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Robert W. Yost  
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Wind Harp June 2023  
In the hills overlooking Vettnik, Slovenia

**Photographer:**  
Robert Yost  
Teaching Professor  
Department of Biology  
Indiana University-Purdue-University  
Indianapolis

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Khadijak Makky, Marquette University, Program chair

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# ***Bioscene: Journal of College Biology Teaching***

## **Volume 49(2) December 2023**

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### **Bioscene Editors**

**Robert W. Yost**, Editor-In-Chief,  
Department of Biology  
Indiana University – Purdue University Indianapolis  
723 W. Michigan St., Indianapolis, IN 46202  
Telephone: 317-278-1147  
FAX: 317-274-2846  
Email: ryost@iupui.edu

**Ashley Driver**, Associate Editor  
Department of Biology, LC375  
University of Scranton  
800 Linden St. Scranton, PA 18510

**Denise Slayback-Barry**, Associate Editor  
Department of Biology  
Indiana University – Purdue University Indianapolis  
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## Articles

### Microplastics Are All Around Us: An Engaging Activity for Undergraduate Biology Majors

Shem D. Unger\*, Mark Rollins

Biology Department, Wingate University, Wingate, NC 28174

\*corresponding author: s.unger@wingate.edu

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#### Abstract

When teaching undergraduate biology courses, developing engaging activities which allow students to learn using a hands-on pedagogy can provide impactful learning moments for students. This activity helps students make connections between lecture content on environmental issues, e.g., the use of microplastics (MPs) and their own day-to-day activities by learning whether or not the products they use regularly contain microplastics or skeptical microplastics. Using a smartphone app (Beat the MicroBead) and personal care products, students quantify the type and number of potential microplastics and complete a post-activity survey. Overall, students found the activity engaging, fun, and informative. Students also report on not realizing the products they frequently use may contain plastics, with the majority finding various forms of microplastics or skeptical microplastics. Students identified Dimethicone, Carbomer, and Acrylates Copolymer as the most abundant MPs, and Cyclopentasiloxane, Polysorbate 20, and Cetareth-20, as the most abundant skeptical MPs present in their personal care and cosmetic products. Students found on average at least 4/10 and 5/10 products containing MPs or skeptical MPs, respectively, with most students responding they had not heard of MPs before this activity. This activity can be incorporated into any college biology or environmental biology course.

**Keywords:** teaching, pedagogy, undergraduate biology, plastic pollution, microbeads, conservation

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#### Introduction

Plastic pollution of aquatic environments, both freshwater streams and marine habitats, is an emerging environmental issue (Conkle et al., 2018). This issue is among the world's major threats to ecosystems given the long-term persistence of many plastic products in the environment (Geyer et al., 2017). Microbeads, which have previously been recognized as a problem as their wide-stream use in cosmetics and other products were largely banned in the US with the Microbead-Free Waters Act of 2015 (Strifling, 2016). However, microplastics (MPs), plastic particles under 5 mm in size, are still found as ingredients in a variety of personal care and cosmetic products (Guerranti et al., 2019), including toothpaste, deodorant, soap, lipstick, moisturizers, etc. (Bhattacharya, 2016; Ustabasi & Baysal, 2019). Moreover, these personal care and cosmetic products (PCCPs), contain potentially harmful microplastics and are widely

available for purchase, and are also present in the environment (Bashir et al., 2021; Zhou et al., 2023). These PCCPs containing MPs can be a source for persistent, potentially harmful plastic materials released into aquatic environments (Fendall & Sewell, 2009), due to being hydrophobic, dense, and small, these MPs from PCCPs have the potential to only be partially captured in wastewater treatment processing plants, thus potentially entering the aquatic ecosystem as effluents (Habib et al., 2022). Therefore, educating the public on the risk of MP-containing products entering aquatic ecosystems is of the utmost importance (Bhattacharya, 2016), with science students as an ideal cohort for environmental education.

Methods for teaching students about microplastic pollution in the environment include the use of board games (Adjiningsih & Sriwattanarothai, 2022), websites (Reed & Chen, 2022), or active learning approaches in outreach

education to teach the chemical structures and how plastics break down in the ocean (Schiffer et al., 2020). Additional approaches include developing learning modules emphasizing sources of MP and linking these to the environment and potential purchasing decisions (Raab & Bogner, 2020). Subsequently, connecting consumer information about microplastics may be linked to a willingness by individuals to adopt pro-environmental behavior (Garcia-Vazquez & Garcia-Ael, 2021). Incorporating a method that increases awareness about this issue while making the connection between students and their role as a consumer of products is therefore needed in an undergraduate course for promoting future citizen scientists. Likewise, developing investigatory, engaging, and effective inquiry opportunities for students to collect data related to the presence of MPs in PCCPs is vital for furthering the understanding of MPs and their potential environmental impact as part of an undergraduate biology curriculum.

The activity described below was incorporated into a first-year organismal biology course for biology majors (Fall 2022 and Spring 2023) and was assigned online via the learning management system, Canvas. This activity was an opportunity for students to make connections to lecture content on environmental issues by collecting data on potential plastic ingredients in PCCPs, followed by a short survey, all submitted online, and then a brief in-class reflection. Therefore, this activity can easily be modified, uses a free smartphone application, and requires minimal time, yet based on student responses is an engaging and effective method for teaching about microplastics.

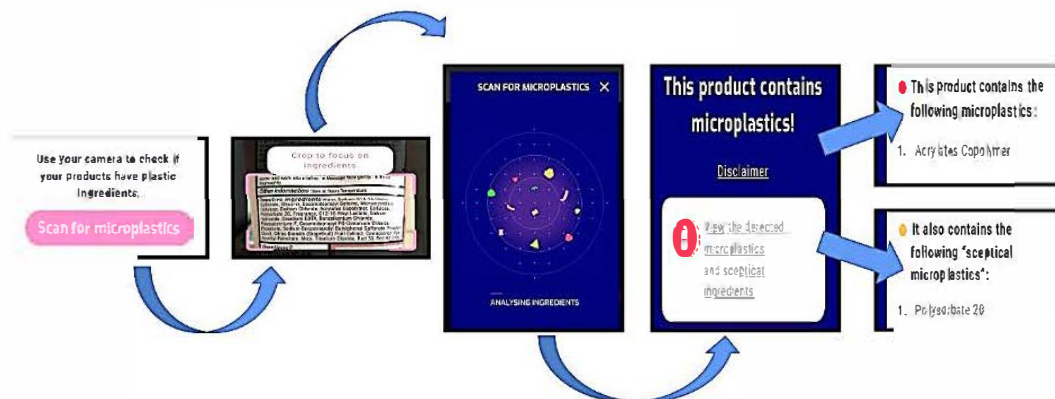
### **Procedure**

Students are first required to read a short article on microplastics, their ubiquity in the environment, and their potential impacts on wildlife (Lim, 2021). The article was selected for students to read prior to this activity because it summarizes the environmental problem of MPs and presents findings of current research into microplastics. Students then continued the

microplastic activity where they follow instructions and download an application on their smartphone (available for free on Apple App or Google Play Stores) and begin scanning home care products for the presence of microplastics of skeptical microplastics (Figure 1). The application was developed by a Dutch environmental group called Plastic Soup Foundation and allows students to scan personal care products using their smartphone. Alternative for students who do not use a smartphone could involve looking up product information on the website associated with the application using the "check your products" search. Student participants then completed a brief survey on the activity. For the application, Beat the Microbead ([www.beatthemicrobead.org](http://www.beatthemicrobead.org)), students were instructed to use the app to scan the ingredients list from a variety of products that are likely to include microplastics such as conditioner, shampoo, soap, exfoliants, shaving gel, deodorant, hand sanitizer, toothpaste, aftershave, facial cleanser, sunscreen, and any cosmetics including mascara, lipstick, foundation, etc. Students were further instructed to contact the instructor for assistance if they ran into any technical issues with the application and informed that they could delete the application following the activity.

The app students utilized for this take-home activity scans for more than 500 potential microplastics, and many brands in the database often contain some form of microplastics (Diewertje et al., 2022). Students then filled out a table and reported how often they used the product (daily, weekly, etc.) and one of three options for the product scanned, either "no microplastics found", or if microplastics were found they were labeled as either "microplastic" (synthetic polymers commonly considered as microplastics including polyethylene, polypropylene, nylon, acrylates copolymer, etc.) or "skeptical microplastic" (synthetic polymers for which there is not enough information available). Students were further instructed to report on the chemical name of the specific item found in each product scanned and continue

**Figure 1.** Workflow Diagram of application used for identifying potential microplastics and skeptical microplastics.



until they successfully scanned a total of 10 PCCPs for MPs.

Lastly the activity was assessed by way of an online survey containing six questions. See Tables 1 and 2 for details. Questions 1, 2, and 5 required simple “yes/no” responses. A Likert Scale accompanied questions 3 and 4. Finally, question 6 invited additional comments from participants. Prior to analysis of responses, names were removed from the surveys to maintain confidentiality. Standard statistical analysis including primarily descriptive statistics (mean, standard deviation, and median) was used to report student data submitted for this activity. A chi-squared analysis was performed for Likert questions (Q3, Q4) to examine the hypothesis that observed responses would differ from expected responses (no pattern) using 0.05 as our significant level.

#### Assessment

In total, there were 102 student participants (biology majors) enrolled in a first-year organismal biology course across Fall 2022 and Spring 2023 semesters at Wingate University, a small liberal arts university outside Charlotte, North Carolina. The majority of students (59.8 %) responded to the survey question Q1 that they had not heard of MP before completing this activity (Table 1). The majority of students (77.5%) responded that they expected to find

microplastics in their PCCPs (Q2), likely after reading the article prior to completing the activity. We found significance for Q3 responses,  $X^2(1, N = 102) = 26.51, p < 0.001$  (Mean Likert response =  $4.55 \pm 0.623$  S.D.), and also for Q4,  $X^2(1, N = 102) = 62.97, p < 0.0001$  (Mean Likert response =  $4.68 \pm 0.645$  S.D.), indicating that this activity likely helped students understand MPs in PCCPs and that the application was both engaging and easy for students to use. Responses for Q5 indicate 96.1% of student participants feel more informed about MPs after completing this activity.

Comments from student participants in Table 2 and below were selected which represent overall feedback. Responses to Q6 (additional comments), were overwhelmingly positive, indicating that students enjoyed the activity, found it engaging, and appreciated the hands-on approach to learning about MPs. Representative comments (Table 2), that illustrate effectiveness of this activity were varied. Additional comments by student participants in addition to those already listed in Table 2 included “ I realized when doing this activity that a large amount of the personal care products I use or see every day contain microplastics”, “I felt as though this assignment was timely, and fun and I enjoyed it. I felt engaged the whole time working on it.”, “It was definitely eye-opening which products have microplastics and which ones don’t”, “It was

**Table 1.** Representative responses to survey questions Q1-5, (N = 102).

Survey Question	Survey Results
Q1: "Have you Heard of microplastics before?"	40.2% YES: 59.8% NO
Q2: "Did you expect to find microplastics in your personal care products?"	77.5% YES: 22.5% NO
Q3: "This activity helped me understand how common microplastics are in everyday personal care products."	Mean = 4.55 ± 0.623 S.D. Median = 5
Q4: "Using the Beat the Microbead App was a straightforward, engaging way to learn about microplastics."	Mean = 4.68 ± 0.645 S.D. Median = 5
Q5: "After completing this activity, do you feel more informed regarding the personal care products you use that potentially contain microplastics?"	96.1% YES: 3.9% NO

shocking to me that even products like hand soap that we wash down into the water cycle every day contain microplastics. This activity showed that we have to invest time into looking at the ingredients of the products that we use every day and that we put on our skins."

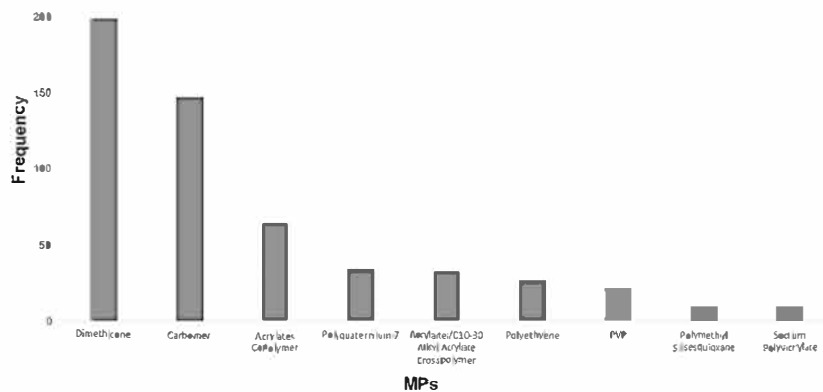
Additional comments that included a different perspective on this utility of this smartphone activity included "Even though I've seen the different microplastics in my personal care products I use daily, I may still use them" and also "It would be beneficial to use website more" and "not really enjoying this activity", which indicate some students either were likely to continue to use the product, had a preference for use of website, or in one participant's case, didn't enjoy the activity. Therefore, while most comments by student participants were positive, this activity could be improved by possibly introducing more of the features of this application in class or ensuring students didn't run into potential issues using and understanding smartphone application and selected appropriate personal care products.

