First Steps of Putting Research into Practice: Utilizing Concept Inventories to Identify Biochemistry Misconceptions and the Development of a Guided Inquiry Activity to Correct the Identified Misconceptions

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Dedication

This thesis is dedicated to my mother, Theresa Humphreys Mattox, who raised both me and my sister as a single parent for most of our childhoods. She made many sacrifices for us and is an excellent role model for me. I am a strong, independent, outspoken woman today because of her love and support.
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Abstract

It is known that students leave science classes with an incomplete or incorrect understanding of some of the concepts covered in the courses. Identification of these misconceptions is difficult, as it usually involves conducting an hour long one-on-one interview with a student. Concept inventories were developed as a way for professors to identify misconceptions in their classroom in an efficient manner. However, there is no collection of biology, chemistry and biochemistry concept inventories and there has been no analysis of the quality of these inventories. One goal of the research was to collect these science inventories and do the much needed analysis. Fifty-two concept inventories were collected and they were analyzed to determine if research was done with the target population and what form of validity and internal consistency are most commonly reported.

Previous research has indicated that biochemistry students have a difficult time with enzyme-substrate interactions and the Enzyme-Substrate Interactions Concept Inventory (ESICI) has been developed to determine the most common misconceptions related to this topic. Using the ESICI, misconceptions were identified in a one-semester biochemistry course and an activity was constructed for use in the classroom as a replacement for the lecture-based method of teaching. This activity will help students to better incorporate enzyme-substrate interactions into their long-term memory as well as give them the chance to discuss their ideas with fellow students. The guided-inquiry activity was also tested for effectiveness as both an in-class activity and as a homework activity in order to determine for which setting this activity is best suited.
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CHAPTER 1: REVIEW OF THE RELEVANT LITERATURE AND PURPOSE OF THE STUDY

This chapter reviews the relevant literature pertaining to the learning theory that shapes this research and what is known about meaningful learning. Literature regarding student misconceptions, the identification of misconceptions, and how misconceptions can be corrected will be reviewed as well. The purpose of this study will also be explored.

1.1 Constructivism

Constructivism is based on the theory that students connect new knowledge to their previous knowledge and focuses on “the meaning-making activity of the individual mind”\(^1\) (p. 58). If a student is lacking the certain prior knowledge, or has incorrect prior knowledge, they will have nothing to connect with the new information or will correct the new knowledge with an incorrect idea, and meaningful learning will not be achieved. The driving idea behind constructivism is that knowledge is constructed in the mind of the learner. However, knowledge is not transferred intact from the teacher to the learner, and there are more factors involved in how students understand the information and incorporate it into their own schema. The web of knowledge is the long-term memory of a student, and it is made up of knowledge that students have learned from their prior classes and life experiences.\(^2\)

There are different versions of constructivism depending on the perspective of the learner and the learner’s interactions with the environment. Social constructivism focuses on how students use their previous experiences with the material and any social interactions with others to create a better connection to the material. Students bring different experiences to their learning process, and by having students work in groups, they have the chance to learn from other people’s experiences.\(^3\) Because students bring
different lenses to their views of knowledge, it is important that they share their experiences.

Piaget’s knowledge of developmental psychology factored into the creation of his theory of cognitive development. According to Piaget, children have preconceived ideas of how things work and they are constantly revising their mental models based on new experiences. The revisions to their mental models come from interactions with the physical environment. However, there are researchers who believe that the interactions that children have with other people also affect their mental models. Vygotsky included these interactions in his theory of cognitive development and meaningful learning.\textsuperscript{4} The zone of proximal development as defined by Vygotsky is “the distance between the actual developmental level as determined by independent problem solving and the level of potential development as determined through problem solving under adult guidance or in collaboration with more capable peers”\textsuperscript{5} (p. 33). Although the zone of proximal development was originally developed and proven in children, learning is a lifelong process. New knowledge is constantly being incorporated into people’s short-term memories, and it is often influenced by how a person interacts with the subject matter. The zone of proximal development is the area in learning where students form new concepts based on their interactions with other students and with the instructor. Cooperative learning allows students to form concepts through these crucial interactions and allows for meaningful learning to occur.\textsuperscript{5}

\subsection{1.2 Meaningful learning}

Rote learning is a manner of learning in which students memorize the new concepts instead of incorporating them into their web of knowledge. In contrast to rote
learning, meaningful learning occurs when a student connects the new information to their existing knowledge and can utilize the new concepts in new contexts. Three requirements must be met in order for meaningful learning to happen: 1) the student must have some prior knowledge that is related to the new content, 2) the new content must be meaningful, meaning that it contains ideas relatable to the student’s existing knowledge, and 3) the student must choose to incorporate these new concepts into their prior knowledge. Determining when meaningful learning occurs can be in the control of the professor or the person who is passing on the knowledge, but there are other factors that determines if the learning will occur. The student is in control of two of the three requirements of meaningful learning, so they are ultimately the determining factor in whether meaningful learning will occur or not. Students have to have some relevant prior knowledge and they have to choose to incorporate the new concepts into their web of knowledge.³

When Piagetian and Vygotskian theories are combined, the resulting theory is learner-centric, includes both environmental and individual interactions, focused on the process, and allows for inclusion of differences in the learning environment. Piaget’s requirement for changing a student’s misconceptions is disequilibration.⁴ In order for existing knowledge schemes (working memory) to be modified and edited, there has to be a reason for the modification. There must be a transition from a state of mental equilibrium to disequilibrium and back to equilibrium.⁵ By creating cognitive dissonance, or mental stress created by conflicting ideas, between their current understanding and application of an idea and the correct understanding and application, students are more likely to incorporate the new understanding into their web of
knowledge. There are other factors that influence how effectively a student learns a concept, including their social interactions concerning the concept. Cooperative learning is one method that can be used to encourage student interaction and discussion regarding the concept being taught.

1.3 Inquiry in Education

When creating inquiry-based learning opportunities for students, there are four different levels of inquiry that are used as teaching methods. The first level of inquiry is confirmation inquiry. This level of inquiry has students work through an activity in which the results are already known. In order to work through this activity, a procedure or set of step-by-step instructions is given. Activities at this inquiry level are generally called “cookbook” activities. The second level of inquiry is structured inquiry. In this level of inquiry, students investigate a question through a procedure given to them by the teacher. The third level of inquiry is guided inquiry. Guided inquiry activities allow students to create their own procedure to investigate a presented question. The fourth, and most complex, level of inquiry is open inquiry. In open inquiry activities, student create their own question and their own procedure to answer the question.

1.4 Guided Inquiry

Guided inquiry activities are designed to walk students through a scaffolded activity that allows student to explore the targeted material in a unique way. The most commonly utilized form of guided inquiry are Process Oriented Guided Inquiry Learning (POGIL) activities. POGIL activities are designed to allow students to build on their prior knowledge while working in teams and managing themselves. The instructor serves as a facilitator for the class, instead of as a traditional lecturer. The facilitator is there to
listen to the groups and to provide guidance for students if necessary. Effective facilitation requires the ability to redirect and rephrase questions as well as managing student frustration.\textsuperscript{9}

One important aspect of POGIL activities is the scaffolding of the activity to follow the 3 phase learning cycle, as shown in Figure 1.1. In the first phase of the learning cycle, students \textit{explore} a concept through a model that is related to their prior knowledge in the subject and look for patterns. They may also be asked to explain any patterns that arise or the relationships that they see in the model. In the second phase of the learning cycle, students continue \textit{forming the concept} through further exploration of a model and continuing explanation of patterns. In the third phase, students are asked to \textit{apply} the concept they have developed from the first two phases to a new situation or relationship.\textsuperscript{10} POGIL activities are scaffolded in this manner to allow students to invent concepts through a series of guided questions.

\begin{figure}
\centering
\includegraphics[width=\textwidth]{3-phase-cycle.png}
\caption{The 3-phase learning cycle utilized in guided inquiry activities}
\end{figure}

In addition to scaffolding the activity so that the three-phase learning cycle is followed, there are four essential elements that distinguish a guided inquiry activity from a non-guided inquiry activity. The first element is small groups of students who self-manage. In these groups, all of the students must be able to share their views on the concept and have a voice in determining the answers to the activity. The second element
is the use of the instructor as the facilitator. The instructor must be willing to step aside and guide students, instead of spoon-feeding them the information. The third essential element of guided inquiry activities is a specially designed activity that follows the learning cycle. All three phases of the cycle must be incorporated into the activity for it to be called guided inquiry. Finally, the activity must place an emphasis on the development of process skills and mastering course content. Process skills include problem solving, deductive reasoning, communication, and self-assessment. By developing these skills through guided inquiry activities, students gain the ability to transfer these critical skills to other learning environments, such as lab.\(^8\)

The other important aspect of POGIL is promoting the sharing of student perspectives. To this end, students are typically assigned specific roles in the group. Each group only submits one set of answers at the end of the class period. Each group of students working on the activity should have no more than four students so that everyone has a chance to share their knowledge. There are four different roles that should be assigned amongst the students of the group. The first role is the manager. The manager is responsible for keeping the group focused on the task at hand, assigning responsibilities to the other team members, and ensuring that all members of the group have a chance to participate. The second role is the role of the spokesperson/presenter. The spokesperson is responsible for presenting the reports and discussion results to the class. The recorder is the third role, and that person keeps a record of the assignments and prepares the report that will be turned in. The last role is that of the reflector who identifies the strengths and weaknesses of the group and prepares a report on how well the group worked to give to the facilitator.\(^{10}\)
1.5 Misconceptions

In order for guided inquiry to be effective at correcting misconceptions, prior knowledge must be identified. Students enter science classes with certain prior knowledge based on their previous classes. Their existing ideas may be inconsistent with the generally accepted knowledge held by professors, which influences how they understand new concepts presented to them in future science classes.\(^\text{11}\) These inconsistent ideas are called many different names, including misconceptions, preconceptions, or alternative conceptions. Student knowledge about a topic is constructed from what they are taught in class and what they learn from their peers in a social context.\(^\text{12}\) Once misconceptions have been incorporated into long-term knowledge, students are hesitant to challenge them or try to correct them. In order for new concepts to be incorporated into student knowledge, interaction is crucial. Without interaction, the new concepts are significantly harder for students to understand. When a student is shown new material that contradicts their previous ideas, it is possible for them to ignore that new information. They will continue with their previously formed incorrect idea if they are frustrated by the disconnect. By allowing students the chance to interact with the material, they are more likely to persist if a disconnect happens.\(^\text{13}\) These incorrect ideas can be discerned through student interviews and, more recently, through the use of concept inventories developed using previously identified misconceptions.

1.6 Biochemistry Misconceptions

Biochemistry is one of the more historically difficult upper-level chemistry classes. Students are required to fit knowledge from biology and chemistry together after having been told for two years that the classes are separate. The concepts that are taught
are considered to be higher-order and require more critical thinking skills. Previous research has resulted in the development of tools to measure student misconceptions in foundational concepts for biochemistry and general biochemistry topics.\textsuperscript{14-15} The first concept inventory that focuses on a specific topic within biochemistry covers enzyme-substrate interactions.\textsuperscript{13}

Enzyme-substrate interactions are introduced to students first during their high-school biology courses and then again in their university biology and chemistry courses. When students enter a survey of biochemistry course, they do so from a variety of different levels. Some students may have learned more about how enzymes interact with their substrates or inhibitors than others. As discussed earlier, students remember different material from courses based on their perspective and their experiences. Since there is no standardized science curriculum in the United States, it can be difficult to teach the more complex enzyme topics at a level from which all of the students can understand and learn. It is important for educators to be able to determine every student’s level of comfort with the topic and to be able to determine if there are any common misconceptions held by many of the students.\textsuperscript{16} Concept inventories are one method of determining what misconceptions are held by a large number of students in a short amount of time, and a concept inventory has been developed that is specific to enzyme-substrate interactions.\textsuperscript{13}

The most common models that students are shown of enzyme-substrate interactions are the lock and key model and the induced fit model, which also include complex ideas about charge interaction and shape specificity. When asked about enzyme-substrate interactions, students often revert to the familiar model, the first picture they
were shown of how an enzyme and a substrate interact. Enzymes are covered in most biochemistry courses, and students are expected to retain all of the concepts. The courses are not specifically designed to correct student misconceptions. Instead of studying to learn, students study to pass the exam. In doing so, they may form misconceptions about enzyme-substrate interactions that are not always tested or corrected. A proper understanding of how enzymes function is critical to understanding more biochemistry topics. Enzyme-substrate interaction is related to many biochemistry ideas, such as the Krebs cycle, metabolism, and DNA synthesis, and is central to many health topics that doctors and nurses need to understand.

A concept inventory was created after interviewing many students and determining what topics were most difficult. There were five categories of misconceptions that were focused on in the creation of the Enzyme-Substrate Interactions Concept Inventory (ESICI). Students were asked to talk about enzymes and substrates in their own terms and describe what is happening in several figures. The ESICI uses the students’ own language in order to remove that barrier sometimes created by using larger “more scientific” words. The distractors in the concept inventory also were created from the student interviews.13

1.7 Purpose of the Study

The goals of this project are to (1) analyze the quality of concept inventories available for biology, chemistry, and biochemistry; (2) develop a guided inquiry activity to improve students’ misconceptions about enzyme-substrate interactions; and (3) compare the effectiveness of a guided inquiry activity as homework and as an in-class activity. There have been analyses of some concept inventories completed in the past,
but there has not been an exhaustive collection of biology, chemistry, and biochemistry concept inventories. While it is known amongst professors that concept inventories exist, it is difficult for people to analyze the quality of these inventories.

Once misconceptions have been identified in a population, the professor should use the results to inform their instruction. However, there are different methods that can be used to change the instructional techniques. One of the more common methods is through guided inquiry activities. Students work through a scaffolded activity together in small groups to form new concepts and apply them to provided models. The current guided inquiry activities for biochemistry do not focus on enzyme-substrate interactions, and have not been developed with the specific intent to correct identified misconceptions. Through the development of a guided inquiry activity that targets specific misconceptions, student misconceptions will be corrected and students will develop higher-order thinking processes.

The guided inquiry activities that exist currently for biochemistry are specifically designed to be utilized in the classroom. There may be a homework portion as the application section of the activity, but none have been created to be specifically homework. When the facilitator and student interaction is removed from a guided inquiry activity, will the activity still be effective at correcting student misconceptions if the information is scaffolded to guide students through models? It is not known if guided inquiry utilized as homework and as an in-class activity are equally effective at correcting misconceptions and guiding students to incorporate new information. By comparing the effectiveness of guided inquiry in both situations, it can be determined if the guiding questions are helpful for students in understanding new material.
1.8 References


Chapter 2: A META-ANALYSIS OF CONCEPT INVENTORIES

2.1 Introduction

When students leave a class, they often have an incorrect or incomplete understanding of the concepts covered in class, as knowledge is not transferred intact from teacher to student.\(^1\) The end of course finals are supposed to be able to determine how much information students have processed and retained, but students often cram for those tests. When students cram, they can retain that information for about two weeks, but the information is forgotten after that. It is not incorporated into their long-term memory, and so they cannot recall that information when they progress to the advanced classes.\(^2\) In advanced classes, new material is introduced that requires a complete understanding of previous topics. New information is built on the older material and then incorporated into a student’s long-term knowledge. Without the firm foundation of prior knowledge, the newer information may not be incorporated.

