**1. Project Title:**

Improving techniques for estimating abundance and habitat use in nearshore marine habitats using environmental DNA.

**2. Investigators:**

Ryan Kelly (University of Washington)

**3. Project Duration and Requested Funds:**

8/1/16 – 7/31/17, $102,000

**4. Summary:**

Organisms of all kinds shed cells containing diagnostic DNA into the environment, which can be recovered and assigned to a taxon based upon its match to known sequences. Because DNA degrades under most ambient environmental conditions—the half-life of DNA in fresh- and saltwater is approximately 24-48 hours (*1,2*)—this environmental DNA (eDNA) provides a snapshot of the species recently present in the sampled habitat. However, while it is widely accepted that DNA can be collected and identified from a range of environmental samples, connecting field collections of eDNA with abundance surveys remains largely unexplored.

Here, we propose to develop eDNA survey methods to quantify fish communities (with a focus on salmon, herring, and smelt species) in a nearshore estuarine habitat in Puget Sound. To compare the efficacy of eDNA and traditional methods, we will collect water samples in parallel with collections made via three traditional net sampling methods, targeting nearshore fish communities that provide a range of spatial sampling scales (from meters to 100s of meters). We will use both quantitative PCR (qPCR) and massively parallel DNA sequencing technologies to provide eDNA data. We will then apply a newly developed statistical framework to provide field estimates of the relationship between species abundance and eDNA. Our replicated sampling design—using three field methods at three spatial scales—provides an opportunity to understand the appropriate spatial scale for eDNA sampling, and the potential value and pitfalls of eDNA surveys for understanding patterns of fish abundance.

**5. Scope, Objectives, Merit:**

Early life-history is a critical period for most commercially important fishes. For anadromous salmonids, the transition from freshwater to the marine environment is a key determinant of marine survival and fisheries productivity. Similarly, ecologically important forage fish (e.g. herring and smelt) use nearshore and estuarine habitats as spawning and rearing grounds. Thus, accurate estimates of fish abundance in nearshore estuarine areas are critical to understanding early life-history survival and play an important, poorly understood role in driving stock assessment models. However, estimates of fish abundance in the nearshore habitats are difficult and expensive to obtain using traditional sampling methods; shallow water and vegetation interfere with acoustic surveys, turbid water often hinders visual surveys, and the presence of vegetation and other structures restricts the efficacy of some net survey techniques. Nonetheless, estimating salmon and other fish species’ abundance is especially important in light of (a) continued loss of foundational vegetated habitats such as seagrass beds, salt marshes, and other coastal wetlands, and (b) restoration efforts intended to mitigate such losses. Assessing the importance of nearshore habitats to salmonids and forage fish —and the success of nearshore restoration efforts in particular—requires efficient methods for quantifying abundance in these habitats. **We propose to apply recently developed environmental DNA (eDNA) survey techniques to assess the fish communities across three habitats used at different points during the salmon migration from freshwater to the ocean.** We will characterize the abundance of salmonids and other commercially valuable and ecologically important fish in three distinct estuarine habitats; these three habitats offer the additional benefit of comparing results across three different spatial sampling techniques. If successful, this project would adapt a rapidly-developing, innovative technology that could inform stock assessments nationwide.

This project has three main objectives: **(1)** Validate and improve eDNA methods for rapidly detecting the occurrence and abundance of ecologically important coastal fish; **(2)** Assess the efficacy of eDNA methods by comparing estimates of occurrence and abundance from traditional net sampling and eDNA methods across three existing sampling methodologies; **(3)** Compare costs and relative benefits of eDNA methodologies relative to traditional sampling methods. The project will improve abundance estimates for species that are ecologically and commercially important but difficult to survey, including juvenile salmon (*Oncorhynchus* spp.) and forage fish (e.g. herring (*Clupea pallasi*) and smelt (family *Osmeridae*)). Herring and smelt have been estimated as the most numerically abundant fish but exhibit recent declines, while Chinook salmon populations in this river system are ESA-listed, but suffer from poor quantification (*3,4*).

