

## SESSION D – GENETICS TOWARD LONGEVITY AND PRODUCTIVITY

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Co-Authors: Yuehuan Zhang

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Co-Authors: Hannah K. Jaris, Marta Gomez-Chiarri, David Steven Brown

**Dr. Hong Yu** – MENDELIAN INHERITANCE OF GOLDEN SHELL COLOR IN THE PACIFIC OYSTER *CRASSOSTREA GIGAS*  
Co-Authors: Qi Li, Hong Yu, Jianlong Ge, Lingfeng Kong

## PROFILING DIFFERENTIAL GENE EXPRESSION BETWEEN GRANULOCYTES AND HYALINOCYTES OF THE PACIFIC OYSTER *Crassostrea gigas*

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Circulating Hemocytes possess strong phagocytosing and killing abilities against invading microorganisms. Therefore, these cells are primarily responsible for host defense in the Pacific oyster, *Crassostrea gigas*. Two subpopulations of *C. gigas* hemocytes are classified on the basis of forms and functions: granulocytes and hyalinocytes. Little is known, however, about differences of transcriptome between granulocytes and hyalinocytes. In this study, we performed to facilitate a transcriptome analysis of the hemocytes in *C. gigas* using high-coverage expression profiling (HiCEP) method, to assess the genes that are involved in responses to environmental stresses and infection. To separate subpopulations, we carried out discontinuous density gradient centrifugation with Percoll. As a result, we could obtain 90-95% pure fractions of both granulocytes and hyalinocytes. Total RNAs were prepared from these hemocytes, and then gene expression was analyzed using the HiCEP method. Among the 5,344 genes that exhibited differential expression profiles between the two subpopulations, 2,504 were upregulated by granulocytes. We could sequenced and annotated 16 transcript-derived fragments (TDFs) among the higher-expressed genes in the granulocytes. Using quantitative PCR, we also confirmed that the TDFs were actually expressed at higher levels on granulocytes, and, to a lesser degree, in hyalinocytes. Two of the sequences of the TDFs showed resemblance to known genes, metallothionein IV and WIPF1, but most showed no similarity to any genes in the databases of other animals.

# EFFECTS OF INTRASPECIFIC DIVERSITY ON GROWTH, SURVIVORSHIP, AND RECRUITMENT OF THE EASTERN OYSTER ACROSS ENVIRONMENTS

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Intraspecific diversity, particularly of foundation species, can significantly affect population, community, and ecosystem processes. Examining how genetic diversity relates to demographic traits provides a key mechanistic link between genotypic and phenotypic variation for taxa with complex life histories. We conducted a field experiment to assess how two metrics of intraspecific diversity (“cohort” diversity: the number of independent juvenile cohorts created from different source populations; and genetic relatedness: genetic similarity among individuals) affect the growth, recruitment, and survivorship of the eastern oyster, *Crassostrea virginica*. At two sites differing in resource availability and predation intensity, we manipulated juvenile oyster diversity and predator exposure (presence/absence of a cage) to compare the effects of cohort diversity and genetic relatedness on oyster demographics under different environmental conditions. We found that differences in predation pressure between sites largely determined post-settlement survivorship of oysters. However, in the absence of predation (i.e., presence of a cage), one or both metrics of intraspecific diversity strongly influenced growth, recruitment, and survivorship, indicating that different diversity metrics have distinct yet interdependent effects on vital rates. Our results demonstrate that intraspecific diversity can affect multiple demographic traits of this important foundation species, yet the magnitude and direction of these effects depend on the diversity metric, as well as the environmental context. Given the global loss of oyster reef habitat and rapid decline in oyster population size, our results are particularly relevant to management and restoration.