In order to be able to identify what concepts students are missing, interviews can be conducted. Researchers also typically try to collect information from professors about what they have noticed students having difficulty with in the past. Educators are aware of what students struggle with in a course, so they are a valuable resource. Researchers who are looking into student misconceptions should also look at the previous research done on this topic. So concept inventories were created as a way to measure student understanding.

In order to understand why students have difficulty integrating new information into their long-term memory, prior incorrect knowledge must be identified. By interviewing students, prior knowledge can be elucidated and analyzed to see where
incorrect ideas have been incorporated into the student’s understanding of a specific
topic.

Concept inventories were developed as a means of formative assessment. They
usually consist of multiple choice questions that have answers developed from incorrect
ideas that students expressed in interviews. Based on which answer option is selected,
the professor can determine which incorrect idea (or misconception) the student has.
Once the assessments are scored for the entire class, the professor can adjust their
teaching based on any incorrect prior knowledge or misconceptions created by the
lecture. Unfortunately, many teachers and professors are not aware that concept
inventories exist, especially since there is not a centralized database for concept
inventories.

The next goal of this project was to analyze the quality of the biology,
biochemistry, and chemistry concept inventories that were collected. There were four
specific aspects of concept inventories analyzed. The first aspect selected was how the
questions/statements were developed. There are two different approaches to developing a
concept inventory. The first method is the “top-down” approach. Using this method,
information about misconceptions is gathered from educators who teach the target topic
and questions and/or statements are developed using the language of educators.
Educators have a higher knowledge level than students, and a different vocabulary.3
When concept inventory items are created through this approach, low student scores
could be related to misconceptions or difficulty in understanding the language of the
question.
The other concept inventory development method is the “bottom-up” approach. Misconception identification is done through student interviews, in addition to literature research and educator input. When questions and/or statements are created, they use the same language that the students did during their interviews. By using language that students are familiar with, it allows researchers to rationalize that any incorrect responses are due to misconceptions, not interpretation difficulty with the questions. How did the creator(s) of the inventory decide what topics to focus on and what the misconceptions were in their target population? There are many different ways that researchers can select misconceptions and develop an assessment but only 4 were focused on in this study.

2.2 Quality Criteria

The first way that misconceptions can be identified is through a review of all relevant literature about the specific topic on which the researcher wishes to focus. The second way that misconceptions can be identified is through research with the target population. Research with the target population can be conducted through student interviews and through a pilot study. By interviewing students, researchers can use student language in the development of assessment items. The use of student language in item development follows best practices for the “bottom-up” approach to concept inventory creation. A pilot study for the instrument allows researchers to check for any issues with question wording and to identify any questions that students are not answering as expected.

The validity of the instrument is another important factor in determining the quality of the inventories. The validity of an instrument is equivalent to the accuracy of an instrument, or a measure used to determine if the instrument measures what it claims
to measure. As shown in Figure 2-1 below, there are 2 different categories of validity: construct and criterion. Construct validity asks if the instrument created actually measures what it is supposed to measure, and the four different kinds of construct validity are content, face, convergent, and discriminant. Content validity is a measure of the accuracy of the instrument. It looks at the information contained within the instrument. Face validity is used to measure if the instrument appears to measure the target concept. Both content and face validity are measured by a panel of experts in the field and are subjective measures. Convergent validity shows that assessments that should be related are related. Discriminant validity shows that students’ scores on the inventory are not related to their abilities to do or understand anything except the concept covered on the inventory.

The other category of validity is criterion validity. Criterion validity asks if the instrument is related to a measure in the real world, such as homework or a test covering the targeted topic. The two different types of criterion validity are predictive and concurrent. Predictive validity means that the assessment can predict how well a student is going to score on an external measure. Concurrent validity is the measure of the ability of an instrument to measure what it is supposed to measure. The instrument should be able to distinguish between the students who understand the material and students who do not understand the material.
In addition to validity, reliability is also considered when assessing the quality of a concept inventory. The reliability of an instrument is equivalent to the precision of an instrument, or a measurement used to determine if the instrument produces the same results each time it is used. There are two different categories of reliability investigated here: internal consistency and reproducibility, as shown in Figure 2-2 below.

Internal consistency is measured by either split-half reliability, Kuder-Richardson 20, or Cronbach $\alpha$. Split-half reliability takes the data set and splits the cases within the set into two different groups. The correlation coefficients of the two groups are then compared to see the consistency between the two groups and therefore the items on the assessment. Split-half reliability gives multiple values for the correlation coefficient, all of which are different. The values change based on how the data is divided into the two groups, and that can lead to large variations in the values. Kuder-Richardson 20 (KR-20) is a statistic that runs all of the possible split-half combinations. It is done with a computer, and is considered a better method for determining the internal consistency of an assessment as more comparisons are run. KR-20 can be used only for dichotomous
items, for example, yes-no questions.\(^5\) Cronbach \(\alpha\) is an alternative method for assessing the internal consistency of an assessment. First put forth by Cronbach in 1951, this test statistic is the same as KR-20, but it can be used for both dichotomous and liker-scale items.\(^6\) Once the internal consistency of the item has been tested, the next step is to check that students answer questions in the same manner when given the instrument twice without any instructional intervention.

Test-retest reliability is used to measure the reproducibility of the instrument. The assessment is given to the same population twice with an interval of time in between the two administrations. The length of time between the administrations is crucial so that students do not memorize the questions and repeat their answers from the first time. If students repeat their answers from the first administration, it is difficult to tell if the misconceptions are in students’ long-term memory. If too long of a gap is left between the two administrations, students may change their answers due to a change in their understanding of the concepts being measured. The correlation coefficient between the results is an estimate of the reproducibility of the instrument.
Figure 2-2. Representation of the different types of reliability that are used to evaluate concept inventories for quality.

When considering the quality of the available concept inventories, it is important to identify the different methods of item/statement development and the forms of validity and reliability that are reported. The more methods of item/statement development that are reported, the higher quality the inventory. The same rule applies with regards to the different forms of validity. Higher quality concept inventories report one or more form of internal consistency and a test-retest statistic for reliability. In the early years of concept inventory development, it was considered best practices to report the split-half reliability for the inventory. Since the development of alternate forms of reliability, such as Cronbach $\alpha$ and Kuder-Richardson 20, split-half reliability is no longer considered the best statistic to measure internal consistency. In order to determine the quality of the collected inventories, the articles about the inventories were collected and the data provided about the inventories in the articles was coded for analysis.
2.3 Methodology for the Collection of Inventories

The first part of this project focused on the collection of concept inventories targeted towards undergraduate students. The search was conducted from December 2013 until December 2014, and involved the use of six different search engines (Google Scholar, ProQuest, ACS Publications, Wiley, EBSCOHost, and ScienceDirect). Based on previous research into the identification of the different terms researchers use to classify their instruments, a list of search terms was devised and inputted into the search fields using Boolean operators to ensure all terms where present in the results. Table 2-1 shows the full list of search terms used to compile the inventories.

<table>
<thead>
<tr>
<th>Alternative Conceptions</th>
<th>Conceptual Knowledge</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alternative Concepts</td>
<td>Conceptual Survey</td>
</tr>
<tr>
<td>Concept Inventory</td>
<td>Diagnostic Test</td>
</tr>
<tr>
<td>Concept Survey</td>
<td>Foundational Concepts</td>
</tr>
<tr>
<td>Conceptual Assessment</td>
<td>Misconception</td>
</tr>
<tr>
<td>Conceptual Evaluation</td>
<td>Student Conceptions</td>
</tr>
<tr>
<td>Conceptual Inventory</td>
<td></td>
</tr>
</tbody>
</table>

Overall, 51 concept inventories were found for science topics. Science topics include physics\textsuperscript{8-18}, astronomy\textsuperscript{19-22}, biology\textsuperscript{23-41}, biochemistry\textsuperscript{54-56}, chemistry\textsuperscript{43-53}, and science\textsuperscript{7}. The science category includes an inventory developed to check for misunderstanding in all science topics covered in high school. The criteria for inclusion was focused on four specific areas. The first requirement was that the concept inventory was discussed in either an article, a dissertation, or a thesis. Articles, dissertations, and theses discuss the development of instruments in detail, have already created the instrument, and provide
inventories that are based in research. Inventories that were only discussed in conference abstracts were not included because there was not information about the development of the inventory, which is another of the criteria for inclusion. If the source did not contain information about the item development, test reliability or test validity, it was not included in the final collection of inventories for analysis. Two proposed inventories were not included in this analysis because they did not discuss the creation of an inventory. The articles mentioned a need for misconceptions in a certain subject to be investigated, but no subsequent articles were found containing more information.

Another factor in selecting the inventories for inclusion was the target population. The inventories needed to be targeted towards an undergraduate population. As a researcher in a university setting, collecting inventories targeted at undergraduate students could be useful to help professors with identifying misconceptions in their classroom. There was a concurrent project in the laboratory focused on the analysis of concept inventories targeted towards high-school students, so they were not included in this analysis. The final inclusion criterion was how the instrument was developed. Inventories were included only if they were targeted towards specific misconceptions and research-based. There are instruments labelled concept inventories that are just tests for a class. Teachers create a test for a specific unit in their classroom, and the instrument reported is only relevant to their classroom. The topics covered in the “inventory” have not been shown to be generalizable to the majority of students who take that class all over the country and were not included in this analysis. If an inventory meets all four of the criteria for inclusion, it is included in the meta-analysis.
2.4 Results of the Inventory Collection

In order to analyze the quality of the concept inventories that are available, a preliminary analysis of the science topics that inventories have been created for must be completed. Figure 2-3 shows the number of concept inventories found for each science topic included. Engineering, technology, and mathematics were excluded, since a previous study has analyzed these topics. The Science Concept Inventory (the only inventory in the science category) covers both chemistry and physics topics, so it is its own category. The Science Concept Inventory was created specifically for students entering university for the first time, and consists of 84 true-false questions. There have been 19 biology concept inventories created, which is more than have been created for other science subjects. But when you consider all of the topics covered in biology courses, the number of inventories does not come anywhere close to covering all of the topics. There were 11 physics concept inventories found. Physicists started developing concept inventories much earlier than the rest of the sciences, but they have slowed down in the identification of misconceptions and the creation of concept inventories to identify these misconceptions in the classroom. Four astronomy inventories were found, and all of these cover different topics in an astronomy class. Thirteen chemistry concept inventories were found, and the first one was created in 1999. The three concept inventories found for biochemistry were all focused on different concepts covered in biochemistry courses. The inventories were divided into groups based on the classroom in which the inventory was tested.
As shown in Figure 2-4, concept inventories have slowly become a more popular field of research. The first inventory found was created in 1941, and covered all topics of science. It was designed to test the science knowledge of men entering The Citadel as freshmen. There was no more research done into measuring student understanding through concept inventories until 1992. The Force Concept Inventory (FCI) was the next inventory created, and it was targeted towards physics students. Slowly, more and more concept inventories were developed and the focus expanded from physics to other science disciplines. More chemistry and biology concept inventories were developed over the years. The decision was made to focus on biology, biochemistry, and chemistry concept inventories as there has been no thorough analysis of these disciplines yet. The next step was to look at the subtopics covered by the biology concept inventories.
Figure 2-4. A timeline of the development of concept inventories from 1941 to present.

The biology concept inventories were broken down by topic in order to see the number of topics that the biology concept inventories cover (Figure 2-5). There are more concept inventories developed for genetics content knowledge than any other topic in biology and two of the inventories are focused on very specific sections of genetics. There is some content overlap with molecular biology concept inventories and biochemistry content inventories, but they were separated by the class that the inventory was tested in (molecular biology classrooms or biochemistry classrooms).

The concept inventories that were tested in a molecular biology classroom are the Introductory Molecular and Cell Biology Assessment (IMCA)\textsuperscript{39}, the Biology Concepts Instrument (BCI)\textsuperscript{29}, and the Meiosis Concept Inventory (Meiosis CI)\textsuperscript{30}. The IMCA has twenty-four multiple choice questions that cover multiple topics covered in molecular biology, including evolution, cell structures and features, active a passive transport, genetics, and gene expression. Even though the IMCA includes some questions related to genetics, it was created for and tested in a molecular biology classroom. The BCI has thirty multiple choice questions that cover diffusion and drift, energetics and interactions, molecular properties and functions, genetic behaviors, evolutionary processes, and
experimental designs. It was tested in both introductory and advanced microbiology classes. The Meiosis CI has seventeen multiple choice questions that target student misconceptions about ploidy, DNA replication, the timing of events during meiosis, gamete formation, what happens to chromosomes during replication, and what “counts” as a chromosome.

![Figure 2-5. The different topics covered by biology concept inventories (N = 19).](image)

Overall, there were 19 concept inventories collected for biology concepts, as shown in Table 2-2. Most of the researchers created their own abbreviations for the inventories. For the inventories without specific abbreviations, ones were created. Both the Biomechanics Concept Inventory and the Biology Concepts Instrument used the abbreviations BCI, so the Biomechanics Concept Inventory will be referred to as the BMCI. Four of the concept inventories were unpublished theses, so the concept inventory is included with the process of the development and the testing of the inventory.
<table>
<thead>
<tr>
<th>Title (Abbreviation)</th>
<th>Subject</th>
<th>Topic</th>
<th>Number of Items</th>
<th>Type of Items</th>
</tr>
</thead>
<tbody>
<tr>
<td>Introductory Molecular and Cell Biology Assessment (IMCA)</td>
<td>Biology</td>
<td>Molecular Biology</td>
<td>24</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Biomechanics Concept Inventory (BCI) in paper but I referred to as BMCI</td>
<td>Biology</td>
<td>Kinesiology</td>
<td>24</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Genetics Concept Assessment (GCA)</td>
<td>Biology</td>
<td>Genetics</td>
<td>25</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Genomic Nursing Concept Inventory (GNCI)</td>
<td>Biology</td>
<td>Genetics</td>
<td>31</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Conceptual Inventory of Natural Selection (CINS)</td>
<td>Biology</td>
<td>Evolution</td>
<td>20</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Evolutionary Development Concept Inventory (EvoDevoCI)</td>
<td>Biology</td>
<td>Evolution</td>
<td>11</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Biology Concepts Instrument (BCI)</td>
<td>Biology</td>
<td>Molecular Biology</td>
<td>30</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Osmosis and Diffusion Conceptual Assessment (ODCA)</td>
<td>Biology</td>
<td>Diffusion</td>
<td>16</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Genetics Literacy Assessment Instrument (GLAI)</td>
<td>Biology</td>
<td>Genetics</td>
<td>31</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Greenhouse Effect Concept Inventory (GECI)</td>
<td>Biology</td>
<td>Sustainability</td>
<td>26</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Sustainable Energy Concept Inventory (SECI)</td>
<td>Biology</td>
<td>Sustainability</td>
<td>27</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Host Pathogen Interactions Concept Inventory (HPICI)</td>
<td>Biology</td>
<td>Microbiology</td>
<td>18</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Measure of Understanding of Macroevolution (MUM)</td>
<td>Biology</td>
<td>Evolution</td>
<td>28</td>
<td>Multiple Choice and Open-Ended</td>
</tr>
<tr>
<td>Diffusion and Osmosis Diagnostic Test (DODT)</td>
<td>Biology</td>
<td>Diffusion</td>
<td>12</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Genetics Concept Inventory (GCI)</td>
<td>Biology</td>
<td>Genetics</td>
<td>38</td>
<td>Multiple Choice, Open-Ended and Matching</td>
</tr>
<tr>
<td>Meiosis Concept Inventory (Meiosis CI)</td>
<td>Biology</td>
<td>Molecular Biology</td>
<td>17</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Dominance Concept Inventory (DCI) but I use DoCI</td>
<td>Biology</td>
<td>Genetics</td>
<td>16</td>
<td>MC, Open-Ended and Matching</td>
</tr>
<tr>
<td>Genetic Drift Inventory (GeDI)</td>
<td>Biology</td>
<td>Genetics</td>
<td>22</td>
<td>True-False</td>
</tr>
<tr>
<td>Biogeochemistry Concept Inventory (BG-CI)</td>
<td>Biology</td>
<td>Geology</td>
<td>32</td>
<td>Multiple Choice</td>
</tr>
</tbody>
</table>
The most concept inventories have been developed for general chemistry because of the large number of concepts covered in that course, as shown in Figure 2-6. There have been very few concept inventories developed for the higher level chemistry classes. Students are expected to have a more complete knowledge of chemistry by the time they leave these classes, but not much research has been done into whether they actually have this knowledge or not. The first physical chemistry concept inventory was released in 2014, and is the first concept inventory to look at student knowledge above a biochemistry level.  