We will focus on developing rigorous eDNA methods in a single estuary and will compare results from eDNA samples with three widely used net sampling techniques (Fyke net, beach seine, surface trawl). Each of these sampling methods occurs in distinct habitats and samples different cumulative areas (increasing from the scale of ~1 to 100s of meters); as such, we expect the match between eDNA and net sample to vary with each sampling method. Concordance should be highest for Fyke nets and lowest for surface trawls. However, the rate at which the concordance between eDNA and sampling scale changes is itself a useful metric because it has direct implications for the appropriate scale at which to apply eDNA methods in the field. Beyond individual sample-to-sample comparisons, we will calculate aggregative measures of fish density from traditional and eDNA methods. Such sample aggregation is a critical step in the development of abundance indices that feed directly into most stock assessments. Thus our research plan provides information on two pressing questions for the future use of eDNA in stock assessments: (1) at what spatial scale can eDNA accurately reflect local abundance? and (2) can eDNA provide integrated metrics of abundance on scales useful for management?

**6. Technical Approach:**

Our approach has three components: 1) Field collection of eDNA samples conducted in parallel with existing nearshore sampling using three sampling techniques. 2) Laboratory processing of eDNA samples to produce quantitative estimates of DNA abundance and 3) application of novel statistical approaches to generate defensible estimates of biomass from eDNA and comparison of estimates from eDNA to those derived from traditional sampling methods. We discuss each in turn.

***Field collections*.** This project will align with the Skagit River Intensively Monitored Watershed Project, which tracks status and trends of species in the Skagit River estuary and Bay (Fig. 2), including all species of Pacific salmon and several forage fish species (*12*). The focal species is Chinook salmon, which is listed as threatened under the Endangered Species Act. Small (<50 mm) Chinook leave freshwater and rear in the estuary before migrating to the Pacific Ocean (*3*). Substantial variation in migration timing and fish movement complicates traditional estimations of abundance; nevertheless these data represent the longest time series of juvenile abundance Puget Sound salmon and are vital for determining stock status, trends, and responses to habitat restoration efforts.

The Skagit Intensively Monitored Watershed Project counts Chinook salmon at four life stages. We will focus on three stages: estuary residence in channels and impoundments, nearshore intertidal residence along beaches and in lagoons, and subtidal residence in Skagit Bay (Fig. 2, Table 1). Sampling in each habitat requires different sampling gear and complicates abundance estimates across habitats. In estuary channels and nearshore lagoons, capture efficiency can be very high (>50%), while efficiency in other habitats is much lower (< 10%). These various sampling procedures highlight the potential utility of eDNA for estimating local abundance, calibrating eDNA to each procedure individually.

Beyond salmonids, Pacific herring, surf smelt, stickleback, and Pacific sandlance reside in Skagit Bay. The multi-species eDNA techniques we propose will capture these and other species inhabiting estuarine and nearshore habitats at various times of the year.

We will collect three 1L water (eDNA) samples in each of three seasons (February-April, May-July, and August-October). The first two seasons represent prime sampling windows when Chinook salmon and forage fish are abundant in the estuary and bay; fall sampling will serve as negative controls. Water samples will be taken in triplicate at each location. We will record environmental covariates to assess their effects on concordance between eDNA and net sampling.

***Environmental DNA methods.*** There are two distinct approaches for eDNA analysis. In the first, the amount DNA from a single target taxon is quantified using quantitative PCR (qPCR; *8*), by comparing the amplification rate of a field sample to one of a standard of known concentration. This protocol quantitatively assesses changes in single target-species’ DNA concentrations in the field (*13,14*).

In the second technique a single locus is PCR-amplified from all genomes present in a sample, the resulting products (amplicons) are sequenced, and the resulting sequences are matched to those of known species in a large database (*15*). Amplicon sequencing can provide data for a hundreds of taxa in the sampled community, but only provides information about relative abundance of DNA in the sample.

We propose to combine these two methods, using 1) qPCR to quantify the abundance of key species, and 2) amplicon sequencing to provide the relative abundance for dozens of species in the community, and then 3) linking qPCR and sequencing results to provide the first quantitative survey of an entire community (*16*).

For our focal species (salmonids and forage fish), there is sufficient published and unpublished genetic information to reliably identify taxa to the species level with both qPCR and sequencing approaches. *14* provides qPCR primers for Chinook salmon and colleagues at the US Geological Survey have developed qPCR primers specific for eight salmonid species (J. Duda pers. comm.). We will choose the three species most frequently observed in the previous year’s net surveys for qPCR, and quantify eDNA using replicate qPCRs from each of the triplicate water samples.

