

POINT LOMA NAZARENE UNIVERSITY

**Mechanisms and Outcomes of Genetic Variation as a Threshold Concept for
Understanding Evolution by Natural Selection.**

A thesis submitted in partial satisfaction of the
requirements for the degree of

Master of Science

in General Biology

by

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The thesis of Karoly Tippetts-Russell is approved, and it is acceptable in quality and form for publication:

Chair

Point Loma Nazarene University

2015

I dedicate this thesis to my witty husband, Patrick,
and our wonderful children, Colby and Finley,
for loving and supporting me through it all.
I could not have accomplished this without you.

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Abstract

Higher education science researchers have postulated that understanding variation is a threshold concept in biology. Variation is evident in all living organisms yet is often overlooked as important or impactful in the process of natural selection. This study explores the idea that understanding the mechanisms and outcomes of genetic variation is a threshold concept for understanding the consequences of that variation, in particular evolution by natural selection. A group of undergraduate students (N=104) from a small, liberal arts university in California answered questions about the mechanisms of genetic variation, the outcomes of genetic variation and the consequences of genetic variation to determine which, if any, of the topics were mastered by students. All students had recently completed one of three introductory biology courses that taught evolution by natural selection. One of the courses was composed of science majors while the other two courses were comprised of non-science majors. If students earned an 80% or higher average on any one of the topics, the author considered that mastery had been achieved. A Kendall's tau was calculated for each pair of topics: mechanisms/outcomes, outcomes/consequences and consequences/mechanisms. It was determined that there was only a statistically significant positive correlation between non-science majors understanding of the mechanisms and consequences of genetic variation. These exploratory findings suggest that mechanisms and outcomes of variation may not be a threshold concept for consequences of variation for this participant group.

Introduction

The process of evolution by natural selection is a paramount concept for introductory undergraduate life science students to comprehend as evidenced by the inclusion of evolution in the International Baccalaureate (IB) Biology curriculum (Conley & Ward, 2009) and as the first “Big Idea” in the Advanced Placement (AP) Biology curriculum framework (College Board, 2011). Additionally, evolution is listed as the first “Core Concept for Biological Literacy” in Vision and Change, a guiding framework for undergraduate biology education (Brewer & Smith, 2011). All three programs focus on learning expectations for students who choose to pursue undergraduate biology courses at universities in the United States. Students are introduced to the concept of evolution by natural selection beginning in middle school and are expected to fully understand its key mechanisms in high school, in California (California Department of Education, 2013). However, undergraduate biology students continue to have a difficult time constructing a scientific understanding of the processes involved in evolution. Students struggle to relinquish naïve conceptions in favor of scientifically accurate ideas (Mbajiorgu, Ezechi & Idoko, 2007), make systems-level connections (Dauer, Momsen, Speth, Makohon-Moore & Long, 2013), and incorporate the molecular basis of variation into their understanding of evolution by natural selection (Speth, Shaw, Momsen, Reinagel, Le, Taqieddin & Long, 2014). Identifying specific prior knowledge requisite for comprehending the intricacies of evolution by natural selection would aid instructors in more successfully guiding undergraduate students in the knowledge construction process.

Each student builds knowledge in a unique manner, yet, it is widely accepted that there are some foundational information pieces that students must possess in order to understand more complex concepts. This is certainly true if students are to understand the complexities of

evolution by natural selection. One key piece of information for understanding the theory of evolution is that natural selection acts on variation, and therefore students need to know the meaning of variation, how it arises in living organisms and recognize that variation exists in all species. The biological basis for variation, resulting in the diversity of life found on Earth, occurs at the genetic level. Discrete changes (mutation, recombination) in the genome (DNA) generate genetic variation among individuals of a species. That genetic variation may be expressed as phenotypic variation upon which selection occurs within a population. Cumulative selection for the most advantageous of these slight individual differences results in changes to populations over time, ultimately leading to descent with modification and speciation. Consequently, one could argue that understanding the causes of variation at a molecular level and succeeding phenotypic changes are foundational for explaining how the many forms of life emerged over time. This study assesses the impact of undergraduate student understanding of the mechanisms of genetic variation (mutation, genetic recombination) and outcomes of genetic variation (genotypic/phenotypic changes) on one's understanding of biological evolution.

The purpose of this study was to test the hypothesis that a scientific understanding of both the mechanism (at a molecular level) and the outcomes (individual level) of variation are foundational for understanding the results and consequences of variation (population/community level). In other words, is it possible to accurately explain the immense biodiversity found on Earth resulting from evolution without possessing a knowledge structure of where variation begins at the genetic level and the ensuing phenotypic changes? If a scientifically accurate understanding of the mechanism and outcome of molecular variation opens up a new and previously inaccessible way of thinking about the consequences of variation in a population and

community, e.g. evolution, then students will demonstrate an inaccurate, incomplete or naïve conception of evolution until the former ideas are accurately understood.

Literature Review

Theoretical Framework

Learning is an active process of internal knowledge construction (Piaget, 1963, reprinted 2003), and students' understandings are "composed of many interrelated elements that can change in complex ways" (Smith, diSessa & Roschelle, 1994, p. 117). This idea of individual knowledge building is demonstrated when students have the 'aha' moment in which they integrate new knowledge into an existing cognitive structure or create a new and unique schema. Students' internal knowledge structures can be uncovered through specific questions and tasks that require students "to describe their reasoning" (Stewart, Cartier & Passmore, 2005, p. 556). As such, the literature base on student conceptions of natural selection and genetic variation informed this study.

Natural Selection

As alluded to in Theodosius Dobzhansky's 1973 essay "Nothing in biology makes sense except in light of evolution," the study of evolution is a unifying theme that connects the many fields within biology. Later, Ernst Mayr partitioned "Darwin's evolutionary paradigm into five theories" (1991, p. 36), the last of which was Natural Selection. Many education studies have examined undergraduate knowledge of evolution by natural selection, and numerous assessments have been developed to determine undergraduate students' conceptions about evolution by natural selection (Anderson, Fisher & Norman, 2002; Bishop & Anderson, 1990; Nehm & Reilly, 2007; Nehm & Ridgeway, 2011; Opfer, Nehm & Ha, 2012). The Concept Inventory of Natural Selection (CINS), originally published by Anderson *et al.* (2002), and revised in 2013 (Evans & Anderson, 2013) is a closed-response, diagnostic tool utilized by college biology

instructors to identify alternative conceptions students may possess. The closed-response nature of the CINS means it can be implemented with many students simultaneously to assess understanding of natural selection before, during and/or after instruction. Nehm and colleagues have published numerous studies focusing on participant understanding of natural selection or evolution (Nehm & Reilly, 2007; Nehm & Ridgeway, 2011; Opfer, Nehm & Ha, 2012). These studies have produced open response instruments including the ACORNS (Assessing Contextual Reasoning about Natural Selection) (Opfer *et al.*, 2012). Table 1 contains a list of common alternative conceptions about natural selection identified by both Anderson and Nehm.

| Table 1 | |
|---|--|
| <i>Common alternative student conceptions about natural selection</i> | |
| <i>Author (Year)</i> | <i>Common Alternative Conceptions of Students</i> |
| Anderson <i>et al.</i> (2002)* | <ol style="list-style-type: none"> 1. Organisms can always obtain what they need to survive. 2. Fitness is equated with strength, speed, intelligence or longevity. 3. Organisms can intentionally become new species over time. 4. Mutations are intentional: an organism tries, needs, or wants to change genetically. |
| Nehm & Schonfeld (2008) | <ol style="list-style-type: none"> 1. Use and disuse 2. Inheritance of acquired traits 3. Intention/need |
| Nehm & Ha (2011) | <ol style="list-style-type: none"> 1. Needs/goals 2. Use and disuse 3. Intentionality 4. Adapting and acquiring traits 5. Deliberate energy allocation 6. Pressure as a direct cause of change |
| Nehm & Ridgeway (2011) | <ol style="list-style-type: none"> 1. Needs drive evolutionary change. 2. Pressure forces change. 3. Use and disuse explain change. 4. Acclimation is the same as adaptation. 5. Inheritance of acquired traits. 6. Intentionality explains change. |
| <i>Note.</i> *The four most commonly identified alternative conceptions are provided. | |

Mayr (1982) described two main beliefs that lead people to misunderstand or fail to accept natural selection: teleology and essentialism. Teleology is the belief that traits are formed for a specific purpose by a creator. Essentialism is the belief that individual variability between individuals within a species is negligible and of no consequence. In fact, without small variations among individuals there is no difference upon which natural selection can act. In a formative paper on student conceptions of natural selection, Bishop and Anderson (1990) noted that “students possessing naïve conceptions did not view variability as important to evolution” (p. 422). While this is one of many studies focused on the process of evolution, there has been little work on understanding what students know specifically about the mechanisms and outcome of variation. Yet, among biologists it is accepted that variation plays an essential role in the process of natural selection.

Understanding Variation

The mechanisms of genetic variation are primarily through the processes of mutation and genetic recombination. Mutations can take one of the following forms: missense point, nonsense point, silent point, insertion, deletion or duplication. Genetic recombination can include crossing over of homologous chromosomes during Metaphase I, independent assortment of homologous chromosomes and random fertilization. Any one of these chromosome ‘shuffles’ of existing alleles can result in new combinations that form a unique offspring. Consequently, both mutation and genetic recombination are crucial maintaining the variation found among individuals of the same species. Some cognitive research however, has found that in evolution education “most individuals appear to doubt that species members can, and do, vary on virtually all dimensions” (Shtulman & Schulz, 2008). In other words, many students deny that traits characteristic of a species vary among individuals of the species, e.g. the relative size of ears on individual bat-

eared foxes. These researchers also concluded that “it seems likely that students must recognize the prevalence of within-species variation before they can learn a concept predicated on this recognition” (Shtulman & Schulz, 2008), i.e. evolution. To ultimately understand evolutionary theory, a thorough grasp of the genetic underpinnings of variation seems necessary.