![Figure 2-6. The breakdown of chemistry concept inventories by subject and topic.](image)

A breakdown of the different chemistry topics for which concept inventories have been developed is shown in Figure 2-6. The most concept inventories have been developed for bonding/structures or are multi-topic. However, there are only 3 inventories for each of those categories. There are a wide range of topics covered by chemistry concept inventories, but there are more topics still taught in chemistry classes. There are a wider range of topics covered by chemistry concept inventories than biology.
concept inventories. So even though more concept inventories for biology exist, they
cover the same seven topics. For example, there are six concept inventories devoted just
to genetics knowledge.

As shown in Table 2-3, there were 12 inventories collected for chemistry and
three collected for biochemistry. There are two different chemistry concept inventories
that use the CCI abbreviation. Both of these inventories are also designed for a general
chemistry target population and are multi-topic. However, the current literature uses CCI
for the Chemical Concepts Inventory designed by Mulford and Robinson and uses ChCI
for the Chemistry Concept Inventory designed by Krause et al. To be consistent, the same
abbreviations are used here.

After collecting concept inventories, an analysis of the quality of each biology,
chemistry, and biochemistry concept inventory was completed. The process through
which misconceptions were identified was considered, as well as the types of reliability
and validity reported for each instrument. The tables used for analysis of the inventories
are included below.
Table 2-3. A List of the Biochemistry and Chemistry Concept Inventories Collected

<table>
<thead>
<tr>
<th>Title (Abbreviation)</th>
<th>Subject</th>
<th>Topic</th>
<th>Number of Items</th>
<th>Type of Items</th>
</tr>
</thead>
<tbody>
<tr>
<td>Enzyme-Substrate Interactions Concept Inventory (ESICI)</td>
<td>Biochemistry</td>
<td>Enzymes</td>
<td>15</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Molecular Life Sciences Concept Inventory (MLS-CI)</td>
<td>Biochemistry</td>
<td>Biochemistry</td>
<td>26</td>
<td>True-False</td>
</tr>
<tr>
<td>Foundational Concepts for Biochemistry (FCBC)</td>
<td>Biochemistry</td>
<td>Biochemistry</td>
<td>24</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Chemical Concepts Inventory (CCI)</td>
<td>Chemistry</td>
<td>Multi-topic</td>
<td>22</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Bonding Representations Inventory (BRI)</td>
<td>Chemistry</td>
<td>Bonding/Structures</td>
<td>23</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Thermochemistry Concept Inventory (TCI)</td>
<td>Chemistry</td>
<td>Thermochemistry</td>
<td>12</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>General, Organic and Biological Chemistry Topic Inventory (GOBCTI)</td>
<td>Chemistry</td>
<td>Multi-topic</td>
<td>45</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Chemistry Concept Inventory (ChCI)</td>
<td>Chemistry</td>
<td>Multi-topic</td>
<td>20</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Implicit Information from Lewis Structures Instrument (IILSI)</td>
<td>Chemistry</td>
<td>Bonding/Structures</td>
<td>1</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Organic Chemistry Students' Alternative Conceptions Related to Acid Strength (ACID I)</td>
<td>Chemistry</td>
<td>Acid/Base</td>
<td>9</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Redox Concept Inventory (ROXCI)</td>
<td>Chemistry</td>
<td>Redox</td>
<td>18</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Flame Test Concept Inventory (FTCI)</td>
<td>Chemistry</td>
<td>Atomic Emission</td>
<td>19</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Acid-Base Reactions Concept Inventory (ABCI)</td>
<td>Chemistry</td>
<td>Acid/Base</td>
<td>28</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Molecular Attractions Concept Inventory (MACI)</td>
<td>Chemistry</td>
<td>Bonding/Structures</td>
<td>24</td>
<td>Multiple Choice</td>
</tr>
<tr>
<td>Quantum Chemistry Concept Inventory (QCCI)</td>
<td>Chemistry</td>
<td>Quantum Mechanics</td>
<td>12</td>
<td>Multiple Choice</td>
</tr>
</tbody>
</table>
Table 2-4. Information on the development of biology concept inventories.

<table>
<thead>
<tr>
<th>Literature Basis</th>
<th>Question/Statement Development</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Literature Basis</td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td>IMCA</td>
<td>X</td>
</tr>
<tr>
<td>BMCI</td>
<td>X</td>
</tr>
<tr>
<td>GCA</td>
<td>X</td>
</tr>
<tr>
<td>GNCI</td>
<td>X</td>
</tr>
<tr>
<td>CINS</td>
<td>X</td>
</tr>
<tr>
<td>EvoDevoCI</td>
<td>X</td>
</tr>
<tr>
<td>BCI</td>
<td>X</td>
</tr>
<tr>
<td>ODCA</td>
<td>X</td>
</tr>
<tr>
<td>GLAI</td>
<td>X</td>
</tr>
<tr>
<td>GECI</td>
<td>X</td>
</tr>
<tr>
<td>SECI</td>
<td></td>
</tr>
<tr>
<td>HPICI</td>
<td>X</td>
</tr>
<tr>
<td>MUM</td>
<td>X</td>
</tr>
<tr>
<td>DODT</td>
<td>X</td>
</tr>
<tr>
<td>GCI</td>
<td>X</td>
</tr>
<tr>
<td>Meiosis CI</td>
<td>X</td>
</tr>
<tr>
<td>DoCI</td>
<td>X</td>
</tr>
<tr>
<td>GeDI</td>
<td>X</td>
</tr>
<tr>
<td>BGC-CI</td>
<td>X</td>
</tr>
</tbody>
</table>
Table 2-5. Information about the validity and reliability of biology concept inventories.

<table>
<thead>
<tr>
<th>Construct</th>
<th>Criterion</th>
<th>Internal Consistency</th>
<th>Test - Retest</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Face</td>
<td>Content</td>
<td>Discriminant</td>
</tr>
<tr>
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<td>X</td>
<td></td>
</tr>
<tr>
<td>BMCI</td>
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<td></td>
<td></td>
</tr>
<tr>
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<td>X</td>
<td>X</td>
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</tr>
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<tr>
<td>CINS</td>
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<td></td>
</tr>
<tr>
<td>EvoDevoCI</td>
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<td>X</td>
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<td>X</td>
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</tr>
<tr>
<td>DODT</td>
<td>X</td>
<td></td>
<td></td>
</tr>
<tr>
<td>GCI</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Meiosis CI</td>
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<td>X</td>
<td></td>
</tr>
<tr>
<td>DoCI</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>GeDI</td>
<td>X</td>
<td></td>
<td></td>
</tr>
<tr>
<td>BGC-CI</td>
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</table>
Table 2-6. Information about the development of chemistry and biochemistry concept inventories.

<table>
<thead>
<tr>
<th>Literature Basis</th>
<th>Research with Target Population</th>
<th>Author Derived</th>
<th>Educator Informed</th>
</tr>
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<td>Interviews</td>
<td>Pilot Study</td>
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<td>MLS-C1</td>
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</tr>
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<td>FCBC</td>
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<td></td>
</tr>
<tr>
<td>CCI</td>
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<td>X</td>
<td>X</td>
</tr>
<tr>
<td>BRI</td>
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<td>X</td>
<td>X</td>
</tr>
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</tr>
<tr>
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</tr>
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</tr>
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</tr>
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<tr>
<td>TISC</td>
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</tr>
<tr>
<td>IILSI</td>
<td>X</td>
<td>X</td>
<td></td>
</tr>
<tr>
<td>ACID 1</td>
<td>X</td>
<td></td>
<td></td>
</tr>
<tr>
<td>ROXCI</td>
<td>X</td>
<td>X</td>
<td></td>
</tr>
<tr>
<td>FTCI</td>
<td>X</td>
<td></td>
<td>X</td>
</tr>
<tr>
<td>ABCI</td>
<td>X</td>
<td>X</td>
<td></td>
</tr>
<tr>
<td>MACI</td>
<td>X</td>
<td></td>
<td></td>
</tr>
<tr>
<td>GOBCTI</td>
<td>X</td>
<td>X</td>
<td></td>
</tr>
<tr>
<td>QCCI</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Table 2-8. Percentage of biology, chemistry, and biochemistry concept inventories that reported the following sources for their question/statement development (N = 36).

<table>
<thead>
<tr>
<th>Question/Statement Development</th>
<th>Percentage of Inventories Reporting this Source</th>
</tr>
</thead>
<tbody>
<tr>
<td>Literature Basis</td>
<td>77%</td>
</tr>
<tr>
<td>Research with Target Population Interviews</td>
<td>72%</td>
</tr>
<tr>
<td>Pilot Study</td>
<td>52%</td>
</tr>
<tr>
<td>Author Derived</td>
<td>11%</td>
</tr>
<tr>
<td>Educator Informed</td>
<td>47%</td>
</tr>
</tbody>
</table>

Based on the analysis shown in Table 2-8, 77% of concept inventories analyzed reported a literature basis for their question/statement development. In addition, 72% of the researchers also did interviews with their target population. It should be noted that these categories are not mutually exclusive. Researchers often utilize more than one method of question/statement development in order to ensure that any incorrect ideas identified are misconceptions and not the result of a single student’s personal experience with the material. It was disappointing to discover how few researchers took their own educational experiences into account when identifying the misconceptions for specific topics. Based on interviews with both students and educators, researchers might be able to reflect on their own educational experience. Another way to identify common misconceptions among students is to interview the educators. The educators see many students pass through the course, and know what topics students typically have trouble understanding.
Table 2-9. Percentage of biology, chemistry, and biochemistry concept inventories that reported the following psychometrics. (N = 36)

<table>
<thead>
<tr>
<th>Forms of Validity</th>
<th>Percent of Inventories Reporting this form</th>
</tr>
</thead>
<tbody>
<tr>
<td>Item Statistics</td>
<td></td>
</tr>
<tr>
<td>Difficulty</td>
<td>61%</td>
</tr>
<tr>
<td>Discrimination</td>
<td>63%</td>
</tr>
<tr>
<td>Construct Validity</td>
<td></td>
</tr>
<tr>
<td>Face</td>
<td>44%</td>
</tr>
<tr>
<td>Content</td>
<td>63%</td>
</tr>
<tr>
<td>Discriminant</td>
<td>25%</td>
</tr>
<tr>
<td>Convergent</td>
<td>16%</td>
</tr>
<tr>
<td>Criterion Validity</td>
<td></td>
</tr>
<tr>
<td>Concurrent</td>
<td>8%</td>
</tr>
<tr>
<td>Predictive</td>
<td>5%</td>
</tr>
</tbody>
</table>

Not many of the concept inventory development papers analyzed report convergent or discriminant validity, and most mention leaving further psychometrics to future research (see Table 2-9). Of the inventories analyzed, 61% report item difficulty, which represents the percentage of students who answered each item correctly. 63% of inventories analyzed reported discrimination, which expresses how well each item discriminates between students with higher knowledge levels and those with lower knowledge levels. Face and content validity were reported by 44% of the inventories and 63% of the inventories, respectively. Since face and content validity are traditionally checked by experts in the content, they are the easiest forms of validity to check.

Concurrent validity was only reported by 8% of the inventories.

Table 2-10. Percentage of biology, chemistry, and biochemistry concept inventories that reported the above psychometrics.

<table>
<thead>
<tr>
<th>Reliability</th>
<th>Percentage of Inventories Reporting this Form</th>
</tr>
</thead>
<tbody>
<tr>
<td>Internal Consistency</td>
<td></td>
</tr>
<tr>
<td>Cronbach alpha</td>
<td>52%</td>
</tr>
<tr>
<td>K-R 20</td>
<td>11%</td>
</tr>
<tr>
<td>Split-half</td>
<td>2%</td>
</tr>
<tr>
<td>Test-Retest</td>
<td>47%</td>
</tr>
</tbody>
</table>

More inventories (41%) reported a Cronbach $\alpha$ than a Kuder-Richardson 20. Perhaps that is because running the calculations for the KR-20 statistic requires researchers to
convert their data into right-wrong answer selections. This prevents researchers from focusing on the misconception identification that can be determined by analysis of the selection of incorrect options for each student and for the class overall. Since split-half reliability is only one comparison of the divided data set rather than multiple comparisons of all of the possible groupings (as with Cronbach $\alpha$ and KR-20), it is logical that it is reported less often than the multiple comparison options. Only 47% of the inventories analyzed reported a test-retest statistic. This means that researchers are checking to ensure that students answer the questions in the same manner over time.

2.6 Conclusions & Implications

There were 51 science inventories gathered through various sources and 36 were analyzed to determine the quality of biology, chemistry, and biochemistry inventories that are available. While there are more biology inventories developed than chemistry inventories, the chemistry inventories cover a wider range of topics. Overall, there are more inventories that take previously identified misconceptions from the literature into consideration than inventories that utilized the author’s own misconceptions. Less than half of the concept inventories analyzed reported a test-retest statistic.

Very few of the concept inventories analyzed follow best practices in the development and testing. When concept inventories were being developed in the beginning of the movement, it appears that researchers were more focused on creating the instruments than running all the analyses. Most of the inventories have only been found to be reliable and valid for one population, even if they claim to cover concepts taught in multiple courses. One of the inventories even takes some questions from a previous inventory on the same topic. The lack of reported concurrent and predictive validity
show a need for researchers to check into a relationship between the concepts measured on inventories and those measured by classroom tests.

Some of the inventories collected mentioned that further psychometric analysis was to be completed in the future or done by other people. In most of these cases, no future psychometric analysis was completed, but there are some cases where people looked at the inventory again. More analysis of most of these inventories should be completed. Researchers need to conduct these analyses themselves in order to ensure that the inventory they developed is up to the same standards as the rest of the inventories that are available.