## **EVIDENCE FOR AND AGAINST LOCAL ADAPTATION IN JUVENILE OYSTER (*CRASSOSTREA VIRGINICA*) TRANSPLANTS ACROSS MULTIPLE SITES IN THE SOUTHEASTERN U.S.**

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Evidence for ecological effects of intra-specific diversity stems primarily from studies of variation within populations, limiting our understanding of the role of between-population variation and evolutionary processes. We conducted a biogeographic reciprocal transplant experiment at three sites across the southeastern U.S. to test the potential for local adaptation among oyster (*Crassostrea virginica*) populations. We produced juvenile oysters from six source sites within the region and reared them under identical conditions in a single hatchery until they were transplanted. Distance from the source site to the experimental transplant site was a significant predictor of survival, growth, and disease prevalence, but the direction and strength of this relationship differed across responses and experimental sites. Oyster survival increased as the distance between source and experimental site increased (i.e., maladaptation). In contrast, oyster growth was highest for source sites closest to two of the experimental sites (i.e., adaptation), with no relationship at the third. At the one site where the disease MSX was prevalent, oysters from nearby source sites had highest prevalence, indicating maladaptation. These results highlight the potential importance of oyster intra-specific diversity for both aquaculture and restoration efforts.

## INTERSPECIFIC BACKCROSSES BETWEEN THE HYBRIDS OF *Crassostrea hongkongensis* ♀ × *C. gigas* ♂ AND THE TWO PARENTAL SPECIES

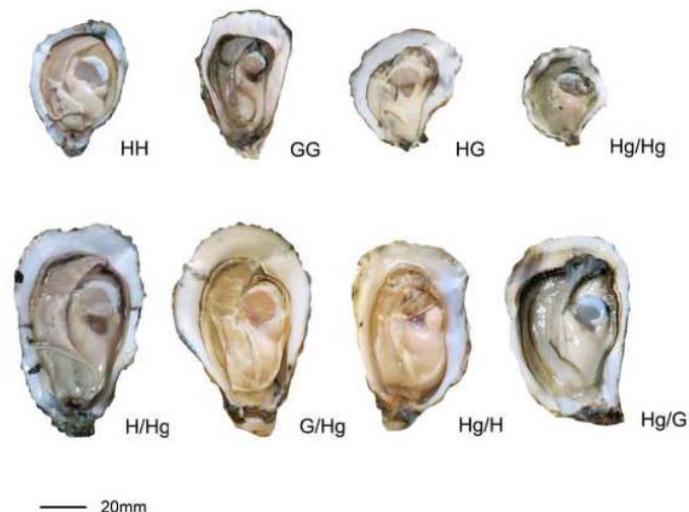
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To explore the possibility of performance improvement in the hybrids of *Crassostrea hongkongensis* ♀ × *C. gigas* ♂, the backcross progeny and F2 hybrids were produced and evaluated. Three replicates were successfully generated, each one consisted of single crosses (HH, GG and HG), two female backcrosses (between female hybrids and males of the two parents species: Hg/H and Hg/G), two male backcrosses (between male hybrids and females of the two parents species: H/Hg and G/Hg), and F2 hybrids (Hg/Hg).

Phenotypic traits of these experimental groups were evaluated for fertilization, survival, growth and gonad development. All backcross groups exhibited high fertilization levels; however, the female backcrosses (Hg/H and Hg/G) and F2 hybrids showed a low hatching rate. Survival and growth superiority of the backcross progeny were observed from larval stage to adult, although outbreeding depression of the F2 hybrids occurred. Interestingly, all backcross progeny and F2 hybrids were fertile with the production of functional gametes and normal sex ratios when fully matured.

Our results revealed that artificial interspecific backcrosses can be successful. Furthermore, backcross progeny are viable, fertile, and fast-growing, and therefore hold promise for potential utilization in aquaculture as a new oyster stock resource.