Molecular Genetics

Kalinowski, Leonard and Andrews (2010) argue in their essay that students would have increased comprehension of evolution if students were explicitly presented with the genetic basis underlying evolution. A number of studies have identified and/or attempted to address biology students’ challenges in comprehending various foundational aspects of genetics (Dauer *et al.*, 2013; Duncan & Reiser, 2007; Smith, Wood & Knight, 2008; Speth *et al.*, 2014). Confusion about one of the origins of variation, mutation, was one of a few substantial conceptual difficulties noted by Smith and Knight (2012) when creating the Genetics Concept Assessment for undergraduates. One area of challenge was “the nature of mutations and their effects” (Smith & Knight, 2012, p.24) and specifically that “mutations do not have to affect proteins to be called mutations” (p.24). Without a holistic understanding of the mechanisms and outcomes of genetic variation, students may “use surface features to construct their explanations” (p.4) and “do not recognize underlying concepts common” (p.4) to many types of organisms (Prevost, Knight, Smith & Urban-Lurain, 2013). For example, Prevost *et al.* (2013) noted a disparity between how often students identified mutations as sources of new alleles in animal and bacterial populations; with mutations defined significantly more often in animal populations. It is incumbent upon instructors to determine if students have constructed a foundational understanding of the molecular processes that result in slight phenotypic variations upon which natural selection can act.

Threshold Concepts

Just as a keystone is needed to transform a decorative arch into a load bearing structure, some concepts are essential for students to understand if they are to recognize subject matter complexities and progress in their learning. Meyer and Land proposed the idea of *threshold concepts* in higher education research in a 2003 report for the Enhancing Teaching-Learning Environments in Undergraduate Courses Project. Meyer and Land (2003) identified threshold concepts as causing a shift in perspective (transformative), making it difficult to return to the previous way of thinking (irreversible), exposing previously hidden interrelatedness (integrative), sometimes bordering new conceptual areas (bounded) and causing cognitive dissonance (troublesome). According to Meyer and Land, threshold concepts are “core concepts that once understood, transform perceptions of a given subject” (p.5). When understood, threshold concepts are “akin to a portal, opening up a new and previously inaccessible way of thinking about something” (p.1). For example, once Dorothy sees the man behind the green curtain she can never again think of Oz as all powerful; her knowledge forever transformed her perspective. If threshold concepts must be understood to continue knowledge acquisition and accommodation, these concepts should help inform the learning progressions written for content areas. Threshold concepts could be seen as important check points in learning progressions where instructors assess student understanding. Threshold concepts have been proposed in many scientific fields including economics (Davies & Mangan, 2007), chemistry (Park & Light, 2008), biochemistry (Loertscher, Green, Lewis, Lin & Minderhout, 2014) and biology (Taylor & Meyer, 2010).

Threshold Concepts in Biology

It has been postulated that threshold concepts are especially difficult to identify in biology because “it is perhaps easier in the discipline of biology for students to achieve ‘success’ through learning ritual responses to definitional questions which have become instituted and resistant to change, while still maintaining significant misconceptions about key biological concepts” (Ross, Taylor, Hughes, Kofod, Whitaker, Lutze-Mann & Tzioumis, 2010). Some threshold concepts proposed in biology are hypothesis testing (Taylor & Meyer, 2010), variation, probability and randomness, proportional reasoning, predictive reasoning, and thinking at the subcellular level and integrating these observations with the macroscopic (Ross *et al.*, 2010). The focus of this study incorporates two of the threshold concepts offered by Ross *et al.* (2010): variation and integration between the subcellular and macroscopic levels.

The subcellular level considered in this study—mechanisms of variation—is at the molecular level, and refers to changes in the DNA structure and the allelic combinations found in an organism, respectively. These subcellular changes can result in minute variations between individuals (outcomes) that confer a survival or reproductive advantage to one individual over another. It is these slight changes that, over hundreds of thousands of years, result in one population of a species branching out into numerous phylogenetic lines; some that thrive and continue changing, others that disappear in extinction and still others that do not change. I hypothesize that to have a thorough understanding of evolutionary change, which is the consequence of variation acted upon by natural selection, one must have a detailed understanding of the mechanisms that caused genetic variation and outcomes of that variation (Figure 1).

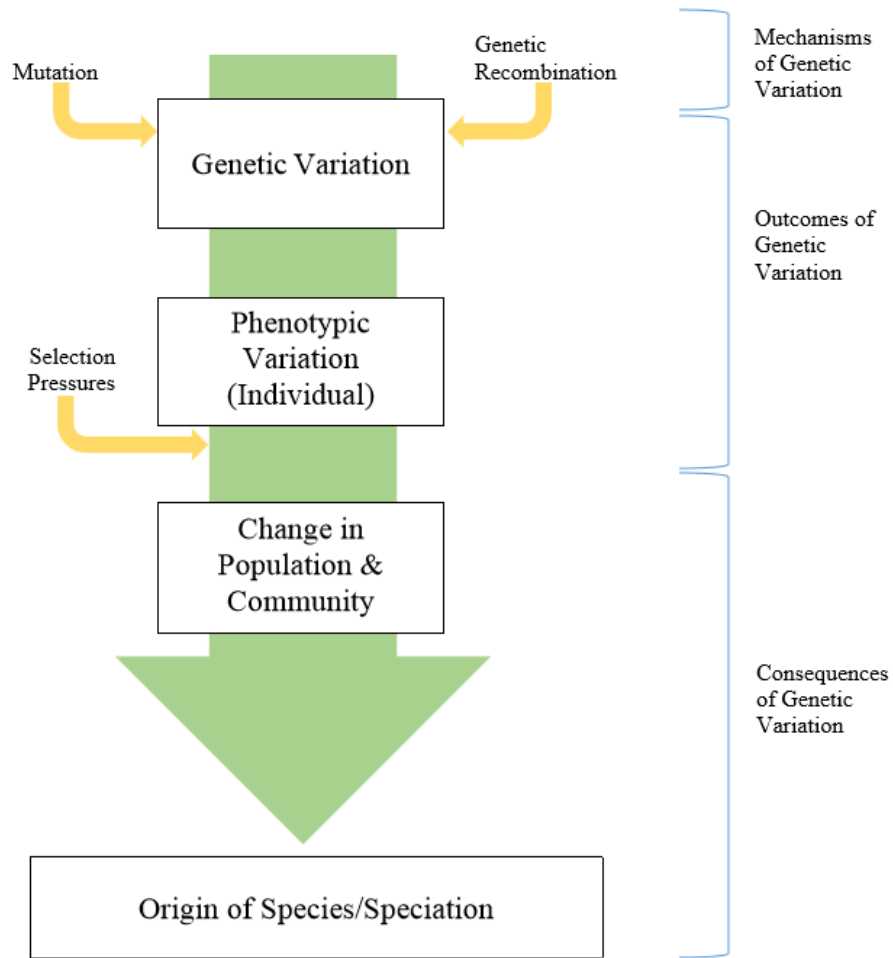


Figure 1. Connection between the mechanisms of variation, outcomes of variation and the consequences of variation.

Research Question

This study will answer the following research question: Is a molecular level understanding of the mechanisms and outcomes of variation a threshold concept for understanding the process of evolution by natural selection for college science students?

Methodology

Research Design

Modified from Creswell and Plano Clark's (2007) triangulation design, a convergence model was utilized as the mixed methods approach to answer the research question (Figure 2).

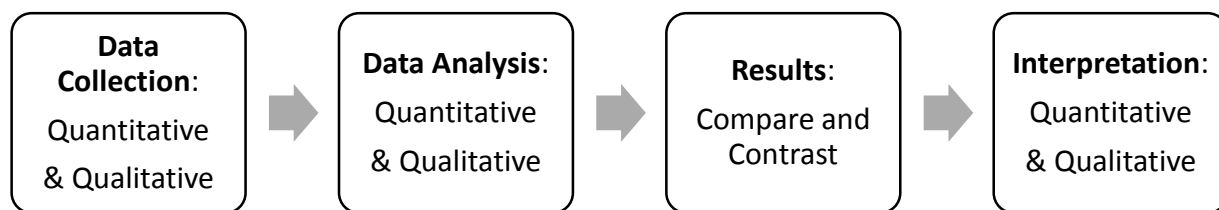


Figure 2. Convergence model of triangulation mixed methods design. This figure illustrates the mixed methods approach used in this study.

Students were given open response (qualitative) and closed response (quantitative) questions to determine what they understood about how variation that begins at the molecular level of an individual organism ultimately results in the great biodiversity found on Earth. Specifically, students were asked to identify the mechanisms of genetic variation, the outcomes of genetic variation, and the consequences of genetic variation.