Students identified a total of 42 MPs and 135 skeptical MPs in their personal care products as part of this activity, with a mean of  $4.07 \pm 1.67$  S.D.,  $5.37 \pm 1.82$  S.D., and  $3.30 \pm 1.79$  S.D., for how many products out of ten either contained MP, skeptical MP, or did not contain any MP, respectively. PCCPs scanned by students when analyzed for the specific type of MP or skeptical MP, on average, contained a total of  $4.02 \pm 1.95$  S.D. unique MPs and  $8.94 \pm 4.35$  S.D. unique skeptical MPs, respectively. The maximum number of unique MPs and unique skeptical MPs were 11 and 21 across ten products scanned with the application. Among the most common MP identified in students' PCCPs included Dimethicone, Carbomer, and Acrylates Copolymer (Figure 2), while the most common skeptical MP included Cyclopentasiloxane, Polysorbate 20, and Cetareth-20 (Figure 3). A wide variety of MPs identified in this activity also include several chemical forms of acrylates, Polyethylene, Polyquaternium, and PVP, are reported in Table 3, albeit at lower frequencies.

**Table 2.** Representative responses to survey question Q6, (N = 102).

Survey Question	Survey Results
Q6: "Additional comments regarding the activity on microplastics?"	Representative comments <p>"I never thought microplastics could be in the things I use daily."</p> <p>"It was easy to how common microplastics are in our everyday lives"</p> <p>"This activity was extremely fun and I will use it on all of my products in the future"</p> <p>"This activity was fun and very straightforward. I think it is an important assignment for all students. It would be fun to study more in-depth and scan more products."</p> <p>"The opp was really easy to use and helpful because without the opp, I wouldn't have known that I was using microplastics on my skin. I really enjoyed this activity"</p> <p>"This was a great way to explore products containing microplastics"</p> <p>"This activity was fun and informative. I am glad it was assigned."</p> <p>"Out of the ten different products I scanned, only three did not contain microplastics of any kind"</p>

Figure 2. The most frequently identified microplastics identified using the application.



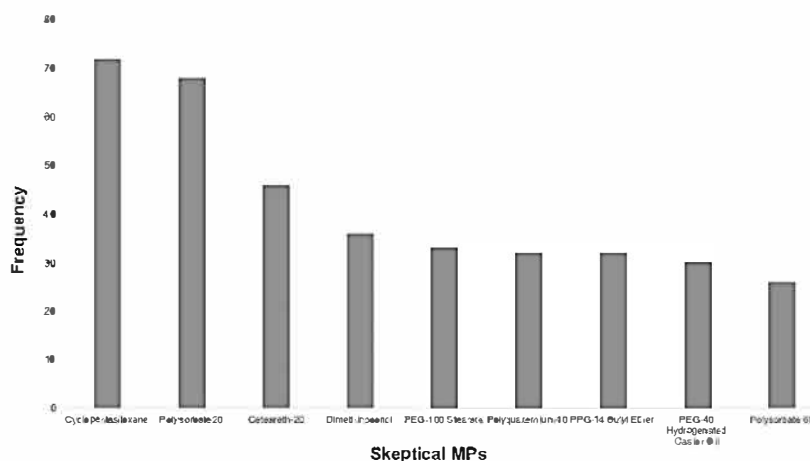
**Discussion**

Students identified a wide range of MPs and skeptical MPs while being engaged when performing this activity. Among these, Dimethicone is a non-biodegradable silicone-based polymer frequently found in PCCPs including handwash, shampoo, conditioner, moisturizing, and face cream (Nawalage & Bellanthudawa, 2022). Acrylate copolymer, a common ingredient of lotions and creams (Goddard & Gruber 1999), is an MP found in marine sediments (Cicinelli et al., 2021). Acrylate copolymer and carbomer when in large concentrations have the potential to inhibit the bioluminescence of the bacterium *Allibibrio fisheri*, reduce the growth rate of microalga, and

mobility of the crustacean *Daphnia* (Rozman & Kalcikova, 2021). Cosmetic products containing carbomer and acrylate copolymer, both of which were commonly reported by students across multiple products, have been found to enter aquatic ecosystems from wastewater treatment plant effluent (Mairinger et al., 2021). Another commonly identified skeptical MP, Cyclopentasiloxane from both antiperspirants and skin care products, is highly volatile and released into the atmosphere (Montemayor et al., 2013), and therefore may be unlikely to “go down the drain” or be released into the water supply.

Student participant responses indicated an overwhelming level of engagement and in many

Figure 3. The most frequently identified skeptical microplastics identified by students during this activity.





**Table 3.** Full List of MPs identified by students during this activity.

<i>Microplastics</i>	<b>Frequency</b>
Dimethicone	199
Carbomer	147
Acrylates Copolymer	65
Polyquaternium-7	34
Acrylates/C10-30 Alkyl Acrylate Crosspolymer	33
Polyethylene	27
PVP	22
Polymethyl Silsesquioxane	10
Sodium Polyacrylate	10
Polyisobutene	7
Trimethylsiloxysilicate	7
Polybutene	6
Styrene	6
Dimethicone Crosspolymer	5
Polyacrylate-13	5
Cetyl Dimethicone	4
Acrylates/C12-22 Alkyl Methacrylate Copolymer	3
Glyceryl Polyacrylate	3
Hydroxyethyl Acrylate/sodium	3
Polyacrylate-2	3
Polyquaternium-6	3
Polyvinyl Alcohol	3
Sodium Acrylate/Sodium Acryloyldimethyl Taurate Copolymer	3
Ethylene/Propylene/Styrene Copolymer	2
Polyester-7	2
Polyisoprene	2
Polymethyl Methacrylate	2
Polyurethane-35	2
Acrylamidopropyltrimonium Chloride	1
Acrylates/Hydroxyesters	1
Butylene/Ethylene/Styrene Copolymer	1
Ethylene/VA Copolymer	1
Ethylenediamine/Stearyl Dimer Dinoleate Copolymer	1
Nylon-12	1
Octylacrylamide/Acrylates/Butylaminoethyl Methacrylate Copolymer	1
Polyacrylamide	1
Polyester-111	1
Polyester-3	1
Polysilicone-2	1
Polyurethane-14	1
Simethicone	1
VA/Crotonates/Vinyl Neodecanoate Copolymer	1

cases students expressed future use of the application and an increased awareness of their own impact and realization that the products they regularly use contained either MPs or skeptical MPs. We recommend additional activities which focus on collecting environmental data on MPs as part of a laboratory that can further introduce undergraduates to research-based MP surveys for sampling soil (Rowe et al., 2019) or other available and easily accessible habitats, e.g., campus water sources, nearby beaches, etc. Public attitudes are vital to reduce MP pollution, and increased knowledge is correlated to an increased willingness to support environmentally friendly MP behaviors (Deng et al., 2020). Accordingly, enacting short effective outside-of-the-classroom activities using an app, as we utilized in our study, can scaffold to non-science majors, high school level students, and even the general public.

While there is increasing attention paid to pollution in aquatic environments, there is still a need for increased awareness and education on plastic pollution (Dalu et al., 2020). Current novel research is reporting on plastic particles being ubiquitous as pollutants in the environment and even present in human blood (Leslie et al., 2022) and lung tissue (Amato-Lourenco et al., 2021), highlighting the potential connection for students and all citizen scientists to be informed on MPs. MP awareness delivered via education programs mass media or policy enforcement as well as eco-friendly alternatives can provide a method for reducing the impact of these persistent MPs (Dowarah et al., 2022). Therefore, we recommend this activity be integrated into first-year environmental or biological courses at the college level and potentially included in high school science courses allowing students to collect data on MPs. Students were engaged and mentioned that they were increasingly informed about MP. While this activity emphasized the use of the smartphone app, students without a smartphone could either partner up with other students or look up information by searching for products online in order to complete the activity

([www.beatthemicrobead.org/product-lists/](http://www.beatthemicrobead.org/product-lists/)).

Further use of this application and activity could incorporate having students conduct follow-up research on the specific MPs and skeptical MPs they identified, possibly searching their potential impact on the environment, why they are used in PCCPs, and potential alternatives.

### Acknowledgments

We thank students who participated in this activity during a first-year organismal biology course. Research followed approved protocols for collection, storage of data, and student participation, #SU09122, of the Wingate University Research Review Board.

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## Innovations

### rDNA Technology and Genomic Library Construction Project in an Undergraduate Molecular Cell Biology Laboratory Class

Subir Ghosh, Ph.D.

Division of Natural Sciences, College of Natural & Applied Sciences

University of Guam,

UOG Station, Mangilao, Guam 96923, USA.

E-Mail: sghosh@triton.uog.edu

Tel. No. (671) 735-2789

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#### Abstract

Undergraduate students of a molecular cell biology laboratory class learn basic recombinant DNA (rDNA) technology by engaging in a Genomic Library Construction Project. Significantly, in this lab project students learn experimental design strategy. The challenge for students is to determine the strategy that they can take to efficiently screen for the success of the genomic library construction. The collaborative rDNA technology project requires basic equipment and reagents available in the molecular biology teaching lab and yet provides an advanced molecular cell biology learning experience and sparks enthusiasm for the molecular life sciences and biotechnology.

Keywords: Undergraduate; Genomic Library Project; Experimental Design Strategy

Running Title: Undergraduate Genomic Library Project.

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#### Introduction

Engaging students in hands-on experimental work in a molecular cell biology laboratory class is an effective way to spark enthusiasm for the molecular life sciences and biotechnology. It is well documented that learning is more effective when students work together collaboratively in problem solving activities (Frey et al., 2022; Ghosh, 2019; Premo et al., 2018; Allchin, 2017; Bierema et al., 2017; Freeman et al., 2014; Jacobs, 2014; McLaughlin and Metz, 2016; Waldrop, 2015). The National Academies of Sciences, Engineering, and Medicine in their 2017 report has included course-based experiences as part of UREs (undergraduate research experiences) for STEM students.

A Genomic Library Construction Project has been designed as part of an upper-division level cellular and molecular biology laboratory coursework to introduce rDNA technology to undergraduate juniors and seniors who have completed the pre-requisite Genetics course. The 1 unit (three hours per week) Cellular and Molecular Biology laboratory class enrolls a

maximum of 20 students. Two laboratory class sections are conducted every Spring semester for the 3 credit Cellular and Molecular Biology lecture course. In this cloning project performed collaboratively, students learn fundamental techniques used in rDNA technology. The students work in pairs and at every step of the multi-step project share their observation and results as well as difficulties encountered with other student groups and Instructor. This activity enables students to develop skills in troubleshooting experiments and the opportunity to review the science involved in the techniques. The multi-step project involves construction of a genomic library starting with genomic DNA fragments prepared from high molecular weight yeast genomic DNA that is recombined into a plasmid cloning vector. The recombinant plasmids are transformed into host *E. coli* bacteria and plated on selective bacterial media plates. The challenge the students face is to distinguish on the media plates between host bacterial colonies that have taken up the recombinant plasmid in contrast to bacteria that

have incorporated the plasmid cloning vector only without the genomic DNA inserts. This provides an opportunity for all students to design a strategy that can indicate the success of the Genomic Library Construction Project directly by observing the colonies growing on the media plates without having to perform any further experimental steps that would involve taking up more lab class sessions and reagents.

In this cloning project, students learn first-hand gene cloning techniques and analytical thinking skills critical for designing experimental strategies. In the prerequisite genetics course, students learn the mechanism of transformation, a genetic event that generates variation in prokaryotic bacteria. The Genomic Library Cloning Project, conducted in the cellular and molecular biology laboratory course, provides students the opportunity to apply their prior knowledge of transformation towards a major cloning endeavor. Students learn that the knowledge of science is critical for applications that directly benefit the further progress of science and contribute to solving practical problems. All other techniques in the multi-step cloning project are introduced to the students for the first time in their undergraduate biology curriculum. The multi-step Genomic Library Construction Project activity, and strategy taken by students to evaluate success of the cloning project is discussed in this paper.

#### **rDNA Technology Learning Objectives from the Genomic Library Construction Project**

rDNA technology has been the foundation for the rapid advances in investigating and understanding life at the molecular level. In the Cellular and Molecular Biology lecture class, students learn of molecular cloning techniques that enable scientists to identify genes and investigate their function in health and disease. The Genomic Library Construction Project laboratory class activity, performed collaboratively, provides undergraduate students with first-hand experience identifying, isolating, and manipulating biomolecules. The students learn to isolate genomic DNA from a

species; isolate plasmid DNA from bacteria; restriction analysis of DNA by gel electrophoresis; purification of DNA bands from the agarose gel, post electrophoresis; perform ligation reaction to clone genomic DNA fragments into plasmid cloning vector DNA; transform host cloning E. coli bacteria with the recombinant DNA molecules and plate onto selective media. An important task the students perform is to examine the plasmid cloning vector map and learn of the important components that constitute a cloning vector. Students learn of the multiple cloning sites that enable insertion of DNA sequences into the plasmid vector; of the origin of replication that enables the recombinant plasmid to autonomously replicate in the transformed cloning host bacteria; and the presence of the selectable marker gene that is critical for growth of only transformed bacteria on selective media. Students also learn that the optimum size of a library of completely random fragments of genomic DNA necessary to ensure representation of a particular sequence of interest is dictated by the size of the cloned fragments and the size of the genome. They further learn to use a statistical equation to determine the number of independent clones,  $N$ , that must be screened to isolate a particular sequence with probability  $P$  (provided in Materials & Methods). Significantly students learn experimental design and molecular strategies. Students are given the challenge of determining the strategy that can be taken to determine successful construction of genomic library by differentiating directly on the media plate, bacterial transformant colonies that have the genomic DNA fragments cloned as opposed to bacteria that have taken the plasmid vector DNA that did not ligate successfully with the genomic DNA inserts.