During the collection of these concept inventories, a decision was made to create a database of created concept inventories for biology, chemistry, and biochemistry. The accessibility of these inventories is a major roadblock that prevents more concept inventories from being implemented in the classroom. The database will provide a link to the article discussing the development of the inventory or, if an article has not been published, there will be contact information. The actual inventory itself will not be included in the database because many inventories are copyrighted. Additionally, if inventories can be found by students, it lessens the validity of the inventory. If a student can google an inventory and get the answers, then the inventory is no longer measuring student knowledge.

Concept inventories are only to be used to inform instruction. The incorrect answer options focus on incorrect ideas that students commonly hold. Due to the formulation process of answer options, students are more likely to select incorrect answers. These inventories should not be used to assign grades to students. The results
of concept inventories should be used to create new methods of addressing the concepts or to determine material that should be explained in more detail. By providing a central location of concept inventories, more chemistry education research can be put into practice and more misconceptions can be corrected.
2.7 References


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Chapter 3: DEVELOPMENT OF A GUIDED INQUIRY ACTIVITY TO ADDRESS STUDENT MISCONCEPTIONS ABOUT ENZYME-SUBSTRATE INTERACTIONS

3.1 Introduction

Enzymes are one of the most prevalent topics covered in biochemistry courses, as the topic is foundational for more advanced topics such as enzyme kinetics and metabolism. Therefore having a solid understanding of this topic is paramount.\(^1\) Students are often first taught the lock and key model of enzyme interaction in their high school or introductory biology classes. Upon reintroduction to enzymes in a biochemistry course, students rely on this prior knowledge of the lock and key model for an explanation of the detailed nature of enzyme-substrate interactions, so much so, that they often refuse to adopt more scientifically accepted explanations of the interactions into their mental model, resulting in a range of misconceptions.\(^2\)

To further confound the issue of students’ understandings of enzyme-substrate interactions is the extent to which the field of biochemistry relies on representations to communicate the subject.\(^3,4\) Biochemistry requires students to develop their skills of interpreting and communicating through the use of representations (i.e. visual literacy).\(^5\) Many studies have investigated the impact of representations on biochemistry students understanding and have shown that students have great difficulty when interpreting representations which often results in significant misconceptions.\(^6\text{-}12\) In order to more efficiently measure the impact of representations and the misconceptions students bring into the biochemistry classroom, several concept inventories have been developed looking at foundational concepts, general topics of biochemistry and molecular biology, and enzyme-substrate interactions.\(^6,13,14\)
The primary focus of biochemistry education research up to this point has been the diagnosis and measurement of misconceptions; however, research needs to now move towards determining how to correct the misconceptions. One way to do this is to provide students with a social constructivist learning environment where students use their previous experiences with the material and any social interactions with others to create a better connection to the material.\textsuperscript{15} Process-Oriented Guided-Inquiry Learning (POGIL) activities are designed to allow students to build on their prior knowledge while working in teams and managing themselves. The instructor serves as a facilitator for the class, instead of as a traditional lecturer.\textsuperscript{16} The facilitator is there to listen to the groups and to provide guidance for students if necessary. Effective facilitation requires the ability to redirect and rephrase questions as well as managing student frustration.\textsuperscript{17}

POGIL activities follow the three phase learning cycle.\textsuperscript{18} In the first phase of the learning cycle, students \textit{explore} a concept through a model that is related to their prior knowledge in the subject and look for patterns. They may also be asked to explain any patterns that arise or the relationships that they see in the model. In the second phase of the learning cycle, students continue \textit{forming the concept} through further exploration of a model and continuing explanation of patterns. In the third phase, students are asked to \textit{apply} the concept they have developed from the first two phases to a new situation or relationship. A complete book of biochemistry POGIL activities have been developed, but only a few activities address the concept of enzymes and there are no POGIL activities that specifically address enzyme-substrate interactions.\textsuperscript{19}

Therefore, the goal of this project was to create a guided inquiry activity that would more effectively correct students’ misconceptions about enzyme-substrate
interactions than traditional lecture. The authors received Institutional Review Board approval for all parts of this research, and all applicable rules and regulations were followed. Students who participated in this study also signed informed consent forms (Appendix B). The specific research questions guiding this manuscript were: (1) What misconceptions do biochemistry students have regarding enzyme-substrate interactions following traditional instruction? and (2) How effective is the developed activity at correcting these misconceptions compared to traditional lecture?

3.2 Activity Design and Implementation

3.2.1 Identification of Misconceptions in Traditional Instruction

In order to identify the most persistent misconceptions related to enzyme-substrate interactions that could be addressed in the activity, the Enzyme-Substrate Interactions Concept Inventory (ESICI) was administered to students in a one-semester biochemistry survey course for non-biochemistry majors using a pre-post instruction design during the spring of 2014 at a large comprehensive university in the United States. The class was composed of 54.72% females and 18.87% African American, 5.66% Asian/Pacific Islander, 3.77% Hispanic, 64.15% Caucasian, 1.89% Middle Eastern, and 5.66% other. The professor was observed in class during instruction of the enzyme unit to determine the depth and breadth of the topic covered. The professor utilized PowerPoints as their teaching medium. The progression of unit topics are as follows: types of enzymes, kinetics, inhibition, and serine proteases.

Unfortunately the semester was interrupted by snow days and the professor lost a week of class time in the middle of allosteric regulation resulting in the unit lasting 4 class periods as opposed to 6 class periods. As enzyme-substrate interactions were
introduced, the professor stated that the lock and key model had been disproven and that induced fit is the correct model for these interactions. Active sites were identified and described in detail, but specificity pockets were not mentioned until serine proteases were covered. Specificity pockets were later referred to as “substrate pockets” and “active-site binding cavities”. The post administration of the ESICI occurred two weeks after examination over the enzyme unit, in order to determine what misconceptions remained after both instruction and assessment of the unit. To establish validity of the data obtained from the ESICI, students ($N = 4$) from the course were interviewed using a think-aloud protocol to ensure they were answering the questions as originally intended (Appendix B). These students were the ones who volunteered for interviews. Based on the student interviews and persistent misconceptions identified from the ESICI, 3 specific misconceptions were selected to be the focus of the guided-inquiry activity: (1) water cannot be a substrate, (2) the difference between an active site and specificity pocket, and (3) enzyme-substrate specificity.

### 3.2.2 Activity Design

Guided inquiry activities revolve around model exploration; therefore, the importance of model selection cannot be underestimated. The major consideration for the model in the activity was that the model had to be already covered in a biochemistry survey course so not to take away from the content of the course. In addition, the model had to exemplify the topic areas that were to be focused on in the activity. As such, chymotrypsin was selected due to it fitting both criteria. Water serves as the second substrate in the mechanism of chymotrypsin and the enzyme has a specificity pocket and an active site, which contribute to the substrate specificity of the enzyme.
Chymotrypsin was selected as the model for this activity because it is taught in many biochemistry courses. As a serine protease, chymotrypsin contains an active site that is conserved across the class of enzymes and the mechanism of the enzyme is also conserved.

The activity was designed with the expectation that students had completed the assigned reading presenting enzymes and enzyme mechanisms prior to class and knew amino acid structures. The exploration and formation of concept stages of the activity were created to be completed in a 50 minute class period with a four-question application portion to be assigned as homework, and is included in Appendix C. There were 3 models designed to encourage critical thinking by the students. Model 1 is used to introduce students to the mechanism of chymotrypsin and to help students identify water as a possible substrate for an enzyme. In addition, students are asked to identify the amino acids in the active site (depicted in a figure) and describe what happens in each step of the reaction. Model 2 provides students the opportunity to explore the specificity pocket of chymotrypsin by considering what types of interactions occur in the specificity pocket. Based on the type of interactions identified, different amino acids and substrates are presented for identification as possible substrates. In model 3, students compare the active site to the specificity pocket in chymotrypsin. In addition to looking at the differences in the active site and specificity pocket, students are asked to consider what type of reactions or interactions occur in each location.

The application section is designed for students to be given the opportunity to apply their newly acquired knowledge to a new system. The enzyme trypsin was chosen for this task as it is in the serine protease family of enzymes providing the same reaction
mechanism but different substrate specificity. Students are given the specificity pocket for trypsin and asked to describe the mechanism based on the mechanism for chymotrypsin. This shows the importance of the conserved catalytic triad in the active site. In order to show students that specificity pockets are different for different enzymes, they are also asked to compare the specificity pocket for trypsin to the specificity pocket for chymotrypsin, as well as determine which amino acids will fit in the specificity pocket for trypsin.

Once developed, the activity was sent to eight biochemistry professors at universities across the United States to ensure content validity, accuracy, and that the activity was at the level of a biochemistry survey course. Based on their feedback, questions were edited for content and certain questions were eliminated to accommodate the activity being completed in the time allotted.

As further checks of validity and brevity of the activity, two focus groups were conducted (see Appendix B for interview protocol). One focus group consisted of three chemistry graduate students who had taken biochemistry during their undergraduate careers and who had all of the applicable knowledge required to complete the activity. They served as a check that the models utilized in the activity contained information that would be covered in a biochemistry survey course, as well as to check the clarity of the questions. The second focus group consisted of undergraduate students who had not taken a biochemistry course previously and were not currently enrolled in such course. These students did not have all of the relevant prior knowledge that the biochemistry students would have, so some information was provided to them such as a list of amino
acid structures and abbreviations. The primary role of this focus group was to check for the clarity of the questions.

3.2.3 Implementation

The activity was implemented in the same one semester survey of biochemistry course primarily for undergraduate non-biochemistry majors during summer 2014. A different professor taught the course but followed similar presentation style as the previous professor. Of the 29 students enrolled in the class, 24 were undergraduates and 3 were graduate students. The class consisted of 72.41% females and was 34.48% African American, 6.90% Asian/Pacific Islander, 10.34% Hispanic, 44.83% Caucasian, and 3.45% other. The activity was implemented as a replacement for the chymotrypsin lecture during the enzyme unit which was taught as the second unit of the course. The ESICI was again administered pre and post instruction of the enzyme unit.

On the day when the activity was scheduled to be utilized in class, the professor started the class with lecture. Since the course was during the summer, the class was two and a half hours long. The plan was for the professor to continue with his lecture from the previous class, and end 50 minutes early so that the activity could be distributed; however, due to the lecture taking more time than expected, the students were only given 20 minutes to complete the activity in class. During this time, the professor and the author, both novices at facilitation of guided inquiry activities, facilitated the activity using guided questions. Students were divided into groups of three to four students in order to ensure that all students could be heard. The majority of students were able to get through Model 1. In order to ensure that students would still benefit from the entirety of the activity even though time was constricted, the remainder of the activity was assigned
as homework. Forty-four percent of students in the class were not present in class on the
day the activity was given, but who did complete the survey both times. This allowed for
a comparison group within the same class.

3.3 Effectiveness of the Activity

3.3.1 Observations and Feedback

During the administration of the activity, students had difficulty determining the
active site and the enzyme in Model 1 which dramatically decreased the amount of
material they could get through in the 20 minutes provided in class. Clarifications have
subsequently been made to the image in Model 1 to correct for this confusion. In
addition, it became clear that students did not read the chapter in the textbook before
coming to the lecture that day because they did not know what an enzyme was or know
the amino acid abbreviations. Even though students did not complete the activity in
class, the professor still gave anecdotal evidence that the students who used the activity
were more knowledgeable than the students who did not use it. For instance, students did
not use active site and specificity pocket interchangeably as they had in the past, and they
used the two terms correctly.

Students also provided feedback on the activity. In the feedback survey (included
in Appendix B), students were asked about the strengths of the activity, areas for
improvement in the activity, and any insights that they gained from the activity. On
average, students reported that the activity was somewhat helpful, rating it a 4 on a scale
from 1-5, with 1 being not helpful and 5 being very helpful. Being able to discuss the
activity with other students in the class was identified as a strength of the activity by 33%
of the students, and 43% reported that the visuals were one of the most helpful aspects of
the activity. One student mentioned that “[d]iscourse between my peers was valuable as we were able to bounce ideas/thoughts off of each other.” Fifty-two percent of students reported that they gained insights about the differences between an active site and a specificity pocket, as well as what helps to determine enzyme specificity. Based on student comments concerning the length of activity, it was shortened and several questions were edited for clarity.

3.3.2 Traditional Instruction’s Impact on Student Understanding of Enzyme-Substrate Interactions

Individual students’ scores on the ESICI who were taught using traditional instruction ranged from 1-10 on the Pretest and 4-12 on the Posttest. As shown in Table 3-1, students’ scores on the inventory were not found to be significantly different from a normal distribution for both the Pretest (Kolmogorov-Smirnov statistic, D (33) = 0.128, p > 0.05), and for the Posttest (Kolmogorov-Smirnov statistic, D (33) = 0.148, p > 0.05). Because both samples meet the appropriate assumptions, parametric techniques were used for data analysis. A paired samples t-test was conducted to evaluate the impact of traditional lecture on students’ score on the ESICI. There was a statistically significant increase in ESCI scores from Pretest (M = 5.67, SD = 2.13) to Posttest (M = 7.97, SD = 2.04), t (32) = 5.89, p < 0.005 (two-tailed). The mean increase in ESICI scores was 2.30 with a 95% confidence interval ranging from 1.51 to 3.10. The eta squared statistic (0.51) indicated a moderate effect size.
Table 3-1. Descriptive statistics and normality tests for the traditional lecture group.

<table>
<thead>
<tr>
<th>Traditional Lecture</th>
<th>N</th>
<th>Mean (M)</th>
<th>Median (MD)</th>
<th>Standard Deviation (SD)</th>
<th>Skewness</th>
<th>Kurtosis</th>
<th>K-S Statistic</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pretest</td>
<td>33</td>
<td>5.67</td>
<td>6.00</td>
<td>2.131</td>
<td>-0.112</td>
<td>-0.393</td>
<td>0.128</td>
<td>0.185</td>
</tr>
<tr>
<td>Posttest</td>
<td>33</td>
<td>7.97</td>
<td>8.00</td>
<td>2.038</td>
<td>-0.216</td>
<td>-0.802</td>
<td>0.148</td>
<td>0.065</td>
</tr>
</tbody>
</table>

Figure 3-1. The percentage of the students who answered correctly on the pre and post inventory for the traditional lecture class.

As discussed above three persistent misconceptions were uncovered based on the ESICI results depicted in Figure 3-1. Question 3 and 7 both address the misconception of enzyme-substrate specificity. Question 3 asks students to select the substrate that could interact with the active site type of structure provided as described in words not chemical structure. Question 7 is similar to question 3 by asking students to consider possible active sites for a given substrate structure, depicting the structure for both the active sites and the substrate. Question 8 asks students to identify which of the given options is not a substrate for an enzyme. Based on the students interviewed, the misconception that water could not be a substrate was selected most often. Question 11 asks students to identify a
representation as either an active site or a specificity pocket, while also selecting an explanation for their choice.