Instrument Details

Science education research has not produced a single assessment to evaluate student understanding of the mechanisms, outcomes, and consequences of genetic variation. Consequently, this study predominantly utilizes questions from other published assessments to gauge student knowledge of these topics. A pilot study refined an instrument to accurately assess undergraduate life science majors' understanding of molecular level variation. The questions related to subcellular variation were combined with more questions that assessed phenotypic variation and understanding of evolution to create the Mechanism, Outcomes, and Consequences of Variation Instrument (MOCVI) used in this study (Appendix A).

Numerous studies have identified variables that can impact how students answer questions and whether the answers truly reflect student knowledge. Opfer *et al.* (2012) stated that evolution assessments should contain a diversity of evolutionary scenarios to determine student

understanding as surface features of questions can impact novice students' reasoning about evolution. Another consideration when choosing questions to elicit student understanding of evolution by natural selection is organism type. As shown in Nehm and Ha (2011) and Heredia, Furtak and Morrison (2012) students may answer questions differently based on the organisms chosen for the questions. Additionally, Nehm and Ha (2011) found that more scientifically-accurate concepts were offered when students were asked about *within* species contexts versus *between* species concepts. Lastly, attention was given to the number of questions about each topic on the assessment. Anderson *et al.* (2002) studied ten scientific concepts related to natural selection and used two closed-response questions to assess student understanding of each of those concepts. Bowling, Acra, Wang, Myers, Dean, Markle, Moskalik and Huether (2008) used one or two closed-response questions to assess each of the seventeen genetics subtopics important for genetics literacy. Klymkowsky *et al.* (2010) used between one and seven questions to assess specific concept areas on the Biology Concept Inventory, and Smith, Wood and Knight (2008) used between one and five questions to assess each learning goal in their study of undergraduate understanding of genetics.

Informed by the above literature, the following considerations were made when creating the instrument. A fruit fly, an elm tree, and a finch were chosen as focus creatures in the open response section because these organisms should be familiar to undergraduate students, yet also represent three vastly different organisms. Careful thought was also given to the types of questions chosen for this study. A gain of function question was chosen to assess understanding of evolution by natural selection in the open response section as trait loss questions tend to elicit more naïve ideas (Nehm & Ha, 2011). Moreover, organismal questions utilized within species contexts to prompt fewer naïve ideas. Furthermore, this current study used at least three or more

questions on each of the three levels of variation to examine student understanding. Lastly, both open and closed response questions about the same topic are employed to ensure that a complete picture of student understanding is revealed. The origin of each question is provided in the instrument found in Appendix A.

The MOCVI was created by selecting questions from the following published assessments: 2014 Advanced Placement Biology Scoring Guidelines, Alberts *et al.* (2013), Anderson and Evans (2013), Bowling *et al.* (2008), Klymkowsky, Underwood and Garvin-Doxas (2008), Opfer, Nehm and Ha (2012) and Smith, Wood and Knight (2008). Table 2 shows the alignment of each instrument task with the topics it covers. The three open response questions were adapted from the 2014 Advanced Placement Biology Scoring Guidelines, Opfer, Nehm and Ha (2012) and Dr. Jan Batzli at the University of Wisconsin, Biology department (personal communication, 2015). Participants wrote out the answers to the three open response questions. The closed response questions consisted of twelve multiple choice and multiple true/false questions adapted from the following published assessments: Alberts *et al.* (2013), Anderson and Evans (2013), Bowling *et al.* (2008), Klymkowsky, Underwood and Garvin-Doxas (2008) and Smith, Wood and Knight (2008).

| Table 2 | | | | |
|---|---------------------------------|---|--|--|
| <i>Question Type and Content on the Instrument</i> | | | | |
| <i>Question Type: OR-open response CR-closed response</i> | Maximum Point Value by Question | <i>Topic of Knowledge Assessed</i> | | |
| | | <i>Mechanisms of Variation (mutation & genetic recombination)</i> | <i>Outcomes of Variation (change in individuals & populations)</i> | <i>Consequences of Variation (origin of species)</i> |
| OR 1a | 5 | | X | |
| OR 1b | 5 | X | | |
| OR 1c | 5 | | | X |
| OR 2 | 10 | X | | X |
| OR 3a | 5 | X | X | X |
| OR 3b | 5 | | X | X |
| | | | | |
| CR 1 | 3 | X | | |
| CR 2 | 3 | | X | |
| CR 3 | 3 | | | X |
| CR* 4a | 1 | X | | |
| CR* 4b | 1 | X | | |
| CR* 4c | 1 | X | | |
| CR* 4d | 1 | X | | |
| CR 5 | 3 | | X | |
| CR 6 | 3 | X | | |
| CR 7 | 3 | | X | |
| CR 8 | 3 | | X | |
| CR* 9a | 1 | X | | |
| CR* 9b | 1 | X | | |
| CR* 9c | 1 | X | | |
| CR* 9d | 1 | X | | |
| CR* 10a | 1 | X | | |
| CR* 10b | 1 | | X | |
| CR* 10c | 1 | | X | |
| CR* 10d | 1 | | | X |
| CR* 11a | 1 | | | X |
| CR* 11b | 1 | | | X |
| CR* 11c | 1 | | | X |
| CR* 11d | 1 | X | | |
| CR 12 | 3 | X | | |

Note. CR-multiple choice questions. CR*-a set of true/false statements about a scenario

Study site & participants

The study was conducted at a small, Christian liberal arts university in southern California. The undergraduate enrollment of the university was roughly 2,600 and over 60% female. The student population is 63% White, 22% Hispanic, 7% Asian American/Native American/Pacific Islander, 6% Multi-Race and 2% Black.

The participants in this study initially consisted of one-hundred seventeen first- and second-year undergraduate students. Students were enrolled in one of the following four-unit courses in Spring 2015: Human Biology and Bioethics, Introduction to Biology, or Ecological and Evolutionary Systems. All three courses meet the Life Science General Education requirement at the university, but the Ecological and Evolutionary systems course is primarily populated with freshman biology majors, while students in the other courses are non-science majors. In addition to different student populations there were also different instructors for each course.

The Human Biology and Bioethics course was taught by a third-year faculty member of the university. This instructor has four years of total teaching experience. Introduction to Biology was taught by a fifteen-year veteran instructor of the university with nineteen years of total teaching experience. Ecological and Evolutionary Systems was taught by an Associate Professor who was an eight-year veteran of the university. This instructor has twenty-four years of total teaching experience.

Data collection

A diagnostic approach was implemented as undergraduate students responded to open response and closed response questions to determine their understanding of the mechanisms, outcomes and consequences of genetic variation. The three open response and twelve closed

response questions were included on the final exams taken by all students enrolled in Human Biology and Bioethics, Introduction to Biology, and Ecological and Evolutionary Systems. The order of questions on each of the final exams was unique meaning that no two classes had the same questions in the same order. This should limit question order bias. Students were assigned a numerical identifier. No personal information was shared with the researcher as only the numerical identifier was included with the responses (PLNU IRB #1405).

Analysis

Student responses were collected and assigned a numeric value to retain participant anonymity. Open response replies were coded for scientifically-based understandings and evaluated using adapted reasoning rubrics (Andrews, Leonard, Colgrove, & Kalinowski, 2011; College Board, 2014; Opfer, Nehm & Ha, 2012). As found in Appendix B, each open response question had a single, unique coding rubric adapted from a single reasoning rubric. Open response questions 1a, 1b, 1c, 3a and 3b were worth a maximum of five points. Participants could earn ten points for open response question 2.

Closed response answers were evaluated for scientifically accurate answer choices. Students could earn a score of three on all closed response, multiple choice questions whereas each scientifically accurate multiple true/false answer choice was worth one point. After all open and closed response questions were assessed, I compared each participant's data in the three categories: mechanism, outcomes, and consequence of genetic variation. Two specific conditions had to be satisfied to support the hypothesis that understanding the mechanisms and outcomes of variation is a threshold concept for the consequences of that variation, e.g. evolution. First, the students who accurately answered the questions about evolution had to accurately answer questions about mutations/genetic recombination and phenotypic changes. Additionally, the

students who incorrectly answered evolution questions also had to incorrectly answer questions about the origin and mechanisms of variation.

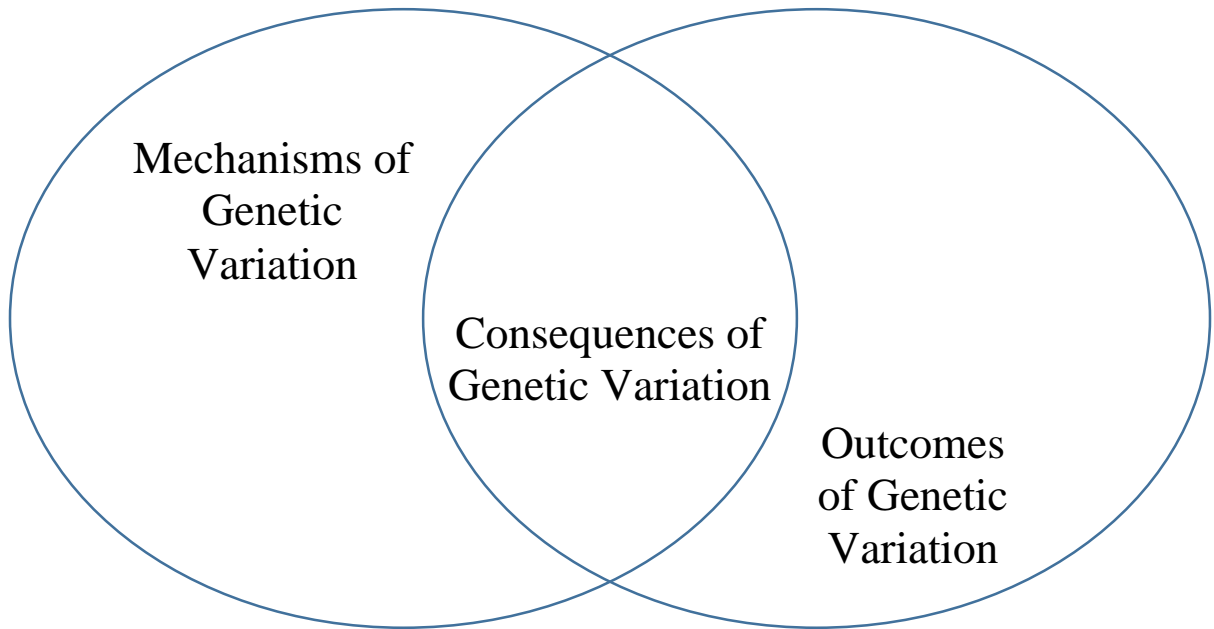


Figure 3. Student response scenario that supports the hypothesis. Without understanding of mechanism **and** outcome, students will not understand consequences.

