

Stream RFP #07 – Supplemental Water Quality Sampling for Combined Reston Mussel Stocking x Stream Monitoring Program

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Executive Summary: We will quantify biogeochemical effects of reintroducing native unionid mussels into two restored urban headwater streams in Reston, Virginia - Snakeden Branch and The Glade. We have scientists with expertise in mussel establishment and population dynamics, biogeochemistry and ecology as experts for each of the key components of the stated needs of Resource Protection Group, Inc. Our study will provide a year and a half of bimonthly sampling at baseflow (June 2024-December 2025) during the ongoing mussel reintroduction as a way to provide a more detailed analysis of how mussel beds in the restoration streams are altering nutrient processing, water quality, and biological communities. We are already measuring and analyzing individual mussel biogeochemistry, and here we proposed to measure water and sediment nutrients in the downstream reaches with the established mussel beds, and at the upstream control stream reach in both streams to scale the mussel nitrogen and phosphorus nutrient cycling effect. Our proposed budget is **\$392,529**

Project performance: May 1, 2024 to October 31, 2025

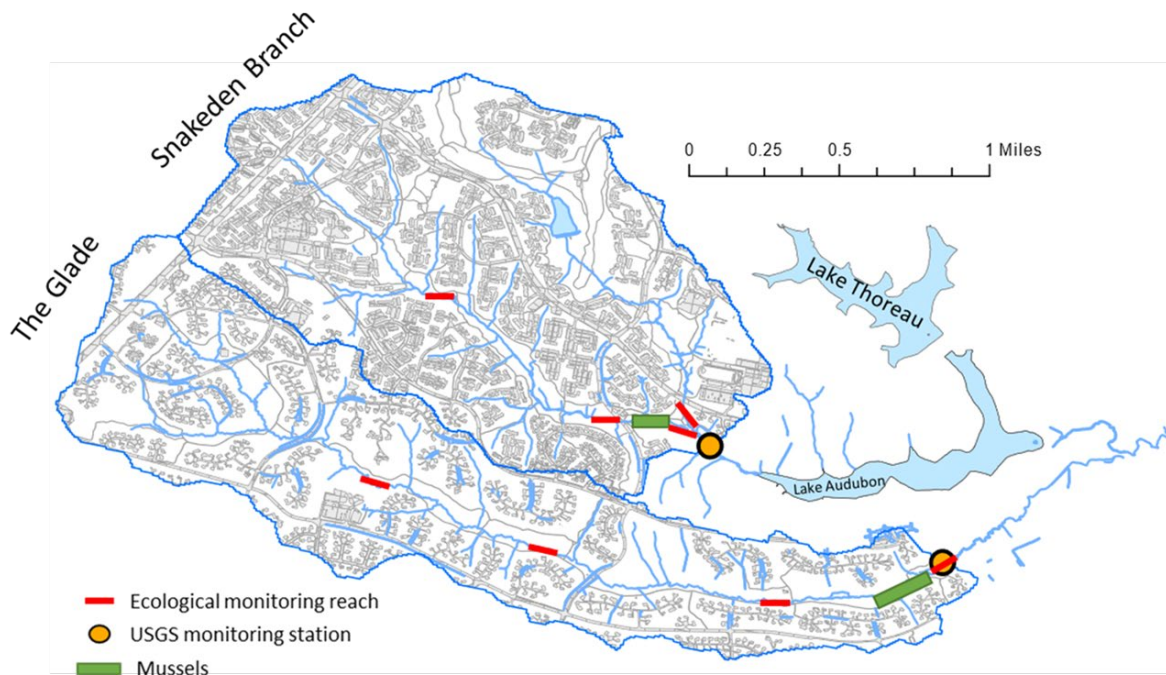
Project Team:

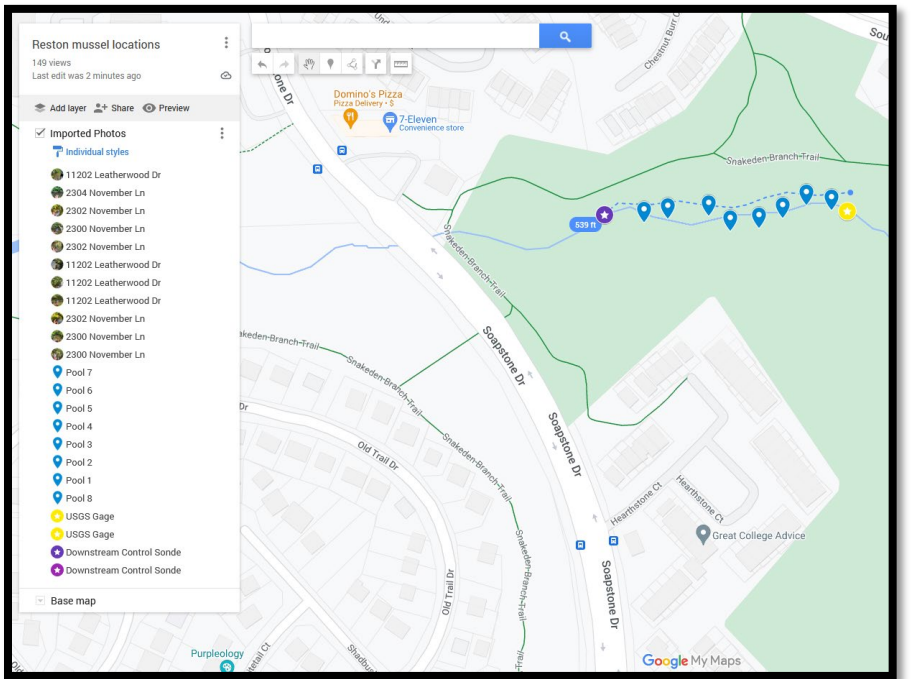
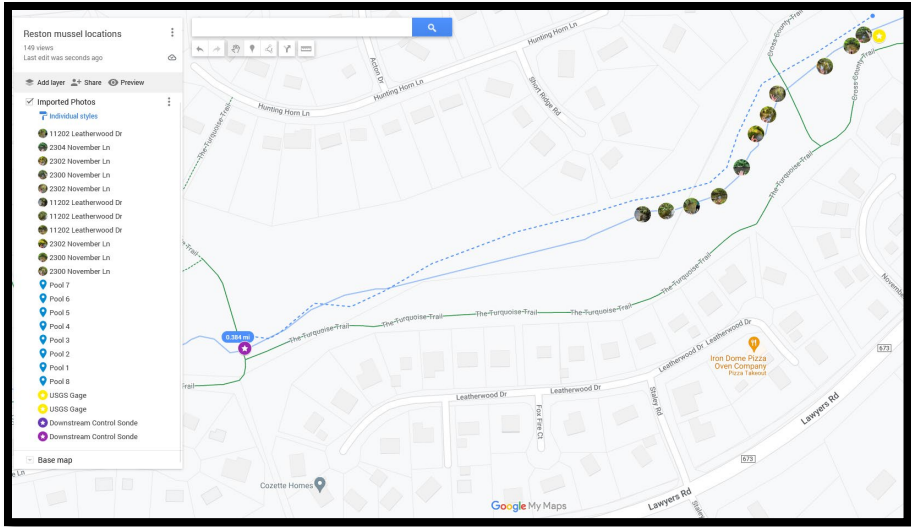
Sally Entrekin, PhD, is an aquatic entomologist and ecosystem ecologist in the department of Entomology at Virginia Tech. She will be the lead PI responsible for management and coordination of all aspects of the project. **Ryan Stewart, PhD, has expertise in soil hydrology and water resource management.** He also manages the VT-SPES Central Soil & Water Quality Lab (CSWQL) that is currently working on the QUAPP associate with this project (appended here) and will be overseeing the water processing proposed here. **Jess Jones, PhD, is a restoration biologist with the US Fish and Wildlife Service stationed at Virginia Tech.** He will be responsible for collecting and/or cultivating mussels, introducing them into the two streams in baskets and free-planted and his crew will monitor the survivorship, movement, and fitness of the mussels throughout the project duration. **Chester Zarnoch, PhD, is a benthic ecologist who studies the structure and functioning of bivalves in estuarine and freshwater habitats, Baruch College CUNY, New York.** He will be responsible for measuring mussel feeding rates in-situ following mussel re-introductions in baskets and in the benthos. He will work closely with others to collate data on mussel filtration rates of suspended particulates as well as the elemental composition (carbon, nitrogen, and phosphorus) of assimilated particulates and biodeposits released to the streambed. These data will be combined with the real time data to upscale dissolved and particulate nutrient dynamics to inform how many mussels are needed to mitigate or augment nutrient retention and flux. **Denise Bruesewitz is a biogeochemist and ecosystem ecologist in Environmental Studies at Colby College, Maine.** She will measure the role of mussels in baskets and benthos on nitrogen removal as a long-term storage via assimilation vs permanent removal via facilitating denitrification in mussel beds with higher organic matter, and nitrate. Dr. Bruesewitz will work in collaboration with Zarnoch to use

sediment core incubations for denitrification. She will work with Zarnoch and Entekin on whole-stream nutrient uptake, metabolism and denitrification. **Paul Angermeier, PhD, is a fishery biologist and Assistant Unit Leader with the USGS Cooperative and a Professor in the Fish and Wildlife Research Unit at Virginia Tech.** He will collaborate with the USGS fish-sampling crew and Jones to develop a fisheries management plan that informs mussel translocations based on existing fish assemblages and outlines management actions to ensure long-term maintenance of mussel populations. **Brendan Foster is a hydrologist at the USGS VA/WV Water Science Center.** They collect water samples for nutrients and sediment using discrete and continuous monitoring approaches. Our proposed work depends on collaboration with USGS who is monitoring water downstream of the two restored streams, collecting benthic macroinvertebrates, and monitoring fish and physical habitat. USGS will expand their monitoring to accommodate additional carbon sampling at the stream monitoring station and longitudinal water-quality surveys throughout the two streams and closer to proposed mussel beds (see map).

Project Description (see map below):

[Link to interactive google map with pool locations](#)





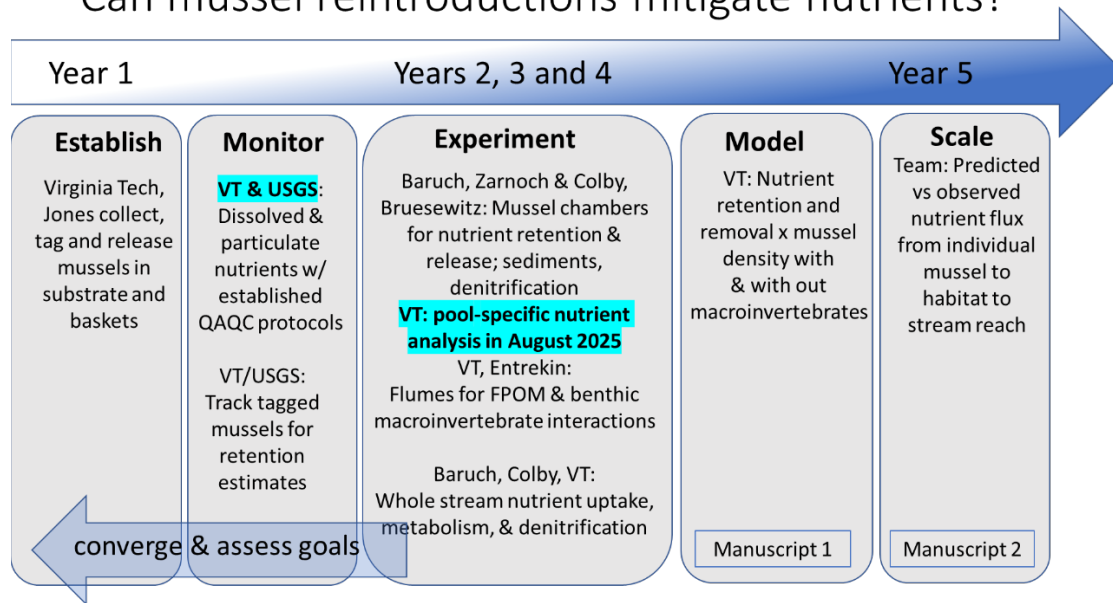
The top map (on page 2) shows the watershed that includes Snakeden and Glad Run. The USGS monitoring gages are orange and the green shows the study reach. The next two figures above are the stream reaches where the control sondes (on the downstream side of the upstream control) and pools with mussels are marked. Yellow symbols are the USGS gage stations. We will take water samples at the upstream control sonde (purple) first and then at the most upstream pool in the mussel-added reach and at the most downstream outflow of the last pool.