We will then use three sets of primers to generate mixed-species amplicons for sequencing. We will use primers with the following target genes and taxa: 16S mtRNA (16S; targeting fish and a diverse range of invertebrate taxa in Puget Sound; *16*), 12S mtDNA (targeting vertebrates; *17*), and we will use the software ecoPrimers (*18*) to develop a novel primer set that will amplify a region of mitochondrial cytochrome c oxidase I (CO1), which varies consistently among the three target taxa chosen for qPCR.

***Linking eDNA and net surveys.*** MatchingeDNA field sampling and existing net sampling protocols will reveal the relationship between fish abundance and eDNA. While there are challenges for translating observations of eDNA into biomass, these are largely analogous to those faced by traditional sampling methods (Fig. 3; *16*); we view eDNA sampling as a new “net” with a unique set of traits that can produce statistical biases in the estimation of abundance.

An unresolved issue is how DNA is shed and disperses in the environment (*14*). Our multi-scale field sampling approach allows us to explore the relationship between eDNA and fish abundance across spatial scales. We expect the strongest eDNA-net correlations to be in estuarine channels, and weakest correlations to be in offshore trawls, consistent with the spatial scales of sampling in those habitats.

**Literature Cited:**

1. Thomsen, P. F. *et al.* Monitoring endangered freshwater biodiversity using environmental DNA. 2012. *Molecular Ecology* 21, 2565–2573.
2. Turner, C. R. *et al.* 2014. Particle size distribution and optimal capture of aqueous macrobial eDNA. *Methods in Ecology and Evolution* 5:676–68.
3. Beamer, E., A. et al. 2005. Delta and nearshore restoration for the recovery of wild Skagit River Chinook salmon: Linking estuary restoration to wild Chinook salmon populations. Supplement to Skagit Chinook Recovery Plan, Skagit River System Cooperative, LaConner, WA. Available at: [www.skagitcoop.org](http://www.skagitcoop.org).
4. Zimmerman, M. S., C. Kinsel, E. Beamer, E. Conner, and D. Pflug. 2015. Abundance, survival, and life history strategies of juvenile migrant Chinook in the Skagit River, Washington. *Transactions of the American Fisheries Society* 144:627–641.
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6. deVargas, C., et al. 2015. Eukaryotic plankton diversity in the sunlit ocean. *Science* *348:* 1261605.
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15. Leray, M., N. Knowlton. 2015. DNA barcoding and metabarcoding of standardized samples reveal patterns of marine benthic diversity. Proc. Natl. Acad. Sci.  112:2076–2081.
16. Shelton, A.O., J.L. O’Donnell, J.F. Samhouri, N. Lowell, G. Williams, R.P. Kelly. *In Revision.* A statistical framework for inferring biological communities from environmental DNA*. Ecological Applications*.
17. Port, J. A. *et al. In Review.* Assessing the vertebrate community of a kelp forest ecosystem using environmental DNA. Molecular Ecology.
18. Riaz, T. *et al.* 2011. ecoPrimers: inference of new DNA barcode markers from whole genome sequence analysis. *Nucleic Acids Research* 39:e145–e145.

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| **Budget** | Amount |
| Salary | $54,108 |
| Benefits, 25.4% | $13,743 |
| Supplies | $667 |
| Contractual Services | $10,000 |
| Total Direct Costs | $78,518 |
| Indirect Costs, 26% | $20,415 |
| Total | $98,933 |
| JISOA - Task 1 fee, 3.1% | $3,067 |
| **Total Project Costs** | **$102,000** |

**16. Budget Justification:**

**Salary**

$54,108 is requested to support a postdoctoral researcher for 12 months who will be conducting field collections, laboratory procedures, and data analyses.

**Fringe benefits**

Fringe Benefits are charged at the standard UW rate of 25.4% (projected benefit rate as of 7/1/16).

**Other Direct Costs:**

**Supply**

Funds in the amount of $667 are requested for field and laboratory work.

**Contractual Services**

Funds in the amount of $10,000 are requested for non-personnel expense being the contract for next-generation sequencing services. While this expense might appear large the power of the technique lies in the tens of millions of sequences that will be generated and the actual cost per sample (and per sequence) is quite low. Funds will defray the costs Illumina MiSeq sequencing runs at the Northwest Genomics Center (run out of the Nickerson Lab at the University of Washington). 3 runs \* $3400/run.