Early in the coding process it was clear that most participants did not provide an accurate answer to questions 3a and 3b (see Appendix B) due to ambiguous wording in the question. Consequently, all of question three in the open response section was removed from analysis. Of the remaining open response questions, there were only three categories into which an answer could be classified: 0, completely incorrect answer, 2.5, the answer is either unclear or is clearly missing part of the answer, or 5, a comprehensive and scientifically-based response is provided. Open response questions were given a point value of five whereas the closed response (multiple true/false) questions were given a value of one point each and the other closed response (multiple choice) questions were given a value of three points. As open response questions tend to be more

challenging, and there are fewer of them, question weighting ensured that neither the open nor closed response questions had vastly more influence on participant results as seen in Table 2.

To determine if there was appropriate coding for the short answer questions, I created a histogram for each short answer question after coding all responses. I created histograms to determine if a majority of scores were in the half credit range (2.5). If any question had a majority of scores in the half credit range, I would need to make a new coding scheme with more categories to get an accurate portrayal of student understanding. None of the short answer questions had a majority of half credit answers, as shown in Figure 4, so there was no need to recode the data.

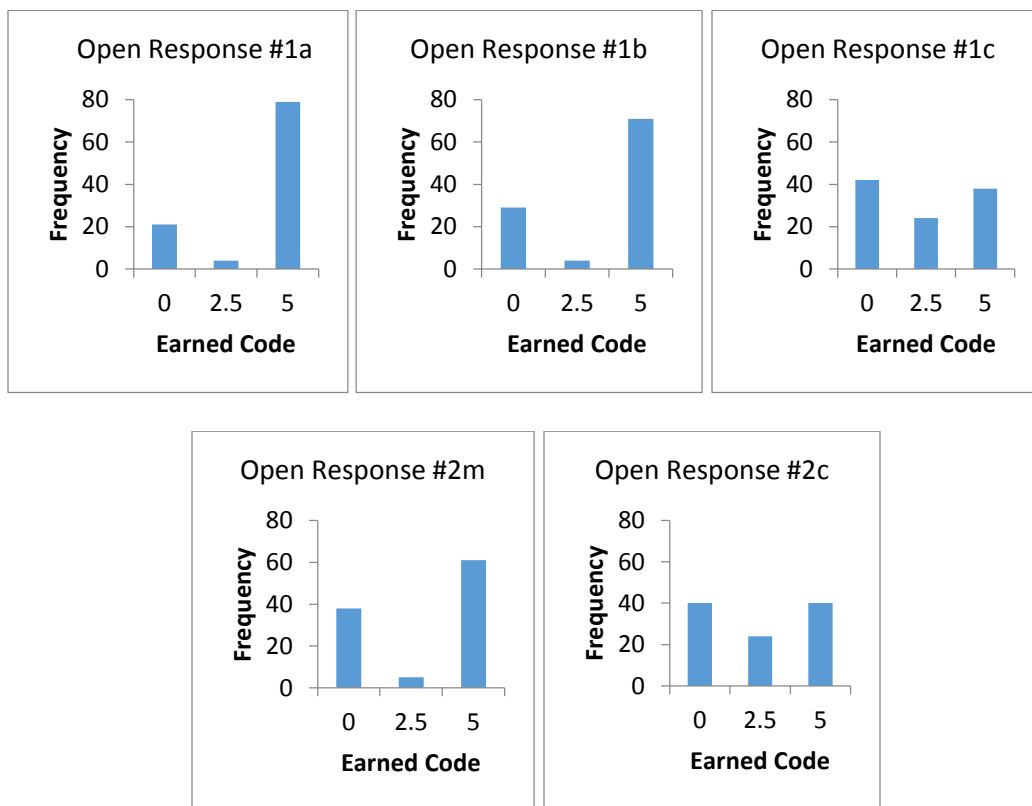


Figure 4. Frequency of earned codes (0, 2.5, 5) for each open response question. Question two had a mechanism component (m) and a consequences component (c).

Fourteen percent of responses were coded by a second researcher to verify the consistency of my coding. This researcher received a random sample of five participants' open

response answers from each of the three different classes (N=15). Using the coding rubrics shown in Appendix B, initial comparison showed eighty-eight percent inter-rater agreement. After discussion of the incongruent responses, the other researcher changed and agreed with the author on three of the nine incongruent codes, the author changed her coding on two of the nine dissimilar responses to match that of the other researcher, both the researcher and the author changed to the same code on two of the dissimilar responses, and the researcher and author continued to disagree on two of the sample coded questions. Post discussion, there was ninety-seven percent agreement on coding between the researcher and the author (Cohen's kappa = 0.938).

Results

At the outset, there were one-hundred seventeen participants in this study. Six students from Ecological and Evolutionary Systems, six students from Introduction to Biology and one student from Human Biology and Bioethics did not answer all closed response and open response questions. Consequently, I did not feel there was enough data to assess their understanding and they were removed from data analysis. The remaining one-hundred four participants' data was analyzed: 39 students from Ecological and Evolutionary Systems, 32 students from Introduction to Biology and 33 students from Human Biology and Bioethics.

Student responses were evaluated for mastery. In this study, mastery was considered earning an average of at least eighty percent on each section: Mechanism, Outcomes, or Consequences. Fifty percent of the time students might guess the correct answer and a seventy-five percent is considered passing in many courses. Consequently, eighty percent was chosen as the threshold for mastery to err toward being more conservative in evaluating student understanding.

Initially, I classified the students as one group: undergraduate students taking an introductory biology class. Upon review of students' responses I noticed a disparity between the responses from participants in Ecological and Evolutionary Systems (an introductory course for biology majors) and the other two courses, comprised of non-science majors (Figure 5). Over sixty percent of science majors “mastered” the Consequences section, while fewer than twenty percent of the non-science majors “mastered” the Consequences section. The Consequences section shows student understanding of evolution by natural selection, a topic taught in all three courses and a focus of this study.

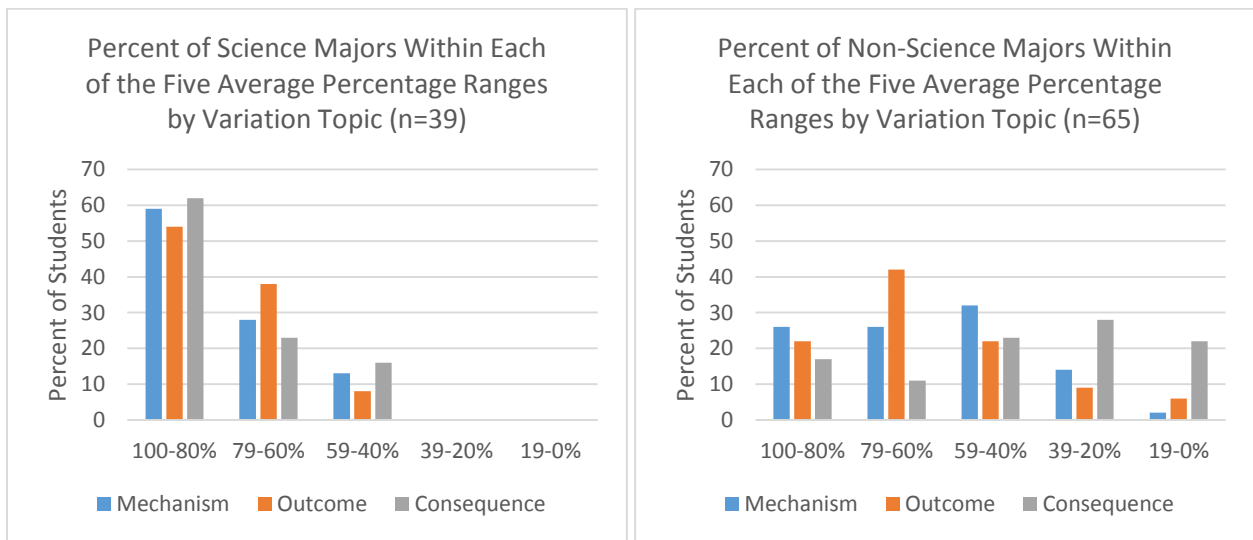


Figure 5. Percentage distribution by topic of science majors and non-science majors.

The “mastery” of participants in the other two classes (non-science majors) is shown in Figure 6. The bars on each histogram is unique indicating that even between the non-science majors the data was not distributed in a similar manner.