Objectives:

Q1. Are changes in nutrient retention, release, and removal measurable at the bed, reach, and stream scales? As per the RPG RFP, we will develop and implement a supplementary water quality field sampling and laboratory analysis program in the Snakeden and Glade Run to gather temporally paired samples above and below the mussel stocking reaches for the following parameters: Water Total-N, NH₃-N, and NO₂+NO₃-N, Water Total-P and Ortho-P, Water Total Suspended Solids (TSS). For these samples, we will provide RPG with data sets within 30 days of completed analyses for sampling events following proposer’s internal QA/QC protocol clearance. As called for by the proposal RFP, we will also continue to measure instream field pH, temperature, dissolved oxygen and specific conductance. We propose *additional* sampling for Water Total and Dissolved Organic C (TOC & DOC), Water (suspended) Chlorophyll-a, Benthic Chlorophyll-a, Sediment NH₃-N, and Sediment P seasonally for 3 of the mussel pools per mussel-added reaches to understand more fully how mussels are influencing the stream biogeochemistry. Finally, we also propose one *additional*, spatially intensive water quality sampling event in all mussel-present pools, each sampled at the upstream and downstream in August 2025 when all PIs are on-site for the *in-situ* mussel chamber experiments that will measure nutrient exchange and feeding rates.

Timeline below is from our original proposal. The highlighted light blue indicates the monitoring associated with this proposal.

Can mussel reintroductions mitigate nutrients?



Preliminary Studies (if applicable) that are a result of our current work on this project: A total of 2,816 adult mussels of the Eastern Elliptio (*Elliptio complanata*) were collected in the spring and early summer of 2023 by hand via snorkeling in the mainstem of the Potomac River and several of its tributaries. Mussels were added to Snakeden and Glade Run in July 2023 and as of December 21st, 98% of the mussels have survived and are mostly in the pools that are shown above (see map above). In September 2023, we completed mussel chamber experiments in-situ

to measure their contribution to nutrient cycling and metabolism. We also measured their filtering rates, biodeposition and excretion using side-channel experimental chambers that sourced stream water. In addition, the USGS has continued to maintain gaging stations for basic water quality parameters in addition to adding fine spatial scale measurements of nitrogen and conductivity in fall and winter. Although we have no nutrient data from the chamber experiments to present here, we will be presenting these data and the Society for Freshwater Science meeting in June 2024. Our research group is on-target (and slightly ahead) of our proposed timeline shown below. Our next steps will be spring benthic sampling for macroinvertebrates and fish, servicing of the deployed sondes for water quality and preparation for the summer field season. More mussels will be added this summer, in addition to more in-situ mussel sampling to quantify their contribution to nutrient cycling and biodeposition. For the in-situ measures of water and sediment, we are using methods that are described in the listed manuscripts (1-5) that were also referenced in our original proposal. In addition, we are following x water collection protocols. The proposed high frequency sampling will allow us to identify if mussels are having a measurable effect on stream nutrients *in-situ*.

Experimental Procedures/Methodologies:

Bi-monthly water quality sampling at the upstream control reach and downstream mussel reach in each of the two streams.

We currently have 2 EXO Hydrolab Sondes deployed and taking hourly measurements for field pH, temperature, dissolved oxygen and specific conductance at upstream control reaches and the USGS has gaging stations at the bottom of the mussel reaches taking these same measurements. All sampling will occur at least 10 days apart and 48 hours following any >0.5 inch/24-hour rain event. We will take water samples twice each month at seasonal baseflow for 18 months and analyze: Water Total-N, NH₃-N, and NO₂+NO₃-Nm, Water Total-P and Ortho-P, Water Total Suspended Solids (TSS), Water Total and Dissolved Organic C (TOC & DOC). At each sampling event, we will take grab samples using the protocol followed by our USGS partners. The method they follow is in the [USGS National Field Manual Chapter A4 section 4.1.3.B](#), which is technically called Dip sampling. This sampling method involves depth and width integration where we will “immerse a hand-held, narrow-mouth bottle at the centroid of flow or at multiple locations along a cross section.” All samples will be taken at each upstream control and downstream location where mussels have been established to detect the mussel signature (see Table 1 for sample numbers). Sample bottles will be rinsed three times with stream water prior to sample collection. Dissolved nutrients and TSS will be filtered with a 0.45 um glass fiber filter in the field using a syringe filter. Salt slugs or a dye tracer will be used in each stream to figure out solute travel times so that dip samples are taken to try to pair upstream/downstream water samples. Both streams will be sampled on the same day. We designed the upstream control and downstream sampling protocol to maximize the likelihood that the sample water is most likely influenced by the mussel beds.

Additional water quality variables (not in the RFP) that will be measured to understand how mussels influencing algae, sediment carbon and dissolved organic carbon.

We also propose to sample **suspended and benthic chlorophyll a** and nutrients and organic carbon from sediment at **3 pool locations** in each reach 4 times across a single year as a supplement to understanding how mussels are affecting nutrients and basal resources in the streams. Composite sediment samples will be collected from the top 5 cm of sediment in each pool. Samples will be stored in a cooler for transport back to the laboratory. Chlorophyll and TOC/DOC samples will be stored in amber bottles and transported back to the lab on ice.

Table 1. Below shows the water quality sample breakdown taken in the two restored streams for 18 months.

| Stream | Reach | Sample #/event | Total Samples |
|-----------------------|-----------|----------------|---------------|
| Snakeden Run | Control | 1 | 36 |
| | Treatment | 1 | 36 |
| Glade Run | Control | 1 | 36 |
| | Treatment | 1 | 36 |
| Total sample # | | | 144 |

One-time, spatially intensive water quality sampling in pools with mussels (not in the RFP).

The same water quality variables listed above will be sampled in the upstream and downstream of all of the pools that have mussels. This pool-specific sampling will occur in August 2025 at the same time the in-situ mussel respiration, denitrification and feeding experiments are being conducted (Table 2).

Table 2. Below breaks down the sample number in each habitat for the one-time, spatially intensive upstream/downstream pool sample collection.

| Stream | Reach | Pools | Sample #/pool | Total Sample # /reach |
|-----------------------|-----------|-----------|---------------|-----------------------|
| Snakeden Run | Control | 1 | 2 | 2 |
| | Treatment | 5 | 2 | 10 |
| Glade Run | Control | 1 | 2 | 2 |
| | Treatment | 12 | 2 | 24 |
| Total sample # | | 19 | 8 | 38 |

Overall project coordination: Field sampling will be conducted by a combination of one of our two lab technicians, a graduate student funded on this project, an undergraduate student supported on this project and PI Entekin. Lab sub-sampling, analyses and the associated QAQC will be conducted by the VT-SPES Central Soil & Water Quality Lab (SWQL). Eric Smith (RPG Consultant) will conduct initial paired contrast and temporal analyses on mussel instream nutrient effects and the graduate student will examine how mussels are influencing primary production and carbon sediment dynamics.

Literature Cited:

1. Atkinson, C. L., & Forshay, K. J. (2022). Community patch dynamics governs direct and indirect nutrient recycling by aggregated animals across spatial scales. *Functional Ecology*, 36(3), 595-606.
2. Bruesewitz, D. A., Tank, J. L., Hamilton, S. K. (2009). Seasonal effects of zebra mussels on littoral nitrogen transformation rates in Gull Lake, Michigan, USA. *Freshwater Biology* 54(7), 1427-1433.
3. Bruesewitz, D. A., Gardner, W. S., Mooney, R. F., Pollard, L., Buskey, E. J. (2013). Estuarine ecosystem function response to flood and drought in a shallow, semiarid estuary: Nitrogen cycling and ecosystem metabolism. *Limnology and Oceanography*, 58(6), 2293-2309.
4. Hoellein, T. J., Zarnoch, C. B., Grizzle, R. E. (2015). Eastern oyster (*Crassostrea virginica*) filtration, biodeposition, and sediment nitrogen cycling at two oyster reefs with contrasting water quality in Great Bay Estuary (New Hampshire). *Biogeochemistry* 122(1), 113-129.
5. Hoellein, T. J., Zarnoch, C. B., Bruesewitz, D. A., & DeMartini, J. (2017). Contributions of freshwater mussels (Unionidae) to nutrient cycling in an urban river: filtration, recycling, storage, and removal. *Biogeochemistry*, 135(3), 307-324.

Scope of Work:

Issue Identification: This project is complimenting ongoing research to measure the effects of freshwater mussel restoration on water quality and stream ecosystem health in two urban, restored streams. Specifically, this project examines the pool habitat to stream reach impact of mussel reintroduction. We will be able to measure these pools to reach scale indicators of stream response to mussels bimonthly, giving us the ability to document how the ecosystem response to mussels' changes over the course of the seasons during baseflow conditions. Additionally, we will have additional student research capacity during the summer months to enhance sampling capabilities and to assist with mussel stocking.

Work Tasks:

| Task (Personnel) | Descriptions | Time Allocation |
|---|--|--|
| 1. Water sampling (VT technician, graduate student and S. Entrekin) | Bi-monthly water and sediment collection and Glade Run and Snakeden Run. This will include bottle preparation, labeling, sample inventory and travel to Reston, VA that will include one overnight trip. | 8 days/month 5/1/2024-10/31/2025 |
| 2. Transport (same as above) | Transport to the VT-SPES Central Soil & Water Quality Lab (SWQL), run by Ryan Stewart , for water and sediment analysis. | 3 days/month 5/1/2024-10/31/2025 |
| 3. Lab processing (SWQL) | Ryan Stewart will provide oversight of the lab processing for samples and Athena Tilley will communicate with PI Entrekin and graduate student and technician to prefer for each field trip. | 2 weeks/month 5/1/2024-10/31/2025 |
| 4. Data analysis (Graduate student, Entrekin, Brusewitz, Zarnoch,) | Bi-monthly data, and linking these data streams to data from the individual to mussel bed scale. | 2 weeks/month |
| 5. Enhanced summer sampling and experimentation: Undergraduate student summer experience (Entrekin, Brusewitz, Jones) | Two on-site undergraduate students who can conduct supplementary sampling. Two undergraduates will provide additional capacity to sample water chemistry parameters during or after storm events and other sampling needs identified by our data. For example, after our experiments in August 2023, we determined we would like some sampling effort on a small tributary that runs into our reach. Undergraduate students will also support ongoing mussel stocking efforts, mussel movement within the streams, sediment and benthic invertebrate sampling, and support of mussel feeding and nutrient cycling experiments. | 2 months/year 6/15 to 8/15 each summer 2024 and 2025 |

Quality Assurance/Quality Control: See appended document.

Determination of Goals: We will send all data to Eric Smith to conduct the primary statistical analysis of the paired data sets and then use that to further address the variability observed and predicted by our USGS collaborators. The rest of the Team will use the data to address questions posed in the primary (first) proposal on the extent to which mussels change stream biogeochemistry.