The Genomic Library Construction Project requires standard molecular biology equipment and reagents available in most institutions. Student fees for the laboratory course are available for the purchase of lab supplies and kits used for the molecular biology project. The multi-step project can be completed in four

three-hour long laboratory class sessions with active supervision and assistance from the instructor.

### **Genomic Library Construction Project Implementation Timeline:**

#### **Laboratory class schedule for the cloning project**

Each lab class is 3 hours long; Students work in pairs; Handout explaining cloning project and protocols for all procedures provided to students.

Lab Class 1: Isolation of Genomic DNA & Plasmid DNA; Restriction Endonuclease digestion of Genomic and Plasmid DNA.

Lab Class 2: DNA Agarose Gel Electrophoresis of Restriction Endonuclease digested Genomic and Plasmid DNA; Purification of DNA from Gel.

Lab Class 3: Ligation Reaction; Transformation of E. coli with Ligation Mix; Plating of E. coli on Selective Media plates.

Lab Class 4: Strategy to evaluate success of Genomic Library Construction Project; Examination of Selective Media plates to detect bacterial colonies transformed with the recombinant plasmid containing a genomic DNA insert fragment.

Genomic Library Construction Project Experimental Design

#### **Yeast Genomic DNA preparation to generate inserts for cloning into vector**

Standard molecular biology protocols for construction of the genomic DNA library are followed as provided in Molecular Cloning: A Laboratory Manual, 3<sup>rd</sup> Edition; and as per manufacturers protocols for the molecular biology kits and reagents used in this cloning project. Students isolate high molecular weight genomic DNA from yeast (*Saccharomyces cerevisiae*) using the Monarch Genomic DNA Purification Kit protocol (New England Biolabs, NEB). The enzyme Lyticase (Sigma-Aldrich) is used for enzymatic lysis of yeast cells to prepare genomic DNA. [In the event that students are not successful in isolating genomic DNA, the

instructor can provide prepared yeast genomic DNA, Sigma-Aldrich]. Next, they restrict the yeast genomic DNA with a restriction endonuclease enzyme (NEB) to generate fragments of varying length. The specific restriction endonuclease enzyme used is revealed to the students during the last stage of the project where the strategy to evaluate success of the Genomic Library Construction Project is formulated. The restricted genomic DNA is electrophoresed in a 1% DNA agarose gel in Tris-Acetate-EDTA (TAE) electrophoresis buffer to obtain a range of DNA fragments from high to low molecular weight and visualized as a smear by staining the gel with DNA specific dye SYBR<sup>TM</sup> Green (Thermo Fisher Scientific) and illuminated with a Syngene<sup>TM</sup> blue light transilluminator (Fisher Scientific). For safety reasons, the instructor (not the students) excises different parts of the restricted genomic DNA smear in the gel representing a specific molecular size range using a razor blade. Collectively all the student groups in the class approximately cover the entire molecular size range of the restricted yeast genomic DNA. The genomic DNA fragments are purified from the agarose gel by the students using Freeze 'N Squeeze<sup>TM</sup> DNA gel extraction spin columns (Bio-Rad) to generate a large population of yeast genomic DNA fragments of varying length for insertion into a plasmid cloning vector to prepare a yeast genomic library.

#### **Plasmid Cloning Vector preparation**

Students isolate using the Aurum<sup>TM</sup> Plasmid Mini Kit (Bio-Rad), the covalently closed circular plasmid cloning vector DNA (pGlo) from E. coli bacteria that contains the plasmid (Source: Instructor; prepared using the Bacterial Transformation Kit, Bio-Rad). The isolated plasmid DNA is restricted with the same restriction endonuclease used to generate yeast genomic DNA fragments. This results in linearization of the plasmid with compatible cohesive ends that would allow insertion of the genomic DNA fragments in to the plasmid cloning vector. The restricted plasmid is electrophoresed on a 1% DNA agarose gel to separate completely

restricted plasmid from unrestricted plasmid. The linearized plasmid forms a sharp 5.3 kilobase pair DNA band and is purified from the gel, using Freeze 'N Squeeze™ DNA gel extraction spin columns (Bio-Rad).

#### **Ligation Reaction to insert Yeast Genomic DNA fragments into Plasmid Cloning Vector**

The purified population of linear double-stranded yeast genomic DNA fragments are inserted into the linearized double-stranded plasmid cloning vector DNA in a ligation reaction using the T4 DNA Ligase enzyme and protocol provided by NEB. The ligation reaction is performed at room temperature for an hour. During this incubation time, the mechanism of the ligation reaction is discussed, and students are quizzed on the expected final products of the joining reaction. In the quiz students are asked to draw the starting substrates in the ligation reaction and then predict all the possible outcomes at the end of the joining reaction. Typically, most of the students provide diagrams of the recombinant plasmid cloning vector containing genomic DNA inserts of varying length. The students are quizzed again and asked to formulate a strategy to determine the success of their ligation reaction. Students find it challenging and are unable to answer this question. The students are informed that after completion of the ligation reaction, an aliquot of the ligation reaction can be electrophoresed in an agarose gel and after DNA staining the products of the joining reaction can be examined to determine the success of the ligation reaction. Students are provided a hint by discussing the science of DNA staining in an agarose gel and the fact that the intensity of the stain indicates the amount of DNA present in the DNA band observed in the gel. The students are asked to generate a hypothesis for the expected result of a successful ligation when the completed ligation reaction is examined in a DNA gel after electrophoresis. The students find this question challenging and are unable to provide the correct answer. The students learn that the staining intensity of the plasmid vector DNA band would decrease if successfully ligated to

the large population of genomic fragments of varying sizes. The entire class benefits from the discussions since they learn analytical thinking that leads to developing scientific hypothesis.

#### **Transformation of Cloning Host E. coli with the Ligation Reaction Mix**

Students introduce the ligated recombinant cloning vector plasmid containing the yeast genomic DNA fragments into cloning host HB101 strain of E. Coli bacteria using the Calcium Chloride Transformation procedure (Hanahan, 1983; Bacterial Transformation Kit, Bio-Rad). Next, the E. coli bacteria are plated on antibiotic selection media plates (LB agar nutrient media plates containing the ampicillin antibiotic, Bio-Rad) and incubated overnight at 37 °C to select for transformed bacteria that potentially contain the recombinant plasmids into which the yeast DNA genomic fragments have been cloned. In the final step of the cloning project, the students address the challenging task of evaluating the success of the Genomic DNA Library Construction Project and this exercise is the highlight focus of this paper and is discussed in the next section.

#### **Strategy to Evaluate Success of the Genomic Library Construction Project**

A highlight of the Genomic DNA Library Construction Project is for students to learn molecular strategies taken in designing experimental approaches. Cloning of the genomic fragments into a standard plasmid cloning vector results in recombinant plasmids bearing genomic DNA inserts. However, during the ligation reaction a population of plasmids do not take up the genomic inserts and self-seal to recreate the original plasmid. When plated onto antibiotic selection media plates, both host E. coli containing the recombinants as well as bacteria bearing only the plasmid vector are observed to grow on the media plates. The bacterial colonies are indistinguishable, and the students cannot determine if they have been successful in construction of the genomic library.

Students are given the challenging task of determining a strategy that can be taken to

evaluate the success of constructing the genomic library. Rather than use a standard plasmid cloning vector, students are provided the pGlo recombinant plasmid (Bio-Rad) to be used as a cloning vector for yeast genomic DNA library construction. The pGlo plasmid contains a jellyfish Green Fluorescent Protein coding gene (Prasher et al., 1992). The plasmid has a cloning site bearing multiple restriction endonuclease recognition sites and has restriction sites at other regions of the plasmid including in the antibiotic selection gene, the origin of replication, and within the GFP gene sequence. Students are provided a map of the pGlo plasmid (Bio-Rad) and given the task of determining a suitable strategy that can be taken to differentiate *E. coli* host bacteria that are transformed with recombinant plasmids containing yeast genomic fragments in contrast to *E. coli* transformed with the pGlo plasmid alone that self-sealed during ligation and failed to insert the genomic fragments. This task is given as homework to the students followed by a lab class meeting where the strategy is discussed and the final step of evaluation of the success of the cloning project is completed.

The strategy taken to screen transformed bacterial colonies on selective media plates for determining success of the construction of the genomic DNA library is a highlight of the project and presents a challenge to all students. Almost all students state that the yeast genomic DNA fragments should be cloned in the multiple cloning site of the pGlo plasmid leaving the GFP coding sequence intact. Thus, their strategy to evaluate the success of the Genomic Library Construction Project was that all bacterial colonies that were transformed with the recombinant pGlo plasmids containing genomic inserts would result in bacterial colonies glowing green. The strategy was reviewed, and students learn that their approach would not be able to evaluate the success of the cloning project since even the pGlo plasmids that did not contain the genomic inserts would contain the intact GFP coding sequence and would also result in glowing green transformed bacterial colonies.

The students also learn that the yeast genomic DNA fragments should not be inserted into the "ori" site (origin of replication) of the pGlo plasmid since the origin of replication site enables the recombinant plasmid to autonomously replicate and amplify in the transformed cloning host bacteria. The antibiotic selectable marker gene present in the pGlo plasmid should also be intact since it is critical for growth of only the transformed bacteria on selective media.

Thus, the strategy that should be taken is to clone the yeast genomic DNA fragments directly into the coding sequence of the GFP gene via a unique restriction site, Xho1, present in the gene and not anywhere else in the pGlo plasmid. This would result in inactivation of the GFP gene and expression of the GFP protein would be lacking in bacteria transformed with recombinant pGlo plasmids containing the genomic DNA inserts. Thus, all transformed bacteria glowing green on the media plates would reflect bacteria containing plasmid lacking genomic inserts. Only the bacterial colonies not glowing green would reflect the success of the Genomic Library Construction Project. The cloning project provided the opportunity to all students to learn experimental molecular strategy.

### Discussion

The Genomic Library Construction Project provides undergraduate students the opportunity to learn first-hand gene cloning and analytical thinking required for developing experimental strategies. Students collaborate and complete the multi-step cloning project to construct a genomic DNA library, learning fundamental molecular biology concepts and rDNA technology techniques as they perform each step of the project. Students performed a wide range of molecular techniques – DNA isolation; restriction endonuclease mapping of DNA; DNA analysis by DNA gel electrophoresis; purification of specific DNA fragments from the agarose gel; cloning vectors; ligation reaction to create recombinant DNA; transformation of cloning host bacteria; and screening of bacterial transformants on selective media plates.



Students further learnt of library screening procedures using labeled molecular probes to discover novel genes involved in a cellular process under investigation by scientists. The Genomic Library Construction Project requires standard molecular biology equipment and reagents available in most institutions. The project involves multiple steps taking four three-hour long laboratory class sessions with active supervision and assistance from the instructor. Standard safety procedures and rDNA technology guidelines as established by the NIH are followed in the laboratory class (The National Institutes of Health Guidelines for Research involving recombinant or synthetic Nucleic acid molecules, NIH, April 2019). Students collaboratively ensure success of each step thereby learning responsibility in undertaking projects. It is anticipated that active engagement in projects such as the genomic DNA library cloning exercise will go a long way in motivating students and raising enthusiasm for the molecular life sciences and future careers in research and the health field.

#### Methods

- The NIH guidelines (The National Institutes of Health Guidelines for Research involving recombinant or synthetic Nucleic acid molecules) are followed for construction and handling of recombinant DNA molecules and host bacteria containing these molecules (<https://osp.od.nih.gov/biotechnology/nih-guidelines/>).
- Standard molecular biology protocols for construction of the genomic DNA library are followed as provided in Molecular Cloning: A Laboratory Manual, 3<sup>rd</sup> Edition; and as per manufacturers protocols for the molecular biology kits and reagents.
- Students learn that the size of a library of completely random fragments of genomic DNA that is necessary to ensure representation of a particular sequence of interest is dictated by the size of the cloned fragments and the size of the genome. The number of independent clones, N,

that must be screened to isolate a particular sequence with probability P is given by

$$N = \ln(1-P) / \ln(1-(l/G)),$$

where, l, is the size of the average cloned fragment, in base pairs, and G is the size of the target genome, in base pairs. In order to have a 99% chance of isolating a desired sequence, the number of clones screened should be such that the total number of base pairs present in the clones screened (l x N) represents a 4.6-fold excess over the total number of base pairs in the genome (Molecular Cloning: A Laboratory Manual, 3<sup>rd</sup> Edition).