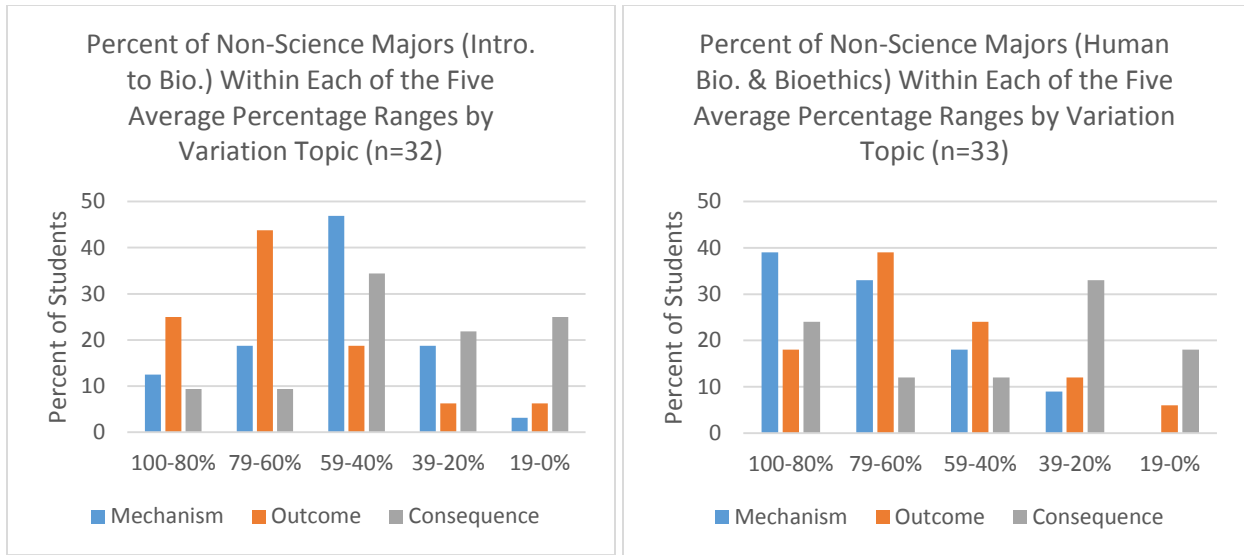


Figure 6. Percentage distribution by topic of each non-science major course.

Table 3 displays the percentage of participants who earned a score within a given percent range on each of the three sections of Mechanism, Outcomes and Consequences of genetic variation. These values were calculated by adding the total number of points earned for each section, separated by course, divided by the maximum possible points for that section, by course. Five percent ranges were created where bolded, highlighted cells show the percentage of each student group who “mastered,” earned an eighty percent or greater, the Mechanism, Outcome, or Consequences section. The unshaded quantities in Table 3 are not shown graphically in any figures. Each green shaded box in Table 3 is displayed graphically in Figure 6 or the left graph in Figure 5.

Table 3

Participant Data for Open Response (OR), Closed Response (CR) and Open/Closed Mean

| Student Group | Earned Percent Range | Mechanism | | | Outcome | | | Consequences | | |
|--|----------------------|-----------|-----|------------|---------|-----|------------|--------------|-----|------------|
| | | OR | CR | OR + CR | OR | CR | OR + CR | OR | CR | OR + CR |
| Science Majors (n=39) | 0-19% | 8% | 3% | 0% | 10% | 0% | 0% | 0% | 0% | 0% |
| | 20-39% | 0% | 0% | 0% | NA | 0% | 0% | 5% | 0% | 0% |
| | 40-59% | 28% | 8% | 13% | 5% | 15% | 8% | 28% | 5% | 15% |
| | 60-79% | 5% | 26% | 28% | NA | 54% | 38% | 10% | 21% | 23% |
| | 80-100% | 59% | 64% | 59% | 85% | 31% | 54% | 56% | 74% | 62% |
| Non-Science Majors: Intro. To Bio. (n=32) | 0-19% | 28% | 0% | 3% | 16% | 9% | 6% | 41% | 6% | 25% |
| | 20-39% | 3% | 13% | 18% | NA | 9% | 6% | 25% | 13% | 21% |
| | 40-59% | 44% | 44% | 46% | 3% | 44% | 18% | 16% | 22% | 34% |
| | 60-79% | 6% | 34% | 18% | NA | 31% | 43% | 16% | 28% | 9% |
| | 80-100% | 19% | 9% | 12% | 81% | 6% | 25% | 3% | 31% | 9% |
| Non-Science Majors: Human Bio. & Bioethics (n=33) | 0-19% | 6% | 0% | 0% | 36% | 3% | 6% | 52% | 3% | 18% |
| | 20-39% | 3% | 6% | 9% | NA | 18% | 12% | 6% | 9% | 33% |
| | 40-59% | 39% | 30% | 18% | 3% | 36% | 24% | 12% | 21% | 12% |
| | 60-79% | 3% | 40% | 33% | NA | 33% | 39% | 21% | 15% | 12% |
| | 80-100% | 49% | 24% | 39% | 61% | 9% | 18% | 9% | 52% | 24% |

Note. Bolded, highlighted percentages indicate mastery for each course in each topic area. NA indicates a value of zero as that percent range could not be calculated for Outcome section due to the coding scheme utilized. Green shading indicates the quantities graphed in Figures 5 and 6.

The science majors had the most participants attain the “mastery” range for all three sections of the instrument (Mechanism—59%, Outcome—54%, Consequences—62%). The non-science majors had a mastery percent of 26% for Mechanism, 21% for Outcome and 17% for Consequences sections. Based solely on the “mastery” percentage for each section,

understanding the consequences of genetic variation over time was the most challenging for non-science majors. Although most non-science majors did not demonstrate “mastery” on any one section, the larger percentage of participants attaining “mastery” on specific sections fluctuated between the two courses (Table 3). It is interesting to note that every group demonstrated more mastery of the Consequences section in the closed response category than the open response category. No such trend was found with Mechanism or Outcome.

The open response (OR) for the outcomes average did not have any participants score in the 20-39% or 60-79% ranges because there was only one question that formed the “average.” Students could either earn a zero, fifty percent or one hundred percent leaving the other two percentage ranges empty.

The data was not normally distributed as evidenced by the variation between the mean and median of each group and the skewed distribution of the science majors earned means. Figure 7 contains scatterplots of the means of each of the three sections for the science majors and non-science majors groups. To determine if there was a relationship between understanding of the mechanisms, outcomes and consequences of genetic variation, I ran the non-parametric Kendall’s *tau* Rank Correlation for participant groups: science majors and non-science majors. Those results are displayed in Table 4.

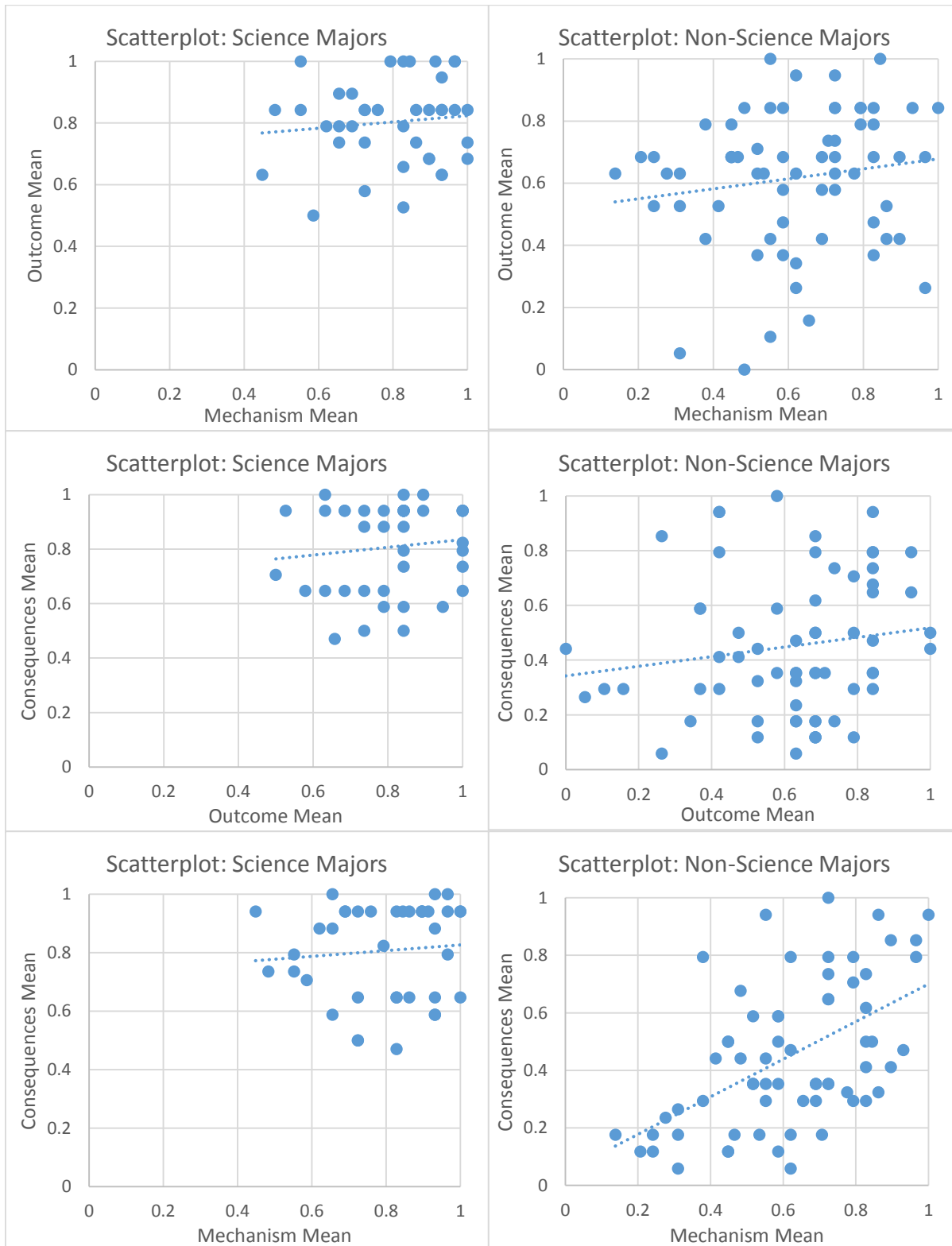


Figure 7. Scatterplots of all possible pairings of mechanism, outcome and consequences for both science majors and non-science majors. Linear trend lines added to each scatterplot.