**Indirect Costs**:

Facilities and Administrative costs are calculated at the standard UW rate of 26% of MTDC for off campus project.

Task I Fee: Per NOAA's guidelines on cooperative institutes, a 3.1% fee for Task I costs is included in all projects funded under JISAO. These funds will be used for education, outreach and administration at JISAO.

**Curriculum Vitae for Ryan P. Kelly:**

**Education:**

**J.D.** 2011, University of California, Berkeley. School of Law (Boalt Hall).

**Ph.D.** 2006, Columbia University, New York. Ecology, Evolution, and Environmental Biology.

**Certificate of Environmental Policy** 2005, Columbia University, New York. Ecology, Evolution, and Environmental Biology.

**M.Phil.** 2005, Columbia University, New York. Ecology, Evolution, and Environmental Biology.

**M.A.** 2003, Columbia University, New York. Ecology, Evolution, and Environmental Biology.

**B.Sc.** 2000, University of California, Los Angeles. Ecology and Evolution. *cum laude*.

**Employment History**

March 2013 – Present: Assistant Professor, University of Washington, School of Marine and Environmental Affairs

September 2011 – March 2013: Fellow, Center for Ocean Solutions, Stanford University.

June 2010 – May 2011: Affiliated researcher, Center for Ocean Solutions, Stanford University.

August 2009 – May 2011: Graduate Student Instructor, UC Berkeley.

May – August 2009: Sierra Club Environmental Law Program, Summer Associate.

September 2006 – August 2008: Postdoctoral researcher, Stanford University.

August 2002 – May 2006: Graduate student and teaching assistant, Columbia University.

August 2003 – May 2005: Science enrichment teacher, 7th and 8th Grades, East Side Middle School and East Side Community High School, Manhattan.

Spring 2002: Staff Assistant for United States Senator Barbara Boxer, Hart Senate Office Building, Washington, D.C.

September 2000 – June 2001: Graduate Teaching Fellow, UCLA.

**Research Interests**

My interests span the divide between hard scientific data and policymakers’ use of those data. My more applied research joins genetic and ecological research with real-world implementation in law and policy, particularly with respect to environmental monitoring, resource management, endangered species, and ocean acidification.

**Research Experience**

March 2013 – Present: Principal Investigator, University of Washington. Research focuses on environmental monitoring using environmental DNA, ocean acidification policy, environmental management using ecosystem thresholds, and related topics.

August 2008 – March 2013: Independent and collaborative work at Stanford University and U.C. Berkeley on ocean policy, marine genetics, and the intersection of scientific data and law.

September 2006 – August 2008: Postdoctoral work at Stanford University’s Hopkins Marine Station in the laboratory of Stephen R. Palumbi. This work focused on the comparative population genetic patterns of Pacific nearshore invertebrates as these relate to a variety of ecological species traits. In addition, my co-authors and I devised and tested a new statistic for population genetic analysis.

2002 – May 2006: Doctoral research at Columbia University in the laboratory of Rob Desalle at the American Museum of Natural History in New York City, investigating the molecular phylogeny and population-level genetic structure of chitons (Polyplacophora) and other marine invertebrates from the Pacific coast of North America.

1998 – 2001: Molecular research in the laboratory of David K. Jacobs at UCLA into the ecology and phylogeography of the genus *Transennella*, a group of small bivalves prevalent along the Pacific coast of North America.

**Teaching Experience**

2013 - Present: Assistant Professor, University of Washington

*Classes Designed and Taught*: Environmental Studies 250, Data Types and Methods (Fall 2013, 2014, 2015); SMEA 515 Ocean and Coastal Law (Winter 2014, Spring 2015, Spring 2016); SMEA 550B Marine Biodiversity Science, Law, and Policy (Spring 2014); SMEA/OCEAN 591 Marine Science in the Coastal Zone (Winter 2015, 2016).

2012: Instructor, Stanford University

*Class Designed and Taught*: Earth Systems 174/274, Marine Biodiversity Science, Law, and Policy (Fall 2012)

2009 - 2011: Graduate Student Instructor, UC Berkeley.

*Classes Taught*: Biology 1B Field Research Section (Fall 2009 and 2010); Earth and Planetary Sciences C51, Big History (Spring 2010 and 2011)

Fall 2005: Teaching Fellow, Columbia University.