#### Reagents

New England Biolabs (NEB): Monarch Genomic DNA Purification Kit, Catalog # T3010S; Xho1 Restriction endonuclease, Catalog # R0146SVIAL; DNA Molecular Weight Marker, Catalog # N3012S; and Quick Ligation Kit, Catalog # M2200S.

Sigma: Lyticase, Catalog # L2524 (for enzymatic lysis of yeast cells to prepare genomic DNA); High Molecular Weight Yeast Genomic DNA, Catalog # 69240-M.

Bio-Rad: Aurum plasmid Mini Kit, Catalog # 7326400EDU; Freeze 'N Squeeze™ DNA gel extraction spin columns, Catalog # 7326165EDU; Bacterial Transformation Kit (includes pGlo plasmid & restriction endonuclease map; cloning host HB101 strain of E. coli; Calcium Chloride Transformation solution; LB liquid and solid nutrient agar media; selective LB media agar plates prepared by adding ampicillin antibiotic, and arabinose sugar to induce GFP gene expression – provided in kit), Catalog # 1660003; DNA grade agarose, Catalog # 1613100EDU; nucleic acid electrophoresis buffer, Catalog # 1610743EDU; DNA Electrophoresis Sample Loading Dye, Catalog # 1660401EDU. [EDU – educational discount provided by Bio-Rad].

Thermo Fisher Scientific: SYBR™ Safe DNA Gel Stain, Catalog # S33111

Fisher Scientific: Syngene™ Slimline LED Blue Light Transilluminator, Catalog # 01-257-274.

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### Conflict of Interest

The author declares no conflict of interest.

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## Perspectives

### Why Fractal Features Flourish: Measurement, Modeling, and Morphogenesis

John R. Jungck, Professor of Biological Sciences and Mathematical Sciences

Delaware Biotechnology Institute, 15 Innovation Way, University of Delaware, Newark, DE 19716

email: [jungck@udel.edu](mailto:jungck@udel.edu)

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#### Abstract

Why should our life science colleagues adopt, adapt, and implement fractal thinking into their research and teaching paradigms? While numerous articles and books have been written about the utility, power, and insight that can be gained by using fractal analysis of data and developing fractal models, other than just appreciating the beauty of fractal biological images, (1) how do we help our life science colleagues to know **why** they should learn about fractals, (2) **where** and **when** fractal analysis and modeling are most applicable, (3) **which** tools should be applied, (4) what **would** a fractal perspective offer them in terms of potential for more deeply understanding their problem, and (5) **how** do these complement and/or contradict their current practices?

**Key words:** fractals, dimension, measurement, modeling, morphogenesis

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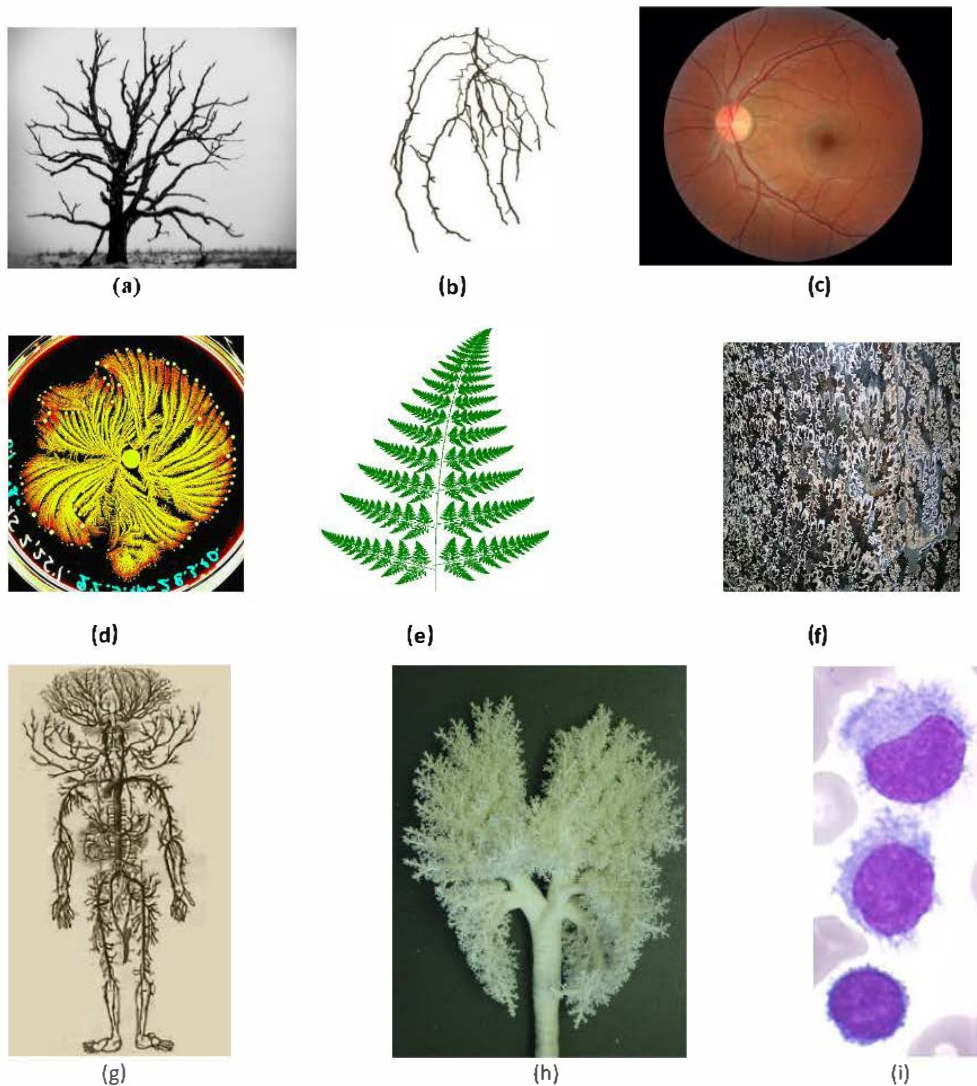
What properties do the fractal images in Figure 1 have in common? Obviously, they are distinct from the structural patterns of Classical or Euclidean geometry that introduced us to triangles, squares, trapezoids, parallelograms, circles, spheres, and polyhedra which are used for measuring and examining symmetric properties. However, the fractal images in Figure 1 are not Euclidean figures, they are typically described as fractured, fissured, fingered, irregular, asymmetrical, or broken. I believe that these images are beautiful, but how can they be useful for doing pattern recognition, image analysis, modeling, and hypothesis testing?

So, what are fractals? Fractals are characterized by three primary features: (1) self-similarity; (2) iterative construction; and (3) non-integer dimensions. Self-similar patterns are ones that repeat forever such that every part of the fractal image looks very similar to the whole image, regardless of how much you zoom in or zoom out. That is, when you look at a piece of an image it looks like the whole image as if the twig of a tree is similar in shape to a full tree. Through iterative construction, fractal patterns are generatable by repeatedly using simple rules over and over to form complex structures. For example, if I can make a rule to draw a twig, I might be able to accumulate enough twigs to

completely generate a whole tree. What does it mean that a structure could have a non-integer dimension? Biologists are used to measuring biological specimens in terms of Euclidean geometry by determining length, width, height, area, and volume. The integer dimensions in Euclidean geometry are zero for points, one for lines, two for planes, and three for solids. Fractals can be measured to have non-integer dimensions because a squiggly line does not have a dimension of one because it fills up part of the plane. The squiggly lines have fractal dimensions greater than one for a straight line but less than two for a full plane. Similarly, a crunched-up planar sheet to make a ball will be crumpled, creased, irregularly shaped and will incompletely fill a sphere. It has fractal dimensions greater than two for a plane but less than three for a full sphere. I elaborate on these fractal features as another mathematical tool.

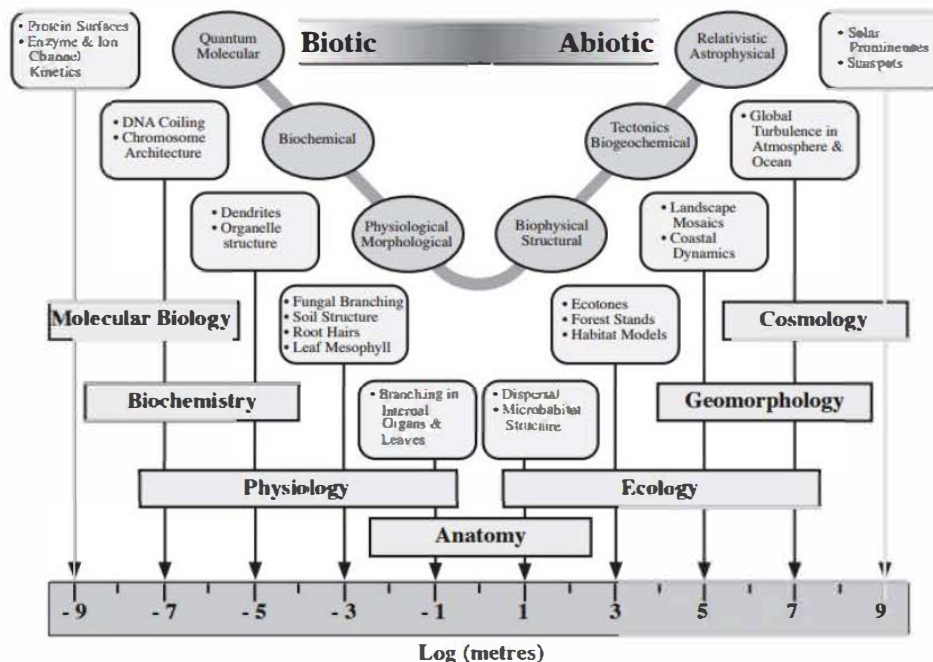
Why should our life science colleagues adopt, adapt, and implement fractal thinking into their research paradigm? While numerous articles and books have been written about the utility, power, and insight that can be gained by using fractal analysis of data and developing fractal models, other than just appreciating the beauty of fractal biological images (Figure 1), how do we help our life science colleagues to

Figure 1. Biological images with fractal aspects



**(a)** Winter tree (Ann Arbor Photography, personal permission); **(b)** White pine seedling root (Mitchell, H. H. (1939). "The Growth and Nutrition of White Pine Seedlings in Cultures with Varying Nitrogen, Phosphorus, Potassium and Calcium." *The Black Forest Bulletin* 9: 70); **(c)** Retinal image of blood vessels (Fundus photograph of a healthy left eye (OS) from a myopic Asian male patient Age 23, Wikicommons); **(d)** Fractal bacteria (*Paenibacillus vortex*) grown on hard agar with very few nutrients created at Prof. Ben-Jacob's lab, at Tel-Aviv University, Israel (Wikicommons); **(e)** Barnsley fern mutated -*Leptosporangiate* fern (Wikicommons); **(f)** *Placenticeratid* ammonite - Pierre Shale, Upper Cretaceous; Meade County, South Dakota, Wikicommons); **(g)** Fractal human circulatory system 19<sup>th</sup> century copper (*Encyclopédie, ou dictionnaire raisonné des sciences, des arts et des métiers*) / par une société de gens de lettres. Recueil de planches sur les sciences, les arts libéraux et les arts mécaniques, avec leur explication. [Anon]. Date:1762 Wikicommons: <https://wellcomecollection.org/works/bza78pcs>); **(h)** Cast of lung from a dog: (<<http://classes.yale.edu/fractals/Panorama/Biology/Physiology/AnimalLungs/AnimalLungs.html>>); **(i)** Hairy Cell Leukemia (Franck Genevieve, Laboratory of Hematology, Academic Hospital of Angers, Angers, Pays de la Loire, France.).

Figure 2. From Kenkel and Walker (1996).



“Examples of fractal patterns at various spatial scales (rounded boxes). Ovals summarize general processes operating at each scale: biotic processes predominate at finer spatial scales, abiotic processes at coarser scales. Rectangles indicate the scientific disciplines at which these patterns and process are normally studied.”

know why they should learn about fractals, where and when fractal analysis and modeling are most applicable, which tools are most appropriate and easy to apply, what would a fractal perspective offer them in terms of potential for more deeply understanding their problem, and how do these complement and/or contradict their current practices?

Many life science colleagues challenge the utility of fractals by frequently dismissing fractals as an ephemeral fascination with our latest new toy, of only peripheral importance to most primary biological and medical phenomena, and view it as yet one more exotic mathematical and theoretical curiosity of little practical utility to them. I believe that this perception not only should be directly addressed, but in doing so, we need to civilly address these concerns by providing very practical advice that humbly notes when a fractal approach should make a

significant difference to their particular problems, practices, and philosophical perspectives. Thus, by focusing on the practical utility of fractals to three research endeavors, namely, measurement, modeling, and morphology, I believe that we can enlarge the conversation with our life science colleagues.

Obviously, the use of mathematics in medicine is one area that many colleagues feel justifies their attention, thus, I frequently begin by quoting John *et al.* (2015):

“Fractal analysis has the potential to allow physicians to make predictions about clinical outcomes, categorize pathological states, and eventually generate diagnoses,” and, then following up with examples that instantiate such a claim. Among the medical problems that have been investigated are differential diagnoses by analyzing shapes of tumors in tissues and cells, microvessel networks, platelet activation,

microvascular patterns in oral mucosa, EEG/ECG signals, brain images, bone texture, and mammography.