| Table 4 | | | | |
|---------------------------------------|-----------------------|-------------------------------|--|---|
| <i>Kendall's tau Rank Correlation</i> | | | | |
| | <i>Science Majors</i> | <i>All Non-Science Majors</i> | <i>Non-Science Majors: Intro. To Biology</i> | <i>Non-Science Majors: Human Bio. & Bioethics</i> |
| Mechanism & Outcome | 0.059 | 0.122 | 0.342 | -0.032 |
| Outcome & Consequences | 0.083 | 0.140 | 0.198 | 0.092 |
| Mechanism & Consequences | 0.104 | 0.382 | 0.547 | 0.327 |

Notes. All correlations are 2-sided. Highlighted scores have 2-sided p-values <0.05.

Kendall's *tau* Rank Correlation was used to test the relationship between the variable sets of mechanism and outcome, outcome and consequences and mechanism and consequences. Kendall's *tau* determines if there is a strong or weak relationship between two variables but does not show causation.

In addition to running correlations for the classes, I also categorized individuals based on whether they demonstrated "mastery" in each of the variation categories (Table 5) as a way to test my hypothesis. "Mastery" was considered earning an 80-100% mean for each of the three sections and was denoted with a plus (+). Earning below an eighty percent indicated "mastery" had not been reached and was denoted with a minus (-). Based on the pattern of plusses and minuses for an individual, each participant was placed in one of the categories in Table 5.

Category A supports my hypothesis because if students understand mechanisms of genetic variation they also show understanding of the outcomes of genetic variation, e.g. phenotypic variation, and understand the consequences of genetic variation, e.g. evolution by natural selection. Category B supports my hypothesis because if students do not understand the mechanisms of genetic variation, they should not understand the outcomes of that variation nor the consequences of the genetic variation. Categories C and D do not support my hypothesis.

Category C is composed of students who do not show an understanding of mechanisms but understand outcomes of variation, and thus mechanism of variation is not a threshold concept. Whether they understand evolution or not does not matter as the data is already contrary to my hypothesis. Category D is composed of students who do not show an understanding of the outcomes of genetic variation but do show an understanding of evolution by natural selection, again suggesting that outcomes of variation are not a threshold concept. Whether they understand mechanisms of variation or not, the data still opposes my hypothesis.

| Table 5 | | | |
|--|------------------|----------------|---------------------|
| <i>Categories of Student Understanding</i> | | | |
| <i>Category</i> | <i>Mechanism</i> | <i>Outcome</i> | <i>Consequences</i> |
| A | + | + | + |
| B | - | - | - |
| <i>C</i> | - | + | - |
| | - | + | + |
| <i>D</i> | + | - | + |
| | - | - | + |
| | + | + | - |
| | + | - | - |

Notes. + indicates section mastery. – indicates no section mastery. To further test the hypothesis, each student was placed in a category depending on sections mastered. Categories A and B support the hypothesis. Categories C and D (*italics*) refute the hypothesis. The two rows without a category title neither support nor refute the hypothesis.

There was a category of participants with data that neither supported nor refuted my hypothesis. These participants demonstrated understanding of the mechanisms of genetic variation but either did not show understanding of the outcomes of genetic variation or the outcomes and consequences of genetic variation. These students are in the learning process but have not yet assimilated evolution by natural selection into a cognitive structure to the point they can demonstrate mastery.

Table 6 shows the number of participants in each category split by science majors and non-science majors. Twelve (31%) of the science majors fell in either Category A or Category B, which supports my hypothesis that to understand the consequences of genetic variation, one must first understand the mechanisms and outcomes of that variation. Eighteen (46%) science majors were classified as Category C or D, and reject my hypothesis, while the remaining nine (23%) are possibly on my hypothesized track for understanding evolution but have not yet mastered that topic. The non-science majors showed different trends with thirty-nine (60%) falling in either Category A or B, sixteen (25%) in either Category C or D and ten (15%) uncategorized participants. All participants were equally motivated to perform well on these questions as they were part of a final exam for a class.

| Category | Science Majors | Non-Science Majors |
|--------------|----------------|--------------------|
| A | 8 | 1 |
| B | 4 | 38 |
| C | 9 | 9 |
| D | 9 | 7 |
| Neither | 9 | 10 |
| TOTAL | 39 | 65 |

The chi-square (χ^2) value calculated for the science majors (N=39) is 2.502. The one-sided χ^2 critical value for science majors with $\alpha=0.05$ ($df=38$) is 53.384, indicating the null hypothesis (the number of students is equal among the categories) is the most attractive explanation for the observed differences. The one-sided χ^2 critical value for non-science majors with $\alpha=0.05$ ($df=64$) is 83.675. The chi-square for non-science majors (N=65) is 116.515, indicating the null hypothesis is not the most attractive explanation for the distribution of responses.

It is generally accepted in science education research that open response questions elicit a more comprehensive or accurate portrayal of student understanding. Consequently, I also looked just at student responses to the first open response question, which had the three parts: mechanism, outcome and consequences of variation (Table 7). The categories W, X, Y and Z correspond to the level of student understanding described for categories A, B, C and D, respectively. Results can be found in Table 8.

| Table 7 | | | |
|---|------------------------------------|----------------------------------|---------------------------------------|
| <i>Categories of Student Understanding for Open Response Question 1</i> | | | |
| <i>Category</i> | <i>Mechanism: Open Response 1b</i> | <i>Outcome: Open Response 1a</i> | <i>Consequences: Open Response 1c</i> |
| W | + | + | + |
| X | - | - | - |
| Y | - | + | - |
| | - | + | + |
| Z | + | - | + |
| | - | - | + |
| | + | + | - |
| | + | - | - |
| <i>Notes.</i> + indicates question mastery. – indicates no question mastery. To further test the hypothesis, each student was placed in a category depending on sections mastered. Categories W and X support the hypothesis. Categories Y and Z (<i>italics</i>) refute the hypothesis. The two rows without a category title neither support nor refute the hypothesis. | | | |

| Table 8 | | |
|--|-----------------------|---------------------------|
| <i>Participants in Each Category by Major for Open Response Question 1</i> | | |
| Category | Science Majors | Non-Science Majors |
| W | 20 | 4 |
| X | 1 | 7 |
| Y | 7 | 17 |
| Z | 2 | 6 |
| Neither | 9 | 31 |
| TOTAL | 39 | 65 |

The two-sided χ^2 critical values for science majors with $\alpha=0.05$ ($df=38$) are 22.878 and 56.896. The calculated χ^2 value of 53.474 indicates that the null hypothesis is the more attractive explanation for the response score distribution of science majors. The two-sided χ^2 critical values for non-science majors with $\alpha=0.05$ ($df=64$) are 43.776 and 88.004. The calculated χ^2 value of 22.854 indicates that we can reject the null hypothesis that student open response scores are due to random distribution.

An unexpected finding was the correlation seen for the Introduction to Biology class. Interestingly, the participants from this course earned the lowest mean scores, yet showed the highest degree of positive correlation between the three sections, and support of my hypothesis, especially when correlating mechanism and consequences (Kendall's $\tau = 0.382$; p -value= 1.6×10^{-5}).

Discussion and Conclusion

My original hypothesis was that to have a thorough understanding of evolutionary change, which is the consequence of variation acted upon by natural selection, one must have a detailed understanding of the mechanisms that caused genetic variation and outcomes of that variation. Based in the data collected in this study, I have to reject my original hypothesis. As identified by Meyer and Land (2003), threshold concepts are likely to be transformative, irreversible, integrative, bounded and troublesome. Although variation can be seen as integrative, it does not seem to be transformative, irreversible or troublesome as students' demonstrated mastery of evolution with and without demonstrating mastery of mechanisms or outcomes of variation.

Correlation

As shown in Table 4, a significant correlation is only found between the mechanisms and consequences of genetic variation for non-science majors. This indicates that if a non-science major participant understands the mechanisms of variation, they could understand the consequences of that variation. It is not clear if there really is no correlation between either mechanism and outcomes or between outcomes and consequences or if other factors are contributing to this non-significant result. The instrument used in this study (MOCVI) may not be sensitive enough or comprehensive enough to delineate the actual differences between those sections. A limitation of this study was the use of previously published questions; only two of the twelve closed response questions were written specifically for this instrument. Questions written and validated for this study would increase instrument specificity. Additionally, increasing the number of questions assessing each of the three sections or increasing the number of open response questions might elicit different results.