3.3.3 Activity’s Impact on Student Understanding of Enzyme-Substrate Interactions

Individual students’ scores on the ESICI who used the activity ranged from 1-9 on the Pretest and 3-12 on the Posttest. As shown in Table 3-2, students’ scores on the inventory were found to be significantly different from a normal distribution for the Pretest (Kolmogorov-Smirnov statistic, D (29) = 0.171, p < 0.05), but were not found to be significantly different from a normal distribution for the Posttest (Kolmogorov-Smirnov statistic, D (29) = 0.135, p > 0.05). Therefore, due to the lack of ability for the pretest sample to meet the assumption of normality, nonparametric techniques were used for data analysis including pretest scores. A Wilcoxon signed-ranks test indicated a significant difference in the Pretest and Posttest scores for the group that used the activity as homework (z = -3.347, p < 0.005).

Table 3-2. Descriptive statistics and normality tests for the activity as homework group

<table>
<thead>
<tr>
<th>Activity as Homework</th>
<th>N</th>
<th>Mean</th>
<th>Median</th>
<th>Standard Deviation</th>
<th>Skewness</th>
<th>Kurtosis</th>
<th>K-S statistic</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pretest</td>
<td>29</td>
<td>5.55</td>
<td>6</td>
<td>2.080</td>
<td>-0.475</td>
<td>-0.581</td>
<td>0.171</td>
<td>0.030</td>
</tr>
<tr>
<td>Posttest</td>
<td>29</td>
<td>7.52</td>
<td>8</td>
<td>2.309</td>
<td>-0.070</td>
<td>-0.362</td>
<td>0.135</td>
<td>0.192</td>
</tr>
</tbody>
</table>
Figure 3-2. The percentage of students who answered correctly on the pre and post inventory for the activity as homework class.

Based on the results of a pre and post survey shown in Figure 3-2, the activity did correct the misconception that water cannot be a substrate for an enzyme. Although the results for items 8 and 11 are still not as high as hoped the item analysis for these questions provides additional information. When looking at the item analysis for question 8, 37.50% of students who used the activity shifted away from the idea that water could not be a substrate for an enzyme, compared to only 23.08% of students who did not use the activity. Out of the students who utilized the activity, 31.25% of them selected that a large protein could not be a substrate, but only 7.68% of students who did not utilize the activity selected this response. There was no change in the percent of students that selected the correct answer for either group.

Students were also more able to differentiate between the specificity pocket and the active site of an enzyme, based on the results of the survey. There was no change in the percentage of students who selected the correct answer, but there was a 6.25% increase in the percent of students who selected the answer that identifies the figure as a
specificity pocket for the wrong reason. For the students who did not use the activity, there was a 15.38% decrease in selection of the correct answer, and a 23.08% decrease in selection of the other answer option that identified the structure as a specificity pocket. In fact, 38.46% of student who did not use the activity incorrectly identified the structure as an active site.

3.3.4 Comparing the Impact on Student Understanding

In order to determine the effectiveness of the activity at correcting misconceptions, the student data was compared to the data for the students who had traditional lecture. According to the Mann-Whitney U tests that were conducted, there were no significant differences between the Pretest scores for the traditional lecture group (MD = 6, N = 33) and the activity as homework group (MD = 6, N = 29), (U = 473, z = -0.079, p = 0.937), and there were no significant differences between the Posttest scores for the traditional lecture group (MD = 8, N = 33) and the activity as homework group (MD = 8, N = 29), (U = 417, z = -0.877, p = 0.381). Because there seems to be no significant difference between the pre and post test scores between the students in each instructional group, the normalized change in percent correct was compared for each group.

Figure 3-3 shows the normalized change in the percent correct for each item on the inventory. The misconceptions that are the focus of the activity are contained within questions 3, 7, 8, and 11. Comparatively, the activity improved student’s ability to interpret enzyme-substrate specificity according to the results for questions 3 and 7. Figure 3-3 also indicates that questions 8 and 11 need additional analysis to explain the impact of the activity on student understanding, as students who used the activity scored
worse on the posttest for these questions. The item analysis of question 8 shows that the students who used the activity shifted away from the misconception that water cannot be a substrate for an enzyme. However, students did not correct their misconception about possible substrates entirely. Instead, a new misconception was created that a large protein could not be a substrate for an enzyme.

**Figure 3-3.** The normalized change in percent correct on the concept inventory for the traditional lecture class and the class that utilized the activity.

The item analysis for question 11 indicates that while students who used the activity did not select the correct answer after using that activity, they did select the most “correct” wrong answer. They were able to identify the depiction as a specificity pocket, but their rationale was incorrect. The activity aimed to introduce students to the phrase “scissile bond” and provide a definition as well as the function of the bond in relation to how the substrate interacts with the enzyme.
3.4 Discussion and Conclusions

The goal of this project was to design a guided inquiry activity to correct selected misconceptions identified from a survey of biochemistry students in a traditional lecture-based course. The results showed students do learn from the developed activity. Although the scores did not increase significantly for the students who used the activity, the questions that contained the target misconceptions did show a shift in the answer selections.

A limitation of this study was the assumption that students did their reading before the lecture. Since the reading was assigned in the syllabus, the researchers believed that the students would do the reading. It was also assumed that students knew their one letter amino acid abbreviations. Since this activity was designed for a one semester, general biochemistry course, future work needs to be done on the effectiveness of the activity in a two semester biochemistry course.

Although there is no significant difference in the post survey score for the students who had traditional lecture and those who used the activity, the activity did correct the selected misconceptions. Students were able to identify that water can be a substrate for an enzyme, as well as differentiate between an active site and a specificity pocket. The activity does correct the targeted misconceptions more effectively than the traditional lecture method of instruction, as shown by the normalized change in percent correct for each instructional method.

3.5 Implications for Future Research and Practitioners

There has not been much research into the use of guided inquiry activities as homework since they are typically designed to be used in class in groups with a facilitator
Due to the time constraint with the first administration of the activity, future research studies will look at the effectiveness of guided inquiry as an in-class activity with a facilitator vs. as a homework assignment. When these activities are utilized as homework, students are required to do more independent thinking due to neither a facilitator nor other classmates being available for discussion. Guided inquiry learning is based on student discussion and collaboration, and homework does not usually allow for that type of interaction. Therefore, additional study is warranted to see if the same outcomes can be reached when the curriculum is used in this unintended way.

As professors, one of our goals is to communicate concepts effectively to students. By constantly revising instructional methods, we can improve how we teach. Multiple students do not always retain the same information from a lecture. By presenting students with a consistent representation of the material and encouraging collaboration, this activity will be useful for biochemistry professors who cover enzymes in their courses. In addition to helping professors teach the material more effectively, the activity also introduces students to utilizing discussion as a learning method. By sharing their ideas with each other, students gain a better understanding from multiple perspectives. Guided inquiry activities are not an assessment, but rather an alternative teaching strategy. As researchers work to improve the assessment of students’ knowledge, they also must work to develop new and effective instructional methods. This activity can open the door into future exploration of how students understand enzyme-substrate interactions. Previous investigations have provided a way to determine if students understand enzyme-substrate interactions, but not many have looked at improving the instruction method.
3.6 References


Chapter 4: GUIDED INQUIRY ACTIVITIES AS HOMEWORK AND IN CLASS

4.1 Introduction

Professors do not always have time to devote from their traditional lecture planning to create POGIL activities for use in their classrooms. By looking at the use of guided inquiry activities as homework, we hope to show that guided-inquiry activities can be used to increase the amount of independent learning that students are able to do. The development of the guided inquiry activity that addresses student misconceptions about enzyme-substrate interactions is included in Chapter Three. Once the initial testing of the activity was complete, the activity was revised based on recommendations from students and focus groups. There are no significant differences between the version of this activity that was given as homework and the revised version of the activity. There are some wording changes, but the content is the same. Two universities were selected as pilot test locations. The first university is a large, public southeastern university with an average undergraduate population of 23,000 students. This institution used the activity as homework one semester and used it as an in-class activity for the second semester. The second university to use the activity is a small private northeastern university. Both universities offer a one-semester survey of biochemistry course for non-biochemistry majors, and the developed activity was administered in that class.

The goal of this project was to determine if the guided inquiry activity is effective at correcting the targeted misconceptions when used in class, as it was previously determined to be effective as a homework assignment. The authors received Institutional Review Board approval for all parts of this research at both universities (Appendix A), and all applicable rules and regulations were followed. The research questions guiding
this chapter were: (1) Is the developed activity effective at correcting the targeted misconceptions when used as an in-class activity?, (2) Which instructional method is more effective at correcting the targeted misconceptions? and (3) Is this activity effective at correcting misconceptions in other populations when utilized as homework?

4.2 Guided Inquiry Outside of the Classroom

Professors have seen success with guided inquiry laboratory activities in physics, chemistry, biology, and physiology.\(^1\) The laboratory experiments require students to think beyond the “cookbook” level on which some labs are written. Instead of giving students every single step, students are asked to work together and figure out how to run the experiment and what they need to do. There has been some use of guided inquiry activities as homework, but the homework assignments typically involve online simulations. Online simulations do not provide scaffolding, meaning that students do not have a clear set of instructions to follow. Guided inquiry activities are created to accompany these online simulations so that scaffolding is still built into the exercise. The activities are built up to gradually guide students to conclusions while also allowing them to form their own conclusions, and do not require an instructor present.\(^5\)

4.3 Guided Inquiry Activity Comparison

Guided inquiry activities have been developed for use in biochemistry classrooms, but no one has yet looked at the effectiveness of guided inquiry activities used as homework in a biochemistry classroom.\(^6\) There have been no studies that compare the effectiveness of a guided inquiry activity in the classroom and as homework. Occasionally guided inquiry activities include a homework portion as the application part of the learning cycle, but there are no activities created to be offered strictly as
homework. One of the guiding principles of guided inquiry is that students must work together to encourage cooperative learning. In the process of converting a POGIL activity into a guided inquiry homework activity, the student roles are removed from the activity, which removes the cooperative learning portion of the activity. The facilitation role is also removed. The scaffolding that helps students to explore, invent, and apply new concepts is still in place.

4.4 Methodology

The activity was implemented in the same one semester survey of biochemistry course primarily for undergraduate non-biochemistry majors during fall 2014 at University 1. The same professor who used the activity as homework taught the course. Of the 40 students who participated in the activity in the class, 38 were undergraduates. The class consisted of 70% females and was 57.5% Caucasian, 25% African American, 7.5% Hispanic, 5% Asian/Pacific Islander, and 5% Indian/Middle Eastern. The activity was implemented as a replacement for the chymotrypsin lecture during the enzyme unit which was taught as the second unit of the course. The Enzyme-Substrate Interactions Concept Inventory (ESICI) was again administered pre and post instruction of the enzyme unit.

4.5 Effectiveness of the Activity Used In Class

Individual students’ scores on the ESICI who used this activity in-class ranged from 0 to 9 on the Pretest and ranged from 3-12 out of total of 15 on the Posttest. As shown in Table 4-1, students’ scores on the inventory were found to be significantly different from a normal distribution for the Pretest (Kolmogorov-Smirnov statistic, $D(40) = 0.182$, $p < 0.05$), but were not found to be significantly different from a normal
distribution for the Posttest (Kolmogorov-Smirnov statistic, D (40) = 0.115, p > 0.05).

Therefore, due to the pretest scores failing to meet the assumption of normality, nonparametric techniques were used for data analysis including pretest scores. A Wilcoxon signed-ranks test indicated a significant difference in the Pretest and Posttest scores for the students who used the activity in-class (z = 3.436, p < 0.005).

**Table 4-1.** Descriptive Statistics and Normality Tests for the Activity as Homework Group

<table>
<thead>
<tr>
<th>Activity Group</th>
<th>N</th>
<th>Mean</th>
<th>Median</th>
<th>Standard Deviation</th>
<th>Skewness</th>
<th>Kurtosis</th>
<th>K-S statistic</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pretest</td>
<td>40</td>
<td>5.23</td>
<td>6.00</td>
<td>2.293</td>
<td>-0.385</td>
<td>-0.492</td>
<td>0.182</td>
<td>0.002</td>
</tr>
<tr>
<td>Posttest</td>
<td>40</td>
<td>7.23</td>
<td>7.00</td>
<td>2.154</td>
<td>0.212</td>
<td>0.034</td>
<td>0.115</td>
<td>0.198</td>
</tr>
</tbody>
</table>

**Figure 4-1.** The percentage of students who answered correctly on the pre and post inventory for the activity in-class.

Based on the data shown in Figure 4-1, using the activity in class corrected the misconception that water cannot be a substrate for an enzyme. It also showed that students can distinguish between an active site and a specificity pocket. Questions 3 and 7 require further analysis to determine what occurred with these misconceptions. When
looking at the item analysis for question 3, 10% of students shifted away from the correct idea that the size of a substrate is a determining factor in the interactions between an enzyme binding site and a substrate. Students focused instead on the idea that charge is the most important factor in determining if a substrate will bind to an enzyme. The item analysis for question 7 shows that 10% of students shifted away from this same correct idea. Students selected the answer option that corresponds with the misconception of “like dissolving like.” Students were more able to differentiate between an active site and a specificity pocket based on the results for item 11 from the survey. Not only did 7.5% of the students shift towards the correct answer, which identifies the depiction as a specificity pocket, they were also able to identify a scissile bond. Additionally, 15% of students shifted away from identifying the depiction as an active site towards identification of the depiction as a specificity pocket.

4.6 Instructional Technique Comparison

In order to determine which of the instructional methods tested is the most effective, the student data was compared between the groups who utilized traditional lecture (Spring 2014), the activity as homework (Summer 2014), and the activity in class (Fall 2014). According to the Kruskal-Wallis tests conducted, there were no significant differences in the Pretest scores for the traditional lecture group (MD = 6, N = 33), the activity as homework group (MD = 8, N = 29), and the activity in class group (MD = 6, N = 40) ($\chi^2 (2) = 0.601$, $p > 0.05$), and there were no significant differences between the Posttest scores for the traditional lecture group (MD = 8, N = 33), the activity as homework group (MD = 8, N = 29), and the activity in class group (MD = 7, N = 40) ($\chi^2 (2) = 2.603$, $p > 0.05$). Because there seems to be no significant difference between the
pre and post scores between the students in any instructional group, the normalized change in percent correct was compared for each of the groups.

Figure 4-2 shows the normalized change in the percent correct for each item on the inventory. The targeted misconceptions for the activity are represented by questions 3, 7, 8, and 11. As shown in the previous chapter, the activity utilized as homework improved students’ abilities to interpret enzyme-substrate specificity (represented by questions 3 and 7) better than traditional lecture. The activity used in class better corrected the misconception related to the difference between active sites and specificity pockets, as represented by question 11. Students moved toward selection of the correct answer and selection of the most correct wrong answer, both of which identify the depiction as a specificity pocket. Since one aim of the activity is to introduce the term “scissile bond,” the shift of students towards the correct answer shows that the activity makes students aware of the phrase.

Figure 4-2. The comparison of normalized change for the students who learned through traditional lecture, utilized the activity as homework, and utilized the activity in class.
However, the traditional lecture appears to be better at correcting the misconception that water cannot be a substrate for an enzyme according to Figure 4-2. Item analysis for question 8 for all three groups showed that all three types of instruction, students moved away from selecting that water could not be a substrate. They did not shift towards the correct answer, which is why the normalized change for question 8 cannot provide all the information needed.