Also, I like sharing the image from Kenkel and Walker (1996) (Figure 2) as it nicely lays out how fractals apply to biological patterns from the molecular to ecological level in a visually succinct fashion.

Over the past thirty-seven years, I have lead faculty professional development and curriculum development workshops on quantitative reasoning, visualization, and modeling through the BioQUEST Curriculum Consortium (<http://bioquest.org>; now subsumed into <http://QUBESsub.org>), the National Institute for Mathematical and Biological Synthesis Center (<http://www.nimbios.org/>), and the Society for Mathematical Biology (<https://www.smb.org/>). Even though many participants have enjoyed the beauty of fractals, the questions raised above have come up frequently. I have found that it has been important to not only focus on advocating for the power of fractals, but to put them into a larger context of mathematical biology and to direct them to practical tools that apply to their data. The silver lining of their challenges has been to expand discussions of a breadth of approaches to measurement, modeling, and morphogenesis to introduce other considerations about learning more generally about why they should learn a variety of approaches, where and when a particular approach to analysis and modeling is most applicable, which tools to apply, what a particular perspective would offer them in terms of potential for more deeply understanding their problem, and how these complement and/or contradict their current practices.

### Measurement

Fractal measurement has enormous utility in examining highly irregular forms and structures that are self-similar across numerous scales. The famous Mandelbrot coastline of England example has been repeated in examining biological patterns of morphologies from the molecular (DNA fractals) to subcellular (fractal

globules in chromosome packing) to cellular (hairy cell leukemia) to multicellular (bacterial colonies) to organ (kidney tubules, bronchial branching in lungs) to organismal (circulation system, lymph system, nervous system, tree architecture) to ecosystems (fragmented forests) to just name a few. Whenever a fragmented, fissured, folded, or filamentous feature appears, we have promoted the utility of fractals. Also, we assert to our colleagues that whenever a mean length, area, or volume is not robust to changes in the scale of our measuring device that then fractal measurement is warranted and appropriate. The standard Richardson plot of the log-log transform of the number of different size boxes to cover an image versus the size of boxes employed to determine a fractal dimension needs explanation beyond a simple equation to be meaningful to most biologists. To most biologists, the visualization of space filling versus fractal dimension is usually most productive.

To extend this, let us compare some alternative geometries and topology and when are they most useful. Euclidean geometry is appropriate when one wants to measure parameters such as length, area, volume, and angles. Fractal geometry is appropriate when one wants to measure irregular edges, surfaces, volumes, and self-similar structures. Their applications and available software for making measurements are listed in Table 1.

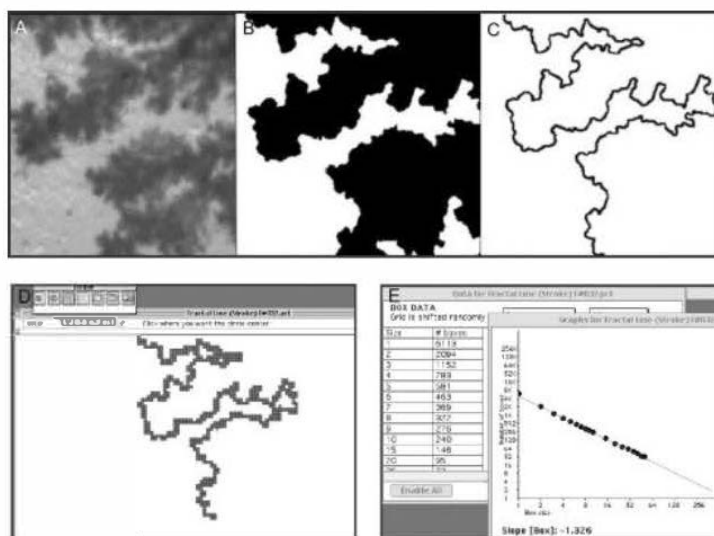
Two mathematical tools are particularly useful for determining fractal dimensions: the Hutchinson metric and a Richardson plot (For a walk through of actually processing a biological image and constructing a Richardson plot to determine a fractal dimension, see Jungck, Stanley, and Fass, 2003; Figure 3).

As useful as Euclidean geometry and fractal geometry approaches are, it is important to share that there more mathematical alternatives for making measurements on biological images (Table 2). It helps us to choose tools appropriate to a particular problem. Just as Euclidean geometry was insufficient to help us measure properties of fractal images, there are other

**Table 1.** Comparison of Euclidean Geometry and Fractal Geometry and their applications

Kind of Geometry	Types of Measurement	Applicable Software
Euclidean Geometry	length, area, volume, and angles; patterns of neighbors	<b>Image J</b> (< <a href="https://imagej.net/ij/">https://imagej.net/ij/</a> >) <b>Fiji</b> (< <a href="https://imagej.net/software/fiji/downloads">https://imagej.net/software/fiji/downloads</a> >) <b>Cell Profiler</b> (< <a href="https://cellprofiler.org/releases">https://cellprofiler.org/releases</a> >) <b>Ka-me: A Voronoi Image Analyzer</b> (< <a href="https://qubeshub.org/resources/kame">https://qubeshub.org/resources/kame</a> >)
Fractal Geometry	<b>Fractal Dimensions via:</b>  Box-counting: (< <a href="https://en.wikipedia.org/wiki/Minkowski-Bouligand_dimension">https://en.wikipedia.org/wiki/Minkowski-Bouligand_dimension</a> >)	FracLac plug-in for Image J (< <a href="https://imagej.nih.gov/ij/plugins/fractalac/FLHelp/Introduction.htm">https://imagej.nih.gov/ij/plugins/fractalac/FLHelp/Introduction.htm</a> >) FracOut! (< <a href="https://code.google.com/archive/p/frakout/">https://code.google.com/archive/p/frakout/</a> >) MULTIFRAC: An ImageJ plugin for 3D stack images (< <a href="https://doi.org/10.1016/j.softx.2020.100574">https://doi.org/10.1016/j.softx.2020.100574</a> >) Others: AnaMorf; IJBlob; BoneJ; SoilJ; Hypha tracker; Comstat 2; and 3D Fractal Tree
	Concentric circles: mass dimension, sometimes referred to as the sandbox dimension	Benoit (< <a href="https://www.trusoft-international.com/benoit.html">https://www.trusoft-international.com/benoit.html</a> >); (< <a href="http://what-when-how.com/biomedical-image-analysis/estimation-techniques-for-the-fractal-dimension-in-binary-images-biomedical-image-analysis/">http://what-when-how.com/biomedical-image-analysis/estimation-techniques-for-the-fractal-dimension-in-binary-images-biomedical-image-analysis/</a> >)

**Figure 3.** Five steps in a fractal analysis of a bacterial colony edge



(A) Original photo image converted to gray scale. (B) A threshold value is set to convert the image to just black or white. (C) Edge detection is used to construct the perimeter. (D) A series of square-gridded graph of various sizes are placed over the image and the number boxes that contain a piece of edge are determined. (E) The fractal dimension is computed from the slope of the line produced by plotting the log of the size of boxes in the grid to the number of boxes occupied by the sample edge (a Richardson plot). Basic models for how these patterns form are well reviewed by Othmer *et al.* (2009). Their spatiotemporal reaction-diffusion models consider a wide variety of bacterial behaviors: (1) consumption of nutrients, (2) production of chemoattractants, (3) tactic movement toward attractants, (4) cell growth, and (5) loss of motility.

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ways of examining biological patterns. For example, the hyperbolic crocheted coral reef (<https://crochetcoralreef.org/>) is a wonderfully beautiful project that combines ecological activism by a community of women crafters along with higher mathematics. When we view 3D models of biomolecular structures, typical software allows us to zoom in or zoom out, rotate, and translate, but not fly through a molecule. Paul McCreary (personal communication) has illustrated the use of hyperbolic geometry to fly through 3D models of biomolecular structures in several of our BioQUEST workshops.

When these multiple kinds of geometry and distinctions between geometry and topology are offered, the critique of fractals *per se* diminishes as participants in workshops perceive that they are aware of and use different tools for different problems. In other words, just because you have a hammer, you haven't turned all problems to concern nails.

On the other hand, we also have to address the problem of neophytes presuming that their excitement about fractals doesn't go too far. In the classic meta-analysis article by Avnir, Biham, Lidar, and Malcai (1998): "Is the Geometry of

Nature Fractal?," after examining papers published over a seven year span, they raise an important caveat:

Do power laws that are limited in range represent fractals? Is it justified to term them as such? Regardless of the question of fractality, a more basic question should be asked: Is this presentation useful? The very existence of so many reports by competent researchers who are well aware of the problematics of declaring fractality for experimental results that span only one order of magnitude suggests that experimentalists seem to gain from the resolution analysis and from the fact that the result of such analysis is often a power law. The usefulness is in the following points: (i) The power law condenses the description of a complex geometry. (ii) It allows one to correlate in a simple way properties and performances of a system to its structure and to the dynamics of its formation. (iii) In many instances, the choice is either to use the limited-range data or to discard it altogether and not have even an approximate picture of the studied object. Opting for the former can be emphatically understood. (iv) Fractal geometry provides a proper language and symbolism for studies of ill-defined geometries.

**Table 2.** Alternative mathematical approaches for biological image analysis

<b>Spatial approach</b>	<b>Types of Measurement</b>	<b>Applicable Software</b>
Graph Theory (topology not geometry)	Number of vertices, edges, faces; lengths of minimal spanning trees, Hamiltonian paths of vertices, and Eulerian paths of edges; Existence of cycles, tours, trees,	<b>Ka-me:</b> A Voronoi Image Analyzer ( <a href="https://qubeshub.org/resources/kame/">https://qubeshub.org/resources/kame/</a> )
Taxi Cab Geometry	Distances restricted to traveling on grids; Manhattan distances	GeoGebra ( <a href="https://www.geogebra.org/m/k23tpEYX">https://www.geogebra.org/m/k23tpEYX</a> )
Network Analysis	Whether small world, scale free, or random network; consistency index; diameter; centrality; modularity; betweenness; assortment coefficient; motifs; triad profile analysis	Cytoscape ( <a href="https://cytoscape.org/">https://cytoscape.org/</a> ) TouchGraph ( <a href="https://www.touchgraph.com/">https://www.touchgraph.com/</a> ) BioGrapher ( <a href="https://www.worldscientific.com/doi/abs/10.1142/9789813141919_0008">https://www.worldscientific.com/doi/abs/10.1142/9789813141919_0008</a> )
Hyperbolic geometry	Negative curvature of surfaces (See: Evans, Myfanwy E., and Gerd E. Schröder-Turk. "In a material world: Hyperbolic geometry in biological materials." <i>Asia Pacific Mathematics Newsletter</i> 5, no. 2 (2015): 21-30.	HWT: a hyperbolic wavelet transforming base ( <a href="https://www.researchgate.net/publication/232063137_The_hyperbolic_wavelet_transform_An_efficient_tool_for_multifractal_analysis_of_anisotropic_fields">https://www.researchgate.net/publication/232063137_The_hyperbolic_wavelet_transform_An_efficient_tool_for_multifractal_analysis_of_anisotropic_fields</a> )



From this quote, I infer and share with participants that on technical grounds they may have exceeded the technical expectations for aptly applying a fractal analysis, but that heuristically they may have benefitted by developing new insights simply by analyzing their problem from a different perspective.

### **Modeling, Morphogenesis, and Morphology**

Fractal biological systems have been modeled by a variety of approaches that range from bottom-up deterministic approaches like cellular automata models to top-down homogeneous and continuous differential equation models. Most of these models have focused on using the self-similarity of fractal forms to develop algorithms and heuristics that are dependent on a very minute set of variables. The resulting scale invariance means that an enormous diversity of biological shapes and patterns can be modeled with fractals. Five different modeling approaches are widely used to model fractal morphogenesis: (1) Diffusion-limited aggregation has been used to model bacterial colony formation, flocking of marine organisms, and, Hele-Shaw growth patterns; (2) Cellular Automata have been used to model polarized movement of cells, chemotaxis, rippling in bacteria colonies, cell alignment, swarming and flocking, and, clustering limb bud cells; (3) Lindenmayer systems have been used to model phytoarchitecture, neural growth, physiological systems, and fossil fronds; (4) Reaction-Diffusion equations have been used to model cancer tumor growth and bacterial colony patterns; and, (5) Blinking fractals have been used to model studying seasonal changes during the growth of biological systems and the growth of a forest. Thus, as with visualization and measurement of fractals, there are a variety of modeling approaches to generating fractal forms (morphogenesis) and users need to be aware of which one is most appropriate or simply easily applicable to their object of inquiry.