*Class Taught*: Life Science and Evolution for undergraduate non-majors

Fall 2003 – Spring 2006: Science enrichment teacher, 7th and 8th grades, Manhattan.

*Description*: Taught weekly science lessons in two public junior high schools as part of the National Science Foundation’s GK-12 teaching fellowship program, on topics ranging from human physiology to systematics.

Fall 2000 – Spring 2001: Teaching Assistant, University of California, Los Angeles.

*Classes Taught*: Evolution; Plant Physiology; Physiology of Marine Phytoplankton

**Awards, Grants, and Fellowships**

2015 – 2016: University of Washington Royalty Research Fund, Making eDNA Quantitative

2012 – 2014: Packard Foundation: Environmental DNA as Next Generation Monitoring Tool

2010 – 2011: UC Berkeley Law School Martin S. & Estelle D. Depper Scholarship

2010: UC Berkeley Law School Alexander Marsden "Captain" Kidd Scholarship

2009: UC Berkeley Department of Integrated Biology Outstanding Graduate Student Instructor

2008 – 2010: UC Berkeley Law School Scholarship

2003 – 2006: National Science Foundation GK-12 Teaching Fellowship, New York City

2005: American Museum of Natural History Lerner-Gray Grant for Marine Research

2005: University of California at Davis Genetics Research Conservation Program Grant

2002 – 2003, 2005-2006: Faculty Fellowship, Columbia University, New York City

2000 – 2001: Teaching Fellowship, UCLA; Distinguished Teaching Assistant Award

**Publications**

**Kelly, R.P.,** J.L. O’Donnell, N.C. Lowell, A.O. Shelton, J.F. Samhouri, S.M. Hennessey, B.E. Feist, and G.D. Williams. 2016. Urbanization, Ecological Diversity, and Two Views of the Cathedral. (*submitted*).

**Kelly, R.P.** 2016. Making Environmental DNA Count. Molecular Ecology Resources 16: 10-12.

Battista, W., **R.P. Kelly**, A. Erickson, and R. Fujita. 2016. A Comprehensive Method for Assessing Marine Resource Governance: Case study in Kāne‘ohe Bay, Hawai‘i. (*in press*, Coastal Management)*.*

Lowell, N. and **R.P. Kelly.** 2016. Evaluating Agency Use of “Best Available Science” Under the United States Endangered Species Act Biological Conservation 196:53-59.

Port, J.A., J.L. O’Donnell, O.C. Romero-Maraccini, P.R. Leary, S.Y. Litvin, K.J. Nickols, and **R.P. Kelly**. 2016. Assessing the Vertebrate Community of a Kelp Forest Ecosystem using Environmental DNA. Molecular Ecology 25(2):527-541.

O'Donnell, J.L., **R.P. Kelly**, N. Lowell, and J.A. Port. 2016. Indexed PCR Primers Induce Template-Specific Bias in Large-Scale DNA Sequencing Studies. PLoS One 11(3): e0148698.

Shelton, A.O., J. L. O'Donnell, J.F. Samhouri, N. Lowell, G.D. Williams, and **R.P. Kelly**. 2016. A Framework for Inferring Biological Communities from Environmental DNA (*in press,* Ecological Applications).

Gattuso, J.-P., A. Magnan, R. Billé, W.W.L. Cheung, E.L. Howes, F. Joos, D. Allemand, L. Bopp, S. Cooley, C.M. Eakin, O. Hoegh-Guldberg, **R.P. Kelly**, H.-O. Pörtner, A.D. Rogers, J.M. Baxter, D. Laffoley, D. Osborn, A. Rankovic, J. Rochette, U.R. Sumaila, S. Treyer, C. Turley. 2015. Contrasting Futures for Ocean and Society from Different CO2 Emissions Scenarios. Science 349 (6243): aac4722.

Marshall, K., A. Stier, J. Samhouri, **R.P. Kelly**, E. Ward. 2015. Conservation Challenges of Predator Recovery. Conservation Letters 9(1): 70-78.