Budget and Budget Details:

| Project Title: Mussel reestablishment as a best management practice: scaling their nutrient effects from the individual to the stream. | | | | |
|---|---|----------------------|--|---------------------------------|
| Principal Investigator: S. Entekin | | | | |
| Organization: Virginia Polytechnic Institute and State University (Virginia Tech) | | | | |
| Requested Duration in Months: 18 Months | | | | |
| Item | Unit Rate (A) | Units (B) | Quantity (C) | Cost (D = A x C) |
| Salaries GRA_AY 1 GRA_SMR 1 GRA_AY 2 GRA_SMR 2 | AY 1-\$25,787 Summer 1-\$8,208 AY 2-\$8,978 Summer 2-\$8,618 | %FTE | 9 academic months 3 summer months 3 academic months 3 summer months | \$51,591 |
| Wage Students (3) | Calendar Y1-\$18,375 Calendar Y2-\$7,875 | %FTE | 12 CY months each 2 @ 6 CY months each 1 @ 3 CY months | \$26,250 |
| Lab Specialist – staff | Calendar Year 1- \$22,969 | %FTE | 6 CY months 0.6 CY months | \$23,332 |
| Research Faculty | Calendar Year 2- \$2,363 | %FTE | 6 CY months 3 CY months | \$49,467 |
| | Calendar Year 1 - | | | |

| | | | | |
|-------------------------------|---|---------------------|--|-----------|
| | \$32,667 Calendar Year 2 - \$7,875 | | | |
| Benefits GRA AY GRA SMR | 9.19% through 6/30/24, then 9.85% | Negotiated Rates | 9 academic months 3 summer months 3 academic months 3 summer months | \$5,037 |
| Wage Student | 6.34% through 6/30/24, then 6.65% | Negotiated Rates | 12 CY months each 6 CY months each | \$1,736 |
| Lab Specialist – staff | 48% through 6/30/24, then 47.72% | Negotiated Rates | 6 CY months 0.6 CY months | \$12,100 |
| Research Faculty | 35.52% through 6/30/24, then 35.10% | Negotiated Rates | 6 CY months 3 CY months | \$17,386 |
| Tuition | AY 1-\$16,141 AY 2-\$5,615 | LS LS | 9 academic months 3 academic months | \$21,756 |
| Publication Fees | Year 1 - \$0 Year 2 - \$1000 | Publication | 2 total | \$2000.00 |
| Supplies | Calendar Y 1-\$24,930 Calendar Y 2-\$6,415 | LS | 12 CY months 6 CY months | \$31,345 |
| Equipment | Calendar Y 1- \$25,000 Calendar Y 2-\$0 | LS | 12 CY months 6 CY months | \$25,000 |
| Travel | Calendar Y 1-\$18,916 Calendar Y 2-\$10,958 | LS | 12 CY months 6 CY months | \$29,874 |
| Other Direct Cost | Calendar Y 1-\$15,370 Calendar Y 2- \$6,935 | LS | 12 CY months 6 CY months | \$22,305 |
| Total Direct Cost | CY 1-\$235,343 CY 2-\$85,836 | | | \$321,179 |

| | | | | |
|---------------------|---|---------------------|-----------------------------|-----------|
| Indirect Cost (26%) | Calendar Y 1- \$50,439 Calendar Y 2- \$20,857 | Negotiated Rates | 12 CY months 6 CY months | \$71,350 |
| Total Cost | Calendar Y 1- \$285,836 Calendar Y2- \$106,693 | | | \$392,529 |

Budget Narrative

Senior PERSONNEL

Sally Entekin – Dr. Entekin is a Principal Investigator (PI) and will oversee the water sampling, data collation and analysis. She is also managing the graduate student, undergraduate students and the technician. No funds requested.

Ryan Stewart – Dr. Stewart is a Co-Principal Investigator (Co-PI) and will oversee the water quality analysis efforts. Dr. Stewart will supervise the Research Faculty and Student Wage Employee. No funds requested.

Paul Angermeier – Dr. Angermeier will continue to be involved in the execution of this work. No funds are requested.

Jess Jones – Dr. Jones will continue to be involved in the execution of this work. No funds are requested.

OTHER FUNDED PERSONNEL

Graduate Students – Funds are requested to support 100% time for a graduate student that will work in the Virginia Tech Entomology department. This individual is responsible for collecting, analyzing, and developing additional novel research questions in the context of this high-frequency water sampling. We request 9 months of their academic salary and 3 months of summer salary in period 1-, and 3-months academic salary plus 3 months summer salary in period 2.

| Salary | % Effort | Duration | Funds Requested |
|---------------|----------|-----------|-----------------|
| \$2,736/month | 100% | 18 months | \$51,591 |

Research Faculty – Funds are requested to support 50% time for a research faculty member that will work in the Virginia Tech Central Soil and Water Lab (VT-CSWL). This individual is responsible for completing sample chain of custody forms, overseeing all water quality analyses performed in the VT-CSWQL, maintaining appropriate quality assurance procedures, and

reporting water quality analysis results.

| Salary | % Effort | Duration | Funds Requested |
|----------|----------|-----------|-----------------|
| \$64,000 | 50% | 18 months | \$49,467 |

Wage Employees – Three undergraduate students will be hired each year to help with project activities during the academic year and for summer period 1 and period 2.

| Salary | % Effort | Duration | Funds Requested |
|---------|-----------|-----------|-----------------|
| \$6,000 | 300%/250% | 18 months | \$26,250 |

Lab Specialist –The lab specialist will help with sample collection, data entry and project coordination.

| Salary | % Effort | Duration | Funds Requested |
|----------|----------|-----------|-----------------|
| \$45,000 | 50%/10% | 18 months | \$25,332 |

For budget preparation purposes only, and per Virginia Tech policy, an escalation factor of 5% has been added to all budgeted personnel funds, effective December 1 of each year for the wage, staff, and faculty positions, and effective August 16 of each year for graduate students. Actual salaries and wages will be charged to the project.

FRINGE BENEFITS

| Personnel Category | Rate | Funds Requested |
|--------------------|---------------|-----------------|
| Graduate Student | 9.19%/83% | \$5,037 |
| Research Faculty | 35.53%/35.10% | \$17,386 |
| Lab Specialist | 48%/47.72% | \$12,100 |
| Wage | 6.34%/6.65% | \$1,227 |
| Total | | \$36,259 |

Fringe Benefits are calculated in accordance with Virginia Tech’s federally negotiated fringe rate agreement. Rates are as follows:

| FRINGE RATES | Through 6/30/24 | On/After 7/1/24 |
|---------------------------|--------------------|--------------------|
| Regular Faculty | 32.78% | 32.96% |
| Special Research Faculty | 35.52% | 35.10% |
| Part Time Faculty | 26.41% | 27.50% |
| SMR Faculty/Wage Employee | 6.34% | 6.65% |
| GRA | 9.19% | 9.83% |
| Classified Staff | 48.00% | 47.72% |

TRAVEL

Period 1 will require 2 people to travel to Reston, VA from Blacksburg, VA 24 times for a total mileage of $(258 \times 2) \times 24$ trips @ \$0.51/mile for a total of \$6,315.84. Each of the trips will require overnight expenses that will be \$200/room @ 2 rooms x 24 trips for a total of \$9,600. We also request \$1500 for one project member to attend and present at the Society for Freshwater Science in Period 1 and \$500 for Tim Hoellein, a project team member, to travel from Chicago to Reston, VA for help in experimentation.
Total=\$18,915.84

Period 2 will require 12 trips for 2 people to travel to Reston, VA from Blacksburg, VA for a total mileage of $(258 \times 2) \times 12$ trips @ \$0.51 for a total of \$3,157.92. Each of the trips will require overnight expenses that will be \$200/room @ 2 rooms x 12 trips for a total of \$4,800, We also request \$1500 for the graduate student to attend and present at the society for Freshwater Science in Period 2 and \$500 for Tim Hoellein, a project team member, to travel from Chicago to Reston, VA for help in experimentation.
Total=10,957.92

Total: \$29,873.76

EQUIPMENT

Another EXO YSI Sonde is requested for water quality, hourly measurements that will be used to replace PI Entrekin's lab sonde that is currently being used but is needed for other projects.
Total: \$20,000 in period 1

These costs also include the purchase of one field laptop that will be exclusively used for this project. Having a field laptop for data download is critical to this project due to the nature of the work and the amount of data used.

Total: \$5,000 in period 1

SUPPLIES

For Dr. Entrekin's work on this project, supplies are requested for sensor replacement, batteries, macroinvertebrate nets, and other costs associated with the upkeep and maintenance of sensors. Total material cost is \$12,000 in period 1.

For Dr. Stewart's work on this project, supplies are requested for reagents and materials needed to analyze water samples for NH₃, NO₂+NO₃, ortho-P, TSS, TOC + TN, DOC, Total P, and other requested analytes. Total material cost is \$19,345 (\$12,930 in period 1, and \$6,415 in period 2).

PUBLICATION COSTS

Publication fees for 2, open access manuscripts are requested for Period 2 for a total cost of \$2000.

OTHER DIRECT COSTS - CONTRACTUAL SERVICES

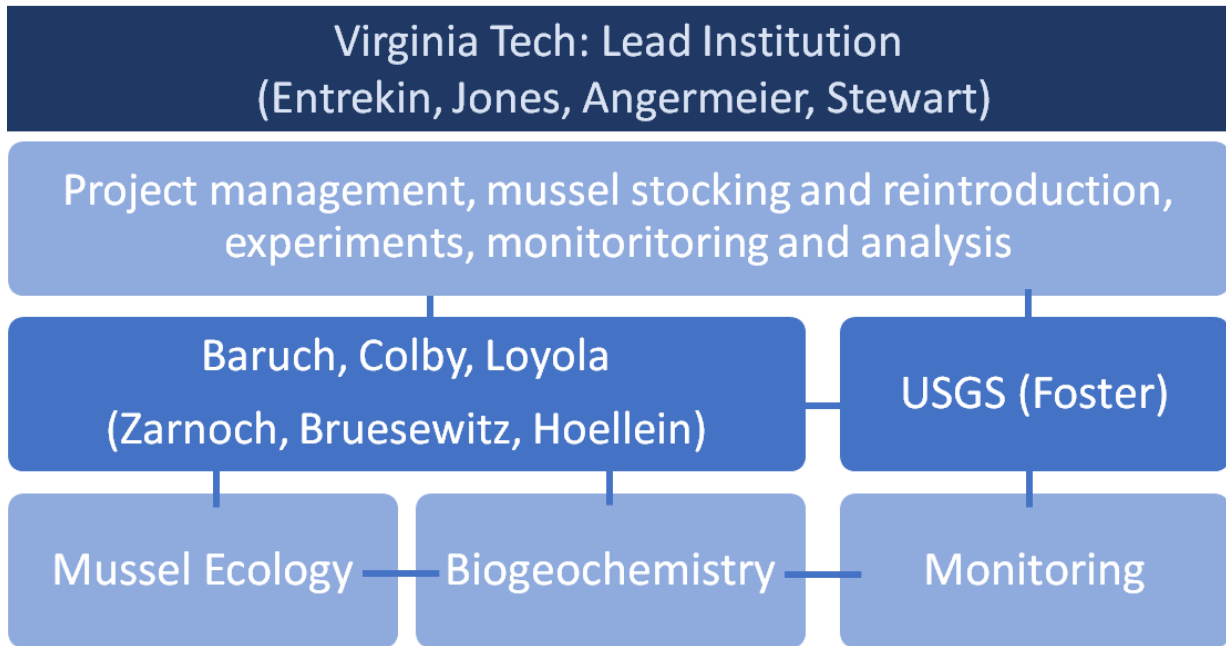
For Dr. Entrekin's work on the project, funds are requested to pay for per-sample charges associated with the VT Fleet Services cost center to maintain and repair the single lab truck. This truck will be used for this project 70% of the time over the 18-month time period. The rate for each analysis is approved by the university Controller's Office. The total request for contractual services is \$4500 (\$3500 in period 1 and \$1000 in period 2).

Funds are requested to pay for per-sample charges associated with the CSWQL cost center and other cost center laboratories on the Virginia Tech campus (e.g., Civil and Environmental Engineering). The rate for each analysis is approved by the university Controller's Office. The total request for contractual services is \$17,805 (\$11,870 in period 1, and \$5,935 in period 2).

INDIRECT COSTS

Virginia Tech's federally-negotiated rate for off-campus research is 26% of modified total direct cost (MTDC). A copy of VT's current federally negotiated indirect rate agreement can be found at <https://osp.vt.edu/resources/rates/indirect-costs.html>. The total amount of indirect requested is \$71,350.

Organizational Chart: Provide an organizational chart depicting the structure of your team.



