparent only at *P. marinus* intensities above moderate intensity (Kennedy et al. 1995; Dittman et al. 2001; Ford and Smolowitz in press)**
- → **Large proportions of even dermo-exposed oysters in all size classes should remain healthy enough to reproduce (which histology does support)**

- → ***These data challenge the assumption that only small, pre-dermo oysters are locally reproductive within dermo-enzootic VA waters***

Acknowledgments

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