4.7 Does the Activity as Homework Work at a Different University?

The activity was implemented in a one semester survey of biochemistry course primarily for undergraduate non-biochemistry majors during fall 2014 at a second university (University Two). The professor who taught the class utilized the activity as homework. All 12 of the students who utilized the activity as homework were undergraduates. The class consisted of 58.33% females and was 50% Caucasian, 16.67% Indian/Middle Eastern, 16.67% Hispanic, and 8.33% Asian/Pacific Islander. The activity was implemented as a replacement for the chymotrypsin lecture during the enzyme unit. The ESICI was again administered pre and post instruction of the enzyme unit.

Individual students’ scores on the ESICI ranged from 3-10 on the Pretest and 5-12 out of a total of 15 on the Posttest. As shown in Table 4-2, students’ scores on the inventory were found to be significantly different from a normal distribution for the Pretest (Kolmogorov-Smirnov statistic, $D (12) = 0.269, p < 0.05$), but not for the Posttest (Kolmogorov-Smirnov statistic, $D (12) = 0.157, p > 0.05$). Because one of the two samples does not meet the assumption of normality, nonparametric techniques were used for the data analysis. A Wilcoxon signed-ranks test indicated a significant difference between the Pretest and Posttest scores for the students who used the activity as
homework at University Two (z = -2.330, p < 0.05). This means that there is a significant difference in student scores on the Pretest and Posttest. Based on the results shown in Figure 4-4, the activity utilized as homework was effective in correcting the targeted misconceptions represented by questions 3, 7, 8, and 11. There was a higher percentage of students who answered correctly for 3, 7, 8, and 11 on the Posttest.

Table 4-2. Descriptive statistics and normality tests for the activity as homework group.

<table>
<thead>
<tr>
<th>Activity as Homework</th>
<th>N</th>
<th>Mean (M)</th>
<th>Median (MD)</th>
<th>Standard Deviation (SD)</th>
<th>Skewness</th>
<th>Kurtosis</th>
<th>K-S Statistic</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pretest</td>
<td>12</td>
<td>6.92</td>
<td>8.00</td>
<td>2.234</td>
<td>-0.462</td>
<td>-1.069</td>
<td>0.269</td>
<td>0.016</td>
</tr>
<tr>
<td>Posttest</td>
<td>12</td>
<td>9.08</td>
<td>9.50</td>
<td>2.275</td>
<td>-0.846</td>
<td>-0.072</td>
<td>0.157</td>
<td>0.200</td>
</tr>
</tbody>
</table>

Figure 4-4. The percentage of students who answered correctly on the pre and post inventory for university 2. This group of students used the inventory as homework.

4.8 Comparison of the Homework Groups

There were two different universities that utilized the activity as homework. Neither of the two groups met the assumptions for normality, so nonparametric tests were used to compare the Pretest and Posttest scores of the two universities. According to the Mann-Whitney U test that was conducted, there was no significant difference between
the Pretest scores for University One (MD = 6, N = 29) and University Two (MD = 8, N = 12), (U = 110, z = 1.852, p = 0.068). The lack of significant difference between the Pretest scores for the two universities allows for a comparison of the Posttest scores in order to determine if there is a significant difference. According to the Mann-Whitney U test that was conducted, there was a significant difference between the Posttest scores for University One (MD = 8, N = 29) and University Two (MD = 9.5, N = 12), (U = 103, z = 2.052, p = 0.042). Because there is a significant difference in Posttest scores for University One and University Two, the normalized change in percent correct was compared for both groups.

Figure 4-5 shows that the activity corrected student misconceptions about enzyme-substrate specificity at both universities (as represented by questions 3 and 7). The activity better corrected student misconceptions about possible substrates for enzymes (question 8) and differentiation between active sites and specificity pockets (question 11) at University Two. As shown in Chapter 3, students from University One shifted from the misconception that water cannot be a substrate towards the idea that a large protein cannot be a substrate. They did not shift towards the correct answer, as students from University Two did. Based on the results shown in Figure 4-5, it appears that students at University Two incorporated the phrase “scissile bond” into their vocabulary, as more students from University Two selected the correct answer containing that phrase.
4.9 Discussion and Conclusions

The goal of this project was to determine if the activity was more effective when utilized as homework or as an in-class activity. The results show that the activity is effective at correcting different misconceptions depending on what form is used. The activity utilized as homework is better at improving students’ ability to interpret enzyme-substrate specificity. The activity utilized in class better improved students’ ability to distinguish between an active site and a specificity pocket. It is possible that allowing students the time to explore the representations by themselves allows for students to better analyze those representations. The environment within the classroom may not be conducive to as much student discussion as expected.

One limitation of this study is the size of the samples. The groups at University One are under 50 students each, and the group at University Two is 12 students. Because the activity was only tested at 2 universities, more work needs to be done to determine if

Figure 4-5. The comparison of normalized change for the students at University One and University Two who utilized the activity as homework.
the results are generalizable to other one-semester biochemistry courses. However, this activity does correct the targeted misconceptions at both universities. Even though it appears that students at University One do not move towards the correct answer for question 8, it is shown that students do move away from the incorrect idea that water cannot be a substrate for an enzyme. Instead of directing students towards the correct idea, the activity or the instruction identifies a misconception amongst students that a large protein cannot be a substrate for an enzyme. Both forms of the activity appear to be effective at correcting the targeted misconceptions, but there are some instances where additional misconceptions were identified.

4.10 Implications for Future Research and Practitioners

Most guided inquiry activities that are designed to be used as homework utilized computer simulations as a way to guide students through the activity. Utilization of this activity as homework without a computer simulation opens the door for future creation of activities in a similar style. Even though guided inquiry activities are generally designed to foster discussion and group work, similar correction of misconceptions can be achieved through the use of homework activities.

This activity communicates the targeted information to students effectively. In the future, more testing of the activity needs to be done to determine its effectiveness at correcting misconceptions across the broad biochemistry curriculum. Future work should also investigate any underlying misconceptions, if any should be identified. When one misconception is corrected, others may be found. Future studies will look at the generalizability of the activity to one-semester biochemistry courses at other universities.
4.11 References


Chapter 5: CONCLUSIONS, IMPLICATIONS, AND FUTURE WORK

The goals of this research were to (1) analyze the quality of concept inventories available for biology, chemistry, and biochemistry; (2) develop an activity to improve students’ misconceptions about enzyme-substrate interactions; and (3) compare the effectiveness of a guided inquiry activity as homework and as an in-class activity.

5.1 Concept Inventory Meta-Analysis

While there have been analyses of concept inventories completed in the past, this is the first comprehensive analysis of biology, chemistry, and biochemistry inventories. Concept inventories have become a more popular research topic in science education from 1941 to present. In the span of one year, 51 concept inventories in science, physics, astronomy, biology, chemistry, and biochemistry were collected. Even though physics concept inventories were among the first developed, more and more science disciplines are working on identifying student misconceptions about specific topics. The biology, chemistry and biochemistry concept inventories were analyzed to determine the most commonly reported development methods and the most commonly reported forms of validity and reliability. The majority of concept inventories consider previous work done with regards to student misconceptions in specific topics as well as interviewing the target population to uncover misconceptions when developing items.

The analysis of the biology concept inventories revealed that there are many inventories for genetics, and quite a few of them focus on very specific aspects. However, none of the molecular biology or the biochemistry concept inventories discuss the kinetics of enzymatic reactions or rate order calculations. These are a few of the harder topics in biology courses, and researchers have not taken the time to identify
common misconceptions that students may have. The analysis of the chemistry concept inventories showed that researchers have focused on misconceptions in general chemistry courses. The higher level chemistry courses have not been researched as much, perhaps due to the specialized nature of the courses. However, upper level chemistry courses traditionally teach more difficult concepts. Perhaps researchers are focusing on the base chemistry concepts in the hopes that by ensuring that students learn the correct information from the start, they will learn the correct information in the upper level courses.

After development, best practices call for the instrument to be validated for the specific target population and the reliability of the instrument should be checked as well. According to the meta-analysis performed, face and content validity are the most commonly reported forms of validity. Both face and content validity can be determined for the instrument and require experts in the content area. Just over half of the inventories analyzed reported Cronbach $\alpha$ as their form of reliability for the developed instrument. Cronbach $\alpha$ is reported more often since it can be used for items that are not dichotomous. One common theme identified amongst the inventories analyzed was the suggestion that further analysis of the validity and reliability would be completed by future researchers. It appeared that developers were more interested in completing the concept inventory than in following the standards that are generally agreed upon as the best practices for development. In order for concept inventories to be useful to professors, the analysis should be completed so that the quality of an inventory can be determined. If the inventory identifies misconceptions that are found in most classrooms, the inventory can be used to develop an activity to correct those misconception or
otherwise inform instruction. By creating quality inventories, researchers can help professors adjust their teaching.

5.2 Development and Implementation of a Guided Inquiry Activity

The developed guided inquiry activity did correct the misconceptions of students as a homework assignment. Although the activity did not include a facilitator, as guided inquiry activities are supposed to, students who utilized the activity were more able to differentiate between an active site and specificity pocket. Students were also able to recognize that water is a possible substrate for an enzyme, even though students did not select the correct answer for that specific question on the inventory. They did shift away from the selection that water cannot be a substrate for an enzyme, but they shifted towards the idea that a large protein cannot be a substrate. Based on the results from the given survey, students found the activity to be somewhat helpful and the visual representations of the mechanism were one of the most helpful aspects of the activity.

When the effectiveness of the activity as homework and as an in-class activity is compared, there is a difference in which misconceptions were corrected. When the activity is utilized as homework, students are better able to interpret enzyme-substrate specificity than when traditional lecture is used at University One. Students did move away from the misconception that water cannot be a substrate for an enzyme, but a new misconception was discovered. Students believed that a large protein could not be a substrate for an enzyme. However, the data shows that students were better able to distinguish between an active site and a specificity pocket when the activity was utilized in-class at University One. The presence of a facilitator may have allowed for students to question their prior knowledge more easily than working outside of class. It seems that
students were more able to identify the difference between an active site and a specificity pocket, but the activity did not integrate the phrase “scissile bond” into their vocabulary.

The students who used the activity as homework at University One were then compared to the group of students who used the activity as homework at University Two. Based on the normalized change for both groups, the activity corrected student misconceptions about enzyme-substrate specificity at both University One and University Two when used as a homework assignment. The activity better corrected the misconception that water cannot be a substrate for an enzyme at University Two, and the misconception that a large protein cannot be a substrate for an enzyme was not created based on the item analysis of student answers. Students were better able to differentiate between an active site and a specificity pocket at University Two. Additionally, the activity incorporated the phrase “scissile bond” into student vocabulary at University Two.

The difference in the corrected misconceptions may shed some insight into the time students need to process information. When the activity was given as homework, students were able to process the models and information at their own pace. They could explore Model One in more detail, and that may have helped them better learn enzyme-substrate specificity. However, the students who were able to discuss ideas with each other when the activity was used in class could better differentiate between the active site and specificity pocket for an enzyme. The group discussion may have also encouraged students to use the new vocabulary included in the activity.
5.3 Implications

5.3.1 Instruction in the Classroom

Professors constantly have to adjust their instructional methods for each new class of students. By allowing professors to see the misconceptions students have from their prior classes, professors can shape instruction so that misconceptions can be corrected. Concept inventories provide a way for professors to measure prior knowledge. There are a variety of instruments out there that can be used for formative assessment of students. It can be difficult for people who are new to chemistry education to assess the quality of these instruments. By providing a reference for professors that itemizes the development, validity, and reliability of biology, chemistry, and biochemistry concept inventories, it is more likely that professors will incorporate concept inventories in their classrooms. The creation of an online database hosting links to concept inventories also allows researchers to incorporate research into the classroom.

The language used by educators during instruction plays an important part in transmitting knowledge to students. It has been shown that students do not recall everything discussed in lecture and that different students retain different information. Students learn material through different perspective and social interactions. By fostering a community of discussion, students gain a better understanding of concepts through multiple viewpoints. Guided inquiry activities are one such medium that educators can utilize to encourage student discussion.

In guided inquiry, students become the ones responsible for learning. The burden is not on the educator to provide every single detail related to a concept. Students have the opportunity to explore models and invent concepts in their own language. Guided
inquiry traditionally requires a facilitator to ensure that students stay on track and to provide guidance when there are questions. The use of guided inquiry as homework removes that aspect, as well as the student interaction. However, the scaffolding and the opportunity to create concepts in their own vocabulary appears to work better than traditional lecture to teach material. As educators look for ways to better engage students in the classroom, guided inquiry provides an alternative instructional method that has been proven to work.

5.3.2 Chemistry Education Research

As research into biochemistry and chemistry education increases, there is a need to “bridge the gap” between research and practice. Understanding how students learn cannot be useful unless it is implemented in the classroom. Guided inquiry activities can be used to challenge incorrect prior knowledge that students may have and allow for the incorporation of correct knowledge through engaging students. Every student learns different material from the same representation. Student discussion can lead to a more complete understanding of the representation because they will be forced to look at the image from different perspectives.

Guided inquiry requires facilitation and discussion, but both of these aspects are removed when guided inquiry is used outside the classroom. It is possible that the scaffolding of questions is what allows students to think more critically and incorporate new knowledge into their long-term memory. By allowing students to take the activity home and work through it at their own pace, it may allow a more thorough investigation of the models. The Enzyme-Substrate Interactions activity could be used to correct misconceptions in not only biochemistry classrooms, but also in students’ dorm rooms.
5.4 Future Work

The collection of concept inventories was used to create an online database to provide educators with a central location for concept inventories. Often teachers and professors do not know that these tools exist or they have no idea where to find them. The collection of concept inventories also allows future researchers to see where there is a gap in the literature. Additionally, by making concept inventories more available to educators, researchers can determine if the misconceptions in each inventory are even more common across the world.

During the initial interviews, students were adamant that water could not be a substrate for enzymes, but could not provide an explanation. Water is shown as a substrate for many enzymatic reactions, but may not be referred to as a substrate during instruction or in the textbook used in the course. Since this misconception was so common among students, identifying the source of this incorrect idea might help to correct the idea earlier. In general chemistry, students are taught that acid-base reactions produce water. It is possible that this leaves students with the impression that water is a product, not a reactant. Tracing this misconception to the source could help to improve students’ understanding earlier in their science career.

Guided inquiry activities have rarely been utilized as homework in the literature, and even more rarely used without an interactive component on a computer. It is the author’s hope that this investigation will open the door into future development of guided inquiry activities for use outside of the classroom. However, homework is typically used as a summative assessment of students’ understanding instead of formative assessment. Through the use of guided inquiry activities, professors can correct misconceptions and
check student understanding. It is a checkpoint for student knowledge, and a way to engage students in chemistry.
September 15, 2014

Ms. Ellen Humphreys
Kennesaw State University
Department of Chemistry and Biochemistry
370 Paulding Avenue NW, MD#1203
Kennesaw, GA 30144

Dear Ms. Humphreys:

As co-chair of the Institutional Review Board (IRB), I have reviewed your proposal, Improving Biochemistry Students’ Understanding of Enzyme-Substrate Interactions, (Submission #090714), and the committee has granted your proposal exemption.