Sally Ann Entrekin

Contact information

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5978
Entomology
Blacksburg, VA

Phone: (540) 231-

E-mail: sallye@vt.edu

Education

| | | | |
|-------|---|------------|------|
| Ph.D. | University of Notre Dame, Notre Dame, IN | Biology | 2008 |
| M.S. | University of Georgia, Athens, GA | Entomology | 2000 |
| B.S. | Georgia Southwestern State University, Americus, GA | Biology | |
| | 1996 | | |

Appointments:

Associate Professor, Dept. of Entomology, Virginia Tech, 2018-present
Adjunct Faculty, Dept. of Biology, University of Central Arkansas, 2018-present
Associate Professor, Dept. of Biology, University of Central Arkansas, 2014-2018
Adjunct Faculty, Dept. of Biological Sciences, University of Arkansas, 2014-present
Assistant Professor, Dept. of Biology, University of Central Arkansas, 2008-2014
Research/Teaching Assistant, Dept. of Biological Sciences, University of Notre Dame, 2003-2008
Project Coordinator, Institute of Ecology/Entomology, University of Georgia, 2001-2003
Biologist/Coordinator, Dept. of Biology, Murray State University, 2000-2001
Research/Teaching Assistant, Dept. of Entomology, University of Georgia/Jones Ecological Research Center, 1997-2000
Research Technician, Jones Ecological Research Center, 1996-1997

Recent Research grants

08/2020-05/2021- A Case Study for Prioritizing Stream Restoration Efforts in the City of Roanoke. Roanoke City, Virginia. \$68,250. T. Thompson, C. Hession. S. Entrekin
08/16/2020-08/15/2025- Effects of BMPs and land use on stream macroinvertebrates and fishes in the Chesapeake basin, US Geological Survey. \$635,929. P. Angermeier and S. Entrekin.
07/01/2019-06/29/2020 - June 30, 2020. Interdisciplinary Seed Grant RFP, Integrative Science and Solutions for Freshwater Systems, Fralin Life Science Institute in support of the Global System Sciences Area at Virginia Tech \$8,400. S.A. Entrekin, E.R. Hotchkiss, D.L. McLaughlin, C.E. Zipper, A.J. Timpano, and T.R. Cianciolo. Using benthic and emergent insect biomass as a metric of stream impairment across a salinity gradient in central Appalachian headwater streams. Grant # 120254
03/01/19 - 07/30/20-Macroinvertebrate secondary production in 3 watersheds before stream liming. Shenandoah Trust Research Grant Program. \$15,000. Principal Investigator.
12/17-12/19- Metlife Foundation Study Abroad Innovation Grant through the 100,000 Strong in the Americas program. *Summer Language and Culture Immersion through Ecological Sustainability*. \$56,000. Co-Investigator.

10/17-07/20-Arkansas Natural Resources Commission. *Lake Conway-Point Remove Watershed Monitoring and Alliance (LCPRWA)*. \$118,774. Co-Investigator.
03/18-03/19-USGS 104B, *Do stream phosphorus dynamics correspond with biological condition in the Lake Conway Point Remove Watershed, Arkansas?* \$23,432. Principal Investigator.
08/16-07/19-Arkansas Department of Environmental Quality, *Water quality education and Stone Dam Creek planning grant*. \$40,000.00. Co-Investigator.
03/16-03/17- USGS 104B, *Biological and ecological consequences of sub-lethal ion concentrations on microbial and macroinvertebrate detritivores*. \$13,000. Principal Investigator.

Entrekin selected publications (* student)

Gruntz*, C. P., Entrekin, S. A., Evans-White, M. A., & Clay, N. A. (2022). Too much of a good thing: Evidence of sodium stress in an inland subtropical riparian detrital system. *Applied Soil Ecology*, 169, 104194.

Vanlandingham*, A. L., Walker, R. H., Alford, A., & Entrekin, S. A. (2021). Intermittency mediates macroinvertebrate and crayfish effects on leaf breakdown in temperate headwater streams. *Freshwater Science*, 40(1), 21-38.

Trentman*, M.T., Tank, J.L., Braund*, D. and Entrekin, S.A., 2021. Agricultural layering explains variation in sediment P dynamics in streams draining two distinct agricultural biomes. *Aquatic Sciences*, 83(1), pp.1-11.

Bosio, S.F., Shirey, P.D., **Entrekin, S.A.**, Hoellein, T.J., Moerke, A.H., Rosi, E.J., Tank, J.L. and Lamberti, G.A., 2021. Dynamics of large wood added to Midwestern USA streams. *River Research and Applications*.

Entrekin, S.A., EJ Rosi, JL Tank, TJ Hoellein, GA Lamberti. 2020. [Quantitative Food Webs Indicate Modest Increases in the Transfer of Allochthonous and Autochthonous C to Macroinvertebrates Following a Large Wood Addition to a Temperate headwater](#) stream. *Frontiers in Ecology and Evolution* 8, 114

Stepanian, P.M., **Entrekin S.A.**, Wainwright C.E., Mirkovic D., Tank J.L. and Kelly J.F. 2020. Declines in an abundant aquatic insect, the burrowing mayfly, across major North American waterways. *Proceedings of the National Academy of Science*.

Halvorson*, H.M., Fuller*, C.L., **Entrekin, S.A.**, Scott, J.T. and Evans-White, M.A., 2019. Interspecific homeostatic regulation and growth across aquatic invertebrate detritivores: a test of ecological stoichiometry theory. *Oecologia*, 190(1), pp.229-242.

Entrekin, S.A., Clay, N.A., Mogilevski*, A., Howard-Parker, B. and Evans-White, M.A., 2018. Multiple riparian–stream connections are predicted to change in response to salinization. *Philosophical Transactions of the Royal Society B*, 374(1764), p.20180042.

Halvorson, H.M., Fuller*, C.L., **Entrekin, S.A.**, Scott, J.T. and Evans-White, M.A., 2018. Detrital nutrient content and leaf species differentially affect growth and nutritional regulation of detritivores. *Oikos*.

Entrekin, S., Howard-Parker, B., Evans-White, M. and Clay, N., 2018. Biological and Ecological Consequences of Sub-Lethal Ion Concentrations on Microbial and Macroinvertebrate Detritivores. *Arkansas Bulletin of Water Research*.

Entrekin, S.A., Austin, B.J., Evans-White, M.A. and Haggard, B.E., 2018. Establishing the linkage among watershed threats, in-stream alterations and biological responses remains a challenge: Fayetteville Shale as a case study. *Current Opinion in Environmental Science & Health*.

Entrekin, S., Trainor, A., Saiers, J., Patterson, L., Maloney, K., Fargione, J., Kiesecker, J., Baruch-Mordo, S., Nicot, J.P., Konschnik, K., Wiseman, H., Ryan, J. 2018. Water stress from high volume hydraulic fracturing threatens aquatic biodiversity and ecosystem services in Arkansas, U.S. *Environmental Science and Technology*.

Baker, L.* , Evans-White, M., and **Entrekin, S.** 2018. Basin vulnerability explains patterns of biological degradation in small streams in the Fayetteville Shale, AR. *Ecological Indicators*.

Craig, L., Olden, J., Arthington, A., **Entrekin, S.** Hawkins, C., Kelly, J., Kennedy, T., Maitland, M., Rosi, E., Roy, A., Strayer, D., Tank, J., West, A., Wooten, M. 2017. The science and management of multiple, interacting threats in freshwater ecosystems. *Elem Sci Anth*, 5.

Kelso, J.* and **Entrekin, S.** 2018. Intermittent and perennial macroinvertebrate communities had similar richness but differed in species trait composition depending on flow duration. *Hydrobiologia*, 807(1), 189-206.

Halvorson*, H. E. Scott*, **S. Entrekin**, M. Evans-White, T. Scott. 2016. Light and dissolved phosphorus interactively affect leaf litter microbial metabolism, stoichiometry and decomposition. *Freshwater Biology* 61 (6): 1006-1019.

Jess Jones

U.S. Fish and Wildlife Service
106a Cheatham Hall
Department of Fisheries and Wildlife Sciences
Virginia Tech University
Blacksburg, Virginia 24061-0321

Office number: (540)-231-2266
Lab number: (540)-231-7241
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Email address: Jess_Jones@fws.gov

Education:

2004-2009 Ph.D. Fisheries Science, Virginia Polytechnic Institute and State University
2001-2004 M.Sc. Fisheries Science, Virginia Polytechnic Institute and State University
1993-1996 B.Sc. Fisheries Science, Virginia Polytechnic Institute and State University

Background and Work Interests:

I am a Restoration Biologist with U.S. Fish and Wildlife Service, stationed in the Department of Fish and Wildlife Conservation at Virginia Tech University, Blacksburg, where I also serve as an Associate Professor and Director of the Freshwater Mollusk Conservation Center.

My research focuses on conservation biology of freshwater mussels, with an emphasis on: (1) restoration ecology, (2) population demography and modeling, (3) population genetics and phylogenetics, (4) ecotoxicology, (5) conservation aquaculture, and (6) quantitative monitoring and field techniques. I am particularly interested in improving field and laboratory techniques to restore mussel populations to enhance the capability of the Department of Interior Natural Resource Damage Assessment and Restoration (NRDAR) program, including applying small- and large-scale mesocosms to improve captive growing and experimental conditions for endangered mussels and fishes.

Employment:

2004-Present Restoration Biologist, U.S. Fish and Wildlife Service, Department of Fish and Wildlife Conservation, Virginia Tech, Blacksburg. The primary duties of this position include:

- Provide project oversight and conduct contracting with university, state and non-governmental cooperators to restore endangered mussel and fish populations in fulfillment of the restoration goals of two multi-million dollar NRDAR case settlements.
- Design and implement complex studies and projects to quantitatively monitor and restore mussel populations in the Clinch and Powell Rivers, including ecotoxicological assessments of damaged and degraded sections of each river.
- Provide technical assistance to federal and state agencies that participate in the restoration and conservation of mussels in the southeastern United States.
- Disseminate project results to agency cooperators, the scientific community and public through written reports, publications, presentations and briefings

Instruction, Employee Supervision and Graduate Student Advising at Virginia Tech:

In 2006, I developed and have co-taught every 2 years thereafter, the graduate course *Applied Conservation Genetics* (6984) and on request, provided guest lectures to other departmental courses at Virginia Tech, such as *Fisheries Techniques* (3114), *Aquatic Entomology* (4354), *Environmental Problems, Population & Development* (3104), *Conservation Biology* (4314), *Aquaculture* (3114), and *Ichthyology* (4514).

I provide supervision and training of FMCC staff to propagate, culture, monitor and conduct research on mussel populations in the Clinch and Powell rivers of southwestern Virginia and Northeastern Tennessee. Staff typically includes 8-12 employees at various stages of professional development, from post-doctoral scientists, Ph.D. and Master level graduate students, full-time technicians, and part-time undergraduate students. All FMCC employees are involved in meeting the assessment and restoration goals of NRDAR case projects. Under a Memorandum of Agreement between the Service and Virginia Tech University, I serve as a liaison officer between the Service and University. In this capacity, I serve on the Virginia Tech faculty at the rank of Associate Professor to cooperatively manage FMCC and communicate the NRDAR project goals to staff and faculty. I have advised a total of 17 graduate students at Virginia Tech and as a committee member as an adjunct faculty member at other universities.

Engagement and Outreach:

Every year, I assist with numerous tours of the FMCC to educate students and the public about aquatic endangered species and the role that conservation aquaculture plays to recover species. I regularly provide technical assistance to state and federal cooperators and professional colleagues on mussel propagation and restoration techniques, pre-review of proposals and manuscripts, as an invited referee to scientific journals, and as a subject Editor for two journals. I have also been involved in numerous public outreach events to showcase FMCCs mussel release efforts. For example, our 2012 mussel release event in the Powell River, Tennessee was covered by local television stations in Tennessee and Kentucky, Lincoln Memorial University, media outlets in Regions 4 and 5 of the U.S. Fish and Wildlife Service, was picked-up by the Associated Press and USA Today, and covered by a reporter from *Science Magazine*, which culminated in a featured article and video documentary in the journal about mussel conservation in the United States, highlighting efforts at Virginia Tech. I have also been involved in academic exchanges with international colleagues from South Africa, Brazil and China, providing technical assistance more broadly.

Awards and Honors:

2013 U.S. Fish and Wildlife Service Rachel Carson Award for Excellence in Science
2011 U.S. Fish and Wildlife Service STAR Award for exemplary performance
2007 U.S. Fish and Wildlife Service STAR Award for exemplary performance
2005 William Preston Thesis Award – The award is for the best original research with the potential to benefit all people. This annual award is the highest academic award for master's degree students given by Virginia Tech University.

Research, Scholarly Publications, and Grants (Career Totals):

Papers in refereed journals: 68

Submitted manuscripts: 4

Peer-Edited Technical Reports: 48

Conference and technical presentations: 122

Sponsored Research: 60 grants and cooperative agreements, total=\$4,885,492.00

Denise Ann Bruesewitz

Environmental Studies Department
Colby College
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Waterville, ME 04901 U.S.A.