Albright, R., D. Alongi, K. Anthony, M. Baird, M. Byrne, C. Collier, S. Dove, K. Fabricius, T. Fyffe, K. Gale, C. Hanratty, O. Hoegh-Guldberg, **R.P. Kelly**, J. Lough, M. Mongin, J. Monks, P. Munday, J.K. Oliver, R. Pears, M. Rodgers, B. Russell, B. Tilbrook, E. Abal. 2015. Ocean Acidification: Linking Science to Adaptive Management Solutions (*revisions* *submitted* *to* Global Change Biology).

Selkoe, K.A., T. Blenckner, M.R. Caldwell, L. Crowder, A. Erickson, T. Essington, J. Estes, R. Fujita, B.S. Halpern, M. Hunsicker, C.V. Kappel, **R.P. Kelly**, J.N. Kittinger, P.S. Levin, J. Lynham, M. Mach, R. Martone, L. Mease, A. Salomon, J. Samhouri, C. Scarborough, A. Stier, C. White, J. Zedler. 2015. Principles for Managing Marine Ecosystems Prone to Tipping Points. Ecosystem Health and Sustainability 1(5):17.

**Kelly, R.P.**, A.L. Erickson, and L.A. Mease. 2015. How Not to Fall Off a Cliff, or, Using Tipping Points to Improve Environmental Management. 41 Ecology Law Quarterly 843.

**Kelly, R.P.** 2015. Will More, Better, Cheaper, and Faster Monitoring Improve Environmental Management? 44 Environmental Law 1111-1147.

**Kelly, R.P.**, J.A. Port, K.M. Yamahara, R.G. Martone, N. Lowell, P.F. Thomsen, M.E. Mach, E. Prahler, M.R. Caldwell, and L.B. Crowder. 2014. Harnessing DNA to Improve Environmental Management, Science 344 (6191): 1455-1456. doi:10.1126/science.1251156.

**Kelly, R.P.**, J.A. Port, K.M. Yamahara, and L. Crowder. 2014. Using Environmental DNA to Census Marine Fishes in a Large Mesocosm. PLoS ONE 9(1): e86175. doi:10.1371/journal.pone.0086175.

A.L. Strong, K.J. Kroeker, L.T. Teneva, L.A. Mease, and **R.P. Kelly**. 2014. Ocean Acidification 2.0: Managing Our Changing Coastal Ocean Chemistry. BioScience: doi: 10.1093/biosci/biu072.

**Kelly, R.P.**, A. Erickson, L. Mease, W. Battista, J. Kittinger, R. Fujita. 2014. Embracing Thresholds for Better Environmental Management. Philosophical Transactions of the Royal Society B 370: 20130276.

**Kelly, R.P.**, S.R. Cooley, T. Klinger. 2013. Narratives Can Motivate Environmental Action: The Whiskey Creek Ocean Acidification Story. Ambio 43(5): 592-599.

**Kelly, R.P.**, and M. Caldwell. 2013. “Not Supported by Current Science” : The National Forest Management Act and the Lessons of Environmental Monitoring for the Future of Public Resources Management. 32 Stanford Environmental Law Journal 151.

**Kelly, R.P.**, and M. Caldwell. 2013. Ten Ways States Can Fight Ocean Acidification (and Why They Should). 37 Harvard Environmental Law Review 57.

Billé, R., **R.P. Kelly**, A. Biastoch, E. Harrould-Kolieb, D. Herr, F. Joos, K. Kroeker, D. Laffoley, A. Oschlies, and J.-P. Gattuso. 2013. Taking Action Against Ocean Acidification: A Review of Management and Policy Options. Environmental Management 52(4): 761-779.

Caldwell, M. R., Hartge, E. H., Ewing, L. C., Griggs, G., **Kelly, R. P.**, Moser, S. C., Newkirk, S. G., Smyth, R. A., & Woodson, C. B. 2013. Chapter 9: Coastal Issues. In: Garfin, G., Jardine, A., Merideth, R., Black, M., & LeRoy, S. (Eds.), Assessment of Climate Change in the Southwest United States: a Report Prepared for the National Climate Assessment. A report by the Southwest Climate Alliance. Washington, DC: Island Press.

**Kelly, R.P.**, and M.R. Caldwell. 2012. The Limits of Water Quality Criteria. The Environmental Forum 29(6): 34-38.

**Kelly, R.P.**, and J. Grote Stoutenburg. 2012. Washington State's Legal and Policy Options for Combating Ocean Acidification in State Waters. Center for Ocean Solutions. (Available at: http://www.ecy.wa.gov/water/marine/oa/2012report\_app8.pdf).