## **ASSESSING THE CONTRIBUTION OF AQUACULTURE AND RESTORATION TO WILD OYSTER POPULATIONS IN A RHODE ISLAND COASTAL POND**

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The decline of wild eastern oyster populations has resulted in expanded restoration and aquaculture efforts. Recent field surveys suggest that wild populations inhabiting Rhode Island coastal ponds are growing, yet the factors contributing to this increase in population size are unknown. A population genetic analysis, based on genotypes from 13 microsatellite loci, was conducted to determine if the growth of wild populations is a consequence of self-seeding or recruitment from nearby aquaculture and restoration sites. Samples were collected from eight populations within Ninigret Pond, a large coastal lagoon highly influenced by human activity, and a ninth control population outside the pond where oyster culture is absent. Genetic parameters were measured and compared among two restored, three farmed, and four wild populations located varying distances from restoration and aquaculture activities. As expected, genetic diversity (allelic richness) was greater within wild populations (11.7) than farmed (6.9) or restored (7.5) populations. Moderate to high levels of genetic differentiation were also detected among most cultured populations ( $F_{ST} = 0.038 - 0.230$ ) likely reflecting differences in source broodstock and hatchery practices. A lesser degree of genetic differentiation was detected among wild/cultured comparisons ( $F_{ST} = 0.035-0.148$ ) and wild populations were not significantly different from one another. A STRUCTURE analysis assigned sampled individuals to one of two groups which corresponded to wild and cultured populations; however, each wild population contained some proportion of individuals that were genetically more similar to the cultured group. The evidence of admixture among wild and cultured groups has important implications for the management of wild, farmed, and restored oyster populations.

# MENDELIAN INHERITANCE OF GOLDEN SHELL COLOR IN THE PACIFIC OYSTER *CRASSOSTREA GIGAS*

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## ABSTRACT

Shell coloration in many molluscs is highly variable. A shell color segregation study with progenies from a full factorial cross generated among Pacific oysters exhibiting distinct shell colors (golden, white and black; Fig. 1) was conducted to investigate the inheritance of the golden shell color and its correlation with dark pigmentation. Random samples from twenty-three full-sib families were obtained and the shell coloration of offspring within each family was recorded. Results revealed that golden coloration was inherited in a different pattern from dark pigmentation, indicating its different genetic basis. Dark pigmentation was identified as a foreground color while golden or white color were background ones. The locus controlling background colors has two alleles with the allele for golden background being dominant to the allele for white background. In addition, the overlying foreground pigmentation of shells with a golden background was significantly lighter than that of shells with a white background, which suggested an epistatic effect of background color on shell foreground pigmentation. All these findings will facilitate the selection of elite oyster lines with desired shell coloration for aquaculture.

**Keywords:** Pacific oyster; *Crassostrea gigas*; Shell color; Inheritance; epistatic

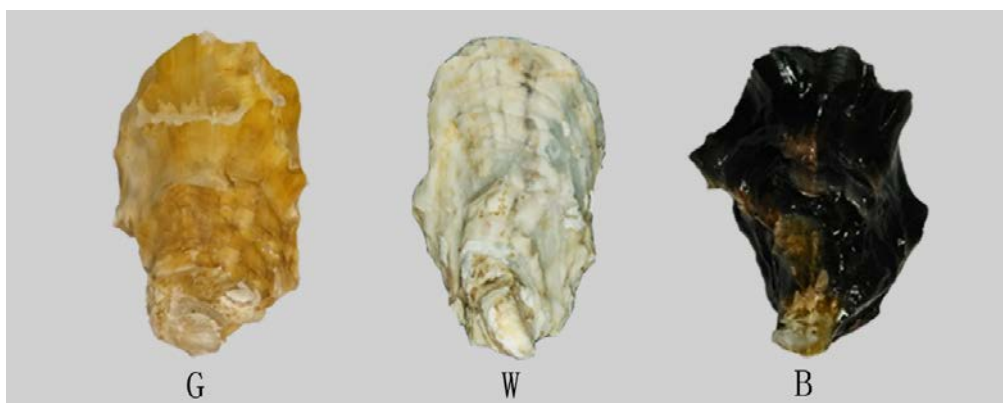


Fig. 1. Representative parents of three shell coloration patterns. G = Golden; W= White; B = Black.