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E-mail: dabruese@colby.edu

EDUCATION

Ph.D.

2008. Department of Biological Sciences, University of Notre Dame, Notre Dame, IN
Dissertation: The effects of invasive zebra mussels (*Dreissena polymorpha*) on nitrogen cycling in freshwater ecosystems of the Midwestern United States.
Advisor: Dr. Jennifer L. Tank. Advising committee: Drs. Gary Lamberti, David Lodge, and Stephen Hamilton

B.S. *summa cum laude*

2001. Biology, Winona State University, Winona, MN
Undergraduate Thesis: Association between annual hydrological patterns and invertebrate dynamics in a large river.
Advisor: Dr. Michael Delong

ACADEMIC AND PROFESSIONAL EMPLOYMENT

Associate Professor & Chair of Environmental Studies
July 2019-present

Associate Professor & Associate Chair of Environmental Studies
January 2019-July 2019

Head Faculty-in-Residence & Faculty Liaison to The Office of Civic Engagement and Community Partnerships, Colby College *2018-2020*

Assistant Professor

September 2012-December 2018. Environmental Studies Program, Colby College.

Research Associate

November 2011-August 2012. Marine Science Institute, University of Texas at Austin with Drs. Ed Buskey and Wayne Gardner

Postdoctoral Research Fellow

June 2010-November 2011. Marine Science Institute, University of Texas at Austin with Drs. Ed Buskey and Wayne Gardner

Postdoctoral Research Fellow

June 2008- May 2010. Lake Ecosystem Restoration New Zealand (LERNZ), Department of Biological Sciences, University of Waikato, New Zealand with Dr. David Hamilton

SELECTED FUNDED GRANTS

“RII Track-1: Molecule to Ecosystem: Environmental DNA as a nexus of Coastal Ecosystem Sustainability for Maine (Maine-e-DNA) as senior personnel and lead at Colby College to the National Science Foundation \$20,000,000 8/1/2019-7/31-2023

“RII Track-2 FEC: Computational Methods and Autonomous Robotics Systems for Modeling and Predicting Harmful Cyanobacterial Blooms” as senior personnel and lead at Colby College to the National Science Foundation \$5,989,336 9/1/2019-8/31/2023

“Evaluating the potential for mutualistic species interactions to enhance restoration success and ecosystem services in urban salt marshes” as co-PI (PI Mary Alldred) to the Hudson River Foundation \$157,224, 1/1/2020-12/31/2021

"CUE Ethics: Collaborative Research: Evaluating Frameworks for Incorporating Computing Across the Curriculum," as a collaborator with PIs Bruce A. Maxwell, Ying Li, and Stephanie R. Taylor to National Science Foundation \$121,689, 2/1/2020-7/31/2021

‘Undergraduate Research into the Cultural, Economic, and Ecological Significance of Church Forests in South Gondar, Ethiopia as co-PI with Drs. Travis Reynolds and Margaret Lowman to National Science Foundation SMA SBE Office of Multidisciplinary Activities. Co-PI from 2016-2018 (\$406,038.00)

‘Plankton N cycling and detection of cyanobacterial blooms across a gradient of lake trophic state’ as PI with Drs. Whitney King and Pete Countway to Maine Water Resources Research Initiative, April 2015 (\$15,000)

Mechanisms of nutrient retention in restored salt marshes: Will marsh restoration in eutrophic ecosystems provide ecosystem services of nitrogen removal and carbon sequestration?” as co-PI with Drs. Hoellein and Zarnoch to Hudson River Foundation, July 2015 (\$33,640)

‘Ecosystem response to enhanced nutrient loadings following Hurricane Sandy in the Long Island South Shore Estuary as co-PI with Drs. Hoellein and Zarnoch to New York Sea Grant, January 2013 (\$24,731)

‘The influence of oyster restoration on nitrogen cycling in the Hudson Raritan estuary as co-PI with Drs. Hoellein (PI), Zarnoch, Branco and Gardner awarded by The Hudson River Foundation, March 2011 (\$152,006)

SELECTED EXTERNAL SCHOLARLY ACTIVITIES & COMMUNITY ENGAGEMENT

Elected to Maine Innovation Economy Advisory Board, Spring 2022-*ongoing*
Kennebec Water District Trustee representing city of Waterville (2018-*ongoing*; 3-year elected term)

Seven Lakes Alliance Science Advisory Board (2017-2019)

Co-developed and co-organized regional meetings of “Northeast GLEON” for undergraduate students at the Carey Institute for Ecosystem Studies, Millbrook NY; annual conference starting in April 2015.

SELECTED INVITED SEMINARS & PANELS

| | |
|------|--|
| 2019 | Panel Discussion: Artists Need to Create on the Same Scale that Society has the Capacity to Destroy, Colby Museum of Art and Lunder Institute for American Art |
| 2017 | Marine Biological Lab (MBL), Woods Hole, MA |
| 2015 | Northeast GLEON plenary speaker |
| 2014 | Virginia Tech (October 2014) |
| 2014 | University of Maine Biology Department (March 2014) |
| 2014 | Belgrade Lakes Association, Belgrade Maine |
| 2014 | Lake Sunapee Protective Association Board Meeting (January 2014) |

Paul L. Angermeier

Current position (since 2006): Research Scientist (GS-15), US Geological Survey, Cooperative Fish and Wildlife Research Unit; Professor, Department of Fish and Wildlife Conservation, Virginia Tech, Blacksburg, VA 24061-0321
Telephone: (540) 231-4501 Fax: (540) 231-7580 Email: biota@vt.edu

Previous position (1996 - 2006): Research Scientist, USGS, Cooperative Fish and Wildlife Research Unit; Associate Professor, Department of Fisheries and Wildlife Sciences, Virginia Tech.

Education: BS Purdue University, Environmental Science, 1975

MS University of Illinois, Ecology, 1979; PhD University of Illinois, Ecology, 1983

Team Teaching Experience at Virginia Tech (since 2010):

- 2010 – 2011 - Constructing Sustainability
- 2013 – 2014 - Biodiversity Conservation & Environmental Sustainability
- 2019 – 2020 - Interfaces of Global Change Advanced Seminar

Selected Professional service (since 2010):

Editorial Board, *Freshwater Biology*, 1997-present

Editorial Board, *Conservation Biology*, 2000-present

American Fisheries Society liaison to Society for Conservation Biology, 2007-2020

USFWS Aquatic Systems Community of Practice for Appalachian Landscape Conservation Cooperative, 2013-2016

Selected Awards (since 2010):

U. S. Department of the Interior Annual Performance Award: 2017, 2019

Selected Grants as Principal or Co-principal Investigator (since 2017):

Development of an eDNA protocol for detecting candy, variegated, and Kanawha darters. Virginia Department of Game and Inland Fisheries. \$20,500.

Development of a landscape model to predict reach-level sources of Roanoke logperch larvae in the upper Roanoke River system. National Fish and Wildlife Foundation. \$125,000.

Farm Bill conservation practice (BMP) efficacy in mitigating livestock impacts in the Copper Creek watershed. Natural Resources Conservation Service. \$70,000.

Assessment of the dynamics and biotic impacts of fine sediment to assist conservation of stream fishes in the Dan and Roanoke river basins. National Fish and Wildlife Foundation. \$189,999.

Ecosystem services in the Roanoke River basin. National Fish and Wildlife Foundation. \$89,560.

Development of a structured decision-making framework to guide translocation of imperiled aquatic species in the Roanoke and Dan river basins. National Fish and Wildlife Foundation. \$94,000.

Evaluating efficacy of agricultural BMPs in the upper Clinch, Powell, and Holston river drainages. Natural Resources Conservation Service. \$121,000.

Effects of BMPs and land use on stream macroinvertebrates and fishes in the Chesapeake basin. U.S. Geological Survey. \$635,929.

Developing a predictive model for in-stream embeddedness to link physical processes with biotic responses. Virginia Tech Global Change Center. \$15,000.

Development and application of a multiscale model of habitat suitability for Candy Darter. Virginia

- Department of Wildlife Resources. \$203,615.
- Targeted surveys to prevent the extinction of Slender Chub *Erimystax cahni*. U.S. Fish and Wildlife Service. \$60,000.
- Coupling social science and watershed modeling to improve ecological health of streams in agricultural landscapes. Virginia Tech Global Change Center and Institute for Society, Culture and Environment. \$15,000.
- Continuation of evaluating efficacy of agricultural BMPs in the upper Clinch, Powell, and Holston river drainages. USDA Natural Resources Conservation Service. \$49,701.

Selected refereed publications since 2017 (out of >140 total):

- Villamagna, A.M., B. Mogollón, and P.L. Angermeier. 2017. Inequity in ecosystem service delivery: socioeconomic gaps in the public-private conservation network. *Ecology and Society* 22 (1):36. [online] <http://www.ecologyandsociety.org/vol22/iss1/art36/>.
- Argentina, J.E., P.L. Angermeier, E.M. Hallerman, and S.A. Welsh. 2018. Spatial extent of analysis influences observed patterns of population genetic structure in a widespread darter species (Percidae). *Freshwater Biology* 63: 1185–1198.
- Buckwalter, J.D., E.A. Frimpong, P.L. Angermeier, and J.N. Barney. 2018. Seventy years of stream-fish collections reveal invasions and native range contractions in an Appalachian (USA) watershed. *Diversity and Distributions* 24: 219-232.
- Dunham, J.B., P.L. Angermeier, S.D. Crausbay, A.E. Cravens, H. Gosnell, J. McEvoy, M.A. Moritz, N. Raheem, and T. Sanford. 2018. Rivers are social-ecological systems: time to integrate human dimensions into riverscape ecology and management. *WIREs Water*. DOI: 10.1002/wat2.1291.
- Scott, L.N., A.M. Villamagna, and P.L. Angermeier. 2018. A new modeling approach to prioritize riparian restoration to reduce sediment loading in two Virginia river basins. *Environmental Management* 62: 721–739. <https://link.springer.com/article/10.1007/s00267-018-1078-6>.
- Angermeier, P.L. and J.R. Karr. 2019. Ecological health indicators. *Encyclopedia of Ecology* (2nd Edition) 1: 391-401 and online Reference Module in Earth Systems and Environmental Sciences. Elsevier, Oxford. <https://doi.org/10.1016/B978-0-12-409548-9.10926-1>.
- Dunn, C.G. and P.L. Angermeier. 2019. Remaining populations of an upland stream fish persist in refugia defined by habitat features at multiple scales. *Diversity and Distributions* 25: 385–399. DOI:10.1111/ddi.12866.
- Govenor, H., L.A.H. Krometis, L. Willis, P.L. Angermeier, and W.C. Hession. 2019. Macroinvertebrate sensitivity thresholds for sediment in Virginia streams. *Integrated Environmental Assessment and Management* 15: 77-92. DOI: 10.1002/ieam.4086.
- Buckwalter, J.D., P.L. Angermeier, J. Argentina, S. Wolf, S. Floyd, and E.M. Hallerman. 2019. Drift of larval darters (Family Percidae) in the upper Roanoke River basin, USA, characterized using phenotypic and DNA barcoding markers. *Fishes* 4(4), 59; [doi:10.3390/fishes4040059](https://doi.org/10.3390/fishes4040059).
- Buckwalter, J.D., E.A. Frimpong, P.L. Angermeier, and J.N. Barney. 2020. Species traits predict stream-fish invaders in an Appalachian (U.S.A) river basin. *Freshwater Biology* 65: 552-564. <https://doi.org/10.1111/fwb.13453>.
- Martin, Z.P., P.L. Angermeier, S. Ciparis, and D.J. Orth. 2021. Coal-mining intensity influences species and trait distributions of stream fishes in two Central Appalachian watersheds. *Ecology of Freshwater Fish* 30: 347-365. DOI:10.1111/eff.12588.
- Angermeier, P.L., L.A. Krometis, M.J. Stern, and T.L. Hemby. 2021. Exploring relationships among stream health, human well-being, and demographics in Virginia, USA. *Ecological Indicators* 121 107194. DOI.org/10.1016/j.ecolind.2020.107194.
- Sleezer, L.J., P.L. Angermeier, E.A. Frimpong, and B.L. Brown. 2021. A new composite abundance metric detects stream fish declines and community homogenization during six decades of invasions. *Diversity and Distributions* 27: 2136-2156. <http://doi.org/10.1111/ddi.13393>

Chester B. Zarnoch

Professor, Environmental Studies
Baruch College, City University of New York
17 Lexington Ave, New York, NY 10010

(A) Professional preparation:

| Institution | Location | Major | Degree/Year | |
|--|-----------------|------------|-------------|------|
| Southampton College, Long Island Univ. | Southampton, NY | Biology | BS | 2000 |
| Brooklyn College, CUNY | Brooklyn, NY | Biology | MA | 2004 |
| Graduate School and Univ. Center, CUNY | New York, NY | Phil.Biol. | MA | 2004 |
| Graduate School and Univ. Center, CUNY | New York, NY | Biology | PhD | 2006 |

(B) Appointments:

1. 2019-Present- Baruch College, City University of New York (CUNY) – Professor of Environmental Studies
2. 2014-2019- Baruch College, City University of New York (CUNY) – Associate Professor of Environmental Studies
3. 2014-Present- Baruch College/ Graduate Center, CUNY – Graduate Deputy Chair in Biology
4. 2008-Present - Graduate Faculty, Ecology, Evolutionary Biology and Behavior - Biology Doctoral Program, Graduate Center, CUNY
5. 2007-2014 - Baruch College, CUNY – Assistant Professor of Environmental Studies
6. 2006-2007 - Brooklyn College, CUNY, Aquatic Research and Environment Assessment Center – Associate Director
7. 2000-2006 - Brooklyn College, CUNY, Aquatic Research and Environment Assessment Center- Graduate Assistant

(C) Relevant Publications:

Abbas, A., Cardenas, A., LaFond, D., Guasaquillo, B., Alldred, M., & Zarnoch, C. (2023). Loss of saltmarsh plants impacts ribbed mussels (*Geukensia demissa*) size, density, and influence on sediment nitrogen cycling. Wetland Ecology and Management. 31: 367-380.