**Kelly, R.P.**, and M. Caldwell. 2012. Why Ocean Acidification Matters to California, and What California Can Do About It. Center for Ocean Solutions. (Available at: http://www.centerforoceansolutions.org/publications/all).

**Kelly, R.P.**, M.M. Foley, W. Fisher, R. Feely, B.S. Halpern, G.G. Waldbusser, and M.R. Caldwell. 2011. Mitigating Local Causes of Ocean Acidification with Existing Laws. Science 332: 1036-1037.

**Kelly, R.P.** 2011. Spineless Wonders: How Listing Marine Invertebrates and their Larvae Challenges the US Endangered Species Act. 19 Penn State Environmental Law Review 1-53.

D.J. Barshis, E.E. Sokta, **R.P. Kelly**, A. Sivasundar, B.A. Menge, J. Barth, and S.R. Palumbi. 2011. Coastal Upwelling May Drive Sweepstakes Recruitment in the Acorn Barnacle *Balanus glandula* Marine Ecology Progress Series 439: 139-150.

**Kelly, R.P.** 2010. The Use of Population Genetics in Endangered Species Act Listing Decisions. 37 Ecology Law Quarterly 1107-1159.

**Kelly, R.P.**, and S.R. Palumbi. 2010. Genetic Structure Among 50 Species of the Northeastern Pacific Rocky Intertidal Community. PLoS ONE 5(1): e8594. doi:10.1371/journal.pone.0008594

**Kelly, R.P.**, T.A. Oliver, A. Sivasundar, and S.R. Palumbi. 2010. A Method for Detecting Population Genetic Structure in Diverse, High Gene-Flow Species. Journal of Heredity 101(4): 423-436.

**Kelly, R.P.**, and S.R. Palumbi. 2009. General-use polymerase chain reaction primers for amplification and direct sequencing of enolase, a single-copy nuclear gene, from different animal phyla. Molecular Ecology Resources 9: 144-147.

**Kelly, R.P.**, and D.J. Eernisse. 2008. Reconstructing a Radiation: The Chiton Genus *Mopalia* in the North Pacific. Invertebrate Systematics 22: 17-28.

**Kelly, R.P.**, and D.J. Eernisse. 2007. Southern Hospitality: A Latitudinal Gradient in Gene Flow in the Marine Environment. Evolution 61(3): 700-707.

**Kelly, R.P.**, I.N. Sarkar, D.J. Eernisse, and R. Desalle. 2007. DNA Barcoding Using Chitons (genus *Mopalia*). Molecular Ecology Notes 7: 177-183.

**Kelly, R.P.** 2006. Genetics and Geography in Pacific Coast Chitons (Mollusca; Polyplacophora). PhD Dissertation, Department of Ecology, Evolution, and Environmental Biology. Columbia University, New York City. 276pp.

**Professional Affiliations**

California Bar Association (number 278958; admitted 2011)

Articles Editor, Ecology Law Quarterly (2010 - 2011)

Society for the Study of Evolution

Western Society of Naturalists

**Graduate Students and Postdocs Supervised**

Natalie Lowell (SMEA Master’s Student, 2013 – present)

Mikaela Freeman (SMEA Master’s Student, 2013 – present)

Colleen Crotty (SMEA Master’s Student, 2013 – present)

Erin Ryan-Penuela (SMEA Master’s Student, 2014 – present)

Sebastien Clos-Versailles (SMEA Master’s Student, 2014 – present)

Gretchen Thuesen (SMEA Master’s Student, 2014 – present)

Max Mossler (SMEA Master’s Student, 2015 – present)

James L. O’Donnell (Postdoc, 2014 – present)

**Master’s Committees**

Raz Barnea (SMEA, graduated 2014)

Sarah Towne (SMEA, class of 2015)

Alex Tanz (SMEA, class of 2016)

Annie Hillier (SMEA, class of 2016)

**PhD Committees**

Audrey Ragsac, UW Biology (Richard Olmstead, PI) 2014 - present

Eleni Petrou, UW SAFS (Lorenz Hauser, PI) 2014 - present

Mike Tillotson, UW SAFS (Tom Quinn, PI) 2014 – present

William King, UW Biology (Ken Sebens, PI) 2015 – present