Morphology is the study of the size, shape, and structure of biological structures. Weibel (2011) has argued that fractals provide a design principle: "Many patterns of nature are too fragmented and irregular to be described in such terms and therefore appear "amorphous." ... fractal geometry as a-perhaps superimposed-

design principle of living organisms is heuristically very productive in relation to many questions." Over the past thirty years, many morphologists have argued that fractal geometry for the quantitative analysis and description of the geometric complexity of natural systems has been a major paradigm shift. One particular aspect of this shift has been to focus on the importance of self-organization rather than a blueprint approach that dictates the development and evolution of every structure. As one of my mentors, Evelyn Fox Keller (2009), has argued: self-organization "denote[s] a paradigm shift or revolution in scientific thinking about complex systems. The developments responsible for this claim began in the late 1960s and came directly out of the physical sciences. They rapidly attracted wide interest and led to yet another redrawing of the boundaries between organisms, machines, and naturally occurring physical systems (such as thunderstorms). In this version of self-organization, organisms are once again set apart from machines precisely because the latter Fractal geometry also has contributed to our understanding of evolutionary trajectories of morphological change". In a recent example, Lemanis and Zlotnikov (2023) examined fossil ammonoids by determining the fractal dimension of the sutures.

"Fractal-like, intricate morphologies are known to exhibit beneficial mechanical behavior in various engineering and technological domains. The evolution of fractal-like, internal walls of ammonoid cephalopod shells represent one of the clearest evolutionary trends toward complexity in biology, but the driver behind their iterative evolution has remained unanswered since the first hypotheses introduced in the early 1800s. We show a clear correlation between the fractal-like morphology and structural stability. Using linear and nonlinear computational mechanical simulations, we demonstrate that the increase in the complexity of septal geometry leads to a substantial increase in the mechanical stability of the entire shell. We hypothesize that the observed tendency is a driving force toward the evolution of the higher complexity of ammonoid septa, providing the animals with superior structural support and protection against predation. Resolving the adaptational value of

this unique trait is vital to fully comprehend the intricate evolutionary trends between morphology, ecological shifts, and mass extinctions through Earth's history."

Besides mechanical stability, space filling fractal architectures allow the three-dimensional distribution of materials (e.g., oxygen from lungs throughout the arteries and the return of CO<sub>2</sub> through the veins), signals (e.g., nervous system), reception of light (e.g., the phytoarchitectural distribution of limbs and orientation and morphology of leaves), and the foraging patterns for resources (e.g., fractal bacterial colonies). It is this breadth of applications of fractal geometry to morphology that has contributed enormously to our basic understanding of biological morphology. If we then add the pragmatic utility of fractal analysis to medical diagnosis, treatment, and prognosis, we begin to appreciate the revolutionary paradigm shift that has occurred.

### Conclusion

By placing fractal measurement and modeling in a context of multiple alternatives appropriate for approaching patterns, problems, and processes, I believe that our colleagues are potentially more receptive to apply fractal analysis to their own research in appropriate ways with less resistance to a new approach, have a better appreciation for what fractal analysis might contribute, and have some idea of the variety of tools for using fractal measurements and models appropriately and effectively.

If we are to promote this "dialogue across disciplines," I believe that there is an onus on us to demonstrate the diversity within our mathematical modeling approaches to morphogenetic processes and patterns and to demonstrate where and when alternative approaches are most useful.

In summary, fractal geometry offers biologists three major contributions to our understanding of how different morphologies function and the evolutionary pressures under which they have evolved. (1) Pragmatism: the obvious utility of making measurements of fractal dimensions allows us to quantify irregular features and make inferences about evolutionary pressures, mechanical and

informational optimality, and medical practice. (2) Mathematics: by understanding that mathematics is constantly developing rather than simply reifying two-thousand-year-old Euclidean geometry as the only way to make spatial measurements and models, we are open to a much wider suite of potential applications and insights to comprehending biological phenomena. (3) Philosophy: fractal geometry provides another example of reinforcing a paradigm shift started by Darwin in re-interpreting design in nature as being driven by selective biotic and physical forces.

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## ***Bioscene: Journal of College Biology Teaching*** **Submission Guidelines**

### I. Submissions to *Bioscene*

*Bioscene: Journal of College Biology Teaching* is a refereed publication of the Association of College and University Biology Educators (ACUBE). *Bioscene* is published online only in May and in print in December. Submissions should reflect the interests of the membership of ACUBE. Appropriate submissions include:

- **Articles:** Course and curriculum development, innovative and workable teaching strategies that include **some type of assessment** of the impact of those strategies on student learning.
- **Innovations:** Laboratory and field studies that work, innovative and money-saving techniques for the lab or classroom. These do not ordinarily include assessment of the techniques' effectiveness on student learning.
- **Perspectives:** Reflections on general topics that include philosophical discussion of biology teaching and other topical aspects of pedagogy as it relates to biology.
- **Reviews:** Web site, software, and book reviews
- **Information:** Technological advice, professional school advice, and funding sources
- **Letters to the Editor:** Letters should deal with pedagogical issues facing college and university biology educators

### II. Preparation of Articles, Innovations and Perspectives

Submissions can vary in length, but articles should be between 1500 and 5000 words in length. This includes references and tables but excludes figures. Authors must number all pages and lines of the document in sequence. This includes the abstract, but not figure or table legends. Concision, clarity, and originality are desirable. Topics designated as acceptable as articles are described above. The formats for all submissions are as follows:

- A. **Abstract:** The first page of the manuscript should contain the title of the manuscript, the names of the authors and institutional addresses, a brief abstract (200 words or less) or important points in the manuscript, and keywords in that order.
- B. **Manuscript Text:** The introduction to the manuscript begins on the second page. It should supply sufficient background for readers to appreciate the work without referring to previously published references dealing with the subject. Citations should be reports of credible scientific or pedagogical research.

The body follows the introduction. Articles describing some type of research should be broken into sections with appropriate subheadings including Materials and Methods, Results, and Discussion. Some flexibility is permitted here depending upon the type of article being submitted. Articles describing a laboratory or class exercise that works should be broken into sections following the introduction as procedure, assessment, and discussion.

Acknowledgment of any financial support or personal contributions should be made at the end of the body in an Acknowledgement section, with financial acknowledgements preceding personal acknowledgements. If the study required institutional approval such as an Institutional Review Board (IRB), the approval or review number should be included in this section. For example, this study was approved under the IRB number 999999. The editor will delete disclaimers and endorsements (government, corporate, etc.)

A variety of writing styles can be used depending upon the type of article. Active voice is encouraged whenever possible. Past tense is recommended for descriptions of events that occurred in the past such as methods, observations, and data collection. Present tense can be used for your conclusions and accepted facts. Because *Bioscene* has readers from a variety of biological specialties, authors should avoid extremely technical language and define all specialized terms. Other than heading titles, the first word in a sentence or a proper noun, authors should not use capitalization, underlining, italics, or boldface within the text. Authors should not add extra spaces or indentations, nor should they use any hidden from view editing tools. All weights and measures must be given in the SI (metric) system.

In- text citations should be done in the following manner:

Single Author:

"... when fruit flies were reared on media of sugar, tomatoes, and grapes" (Jaenike, 1986).

Two Authors:

"...assay was performed as described previously (Roffner & Danzig, 2004).

Multiple Authors:

"...similar results have been reported previously (Baehr et al., 1999).

- C. References: References cited within the text should appear alphabetically by the author's last name at the end of the manuscript text under the heading references. All references must be cited in the text and come from published materials in the literature or the Internet. Authors should use the current APA style when formatting the reference list.

- D. Example citations are below.

(1) Articles-

(a) Single author:

DeBuhr, L. E. (2012). Using Lemna to Study Geometric Population Growth. *The American Biology Teacher*. <https://doi.org/10.2307/4449274>

(b) Multi-authored three to seven authors:

Green, H., Goldberg, B., Schwartz, M., & Brown, D. D. (1968). The synthesis of collagen during the development of *Xenopus laevis*. *Developmental Biology*, 18(4), 391--400. [https://doi.org/10.1016/0012-1606\(68\)90048-1](https://doi.org/10.1016/0012-1606(68)90048-1)

(c) Mutli-authored more than seven authors

List the first six authors than an ellipsis followed by the last author.

(2) Books-

Bossel, H. (1994). *Modeling and Simulation* (1st ed.). New York, NY: A K Peters/CRC Press. <https://doi.org/10.1201/9781315275574>

(3) Book chapters-

Glase, J. C., & Zimmerman, M. (1993). Population ecology: Experiments with Protistans. In J. M. Beiswenger (Ed.), *Experiments to Teach Ecology* (pp. 39–82). Washington, DC: Ecological Society of America. Retrieved from <https://tiee.esa.org/vol/expv1/protist/protist.pdf>

(4) Web sites-

McKelvey, S. (1995). Malthusian growth model. Retrieved November 25, 2005, from <https://www.stolaf.edu/people/mckelvey/envision.dir/malthus.html>

- E. Tables

Tables should be submitted as individual electronic files in Word (2013+) or RTF format. Placement of tables should be indicated within the body of the manuscript. The editor will make every effort to place them in as close a proximity as possible. All tables must be accompanied by a descriptive legend using the following format:

**Table 1.** A comparison of student pre-test and post-test scores in a non-majors' biology class.

- F. Figures

Figures should be submitted as **high resolution** ( $\geq 300$ dpi) individual electronic files, either TIFF or JPEG. Placement of figures should be indicated within the body of the manuscript. The editor will make every effort to place them in as close a proximity as possible. Figures only include graphs and/or images. Figures consisting entirely of text will not be accepted and must be submitted as tables instead. No figures put together using a cut and past method will be accepted. All figures should be accompanied by a descriptive legend using the following format:

**Fig. 1.** Polytene chromosomes of *Drosophila melanogaster*.

### III. Letters to the Editor

Letters should be brief (400 words or less) and direct. Letters may be edited for length, clarity, and style. Authors must include institution address, contact phone number, and a signature.

#### IV. Other Submissions

Reviews and informational submissions may be edited for clarity, length, general interest, and timeliness. Guidelines for citations and references are the same for articles described above.

#### V. Manuscript Submissions

All manuscripts are to be sent to the editor electronically and must comply with the same guidelines for text, figure and table preparation as described above. *Authors must clearly designate which type of article they are submitting (see Section I) or their manuscript will not be considered for publication.* Emails should include information such as the title of the article, the number of words in the manuscript, the corresponding author's name, and all co-authors. Each author's name should be accompanied by complete postal and email addresses, as well as telephone and FAX numbers. Email will be the primary method of communication with the editors of *Bioscene*.

Communicating authors will receive confirmation of the submission. Manuscripts should be submitted either as a Microsoft Word or RTF (Rich Text File) to facilitate distribution of the manuscript to reviewers and for revisions. A single-email is required to submit electronically, as the review process is not necessarily blind unless requested by an author. If the article has a number of high resolution graphics, separate emails to the editor may be required. The editors recommend that authors complete and remit the [Bioscene Author Checklist](#) with their submission in order to expedite the review process.

#### VI. Editorial Review and Acceptance

For manuscripts to be sent out for review, at least one author must be a member of ACUBE. Otherwise, by submitting the manuscript without membership, the corresponding author agrees to page charges. Charges will be the membership fee at the time of submission per page. Once the authors' membership or page charge status has been cleared, the manuscripts will be sent to two anonymous reviewers as coordinated through the Editorial Board. Reviewer names and affiliation will be withheld from the authors. The associate editors will examine the article for compliance with the guidelines stated above. If the manuscript is not in compliance or the authors have not agreed to the page cost provisions stated above, manuscripts will be returned to authors until compliance is met or the page cost conditions have been met. Reviewers will examine the submission for:

- **Suitability:** The manuscript relates to teaching biology at the college and university level.
- **Coherence:** The manuscript is well-written with a minimum of typographical errors, spelling and grammatical errors, with the information presented in an organized and thoughtful manner.
- **Novelty:** The manuscript presents new information of interest for college and university biology educators or examines well-known aspects of biology and biology education, such as model organisms or experimental protocols, in a new way.

Once the article has been reviewed, the corresponding author will receive a notification of whether the article has been accepted for publication in *Bioscene*. All notices will be accompanied by suggestions and comments from the reviewers. The author must address all of the reviewers' comments and suggestions using the original document and **track changes** for any consideration of a resubmission and acceptance. Revisions and resubmission should be made within six months. Manuscripts resubmitted beyond the six-month window will be treated as a new submission. Should manuscripts requiring revision be resubmitted without corrections, the associate editors will return the article until the requested revisions have been made. Upon acceptance, the article will appear in *Bioscene* and will be posted on the ACUBE website. Time from acceptance to publication may take between twelve and eighteen months.

#### VII. Revision Checklist

Manuscripts will be returned to authors for failure to follow through on the following:

- A. Send a copy of the revised article **using track changes** for text changes back to the associate editor, along with an email stating how reviewers' concerns were addressed.
- B. Make sure that references are formatted appropriately in APA style format.
- C. Make sure that recommended changes have been made or a clear explanation as to why they were not.
- D. Figures and legends sent separately, but placement in manuscript should be clearly delimited.