Ayvazian, S., Mulvaney, K., Zarnoch, C., Palta, M., McNally, S., Reichert-Nguyen, J., Pilaro, M., Jones, A., McCarthy, G. J., Terry, C., & Fulweiler, R. W. (2021). Beyond bioextraction: the role of oyster-mediated denitrification in nutrient management. Environmental Science and Technology. 55 (21): 14457-14465.

Zhu, J., Zarnoch, C., Gosnell, J., Alldred, M. & Hoellein, T. (2019). Ribbed mussels (*Geukensia demissa*) enhance nitrogen-removal services but not plant growth in restored eutrophic salt marshes. Marine Ecology Progress Series. 631, 67-80.

Rosenzweig, B. R., Groffman, P. M., Zarnoch, C., Branco, B. F., Hartig, E. K., Fitzpatrick, J., Forgione, H. M., & Parris, A. (2018). Nitrogen regulation by natural systems in 'unnatural' landscapes: denitrification in ultra-urban coastal ecosystems. Ecosystem Health and Sustainability 4 (9), 205-224.

Hoellein, T. J., Zarnoch, C., Bruesewitz, D. A., & DeMartini, J. (2017). Contributions of freshwater mussels (Unionidae) to nutrient cycling in an urban river: filtration, recycling, storage, and removal. Biogeochemistry, 135(3), 307-324.

Lindemann, S., Zarnoch, C., Castignetti, D., & Hoellein, T. (2016). Effect of eastern oysters (*Crassostrea virginica*) and seasonality on nitrite reductase gene abundance (nirS, nirK, nrfA) in an urban estuary. Estuaries and Coasts, 39, 218-232.

Hoellein, T.J., Zarnoch, C., & Grizzle, R.E. (2015). Eastern oyster (*Crassostrea virginica*) filtration, biodeposition, and sediment nitrogen cycling at two oyster reefs with contrasting water quality in Great Bay Estuary (New Hampshire, USA). Biogeochemistry, 122, 113-129.

Hoellein, T. J., & Zarnoch, C. (2014). Effect of eastern oysters (*Crassostrea virginica*) on sediment carbon and nitrogen dynamics in an urban estuary. Ecological Applications, 24, 271-286.

(D) Synergistic activities:

1) *Student mentoring.* The mentoring of undergraduate research students is a significant responsibility at Baruch College and at CUNY. I have had 72 undergraduate students work in my lab over the last 16 years. Many of these students have worked with me for multiple years and have presented their work at local and national scientific meetings. I have also been active in mentoring high school students particularly from programs supporting minority students interested in STEM fields. I have had 45 high school students work in my lab with many moving to undergraduate programs in the sciences. Lastly, I am very much involved in graduate education at CUNY. I serve as the Graduate Deputy Chair in Biology at Baruch College which requires me to advise Biology PhD students on coursework, teaching responsibilities, and program requirements. I have also mentored and served on the committees for 10 CUNY PhD students.

2) *Professional activities and public service.* I have been an active member of the marine science community in New York. I am a board member of the New York Marine Sciences Consortium since 2008 and have been involved in organizing the consortium's annual conference. I serve on the advisory council for the New York Harbor Foundation's Billion Oyster Project and Harbor School's Aquaculture Program. I am also on the advisory council for Molloy College's Center for Environmental Research and Coastal Oceans Monitoring. I have been a member of the NY/NJ Harbor Estuary Program's Oyster Restoration Working Group since 2011 and have served on the Science and Technical Advisory Committee since 2017. Lastly, I serve on the Science and Technical Advisory Committee for the Long Island Sound Study.

POSITION

VIRGINIA POLYTECHNIC INSTITUTE AND STATE UNIVERSITY Blacksburg, VA
Associate Professor Aug. 2020 – present
Assistant Professor Dec. 2013 – Aug. 2020

EDUCATION

| <u>Institution</u> | <u>Date</u> | <u>Degree</u> | <u>Major</u> | <u>Minor</u> |
|---------------------------|-------------|------------------|----------------------|--------------|
| Oregon State University | 2013 | Post-Doc Scholar | - | - |
| Oregon State University | 2013 | Ph.D. | Water Resources Eng. | Soil Science |
| Oregon State University | 2010 | M.S. | Water Resources Eng. | - |
| Cal Poly, San Luis Obispo | 2002 | B.S. | Mechanical Eng. | - |

RESEARCH PROGRAM

- Quantifying effects of soil health management practices
- Measuring and modeling changes in water quantity and quality from disturbance
- Creating analytical tools for infiltration and runoff modeling
- Characterizing hydrological processes under different soil and land use conditions
- Assessing soil controls on exchange of water vapor and greenhouse gases
- Quantifying root water uptake processes
- Developing novel instrumentation to monitor environmental processes

PUBLICATIONS

- Authored/co-authored 90 peer-reviewed journal articles
- Authored/co-authored 1 book chapter, 4 conference proceedings, and 1 review

TEACHING

CSES 3614 – Soil Physical and Hydrological Properties (3 credits; every Spring)
ENSC 3634 – Physics of Pollution (3 credits; every Fall)
CSES/FREC 5144 – Watershed Hydrology (co-instructor; 3 credits; every Spring)

ADVISING

Served as major advisor for 2 post-doctoral scholars, 4 graduated Ph.D. students, and 4 graduated M.S. students. Currently advising 4 graduate students (2 Ph.D., 2 M.S.). Serving or served on 33 graduate committees (15 Ph.D., 18 M.S.) in multiple colleges and universities. Advising or advised 30 undergraduate research assistants (12 as independent research projects; 18 as research assistants).
Serving as faculty advisor for the Virginia Tech Environmental Student Organization (2014 – present).
Serving as faculty advisor for the Peace Corps Prep program at Virginia Tech (2020 – present).

SELECTED HONORS

- 2022 Fulbright U.S. Scholar Award – Visiting Professor, University of Innsbruck, Austria. Awarded by the U.S. Fulbright Program, Department of State.
- 2021 IMSC Early Career Award – Awarded by the International Soil Modeling Consortium.
- 2020 Inspiring Young Scientist Award – Awarded by the Environmental Quality Section of ASA.
- 2019 Soil Physics and Hydrology Early Career Award – Awarded by the Soil Physics and Hydrology Division of the Soil Science Society of America.
- 2016 Virginia Tech Scholar of the Week – Awarded by the Office of the Vice President for Research.

PROFESSIONAL SERVICE

1. Mentored five graduate students through SSSA Soil Physics and Hydrology Mentoring Program (2015-2019); invited panelist for SSSA Meeting session: "Student Networking Session-Building Professional Relationships Workshop!" (2019).
2. Convener/Moderator for the Soil Science Society of America (SSSA) and American Geophysical Union (AGU) Annual Meetings. Since 2015 I have convened more than 12 technical sessions.
3. Member of the AGU Unsaturated Zone Technical Committee (2018-present).
4. Member of the ASA-CSSA-SSSA committees: S483 Don and Betty Kirkham Soil Physics Award (2016-2017); ACS530 Early Career Board (2019-2022); S471 Soil Science Research Award Committee (2020-2022); ACS449.8 Student Research Symposium Committee (2020-2022).
5. Chair-elect of the SSSA Soil Physics and Hydrology Division (2023).
5. Secretary (2019) and then Chair (2020) for the multi-state project W4188.
6. Member of the NEON Soil Sensor Technical Working Group (2018-present).
7. Peer review: 100+ Manuscript Reviews in 40+ journals; Grant panel member for NSF and USDA programs; Ad-hoc reviewer for NSF, USGS, and FFAR proposals; Associate editor for Soil Science Society of America Journal (2020-present).

PROFESSIONAL AFFILIATIONS

| | |
|--|----------------|
| American Geophysical Union | 2010 – present |
| Soil Science Society of America | 2012 – present |
| American Society of Agronomy | 2016 – present |
| American Association for the Advancement of Science (AAAS) | 2017 – present |
| International Soil Tillage Research Organization | 2019 – present |
| European Geophysical Union | 2020 – present |
| International Soil Modeling Consortium | 2020 – present |

FUNDED RESEARCH

Highlights:

- As PI, secured funding from major federal programs including the USDA Agriculture and Food Research Initiative (AFRI) and the USDA National Resources Conservation Service (NRCS).
- PI or Co-PI on 40+ externally funded grants totaling more than \$9.2 million (\$2.7 million to Stewart research program).

Selected Projects:

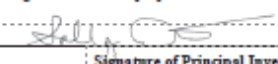
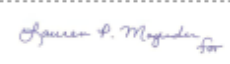
- Stewart, R. D., W. L. Daniels, D. Sample, J. Ignosh, V. Temu, and M. Kering. Determining Impacts of Utility-Scale Solar on Stormwater Runoff and Soil and Water Quality and Providing Design Criteria. Virginia Department of Environmental Quality. 2023-2029. \$3,339,282.
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Attachment A