If you have any questions, please feel free to contact me at 516-877-4344 or e-mail me at altman@adelphi.edu.

Sincerely,

Robert Otto, Ph.D., Co-Chair
Adelphi University Institutional Review Board

Dr. Brian Stockman
Dear Ms. Humphreys:

I have reviewed your application for the new study listed above. This study qualifies as exempt from continuing review under DHHS (OHRP) Title 45 CFR Part 46.101(b)(1) - effectiveness of instructional techniques and (2) - educational tests, surveys, interviews, public observations. The consent procedures described are in effect. You are free to conduct your study.

Please note that all proposed revisions to an exempt study require IRB review prior to implementation to ensure that the study continues to fall within an exempted category of research. A copy of revised documents with a description of planned changes should be submitted to irb@kennesaw.edu for review and approval by the IRB.

Thank you for keeping the board informed of your activities. Contact the IRB at irb@kennesaw.edu or at (678) 797-2268 if you have any questions or require further information.

Sincerely,

Paula Strange, Assistant Director for Research Compliance
KSU Institutional Review Board
6/27/2014

Ellen Humphreys, Student
KSU Department of Chemistry and Biochemistry

RE: Your application dated 6/10/2014, Study #14-447: Improving Biochemistry Students' Understanding of Enzyme-substrate Interactions

Dear Ms. Humphreys:

Your application for the new study listed above has been administratively reviewed. This study qualifies as exempt from continuing review under DHHS (OHRP) Title 45 CFR Part 46.101(b)(2) - educational tests, surveys, interviews, public observations. The consent procedures described within your application are in effect. You are free to conduct your study.

Please note that all proposed revisions to an exempt study require IRB review prior to implementation to ensure that the study continues to fall within an exempted category of research. A copy of revised documents with a description of planned changes should be submitted to irb@kennesaw.edu for review and approval by the IRB.

Thank you for keeping the board informed of your activities. Contact the IRB at irb@kennesaw.edu or at (678) 797-2268 if you have any questions or require further information.

Sincerely,

Christine Ziegler, Ph.D.
KSU Institutional Review Board Chair
9/5/2014

Ellen Humphreys, Student
KSU Department of Chemistry and Biochemistry


Dear Ms. Humphreys:

Your application for the new study listed above has been administratively reviewed. This study qualifies as exempt from continuing review under DHHS (OHRP) Title 45 CFR Part 46.101(b)(2) - educational tests, surveys, interviews, public observations. The consent procedures described in your application are in effect. You are free to conduct your study at Kennesaw State University.

Please note that all proposed revisions to an exempt study require IRB review prior to implementation to ensure that the study continues to fall within an exempted category of research. A copy of revised documents with a description of planned changes should be submitted to irb@kennesaw.edu for review and approval by the IRB.

Thank you for keeping the board informed of your activities. Contact the IRB at irb@kennesaw.edu or at (678) 797-2268 if you have any questions or require further information.

Sincerely,

Christine Ziegler, Ph.D.
KSU Institutional Review Board Chair
Appendix B: Study Consent Form, Demographic Surveys, Interview Consent Form, Interview Protocol, and Surveys about the Activity

Improving Biochemistry Students' Understanding of Enzyme-Substrate Interactions
Informed Consent Form and Participant Survey

Introduction
The purpose of this study is to conduct a thorough investigation into the effectiveness of traditional lecture-based classes in correcting student misconceptions relating to enzyme-substrate interactions. We are interested in learning which misconceptions are corrected through this method of teaching and which misconceptions remain.

The study consists of two stages, and you may be participating in one or both. The first stage consists of a survey that will be given to determine misconceptions that remain after the instruction of the enzyme unit. After this survey is given, individual interviews will be given to determine the validity of this survey as a tool for identifying misconceptions in biochemistry classrooms. The data gathered from these survey responses and validity interviews will be used to create, pilot, and validate a large-scale suite of activities that will be distributed nationally throughout the chemistry community. You are being invited to participate in this study because you are taking a survey of biochemistry course, CHEM 3500.

Description of Procedures
If you agree to participate in stage one of this study, you will agree to have your answers to the enzyme-substrate interactions survey included in the study. You will take this survey as part of your regular course assessment and is expected to require 10-15 minutes to complete. This survey contains 15 questions about enzyme-substrate interactions. The survey will be administered during class time and will count toward your quiz grade. However, choosing to include your survey in the research study will not influence your grade in the course. During class, you can expect the following procedures to be followed:

1. You will be asked to complete a survey regarding your understanding of enzyme-substrate interactions at the beginning of the course.
2. You will be asked to take the same survey two weeks after the enzyme unit has been covered by an exam.
3. You will be asked to provide basic demographic information such as age, gender, race/ethnicity, and class year. You may skip any question that you do not wish to answer or that makes you feel uncomfortable.

Risks
There are no known physical, psychological, social, or medical risks associated with this research. Any information is confidential, and if this work is to be published, your name will not be used.

Benefits
If you decide to participate in this study there will be no direct benefit to you. The researcher will learn more about how professors teach enzyme-substrate interactions. The research will also help the researcher develop a suite of activities to better correct students' misconceptions.

Costs and Compensation
You will not incur any costs from participating in this study. The professor will not offer compensation for participation in this study.
Participant Rights
Your participation in this study is completely voluntary and you may refuse to participate or leave
the study at any time. If you decide to not participate in the study or leave the study early, it will
not result in any penalty or loss of benefits to which you are otherwise entitled.

Confidentiality
Records identifying participants will be kept confidential to the extent permitted by applicable
laws and will not be made publicly available. However, auditing departments of Kennesaw State
University, and the Institutional Review Board (a committee that reviews and approves human
subject research studies) may inspect and/or copy your records for quality assurance and data
analysis. These records may contain private information.

To ensure confidentiality to the extent permitted by law, the following measures will be taken:
You will be asked to provide basic demographic information. All data will be stored in a locked
filing cabinet in the research laboratory at Kennesaw State University. These data will be kept
indefinitely for analysis. If the results are published, your identity will remain confidential.

Inclusion Criteria for Participation
You must be enrolled at students at Kennesaw State University and enrolled in the CHEM 3500
course as students. You must be at least 18 years of age.

Questions or Problems
You are encouraged to ask questions at any time during this study.
• For further information about the study contact Dr. Kimberly Linenberger by phone at
  (770) 423-6278 or by email at klinenbe@kennesaw.edu, or Ellen Humphreys by email at
  ehumphr6@kennesaw.edu.
• If you have questions about the rights of research subjects or research-related injury,
  please contact the IRB Administrator, (770) 423-6738, irb@kennesaw.edu, or Director,
  (678) 797-2268, Office of Research, Kennesaw State University, Kennesaw, Georgia 30144.

Participant Signature
Signing your name below indicates that you voluntarily agree to participate in this study, that the
study has been explained to you, that you have been given the time to read the document, and
that your questions have been satisfactorily answered.

I agree to participate in this research study regarding what students know about enzyme-
substrate interactions. The information I provide may be used for additional research or
publications; however, since my name is not used, my identity is protected. I have had the
opportunity to ask any questions I might have about my participation in this study and they have
been answered to my satisfaction. By signing my name below, I certify that I have read the consent
form, agree to participate in this study and I confirm that I am at least 18 years of age. I reserve the
right to request that my scores not be included in the study at any time during the study.

_________________________   _____________________
Signature        Date

_________________________
Printed Name
Demographic Survey

1. I would best describe myself as (check only one):
   ____ Freshman   ____ Sophomore   ____ Junior   ____ Senior   ____ Graduate Student

2. What is your gender?
   ____ Female   ____ Male

3. What is your race/ethnicity?
   ____ African American/Black
   ____ American Indian/Alaska Native
   ____ Asian/Pacific Islander
   ____ Hispanic
   ____ White/Caucasian
   ____ Other (please specify): _______________________________________________

4. What is your age? _____________________

5. What is your major? ___________________________

6. How many credits are you taking this semester? ___________

7. Would you be willing to take part in an interview that will last approximately 30 minutes at a mutually convenient time after the survey is given the second time? During the interview your responses will be audio recorded. Your participation in the interview is completely voluntary and will in no way affect your grade in CHEM 3500. Depending on the number of students who volunteer for the interview, it may not be possible to interview all volunteers, in which case a sample will be chosen.
   If you do not wish to participate in the interview, please leave your email address blank.
   If you are willing to participate in the interview, please fill in your email address.

   E-mail address: _________________________
Improving Students’ Understanding of Enzyme-Substrate Interactions

Interview Consent Form

I understand that I am being asked to participate in an interview that will last approximately thirty minutes. I understand that I will be asked questions about my understanding of specific questions of enzyme-substrate interactions and my response to those questions. I understand that the researcher is not going to correct me if I have an incorrect understanding of the concept and any head nodding or silence on the part of the researcher does not imply that I am correct. The information gathered from this interview will be used as part of a larger project that is attempting to understand students’ understanding of enzyme-substrate interactions.

I understand that all personally identifiable information will be kept strictly confidential and will not appear in any reports generated using the information gathered from the interview. I understand that participation in this study is completely voluntary. I do not have to answer any questions I do not want to and can stop the interview at any time and withdraw from the study. Withdrawing from the study will in no way affect my grade in CHEM 3500. My professor for CHEM 3500 will not know if I participate.

I understand that participation in this study will require about 30 minutes of my time during the spring 2014 semester. I understand that at the end of the project, I may be asked to review the findings from my interview. I give my permission for the interview to be audio recorded. I understand that the purpose of the recording is to assure that what I say is represented accurately in the research process. I understand that the audio recording along with any work I generate (e.g., drawings) will be kept by the researcher and destroyed after 2021. I understand that the audio recordings along with any work I generate could be shown at conferences and/or reprinted in articles detailing the results of the study.

I have had the opportunity to ask any questions I might have and they have been answered to my satisfaction. By signing below, I agree to participate in the interview and I confirm that I am at least 18 years of age.

If you have any further questions or concerns, please feel free to contact: Ellen Humphreys, Department of Chemistry and Biochemistry, 1000 Chastain Road MD# 1203, Kennesaw, Georgia 30144 or email at ehumphr6@kennesaw.edu or my research advisor, Dr. Kimberly Linenberger at klinenbe@kennesaw.edu. If you have questions about your rights as human subjects contact the Office of Research (770) 423-6738 or irb@kennesaw.edu.

_____________________________________   ___________ ______
Research Participant                      Date

_____________________________________   _________________
Researcher                                 Date

_____________________________________   _________________
Researcher                                 Date
Enzyme-Substrate Interaction Concept Inventory
Student Validity Interview Guide

If you remember, during the past week, you took the Enzyme-Substrate Interaction Concept Inventory (give interviewee copy of inventory) and I would now like to talk with you about a few of the items on it.

- The Inventory will be divided into sets of 3 questions. Have interviewee read aloud and answer each set of questions explaining why they answered the way they did as they do so. If the interviewee realizes that he/she misread the statement, the interviewer will cross their answer out with a single line and circle the new answer. If interviewee changes their mind as they explain their answers, the interviewer will completely darken out the old answer and circle the new.

*Question Sets:*
Set 1: 1, 2, 3  Set 4: 10, 11, 12
Set 2: 4, 5, 6  Set 5: 13, 14, 15
Set 3: 7, 8, 9

For each set of questions the following questions will be asked.

- Of these questions, are there any that you find confusing? Why?
- Of these questions, is there a question that “best” represents the specific concept? Why?
- Were any topics omitted from this set that you feel need to be included to best represent the specific concept? Why?

After all question sets have been discussed the following questions will be asked.

- Are there any other Concept Inventory items that stand out that you would like to discuss with me? Why?
- Were any concepts left off of the inventory that you feel need to be included to best represent your understanding of enzyme-substrate interactions?
Activity Questionnaire

1. Did you do the Enzyme-Substrate Interactions Activity?
   ___ Yes
   ___ No

2. How long did you spend doing the activity?
   ___ I didn’t do the activity
   ___ 30 minutes to 1 hour
   ___ 1-2 hours
   ___ 2-3 hours
   ___ 3-4 hours
   ___ More than four hours

3. Did you work with anyone on this activity?
   ___ No
   ___ Yes, friends from class
   ___ Yes, friends who have taken the class before

4. Which, if any, of the following resources did you use to help complete the activity?
   ___ None
   ___ Textbook
   ___ Wikipedia
   ___ Google
   ___ Google Scholar
   ___ Powerpoints from class (if applicable)
   ___ Other:

5. On a scale of 1-5, with 1 being not helpful at all and 5 being very helpful, how helpful do you feel that the activity was with teaching you about enzyme-substrate interactions?
   ___ I did not do the activity
   ___ 1 – The activity was not helpful at all.
   ___ 2 – The activity was not very helpful.
   ___ 3 – I have no feeling about whether the activity was helpful or not.
   ___ 4 – The activity was somewhat helpful.
   ___ 5 – The activity was very helpful.

6. Are there any other comments you have on the activity?
Views towards New Activities

For each of the following questions please answer in regards to the new activities that you just completed. Please be as honest as possible as it is very valuable to the development and improvement of the activities.

1. **Strengths**—identify the ways in which the activities were of high quality and commendable. Each strength statement should address what was valuable in the experiment and why this attribute is important.
   1. 
   2. 
   3. 

2. **Areas for Improvement**—identify the changes that can be made in the future, between this semester and next semester, that are likely to improve these activities. Improvements should recognize the issues that caused any problems and mention how changes could be implemented to resolve these difficulties.
   1. 
   2. 
   3. 

3. **Insights**—identify new and significant discoveries/ understandings that were gained concerning the activities. Insights include why a discovery/new understanding is important or significant and how it can be applied to other situations.
   1. 
   2. 
   3. 

4. **Additional Topics**—identify topics covering enzyme-substrate interactions that you feel should be included in the activities to best address your understanding of the topic.
   1. 
   2. 
   3.
Appendix C: Enzyme Substrate Interactions Activity

Enzyme-Substrate Interactions

Students should work in groups of no more than 4 students and go through the activity together during class time.

Outcomes:
1. Identify water as a substrate for enzymes.
2. Differentiate between an active site and a specificity pocket and identify possible substrates based on that knowledge.
3. Select substrates for enzymes based on knowledge of geometric and electronic complementarity.

Model 1:
Chymotrypsin is an enzyme classified as a serine protease, meaning that it breaks down proteins. Chymotrypsin has both an active site and a specificity pocket, as do all serine proteases. The active site is where reactions with a substrate occur, and is conserved across all serine proteases. The specificity pocket helps to line the proper cleavage point up in the active site. Large, uncharged amino acids fit in the specificity pocket of chymotrypsin to position the following amino acids properly in the active site. The peptide is cleaved at the carbonyl group of the amino acid in the active site.

Oxyanion Hole – a region that stabilizes a negatively charged oxygen on the substrate
Scissile Bond – the bond that is cleaved within a protein

For Figure 1, put a star by the bonds being broken and circle the bonds being formed.

Figure 1
1. Label the active site and specificity pocket in Figure 1A and the oxyanion hole in Figure 1B based on the location of the substrate and the information in the above paragraph.

2. Which amino acids are in the active site?

3. What do the solid lines and the dashed lines represent?

Note: The three amino acids in the active site of chymotrypsin are called the catalytic triad. The catalytic triad is conserved across all serine proteases and is where the name for this class of enzymes originates.