#### VIII. Editorial Policy and Copyright

It is the policy of *Bioscene* that authors retain copyright of their published material



# ACUBE

## ASSOCIATION OF COLLEGE AND UNIVERSITY BIOLOGY EDUCATORS

67th Annual Meeting - Where do we go from here?  
October 27-28, 2023  
Hybrid meeting via Zoom  
and in person at Rockhurst University, Kansas, MO

### Our Mission

The mission of ACUBE is to provide professional development in the realm of teaching, research, advising, and scholarship. We achieve this through our peer-reviewed publication *Bioscene*, and our Annual Meeting. Overall, members of ACUBE find each other to be one of the most valuable academic assets they have. Members of ACUBE share ideas and address the unique challenges of balancing teaching, research, advising, administration, and service. We are a supporting and mentoring community that provides professional development opportunities to:

- Develop and recognize excellence in teaching
- Incubate new and innovative teaching ideas
- Involve student research in the biology curriculum
- Advise and mentor students in and out of the classroom
- Enhance scholarship through our national, peer-reviewed journal [Bioscene](#)

### ACUBE Statement on Diversity and Equity in Biology Education

The Association of College and University Biology Educators (ACUBE) recognizes the fundamental importance of equity and inclusion in Biology and other academic fields. Justice demands equal access for all students and would-be students of the life sciences, as well as that instructors and practitioners enact curricula and conduct their scientific endeavors in ways that are maximally inclusive. Biology informs us that the resilience and adaptability of populations of organisms as well as the ecosystems in which they live hinges upon diversity within those systems. The same is true for academic communities, wherein the multiple perspectives of people from differing backgrounds contributes to creativity in approaching problems and interpreting data and serves to check assumptions and biases based on narrow-minded thinking.

ACUBE stands with our fellow scholarly organizations for biology education in our commitment to diversity and equity in the life sciences, including the National Association of Biology Teachers statement on Equity in Science Education and the Society for the Advancement of Biology Education Research statement on Solidarity with the Black Community. Members of marginalized groups deserve to feel safe no matter where they are, and they deserve to feel welcome and valued in our classrooms, laboratories, and field research sites. ACUBE is committed to striving toward better understanding of how we can foster diversity, equity, and inclusion in the life sciences and, we are committed to action toward ensuring that no one is marginalized in our classrooms or campus communities.

The ACUBE Commitment to Decolonizing Biology Education

Biology education has been monopolized by Western thought and a white patriarchal system since its inception. Biology researchers and educators must reimagine what it means to do science and how we educate future biologists. This requires a dismantling of the current higher education system in order to free the mind and spirit of all humans who wish to dream and explore their potential. ACUBE supports this endeavor, and we hope that you will join us to honor and promote all systems and ways of knowing that build knowledge through the use of counter storytelling in your educational practice.

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Thursday, October 26, 7:00 pm Central Time  
 Saturday, October 27, 1:00 pm Central Time

ACUBE's 67<sup>th</sup> Annual Meeting Keynote Speaker

Arrupe Hall 114, Friday, 1:00-2:00 pm

Keynote Speaker, Dr. Desiree Forsythe (she/her) has a background in both the biological sciences and education. She is a STEM education researcher and focuses on how to disrupt oppressions in the sciences through the use of critical, feminist, and queer theorizations. Dr. Forsythe has several lines of research including how those with privileged identities in STEM work at incorporating social justice into pedagogical and professional practice and how STEM students with marginalized identities experience and navigate STEM environments (such as classrooms, advising, office hours, etc.). She holds a Ph.D. from the University of Rhode Island. She is currently a postdoctoral fellow with the Grand Challenges Initiative at Chapman University in Orange, California.



Friday October 27 <sup>th</sup> (all times Central)	
8:30- 9:00	Tea/Coffee/Pastries served (Arrupe Hall 114)
9:00-9:30	Opening welcome (Haswell) (Arrupe Hall 114 & <a href="#">Zoom</a> )
	Morning Concurrent Sessions (presenters in person)
	Arrupe Hall 114   Arrupe Hall 116 & <a href="#">Zoom</a>
9:30-10:10	Dyer - Achieving Workload Equity Among Full-time Faculty (In person only)
10:10-10:30	Salem & Lee - Assessment of underrepresented minority (URM) pre-health student progress toward careers in health care ( <a href="#">Zoom link</a> )
10:30-11:00	Break
	Arrupe Hall 114 & <a href="#">Zoom</a>   Arrupe Hall 116 & <a href="#">Zoom</a>
11:00-11:20	Felzien - Considering student learning, equity, and stress in high stakes assessment design
11:20-12:00	Elsenpeter - Recruiting and Supporting Diverse Students in STEM
12:00-1:00	Lunch (Arrupe Hall 114)
1:00- 2:00	Keynote Speaker – Dr. Desiree Forsythe (Arrupe Hall 114 & <a href="#">Zoom</a> )
	Afternoon Sessions (presenters in person)
	Arrupe Hall 114 & <a href="#">Zoom</a>   Arrupe Hall 116 & <a href="#">Zoom</a>
2:10-2:50	Archibald & Rivers - Building inclusivity and equity into large majors and non-majors biology courses
2:50-3:10	Amin et al. - Examining the features and stability of graph knowledge among undergraduate biology students
3:10-3:50	(poster set up during this time)
4:00-4:45	Poster session (In person only)
4:45-5:30	ACUBE Business Meeting (Arrupe Hall 114 & <a href="#">Zoom</a> )
5:30-7:30	Dinner & awards presentation (Arrupe Hall 114 - bar open until 7 pm) (In person only)

Saturday October 28 <sup>th</sup> (all times Central)	
8:30-9:00	Tea/Coffee served (Arrupe Hall 114)
	Concurrent Morning Workshops and Presentations (presenters virtual)
	Arrupe Hall 114 & <a href="#">Zoom</a>   Arrupe Hall 116 & <a href="#">Zoom</a>
9:00-9:45	Bioscene editorial board meeting (open to all conference participants)
9:50-10:30	Swofford - Engaging online students with interactive Google Slides lessons
10:30-10:50	Break
10:50-11:10	Graves - Evolution of the Course Syllabus
11:10-11:30	Ankrom - Students' perceptions of the impact of gaming on course performance
11:30-12:00	Closing Remarks (Haswell) (Arrupe Hall 114 & <a href="#">Zoom</a> )
12:00-1:00	Lunch (Arrupe Hall 114)
1:00-2:30	Steering Committee Meeting (closed meeting - Arrupe Hall 116)



## Abstracts: (alphabetical by first author)

Examining the features and stability of graph knowledge among undergraduate biology students  
(20-minute presentation)

Nouran Amin, Kal Holder, Eli Meir, Susan Maruca, Joel K. Abraham, & Stephanie M. Gardner  
Purdue University

Arrupe Hall 114, Friday, 2:50-3:10 pm [Zoom link](#)

Graphing is a fundamental skill in science education, particularly for undergraduate biology students. Constructing graphs is a reflective activity that requires students to integrate concepts from biology, statistics, and spatial reasoning. Our earlier work examined introductory biology students' reasoning when constructing versus evaluating pre-made graphs using a performance-based assessment, GraphSmarts. Findings uncovered differences between the graphs students crafted and those they selected from pre-made graph sets. Furthermore, students exhibited a distinct application of statistical concepts while explaining the pre-made graphs they selected compared to the concepts used for reasoning about the graphs they made. This revealed how the application of different pieces of a students' graphing-related knowledge is influenced by the context of the graphing task. However, these analyses were based on students' written open-ended responses in the GraphSmarts assessment. To gain deeper insights into student knowledge and thinking, we have conducted think-aloud interviews with six introductory biology students to date, aligning features of their reasoning with Scherr's Conceptual Dynamics (CD) Framework--which offers a lens to reveal the features and stability of student knowledge through 5 observable properties: determinacy, coherence, context-dependence, malleability, and variability. Using the CD framework as an analytical tool, our preliminary analyses suggest that students who selected graph types they did not make exhibited either uncertainty about their own graph knowledge or the activation of a different set of knowledge. In contrast, a similar set of knowledge was activated and applied by students who created and made similar graphs. Our next steps are to continue investigating the utility of the CD framework by recruiting additional participants from diverse institution types. The products of this study will contribute to effective approaches for teaching graphing skills in the undergraduate biology classroom by highlighting important context-dependent factors that affect how students activate and apply their knowledge for graph construction.

Students' perceptions of the impact of gaming on course performance  
(20-minute virtual presentation)

Joseph Ankrom  
Binghamton University

Arrupe Hall 114, Saturday, 11:00-11:20 am [Zoom link](#)

A switch to active learning has been shown to improve students' outcomes, but buy-in to active learning, by both students and instructors, can be a major obstacle to its implementation. Studies have shown similar skill set requirements in some video games and active learning. Since approximately 75% of college students play video games regularly, exploring these similarities provides a potential avenue to improve student buy-in to active learning. To this end, we surveyed students in a variety of classroom settings from large enrollment introductory biology to Asian-American studies courses, with a varying amount of active learning. Previously, I showed that gamers in large enrollment, introductory biology courses outperformed non-gamers and demonstrated higher buy-in to active learning as indicated by the Exposure, Persuasion, Identification, Commitment scale to assess level of acceptance and commitment to specific behaviors. In this presentation, I will share the results of our qualitative coding of open-ended responses that queried students' perceptions of the impact of gaming on course performance and the similarities they see between gaming and active learning.

Building inclusivity and equity into large majors and non-majors biology courses  
(40-minute presentation)

Jenny Archibald and Trevor Rivers  
University of Kansas

Arrupe Hall 114, Friday, 2:10-2:50 pm [Zoom link](#)

Large classrooms include students with many personal histories, identities, challenges, and strengths. At the University of Kansas, 100-level classes range from 250-800+ students, with a vast range of preparedness among the incoming freshmen. To best support success and a sense of belonging in a variety of students, we strive to build our

courses in ways that deliberately consider that diversity. This starts on day one, with an emphasis on humanizing the instructors' and giving students a chance to start building connections in STEM with their peers and teaching teams. We will discuss how we approach demystifying the hidden curriculum, design assignments to maximize flexibility while maintaining academic rigor, and promote active learning in both traditional face-to-face and hybrid settings. The structure of a course can both help students accommodate learning with other complications in their lives and also make managing the course more feasible for the instructors. We encourage ACUBE participants to contribute to a conversation on ideas, difficulties, and successes.

Build-a-course workshop  
(40-minute presentation)

Rebecca Burton

Alverno College

Arrupe Hall 116, Friday, 11:20-12:00 am [Zoom link](#)

Need to design a new course? Don't just grab the textbook and use the table of contents. This workshop will be a hands-on opportunity to develop course outcomes, criteria, rubrics, and an assessment plan.

Mentoring roundtable  
(40-minute presentation)

Rebecca Burton

Alverno College

Arrupe Hall 116, Friday, 3:10-3:50 pm [Zoom link](#)

This will be an open discussion of all things faculty-related. It will be an opportunity for educators with all levels of experience to share questions, advice and insights about teaching, employment, and other topics

Active Learning in Parasitology: The Use of Learning Tools to Assess Student Comprehension of Learning Objectives  
(20-minute presentation)

J.T. Cornelius, B.S. & Joanna Cielocha, Ph.D.

Indiana University School of Medicine & Rockhurst University

Arrupe Hall 116, Friday, 10:10-10:30 am [Zoom link](#)

Parasitology is commonly taught at the graduate and undergraduate levels. Topics covered often include classification, morphology, distribution, ecology, pathology, and life cycles of various parasitic groups (e.g., Protozoa, Platyhelminthes, Nematoda, etc.). While the content being presented is important, it is essential to understand pedagogical approaches to both faculty delivery and student comprehension. Combining best teaching practices with course content can yield student mastery throughout the course. Teaching methodologies included hands-on activities, modeling, and the use of problem-based approaches. Assessment of student learning objectives is required to adequately measure student knowledge. This poses the question of how to present, assess, and quantify student understanding of core parasitology themes based on learning objectives. Through the conflation of national parasitology learning objectives, 6 central learning objectives were created for the BL4200: Parasitology course at Rockhurst University. To assess student comprehension of these objectives, a pre/post-test assessment was developed and administered to students in Fall 2021 and 2023\*. Additionally, three active learning tools were utilized to further student comprehension. Fall 2021 data provided insight on the relationship between active learning, central learning objectives, and student comprehension over the course of a semester. Fall 2023 implementation is ongoing. This mixed methods study utilizes both formative and summative assessment data to understand student comprehension. A thematic analysis was performed on student reflection data to observe the relationship between student comprehension and active learning tools. Statistical methods were employed to analyze pre/post-test assessments and overall knowledge gaps in specific learning objectives.

\*FA2023 - pre-test assessment data only

### The Impact of Visual Cue Flashcards on Short-term and Long-term Retention: Reducing Opportunity Gaps in Anatomy and Physiology

(poster: in-person only)

Jack Del Vecchio and Konnor Brennan

Rockhurst University

Arrupe Hall 114, Friday, 4:00-4:45 pm

This study aimed to investigate the effectiveness of using flashcards with visual cues to promote interleaved and spaced practice in an undergraduate anatomy and physiology course. Interleaved practice and spacing have been shown to be effective learning strategies that can improve long-term retention of information. However, their effectiveness when used in classroom studies has not been fully explored. This study compared student performance on exams between a control group that used standard flash cards and an experimental group that used flashcards with visual cues (VC) intended to promote interleaved practice and spacing. VC cards were designed to encourage students to alternate between topics and delay concepts consistently being recalled accurately. The study used a pre-test measuring the baseline knowledge of anatomy and physiology, exam scores, and a post exam survey. Results indicated that the use of VC cards to promote interleaved practice and spacing closed the expected performance gap on exam questions targeting concepts covered with the flashcards. This research contributes to the existing literature on the use of flashcards and provides valuable information for instructors looking to enhance student learning in STEM courses by promoting interleaved practice and spacing. Some students in STEM courses have not had the opportunity to build effective study skills. VC cards have the potential to be a simple intervention to allow underprepared students to bridge the opportunity gap in exam performance with students from well-resourced backgrounds.