Stream Research Initiative Program SOLICITATION OFFER AND AWARD

Attachment A

Stream Research Initiative Program
SOLICITATION OFFER AND AWARD

| | | | |
|--|--|---|--|
| Resource Protection Group, Inc. c/o Justin Curtis, Esq. AquaLaw, P.C. 6 South 5 th Street, Richmond, VA 23219 | | 1. FOR INFORMATION CONTACT Name: Justin Curtis, Esq. Phone: 804 716 9021 x204 E-mail: justin@aqualaw.com | |
| 2. SOLICITATION NUMBER Stream RFP #07 – Supplemental Water Quality Sampling for Combined Reston Mussel Stocking x Stream Monitoring Program | | 3. TYPE OF SOLICITATION REQUEST FOR PROPOSALS (RFP) Resource Protection Group – RFP #07 | |
| | | 4. DATE ISSUED January 11, 2024 | |
| SOLICITATION | | | |
| 5. DESCRIPTION OF WORK The Proposer shall furnish all necessary staff, materials, tools, equipment, and supervision to provide the research program and deliverables as described in the referenced RFP and Proposer's Response. We will quantify biogeochemical effects of reintroducing native unionid mussels into two restored urban headwater streams in Reston, Virginia - Snakeden Branch and The Glade. We have scientists with expertise in mussel establishment and population dynamics, biogeochemistry and ecology as experts for each of the key components of the stated needs of Resource Protection Group, Inc. Our study will provide a year and a half of bimonthly sampling at baseflow (June 2024-December 2025) during the ongoing mussel reintroduction as a way to provide a more detailed analysis of how mussel beds in the restoration streams are altering nutrient processing, water quality, and biological communities. We are already measuring and analyzing individual mussel biogeochemistry, and here we proposed to measure water and sediment nutrients in the established mussel beds, and at the upstream control stream reach in both streams to scale the mussel nitrogen and phosphorus nutrient cycling effect. | | | |
| 7. PROPOSED BUDGET \$392,529 | | | |
| OFFER (Offeror must complete in its entirety) | | | |
| 8. PRINCIPAL INVESTIGATOR Name and Title: Sally Entekin, Associate Professor Organization: Virginia Tech Entomology Department Mailing Address: 190 Drillfield Drive, Blacksburg, VA 24061 Telephone: 501-269-2108 Fax: _____ E-mail: sallye@vt.edu PI Assurance: I agree to accept responsibility for the scientific conduct of the project, to provide the required reports, to acknowledge Resource Protection Group, Inc. in any presentations and publications wherein the results of this project are used, and to provide copies of presentation abstracts and publications to RPG. I also agree to allow this proposal to be reviewed by industry and/or academia and that there is no proprietary information in this proposal. Date: 4/11/2024 Signature of Principal Investigator:  | | | |
| 9. PROPOSER'S ORGANIZATION Name: Virginia Polytechnic Institute and State University Address: 300 Turner Street NW, STE 4200, Blacksburg, VA 24061 Federal Tax ID Number: 54-600180 Certifying Representative: James E. Heflin, Co-Interim Executive Director of Sponsored Programs Name and Title: _____ Certification and Acceptance: I certify that to the best of my knowledge, the statements contained herein are complete and true and I accept the obligation to comply with RPG terms and conditions provided an award is made as a result of this submission. Signature of Organization's Certifying Representative:  for Date: 04/12/2024 | | | |
| AWARD (To be completed by Review Entities) | | | |
| 10. APPROVAL AMOUNT | | 11. DATE OF AWARD | |
| 12. APPROVED RESEARCH START DATE | | 13. APPROVED RESEARCH COMPLETION DATE | |
| 14. RECOMMENDATION BY RESOURCE PROTECTION GROUP, INC. Justin Curtis, Esq., Member, Board of Directors Date: _____ <input type="checkbox"/> Approved < Rejected <input type="checkbox"/> Proposed with Conditions: | | | |

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COLLEGE OF AGRICULTURE AND LIFE SCIENCES
SCHOOL OF PLANT AND
ENVIRONMENTAL SCIENCES
VIRGINIA TECH.

DRAFT Quality Assurance Project Plan

Prepared for:

**Virginia Department of Environmental Quality
and will be modified for this proposal**

By:

Ryan D. Stewart

**Virginia Tech, School of Plant & Environmental Sciences
185 Ag Quad Lane, 0404, Virginia Tech
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David J. Sample

**Virginia Tech, Biological Systems Engineering Dept.
Hampton Roads Agricultural Research and Extension Center
1444 Diamond Springs Rd
Virginia Beach, VA 23455
dsample@vt.edu**

April 15, 2024

Project Organization and Schedule

Key project personnel responsible for the monitoring program are listed in Table 1.

Table 1. Key project personnel.

| Title | Name (Affiliation) | Phone Number/E-mail |
|------------------------|---------------------------|---|
| Principal Investigator | Ryan Stewart | (540) 231-0253; rds@vt.edu |
| Lab Manager | Athena Tilley | (540) 231-9795; athena@vt.edu |
| Research Scientist | John Hoben | (540) 231-4490; jph@vt.edu |
| | | |
| | | |
| | | |
| | | |

Quality Control

Measurement Performance Criteria

The overall QA objective for this project is to develop and implement procedures for field sampling, COC, and reporting that will provide scientifically defensible results. Specific procedures for sampling, COC, reporting of data, internal QC, audits, preventive maintenance of field equipment, and corrective action are described in the other sections of this QAPP. Overall data quality objectives are provided in Table 4. These limits are consistent with the U.S. Environmental Protection Agency (USEPA) water quality criteria (USEPA, 1992).

Table 2. Quality Assurance Objectives for Individual Measurements.

| Parameter | Method ¹ | Detection Limit | Precision | Accuracy | Completeness |
|------------------------|--|-----------------|-----------|----------|--------------|
| Total Phosphorus | 4500-P H | 0.02 mg/L as P | 90% | 90% | 80% |
| Dissolved Phosphorus | 4500-P H (after filtering with 0.45 µm filter) | 0.02 mg/L as P | 90% | 90% | 80% |
| Ortho Phosphorus | 4500-P G (after filtering with 0.45 µm filter) | 0.02 mg/L as P | 90% | 90% | 80% |
| Total Nitrogen | 4500-N C | 0.1 mg/L as N | 90% | 90% | 80% |
| Nitrate/ Nitrite | 4500-NO ₃ -I 4500-NO ₂ -B (after filtering with 0.7 µm filter) | 0.05 mg/L as N | 90% | 90% | 80% |
| Ammonia | 4500-NH ₃ H (after filtering with 0.7 µm filter) | 0.05 mg/L as N | 90% | 90% | 80% |
| Total Suspended Solids | 2540 D (after filtering with 0.7 µm filter) | 0.01 g | 90% | 90% | 80% |

¹Refers to American Public Health Association *et al.* (2017).

Precision

Field and laboratory precision will be assessed through the collection and measurement of field duplicates at a rate of one duplicate per 10 analytical samples.

Precision will be assessed through the calculation of the relative percent difference (RPD) for two replicate samples and relative standard deviation (RSD) for three or more replicate samples. RPD is the absolute difference between two results expressed as a percentage of the average result. It is calculated according to the following formula:

$$\frac{S-D}{0.5 \cdot (S+D)} \cdot 100\% \quad (2)$$

where:

S = Original sample value;

D = Duplicate sample value.

The acceptance criteria for RPD will be less than or equal to 20%. Percent RSD is calculated according to the following formula:

$$\frac{s}{\bar{X}} \cdot 100\% \quad (3)$$

where:

s = standard deviation

\bar{X} = mean

1.0 Accuracy

Accuracy of the sample collection and analysis procedures ensures that samples are not affected by sources external to the sample such as sample contamination by ambient conditions. Sampling and analysis accuracy will be assessed by the data from equipment and trip blank samples.

Trip blank samples will provide a measure of potential cross-contamination of samples by volatile organic compounds during shipment and handling.

Trip blank samples should not contain target analytes. Accuracy also will be ensured by adhering to all sample handling procedures, sample preservation requirements, and holding time periods.

2.0 Field Quality Control

Field quality control (QC) samples are used to assess the influence of sampling procedures and equipment used in sampling. They are also used to characterize matrix heterogeneity. For basic water quality analyses, quality control samples to be prepared in the field will consist of equipment blanks, field duplicates, and matrix spikes (when applicable).

Internal QC is achieved by collecting and analyzing a series of duplicate, blank, spike, and spike duplicate samples to ensure that analytical results are within the specified QC objectives. The QC sample results are used to quantify precision and accuracy and identify any problem or limitation in the associated sample results. The internal QC components of a sampling and analyses program will ensure that the data of known quality are produced and documented. The internal QC samples, frequency, acceptance criteria, and corrective action must meet the minimum requirements presented in the following sections.

3.0 Sample Identification

All samples will be identified with a unique number and samples labeled with the following information. See Section 1.2 for terminology.

- Site ID
- Subcatchment ID
- Sampling Point ID (if applicable)
- Date
- Time
- Initials of the sample collector
- Sample type (normal or QC)

4.0 Chain of Custody Forms

All sampling events will employ chain of custody (COC) forms that will record the date and time of sampling, parameters, site and location designations, sampling and transport entity and dates, and receipt entity and processing/preparation applied in the laboratory. A sample COC form is

found in Appendix A. Hard and electronic copies of COC forms will be retained by the lab and attached to all reports.

5.0 Field and Equipment Blanks

At least two field blank samples will be submitted along with storm event water samples for each site each year. The first blank will occur with the initial designated research sampling trip and the second blank will occur after at least three subsequent sampling events in a given year. The blank samples will be comprised of laboratory deionized (DI) water that is transported to the site in a separate decontaminated container and transported to the lab with the same preservation and hold requirements as the research event samples.

6.0 Equipment Blanks

Equipment blanks will be collected and analyzed for all analytes of interest along with the associated environmental samples. Equipment blanks will consist of laboratory-prepared blank water (certified contaminate-free) processed through the sampling equipment using the same procedures used for environmental samples.

7.0 Field Duplicates

Field duplicates will be collected at the rate of 1 per 20 normal samples, or 1 per sampling event, whichever is greater. Field duplicates will be collected at the same time as environmental samples or of two grab samples collected in rapid succession, and will be analyzed along with the associated environmental samples. If the relative percent difference (RPD) of field duplicate results is greater than 25% and the absolute difference is greater than the reporting limit (RL), both samples should be reanalyzed.

8.0 Field Instrument Calibration

Routine field instrument calibration will be performed before instrument use to ensure instruments are operating properly and producing accurate and reliable data. Calibration will be rechecked at a frequency recommended by the manufacturer. Water level sensors will be checked for drift using baseline depth readings through time.

9.0 Laboratory Quality Control

10.0 Laboratory Blanks

11.0 Laboratory Duplicates

12.0 Matrix Spikes

[TO PROVIDE]

13.0 Data Management Procedures

14.0 Documentation and Records

All records generated by this project will be stored on a shared cloud-based drive (e.g., Microsoft Sharepoint). Copies of this QAPP will be distributed to all parties involved with the project, including signatories and field sampling personnel. Any future changes or amendments to the QAPP will be held and distributed in the same fashion. Copies of previous versions of the QAPP will be marked as “superseded by Revision #” so as not to create confusion.

The records of all project information and data used to complete the activities of the project will be retained for at least seven years from the date of sampling, measurement, report, or application.

Copies of field logs, copies of COC forms, original preliminary and final reports, and electronic media reports will be kept for review by Virginia Tech. The field crew will retain the original field logs.

Field data sheets are checked and signed in the field by whoever is performing the sampling. They will identify any results where holding times have been exceeded, sample identification information is incorrect, samples were inappropriately handled, or calibration information is missing or inadequate. Such data will be marked as unacceptable and will not be entered into the electronic database and/or otherwise used for project analysis, reporting, or other purpose.

Concentrations of chemicals and toxicity endpoints, and all numerical biological parameters will be calculated as described in the referenced method document for each analyte or parameter. The data generated will be converted to a standard database format maintained by the responsible party and available for NFWF staff review when requested. This review is for QA

purposes only and will not be used for any other purpose. All project information will remain confidential.

15.0 Data Quality Assessment

Data must be consistently assessed and documented to determine whether project QA objectives have been met, quantitatively assess data quality, and identify potential limitations on data use. Assessment and compliance with quality control procedures will be undertaken during the data collection phase of the project.

16.0 Field & Laboratory Analytical Procedures

All water and soil samples will be analyzed by the Virginia Tech Central Soil & Water Quality Laboratory (CSWQL). The Virginia Tech Extension Soil Testing Laboratory will also be used for plant-available nutrients and metals. A subset of samples will be analyzed at a VELAP-certified independent lab (Enthalpy; Richmond, VA), as discussed in Section 5.2. All laboratories will provide results and quality control data in standardized laboratory reports with appropriate standard data qualifiers if/as needed.

The CSWQL is a university-authorized Service Center that offers a range of analyses to both internal Virginia Tech research programs and external private-sector clients. The SWQL has functioned as an audited cost recovery center for over 15 years. Insofar as possible within an active academic research environment, the SWQL conforms with VDEQ (2020, 2022) field sampling and lab analytical protocols and follows a wide range of rigorous internal Quality Control (QC) and QA measures including:

- Use of established and approved Standard Operating Procedures (SOPs) for all field and laboratory procedures.
- Strict adherence to field sampling, preservation, and hold time protocols and limits.
- Utilization of COC forms for all water and soil samples.
- Consistent labeling with site and sample codes, date, time, & collector noted.
- Inclusion of periodic field trips/equipment blanks.
- Collection and analysis of field split samples (minimum one per 25 stations/events).
- Minimum analytical accuracy and precision of $\pm 10\%$.

