Supplemental Materials and Methods

Strains

All strains (refer to Table 1) were acquired from the *Caenorhabditis* Genetics Center (University of Minnesota), which is funded by the NIH National Center for Research Resources (NCRR). VC1121 and VC194 were generated by the *C. elegans* Reverse Genetics Core Facility at UBC. RB938 and RB911 were generated by the *C. elegans* Gene Knockout Project at OMRF. Both of these centers are part of the International *C. elegans* Gene Knockout Consortium (www.celeganskoconsortium.omrf.org).

***Maintenance of* C. elegans**

C. elegans were maintained on nematode growth media (NGM) plates using OP50 *E. coli* (available from the *Caenorhabditis* Genetics Center) as a food source according to standard methods (Brenner, 1974; Stiernagle, 2006).

Collection of* C. elegans *Genomic DNA, Nested PCR, and Agarose Gel Electrophoresis

Students were required to wear gloves and cautioned against introducing foreign DNA into their samples. Genomic DNA was collected by adding 500 µL of sterile ddH_2O to a 35 mm plate full of mixed-stage *C. elegans* worms, collecting 50 µL into a PCR tube, adding an equal volume of PK Buffer (50 mM KCl, 1.25 mM MgCl, 0.5% NP40, 0.5% Tween 20, 0.2 mg/mL proteinase K and 10 mM Tris HCl, pH 8.3) and freezing at -70^{0}C for 15 minutes. Samples were incubated at 60^{0}C for 4 hours, 95^{0}C for 15 minutes, and then frozen until ready for use in nested PCR.

To perform external nested PCR, the genomic DNA was thawed and diluted 1:10 in sterile ddH_2O. The sequences for primers used in the nested PCR reactions are included in Supplemental Material B. PCR reactions (50 µL final volume) were assembled by adding the following in the order indicated: 10 µL sterile ddH_2O, 5 µL diluted genomic DNA (or 5 µL sterile ddH_2O for a negative control), 5µL of 10 µM external forward primer, 5 µL of 10 µM external reverse primer, and 25 µL of Master Mix. Master Mix was made for the students immediately before class, and stored on ice (2X Promega Taq Buffer, 5 mM $MgCl_2$, 0.4 mM dNTPs, with 0.5 µL of Taq / 25 µL). Assembled PCR samples were mixed thoroughly by

pipetting and then run in a thermocycler with the following program: 92^0C for 3 minutes, and then a loop of the following for 30 cycles: 92^0C for 40 sec., 52^0C for 40 sec., 72^0C for 3 min. One last extension was performed at 72^0C for 10 minutes. Samples were then frozen for future use in the internal nested PCR reactions. Internal PCR reactions were set-up similar to the external reactions except that a 1:10 dilution of the external reactions was used as a template and the internal primer sets were used. Agarose gel electrophoresis was performed by running 15 μL of the internal PCR reactions on 0.8% agarose gels using standard methods (refer to the lab manual in Supplemental Material A).

Feeding RNAi

Feeding RNAi bacterial clones were obtained from the Ahringer feeding RNAi library (GeneService, Ltd, Cambridge, UK). Also note that some feeding RNAi strains are available for free through Cold Spring Harbor Lab (www.silencinggenomes.org). Feeding plates (60 mm) were made similar to the standard NGM plates used for culturing the worms (Stiernagle, 2006), except they had 25 μg/mL carbenicillin, 12.5 μg/mL tetracycline and 0.2% filter-sterilized lactose added (after autoclaving, when NGM had cooled to less than 60ºC). Note that many laboratories use IPTG (1 mM final concentration) instead of lactose because it increases the shelf life of the plates. We opted to use lactose because of its lower cost; and used the plates within two days of making them to avoid issues with storage. Feeding RNAi bacteria were grown in liquid culture, and 50 μL was plated on the feeding plates using standard methods (refer to Lab Manual in Supplemental Material A). Bacteria were allowed to grow for 24 hours in the dark at room temperature, and then were stored at 4ºC until needed. Three days before student observation, larval stage worms were added to the feeding RNAi plates by staff. To obtain larval worms, mixed-staged populations were bleach treated to obtain eggs for a synchronous hatch-off (refer to Protocol 6 in (Stiernagle, 2006) and larval worms were collected 2 days later by rinsing the plates with M9 (recipe in Protocol 6, Stiernagle, 2006). About 30 worms were added per feeding RNAi plate, allowed to feed for 24 hours, and then were removed with aspiration. Two days later students began observing the plates. We estimate that the materials cost per student for performing this activity was approximately $7 per student in 2008, for reagents to perform the RNAi and nested PCR.

Bioinformatics exercises

Students followed a descriptive worksheet to explore bioinformatics information and tools related to their assigned gene. The worksheet (in Supplemental Material E), introduced them to www.Wormbase.org, the National Center for Biotechnology Information's Online Mendelian Inheritance in Man database (www.ncbi.nlm.nih.gov/omim), Geneious DNA analysis software (more information at www.geneious.com), Simple Modular Architecture Research Tool (SMART, smart.embl-heidelberg.de), and an open-source Java viewer for chemical structures in 3D (Jmol, jmol.sourceforge.net). Geneious software is intuitively easy to use bioinformatics software that provides both standard and customized academic licenses. Tutorials and educational tools are also available. Students accessed this software during lab with their lab partners on twelve university-owned laptops. They used the free one-week trial of Geneious Pro on their home computers during the final week when they completed their posters (it is currently a free two-week trial).

References

Brenner, S. (1974). The genetics of *Caenorhabditis elegans*. Genetics *77*, 71-94.

Stiernagle, T. (2006). Maintenance of *C. elegans*. WormBook, The *C. elegans* Research Community, http://www.wormbook.org (accessed April 20, 2012).