4. In one sentence, describe what happens in the first step of this reaction.

5. How has the molecular geometry of the carbon that is now bound to Serine 195 changed?
Note: The steps between the initial substrate and the final product have temporary structures. These are called intermediates. When identifying the intermediates for the chymotrypsin mechanism, the geometry of the carbon bound to Serine 195 determines the name of the intermediate. The intermediate shown in Figure 1B is referred to as a tetrahedral intermediate due to the molecular geometry of the carbon bound to Serine 195. The backbone amines on the Glycine 193 and the Serine 195 help to stabilize the substrate once it has interacted with the active site and the specificity pocket.

For Figure 2, put a star by the bonds being broken and circle the bonds being formed.

6. In one sentence, describe what happens in the second step of this reaction.

7. What new functional groups are being created by the bond cleavage?
8. Based on the definitions presented at the beginning of this activity, what is the name of the bond that will be cleaved in the reaction depicted in Figures 2A and 2B?

9. How has the molecular geometry of the carbon that is now bound to Serine 195 changed from Figure 2A to Figure 2B?

Note: The intermediate shown in Figure 2B is referred to as an acyl-enzyme intermediate due to the change in the functional group on the carbon bound to Serine 195. The carbon bound to Serine 195 is now an acyl group due to the double-bonded oxygen and the presence of two R-groups, one of which is the serine of the active site of chymotrypsin, and the other being the remainder of the original peptide.

For Figure 3, put a star by the bonds being broken and circle the bonds being formed.

Figure 3
10. What happened to the C-terminus of the substrate?

11. Has the intermediate shown in Figure 3A changed from the intermediate shown in Figure 2B?

12. In one sentence, describe what is happening in this step of the reaction.

13. What is the substrate that is utilized in this step of the reaction?

14. What type of interaction does the utilized substrate have with Histidine 57 in Figure 3B?
For Figure 4, put a star by the bonds being broken and circle the bonds being formed.

Figure 4

15. In one sentence, describe what is happening in this step of the reaction.

16. Based on your understanding of the intermediates in Figures 1B and 2B, how would you categorize the intermediate shown in Figure 4B?
For Figure 5, put a star by the bonds being broken and circle the bonds being formed.

Figure 5

17. In one sentence, describe what is happening in this step of the mechanism.

18. Based on your understanding of a protease, identify the substrate(s) for the overall mechanism of chymotrypsin based on Figures 1-5.

19. Based on your answer to question 18, what type of reaction does chymotrypsin catalyze?

20. Draw the two products that are released from the entire reaction shown in Figures 1-5.
21. In 3 sentences (or bullet points), describe the entire reaction that occurred in these diagrams. Make sure to discuss the role of the catalytic triad in the active site.

Model 2:
While all serine proteases have a specificity pocket, they differ in amino acid composition within the specificity pocket. The amino acids in the specificity pocket interact with different substrates based on charge and size. The specificity pocket of chymotrypsin helps to line the proper cleavage point up in the active site. Large, uncharged amino acids at any point in the amino acid chain when sterically favorable can fit in the specificity pocket of chymotrypsin to position the following amino acids properly in the active site. The peptide is cleaved at the scissile bond which is after the carbonyl group of the amino acid in the specificity pocket.

22. Chymotrypsin is an enzyme that has substrate specificity. What determines whether or not a substrate will bind in the active site?

23. Which of the following could cause the interaction between phenylalanine and the amino acids in the specificity pocket as shown in Figure 6? Circle your answer and
explain why you chose that option. Explain why you did not choose each of the other options.

a) Electrostatic interactions (interaction between + and -)

b) Hydrogen bonds

c) Dipole-dipole interactions

d) London dispersion forces/Van der Waals

24. Provide a rationale for why or why not each of the following amino acid “R” groups will or will not interact with the amino acids in the specificity pocket of chymotrypsin.

25. Based on your knowledge of the specificity pocket of chymotrypsin, pick which one(s) of the following molecules will be substrates for chymotrypsin and explain why.
   B. M-N-T-T-R-K-V-K-E-G
   C. M-S-Q-S-M-D-Y-K-D-Q
   D. K-I-Q-S-N-Q-R-G-A-V-M

26. What other substrate(s) will be needed for the reaction to occur?
27. The following peptide is not a substrate for chymotrypsin. Provide possible reasons why it is not.


Model 3:

28. How is the amino acid composition of the specificity pocket shown in Figure 6 different from the catalytic triad that makes up the active site shown in Figures 1-5?

29. Do reactions, interactions, or both occur in the specificity pocket?

30. Do reactions, interactions, or both occur in the active site?

31. What is the difference between a chemical reaction and an interaction between molecules?
Homework:

Application

Figure 7

Trypsin Specificity Pocket

1. Figure 7 shows the specificity pocket of trypsin, another serine protease. What is the difference between the specificity pocket of trypsin as shown here and the specificity pocket of chymotrypsin as shown in Figure 6?

2. Which amino acids will form the strongest interactions with the specificity pocket of trypsin at physiological pH? Explain why.

3. Since trypsin is a serine protease, what part of the enzyme is conserved?

4. Based on your knowledge of the active site and specificity pocket of trypsin and the reaction for chymotrypsin, describe in 3 sentences the reaction for trypsin.
Appendix D: Facilitation Guide for the Enzyme-Substrate Interactions Activity

**Enzyme-Substrate Interactions**

Students should work in groups of no more than 4 students and go through the activity together during class time.

**Outcomes:**
4. Identify water as a substrate for enzymes.
5. Differentiate between an active site and a specificity pocket and identify possible substrates based on that knowledge.
6. Select substrates for enzymes based on knowledge of geometric and electronic complementarity.

**Model 1:**
Chymotrypsin is an enzyme classified as a serine protease, meaning that it breaks down proteins. Chymotrypsin has both an active site and a specificity pocket, as do all serine proteases. The active site is where reactions with a substrate occur, and is conserved across all serine proteases. The specificity pocket helps to line the proper cleavage point up in the active site. Large, uncharged amino acids fit in the specificity pocket of chymotrypsin to position the following amino acids properly in the active site. The peptide is cleaved at the carbonyl group of the amino acid in the active site.

**Oxyanion Hole** – a region that stabilizes a negatively charged oxygen on the substrate
**Scissile Bond** – the bond that is cleaved within a protein

For Figure 1, put a star by the bonds being broken and circle the bonds being formed.

Figure 1
32. Label the active site and specificity pocket in Figure 1A and the oxyanion hole in Figure 1B based on the location of the substrate and the information in the above paragraph.

33. Which amino acids are in the active site?
Aspartic acid, Histidine, and Serine (Asp, His, and Ser)

34. What do the solid lines and the dashed lines represent?
The solid line represent single bonds/covalent bonds and the dashed lines represent hydrogen bonds.

Note: The three amino acids in the active site of chymotrypsin are called the catalytic triad. The catalytic triad is conserved across all serine proteases and is where the name for this class of enzymes originates.

35. In one sentence, describe what happens in the first step of this reaction.
In the first step of this reaction, a substrate has entered the enzyme and has bonded/interacted with the amino acids in the active site as well as in the oxyanion hole.
36. How has the molecular geometry of the **carbon** that is now bound to Serine 195 changed?

There were 3 things bonded to the carbon and now there are four things bonded to it. It went from trigonal planar to tetrahedral.

**Note:** The steps between the initial substrate and the final product have temporary structures. These are called intermediates. When identifying the intermediates for the chymotrypsin mechanism, the geometry of the carbon bound to Serine 195 determines the name of the intermediate. The intermediate shown in Figure 1B is referred to as a tetrahedral intermediate due to the molecular geometry of the carbon bound to Serine 195. The backbone amines on the Glycine 193 and the Serine 195 help to stabilize the substrate once it has interacted with the active site and the specificity pocket.

For Figure 2, put a star by the bonds being broken and circle the bonds being formed.

![Figure 2](image)

37. In one sentence, describe what happens in the second step of this reaction.
In the second step of this reaction, the bond between the amide and the carboxyl group of the substrate in the active site is broken, and the histidine donates a proton to the leaving part of the substrate.

38. What new functional groups are being created by the bond cleavage?
An amino group (the leaving group) and an ester are created

39. Based on the definitions presented at the beginning of this activity, what is the name of the bond that will be cleaved in the reaction depicted in Figures 2A and 2B?
Scissile bond/peptide bond (preferred scissile bond, so redirect them to look back at the definitions from the beginning of the activity)

40. How has the molecular geometry of the carbon that is now bound to Serine 195 changed from Figure 2A to Figure 2B?
The carbon now only has 3 things bound to it again. So it changed from tetrahedral back to trigonal planar.

**Note:** The intermediate shown in Figure 2B is referred to as an acyl-enzyme intermediate due to the change in the functional group on the carbon bound to Serine 195. The carbon bound to Serine 195 is now an acyl group due to the double-bonded oxygen and the presence of two R-groups, one of which is the serine of the active site of chymotrypsin, and the other being the remainder of the original peptide.

For Figure 3, put a star by the bonds being broken and circle the bonds being formed.

Figure 3
41. What happened to the C-terminus of the substrate?
It left the active site.

42. Has the intermediate shown in Figure 3A changed from the intermediate shown in Figure 2B?
No

43. In one sentence, describe what is happening in this step of the reaction.
In this reaction, water is entering the active site of the enzyme and hydrogen bonding with the N on histidine.

44. What is the substrate that is utilized in this step of the reaction?
Water

45. What type of interaction does the utilized substrate have with Histidine 57 in Figure 3B?
Hydrogen bonds/Intermolecular forces (preferably hydrogen bonds, so redirect them to what the dashed lines represent)
46. In one sentence, describe what is happening in this step of the reaction. In this step of the reaction, the water gave a proton/an H to histidine so it could bond with Serine 195 and the OH bonded with the carboxyl group left from the substrate, as did Glycine 193.

47. Based on your understanding of the intermediates in Figures 1B and 2B, how would you categorize the intermediate shown in Figure 4B?
It has 4 things bonded to it, so it looks like the tetrahedral intermediate from Figure 1.

For Figure 5, put a star by the bonds being broken and circle the bonds being formed.

Figure 5

48. In one sentence, describe what is happening in this step of the mechanism.
In this step of the mechanism, the hydrogen bonds between the Glycine 193, the Serine 195 and the carboxyl group break, and the Serine 195 bond to the carbon of the carboxyl group breaks.

49. Based on your understanding of a protease, identify the substrate(s) for the overall mechanism of chymotrypsin based on Figures 1-5.
A peptide/protein and water

50. Based on your answer to question 18, what type of reaction does chymotrypsin catalyze?
There is water involved in the reaction, so a hydrolysis reaction.
51. **Draw** the two products that are released from the entire reaction shown in Figures 1-5.

52. In 3 sentences (or bullet points), describe the entire reaction that occurred in these diagrams. Make sure to discuss the role of the catalytic triad in the active site.

A substrate entered the active site of the enzyme and was bound by the Serine 195 in the active site and the Glycine 193 and Serine 195 amide groups in the oxyanion hole. The amide/carboxyl/scissile bond was cleaved, the cleaved part of the substrate left, and water entered the active site. The water was used to regenerate the enzyme so that it looked like it did in the beginning of the reaction, and then the rest of the substrate left after the OH from the water was bonded onto the carbonyl group.

**Model 2:**

While all serine proteases have a specificity pocket, they differ in amino acid composition within the specificity pocket. The amino acids in the specificity pocket interact with different substrates based on charge and size. The specificity pocket of chymotrypsin helps to line the proper cleavage point up in the active site. Large, uncharged amino acids at any point in the amino acid chain when sterically favorable can fit in the specificity pocket of chymotrypsin to position the following amino acids properly in the active site. The peptide is cleaved at the scissile bond which is after the carbonyl group of the amino acid in the specificity pocket.

![Chymotrypsin specificity](image)

53. Chymotrypsin is an enzyme that has substrate specificity. What determines whether or not a substrate will bind in the active site?
54. Which of the following could cause the interaction between phenylalanine and the amino acids in the specificity pocket as shown in Figure 6? Circle your answer and explain why you chose that option. Explain why you did not choose each of the other options.

e) Electrostatic interactions (interaction between + and -)
No because phenylalanine is not charged. It is polar, but at physiological pH, it will not be charged.

f) Hydrogen bonds
No because both amino acids will have hydrogens at the ends, so they won’t bond to each other.

g) Dipole-dipole interactions
No because the ring on phenylalanine is made of just carbon and hydrogen, so it would not have a dipole moment, induced or otherwise

h) London dispersion forces/Van der Waals
Yes because London dispersion forces/Van der Waals are just interactions. If two molecules get close enough, there will be some sort of interaction.

55. Provide a rationale for why or why not each of the following amino acid “R” groups will or will not interact with the amino acids in the specificity pocket of chymotrypsin.

\[
\begin{align*}
\text{[Structure image]} \\
\text{No – too small to fit all the way in the pocket} & & \text{Yes – it will hydrogen bond with the serine and it is large and uncharged}
\end{align*}
\]

56. Based on your knowledge of the specificity pocket of chymotrypsin, pick which one(s) of the following molecules will be substrates for chymotrypsin and explain why.
A and C will be substrates because they contain large uncharged amino acids that will fit in the specificity pocket.

57. What other substrate(s) will be needed for the reaction to occur?

Water

58. The following peptide is not a substrate for chymotrypsin. Provide possible reasons why it is not.


It doesn’t have a large hydrophobic amino acid present.

Model 3:

59. How is the amino acid composition of the specificity pocket shown in Figure 6 different from the catalytic triad that makes up the active site shown in Figures 1-5?
The specificity pocket has 3 different amino acids than the active site, but they both have 3 amino acids

60. Do reactions, interactions, or both occur in the specificity pocket?
Interactions

61. Do reactions, interactions, or both occur in the active site?
Both reactions and interactions

62. What is the difference between a chemical reaction and an interaction between molecules?
A reaction changes the chemical composition of the substrate and an interaction just allows two molecules to stay near each other.
Homework:

Application

Figure 7

Trypsin Specificity Pocket

5. Figure 7 shows the specificity pocket of trypsin, another serine protease. What is the difference between the specificity pocket of trypsin as shown here and the specificity pocket of chymotrypsin as shown in Figure 6? Chymotrypsin has a serine in the middle of the specificity pocket and trypsin has an aspartic acid.

6. Which amino acids will form the strongest interactions with the specificity pocket of trypsin at physiological pH? Explain why. Lysine and Arginine will form the strongest interactions with the specificity pocket of trypsin because they are positively charged at physiological pH.

7. Since trypsin is a serine protease, what part of the enzyme is conserved? The active site is conserved.

8. Based on your knowledge of the active site and specificity pocket of trypsin and the reaction for chymotrypsin, describe in 3 sentences the reaction for trypsin. The substrate will enter the active site of trypsin and either lysine or arginine will interact with the specificity pocket. The substrate will be cleaved in at the bond between the amide group or the carboxyl group of the amino acid in the active site and then the cleaved part will leave. Water enters the active site (as the next substrate),
bonds with the carboxyl group of the amino acid in the specificity pocket and the histidine of the active site, and then the leftover substrate leaves the enzyme.