- At least daily calibration of all analytical instruments with full range standards.
- Laboratory duplicate runs every 20 samples or fewer.
- Inclusion of laboratory blanks in each sample event run.
- Regular determination of reporting level (RL) and quantification level (QL) for all analytes.
- Inclusion of known concentration standards (or matrix spikes) within each sample x analyte run prepared in a similar background matrix.
- Daily review of all laboratory data by a lab manager and overall data QA/QC flow path review as set forth by (VDEQ, 2022).
- Daily backups of all lab data and fully secured data storage systems.
- Periodic review of field & lab data and procedures by the relevant Principal Investigator.

Table 6 provides a summary of the number of samples, taking into consideration the 42 stations, 24 events per year, field and lab duplicates and blanks, etc.

17.0 Water Quality Parameters

A list of the water quality parameters to be assessed in the laboratory, analytical methods, and their respective maximum hold times is provided in Table 5. Water samples will be immediately chilled in the field, transported on ice, and then preserved in a lab refrigerator at a temperature of 4°C until they are analyzed.

The analytical limits and required hold times in Table 5 are consistent with USEPA (1992; 2015) water quality sampling guidance and/or with methods cited by VDEQ (2022). For this research program, the Reporting Level (RL) is the lowest level that the SWQL (or an external lab) can reliably and consistently quantify for a given parameter. This value is frequently also listed as the Quantification Level (QL). In contrast, the MDL (Method Detection Limit) is the minimum concentration of an analyte that can be measured and reported with 99% confidence that the analyte concentration is greater than zero. Furthermore, the MDLs listed in Table 5 are those cited by VDEQ (2022) which are considerably lower in many cases than those currently cited by the USGS National Water Quality Laboratory (NWQL) and other VELAP labs that we commonly cooperate with.

18.0 Sample Storage and Preservation

Storm samples will be transported to the laboratory as soon as possible after collection due to holding time considerations. The sample hold time is the allowable length of time between sample collection and laboratory manipulation. Sample preservation and maximum hold times are provided in Table 5. Water samples will be preserved in a refrigerator at a temperature below 6°C (42.8°F) before they are analyzed. For a parameter of interest, the analysis sample will be gently shaken to be uniformly mixed and then subdivided into three sub-samples of equal volume. The three sub-samples will be analyzed separately, and the three measured values will be arithmetically averaged and taken as the value of this parameter. All containers will be flushed at least three times using the sub-samples before they are used.

19.0 Sample Storage, Preservation, and Holding Times

Storm event samples will be chilled upon collection in a refrigerated sampler preserved per Table 5 and transported to the laboratory as soon as practical after collection. The **actual field hold time** is defined as the amount of time lapsed from the end of the storm until the sample is filtered and preserved in accordance with Table 5. Appropriate acid preservation requirements will be met by adding the acid to sample bottles before the event whenever possible.

The **required sample hold time** is the allowable length of time between sample collection and laboratory analyses. Required sample hold times are unique for each analyte (Table 5). If the sample hold times cannot be met, the sample will be discarded. Samples for ortho-P will be filtered with a disposable 0.45 µm glass fiber filter (GFF) in the field before transport.

For a parameter of interest, the analysis sample will be gently shaken to be uniformly mixed with the aforementioned churn splitter and then subdivided into three sub-samples of equal volume. The three sub-samples will be analyzed separately, and the three measured values will be arithmetically averaged and taken as the value of this parameter. All containers will be flushed at least three times using the sub-samples before they are used.

Table 3. Laboratory water quality analytical parameters, methods, preservation, hold times, and reporting limits (RL).

| Parameter/Analyte* | Method | Preservation | Hold Time** | Analysis Sample, mL | Container | Reporting Limit*** (units) | Method Detection Limit, mg/L*** |
|-------------------------------------|-------------------------------|---|-------------|---------------------|-----------|----------------------------|---------------------------------|
| Acidity (pH) | Standard meter | Chill to 4° C | 48 hr | | | 0.01 SU | NA |
| Specific Conductance (SC) | EPA 120.1 Conductivity bridge | Chill to 6° C | 28 d | | | 5 µs/cm | NA |
| Particle Size Distribution (PSD) | | Cool to ≤6°C, analyze ASAP, and if necessary, preserve by freezing. | 7 d | 1000 | PG | NA | NA |
| Total Suspended Solids (TSS) | USGS I-3765-85 | Cool to ≤6°C in the field for transport, analyze ASAP, reserve by freezing Chill to 4° C | 7 d | 1000 | P | 1.0 | 1.0 |
| NH ₃ +NH ₄ -N | EPA 350.1 Rev 2.0 | Filter immediately (dissolved), Analyze ASAP. Cool to ≤6°C in the field for transport, then preserve by freezing if not immediately analyzed Chill to 4° C H ₂ SO ₄ to pH < 2 | 28 d | 500 | PG | 0.05 | 0.003 |

| Parameter/Analyte* | Method | Preservation | Hold Time** | Analysis Sample, mL | Container | Reporting Limit*** (units) | Method Detection Limit, mg/L*** |
|--------------------------------------|------------------------------|--|-------------|---------------------|-----------|----------------------------|---------------------------------|
| Total Kjeldahl Nitrogen (TKN) | | Filter immediately (dissolved), Analyze ASAP. Cool to $\leq 6^{\circ}\text{C}$ in the field for transport, then preserve by freezing if not immediately analyzed | 28 d | 500 | PG | | |
| $\text{NO}_2 + \text{NO}_3\text{-N}$ | EPA 353.2 | Filter immediately (dissolved), Analyze ASAP. Cool to $\leq 6^{\circ}\text{C}$ in the field for transport, then preserve by freezing if not immediately analyzed Chill to 4°C H_2SO_4 to $\text{pH} < 2$ | 28 d | 200 | PG | 0.05 | 0.002 |
| Total-N | SM 4500-N-C or ASTM D8083-16 | Filter immediately (dissolved), Analyze ASAP. Cool to $\leq 6^{\circ}\text{C}$ in the field for transport, then preserve by freezing if not immediately analyzed Chill to 4°C H_2SO_4 to $\text{pH} < 2$ | 28 d | 200 | PG | 0.10 | 0.03 |

| Parameter/Analyte* | Method | Preservation | Hold Time** | Analysis Sample, mL | Container | Reporting Limit*** (units) | Method Detection Limit, mg/L*** |
|--------------------|-----------|---|-------------|---------------------|-----------|----------------------------|---------------------------------|
| Total-P | EPA 365.4 | Filter immediately (dissolved), Analyze ASAP. Cool to ≤6°C in the field for transport, then preserve by freezing Chill to 4° C H ₂ SO ₄ to pH < 2 | 28 d | 100 | PG | 0.02 | 0.005 |
| Ortho-P | EPA 365.1 | Filter immediately (dissolved), Analyze ASAP. Cool to ≤6°C in the field for transport, then preserve by freezing Chill to 4° C | 48 hr | 100 | PG | 0.02 | 0.002 |

PG: polyethylene glass; P: polyethylene.

The detection limits are in nitrogen and phosphorus elements, respectively.

*Minimum research data set; other parameters may be added as indicated by field observations.

**Maximum hold time allowable per USEPA/ASTM/AWWA Standard Methods guidance; See Section 5 for a description of laboratory method & calibration study.

*** The Reporting Limit (RL) is the minimum level that a given parameter can be quantified at; also referred to as the QL. The RL is periodically reconfirmed by the laboratory via serial dilutions of the analyte against the same background matrix. The RLs reported here are consistent with documentation provided by the VT SWQL, the USGS NWQL, and several Virginia VELAP labs. The RL is necessarily higher than the Method Detection Limit (MDL) which is the lowest level that a given analyte can be detected as being present (yes/no) in a given matrix with 99% confidence. The MDLs listed here are per VDEQ (2022) and their DCLS catalog.

20.0 Inter-lab Water Quality Methods and Results Comparison (For VT x DEQ Solar Study)

During the first six months of water quality monitoring and sampling, we will select a minimum subset of 30 discrete samples that will be split in the field into two separate sub-samples:

- One sample will be transported to a VELAP-certified laboratory, Enthalpy (Richmond, VA), utilizing their containers, COC forms, etc. This sample will be analyzed via standard methods and hold times as outlined in Table 5.
- A second split sample is to be transported to the VT CSWQL. Once at VT, this sample will be further split into (1) a primary sample that undergoes the standard analyses as outlined in Table 5 and (2) a paired sample that will be immediately frozen and reanalyzed for all parameters again after 45 days. The VT sample splits will also include an additional field SC sample that will be filtered in the laboratory upon receipt rather than immediately in the field. Once the full data sets are available and QA confirmed, we will perform the following statistical analyses on the comparative data sets:
 - Comparison of each analyte x date between the two labs (Enthalpy & VT SWQL) for consistency and agreement.
 - Direct comparison via paired contrasts (or regression) for total-N via SM 4500-N-C (Enthalpy) vs. ASTM D8083-16 (VT CSWQL).
 - Direct comparison of total-P via EPA 365.4 (both labs) vs. VT CSWQL alternative method of direct determination via ICPES combustion.
 - Comparison of ortho-P determinations for field vs. lab filtration.
 - Effect of lab freezing and extended hold time on all parameters.

Following these analyses, we will determine whether adjustments or modifications to the methods and hold times shown in Table 3 are justified for utilization over the remaining field sampling and lab analytical program or if alternative lab protocols are more time and resource-efficient. Any such adjustments will be submitted for approval by VDEQ.

21.0 Laboratory Soil Analyses (Not applicable to M)

All soil analyses will be conducted by the Virginia Tech SWQL and follow methods by SSSA (2022), Maguire and Heckendorn (2019) or NRCS (2022).

22.0 Physical Properties

- Rock fragment content (> 2 mm) via mechanical screening

- Bulk density – oven dry gravimetric with rock fragment correction when needed
- Particle size analysis (≤ 2 mm) for silt + clay fractions via pipette method or laser diffraction

23.0 Chemical Properties

- Total C&N – via automated combustion analysis
- Acidity (pH) – saturated paste or 1:1 soil: water extract
- Specific conductance (SC) – saturated paste extract
- Plant-available Ca, K, Mg, and P – Mehlich I extraction x ICPES
- Plant-available B, Cu, Fe, Mn, and Zn – Mehlich I extraction x ICPES

Table 4. Quality assurance requirements and anticipated number of water samples for each parameter.

| Parameter/Analyte* | Samples per Station | Number of Stations per Site | Total number of Samples per Site | Laboratory Method Blanks | Field Equipment Rinsate Blanks | Laboratory Control Standard | Matrix Spike | Field Duplicates | Laboratory Duplicates |
|-------------------------------------|---------------------|-----------------------------|----------------------------------|--------------------------|--------------------------------|-----------------------------|------------------------|------------------|-----------------------|
| Total Suspended Solids | 20 | 7 | 140 | 1/batch ¹ | 21 ² | 1/batch ¹ | 1/batch ¹ | 14 ³ | 1/batch ¹ |
| NH ₃ +NH ₄ -N | 20 | 7 | 140 | 1/batch ¹ | 21 ² | 1/batch ¹ | 1/batch ¹ | 14 ³ | 1/batch ¹ |
| NO ₂ +NO ₃ -N | 20 | 7 | 140 | 1/batch ¹ | 21 ² | 1/batch ¹ | 1/batch ^{1,4} | 14 ³ | 1/batch ¹ |
| Total-N | 20 | 7 | 140 | 1/batch ¹ | 21 ² | 1/batch ¹ | 1/batch ¹ | 14 ³ | 1/batch ¹ |
| pH | 20 | 7 | 140 | NA | NA | NA | NA | 14 ³ | 1/batch ¹ |
| Total-P | 20 | 7 | 140 | 1/batch ¹ | 21 ² | 1/batch ¹ | 1/batch ¹ | 14 ³ | 1/batch ¹ |
| Ortho-P | 20 | 7 | 140 | 1/batch ¹ | 21 ² | 1/batch ¹ | 1/batch ¹ | 14 ³ | 1/batch ¹ |
| Particle Size Distribution | 20 | 7 | 140 | 1/batch ¹ | 21 ² | 1/batch ¹ | 1/batch ¹ | 14 ³ | 1/batch ¹ |

¹ A laboratory QA batch will consist of no more than 20 samples.

² Rinsate will be sampled 1) from rinse water taken before first event, 2) after first sample is collected, and 3) after the last sample is collected for a total of 3 per monitoring station.

³ Field duplicates will be collected for at least 10% of samples submitted for analysis.

⁴ A matrix spike or spike duplicate will be run for each batch.

24.0 References

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