Category Distribution

The null hypothesis was the most attractive explanation for the distribution of science majors into various categories (Table 9), indicating that random chance might have produced the distribution seen. Thirty-one percent of science majors and sixty percent of non-science majors were classified as belonging in categories A or B (Table 9). These participants support my hypothesis: knowledge of the mechanisms and outcomes of genetic variation are foundational for understanding evolution. However, the vast majority of these non-science majors did not show mastery of any of the three topics of mechanism, outcome and consequences, and fell in category B. Although category B supports my hypothesis, it indicates there is little student understanding of evolution by natural selection or the associated genetic components; a problem discussed by

many other authors (Anderson *et al.*, 2002, Bishop & Anderson, 1990, Kalinowski *et al.*, 2013, Nehm & Reilly, 2007, Opfer *et al.*, 2012). The percentage of participants who were classified as belonging to categories C or D, and do not support my hypothesis, are forty-six percent of science majors and twenty-five percent of non-science majors.

| <i>Category</i> | <i>Percentage of Participants</i> | |
|-----------------|-----------------------------------|---------------------------|
| | <i>Science Majors</i> | <i>Non-Science Majors</i> |
| A/B | 31% | 60%* |
| C/D | 46% | 25%* |
| None | 23% | 15%* |
| W/X | 54% | 17%* |
| Y/Z | 23% | 35%* |
| None | 23% | 48%* |

Notes. Highlighting indicates support of thesis hypothesis. * indicates rejection of null hypothesis for cause of response distribution.

Mastery

An assumption I made in this study was that most university students would come to college with a solid understanding of evolution by natural selection. However, only sixty-two percent of science majors and eighteen percent of non-science majors attained “mastery” on the consequences of genetic variation section, e.g. evolution, questions even after taking a college course in which that content was taught. As shown in Table 3, students in Ecological and Evolutionary Systems outperformed students in other classes on all the topics assessed. It is unclear what caused these students to show a better understanding. One reason for the greater level of mastery of natural selection demonstrated by these students may be that they are all science majors and have taken more science classes on this content. Science majors might also be innately more interested in the topic as it is part of their chosen area of study and,

consequently, perform better. Another reason for the level of mastery may be the content of the course they took. All three courses ‘teach’ evolution but there is more of an emphasis on the process of evolution by natural selection in the science majors’ course, as denoted in the course name. Last, the extensive teaching experience of the instructor of the Ecological and Evolutionary Systems course may have impacted student performance.

The Human Biology and Bioethics and Introduction to Biology courses were combined to determine the average for non-science majors in this study. As noted in the results, the percent of non-science majors demonstrating ‘mastery’ on the sections of mechanism, outcome and consequences were twenty-six percent, twenty-one percent and seventeen percent, respectively. I hypothesized that consequences of genetic variation could only be understood if students understand the mechanisms that cause genetic variation and the outcomes of that variation. The finding that the consequences of genetic variation is the least understood of the three categories for non-science majors’ supports my hypothesis. There should be fewer participants demonstrating mastery for a more complex topic, such as evolution, if there are other areas that must be understood first.

When Charles Darwin published his 1859 book *On the Origin of Species by Means of Natural Selection*, in which he articulated his ideas on how natural selection leads to evolution, Gregor Mendel’s ground-breaking genetics work on pea plants had not been presented. Not until 1866 did Mendel publish his “Experiments on Plant Hybridization” which described the existence of “factors” that controlled the genotypes and phenotypes of plants. Consequently, the people who initially agreed with and supported Darwin’s theory of evolution by means of natural selection knew nothing of Mendelian genetics or genetic variation. It may be that people can understand evolution without understanding the molecular basis of variation as long as they

understand there is variation among individuals upon which natural selection acts; many results in this study were found to be not statistically significant possibly due to this reason.

Limitations

One major limitation of this study was the unequal number of questions on the three different sections: Mechanism, Outcome and Consequences. There were twenty-nine possible points in Mechanism whereas Outcome and Consequences only had thirteen and twenty-three, respectively. An identical number of questions and points for each section could reveal more information and possibly result in different conclusions. Another limitation was the use of published questions. As mentioned previously, the majority of questions on this instrument come from other instruments or assessments each with a unique focus. If questions were written to gauge participant understanding of variation, specifically the mechanisms, outcomes and consequences of genetic variation, it might result in a more complete, and possibly different, view of participant fluency. Another limitation was the participant population used in this study. The demographics of this university do not accurately reflect those of many universities in the United States and, consequently, the results might not be generalizable to other participant groups. Additionally, the non-science major group was composed of students who completed two different courses by two different instructors. One of the instructors might have emphasized evolution by natural selection more than the other.

Future Research

There are a number of avenues for research of threshold concepts in biology. To more accurately determine if the topics of mechanism and outcomes of genetic variation are a threshold concept for consequences of genetic variation, questions could be written for this specific task and administered with follow-up interviews. It could then be evaluated if the new

questions that focus on student understanding of variation at the three levels are valid. If these questions are valid, this study could be run again to see if a different set of questions elicit similar results as those found here. Another direction of future study might be to determine causation of the correlations identified in this study. This might be accomplished by interviewing participants to attain a more complete view of their understanding of these three topics. From these interviews there may be other topics revealed as paramount to knowledge construction of evolution.

Although the hypothesis for this study was rejected, it may prove useful to teachers and researchers looking for biology threshold concepts in the future. For teachers, knowing that students may not have to understand the genetic underpinnings to understand evolution by natural selection can add flexibility to the order of content taught in courses. Other researchers might use the MOCVI as a framework around which to add more questions and delve deeper into the topics addressed here. Additionally, this study adds to the body of work that demonstrates understanding evolution is a challenge for undergraduate students, both science and non-science majors.

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APPENDIX A

Mechanism, outcomes and consequences of variation instrument (MOCVI)

Open Response

Adapted from--Question 8 of the 2014 AP Biology Scoring Guidelines

1. A research team has genetically engineered a strain of fruit flies to eliminate errors during DNA replication. The team claims that this will eliminate genetic variation in the engineered flies. A second research team claims that eliminating errors during DNA replication will not entirely eliminate genetic variation in an actively, sexually reproducing population of engineered flies.

- Provide ONE piece of evidence that would indicate new genetic variation has occurred in the engineered fruit fly strain.
- Describe ONE mechanism that could lead to genetic variation in the engineered strain of flies.
- Describe how genetic variation in a population contributes to the process of evolution in the population.

Adapted from--Opfer et al. (2012)

2. All elm trees produce seeds that have papery wings. How would biologists explain how an elm tree species with winged seeds evolved from an ancestral elm species that did not produce winged seeds?



Personal Communication—Dr. Jan Batzli

3. Body length in a population of Galapagos finch located on Daphne Island is known to vary from 114mm to 160mm in length, based on historic records. As a researcher, you visited Daphne Island in the summer of 2014 and measured body length of the entire finch population. To your surprise, you find that all the bodies are the same length (119mm)- no variation.

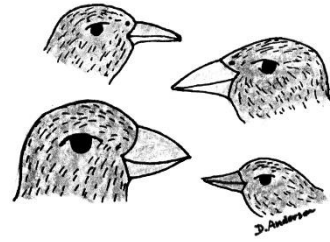
- Describe two mechanisms that could reduce phenotypic variation in body length.
- Briefly describe the factors involved in evolution of body length in finches.

Closed Response

From--Anderson & Evans (2013)

Introduction to Galapagos finches-use this information to answer the next question.

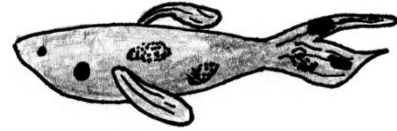
- Finches have been studied on the Galapagos Islands by many scientists.
 - The original finches most likely came to the islands one to five million years ago.
 - Scientists have evidence that 14 species of finches on the Islands evolved from a single species.
 - Species found on the islands have different beak sizes and shapes.
-



1. How did the different types of beaks first appear in the finches?
 - a. Changes in the finches' beak size and shape happened because of their need to be able to eat different kinds of food to survive.
 - b. Changes in the size and shape of the beaks of the finches because of random changes in the DNA.*
 - c. Changes in the beaks of the birds happened because the environment caused beneficial changes in the DNA.
 - d. The beaks of the finches changed a little bit in size and shape during each bird's life, with some getting larger and some getting smaller.

Introduction to South American guppies-use this information to answer the next two questions.

- These are small, colorful fish found in streams in Venezuela.
- Scientists have studied guppies in both natural streams and in lab experiments.
- Males have black, red, blue and reflective spots.
- Brightly colored males are easily seen and eaten by predators, however females tend to choose more brightly colored males.
- In a stream with no predators, the number of males that is bright and flashy increases in the population.
- If predators are added, the number of brightly-colored males gets smaller within about five months (3-4 generations).



-
2. What is the best way to describe the evolutionary changes that happen in the guppy population over time?
 - a. The traits of each guppy in the population change slowly.
 - b. Guppies with certain traits reproduce and become more common. *
 - c. Behaviors learned by certain guppies are passed on to their offspring and become more common.
 - d. Mutations happen in the guppy population to meet the needs of the fish as the environment changes.

3. What could cause populations of guppies in different streams to become different species?
 - a. Groups of guppies could accumulate so many differences that they would not be able to breed with each other, and this would make them different species.*
 - b. All guppies are alike and there are not really different species.
 - c. Guppies that need to attract mates could change their spots in many ways, and this would make them different species.
 - d. Guppies that want to avoid predators in the different streams could change their patterns so they are not so bright, and this would make them different species.