### Achieving Workload Equity Among Full-time Faculty

(40-minute presentation, in-person only)

Jamie Dyer

Rockhurst University

Arrupe Hall 114, Friday, 9:30-10:10 am

With ever-increasing and varying responsibilities for different faculty roles, having a clearly defined policy for workload equity is essential for fairly distributing responsibilities and promoting happiness in the workplace. Different faculty roles vary tremendously, with large deviations in course enrollment numbers, teaching loads, student credit hour generation, service responsibilities, scholarship activities, advising, and leadership roles, among others. Disparities, whether actual or perceived, in workload lead to feelings of unjust working environments and dissatisfaction with careers. In order to encourage equity and retain strong faculty members, regular examination of current practices, development of policies to provide a framework for promoting equivalent workloads, and transparency can promote positive work environments, which can lead to improved faculty and student experiences. In order to facilitate the promotion of workload equity, this session will consist of an active discussion among all participants regarding current practices at the departmental and university level to ensure similar workloads among faculty members, as well as suggestions for improving fair distribution of responsibilities among faculty members.

### Recruiting and Supporting Diverse Students in STEM

(40-minute roundtable)

Ryan L. Elsenpeter, Joanna J. Cielocha, Mayuri Gilhooly, Michael C. Marvin

Rockhurst University

Arrupe Hall 114, Friday, 11:20-12:00 am [Zoom link](#)

Our team was recently awarded a National Science Foundation S-STEM grant to support low-income/high-achieving STEM students through scholarships and a robust university-wide network. This grant will focus on students pursuing B.S. degrees in Biology, Chemistry, and Physics at a small, private, liberal arts institution. As part of our grant, STEM faculty will serve as mentors and advisors for students in the program. STEM faculty mentors will participate in professional development around diversity, equity, and inclusion as well as mental health first aid training. These professional development activities will help prepare faculty for working with a more diverse student population. Faculty mentors will also develop programming to expose students to various career and graduate school options. A pre- and post-assessment will be administered to both STEM faculty and students. Student assessments will measure a sense of belonging in their cohorts, in college, and in STEM. We are in the early stages of implementation and recruitment and are seeking feedback and discussion regarding faculty and peer mentorship of our targeted student cohort population.

Considering student learning, equity, and stress in high stakes assessment design  
(20-minute presentation)  
Lisa Felzien, Connor Senseney, Olivia Wilmsmeyer  
Rockhurst University  
Arrupe Hall 114, Friday, 11:00-11:20 am [Zoom link](#)

The COVID pandemic caused STEM faculty who emphasized traditional high stakes testing to consider different assessment methods. During the shift to online teaching, faculty often designed assessment formats that required different levels of thinking and did not strictly adhere to the timing constraints of a single class period. The dilemma of returning to traditional methods or continuing to adapt remains. In this presentation, we will critique the progression of assessment strategies in an upper-level immunology course, taught by one of the authors and taken by the other two authors. We will share an overview of research findings on test anxiety and the impact of traditional testing on student learning, consider work on biomarkers for stress in test settings, and discuss how alternatives to testing may support deeper learning and help to level the playing field for diverse groups. Finally, we will share how immunology students have been involved in designing experiments that contribute to understanding the impact of in-class testing on the stress response.

Sentiment analysis to determine student perceptions in Statistics for Health Sciences  
(poster: in-person only)  
Mayuri Gilhooly and Emily Dale  
Rockhurst University  
Arrupe Hall 114, Friday, 4:00-4:45 pm

Student survey data are very valuable, yet summarizing free response data can be challenging. This study proposes a way to summarize students' free responses from a Student Assessment of their Learning Gains (SALG) survey in Statistics for Health Sciences. SALG data for two semesters were analyzed using sentiment analysis methods, Valence Aware Dictionary for sEntiment Reasoning (VADER) and Affective Norms for English Words (AFINN). Sentiment analysis is a valuable tool for extracting meaningful insights from text data and understanding the sentiments and opinions expressed by individuals or groups. Using sentiment analysis in Python, students' text data were categorized into positive, neutral, and negative statements based on sentiment scores assigned to words in the dataset. Moreover, a comparison between the two methods (VADER and AFINN) is also generated. This proposed method aims to provide a way for an instructor to identify negative text responses faster and determine ways to address them.

Evolution of the Course Syllabus  
(20-minute virtual presentation)  
James Graves  
University of Detroit Mercy  
Arrupe Hall 114, Saturday, 10:50-11:20 am [Zoom link](#)

A syllabus was a label with some written information in the distant past. More recently, it was basically a list of topics in a college course. The purpose of this study was to examine how the course syllabus has changed over the years. Syllabi for a microbiology lecture course that were made and saved since the availability of the office personal computer were compared. Many characteristics of a microbiology syllabus would probably be the same as a syllabus for other courses. In the past few decades, the length of the syllabus greatly increased. Before the year 2000 the length of the syllabus was only one page. This appeared to have been common at that time. The one-page syllabus contained information about the instructor, grading, text, course objective and a list of lecture topics. After the millennium, the length of the syllabus quickly increased to four pages. Information was increased in existing syllabus parts. New parts such as images, prerequisites, attendance, school support, academic integrity, disabilities and Title IX were added. Moreover, links to the text publisher, professional organizations, Blackboard and websites within the institution were provided. In year 2018, just before the coronavirus pandemic, the syllabus increased in size to seven pages. Copyright, classroom conduct, bias-motivated Incidents and religious observances were included. At the present, there is interest in showing an increased number of course objectives, many tables, the presence of growth mindset and student-centered learning in the syllabus. The appropriate length for a college syllabus may be a matter worth considering.

Exploring the immune system using mini cases and HHMI BioInteractive  
(40-minute presentation)

Melissa Haswell

Delta College

Arrupe Hall 220, Friday, 9:30-10:10 am [Zoom link](#)

This workshop will explore narrative mini cases alongside the HHMI BioInteractive Immune System learning module. The activity introduces the cells of the immune system and walks through the timeline of a typical immune response while comparing it to several real-life immune reactions. Participants will identify and explain the role of memory cells when the body responds to various common antigens by interpreting graphs and images.

Using songbird nestboxes to engage Ornithology and summer research students in CUREs  
(poster: in-person only)

Julie Jedlicka

Missouri Western University

Arrupe Hall 114, Friday, 4:00-4:45 pm

Avian foraging is a focus of my research, and I incorporate Course-based Undergraduate Research Experiences (CUREs) for students in my Ornithology class. I am presenting an example of a CURE experience that can be replicated across other classes and universities. Student research is an integral part of studying avian nest site selection and reproductive success in the 62 songbird nest boxes I established on campus. Nest boxes of 2 different types (predator proof and standard nest boxes) were established in pairs across Missouri Western State University's 700-acre campus in 2022. We analyzed whether native, cavity-nesting birds exhibited a preference for a specific box type and whether number of eggs laid and fledglings produced differed between box types. We recorded the species type, number of eggs laid, number of live young and dead young, nest status, adult and young status and parasitic presence. Students taking Ornithology at MWSU in Spring semester are assigned a pair of nest boxes to begin monitoring after Spring Break. Students were responsible for checking the boxes twice a week, data collection, and data recording. Incorporating students lead to a feeling of student ownership and responsibility over the research. Some students then decided to continue with the project and collect data after the course ended, throughout the summer, leading to more faculty-student research opportunities and engagement.

Plankton exploration at Swan Lake Iris Gardens: A mentoring summer research project for students  
(10 minute virtual presentation)

Dr. Dan Kiernan and Dr. Pearl Fernandes

University of South Carolina Sumter

Arrupe Hall 116, Saturday, 11:10-11:20 am [Zoom link](#)

The project on Plankton Exploration at Swan Lake Iris Gardens was undertaken to provide students on a two-year campus with a summer research experience using the scientific method learned in biology classes and get excited about research in biology. Freshwater ecosystems depend upon the variety and quantity of phytoplankton and zooplankton to survive. Students, who enrolled in the summer research project, identified some common phytoplankton and zooplankton at Swan Lake Iris Gardens, a freshwater ecosystem near the USC Sumter campus. Further, they obtained an indication of what the presence of certain plankton means for the health of this aquatic system. Students collected water samples at varying depths from the lake twice a week in summer for a six-week period. In the laboratory, the students identified the phytoplankton and zooplankton and discovered through their results that Swan Lake is a healthy ecosystem. By creating a brochure on the diversity of plankton, students learned the importance of connecting science with the community. Overall, students gained experience in scientific research and the project helped them solidify their plans to continue to pursue research in science.

How do we get our biology students (and us) to find the joy in our classes?  
(40-minute virtual presentation)

Anne McIntosh  
University of Alberta, Augustana Campus  
Arrupe Hall 116, Saturday, 9:50-10:30 am [Zoom link](#)

Since our return to the classroom post-COVID, I have struggled with engagement from the students in my classes. I teach ecology, plant biology, and statistics for the natural sciences courses and sometimes I feel like the economics professor in Ferris Bueller's Day Off calling out "Anyone... anyone". To combat this and help my students (and me!) find the joy back in the classroom, I have learned some new approaches that I am using in my classes. In this 40-minute workshop I will share some of the things I am doing (e.g., using interactive software questions, incorporating universal design learning principles, and alternative types of assessment) to help create an engaging classroom experience. We will also share ideas from workshop participants about tools and techniques that you have adopted that are creating a positive learning environment in your biology classrooms.

Improving online discussion engagement: Lessons learned in a developmental biology course  
(20-minute virtual presentation)

A.B. Munley and A.M Driver  
The University of Scranton  
Arrupe Hall 116, Saturday, 10:50-11:10 am [Zoom link](#)

With the onset of the pandemic, teaching through online approaches has increased. With this increase comes the question of whether these approaches are effective in generating meaningful interactions. For this session, recent work from a student-faculty partnership addressing student interactions during online discussion activities in a developmental biology course will be presented. Primary issues including student participation and engagement will be discussed. In addition, approaches to begin addressing these issues will be shared. Questions will also be posed to further discuss ways in which the student experience can be improved using online discussion activities within biology courses.

Interdisciplinary biophysics project with fruit fly larvae  
(poster: in-person only)  
Fernando Pascacio, Thomas Butler, and Mayuri Gilhooly  
Rockhurst University  
Arrupe Hall 114, Friday, 4:00-4:45 pm

This project aims to bridge the gap between physics and biology by utilizing an interdisciplinary approach to teaching and learning. We aim to address an interdisciplinary biophysics project that involves students with different backgrounds. This project consists of 3 main stages to study the motion of fruit fly larvae. 1) Hardware set up involves in setting up the camera, LED light sources and preparing a surface for the larvae to crawl. 2) Preparation of fruit fly larvae. 3) Collecting videos of larvae motion. Through video and still image analysis, students can study the random walk of fruit fly larvae with physics principles such as vectors and kinematics. This interdisciplinary approach can provide students an opportunity to make connections between disciplines. Our emphasis will be on hardware setup and questions that can be investigated with this biophysics project.

Assessment of underrepresented minority (URM) pre-health student progress toward careers in health care  
(20-minute presentation)

Laura Salem and Annie Lee  
Rockhurst University  
Arrupe Hall 114, Friday, 10:10-10:30 am [Zoom link](#)

Rockhurst University has a large population of students interested in a wide variety of healthcare careers. As the campus continues to diversify the student population, resources need to be developed to support underrepresented minority (URM) students in achieving their career goals. This talk will describe the development of advising resources for URM pre-health students and the redesign of the Pre-Health Canvas page to include modules and self-assessments that allow students to monitor their progress toward their health professional goals.

Engaging online students with interactive Google Slides lessons  
(40-minute virtual presentation)  
Lynn Swafford  
Wayne Community College  
Arrupe Hall 114, Saturday, 9:50-10:30 am [Zoom link](#)

It is challenging to engage with students in online asynchronous classes and difficult to know what concepts they are struggling with. To address these issues, I have created interactive Google Slides lessons to present the content in my General Biology for non-majors course. These lessons are self-paced, have interactive practice questions, and provide students with the opportunity to discuss concepts with me and their classmates. Come learn how these lessons are structured, what students like about them, and how you can make your own.

Bioscience editorial board meeting  
(40 min virtual presentation)  
Robert Yost, editor-in-chief, Bioscience  
Indiana University-Purdue University Indianapolis  
Arrupe Hall 114, Saturday, 9:00-9:45 am [Zoom link](#)

This meeting is open to all attendees who are interested in ACUBE's journal, Bioscience: Journal of College Biology Teaching, either as an author, reviewer, or reader. Come hear the annual Bioscience report on publications and learn about becoming involved with Bioscience. This session will provide information on journal submissions for 2023 along with an overview of the submission and review process. An open discussion is planned on ways to increase submissions to and expand the audience of Bioscience. Attendees are encouraged to put forth their ideas for the journal during this session.