Conclusions

Strains established in this project are highly recommended for developing similar projects. As those combinations are unusual, students will not be able to easily find expected results online, which contributes to the development of their own observation, data collection, and analysis, and awakens their curiosity, which may increase their interest in the challenging scientific activities.

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Using DGRP sequenced genomes to map heterozygous modifier effects on cell death in Bar eye of Drosophila.

Thompson, James N., jr., Daniel Tinney, Jacob Khoussine, Gary Cox, J. Ross Ogden, Grayson Audette, Saba Bingabr, Danielle Branesky, Andres Gomez, Jason Lauderdale, Cassandra Long, Jacob Mitchell, Sonya Narula, Dennis Nguyen, Thuc-Vi Nguyen, Daniel Pons, Cameron Steele, Garrett Sutton, Tyler Tallman, and Barbara Safiejko-Mroczka.

Department of Biology, University of Oklahoma, Norman, OK.
major mutation exposes a variable range of expression due to secondary modifier loci in the genetic background. These modifier loci must be directly or indirectly relevant to the targeted pathway of the major mutation.

But one cannot introgress a targeted mutation into a sequenced strain without destroying the sequenced background by recombination and segregation. Some experimental designs may benefit from that approach. But drawing upon the exceptional resource offered by the DGRP lines, there is a middle road. In this pilot study, we test a limited set of the more than 200 currently available DGRP sequenced lines to assess their heterozygous modifier effects on mutant expression of Bar, a dominant cell death eye mutation (small duplication) with variable expression. Specifically, we are screening for genomic regions that influence cell death by measuring the number of eye facets in Bar eyes on different heterozygous sequenced genome backgrounds. If successful, the next phase will be to explore whether the same quantitative modifier loci are involved in phenotypic expression of other cell death-related traits such as mutations that cause notches in Drosophila wings.

This pilot experiment used five of the DGRP lines, obtained from the Bloomington Drosophila Stock Center (# 25174, 25175, 25177, 25179, and 25180). Virgin females were collected from the Basc strain, which carries Bar, white-apricot, and scute mutations along with inversions that make it an effective X-chromosome balancer. Basc females were mated to males from a DGRP strain yielding F1 males that carried Bar and were heterozygous for one of the sequenced genomes. Heads were removed and bisected so the eyes pointed up when mounted on an electron microscope plug. Samples were prepared using the protocol in Thompson et al. (2009) and were viewed and photographed with a Zeiss Neon 40EsB electron microscope. The number of successful mounts varied somewhat from one group to another. Future experiments will attempt to standardize the data sample size.

Perhaps surprisingly (or luckily), even just these five representative sequenced genomes demonstrated significantly different influences on cell death in the Drosophila eye (Figure 1). For a sample subset of these data, photographs were scored by up to 15 students, and the replicate variation associated with repeatability among researchers was not significant. Differences were typically only one or two facets in an eye having perhaps 100. Against this “replicate repeatability”, strain effects were easily identifiable (Figure 2; for a comparison of the two extremes in this small sample, mean ± sd for strain 25175 is 105.4 ± 17.4, n = 7; for strain 25174, 141.0 ± 7.7, n = 15, facets per eye). It is clear that even this small sample of DGRP genomes carries cell death modifiers that differ in their effects on Bar as heterozygotes.

A future addition to the analysis will be a measure of fluctuating symmetry (FA) when sufficient data are available from both eyes of an individual. FA is a standardized difference between the left and right sides of a trait that is expected to be symmetrical and for which deviations can be interpreted in terms of developmental stress. This will yield an insight into developmental homeostasis influencing cell death expression.

![Figure 1. Average facet counts for five representative DGRP strains heterozygous for Bar (standard deviations are too small to show). 1, strain 25175; 2, strain 25177; 3, strain 25180; 4, strain 25179; 5, strain 25174.](image-url)
Acknowledgments: This pilot study was done as an experiment designed by students in an advanced Biology lab course and was made possible through the advice and electron microscope supervision of Preston Larson and Gregory Strout, Samuel Roberts Noble Microscopy Laboratory at the University of Oklahoma.


Erratum


Noor and Coyne (1995) reported that hybrid males from a cross between a recently (seven-generations prior) collected strain of *D. p. bogotana* and a stock of *D. pseudoobscura* were sterile. While the conclusion was correct, re-examination of records finds that two pieces were incorrectly described. First, the sample size was five rather than twenty (or no clear record was kept of additional dissections), and second, the hybrid males were tested for fertility 1 day after eclosion rather than 7. I (Noor) apologize for my errors in presentation.