Adapted from--Smith, Wood & Knight (2008)

4. A population of buffalos is isolated such that no new buffalos can come into their territory. Determine if the following **are** primarily responsible (true) or **are not** primarily responsible (false) for the appearance of new alleles in this population.
 - a. True/False: Reassortment of chromosomes during the process of creating sperm or eggs. F
 - b. True/False: Mutations in cells that will become sperm or eggs. T
 - c. True/False: Changes in the environment that favor some buffalo traits over others. F
 - d. True/False: Random mating between the buffalos in the population. F

Adapted from—Bowling et al. (2008)

5. As HIV has spread around the world, we know some human individuals are resistant to the effects of the virus even though they are HIV positive. Why?
 - a. They carry genetic differences that provide the resistance.*
 - b. Genetic changes that provide resistance are produced in response to infection by the virus.
 - c. Natural selection causes genetic differences to be produced that result in resistance.
 - d. The environment in which the individual lives determines resistance.

Adapted from—Bowling et al. (2008)

6. Which of the following is a characteristic of mutations in DNA?
 - a. They are usually expressed and result in positive changes for the individual.
 - b. They are usually expressed and cause significant problems for the individual.
 - c. They usually occur at very high rates in most genes.
 - d. They result in different versions of a gene within the population.*

Adapted from—Klymkowsky et al. (2008)

7. Natural selection produces evolutionary change by ...
 - a. changing the frequency of various versions of genes. *
 - b. reducing the number of new mutations.
 - c. producing genes needed for new environments.
 - d. reducing the effects of detrimental versions of genes.

Adapted from—Klymkowsky et al. (2008)

8. You follow the frequency of a particular version of a gene in a population of **asexual** organisms. Over time, you find that this version of the gene disappears from the population. Its disappearance is presumably due to ...
 - a. genetic drift.
 - b. its effects on reproductive success. *
 - c. its mutation.
 - d. the randomness of survival.

Adapted from—Alberts et al. (2013)

9. Determine which of the following **could** result in genetic variation (true) or **could not** result in genetic variation (false).
 - a. True/False: Mutations within the regulatory DNA of a gene. T
 - b. True/False: Purifying selection. F
 - c. True/False: Gene duplication and divergence. T
 - d. True/False: Recombination of gene versions during fertilization. T

10. Determine which of the following **could** (true) result from fertilization (fusing of egg and sperm) or **could not** result from fertilization (false).
 - a. True/False: New combinations of alleles. T
 - b. True/False: Appearance of new alleles. F
 - c. True/False: Different phenotypes of an organism. T
 - d. True/False: A new species of organism. T

11. Determine which of the following **can** result in the formation of new species (true) or **cannot** result in the formation of new species (false).
 - a. True/False: Random mating between individuals in a population. F
 - b. True/False: A population is separated by a geographic barrier. T
 - c. True/False: Reduced gene flow between groups of individuals in a population. T
 - d. True/False: Reassortment of chromosomes during production of eggs and sperm. F

Adapted from--Smith, Wood & Knight (2008)

12. Suppose that a single DNA base change of an A to a T occurs and is copied during replication. Is this change necessarily a mutation?
 - a. Yes, it is a change in the DNA sequence. *
 - b. Yes, if the base change occurs in the coding part of a gene; otherwise no.
 - c. Yes, if the base change occurs in the coding part of a gene and alters the amino acid sequence of a protein; otherwise no.
 - d. Yes, if the base change alters the appearance of the organism (phenotype); otherwise no.

APPENDIX B

1. a

Adapted from College Board (2014)
 Scoring Guide-5 point maximum

| | Full Credit (5 points) | Half Credit (2.5 points) | Zero (0 points) |
|----------------------------------|---|---|--|
| Piece of evidence | A-- New phenotypes B-- Different DNA sequence C-- New genotypes D-- Chromosomal differences E-- Different mRNA sequence F-- Protein with different amino acid sequence | Answer is unclear, ambiguous or incomplete. | Response does not answer the question. |
| Student Response Examples | <i>“New phenotypes are observed.”</i> | <i>“Active, sexually reproducing population of flies.”</i> It is unclear exactly what the participant understands. More explanation is needed to clarify the depth of the response. | <i>“Reproduction of sexually reproducing flies means there must be genetic variation when fertilization occurs between two haploid gametes.”</i> |

1. b

Adapted from College Board (2014)
Scoring Guide- can earn 5 points maximum

| | <i>Full Credit (5 points)</i> | <i>Half Credit (2.5 points)</i> | <i>Zero (0 points)</i> |
|--|--|---|--|
| Description of Mechanism | <p>A-- Sexual reproduction produces offspring with new combinations of alleles/traits</p> <p>B--Meiosis produces new combinations of alleles/traits</p> <p>C--Crossing over produces new combinations of alleles/traits</p> <p>D--Independent assortment produces new combinations of alleles/traits</p> <p>E--Random fertilization produces new combinations of alleles/traits</p> <p>F--Immigration/gene flow introduces new alleles/gene sequences</p> <p>G--Viral infection inserts DNA into genome</p> <p>H--Nondisjunction causes anomaly in chromosome number</p> <p>I--Chromosomal rearrangements (e.g., large deletions, duplications, translocations, inversions, transposons, etc.) inactivate genes or result in multiple copies of genes</p> <p>J--Radiation or chemicals or mutagens induce mutations/changes in DNA</p> | <p>Answer is unclear, ambiguous or incomplete.</p> | <p>Response does not answer the question.</p> |
| Student Response Examples | <p><i>“Crossing over during meiosis can produce new combinations of genes.”</i></p> | <p><i>“Mutations are one mechanism that could lead to genetic variation in the engineered strain of flies because if the flies develop some sort of mutation during development, it’s possible for that mutation to get passed on.”</i> By mentioning a <i>mutation during development</i>, it is unclear if they understand that mutations in somatic cells are not heritable.</p> | <p><i>“One mechanism could be behavioral isolation.”</i></p> |

1. c

Adapted from College Board (2014)
Scoring Guide- can earn 5 points maximum

| | <i>Full Credit (5 points)</i> | <i>Half Credit (2.5 points)</i> | <i>Zero (0 points)</i> |
|----------------------------------|---|--|--|
| Description of evolution | A--Genetic variation is the basis of phenotypic variation that can be acted upon by natural selection B--Without genetic variation, there is no phenotypic variation on which natural selection can act | Answer is unclear, ambiguous or incomplete. | Response does not answer the question. |
| Student Response Examples | <i>“The traits are inherited by offspring. With unequal reproductive success with variations and not all offspring surviving at same rates will lead to changes in the gene pool of the population over many generations. The population thus evolves over time.”</i> | <i>“W/O variation-there wouldn’t be any diff. in the populations-these differences allow certain indiv. to be more ‘fit’/likely to survive than others-enacting the process of natural selection.”</i> | <i>“New characteristics are developed as needed for survival.”</i> |

2.

Adapted from Opfer et al. (2012)
Scoring Guide- can earn 10 points maximum

| Concept | Coding Description | Student Response Example | Score |
|---------------------------------|--|---|--------------|
| Mechanism of Variation | Mutation or the random change of genetic information; may produce different phenotypes from parent's traits. | <i>"The first tree to have winged seed had this trait as mutation."</i> | 5 |
| | Answer is unclear, ambiguous or incomplete. | <i>"The elm trees species with winged seeds could have evolved from an ancestral elm species that didn't produce winged seeds through a possible mutation during development or through evolution with another species."</i> | 2.5 |
| | Mentioning "trait" but no mentioning about the 'cause' of the trait; incorrect response that doesn't answer the question | <i>"This could be from biogeographical changes in the trees."</i> | 0 |
| Consequence of Variation | The new trait increases fitness and allelic frequency increases in the population. | <i>"Those trees with winged seeds experienced better reproductive success than those without, meaning that the winged-seed trees survived and reproduced much more successfully, while the non-winged trees did not have as much success. It happened over many generations, and eventually the winged-seed trees outcompeted the non-winged and how they are all winged-seeded."</i> | 5 |
| | Answer is unclear, ambiguous or incomplete. | <i>"These seeds were better at spreading and allowed for easier distribution of genes. This mutation was then selected for and more offspring resulted from it."</i> | 2.5 |
| | No mention of increased fitness for the parent or changes in the allelic frequency in the population. | <i>"The population had to adapt...in order to have a higher survival rate."</i> | 0 |

3. a

Adapted from Andrews, et al. (2011)
Scoring Guide- can earn 5 points maximum

| Description | Points |
|--|---------------|
| Environmental conditions affect the survival and spread of existing, heritable traits by creating differential reproduction between individuals. | 2.5 |
| Differential reproduction leads to a change in the proportion of individuals that have a trait in the next generation. | 2.5 |
| Minimal phenotypic plasticity | 2.5 |
| No immigration | 2.5 |
| Geographic isolation | 2.5 |
| Bottleneck effect | 2.5 |
| Small population size | 2.5 |

3.b

Scoring Guide- can earn 5 points maximum

| Description | Points |
|--|---------------|
| Variation: genotypic/phenotypic | 1 |
| Genetic: heritable trait | 1 |
| Selection: stabilizing, sexual, environmental pressure | 1 |
| Time: generations | 1 |
| Adaptation | 1 |